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Inventory

Herpetofauna from a protected area situated in a biogeographic transition zone in Central South America

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Abstract: Several herpetofaunal inventories have been conducted in the Neotropical region. However, many areas remain to be investigated, and this is particularly true of transition zones between distinct ecoregions. Herein we describe the richness, species composition, and abundance of herpetofauna from a scarcely known portion of the Brazilian Cerrado and assess the taxonomic similarities of the assemblages among the sampled habitats and between neighbouring ecoregions. A mid-term herpetological inventory was conducted from May 2009 to January 2011 at Serra das Araras Ecological Station (SAES) using pitfall traps with drift fences in five distinct forested and open habitats. In these and in additional habitats, visual and acoustic searches and occasional encounters were also used to access the local composition of the herpetofauna, together with literature data and examination of specimens deposited in collections. We also compared the species composition at SAES on a regional scale with those of other 29 localities previously studied in nine South American ecoregions. We documented 123 species at the SAES (39 amphibians and 84 reptiles), 112 of which were recorded during the field inventory. Richness was highest in riparian forests and lowest in semi-deciduous dry forest and cerrado woodland. Riparian forests also presented the highest number of exclusive species, while only one exclusive species was found in cerrado woodland. Cerrado *sensu stricto* and cerrado parkland contained 53 and 40 species, including seven and 10 exclusive species, respectively, and showed greater similarity than the sampled forested habitats. In comparison to other localities in the Cerrado (including transition zones), SAES is home to one of the most diverse assemblages of herpetofauna. This may be attributed to continued sampling efforts and to the marked environmental heterogeneity resulting from the topographic profile and confluence of several ecoregions. Amphibian species composition at SAES is more similar to a nearby locality also studied in the Cerrado and to Chiquitano Dry Forest assemblages, while the reptile species composition is nested among the assemblages studied in the Cerrado. Our findings indicate that the herpetofauna of SAES is rich and representative of the regional biodiversity, with species composition evidencing its transitional character.
Keywords: *Anura; biodiversity; Brazilian savanna; ecotone; Reptilia.*

Herpetofauna de uma área protegida situada em zona de transição na porção central da América do Sul

Resumo: Vários inventários da herpetofauna têm sido conduzidos na região Neotropical. No entanto, muitas áreas permanecem por investigar, e isto é particularmente verdade nas zonas de transição entre ecorregiões. Aqui descrevemos a riqueza, composição de espécies e abundância da herpetofauna de uma porção pouco conhecida do

Cerrado brasileiro e avaliamos as semelhanças taxonômicas das assembleias entre os habitats amostrados e entre ecorregiões vizinhas. Um inventário herpetológico de médio prazo foi realizado entre maio de 2009 e janeiro de 2011 na Estação Ecológica Serra das Araras (SAES), por meio de armadilhas de interceptação e queda em cinco hábitats florestados e abertos. Nestes e em habitats adicionais, também foram utilizadas buscas visuais e acústicas e encontros ocasionais para acessar a composição da herpetofauna, juntamente com dados de literatura e exame de exemplares depositados em coleções. Também comparamos a composição de espécies do SAES em escala regional com aquelas de outras 29 localidades previamente estudadas em nove ecorregiões sul-americanas. Documentamos 123 espécies no SAES (39 anfíbios e 84 répteis), das quais 112 foram registradas durante os inventários de campo. A riqueza de espécies foi maior na mata ciliar e menor na floresta seca semidecídua e cerradão. Matas ciliares também apresentaram maior número de espécies exclusivas, enquanto apenas uma espécie exclusiva foi encontrada no cerradão. Cerrado *sensu stricto* e parque cerrado apresentaram 53 e 40 espécies, incluindo sete e dez espécies exclusivas, respectivamente, e mostraram maior similaridade entre si do que com os habitats florestados amostrados. Em comparação com outras localidades já inventariadas no Cerrado, a SAES abriga uma das mais diversas assembléias da herpetofauna. Isto pode ser atribuído a esforços de amostragem continuados e à acentuada heterogeneidade ambiental resultante do perfil topográfico e da confluência de diversas ecorregiões. A composição de espécies de anfíbios no SAES é mais semelhante a uma localidade próxima também estudada no Cerrado e a assembleias da Floresta Seca de Chiquitano, enquanto a composição de espécies de répteis está aninhada entre as assembleias estudadas no Cerrado. Nossos resultados indicam que a herpetofauna do SAES é rica e representativa da biodiversidade regional, com composição de espécies que evidencia seu caráter transitório.

Palavras-chave: *Anura; biodiversidade; ecótono; Reptilia; savana brasileira.*

Introduction

Faunal inventories are direct and reliable sources of information to increase the body of knowledge of different dimensions of biodiversity (Byrne et al. 2016, Karlsson et al. 2020, Vergara-Asenjo et al. 2023). Species lists resulting from inventories can be used in both theoretical (e.g., analyses of biogeographical relationships, as in Nogueira et al. 2011, Rodrigues & Prudente 2011, Valdujo et al. 2012, Bastiani & Lucas 2013, Moraes et al. 2017, 2020) and practical approaches, such as in the establishment and monitoring of conservation areas (e.g., Araújo et al. 2010, Oliveira et al. 2021, Stephenson et al. 2022, Freire et al. 2023). The periodical assessment of species composition of local faunas allows one to evaluate the natural dynamics of communities, as well as the effectiveness of conservation action plans (Argel-de-Oliveira 1993, Sweke et al. 2016).

Due to their ecological diversification and sensitivity to environmental changes, amphibians and reptiles are considered important bioindicators (Simon et al. 2011, Silva et al. 2020, Santana et al. 2021), and may also provide useful information to select priority areas for conservation (Silvano & Segalla 2005). In the Neotropical region, particularly in Brazil—admittedly a megadiverse country regarding these two animal groups (Rodrigues 2005a, Segalla et al. 2021)—several herpetofaunal inventories have been conducted in recent decades. Despite the growing number of herpetofaunal inventories throughout the country (Affonso et al. 2015, Teodoro et al. 2020, Costa-Neto et al. 2022), many areas still remain to be investigated. This is particularly true for transition zones between distinct ecoregions, which may present high species richness, with representatives from the contacting ecoregions (Valdujo et al. 2012, Silva et al. 2013, Dal Vecchio et al. 2016, Santos et al. 2019, Matavelli et al. 2019, Bastos & Zina 2022).

The Brazilian Cerrado, considered the largest (2,000,000 km²), most distinct, most diverse, and highly threatened tropical savanna in the world (Silva & Bates 2002, Colli et al. 2020), is listed as one of the global biodiversity hotspots (Myers et al. 2000). The main threats to the Cerrado are both direct (e.g., continuous modification of natural landscapes into urban, agricultural or pasture areas; wildfires; pollution, erosion and sedimentation; invasive species) and indirect (urban sprawl, cattle raising, crops, mining, pulp mills, transportation infrastructure, electric power, oil and gas), in addition to climate change (CEPF 2023). Nearly 46% of pristine Cerrado vegetation has already been deforested, while less than 20% of the total area remains undisturbed (Strassburg et al. 2017) and only 10.5% is legally protected (Vieira-Alencar et al. 2023).

The Brazilian Cerrado is home to approximately 209 amphibian species (Valdujo et al. 2012) and 267 reptiles (Colli et al. 2002, Nogueira et al. 2011). These numbers represent 17.6 and 31.2%, respectively, of the species richness reported for Brazil (Segalla et al. 2021, Guedes et al. 2023). Due to the immense cryptic diversity and/or the numerous undersampled areas throughout the Cerrado, many species are yet to be described (e.g., Domingos et al. 2017, Guerra et al. 2020, Guedes et al. 2023). Unexplored areas of Cerrado include its westernmost portion, where there are interdigitations with neighbouring ecoregions, including the Pantanal wetlands, moist and dry tropical forests (Dinerstein et al. 2017). Biological assemblages representative of this transition zone are officially protected within the Serra das Araras Ecological Station (SAES) in the southwestern part of the state of Mato Grosso.

Initial studies on amphibians and reptiles from SAES were conducted in the second half of the 1980s (Vanzolini 1986, Nascimento et al. 1988), but only a list of lizard species from SAES was formally published (Nascimento et al. 1988) based on the material collected at that time. A more complete list of lizard species was published nearly two decades later (Nogueira & Rodrigues 2006). Additional publications mentioning material obtained at SAES include notes on the geographic distribution of amphibians (Strüssmann et al. 2012, Pansonato et al. 2017), amphisbaenians (Dorado-Rodrigues et al. 2013), lizards (Barreto et al. 2012), and crocodilians (Mudrek et al. 2021). They also include descriptions of new species of snakes (Lema 2002, Marra-Santos & Reis 2018) and ecological studies on freshwater turtles (Marques et al. 2017, Brito et al. 2018), anurans (Barros et al. 2024), and lizards (Barros et al. 2022).

In this study, we discuss the results of a mid-term herpetofaunal inventory in a mosaic of open and forested habitats in Serra das Araras Ecological Station, and describe the richness, species composition, and abundance of amphibians and reptiles from a scarcely known portion of the Cerrado. We also assess the taxonomic similarities of the assemblages in the sampled habitats.

Material and Methods

1. Study area

Serra das Araras Ecological Station (SAES) is a national protected area of 28,700 hectares situated in the municipalities of Porto Estrela and Cáceres (15°28’– 15°51’ S; 57°03’– 57°19’ W), in the southwestern part of the state of Mato Grosso, Midwestern Brazil (Figure 1). The region encompasses a corridor of parallel mountains running through the Província Serrana, a physically isolated area of residual relief of nearly 400 km × 40 km, which constitutes the watershed between the Paraná-Paraguay River Basin and the Amazonas River Basin (Ross 1991). Evidence from local fish assemblages indicates that the drainage basins of local uplands and adjoining highlands—currently isolated—have a recent shared history (Ribeiro et al. 2013). In fact, the whole region is considered a transition zone between the western portions of the Cerrado and the southern Amazon rainforest (Marques et al. 2020) (Figure 1).

The climate is considered Aw according to the Köppen’s classification, with a markedly seasonal tropical savanna climate, well-defined dry winters (from May to October) and rainy summers (November to April) (Álvares et al. 2013). SAES presents high environmental heterogeneity. The local landscape comprises a mosaic of open and forested vegetation types (ICMBio 2016), with biotic elements that also occur in other areas of Cerrado, Pantanal (the world’s largest continental wetland), southern Amazon moist forests, and tropical dry forests (Gonçalves & Gregorin 2004, Valadão 2012, Vitorino et al. 2018, see also Marques et al. 2020).

During a field study of amphibians and reptiles at SAES, we systematically sampled five vegetation types, classified and defined by Ribeiro & Walter (2008). Two of them are considered open formations (cerrado *sensu stricto* and cerrado parkland), and three are considered forested formations (riparian forest, semi-deciduous dry forest, and cerrado woodland—known in Brazil as cerradão) (Figure 2). For more detailed information about these types of vegetation, see Barros et al. (2022). Occasional samplings were also conducted in deciduous forests established on limestone outcrops (*mata calcária*), rocky fields

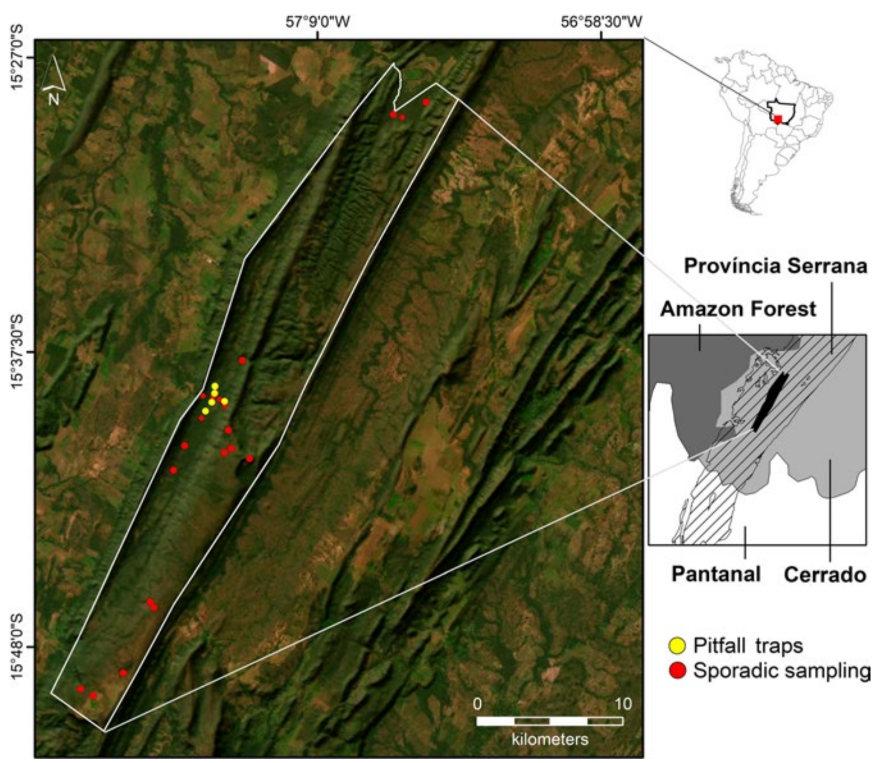

Figure 1. Location of Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil, indicating the herpetofauna sampling sites.

Figure 2. Study area and habitats systematically sampled: a – general view of Serra das Araras Ecological Station (SAES), b – riparian forest, c – semi-deciduous dry forest, d – cerrado woodland, e – cerrado *sensu stricto*, f – cerrado parkland.

(*campo rupestre*), and savannas on rocky soil (*cerrado rupestre*) (see Felfili et al. 2007, Pereira & Fernandes 2022).

2. Data collection

We inventoried amphibians and reptiles at SAES from May 2009 to January 2011, during 21 consecutive monthly field expeditions, with mean intervals of 25 (±6) days between each expedition. On each of the five sampled vegetation types, we installed two sets of pitfall traps with drift fences (as described by Cechin & Martins 2000), with least 200 m apart from each other, totalling ten sampling points. Sets of pitfalls were composed of ten plastic buckets (60-litre each), buried along a straight line at 7.0 m intervals. The openings of the buckets were interconnected by a 0.5 m high plastic drift fence. The buckets were left open and checked daily (in the morning) for five consecutive days each month, totalling 105 non-consecutive sampling days. When not in use, the buckets remained covered. The total sampling effort was 7,500 bucket-days (1,500 bucket-days at each vegetation type).

We also sampled local herpetofauna during active searches, performed along transects slowly surveyed on foot by three to five observers, during the daytime (08:20-10:30h) and at night (18:30 to 23:00h). The sampling effort at each sampling site varied from 4.5 to 20 observer-hours, totalling 210 observer-hours (42 observer-hours at each vegetation type). Crocodilians and freshwater turtles were sampled sporadically, during expeditions along the many water courses existing inside the SAES. As a source of information on the composition of local herpetofauna, we also considered opportunistic encounters and photographs of live or dead specimens found inside SAES, as well as secondary data gathered from the literature or from museum records, based on vouchers in three Brazilian zoological collections: *Coleção Herpetológica da Universidade Federal de Mato Grosso* (UFMT, Cuiabá, Mato Grosso), *Coleção de Anfíbios e Répteis do Museu de Ciências e Tecnologia da Pontifícia Universidade Católica do Rio Grande do Sul* (MCT-PUCRS, Porto Alegre, state of Rio Grande do Sul), and *Coleções de Anfíbios e Répteis do Museu Nacional, Universidade Federal do Rio de Janeiro* (MNRJ/UFRJ, Rio de Janeiro, state of Rio de Janeiro).

All the anurans and reptiles captured in pitfalls, sighted, or heard during the study were recorded, but only a few specimens were collected as vouchers. Specimens were killed using an injectable

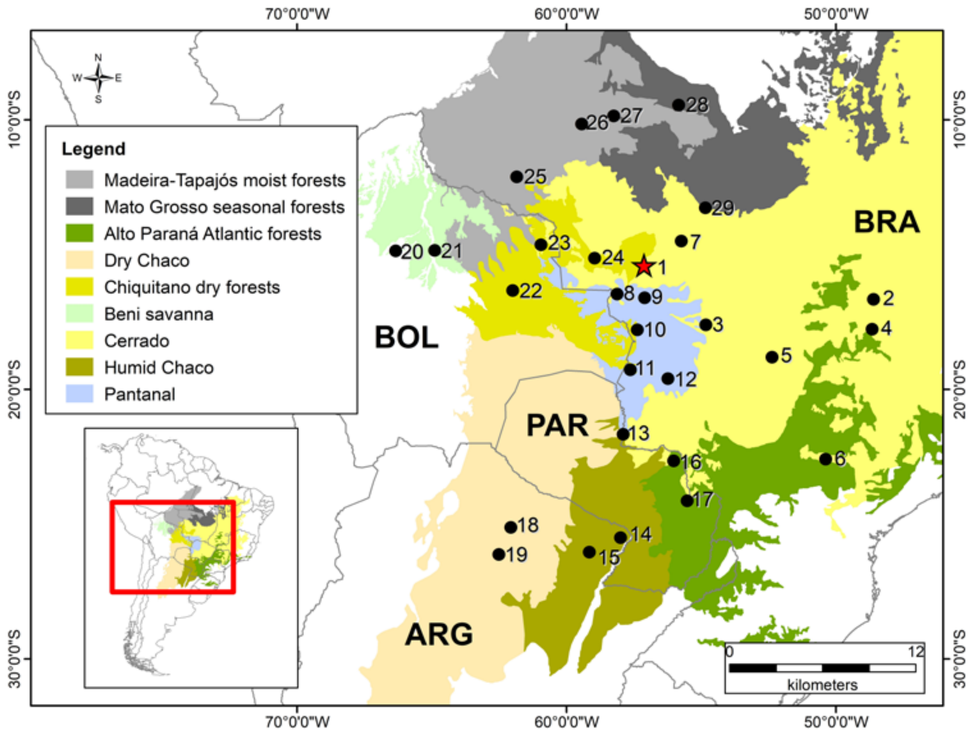

Figure 3. Geographical location of the Serra das Araras Ecological Station (SAES) and other 28 localities previously studied in nine South American ecoregions (sensu Dinerstein et al. 2017): **Cerrado** – 1 (SAES; red star), 2 (Morais et al. 2012), 3 (Silva Jr. et al. 2009), 4 (Ramalho et al. 2019), 5 (Ramalho et al. 2014), 6 (Araujo & Almeida-Santos 2011), 7 (Gonçalves 2023); **Pantanal** – 8 (Pansonato et al. 2011), 9 (Dorado-Rodrigues et al. 2015), 10 (Jesus & Lima 2003), 11 (Piva et al. 2020), 12 (Ferreira et al. 2006); **Humid Chaco** – 13 (Souza et al. 2010), 14 (Álvarez et al. 2009), 15 (Pesci et al. 2018); **Alto Paraná Atlantic Forest** – 16 (Martinéz et al. 2016), 17 (Cacciali et al. 2015); **Dry Chaco** – 18 (Kacoliris et al. 2006), 19. (Perez-Iglesias 2017); **Beni Savanna** – 20 (Eversole et al. 2021), 21 (Rivas et al. 2023); **Chiquitano Dry Forest** – 22 (Schulze et al. 2009), 23 (Watling et al. 2009), 24 (Strüssmann unpublished); **Madeira-Tapajós Moist Forest** – 25 (Ferrão et al. 2012), 26 (São Pedro et al. 2009), 27 (Noronha et al. 2015); **Mato Grosso Tropical Dry Forest** – 28 (Rodrigues et al. 2015), 29 (Rossi et al. 2023).

overdose of lidocaine 2% (Xylestesin®), fixed in 10% formalin, and later preserved in 70% alcohol. Vouchers are deposited at UFMT, and voucher numbers are provided in the results. The nomenclature and systematic arrangement used for amphibians follow Frost (2023) and those for reptiles follow Uetz et al. (2023).

3. Data analysis

To verify sampling efficiency, we created rarefaction curves and confidence intervals (95%). We estimated the number of species and standard deviation based on the number of sampling days, after 1000 runs of the Bootstrap species richness estimator (Colwell et al. 2004), using the EstimateS 8.2.0 software (Colwell 2009). To assess similarities among vegetation types (cerrado *sensu stricto*, cerrado parkland, riparian forest, semi-deciduous dry forest, and cerrado woodland), we performed a cluster analysis based on the Jaccard index.

We also compared the species compositions at SAES (amphibians and reptiles separately) with those of other 29 localities previously studied in nine South American ecoregions (*sensu* Dinerstein et al. 2017)—Alto Paraná Atlantic Forest, Beni Savanna, Cerrado, Chiquitano Dry Forest, Dry Chaco, Humid Chaco, Madeira-Tapajós Moist Forest, Mato Grosso Tropical Dry Forest, and Pantanal (Figure 3)—through a cluster analysis. We only included inventories (28 for amphibians and 24 for reptiles) with similar sampling methods and similar temporal coverage (i.e, samplings during rainy and dry periods). To avoid taxonomic issues, we excluded species mentioned in the original manuscripts as undetermined ("cf.", "gr." and "aff.") or without species identification ("sp."). We computed the Jaccard dissimilarity index for comparisons between assemblages and the Unweighted Pair Group Method with Arithmetic mean (UPGMA) to generate the dissimilarity dendrogram among the localities. We assessed the robustness of generated dendrograms with an alternative unbiased index (AU) calculated from multiscale bootstrap resampling (Suzuki & Shimodaira 2006). We used the R version 3.6 software (R Development Core Team 2023) and the vegan Community Ecology Package version 2.5-7 (Oksanen et al. 2018).

Results

We documented 123 species—39 amphibians and 84 reptiles—at Serra das Araras Ecological Station (SAES). All the amphibians belong to Anura; reptiles include five Amphisbaenia, 48 Serpentes, 26 Sauria, four Testudines, and one Crocodylia (Table 1; Figures 4–9). The 123 species recorded represent 63% of the estimated richness (SD = 186.58 ± 7.46). The rarefaction curve did not reach the asymptote (Figure 10).

Table 1. List of amphibian and reptile species recorded at Serra das Araras Ecological Station (SAES), municipality of Porto Estrela, Mato Grosso, Brazil. Vegetation types: Rf (riparian forest); Sf (semi-deciduous dry forest); Cd (Cerrado woodland); Ce (Cerrado *sensu stricto*); Pc (Cerrado parkland). Sampling method: AS (active searches); OE (opportunistic encounters); P (pitfalls); SD (secondary data: records from the literature or collections). Vouchers (see museum acronyms in “Data Collection”), photographs or references (1 – Marra-Santos & Reis 2018; 2 – Lema 2002; 3 – Strüssmann 1988; 4 – Nascimento et al. 1988; 5 – Nogueira et al. 2009).

Taxa	**N total**	**Vegetational types**					**Method**				**Voucher**
		Rf	**Sf**	**Cd**	**Ce**	**Pc**	**P**	**AS**	**OE**	**SD**	
Amphibia (39)											
Anura											
Bufonidae (3)											
Rhaebo guttatus (Schneider, 1799)	**2**	2					2			x	UFMT 10607
Rhinella diptycha (Cope, 1862)	**31**	17	1	4	1	8	15	1	15	x	UFMT 10606
Rhinella stanlaii (Lötters & Köhler, 2000)	**70**	38	2	26	1	3	50	13	7	x	UFMT 10609
Craugastoridae (2)											
Oreobates heterodactylus (Miranda-Ribeiro, 1937)	**23**	2	21				2	17	4	x	UFMT 10628
Pristimantis dundeei (Heyer & Muñoz, 1999)	**19**	13	1	4		1	8	6	5	x	UFMT 10626
Dendrobatidae (1)											
Ameerega braccata (Steindachner, 1864)	**12**	1	1	4	3	3	8	1	3	x	UFMT 10625
Hylidae (13)											
Boana aff. *geographica* (Spix, 1824)	**2**	2						1	1		UFMT 10620
Boana albopunctata (Spix, 1824)	**2**	1				1		1	1	x	UFMT 374
Boana raniceps (Cope, 1862)	**6**	5				1		5	1		UFMT 10616
Dendropsophus cf. *elianeae* (Napoli & Caramaschi, 2000)	**2**					2		1	1		UFMT 10635
Dendropsophus melanargyreus (Cope, 1887)	**2**	1		1				1	1		UFMT 13843
Dendropsophus minutus (Peters, 1872)	**10**	1	2			7	1	8	1	x	UFMT 10633
Dendropsophus nanus (Boulenger, 1889)	**2**					2		1	1		UFMT 10636
Osteocephalus taurinus Steindachner, 1862	**7**	2	5					6	1		UFMT 13849
Pithecopus azureus (Cope, 1862)	**3**	1				2		2	1		UFMT 13848
Scinax fuscomarginatus (A. Lutz, 1925)	**2**					2		1	1		UFMT 10632
Scinax fuscovarius (A. Lutz, 1925)	**7**	4				3		3	4	x	UFMT 10622
Scinax nasicus (Cope, 1862)	**2**				2			1	1		UFMT 13845
Trachycephalus typhonius (Linnaeus, 1758)	**2**	1			1			1	1	x	UFMT 10617
Leptodactylidae (17)											
Adenomera sp.	**150**	53	15	23	14	45	138	5	7	x	UFMT 10634
Leptodactylus brevipes Cope, 1887	**1**	1							1		UFMT 13846
Leptodactylus elenae Heyer, 1978	**7**		1	1		5	5	1	1	x	UFMT 10621
**Leptodactylus furnarius* Sazima & Bokermann, 1978	**x**									x	UFMT 673
Leptodactylus fuscus (Schneider, 1799)	**4**	1		1		2	2	1	1	x	UFMT 10614
Leptodactylus jolyi Sazima & Bokermann, 1978	**17**	1			1	15	10	5	2		UFMT 10608
Leptodactylus labyrinthicus (Spix, 1824)	**46**	2		1		43	35	8	3	x	UFMT 10627
Leptodactylus macrosternum Miranda-Ribeiro, 1926	**10**	3	2			5	3	5	2		UFMT 10610
*Leptodactylus mystaceu*s (Spix, 1824)	**6**		6					5	1		UFMT 10619
Leptodactylus mystacinus (Burmeister, 1861)	**2**		1			1		1	1		UFMT 10612
Leptodactylus syphax Bokermann, 1969	**5**	1	4					1	4	x	UFMT 10615
Physalaemus albonotatus (Steindachner, 1864)	**5**	4				1	2	1	2		UFMT 10623
Physalaemus centralis Bokermann, 1962	**2**					2		1	1		UFMT 13844

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Taxa	N total	Vegetational types					Method				Voucher
		Rf	Sf	Cd	Ce	Pc	P	AS	OE	SD	
Physalaemus cuvieri Fitzinger, 1826	**104**	2		2	4	96	95	9		x	UFMT 10624
Physalaemus nattereri (Steindachner, 1863)	**2**	1				1		1	1	x	UFMT 546
Pseudopaludicola ameghini (Cope, 1887)	**10**					10		8	2		UFMT 10631
Pseudopaludicola saltica (Cope, 1887)	**16**					16	2	12	2	x	UFMT 10637
Microhylidae (3)											
Chiasmocleis albopunctata (Boettger, 1885)	**53**	1				52	51	1	1		UFMT 10629
Elachistocleis magna Toledo, 2010	**45**					45	43	1	1		UFMT 11993
Elachistocleis cf. *corumbaensis* Piva, Caramaschi & Albuquerque, 2017	**115**	2				113	114	1			UFMT 13847
Reptilia (84)											
Squamata (79)											
Amphisbaenia (5)											
Amphisbaenidae (5)											
Amphisbaena alba Linnaeus, 1758	**2**	1		1					2		UFMT 9872
Amphisbaena brasiliana (Gray, 1865)	**1**					1			1		UFMT9880
Amphisbaena silvestrii Boulenger, 1902	**2**	1	1				1		1		UFMT 8342
Amphisbaena steindachneri Strauch, 1881	**3**	1			1	1			3		UFMT 8726
Amphisbaena vermicularis Wagler in Spix, 1824	**4**	1		1	2		2	1	1		UFMT 8333
Serpentes (48)											
Anomalepididae (2)											
**Liotyphlops ternetzii* (Boulenger, 1896)	**x**									x	MNRJ/UFRJ 7854
**Liotyphlops taylori* Marra-Santos and Reis, 2018	**x**									x	1
Boidae (4)											
Boa constrictor Linnaeus, 1758	**3**	1			1	1		1	2	x	UFMT 8727
Corallus hortulana (Linnaeus, 1758)	**1**	1						1			UFMT 8335
Epicrates crassus Cope, 1862	**4**				2	2	2	1	1		UFMT 8325
Eunectes murinus (Linnaeus, 1758)	**1**	1							1	x	UFMT 9271
Colubridae (36)											
Adelphostigma occipitalis (Jan, 1863)	**8**			1	6	1	8			x	UFMT 8327
**Apostolepis christineae* De Lema, 2002	**x**									x	2
Atractus albuquerquei Cunha & Nascimento, 1983	**11**		1	6	4		10		1		UFMT 8329
Chironius exoletus (Linnaeus, 1758)	**1**			1				1		x	UFMT 118
Chironius flavolineatus (Jan, 1863)	**4**	2	1	1			1	1	2		UFMT 8330
Chironius laurenti Dixon, Wiest & Cei, 1993	**1**	1							1		
Dipsas turgida (Cope, 1868)	**3**	2	1				1	1	1		UFMT 8327
Drymarchon corais (Boie, 1827)	**3**	2		1				1	2	x	UFMT 8336
Dryophylax chaquensis (Bergna & Alvarez, 1993)	**1**					1			1		UFMT 9874
Dryophylax hypoconia (Cope, 1860)	**1**	1						1			UFMT 9878
Erythrolamprus almadensis (Wagler, 1824)	**1**	1							1		UFMT 9873
Erythrolamprus reginae (Linnaeus, 1758)	**5**	2				3	4	1		x	UFMT 8331
Helicops leopardinus (Schlegel, 1837)	**1**	1							1		UFMT 10686
Imantodes cenchoa (Linnaeus, 1758)	**1**		1						1		Photograph

Continue...

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Taxa	N total	Vegetational types					Method				Voucher
		Rf	Sf	Cd	Ce	Pc	P	AS	OE	SD	
Leptodeira annulata Linnaeus, 1758	**2**	1			1			1	1	x	UFMT 8324
Leptophis marginatus (Cope, 1862)	**2**	2						1	1		UFMT 8321
Lygophis dilepis (Cope, 1862)	**1**	1							1		UFMT 9875
Mastigodryas boddaerti (Sentzen, 1796)	**3**	2		1				1	2		UFMT 8729
Oxybelis fulgidus (Daudin, 1803)	**2**	1		1					2		UFMT 9871
Oxyrhopus melanogenys (Tschudi, 1845)	**1**				1		1				UFMT 8332
Oxyrhopus rhombifer Werner, 1909	**1**				1			1			UFMT9877
**Palusophis bifossatus* (Raddi, 1820)	**x**									x	UFMT 173
Phalotris matogrossensis De Lema, D'Agostini & Cappellari, 2005	**1**				1			1		x	UFMT 10139
Phalotris nasutus (Gomes, 1915)	**1**				1				1		Photograph
Philodryas agassizii (Jan, 1863)	**9**	2		1	4	2	8		1		UFMT 8326
Philodryas nattereri Steindachner, 1870	**4**	1			2	1	2	1	1		UFMT 8343
Philodryas olfersii (Liechtenstein, 1823)	**2**				2		2			x	UFMT 8322
Phrynonax sexcarinatus (Wagler, 1824)	**2**	2						1	1		UFMT 10138
Pseudoboa nigra (Duméril, Bibron & Duméril, 1854)	**4**		1	1	1	1		1	3		UFMT 8338
Pseudoeryx plicatilis Cope, 1885	**1**					1			1		UFMT 10687
Siphlophis compressus (Daudin, 1803)	**1**		1					1			UFMT 8339
Spilotes pullatus (Linnaeus, 1758)	**3**	1	1	1				1	2	x	UFMT 119
Spilotes sulphureus (Wagler, 1824)	**1**	1							1	x	UFMT 2428
Tantilla melanocephala (Linnaeus, 1758)	**1**				1				1	x	UFMT 194
Xenodon merremii (Wagler, 1824)	**2**	1		1					2	x	UFMT 8731
**Xenodon severus* (Linnaeus, 1758)	**x**									x	UFMT 200
Elapidae (1)											
**Micrurus frontalis* Duméril, Bibron & Duméril, 1854	**x**									x	UFMT 267
Leptotyphlopidae (1)											
Epictia albifrons (Wagler, 1824)	**1**		1						1		UFMT 8337
Typhlopidae (1)											
Amerotyphlops brongersmianus (Vanzolini, 1976)	**4**	4					4				UFMT 8341
Viperidae (3)											
**Bothrops mattogrossensis* Amaral, 1925	**x**									x	3
Bothrops moojeni Hoge, 1966	**7**	3	1	1	1	1	4	2	1		UFMT 8334
Crotalus durissus Linnaeus, 1758	**4**	1	1		2		2	2			UFMT 8323
Sauria (26)											
Anolidae (1)											
Anolis aff. *meridionalis* (Boettger, 1885)	**15**				13	2	13	1	1	x	UFMT 8311
Gekkonidae (1)											
Hemidactylus mabouia (Moreau de Jonnès, 1818)	**1**	1							1		UFMT 8307
Gymnophthalmidae (5)											
Bachia dorbignyi (Duméril & Bibron, 1839)	**3**			1	2		3				UFMT 8304
Cercosaura olivacea Gray, 1845	**41**	1			20	20	40		1	x	UFMT 8317
Cercosaura parkeri Ruibal, 1952	**6**				1	5	5	1			UFMT 8317
Colobosaura modesta (Reinhardt & Luetken, 1862)	**62**	12	6	21	19	4	59	1	2	x	UFMT 8313

Continue...

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Taxa	**N total**	**Vegetational types**					**Method**				**Voucher**
		Rf	**Sf**	**Cd**	**Ce**	**Pc**	**P**	**AS**	**OE**	**SD**	
Micrablepharus maximiliani (Reinhardt & Luetken, 1862)	**40**		1	1	24	14	39	1		x	UFMT 8310
Hoplocercidade (1)											
Hoplocercus spinosus Fitzinger, 1843	**8**	1	4	3			8			x	UFMT 8307
Iguanidae (1)											
Iguana iguana (Linnaeus, 1758)	**2**	2						1	1	x	UFMT 10137
Phyllodactylidae (1)											
Phyllopezus przewalskii Koslowsky, 1895	**2**		2					1	1	x	UFMT 9881
Polychrotidae (1)											
Polychrus acutirostris Spix, 1825	**3**				1	2	1	1	1		UFMT 8730
Scincidae (3)											
Copeoglossum nigropunctatum (Spix, 1825)	**18**	15	2	1			17	1		x	UFMT 8315
Manciola guaporicola (Dunn, 1935)	**9**	1				8	8		1	x	UFMT 8316
Notomabuya frenata (Cope, 1862)	**28**	7	8	6	3	4	27		1	x	UFMT 8314
Sphaerodactylidae (1)											
Gonatodes humeralis (Guichenot, 1855)	**9**	3	3	3			6	2	1	x	UFMT 8312
Teiidae (8)											
Ameiva ameiva (Linnaeus, 1758)	**11**	2	1		5	3	9	1	1	x	UFMT 8305
**Ameivula* aff. *ocellifera* (Spix, 1825)	**x**									x	UFMT 2583
**Kentropyx calcarata* Spix, 1825	**x**									x	4, 5
**Kentropyx paulensis* (Boettger, 1893)	**x**									x	UFMT 183
Kentropyx vanzoi Gallagher & Dixon, 1980	**34**			1	31	2	34			x	UFMT 8308
Salvator merianae Duméril & Bibron, 1839	**6**	1	1	1	1	2		1	5	x	UFMT 8320
Tupinambis quadrilineatus Manzani & Abe, 1997	**6**				5	1	6				UFMT 8319
Tupinambis teguixin (Linnaeus, 1758)	**3**	1		2					3	x	UFMT 203
Tropiduridae (3)											
Stenocercus sinesaccus Torres-Carvajal 2005	**18**	6	3	7	1	1	17	1		x	UFMT 8303
Tropidurus itambere Rodrigues, 1987	**3**		3					3		x	UFMT 8306
Tropidurus lagunablanca Carvalho, 2016	**2**		2					1	1	x	UFMT 193
Testudines (4)											
Chelidae (2)											
Mesoclemmys vanderhaegei (Bour, 1973)	**2**	2						1	1		UFMT 10140
Phrynops geoffroanus (Schweigger, 1812)	**1**	1							1	x	UFMT 10689
Testudinidae (2)											
Chelonoidis carbonarius (Spix, 1824)	**2**	1			1			1	1		Photograph
Chelonoidis denticulatus (Linnaeus, 1766)	**2**	2						1	1		Photograph
Crocodylia (1)											
Allligatoridae (1)											
Paleosuchus palpebrosus (Cuvier, 1807)	**2**	2						1	1		UFMT 9876
Total number of species	**123**	74	36	35	40	53	50	76	88	61	
Total number of individuals	**1268**	266	109	133	188	571	930	182	155		

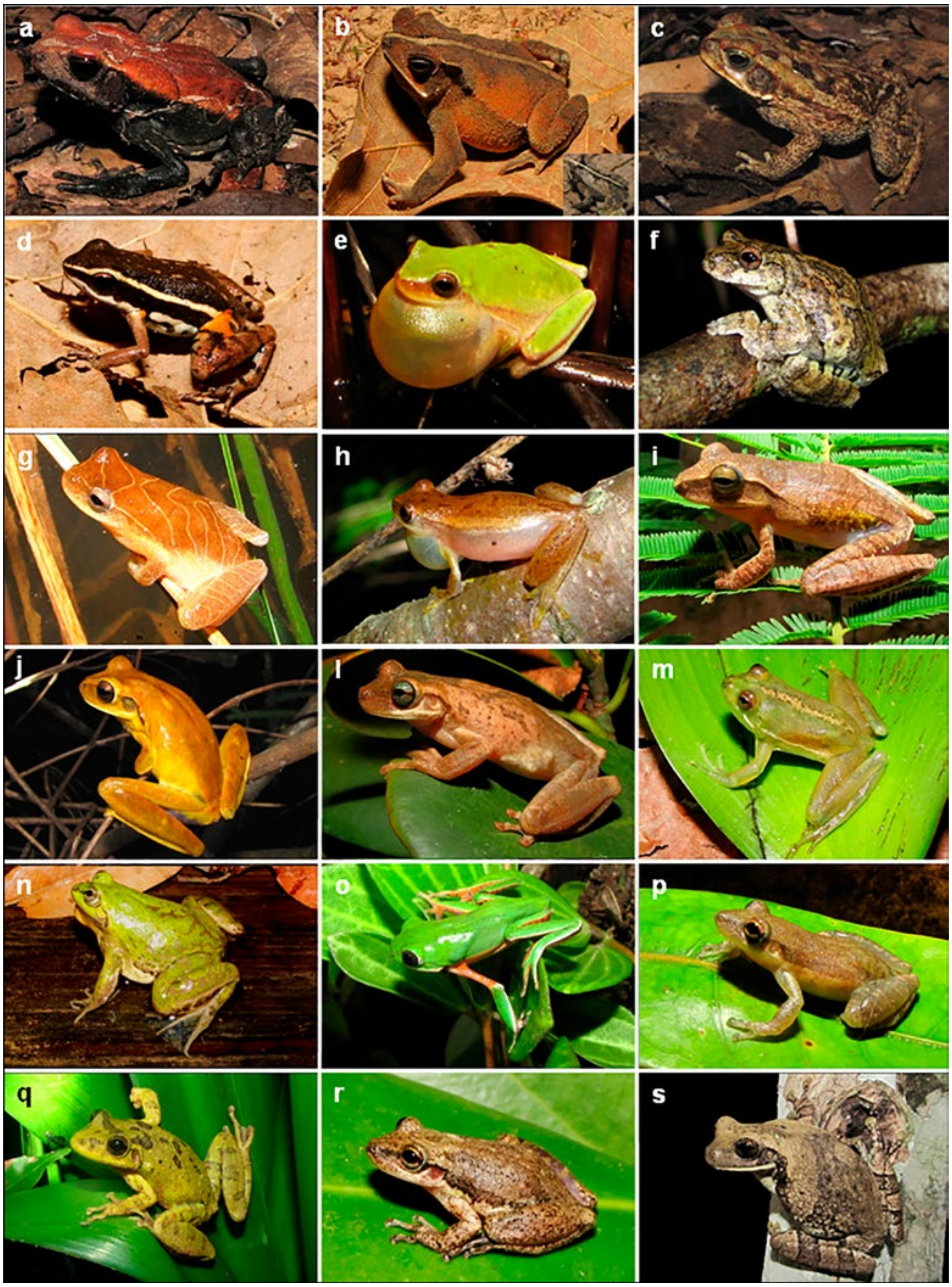

Figure 4. Amphibians recorded at Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil. a – *Rhaebo guttatus*, b – *Rhinella stanlaii*, c – *Rhinella diptycha*, d – *Ameerega braccata*, e – *Dendropsophus* cf. *elianeae*, f – *Dendropsophus melanargyreus*, g – *Dendropsophus minutus*, h – *Dendropsophus nanus*, i – *Boana albopunctata*, j – *Boana raniceps*, l – *Osteocephalus taurinus*, m – *Pseudis limellum*, n – *Pseudis platensis*, o – *Pithecopus azureus*, p – *Scinax fuscomarginatus*, q – *Scinax fuscovarius*, r – *Scinax nasicus*, s – *Trachycephalus typhonius*.

A total of 112 species were recorded during the field study. Pitfalls allowed to record 930 individuals, from 50 species (eight of which were recorded exclusively through this method), while active searches yielded 182 individuals from 76 species (six exclusive), and opportunistic encounters 155 individuals from 88 species (20 exclusive species). Secondary data allowed the recording of 61 species from SAES, eleven of which were listed exclusively based on literature records and museum vouchers (Table 1). Among the records made during the field study, a higher number of species was found in riparian forest (74 species; 20 exclusive), followed by cerrado parkland (53 species; 10 exclusive), cerrado *sensu stricto* (40 species; 7 exclusive), semi-deciduous dry forest (36 species; 7 exclusive), and cerrado woodland (35 species; one exclusive). Nine species were recorded in all the vegetation types (Table 1).

Figure 5. Amphibians recorded at Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil. a – *Physalaemus nattereri*, b – *Physalaemus albonotatus*, c – *Physalaemus centralis*, d – *Physalaemus cuvieri*, e – *Pseudopaludicola ameghini*, f – *Pseudopaludicola saltica*, g – *Leptodactylus macrosternum*, h – *Leptodactylus elenae*, i – *Leptodactylus fuscus*, j – *Leptodactylus labyrinthicus*, l – *Leptodactylus mystaceus*, m – *Leptodactylus mystacinus*, n – *Leptodactylus brevipes*, o – *Leptodactylus jolyi*, p – *Leptodactylus syphax*, q – *Adenomera* sp., r – *Chiasmocleis albopunctata*, s – *Elachistocleis magna.*

Cluster analysis based on the occurrence of the 38 amphibians and 74 reptile species recorded during field studies at SAES resulted in the formation of two groups (AU ≥ 91): (1) a group of three sampled forested formations (similarity 0.62 between semi-deciduous dry forest and cerrado woodland, and 0.73 between these two and the riparian forest); (2) a group of two open formations (cerrado *sensu stricto* and cerrado parkland, which presented a similarity of 0.5) (Figure 11).

The cluster analysis based on the occurrence of 179 amphibian species from 28 localities resulted in the formation of three main groups (AU ≥ 85): (1) a group of 12 localities situated in Alto Paraná Atlantic Forest, Chiquitano Dry Forest, and Cerrado ecoregions; (2) a group of ten localities in the Pantanal and Chaco ecoregions; (3) a group of two localities in the Beni savannas (Figure 12A). SAES is nested within group (1), with its amphibian species' composition being more similar

Figure 6. Amphibians and reptiles recorded at Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil. a – *Elachistocleis* cf. *corumbaensis*, b – *Pristimantis dundeei*, c – *Oreobates heterodactylus*, d – *Chelonoidis carbonarius*, e – *Chelonoidis denticulatus*, f – *Mesoclemmys vanderhaegei*, g – *Phrynops geoffroanus*, h – *Paleosuchus palpebrosus*, i – *Hemidactylus mabouia*, j – *Phyllopezus przewalskii*, l – *Gonatodes humeralis*, m – *Copeoglossum nigropunctatum*, n – *Manciola guaporicola*, o – *Notomabuya frenata*, p – *Anolis* aff. *meridionalis*, q – *Hoplocercus spinosus*, r – *Iguana iguana*, s – *Polychrus acutirostris.*

to those from a nearby locality in the Cerrado and from localities in the Chiquitano Dry Forest.

For reptiles, the cluster analysis based on the occurrence of 274 species from 24 localities indicated four well-supported groups (AU ≥ 82): (1) a group of 12 localities mainly from Cerrado and Pantanal; (2) a group of two localities in the Beni savannas; (3) a group of four localities from Gran Chaco; (4) a group of five localities in moist forests from southern Amazonia (Figure 12B). Our results indicated that reptile species' composition at SAES is more similar to that of five localities previously studied in the Cerrado, being nested within the group (1).

Discussion

Our study revealed the presence of a rich and diverse herpetofauna at the Serra das Araras Ecological Station—SAES, based on our own field inventories (38 amphibians and 74 reptiles) plus the literature/museum data (one amphibian and ten reptile species, nearly 9% of the total richness). The SAES herpetofauna includes 3.2% of the amphibians (Segalla et al. 2021) and 9.8% of the reptiles known in Brazil (6.1% of the amphisbaenians, 11.1% of the snakes, 9% of the lizards, 10.5% of the turtles, and 16% of the crocodilians; Guedes et al. 2023).

Figure 7. Reptiles recorded at Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil. a – *Stenocercus sinesacus*, b – *Tropidurus lagunablanca*, c – *Tropidurus itambere*, d – *Micrablepharus maximiliani*, e – *Colobosaura modesta*, f – *Bachia dorbignyi*, g – *Cercosaura olivacea*, h – *Cercosaura parkeri*, i – *Ameiva ameiva*, j – *Kentropyx vanzoi*, l – *Salvator merianae*, m – *Tupinambis quadrilineatus*, n – *Tupinambis matipu*, o – *Amphisbaena alba*, p – *Amphisbaena silvestrii*, q – *Amphisbaena steindachneri*, r – *Amphisbaena vermicularis*, s – *Amerotyphlops brongersmianus.*

The local herpetofauna represents about 26% of the 471 species found in the state of Mato Grosso (171 amphibians and 300 reptiles; Ávila et al. 2021, Guedes et al. 2023). The inventoried herpetofauna from SAES includes some genera that currently present a low taxonomic resolution with respect to the populations of Cerrado and transitional areas with neighbouring ecoregions (such as *Dendropsophus* and *Ameivula*).

The herpetofaunal diversity at SAES corresponds to 26% of the 476 species known from the Cerrado ecoregion (Colli et al. 2002, Nogueira et al. 2011, Valdujo et al. 2012), even though it is widely recognized that this number is largely underestimated (see Colli et al. 2020 and references therein). SAES can be considered one of the localities with higher species richness among all previously inventoried areas of Brazilian Cerrado (Supplementary Material). However, SAES is inserted in a transition zone between Cerrado and other South American ecoregions, contributing to high environmental heterogeneity, which is likely one of the main reasons for its rich

Figure 8. Reptiles recorded at Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil. a – *Epictia albifrons*, b – *Boa constrictor*, c – *Corallus hortulana*, d – *Epicrates crassus*, e – *Eunectes murinus*, f – *Chironius flavolineatus*, g – *Drymarchon corais*, h – *Leptophis marginatus*, i – *Palusophis bifossatus*, j – *Mastigodryas boddaerti*, l – *Oxybelis fulgidus*, m – *Phrynonax sexcarinatus*, n – *Spilotes sulphureus*, o – *Spilotes pullatus*, p – *Tantilla melanocephala*, q – *Atractus albuquerquei*, r – *Dipsas turgida*, s – *Adelphostigma occipitalis.*

herpetofauna (Marques et al. 2020; e.g., Gonçalves & Gregorin 2004, Valadão 2012, Vitorino et al. 2018).

We sampled five vegetation types which compose a mosaic of contrasting open and forested habitats (Ratter et al. 2003, Ribeiro & Walter 2008). This mosaic can favour the occurrence of species associated with particular types of habitats and microhabitats (Nogueira et al. 2009) and with different ecological attributes, which enables the coexistence of lineages with distinct ecological requirements (Colli et al. 2002, Nogueira et al. 2005, 2009, Barros et al. 2022, 2024). Although not tested herein, the high richness observed may also be the result of local factors, such as herpetofaunal species replacement due to variations in vegetation along the elevational gradient (220 to 860 m a.s.l.), as empirically observed for birds at SAES (Vitorino et al. 2018). The continued sampling effort—210 observer-hours and 7,500 bucket-days, along 21 consecutive months—may also contribute to the observed richness (Supplementary Material). Besides, complementary sampling methods returned exclusive records: a local resident alone was responsible for most of the opportunistic

Figure 9. Reptiles recorded at Serra das Araras Ecological Station (SAES), Porto Estrela, Mato Grosso, Brazil. a – *Phalotris matogrossensis*, b – *Phalotris nasutus*, c – *Pseudoeryx plicatilis*, d – *Philodryas agassizii*, e – *Philodryas nattereri*, f – *Philodryas olfersii*, g – *Oxyrhopus melanogenys*, h – *Oxyrhopus rhombifer*, i – *Pseudoboa nigra*, j – *Pseudoboa nigra*, l – *Pseudoboa nigra*, m – *Siphlophis compressus*, n – *Dryophylax chaquensis*, o – *Dryophylax hypoconia*, p – *Erythrolamprus almadensis*, q – *Erythrolamprus reginae*, r – *Bothrops moojeni*, s – *Crotalus durissus*.

encounters, 16% of which corresponded to exclusive records (mostly snakes). Among the species exclusively recorded for SAES based on literature or museum specimens are the rare fossorial snakes *Apostolepis christineae* and *Liotyphlops taylori*, both originally described after type material obtained at SAES. The holotype of *A. christineae* was collected at the station by a resident (see Lema 2002). Our data reinforce the importance of the constant presence in the area for the occasional record of infrequent species. Indeed, opportunistic encounters have been recognized as a valuable source of occurrence data, both from common (Hartmann et al. 2009) and rare species (e.g., Cechin 1999, Lisboa et al. 2021).

The semi-fossorial snake *Atractus albuquerquei* and the spider-eating snake *Philodryas agassizii* were the most abundant snake species at SAES (11 and nine individuals, respectively; Table 1). Being widely distributed from central and south-eastern Brazil to central Argentina, Uruguay, and Paraguay, the latter is relatively uncommon in zoological collections and specialises in feeding on spiders (Marques et al. 2006). Moreover, there are scarce records for the species in Southwestern

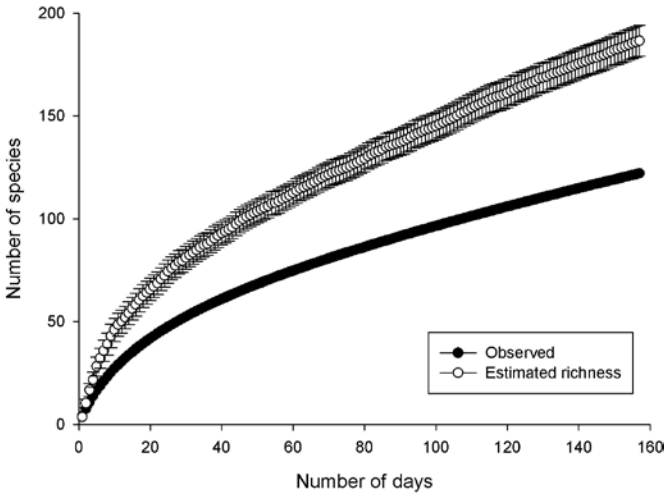

Figure 10. Sample-based species accumulation curves (black spots) and rarefaction curves (white spots) for herpetofauna recorded in Serra das Araras Ecological Station (SAES). Error bars indicate 95% confidence intervals.

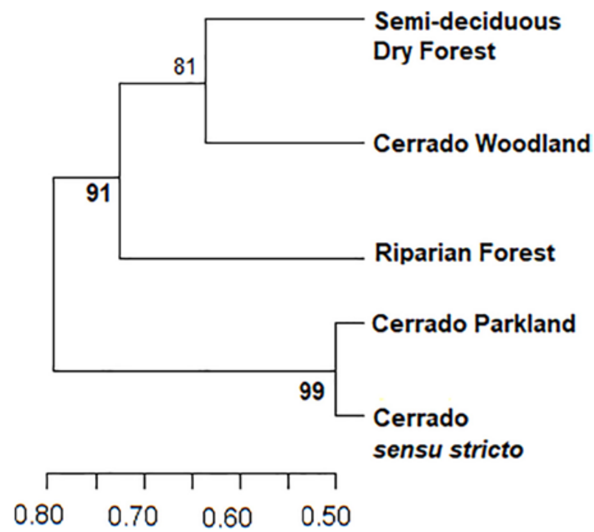

Figure 11. Similarity dendrogram (Jaccard) between systematically sampled areas. Vertical bars indicate clusters. Values around nodes are unbiased bootstrap. Nodes lacking values have bootstrap values < 80.

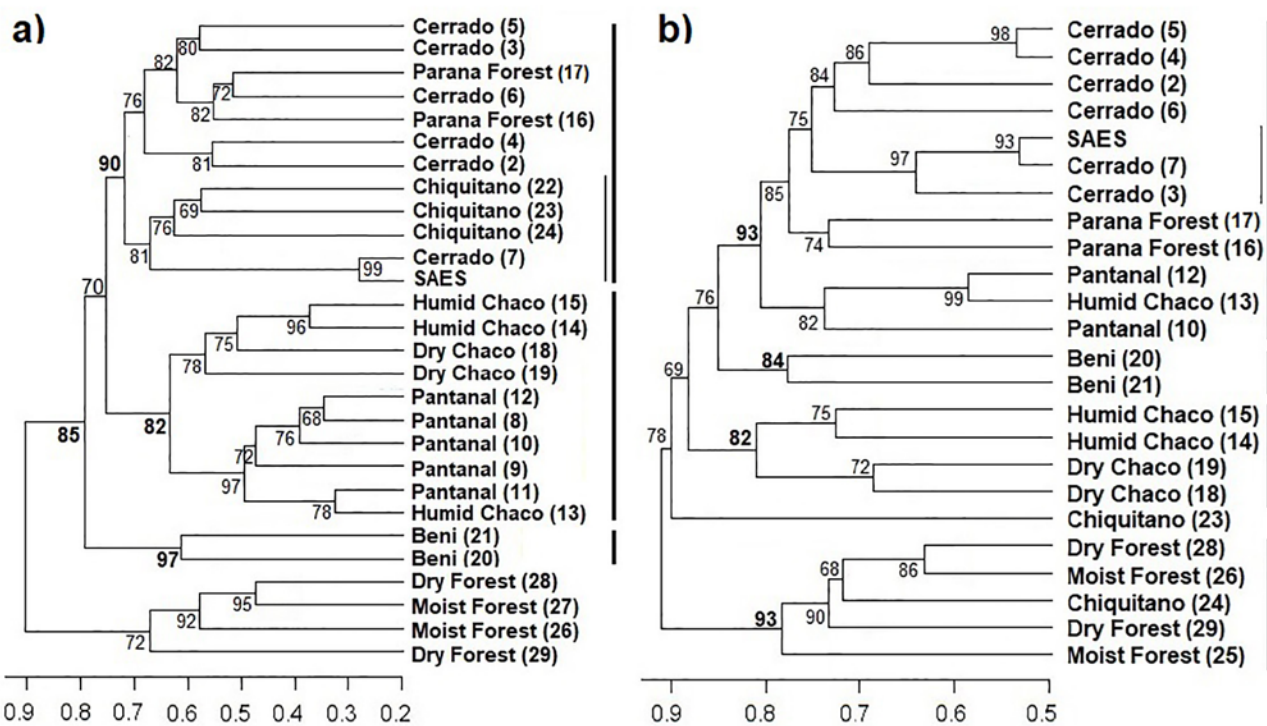

Figure 12. Dissimilarity dendrogram generated by UPGMA from a) amphibian and b) reptile species composition recorded in 29 areas of central South America. Vertical bars indicate clusters. Values around nodes are unbiased bootstrap. Nodes lacking values have bootstrap values < 79. **Cerrado** – 1 (SAES; red star), 2 (Morais et al. 2012), 3 (Silva Jr. et al. 2009), 4 (Ramalho et al. 2019), 5 (Ramalho et al. 2014), 6 (Araujo & Almeida-Santos 2011), 7 (Gonçalves 2023); **Pantanal** – 8 (Pansonato et al. 2011), 9 (Dorado-Rodrigues et al. 2015), 10 (Jesus & Lima 2003), 11 (Piva et al. 2020), 12 (Ferreira et al. 2006); **Humid Chaco** – 13 (Souza et al. 2010), 14 (Álvarez et al. 2009), 15 (Pesci et al. 2018); **Alto Paraná Atlantic Forest** – 16 (Martinéz et al. 2016), 17 (Cacciali et al. 2015); **Dry Chaco** – 18 (Kacoliris et al. 2006), 19. (Perez-Iglesias 2017); **Beni Savanna** – 20 (Eversole et al. 2021), 21 (Rivas et al. 2023); **Chiquitano Dry Forest** – 22 (Schulze et al. 2009), 23 (Watling et al. 2009), 24 (Strüssmann unpublished); **Madeira-Tapajós Moist Forest** – 25 (Ferrão et al. 2012), 26 (São Pedro et al. 2009), 27 (Noronha et al. 2015); **Mato Grosso Tropical Dry Forest** – 28 (Rodrigues et al. 2015), 29 (Rossi et al. 2023).

Cerrado (Nogueira et al. 2019). Except for semideciduous dry forest, this species was recorded at SAES in all vegetation types.

Regarding the sampled habitats, richness was highest in riparian forests (74 species) and cerrado parkland (53 species), and lowest in cerrado woodland (35 species). In our study area, cerrado parkland areas occur in straight contact with riparian forests, resulting in suitable microhabitats for a larger number of species, due to higher humidity and environmental heterogeneity. In the Cerrado, several species of amphibians and reptiles are habitat specialists (Vitt et al. 2007, Nogueira et al. 2009, Ernst et al. 2012, Ramalho et al. 2021), but even those species from open habitats eventually use riparian forests (Rodrigues 2005b). Indeed, these and other habitats with high or permanent availability of water may provide suitable conditions to maintain herpetofauna (e.g., Brasileiro et al. 2005, Valdujo et al. 2011, Gambale et al. 2014, Andrade et al. 2017). The availability of water and humidity is even more important in regions subject to contrasting seasonal climate conditions and to hydrological stress, especially during the dry season (Kopp et al. 2010, Gambale et al. 2014). On the other hand, being established on hydromorphic terrain (Ribeiro et al. 1983), the cerrado parkland also presents higher water availability—for longer periods—than other open Cerrado habitats. At SAES, microhabitats with reproductive aggregations of amphibians were abundant in cerrado parkland areas during the rainy season. In addition to a high species richness, this may lead to a higher proportion of exclusive species in this type of habitat, with reproductive modes depending on particular ecological requirements. As an example, females of *Elachistocleis magna* habitually used tiny depressions filled with water amid the cerrado parkland to deposit egg clutches, and both *Pseudopaludicola ameghini* and *P. saltica* are known to be largely dependent of hydromorphic terrain (Dorado-Rodrigues et al. 2024, Pansonato et al. 2013).

Only one exclusive species (the semi-arboreal colubrid snake *Chironius exoletus*) was found in cerrado woodland, which is structurally more similar to a forest, even though it comprises plant species typical of savannah formations (Ribeiro & Walter 2008). This and the other two forested vegetation types evaluated here (riparian forest and semi-deciduous dry forest) were more similar in species composition. Although forest formations exhibit high heterogeneity and climate stability (Ferreira et al. 2017, Caioni et al. 2020), their structural similarity can result in such overlapping species composition at SAES. The differences found at SAES in the species composition between forest and open habitats (cerrado *sensu stricto* and cerrado parkland) have similarly been reported in other Cerrado areas (e.g., Brasileiro et al. 2005, Nogueira et al. 2005, Gambale et al. 2014, Pacheco et al. 2018), evidencing the complementarity of different vegetation types in the assembly of local herpetofaunal assemblages.

Some of the species recorded at SAES are widely distributed, being present in several neighbouring ecoregions (such as the anurans *Boana raniceps, Dendropsophus minutus, D. nanus, Leptodactylus fuscus, L. labyrinthicus, L. macrosternum, Scinax fuscovarius*; the lizards *Ameiva ameiva, Iguana iguana*, and *Salvator merianae*; the snakes *Boa constrictor, Chironius flavolineatus, C. exoletus, Drymarchon corais, Corallus hortulana, Eunectes murinus*). Overall, the composition of the herpetofauna is more similar to that from a nearby locality (210 km northeast; Cerrado 7 in Figure 3) previously studied in the Cerrado. However, similarity patterns are slightly distinct for SAES amphibian and reptile assemblages. Both share species with other assemblages from Cerrado and Alto Paraná Atlantic Forest. The amphibian assemblage also presents similarities with those from the Chiquitano Dry Forest, while the reptile assemblage presents similarities, also, with assemblages from the Pantanal. These findings confirm the transitional nature of the SAES herpetofauna. The higher sensitivity of anurans to the environmental conditions during dry periods in the Pantanal, together with a greater relative dispersal capability of the reptiles (e.g., Rivas et al. 2021), supposedly would affect anurans at the most and therefore could lead to the above-mentioned differences in similarities with neighbour ecoregions, a hypothesis that could be further investigated.

Although most of the SAES is covered by different open vegetation types (where species typical of the Cerrado have been recorded), there are extensive areas of semi-deciduous dry forest and patches of riparian and gallery forest, more evident and extensive at the bottom of valleys (where species typical of the Amazon Forest were also recorded). In addition, species commonly found in the Pantanal occupy lentic environments adjacent to the alluvial floodplain of the Paraguay river. The presence of faunal elements typical of both Pantanal and Amazonia reinforce the idea that the so-called *Província Serrana* historically was—and still is—a dispersal corridor linking these two ecosystems, providing faunal interchange and genetic influx from one to another (Ribeiro et al. 2013). This interchange and the resulting diverse herpetofauna can be maintained by preserving the extensive riparian forests along river courses and the open habitats along the aligned mountain chains.

Our findings indicate that the herpetofauna of SAES is rich and representative of the regional biodiversity, with species composition evidencing its transitional nature. This high regional representativeness emphasizes the importance of protected areas for the maintenance of regional populations. However, although the SAES is a legally protected area, the surrounding region is under intense pressure from agricultural expansion, limestone mining, and transportation infrastructure, which are rapidly transforming natural areas (ICMBio 2016, Wantzen et al. 2024). Even considering that only one of the recorded species is considered under threat (the lizard *Kentropyx vanzoi,* presently considered as Vulnerable; MMA 2022), we suggest this region deserves special attention, e.g., by establishing a mosaic of conservation units along the entire *Província Serrana* in order to effectively protect this biodiversity corridor.

Supplementary Material

The following online material is available for this article:

Table S1 – Herpetofaunal studies conducted in core areas of Cerrado and transition zones with neighbouring ecoregions (Cerrado-Amazonia, Cerrado-Pantanal, Cerrado-Pantanal-Atlantic Forest, and Cerrado-Caatinga). Sampling method (sampling effort): AS – active searches (observer-hours); OE – opportunistic encounters; P – pitfalls (buckets-day); SD – secondary data.

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Author Contributions

Tainá Figueras Dorado-Rodrigues: Contribution to data collection; Substantial contribution to data analysis and interpretation; Contribution to manuscript preparation; Substantial contribution to critical revision, adding intellectual content.

Rafael Martins Valadão: Substantial contribution in the concept and design of the study; Substantial contribution to data collection; Contribution to data analysis and interpretation, Contribution to critical revision, adding intellectual content.

Luciana Mendes Valério: Contribution to data collection; Contribution to critical revision, adding intellectual content.

Carolina Potter de Castro: Contribution to data collection; Contribution to critical revision, adding intellectual content.

Christine Strüssmann: Substantial contribution in the concept and design of the study; Resources; Contribution to data collection; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Conflicts of Interest

The authors declare no conflict of interest related to the publication of this work.

Ethics

The Instituto Chico Mendes de Conservação da Biodiversidade (ICMBio-SISBIO n° 19518-1) provided collecting and research permits. The Brazilian legislation related to animal ethics and the National Council for Control of Animal Experimentation were both just being implemented when the study began in 2009 and there was no Committee on Ethics in the Use of Animals (CEUA) within the scope of UFMT at the time. Therefore, the collection of specimens followed the guidelines of the Conselho Federal de Biologia and of the Sociedade Brasileira de Herpetologia.

Data Availability

Supporting data is available at: https://figshare.com/articles/dataset/HerpetofaunaSaes_csv/26352553/1.

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Article

restauraRapp – An R package to subsidize forest restoration planning in Riparian Permanent Preservation Areas (PPAs)

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Abstract: The Law No. 12,651/2012 brought changes to the widths of riparian Permanent Preservation Areas (PPAs) to be restored, which now also depend on the size of the rural property in fiscal modules, which in turn, varies depending on the municipality. Because of this, defining the environmental liabilities of these PPAs is complex, requiring geoprocessing procedures and information that are not always readily available. To automate the identification of these areas to be restored, the *restauraRapp* package was created, which presents a set of functions for automating four steps of the procedure: I) Data acquisition; II) Classification of property sizes; III) Processing information on land use and cover data in riparian areas; IV) Cartographical and tabular output generation. This package can support landscape planning focused on the restoration of riparian PPAs, potentially assisting NGOs, municipality governments and landowners.
Keywords: *Environmental policy; Brazilian Forest Act; Landscape management; GIS.*

restauraRapp – Um pacote R para subsidiar o planejamento da restauração florestal em Áreas de Preservação Permanente (APPs) Ripárias

Resumen: A Lei nº 12.651/2012 trouxe alterações nas larguras de Áreas de Preservação Permanentes (APPs) ripárias a serem restauradas, que passam a depender também do tamanho do imóvel rural em módulos fiscais, que por sua vez, varia conforme o município. Por conta disso, a definição do passivo ambiental dessas APPs é complexa, exigindo procedimentos de geoprocessamento e informações nem sempre prontamente disponíveis. Para automatizar a identificação destas áreas de passivo ambiental, foi criado o pacote *restauraRapp*, que apresenta um conjunto de funções para a automatização de quatro etapas do procedimento: I) Aquisição de dados; II) Classificação dos tamanhos dos imóveis; III) Processamento das informações sobre o uso e cobertura do solo em áreas ripárias; IV) Processamento e geração de resultados em formato cartográfico e tabular. O pacote gera resultados que podem auxiliar no planejamento de paisagens focado na recuperação de APPs ripárias, auxiliando ONGs, governos municipais e proprietários de terras.
Palavras-chave: *Políticas ambientais; Código Florestal Brasileiro; Gestão de paisagem; SIG.*

Introduction

The conversion of natural habitats into anthropogenic areas threatens biodiversity and ecosystem services and functions, and is one of the major conservation challenges today (Foley et al. 2005, Tscharntke et al. 2012). Protected areas (PAs) alone are not sufficient to prevent biodiversity decline (Joppa et al. 2008, Williams et al. 2022), and lack of planning and political measures makes the establishment of new conservation units much harder (Bernard et al. 2015, Oliveira et al. 2017). Climate change brought additional threats to PA's ability to guarantee biodiversity conservation, since it alters biophysical conditions, potentially making current PAs unsuitable for future species conservation (Araújo et al. 2011). Therefore, species conservation depends on anthropogenic landscapes (Melo et al. 2013), also because they can facilitate species movement between currently suitable sites to future suitable ones (Huang et al. 2020). Additionally, the synergistic effects of climate change and biodiversity simplification in fragmented landscapes compromise the provision of key ecosystem services (Fahrig et al. 2011). Thus, the proper management of human-modified landscapes is essential for biodiversity conservation and human well-being (Summers et al. 2012).

Brazil has a long history of environmental legislation (Drummond & Barros-Platiau 2006), particularly tackling forest destruction and the protection of rivers, water sources and slopes (Brasil 2010). However, it was not until the 20th century (Brazilian Forest Act 1934 and Brazilian Forest Act 1965) that the environmental legislation started to directly influence the territorial land use and cover. The first Brazilian Forest Act (Decreate 23.794/34, art. 4o) focused on forest destruction on environmentally sensitive areas, such as along rivers and water springs. The concerns with waterway banks were also expressed in the Water Code (24.643/34), however, with an administrative aspect and not aimed at environmental conservation. It was only in 1965 (4.771/65) that the Permanent Preservation Areas (PPAs) were properly defined (Brancalion et al., 2016) and then described as forests and other forms of natural vegetation located along rivers or any other watercourse, in a marginal strip whose minimum width was 5 (five) meters for rivers of less than 10 (ten) meters in width, equal to half the width of the water courses that measure from 10 (ten) to 200 (two hundred) meters of distance among river banks and 100 (one hundred) meters for all courses whose width is greater than 200 (two hundred) meters. The PPAs also encompassed areas around natural and artificial ponds and lakes, water springs, top of the hills, slopes greater than 45°, "restingas" and dunes that hold sandy soil, the edges of plateaus and areas with altitude above 1,800 m (Law no. 4.771/65).

Later in 1986 (Law no. 7,511, July 7th) and in 1989 (Law no. 7,803, July 18th) more detailed definitions of PPAs were provided, including the statement that the PPA width should be measured starting from the maximum water level, and an enlargement of the riparian PPAs, for instance 30 m from rivers with less than 10 m of width, 50 m for rivers between 10 and 50 m of width, 100 m for rivers between 50 and 200 m of width, 200 m for rivers between 200 and 600 m, 600 m of width for rivers wider than 600 m of width, also, a 50 m buffer around the water springs were defined as PPA (Table 1).

After 13 years of debate in the Brazilian Congress and despite warnings from the academic community (e.g. Metzger 2010, Metzger et al. 2010, Nazareno 2012, Sparovek et al. 2012), regarding the interruption of important ecosystem services and the potential erosion of biodiversity due to the proposed changes (Toledo et al. 2010, Develey & Pongiluppi 2010 and a series of manuscript from this special issue, Alarcon et al. 2015), in 2012 a new law was promulgated that regulates the exploitation, conservation and restoration of the native vegetation in the Brazilian territory. The law no. 12,651 was sanctioned with vetoes by President Dilma Roussef on May 25th 2012, and later altered by law no 12,727, on October 17th 2012. One of the main changes in the Brazilian Forest Act (2012) lies in its transitional provisions. These provisions establish priority conditions for the restoration of PPAs in rural properties with consolidated areas. These areas receive differentiated treatment, whereby landowners must restore minimum vegetation buffers along watercourses, with variable widths (5 to 100 meters) depending on the type of watercourse and property and river sizes. This has added complexity to defining the width of riparian areas that require restoration, as it now demands more detailed information and geoprocessing expertise. The process involves partitioning the hydrography and properties data, making it more repetitive due to the need to create, manipulate and merge buffers of varying sizes multiple times, increasing the likelihood of processing errors. In order to help on this task, we built r*estauraRapp,* which is an R package designed to seamlessly delimitate and quantify the riparian PPAs of water bodies, according to property sizes, therefore helping to define areas that should be restored. Here we introduce the *restauraRapp* R package, which aims to facilitate the steps to generate and quantify potential restoration areas along PPAs defined by law:

A) Automate data download and acquisition,
B) Divide property by sizes, based on fiscal module, as defined in the Brazilian Forest Act 2012,
C) Create the buffer zones and clip the areas, according to the width defined by the Brazilian Forest Act 2012, based on property sizes and river widths,
D) Export results in tabular and cartographic formats.

Methods

1. Data

Three datasets are used in order to calculate and define PPAs that are preserved and the ones that are degraded and could be restored (Table 2). These datasets are crucial, as they include all variables necessary to determine the preservation status of PPAs. Land use/cover and hydrography data are available at the municipality scale from the FBDS database, while CAR data is available at the state level and needs to be filtered to align with the other datasets. Although we strongly recommend using data at the municipality scale, the package also supports larger areas (such as stacked municipalities), provided the data sources and function parameters are respected (see below for details on function parameters). Ultimately, it is the user's responsibility to define the study area to which these datasets will be applied during the process.

Table 1. PPA protection law changes since the first Brazilian Forest Act (1965) until the last change in May/2000.

Until 1965	September 15th 1965	July 7th 1986	July 18th 1989	May 26th 2000
Brazilian Forest Act 1934	Brazilian Forest Act 1965	Law no. 7,511	Law no. 7,803	MP no. 1956-50/00
No PPAs, but with some protection, if the area was declared as "protected forest"	Obligation to preserve native forests along watercourses, varying by its size.	Same as 4.771/65, but the native forests along watercourses enlarged, varying between 30 meters to 150 meters.	Riparian areas along intermittent water springs become PPA	PPAs do not need to be covered by native vegetation to be a preservation area.

Table 2. Data description and source. *Fundação Brasileira para o Desenvolvimento Sustentável; **Sistema Nacional de Cadastro Ambiental Rural.

Data	Type	Source	Description
Land use/cover	Shapefile	FBDS*	Spatial dataset with land use and cover information based on RapidEye imagery with 5 meters resolution at spatial resolution 1:10000
Hydrography	Shapefile	FBDS*	Hydrography spatial dataset that includes rivers and other water bodies types (spring and lakes) at spatial resolution 1:10000
CAR	Shapefile	SICAR**	Spatial dataset with properties by state

Table 3. Riparian buffer widths that are considered in the package according to property class sizes.

	Property class sizes in Fiscal Modules (FM)			
Hydrography	**< 1**	**Between 1 e 2**	**Between 2 e 4**	**> 4**
Rivers	5 m	8 m	15 m	20 m
Around the springs	15 m	15 m	15 m	15 m
Around the natural lakes	5 m	8 m	15 m	30 m

The Land use/cover FBDS dataset was mapped through supervised classification of Rapid Eye imagery level 3A (5m resolution) and edited in a scale of 1:10,000 in order to correct classification and discontinuity errors (Rezende et al. 2018). This dataset was chosen because it is a vector created from a raster map with 5 m pixels resolution (5 m × 5 m). This resolution provides the necessary scale to evaluate the area to be restored in the smallest possible PPA width (5 m in properties smaller than one Fiscal Module (FM), see below) and is the best resolution available for land use/cover in Brazil (Rezende et al. 2018). Also, it was made based on supervised classification of images starting in 2013 (FBDS, 2023), first year after the Brazilian Forest Act 2012 promulgation and the nearest map available to mid 2008, the date used as the baseline for the new legislation.

The hydrography spatial dataset is divided in four parts, considering the width and the type of the water body, rivers narrower than 10 meters, rivers wider than 10 meters, lakes and springs. This dataset was created and refined by compiling official cartographic sources at the best available scales and adapting them using RapidEye images at a 1:10,000 scale. Contours generated from the SRTM digital elevation model (30 m × 30 m/pixel) were used as a secondary reference (Rezende et al. 2018). This dataset represents the most refined data currently available.

The CAR (Cadastro Ambiental Rural) is a national electronic public register established by Law No. 12,651 and regulated by Normative Instruction MMA No. 2 of May 5, in 2014. The registration is mandatory for all rural properties and aims to integrate environmental information of the properties (SICAR [s.d]). The CAR is also the first step for the establishment of an Environmental Regularization Program (in portuguese Programa de Regularização Ambiental, PRA), where the objective is to promote the environmental regularization of the property including the recovery, recomposition, regeneration or compensation of environmental liabilities (SICAR [s.d]).

The property polygons are obtained from CAR data (“Perímetro dos imóveis” option) from the official database system (SICAR). This information is the only one that still needs to be accessed manually, all the other cartographic information are automatically retrieved by the functions present in r*estauraRapp* package (see below). Based on this information, property sizes are defined according to the fiscal modules (FM), which vary between municipalities. The Law No. 12.727 alterated the Law No. 12,651, and now it considered four sizes of rural properties, that we named here: micro (up to 1 FM), small I (from 1 to 2 FM), small II (from 2 to 4 FM) and large (greater than 4 FM). The FM is a measurement unit, in hectares, determined by The National Institute for Colonization and Land Reform (INCRA) and different for each Brazilian municipality, in which the value of one FM could vary from 5 to 100 hectares depending on municipality land use aspects (Embrapa [s.d]).

2. *Buffer sizes*

The functions of the package divide properties by size and create a set of buffers according to property class sizes, as shown in Table 3. These sizes define the widths of the areas that, if degraded, should be restored according to the Brazilian Forest Act (2012). The transitional provisions define that regularization using the new PPA buffer zones is only possible when consolidated areas are present. These are areas that were deforested and/or altered before July 22, 2008, within preserved buffer zones. In these cases, the continuity of economic activities is permitted, as long as the property is registered in the CAR. Following registration, the landowner must also adhere to the PRA, which requires a detailed recovery plan for PPAs and Legal Reserves (LR). By completing these steps, the landowner becomes eligible for the benefits of the transitional provisions, including the new PPA buffer widths (ranging from 5 to 100 meters), permission to continue activities in consolidated areas, exemption from previous fines, and other advantages.

For rivers in properties with more than four fiscal modules, buffer width output was standardized to 20 meters, because such buffers can vary in width (between 20 meters and 100 meters), in accordance with the property’s PRA, and may need to be checked individually.

In addition, two alternative scenarios were provided to evaluate municipal regions where no CAR registration is available. These scenarios assume that areas without CAR correspond either to properties smaller than 1 FM (classified as micro properties, narrowed area to be restored) or larger than 4 FM (classified as large properties, wider areas to be restored), applying the respective buffer sizes for each class. Users have the discretion to apply one or both scenarios as needed.

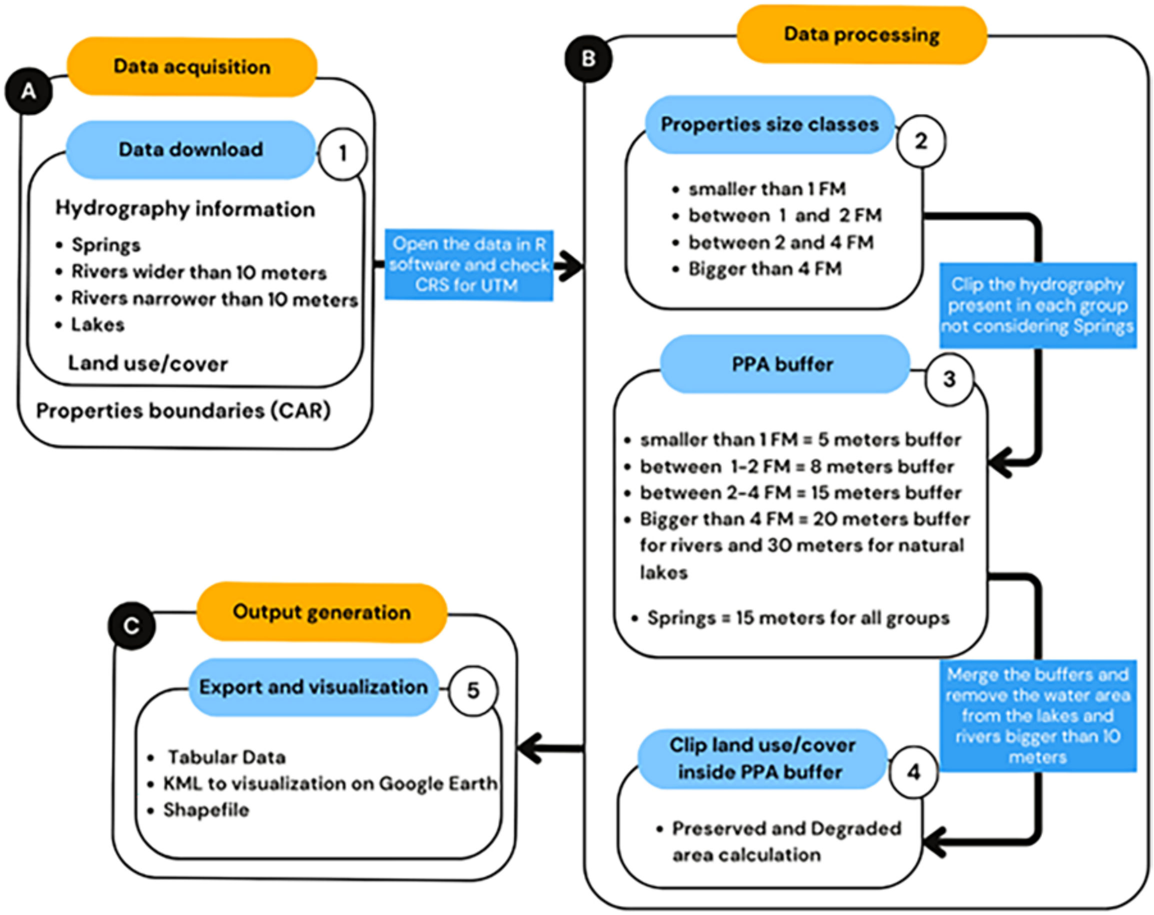

Figure 1. Workflow for assessing preservation status of PPAs, with automated steps using the restauraRapp package. (A) Data acquisition, 1. automated data download. (B) Data processing, 2. splitting properties by size based on fiscal modules defined by the Brazilian Forest Act (2012), 3. generation of PPA buffers according to properties sizes, 4. clipping PPA areas splitting among preserved or degraded. (C) Output generation, 5. exporting results in tabular data, shapefiles, KML formats.

3. *Process development*

All the functions of the *restauraRapp* package (see below) were designed and developed to facilitate the assessment of PPA preservation status. The workflow for buffer creation and PPA evaluation involves a structured process for manipulating spatial datasets (Figure 1). The first step is to download the hydrography (all four files) and land use/cover spatial datasets from FBDS database, which is an automated process (see below), while the properties boundaries datasets from the SICAR database must be manually downloaded. Once the data is ready, manually load the required spatial datasets (Table 2), ensuring that all datasets are checked for Coordinate Reference System (CRS) consistency. Universal Transverse Mercator (UTM) system should be used to enable accurate measurement of PPA's areas. Ensuring that all datasets use the same CRS is crucial for the proper functioning of the functions. This must be verified and, if necessary, adjusted. Otherwise, the process will result in an error. Next, select the property boundaries of the target municipality from the SICAR dataset, and divide in classes by its FM size to determine the area occupied by each class. After classifying the properties, the polygons from each class are used to clip the hydrography spatial dataset (rivers narrowed than 10 meters, rivers wider than 10 meters and lakes), excluding springs. Springs were not included, as their buffer widths do not vary based on property size.

Once the hydrography dataset is divided, buffers are applied according to the PPA width defined by the Brazilian Forest Act (2012) for each property size class (Table 3), generating the corresponding PPA areas. The buffered datasets are then merged. To ensure that only the PPA areas are measured, the internal areas of the polygons representing rivers wider than 10 m and lakes, corresponding water bodies, are cropped out. Finally, the land use/cover spatial dataset is clipped using the buffered areas to obtain the information of the preservation on the preservation status of the PPAs.

With the information on the preservation status of the PPAs, the user can export the results in various formats, such as a tabular file containing the values for each property class or for each property, a shapefile for spatial data storage and analysis in GIS software, and a KML file for visual validation using high-resolution satellite images.

All the development was made using the coding program R v4.0 (R core team 2023) and RStudio v1.4.1743 (RStudio team 2023), using the packages *sf* (Pebesma 2018), *curl* (Ooms 2024), *XML* (Lang 2024), *abjutils* (Lente & Trecenti 2022) and *dplyr* (Wickham et al. 2023).

Table 4. Functions of the package *restauraRapp*, parameters for the execution and function description.

Function	Parameters	Descriptions
resapp_fbds_dados	State (text), municipality (text),	Automatically downloads land use and cover and hydrography datasets, creates a directory with the name FBDS_MUNICIPALITY in your home folder and saves the datasets there
resapp_car_class	The CAR spatial dataset (.shp) downloaded from the SICAR website	Splits property by class sizes, returning a spatial dataset (*.shp) to each class size
resapp_car_info	The CAR spatial dataset (*.shp) downloaded from the SICAR website, municipality name (text), option between table of spatial dataset (text)	Returns the total area and the number of properties by class size for the focal municipality. Outputs can be a table or a spatial dataset (*.shp)
resapp_app_buffer	The hydrography spatial datasets (*.shp) automatically downloaded from the FBDS, the CAR spatial dataset (*.shp) downloaded from the SICAR website (*.shp), the land use and land cover spatial dataset downloaded from FBDS website (*.shp), scenario option (text)	Unifies hydrography spatial datasets and creates the buffer with the widths to be restored by class size, returning the intersection of these buffers with the land uses as a spatial dataset (*.shp) for the chosen scenario (see below for scenario details).
resapp_app_info	The scenarios generated by “resapp_app_buffer” (*.shp), CAR spatial dataset (*.shp) downloaded from the SICAR website (*.shp), option between table and spatial dataset (text)	Uses the outputs from the “resapp_app_buffer” function, unites the different land use/cover in the classes “restore” and “preserved”, returning the spatial dataset (*.shp) or data tables with the areas of these classes (restores and preserved) by property class sizes, for all properties or by property
resapp_app_kml	Scenario 1 (only properties with CAR) from the function “resapp_app_buffer” (*.shp), municipality (text), state (text)	Two options of spatial dataset (*.kml) files i. the entire PPA and ii. the areas to be restored, both by property class size

Results

1. *Package functions*

The *restauraRapp* package currently includes six functions that automate the entire processes, from downloading data and creating property size classes, to clipping the PPAs based on property class sizes and river widths, and finally generating and organizing the results. It is important to note that PPAs sizes also depend on the presence of consolidated areas as of 2008 and the type of activity. Each function is detailed in Table 4.

2. *Downloading the data*

The function `resapp_fbds_dados` (Equation 1, Table 4) downloads land use/cover and hydrography datasets. The parameters are “MUNICIPALITY” which is the name of the municipality in capital letters and, when necessary, separated by underscore, and “UF” which is the state abbreviation from which the municipality belongs. These are the only parameters needed to execute this function, which will download and save the required information in a folder named “data”. This folder is automatically created while running the function within the user’s R project directory. If the function is used outside an existing R project directory, the “data” folder is created in the R base working directory, which can be identified using the base R function “getwd()”. This ensures that the data is stored in an organized, traceable and easy to access manner.

```
resapp_fbds_dados(UF, MUNICIPALITY)
```

Equation 1: Function for downloading land use/cover and hydrography datasets from FBDS website.

So far, the CAR dataset needs to be manually downloaded from the SICAR database (https://www.car.gov.br/publico/estados/downloads), because of the presence of a captcha, preventing access through direct link. The “AREA_IMOVEL” file, downloaded through the option “Perímetros dos imóveis” provides information for the buffer calculation function in the package.

3. *Property sizes classification*

The function `resapp_car_class` (Equation 2, Table 4) classifies properties based on the number of FM, which vary in size depending on the municipality. The input “CAR” refers to the spatial dataset (*.shp) containing property boundaries information. This function returns a spatial dataset (*.shp) with the classified polygons, which can be used directly or as an internal input for other functions within the package.

```
resapp_car_class(CAR)
```

Equation 2: Function responsible for classifying CAR spatial dataset according to property class sizes.

The function `resapp_car_info` (Equation 3, Table 4) classifies the properties by class size, Returns the total area and the number of properties by class size for the focal municipality.

```
resapp_car_info(CAR, mun, type)
```

Equation 3: Function for obtaining information about property sizes distribution.

Where “CAR” is a spatial dataset (*.shp) carrying the properties information. If “mun” is defined, municipality limits will be used to get the portions of the properties that are exclusively within the focal municipality. If NULL, the results will include the entire property limits, also including portions that extrapolate municipality borders. It is important to note that the name of the municipality must be written according to the Brazilian Institute of Geography and Statistics (IBGE) database.

It allows users to select among the total area of the properties or just the areas within the limits of the municipality, according to what is registered in CAR, returning cartographic information (default), or just the data table. To select the areas within the municipality limits, it is necessary to add the name of the municipality to the function (parameter “mun”), which will automatically get the county limits and return the result according to the user’s choice, important to notice that the name of municipality must be written accordingly to Brazilian Institute of Geography and Statistics database. For the output options, the parameter type could NULL (default) for spatial dataset (*.shp) and “df” for table.

4. *Buffering the PPAs*

The function `resapp_app_buffer` (Equation 4, Table 4) creates buffers referring to the PPAs (Table 3) and clips the land use and land cover data (Table 2). This function takes a considerable processing time, depending on municipality scale and hydrography complexity, but is comparable to a geoprocessing process made in R, responding directly to hardware performance. For this reason, currently it has a parameter to be executed in parts, split by property class sizes and/or scenarios (see below). To execute the entire process at once, use option “tudo”, to split the process use: “micro” for properties below 1 FM, “peq12” for properties between 1 and 2 FM, “peq24” for properties between 2 and 4 FM and “grande” for properties above 4 FM. To split by scenario the “tipo” parameter must be “out2” for Scenario 2 or “out3” for Scenario 3 (see below) the option “tudo” (out1) only works for the areas that have a CAR registered.

This function automates the division of properties by size class, the creation of buffers using the Brazilian Forest Act (2012) widths for consolidated areas, and the clipping of these buffers with land use/cover data, followed by the calculation of preserved and deforested areas. With this approach, the user no longer needs to repeatedly create buffers of different sizes, manually calculate property areas, or perform other data manipulations. This reduces the likelihood of processing errors and fosters a more streamlined, standardized workflow.

```
resapp_app_buffer(mapa_NAS, mapa_MDA, mapa_
    RMS, mapa_RMD, CAR, uso, tipo)
```

Equation 4: Function to create the buffers, using the widths defined by law (Table 3), and clips the land use/cover inside it.

Where “mapa_NAS” is an object carrying the information about the springs; “mapa_MDA” carries the information about lakes; “mapa_RMS” have information about the rivers smaller than 10 m; “mapa_RMD” have information about rivers bigger than 10 m; “CAR” are the properties limits; “uso” is the land use/cover of the municipality; “tipo” is the process that need to be made (according to property class size, or all properties).

In some cases, municipalities do not have lakes or rivers bigger than 10 m inside its limits, in these cases the parameters “mapa_MDA” and “mapa_RMD” accept NULL values. If one of the class sizes is selected (in “tipo”), the union of the objects is needed before proceeding in other functions.

Based on the data and the procedures above, three different scenarios can be created:

- Scenario 1 (“out1”): All the properties with CAR are evaluated, and the restoration areas are evaluated based on river width and property size;
- Scenario 2 (“out2”): Only the land that is not covered by CAR is considered, and all the area is treated as properties smaller than 1 FM, *i.e.,* the smaller buffer width that should be restored (5 m);
- Scenario 3 (“out3”): Only the land that is not covered by properties in the CAR is considered, and these areas are treated as large properties (> 4 FM), *i.e.*, the wider buffer possible (20 m);

5. *Compiling and organizing the results*

The function `resapp_app_info` (Equation 5, Table 4) divides the different land uses/cover as “Preserved” (Forest and non-forest native habitats) and “Restore” (other uses, except water) classes. This function has options to return the results as a shapefile (parameter “tipo = tudo”), as tabular data for the municipality (parameter “tipo = df”) or as tabular data for each property (parameter “tipo = prop”).

```
resapp_app_info(out1, out2, out3,
    CAR, tipo)
```

Equation 5: Function responsible for splitting land use/cover data into “Preserved” and “Restore” and creates the shape or tabular output from the results of Equation 4.

Where “out1”, “out2” and “out3” are the results of the function `resapp_app_buffer (Equantion 4)` respectively Scenarios 1, 2 and 3. “CAR” is a spatial dataset with property limits and “tipo” the type of output that will be created (“tudo” for spatial dataset *.shp, “df” for a tabular output for the entire municipality, and “prop” for a tabular output by each property). For the “tudo” option, it is necessary to provide only the output from Scenario 1 (“out1”) from the function `resapp_app_buffer (Equation 4)`. The “df” option needs the information of all three scenarios (“out1”, “out2” and “out3”), and the “prop” option needs the properties limits from the CAR spatial dataset and the Scenario 1 result (“out1”).

Finally, the function `resapp_app_kml (Equation 6, Table 4)` exports the spatial datasets obtained by executing the function `resapp_app_buffer (Equation 4)` function as KML files to be opened in Google Earth, which is a free and an easy to use tool. The final files of this function are the complete PPA polygons based on property sizes and the polygons of the areas to be restored, also splitted by property size class.

```
resapp_app_kml (out1, MUNICIPALITY, UF)
```

Equation 6: Function to export the spatial dataset as a KML file.

Where "out1" is the output from function `resapp_app_buffer (Equation 4)`. "MUNICIPALITY" is the name of the municipality and "UF" is the abbreviation of the state to which the municipality belongs. The "MUNICIPALITY" and "UF" parameters are for file name generation and can be a NULL value.

Discussion

The changes brought by the Brazilian forest Act 2012 aim to facilitate the compliance with the environmental legislation in rural areas (Soares-Filho et al. 2014), however, they also bring additional difficulties to define and monitor environmental legislation observance (Garcia et al. 2013, Soares-Filho et al. 2014, Brancalion et al. 2016). With *restauraRapp*, we show that the areas to be restored in PPAs can be straightforward mapped in a precise and spatially explicit way. This can help initiatives for environmental legislation compliance, since it can automatically identify forest deficits for large areas and help in the planning of the restoration initiatives, allowing restoration costs to be reduced and area prioritization to be done. Since cost and planning are among the biggest challenges in restoration initiatives (Ifthekar et al. 2016, Strassburg et al. 2019, Brancalion et al. 2019, Schimetka, 2024), the package can help restoration actions to gain scale.

Additionally, *restauraRapp* can help validate, mainly through visual analysis, the information present in the CAR system, since the information is self-declared, and incongruities are commonly found in the dataset (Melo et al. 2021). It is noteworthy that watercourses PPAs play a key role within agricultural landscapes, as they provide connections between the native remnants (Rosot et al. 2018, Rother et al. 2018), as well as the protection of water resources (Valera et al. 2019). Supporting decision-making through the approximation of science based solutions and police makers, as well the application of these decisions, are also essential for reconciling agricultural production and conservation (Ferreira et al. 2012, Tavares et al. 2021). In this context, the proposed R package can subsidize the prioritization of areas for ecological restoration, either because they present a higher importance for water resources conservation, or because of forest fragment connectivity, or even because they are located in a much more resilient region of the landscape.

Additionally, *restauraRapp* automates the downloading and standardization of data sources, streamlining access to information and organizing data more efficiently. The system automatically creates directories in the selected project folder, ensuring organized data storage. While the time required to run the package may vary depending on the municipality size and hydrography complexity, it remains comparable to standard geoprocessing tasks in R. However, automation allows for a smoother, more efficient workflow by minimizing manual intervention. By automating the repetitive steps, the package also significantly reduces the risk of common geoprocessing errors, ensuring greater accuracy and consistency in the results.

In summary, *restauraRapp* is a tool developed to facilitate the delimitation of riparian areas that should be restored based on the Brazilian Forest Act (2012). Therefore, it is a tool that can help to plan landscape management actions towards restoration of riparian areas, and can aid the conservation of aquatic and terrestrial environments in private lands in Brazil.

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Alexandre Camargo Martensen: Conceptualization; Resources; Founding acquisition; Methodology; Writing – review & editing.

Conflicts of Interests

The author(s) declare(s) that they have no conflict of interest related to the publication of this manuscript.

Ethics

This study did not involve human beings and/or clinical trials that should be approved by one Institutional Committee.

Data Availability

The functions, as well all the files for the execution of an example of the package run, are available at: https://github.com/NEEDS-LS/restauraRapp.

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Short Communication

Enhancing public policies for Brazilian bullfrog farming

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Abstract: Aquaculture is a globally significant industry involved in the cultivation of aquatic organisms to provide food for the population and boost the economies of producing countries. Brazil stands out in the global production of bullfrogs, but this activity has generated ecological and economic problems. The lack of regulation and control can threaten biodiversity, as these bullfrog's act as vectors for pathogens such as chytrid fungus *Batrachochytrium dendrobatidis* (Bd) and the virus *Ranavirus* (Rv). Both Bd and Rv are associated with declines and mass extinctions of amphibians worldwide. Studies in Brazil have identified a high prevalence of these pathogens in bullfrog farms, posing threats to both the industry and the environment. The absence of effective regulation in bullfrog farming is a challenge, as chytridiomycosis and ranaviruses are diseases that must be reported to the World Organisation for Animal Health, requiring management of impacts on bullfrog production. Therefore, our goal was to promote collaboration among researchers, public entities, and industry leaders, aiming to review existing legislation and propose effective biosecurity measures. We conducted extensive reviews on bullfrog farming in Brazil, participated in and organized meetings with government and industry representatives, and actively engaged in Species Conservation Action Plans. Within this context, efforts have been made to map frog farms and develop biosecurity proposals aimed at pathogen control. We conclude that intersectoral collaboration is crucial for the development and implementation of effective policies. Furthermore, we present a detailed review of the current legislation directly related to the topic to facilitate understanding of the current situation in Brazil. The implementation and maintenance of more comprehensive regulations, along with the adoption of sustainable practices in frog farming, are essential to ensure the industry's sustainability and biodiversity conservation.
Keywords: *Aquarana catesbeiana; biodiversity conservation; biological invasion; chytridiomycosis; inter-sectoral collaboration; ranaviruses.*

Aprimoramento de políticas públicas sobre a ranicultura brasileira

Resumo: A aquicultura é uma indústria global importante na prática do cultivo de organismos aquáticos para fornecer alimentos à população e impulsionar a economia dos países produtores. O Brasil se destaca na produção global de rãs-touro, mas essa atividade tem gerado problemas ecológicos e econômicos. A falta de regulamentação e controle pode ameaçar a biodiversidade, já que essas rãs atuam como vetores de patógenos como o fungo quitrídio *Batrachochytrium dendrobatidis* (Bd) e o vírus *Ranavirus* (Rv). Tanto o Bd, quanto o Rv, estão associados a declínios e extinções em massa de anfíbios em todo o mundo. Estudos no Brasil identificaram alta prevalência desses patógenos em ranários, representando ameaças à indústria e ao ambiente. A ausência de regulamentação eficaz na ranicultura é um desafio, já que a quitridiomicose e a ranavirose são doenças de notificação obrigatória à Organização Mundial de Saúde Animal, exigindo gestão dos impactos na produção de rã-touro. Portanto, nosso objetivo foi promover a articulação entre pesquisadores, órgãos públicos e líderes do setor, visando revisar as legislações existentes e propor medidas eficazes de biosseguridade. Realizamos extensivas revisões sobre a ranicultura no Brasil, participamos e promovemos reuniões com representantes governamentais e produtivos e nos engajamos ativamente nos Planos de Ação para a conservação de espécies. Nesse contexto, esforços foram feitos para mapear ranários e desenvolver propostas de biosseguridade visando o controle de patógenos. Concluímos que a colaboração entre setores é crucial para a elaboração e implementação de políticas eficazes. Além disso, apresentamos uma revisão detalhada da legislação vigente diretamente relacionada ao tema, visando facilitar a compreensão da situação atual no Brasil. A implementação e manutenção de regulamentações mais abrangentes, assim como a adoção de práticas sustentáveis na ranicultura, são essenciais para garantir a sustentabilidade da indústria e a conservação da biodiversidade.

Palavras-chave: *Aquarana catesbeiana; biologia da invasão; colaboração intersetorial; conservação da biodiversidade; quitridiomicose; ranavirose.*

Introduction

Aquaculture, the cultivation of aquatic organisms under controlled conditions, stands out as an industry with a significant global impact on the food supply and the economy (Garlock et al. 2019). According to the Food and Agriculture Organization of the United Nations (FAO), global aquaculture production reached a historic milestone of 114.5 million tons in 2020, with an estimated value of US$263.6 billion (FAO 2024). Fish, crustaceans (especially shrimp), and molluscs, such as oysters and mussels, are commonly cultivated in this context. Furthermore, the practice of frog farming, primarily aimed at breeding the species *Aquarana catesbeiana* (Shaw, 1802), known as the bullfrog, is also part of the aquaculture field and has experienced a notable increase in popularity, particularly in Asia and South America (Ribeiro & Toledo 2022).

The bullfrog, native to North America, has been introduced to various regions of the world for commercial purposes, particularly for meat consumption, becoming one of the most widely distributed invasive species, with significant ecological and economic impacts (Falaschi et al. 2020). This scenario is largely due to escapes, or intentional releases during the breeding process of the species (Both et al. 2011). Brazil stands out as one of the pioneers and largest producers of bullfrogs globally (Ribeiro & Toledo 2022), as well as the country with the greatest amphibian diversity in the world (Frost 2024). However, the absence of regulation and control in the breeding and trade of frogs is emerging as a growing concern for biodiversity conservation.

Bullfrogs are known to carry and serve as vectors for various pathogens, such as the chytrid fungus (*Batrachochytrium dendrobatidis* or Bd), which causes chytridiomycosis (Longcore et al. 1999), and Ranavirus (*Ranavirus* spp. or Rv), responsible for ranavirosis (Mazzoni et al. 2009, Schloegel et al. 2010, O'Hanlon et al. 2018). These pathogens have been associated with mass mortality events and species extinctions on a global scale. While Bd is distributed worldwide, causing declines and extinctions of various amphibian species (Scheele et al. 2019), Rv has an apparent more restricted geographic distribution, but is capable of infecting a greater variety of organisms, including fish, reptiles, and amphibians (Brunner et al. 2015).

In Brazil, ranavirosis has been responsible for mass mortality events among amphibians and fish (Mazzoni et al. 2009, Ruggeri et al. 2019, 2023), while Bd has been associated with various population declines of native amphibian species (Carvalho et al. 2017). Both pathogens have been found in Brazilian frog farms. Studies conducted in the states of São Paulo, Paraná, and Santa Catarina reported a high prevalence of the fungus (Ribeiro et al. 2019, Santos et al. 2020), as well as the detection of high loads of Bd in wastewater reaching natural water bodies (Ribeiro et al. 2019). Similarly, Rv has been detected in Brazilian frog farms causing mass mortality in bullfrogs (Mazzoni et al. 2009, Candido et al. 2019). The presence and evolution of these pathogens in farming environments pose a significant threat to both the frog industry and the environment (Ribeiro & Toledo 2022).

Both chytridiomycosis and ranavirosis are notifiable diseases under the World Organisation for Animal Health (WOAH) (Schloegel et al. 2010), meaning they must be reported to government authorities. This prompts government intervention, official laboratory involvement, and can raise serious ecological, economic, and reputational concerns, particularly within the context of bullfrog production, highlighting the need for effective management. These issues extend beyond the problem of the species invasiveness to broader epidemiological concerns. The introduction and geographic expansion of invasive exotic species, along with pathogen dissemination and inadequate waste management from bullfrog production, pose highly relevant threats that require immediate attention (Ribeiro et al. 2019). Identifying and controlling these pathogens are crucial aspects to ensuring the long-term sustainability and success of the bullfrog farming industry.

The lack of information and coordination among sectors involved in frog farming has posed a challenge to the creation of effective regulations. Although there are controversies regarding environmental regulation, its importance in safeguarding health and the environment is acknowledged (Clausen et al. 2023). The "triple helix" model emphasizes collaboration among industry, regulators, and academics to promote regulatory innovation, showing the need for cooperation in formulating and implementing regulations (Zhou & Etzkowitz 2021). Empirical case studies to examine what worked and what did not are fundamental to better understand the relationship between science and policy (de Mello et al. 2022).

Thus, our aim was to promote collaboration among researchers, public agencies, and industry leaders, with the objective of reviewing existing legislation, proposing effective biosecurity measures, regularly monitoring pathogens, treating contaminated individuals and water, and discussing specific solutions for each priority area. By adopting responsible practices and conservation efforts, we aspire to ensure long-term sustainability and profitability in frog farming, safeguarding animal health and ecosystem quality.

Material and Methods

We conducted a comprehensive review of the history and status of bullfrog farming in Brazil, with a focus on identifying relevant legislation and the government agencies responsible for its enforcement. To ensure a thorough analysis, we consulted various national and state databases, publicly accessible, including legal archives, government reports, and environmental regulations related to aquaculture and wildlife management. Information on bullfrog farmers, occurrences of wild bullfrogs, and the Bd and Rv pathogens was gathered from scientific articles, databases, producer associations, informal networks, and available online sources.

Our online search utilized platforms such as Google, Google Scholar, Web of Science, SpeciesLink, GBIF, YouTube, and social media. To locate bullfrog farms and relevant legislation, we employed a range of Brazilian Portuguese keywords, including aquaculture, ranaculture, bullfrog, bullfrog farm, frog farming, frog meat, frog production, bullfrog trade, environmental regulations, aquaculture legislation, wildlife management, laws, decree, legislation, and regulation. We refined our search by matching these keywords with specific regions, states, or municipalities in Brazil, allowing us to gather detailed information on locations and the relevant legal frameworks.

To address regulatory gaps and reduce the negative impacts of bullfrog farming on native amphibian populations, we organized a series of meetings with key stakeholders. These meetings included representatives from the public sector—at both state and federal levels—as well as from the private sector and academic researchers. The discussions focused on evaluating current regulations, identifying shortcomings, and proposing actionable strategies to improve regulatory oversight and promote sustainable bullfrog farming practices.

In addition to these efforts, we actively participated in the development and implementation of National Action Plans (PANs) and Territorial Action Plans (PATs), coordinated by government agencies. We took a proactive role by proposing conservation-focused actions that align with species protection objectives, ensuring that the ecological impacts of bullfrog farming are considered in the regulatory framework.

Results and Discussion

1. A brief overview of bullfrog farming in Brazil

Within the context of aquaculture, which encompasses the breeding and cultivation of aquatic organisms, lies frog farming, a specialized practice focused on the production of frogs, often for commercial purposes. In Brazil, this activity began in the 1930's with the importation of the first bullfrogs (*Aquarana catesbeiana*) from North America (Ferreira et al. 2002). In 1935, the first commercial bullfrog farm was established in Brazil, located in the state of Rio de Janeiro. Bullfrog farming in São Paulo began in 1939 (Silva et al. 2013). The Brazilian bullfrog market started to gain prominence in the early 1980s. However, many producers abandoned the activity due to inadequate facilities for bullfrog farming and poor management techniques (Braz Filho 2001, Feix et al. 2004, Ribeiro & Toledo 2022).

In 2009, Brazil was ranked as the second largest bullfrog producer, second only to Taiwan (Embrapa 2015. Ribeiro & Toledo 2022). Bullfrog farming presents promising infrastructure, environmental conditions, and market potential in various regions of the country. It is estimated that Brazil once had around 600 established bullfrog farms, along with 15 slaughter and processing industries (Lima et al. 1999). In the Southeast region, there are a total of 144 bullfrog farms distributed across 60 municipalities in the state of São Paulo, 16 in Rio de Janeiro, 10 in Minas Gerais, and three in Espírito Santo (Rodrigues et al. 2010). Currently, 151 bullfrog farms have been mapped, primarily located in the southeastern Brazil, with an estimated production of around 400 tons of bullfrogs (Ribeiro & Toledo 2022).

Brazil has favourable conditions for bullfrog farming, especially due to the climate conducive to the activity. There is significant interest and financial enthusiasm for starting bullfrog farming; however, the production chain for bullfrog farming is still poorly organized, leading to various challenges. These include delays in the environmental licensing process, bureaucratic hurdles in producer registration, and difficulties in product distribution (Cribb et al. 2013). Often, bullfrog farming projects are initiated without adequate prior planning, resulting in a considerable number of frustrated producers who enter and leave the activity year after year (Cribb et al. 2013).

According to Decree No. 62.243, from November 2016, regarding the environmental licensing of aquaculture in the state of São Paulo, any activity involving the cultivation or breeding of organisms with life cycles occurring partially or entirely in an aquatic environment is considered aquaculture (São Paulo 2016; see Table 1 for a review of legal bases for bullfrog farming in Brazil). As a segment of aquaculture, ranaculture or bullfrog farming requires producers entering this sector to register as aquaculturists with the Brazilian Institute of Environment and Renewable Natural Resources (IBAMA). Additionally, rural producers must also register the production with the competent agency of each region. Despite the existence of these regulations, the unification of these registrations or even access to them is unfeasible, complicating research studies and the development of action plans to comply with existing laws.

A significant challenge in bullfrog farming that directly impacts the conservation of native amphibians is the spread of infectious diseases, particularly chytridiomycosis and ranavirus (Mazzoni et al. 2009, Ribeiro et al. 2019, Ruggeri et al. 2023, Santos et al. 2020). Additionally, the introduction of exotic species, such as the bullfrog, into local ecosystems poses serious ecological risks. Bullfrog farming practices have contributed to numerous incidents of invasive in Brazil (Ribeiro & Toledo 2022, Both et al. 2011). The establishment and expansion of bullfrogs in the wild have led to competition with native amphibians, altered ecosystems, and increased pathogen transmission (Falaschi et al. 2020, Melo-Dias et al. 2023, Ribeiro et al. 2019).

In this context, a noteworthy program from the Ministry of Agriculture and Livestock (MAPA) is aimed at the health of frogs produced and commercialized. The Normative Instruction No. 4, from February 2015 (Brasil 2015a), which covers all establishments involved in the cultivation or maintenance of aquatic animals in the national territory, came into effect only in September 2017 (Brasil 2015a) (Table 1). Its purpose is to promote the sustainability of aquatic animal production systems and ensure the health of raw materials from national farms. This instruction establishes guidelines on registration norms, good production practices, prophylactic measures, biosecurity, national and international transportation, quarantine criteria, and slaughter. The objectives of the Normative Instruction, as well as all its included aspects, are of utmost importance for improving the regulation of bullfrog farming. Although it has recently been implemented, significant efforts in promoting and disseminating this instruction will be crucial for its adoption and viability throughout the bullfrog farming production chain.

The challenges faced by bullfrog farming, whether due to gaps in legislation, lack of incentives, dissemination, investment, or organization of the production chain, have resulted in instability within the sector. This is reflected in the scarcity of data on production and

Table 1. Compilation of legislation and regulations pertinent to bullfrog farming at the federal level and in the states where we operate.

Identification of legal basis	Publication date	Most relevant information	Responsible authority	Scope
Lei nº 6.938/1981	August 31, 1981	Establishes the National Environmental Policy, its purposes, and mechanisms for formulation and implementation	Ministry of the Environment (MMA)	Federal
Lei nº 9.433/1997	January 8, 1997	Establishes the National Policy on Water Resources, creates the National System of Water Resources Management	Ministry of the Environment (MMA)	Federal
Lei de Crimes Ambientais nº 9.605/1998	February 12, 1998	Establishes criminal and administrative sanctions derived from conducts and activities harmful to the environment. Practices related to frog farming must be conducted in accordance with the guidelines established in this law	President of the Republic of Brazil	Federal
Portaria IBAMA nº 102/98	July 15, 1998	Regulates commercial production of exotic wildlife	Brazilian Institute of Environment and Renewable Natural Resources (IBAMA)	Federal
Lei Estadual nº 3.239/1999	August 2, 1999	Establishes the State Policy on Water Resources, creates the State System for Water Resources Management, and regulates the State Constitution	Governor of the State of Rio de Janeiro	State: RJ
Lei nº 11.959/2009	June 26, 2009	Establishes the National Policy on Sustainable Development of Aquaculture and Fisheries	Ministry of Fisheries and Aquaculture (MPA)	Federal
Resolução CONAMA nº 413/2009	June 26, 2009	Regulates the environmental licensing of aquaculture	National Environmental Council (CONAMA)	Federal
Resolução INEA nº 78/2013	October 4, 2013	Establishes procedures to be adopted in the environmental licensing of inland aquaculture ventures operating in the state of Rio de Janeiro	State Environment Institute (INEA)	State: RJ
Instrução Normativa nº 23/2014	September 11, 2014	Mandates the requirement of the Animal Transit Guide (GTA) to support the transportation of live aquatic animals and raw materials from aquatic animals originating from aquaculture establishments and destined for establishments registered with an official inspection agency	Ministry of Fisheries and Aquaculture (MPA)	Federal
Decreto nº 3831-R/2015	July 9, 2015	Regulates the environmental licensing of aquaculture in the state of Espírito Santo	State Governor's Office	State: ES
Instrução Normativa nº 4/2015	February 4, 2015	Establishes the National Program for Health of Cultured Aquatic Animals - "Aquaculture with Health"	Ministry of Fisheries and Aquaculture (MPA)	Federal
Portaria nº 20/2015	February 4, 2015	Designates laboratories as institutions qualified and authorized by the Ministry of Fisheries and Aquaculture to provide training on the collection and dispatch of official samples, aimed at certifying legally qualified professionals for the execution of sanitary defence activities related to aquatic animals	Ministry of Fisheries and Aquaculture (MPA)	Federal
Portaria nº 19/2015	February 4, 2015	Defines the list of mandatory notification diseases of aquatic animals to the Official Veterinary Service (SVO)	Ministry of Fisheries and Aquaculture (MPA)	Federal

Continue...

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Identification of legal basis	Publication date	Most relevant information	Responsible authority	Scope
Lei n° 13.123/2015	May 20, 2015	Regulates access to genetic heritage, protection and access to associated traditional knowledge, and benefit-sharing for the conservation and sustainable use of biodiversity	President of the Republic of Brazil	Federal
Decreto n° 8.772/2016	May 11, 2016	Regulates Law No. 13.123/2015	President of the Republic of Brazil	Federal
Decreto nº 62.243/2016	November 1, 2016	Provides rules and procedures for the environmental licensing of aquaculture in the state of São Paulo	Legislative Assembly of the State of São Paulo	State: SP
Nota técnica nº 9/2017/CAQ/DSA/CGSA/DSA	NA	Details the health status of amphibians in Brazil in WAHIS/OIE	Ministry of Agriculture and Livestock (MAPA)	Federal
Portaria SMC nº 103/2018	May 4, 2018	Approves a reference list of aquatic animal species that have been introduced into the national territory and used in agricultural activities	Ministry of Agriculture and Livestock (MAPA)	Federal
Resolução CONABIO nº 07/2018	May 29, 2018	Establishes the National Strategy for Invasive Exotic Species	Ministry of the Environment (MMA)	Federal
Portaria MMA nº 3/2018	August 16, 2018	Implementation Plan of the National Strategy for Invasive Exotic Species	Ministry of the Environment (MMA)	Federal
Instrução Normativa nº 2/2018	September 27, 2018	Provides for the risk analysis of the importation of aquatic organisms and their derivatives	Ministry of Fisheries and Aquaculture (MPA)	Federal
Resolução CONAMA nº 489/2018	October 26, 2018	Defines the use and management of captive wildlife and exotic fauna	Ministry of the Environment (MMA)	Federal
Norma nº 17/2020	January 16, 2020	Establishes procedures for the management, use, and production of bullfrogs in the state of Santa Catarina	Environment Institute (IMA)	State: SC
Resolução SEDEST nº 14/2020	March 5, 2020	Establishes norms and criteria for the environmental licensing of aquaculture enterprises and activities	State Secretariat for Sustainable Development (SEDEST)	State: PR

trade, as well as the difficulty in accessing this information. Thus, it is crucial to contribute to the improvement of bullfrog farming regulations in Brazil, not only to make the laws more applicable and comprehensive for rural producers but also, and most importantly, to facilitate access to information with the aim of conserving native amphibians (see Figure 1, which summarizes the relationship between these challenges, regulatory gaps, proposed improvements, and expected outcomes in Brazilian bullfrog farming).

2. Implications of policy decisions regarding the bullfrog

The bullfrog, introduced to Brazil in 1935 for commercial purposes, is now widely distributed across the country (Ferreira et al. 2002, Melo-Dias et al. 2023). Historically, the southern and southeastern regions of Brazil have had more invasive alien species, but in recent decades, an increase in the number of these species has been observed in the central-western and northern regions (Dechoum et al. 2024). Recognized as one of the most impactful invasive species on biodiversity, control and mitigation actions are essential, especially considering the Aichi Biodiversity Targets established internationally by the Convention on Biological Diversity (CBD).

The rapid and massive loss of global biodiversity demands a swift response from scientific research to guide effective policies. However, Brazilian political leaders have impeded this goal. In 2009, the Brazilian Congress proposed a law to "naturalize by decree" several non-native fish species to promote aquaculture development (Pelicice et al. 2014). Years later, the New Biodiversity Law came into effect (Law No. 13,123/2015; Decree No. 8,772/2016; Table 1). Although presented as a scientific advancement, this legislation imposes bureaucratic demands and severe penalties, significantly hindering biodiversity research in Brazil (Bockmann et al. 2018).

In the context of bullfrog farming, controversy heightened in 2018 with a MAPA ordinance proposing that introduced and established aquatic species in Brazil, including the bullfrog, be considered "native" (Ordinance SMC No. 103/2018, Brasil 2018; Table 1). Approval of this ordinance would permit the unrestricted trade and farming of these species across Brazil. Such a measure could intensify the introduction

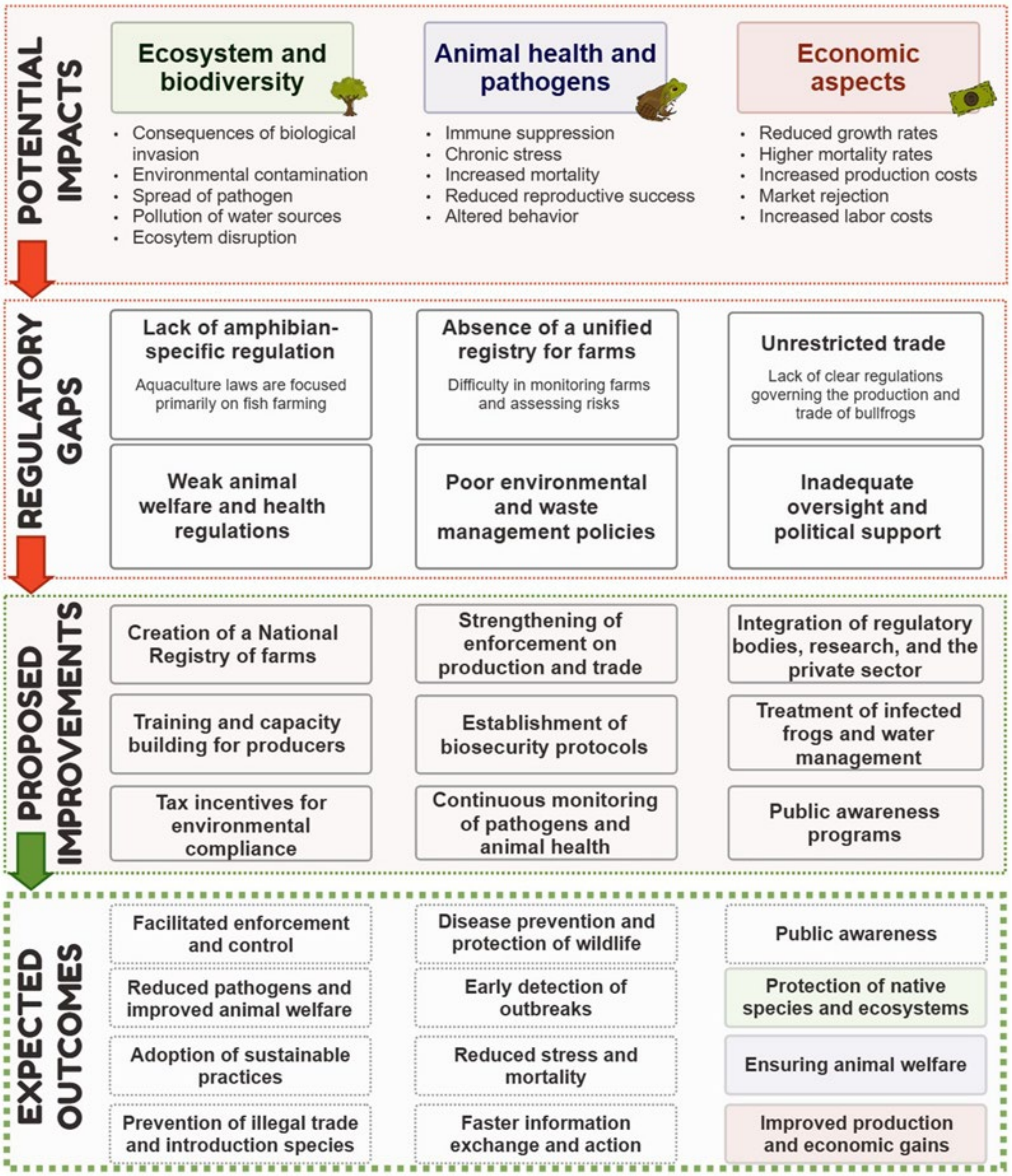

Figure 1. Schematic workflow representing the impacts, regulatory gaps, proposed improvements, and expected outcomes in bullfrog farming in Brazil. The workflow illustrates the overall relationships between components at each level without direct connections between individual elements, as the components operate in an integrated manner, making them difficult to isolate. The expected outcomes reflect the combined implementation of the proposed improvements and will address all identified impacts, whether environmental, related to animal welfare, or economic.

of new populations, leading to the loss of ecosystem services and traditional knowledge about native species (Speziale et al. 2012). Additionally, Brazil shares large river basins with other South American countries, potentially making it a significant source of non-native species for neighbouring nations (Brito et al. 2018).

This setback conflicts with several Aichi Biodiversity Targets, particularly the goal related to the prevention, control, or eradication of non-native species (Lima Junior et al. 2018). Given that Brazil hosts the world's most diverse aquatic fauna and the greatest diversity of amphibians (Padial et al. 2017, FAO 2024), it is crucial for authorities to adopt measures that value and conserve native biodiversity. Without such measures, the rate of introductions in Brazil is likely to exceed the research investigating their negative effects (Brito et al. 2018). To prevent devastating impacts, scientists must advocate for laws that facilitate international collaboration and encourage research, ensuring that biodiversity conservation is not hindered by bureaucratic barriers (Bockmann et al. 2018). Recognizing the issue as a cross-cutting public policy and developing sustained governance practices are fundamental goals for the management of invasive alien species in Brazil (Dechoum et al. 2024).

3. *Promoting engagement of sectors involved in bullfrog farming: initial strategies and discussions for policy formulation*

In December 2018, we initiated a series of meetings aimed at establishing partnerships to drive the objectives outlined in this study. A technical meeting hosted by the Aquatic Animal Coordination (CAQ) of MAPA brought together members from various divisions of MAPA, researchers active in bullfrog farming, amphibian diseases, and conservation, as well as representatives from relevant sectors. The goal was to discuss amphibian health and organize sanitary defence actions for bullfrog farming in Brazil. The discussion centered on the sector's progress, challenges, and the major diseases affecting amphibians, as well as strategies to strengthen public policies related to animal health.

In 2019, at the IX Brazilian Herpetology Congress, the leading event for amphibian and reptile studies in Brazil, we organized a workshop to advance the discussions on bullfrog farming regulation in the country, emphasizing the consolidation of proposals such as mapping bullfrog farms. With around 20 participants, the workshop featured presentations on key topics, including "Frog farms and the conservation of Brazilian amphibians", "Notification of diseases of amphibians on the OIE list", "Action plan for sanitary control in Brazilian frog farming", and "The role of amphibians in the sustainability of Aquaculture, polyculture, and the Horizon 2020 program". This meeting brought together researchers from universities, such as the State University of Campinas (UNICAMP), São Paulo State University (UNESP), University of São Paulo (USP), Federal Rural University of Pernambuco (UFRPE), and Federal University of Paraná (UFPR), as well as representatives from important regulatory bodies such as MAPA, the Ministry of the Environment (MMA), and the Agricultural Defence of São Paulo. This diverse group of stakeholders fostered productive debates aimed at advancing regulatory efforts.

During these discussions, we considered the feasibility of establishing an official registry of producers, initially proposed in São Paulo. Although crucial for amphibian conservation, implementing these proposals requires careful planning and cooperation between researchers, regulatory bodies, and producers.

Bullfrog farming in Brazil faces challenges due to gaps in public policies and a lack of integration between sectors. Key issues include the absence of accurate data on farm numbers, locations, and production, which are essential for understanding the activity's impact and for recommending improvements (Figure 1). In this context, our first step was to conduct a review of Brazilian farms, detailing their production and commercialization processes, and highlighting the necessary improvements to ensure the sustainability of the activity (see Ribeiro & Toledo 2022).

Considering the high loads of Bd present in both bullfrogs and wastewater, along with the lack of treatment for the fungus (Santos et al. 2020, Ribeiro et al. 2019), we emphasize the urgent need to develop biosecurity protocols for infrastructure and commercialization. Developing a best practices manual and control measures at all production stages will help raise awareness among producers and minimize the risk of pathogen spread, protecting both amphibians and ecosystems health.

According to Normative Instruction No. 04/2015, all aquatic animal breeding sites must register with the State Animal Health Defence Agency (OESA) or the State Agriculture Secretariat (Brasil 2015a). Brazil maintains a list of aquatic animal diseases that require mandatory notification, including chytridiomycosis and ranavirus, which must be reported to the Official Veterinary Service (SVO) for control measures (MPA Ordinance No. 19/2015) (Brasil 2015b).

Any citizen can notify suspected diseases through an initial investigation form. After notification, MAPA activates the SVO, responsible for epidemiological investigation, including data and sample collection, as established in Normative Instruction No. 04/2015 (Brasil 2015a). Sample analysis is conducted by laboratories accredited by MAPA. Although there is high demand for analyses, there are few accredited laboratories to perform them. To expedite the analysis process, a standardized manual for sample collection for diagnosing diseases in fish, shrimp, and mollusks was developed in 2013, but not for amphibians (MPA 2013). Only in 2022 was an instruction for sample collection for this group of animals published (MAPA 2022).

The World Organization for Animal Health (WOAH) conducts semi-annual evaluations of sanitary conditions, classified as lack of information, qualitative information, and quantitative information. The first official notification of amphibian diseases in Brazil occurred in 2018. Although Rv had been identified in bullfrog farms (Mazzoni et al. 2003), there were no records of this pathogen in wild amphibians. Regarding the Bd fungus, there were no official records of its presence in captive amphibians; however, studies indicated the presence of this pathogen in several bullfrog farms in Brazil (Santos et al. 2020, Ribeiro et al. 2019). Consequently, we notified the presence of the pathogen in farms, and this information was considered in the WOAH evaluation. Concerning wild amphibians, the presence of the fungus was officially documented across the national territory.

Recognizing that disease notifications are frequently limited to domestic animals, fostering collaboration among various institutions to identify threats and assess the risks associated with pathogen spread is vital. Strengthening epidemiological and zoosanitary surveillance necessitates cooperation across sectors, including the SVO, environmental agencies, research entities, and the private sector. A key element in this effort is establishing a communication channel for promptly reporting suspected or confirmed disease cases. To facilitate this integration, the "Fauna Network" was created as a communication platform for reporting wildlife disease cases, offering a comprehensive understanding of Brazil's current status regarding notifiable diseases (ICMBIO 2023). As representatives of the academic sector specializing in amphibian diseases, we contribute to this network, enhancing communication and ensuring transparency about the distribution of wildlife diseases. This initiative bolsters the surveillance and management of emerging infectious diseases in wildlife, enabling rapid information exchange among key stakeholders for effective monitoring and response efforts that protect public health, domestic animal health, and biodiversity. By adopting this interdisciplinary approach, the "Fauna Network" plays a role in addressing the risks posed by wildlife diseases and improving overall ecosystem health.

The future of bullfrog farming in Brazil hinges on maintaining the health and welfare of farmed animals. The National Health Program for Cultivated Aquatic Animals, known as "Aquaculture with Health" (Normative Instruction MPA No. 04/2015, updated by Normative Instruction MAPA No. 04/2019) (Brasil 2019), has been in place since

2017 and aims to promote sustainable production systems by preventing, controlling, and/or eradicating diseases. This program equips the SVO to respond quickly to disease outbreaks, certify establishments, and regulate quarantine services, ensuring a safer and more sustainable farming system.

Globally, however, amphibian farming and trade face substantial challenges related to invasion, overexploitation, biosecurity, and disease control. The frog leg trade has been characterized by unsustainable practices, resulting in severe consequences for amphibian populations and biodiversity (Auliya et al. 2023). International regulations remain inadequate, with around 98% of amphibian species not covered by CITES, largely due to the lack of a unique identifier in the World Customs Organization system (Auliya et al. 2016). This regulatory gap makes it difficult to monitor trade, track species, and enforce critical health and safety measures, facilitating biological invasions and the spread of pathogens. However, some progress has been made, such as the Bern Convention's recommendation to restrict trade in salamanders and newts in Europe to prevent the spread of *Batrachochytrium salamandrivorans* (Bsal), a pathogen responsible for severe declines in salamander populations (Auliya et al. 2016, Martel et al. 2013, Thomas et al. 2019).

Although the amphibian trade may appear small compared to other global markets, its impact on biodiversity loss is profound (Catenazzi et al. 2010). To address these challenges, stronger international regulations and enforcement are essential, highlighting the need for broader biosecurity measures and comprehensive disease surveillance. While collaborative efforts between organizations like WOAH and CITES are promising, further integration of wildlife disease management into trade regulations is crucial for safeguarding amphibian populations and the ecosystems they support.

In summary, discussions highlighted the impacts of bullfrog farming on native amphibians and ecosystems, emphasizing the need for a specific best practice manual and an action plan for amphibian health, as Brazil is one of the largest producers and exporters of frogs globally (Figure 1). By drawing on international examples, Brazil can collaborate to implement improved regulatory frameworks and proactive disease management strategies, ensuring the long-term sustainability of its bullfrog farming and trade industry. With established partnerships and a network of engaged stakeholders, the regulation of bullfrog farming in Brazil appears increasingly feasible.

4. *Importance of action plans in the development and implementation of public policies for bullfrog farming*

4.1. State level – territorial action plan

Action Plans, whether National (PAN) or Territorial (PAT), serve as strategic tools for public policy aimed at species conservation. These plans are collaboratively developed by representatives from academic, government, non-governmental organizations, and the productive sector, ensuring that priority measures are identified, and guidelines are established to improve the conservation status of target species. PATs focus on specific territories and operate on a state scale, with structured objectives and defined deliverables.

In the development of the *PAT Caminho das Tropas Paraná – São Paulo*, initiated in 2020, planning workshops and thematic meetings were held to promote integrated conservation efforts. This PAT covers 12,474,067 hectares across 163 municipalities in the states of Paraná and São Paulo (Figure 2). The primary objective of this PAT is to implement targeted actions that mitigate threats to species while promoting the integrated conservation of fauna and flora. Two key goals we focus on for amphibian conservation are: (i) Establishing priority areas for the prevention, detection, control, eradication, and best practices in production related to invasive species, such as bullfrogs, and (ii) Planning formative actions from a One Health perspective (Couto & Brandespim 2020).

Regarding the first objective, our focus is on bullfrog invasiveness, proposing actions to identify commercial bullfrog farms and assess impacts, aiming to prevent or control their potential invasion, while also developing a best practices manual for bullfrog farming. Simultaneously, we are engaged in developing a best practices manual for bullfrog farming. Concerning the second objective, focus is on the impacts of bullfrog farming on pathogen spread (*e.g.*, Bd and Rv), evaluating existing sanitary protocols and recommending adjustments for integration into state agency monitoring activities.

Mapping of bullfrog farms and their wild populations has been conducted across São Paulo, Paraná, and Santa Catarina, providing essential data for managing biological invasion. These findings, submitted to PAT coordination in March 2021 (Figure S1), are critical for developing measures to mitigate bullfrog-related impacts on native amphibians. Given the concerning prevalence rates of Bd and Rv in Brazilian farms and incidents of bullfrog escapes, we are steadfast in implementing proposed measures to mitigate negative impacts on native amphibian populations. We emphasize the importance of collaboration among government sectors, researchers, and producers to establish effective public policies regulating bullfrog farming practices in Brazil.

4.2. Federal level – national action plans

At the federal level, amphibian conservation efforts include our participation in two National Conservation Action Plans (PANs): the Southern Herpetofauna PAN and the Southeastern Herpetofauna PAN (Figure 2). Both PANs are in their second implementation cycle and aim to maintain the diversity of amphibian and reptile species in the south and southeastern Brazil, respectively.

In November 2020, we participated in a virtual technical meeting to update regulation number 17/2020 for bullfrog farming in Santa Catarina. Initially established on January 16th of the same year, the regulation aims to manage, use, and oversee bullfrog farming in the state (FATMA 2020). This update is part of the Southern Herpetofauna PAN's objectives, specifically to control bullfrog invasions and minimize pathogen spread. The primary goal is to enhance the regulation, ensuring it includes all necessary measures to reduce the threats posed by bullfrog farming to native populations. Additionally, it may serve as a replicable model for other Brazilian states, helping to reduce the negative impacts of this activity on biodiversity.

This initiative involves collaboration among representatives from government agencies, including the Santa Catarina Environment Institute (IMA) and the Chico Mendes Institute for Biodiversity Conservation (ICMBio), as along with researchers from public institutions, such as the Federal University of Santa Maria (UFSM), the UNICAMP, and the Federal University of Amazonas (UFAM). We highlighted aspects related to authorization and registration for the management and use of the species in captivity, which must adhere to specific criteria for activities such as commerce, acquisition, transportation, cultivation,

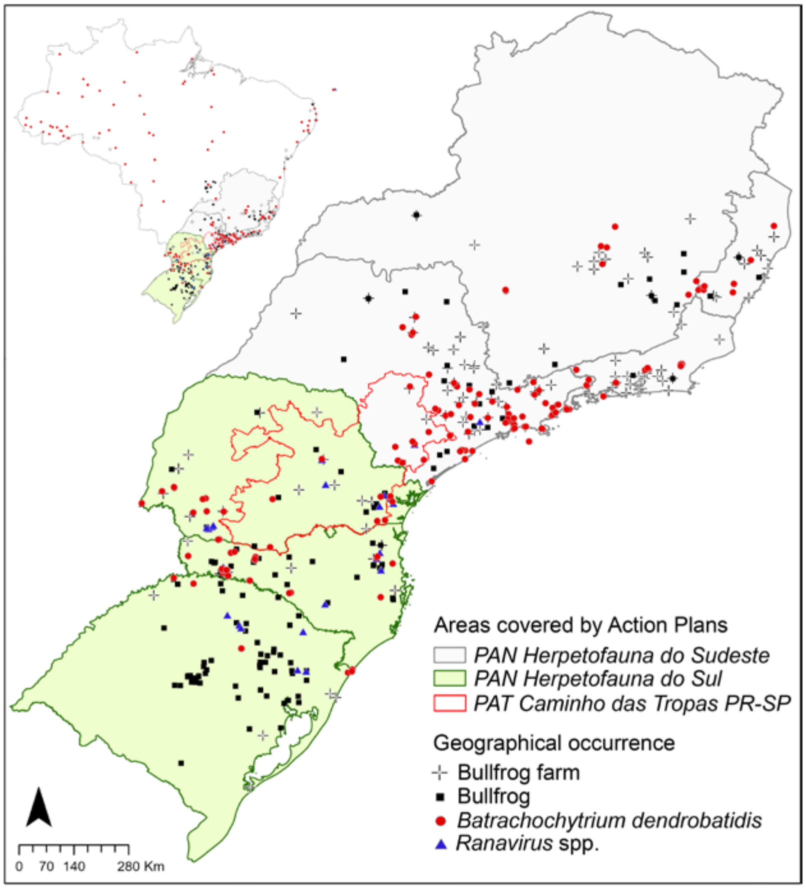

Figure 2. Areas covered by the National and Territorial Action Plans in which we are engaged in amphibian conservation actions, highlighting the occurrence points of major threats to amphibians.

distribution, and propagation. Key measures for controlling bullfrogs and their pathogens were outlined, including the implementation of screens to prevent escapes, the use of finer meshes in pipelines to contain tadpoles, and the adoption of double doors with automatic closures to restrict animal passage.

The need for the control and treatment of animals infected by Bd or Rv, as well as the proper treatment of effluents, was also addressed. The discussions included necessary actions to assess sanitary conditions in bullfrog farms, establish viable protocols, and identify responsible agencies for these measures. To date, we have contributed to the restructuring and review of the regulation, which is currently under approval by different sectors of the IMA before its final publication.

In March 2023, we participated in a workshop for the second cycle of the Southeastern Herpetofauna PAN at the Biodiversity Conservation Training Center (ACADEBio). This PAN encompasses the states of São Paulo, Rio de Janeiro, Minas Gerais, and Espírito Santo, focusing on improving the conservation of target species and their habitats. During the workshop, we expressed concerns about the impacts of bullfrog farming on native amphibians and proposed actions aligned with the PAN's strategies to mitigate these effects. Two specific objectives of the PAN are particularly relevant to this issue. First, the plan aims to strengthen public policies related to environmental regulation. Second, it focuses on controlling invasive species, such as bullfrogs, by identifying wild populations and defining priority areas for control. These objectives underscore the importance of aligning frog farming regulations with broader conservation policies to reduce the negative impacts on native species.

The emphasis on coordinating strategies aligned with broader conservation policies is critical for the regulation of bullfrog farming in Brazil and the conservation of native species and ecosystems. Concentrating efforts on defining specific objectives and developing actionable plans is essential for ensuring the continuity of biodiversity initiatives in these areas. Collaboration among various stakeholders and the adoption of integrated approaches are fundamental for the success of conservation policies, delineating a promising path towards the effective conservation of Brazilian herpetofauna.

Conclusion

Corroborating the principles of the Triple Helix (Zhou & Etzkowitz 2021), our experiences emphasize that collaboration among different sectors involved in regulating an activity is fundamental to the success of regulatory policies. Our direct engagement with public policies highlights that operating at smaller scales, such as the state level, can expedite and facilitate the implementation of actions compared to broader contexts like the federal level. Although our study focuses on the southern and southeastern regions of Brazil, areas most affected by bullfrog farming and the presence of Bd and Rv pathogens (Figure 2; see revised distribution data in Table S1), we suggest that similar challenges might occur in other regions, given the widespread production and trade of bullfrogs across the country (Figure 2; Ribeiro & Toledo 2022, Melo-Dias et al. 2023). Therefore, while we cannot conclusively state the consistency of problems and solutions across all Brazilian states based on the provided data, we believe the strategies proposed to address bullfrog invasion and pathogen spread could be applicable on a national scale. Given the broad distribution of bullfrogs and their associated risks, actions to mitigate these impacts should be considered throughout Brazil.

The sharing of ideas and discussions from different viewpoints is essential for improving the development and execution of species conservation actions. Therefore, we reinforce the crucial importance of cooperation among various public sectors, researchers, and producers to establish effective policies related to the regulation of bullfrog farming in Brazil. It is evident that there is a gap between governmental, academic, and productive sectors that needs to be bridged. Similarly, sharing our research results and disseminating scientific publications play a significant role in serving as a reference and guidance for species conservation initiatives. The regulation of bullfrog farming in Brazil and the conservation of amphibians are topics that demand attention and require a collaborative effort among the involved sectors to implement and maintain bullfrog farming practices sustainably, both economically and environmentally (Figure 1).

Supplementary Material

The following online material is available for this article:

Figure S1 – Geographic distribution of bullfrogs, bullfrog farms, and pathogens (Bd and Rv) in the PAT *Caminho das Tropas Paraná – São Paulo* territory and 50 Km buffer zone, with the PAT territory shown in white and Conservation Units highlighted in green. Brazilian states are listed as follows: SP = São Paulo, PR = Paraná, and SC = Santa Catarina.

Table S1 – Geographic distribution of amphibian threats in Brazil: bullfrogs, bullfrog farms, and Bd/Rv pathogens with coordinates and sources.

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Authors' Contribution

Luisa P. Ribeiro: Conceptualization; Data curation; Methodology; Writing – original draft; and Writing – review & editing.

Luís Felipe Toledo: Conceptualization; Funding acquisition; Supervision; and Writing – review & editing.

Conflicts of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

Ethics

This study did not involve human beings and/or clinical trials that should be approved by one Institutional Committee.

Data Availability

Metadata associated with this study can be found in the Supplemental File.

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Inventory

Floristics of the herbaceous-shrub vegetation in the restinga of Araoca Beach, municipality of Guimarães, Maranhão, Northeast Brazil

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Abstract: The Environmental Protection Area of Reentrâncias Maranhenses is located on the northern coast of the state of Maranhão, covering 16 municipalities and great floristic diversity. However, this biodiversity is threatened by the reduction and even extinction of species aggravated by anthropogenic action. The objectives of this study were (i) to analyze the floristic composition of the herbaceous and shrubby vegetation in the restinga ecosystem of Araoca Beach, located in the municipality of Guimarães; (ii) classify habits; (iii) to analyze pollination and seed dispersal syndromes; (iv) to categorize the physiognomies present and (v) to identify environmental impacts. Field expeditions were carried out between July 2022 to October 2023 (during the dry and rainy seasons) and plant species were recorded using the walking method. In total, 122 herbaceous and shrub species belonging to 95 genera and 50 families were cataloged. The families with the highest species richness were Fabaceae (24), Cyperaceae (10), Rubiaceae (6), and Asteraceae (5), corresponding to 37.2% of the total species collected. Herbaceous species (59), shrubs (29), sub-shrubs (21), lianas and vines (12) accounted for 48.4%, 23.8%, 17.2%, and 9.8% of the species, respectively. The survey also recorded a single species of palm (Arecaceae), corresponding to 0.8% of the species collected. As for pollination, 100 species were categorized exclusively as zoophilic and 15 as anemophilic. Concerning seed dispersal, anemochory accounted for 7.4% of the individuals (9); exclusive zoochory was present in 33.6% of the species (41); and exclusive autochory in 70 species (57.4% of the samples). Only *Pavonia cancellata* (L.) Cav. showed two types of dispersal (zoochory and autochory). The following physiognomies were described: unflooded open field, flooded open field-halophytic, unflooded closed shrubby vegetation, flooded closed shrubby vegetation, and flooded open shrubby vegetation. Araoca's plant diversity indicates a similarity to other restingas in Maranhão. The considerable amount of garbage left in the vicinity of the dunes, formation of trails, and cattle grazing areas alter the dynamics and compromise the balance of this ecosystem. Considering this scenario, the restinga of Araoca Beach is subject to loss of floristic composition, making it necessary to develop studies aimed at maintaining its diversity.
Keywords: *Conservation; environmental impacts; physiognomy; Reentrâncias Maranhenses; floristic similarity.*

Florística da vegetação herbáceo-arbustiva na restinga da Praia de Araoca, município de Guimarães, Maranhão, Nordeste do Brasil

Resumo: A Área de Proteção Ambiental das Reentrâncias Maranhenses está localizada no litoral norte do estado do Maranhão, abrangendo 16 municípios e apresentando grande diversidade florística. Contudo, essa biodiversidade está ameaçada pela redução e até extinção de espécies, situação agravada pela ação antrópica. Os objetivos deste

estudo foram (i) analisar a composição florística da vegetação herbáceo-arbustiva no ecossistema de restinga da praia de Araoca, localizada no município de Guimarães; (ii) classificar hábitos; (iii) analisar síndromes de polinização e dispersão de sementes; (iv) categorizar as fitofisionomias presentes e (v) identificar os possíveis impactos ambientais. As expedições de campo foram realizadas entre julho de 2022 e outubro de 2023 (durante as estações seca e chuvosa) e as espécies vegetais foram registradas usando o método do caminhamento. No total, foram catalogadas 122 espécies herbáceo-arbustivas pertencentes a 95 gêneros e 50 famílias. As famílias com maior riqueza de espécies foram Fabaceae (24), Cyperaceae (10), Rubiaceae (6) e Asteraceae (5), correspondendo a 37, 2% do total das espécies coletadas. Quanto ao hábito, as espécies herbáceas (59) correspondem a 48,4% da amostragem, as arbustivas (29) representaram 23,8%, as subarbustivas (21) equivalem a 17,2% e as lianas/trepadeiras somaram 9,8% das espécies. O levantamento ainda registrou uma única espécie de palmeira (Arecaceae), correspondendo a 0,8% das espécies coletadas. Quanto à polinização, 100 espécies foram categorizadas exclusivamente como zoofílicas e 15 relativas à anemofilia. Em relação à dispersão, a anemocoria representou 7,4% dos indivíduos (9); a zoocoria exclusiva estava presente em 33,6% do levantamento (41); e a autocoria exclusiva em 70 espécies, somando 57,4% das amostras. Apenas *Pavonia cancellata* (L.) Cav. apresentou dois tipos de dispersão, zoocoria e autocoria, que representam 0,8% do total das síndromes de dispersão. Foram descritas fisionomias de campo aberto não inundável, campo aberto inundável-halófilo, frutíceto fechado não inundável, frutíceto fechado inundável e frutíceto aberto inundável. A diversidade vegetal de Araoca indica similaridade de restingas segundo as espécies comumente encontradas nas áreas estudadas no Maranhão. A considerável quantidade de lixo deixado nas proximidades das dunas, formação de trilhas e pastagem de bovinos em áreas de estrato arbustivo, alteram a dinâmica e comprometem o equilíbrio desse ecossistema. Considerando esse cenário, a restinga da Praia de Araoca está sujeita a perda de composição florística, sendo necessário o desenvolvimento de estudos que direcionem a manutenção de sua diversidade.
Palavras-chave: *Conservação; impactos ambientais; fisionomias; Reentrâncias Maranhenses; similaridade florística.*

Introduction

The Reentrâncias Maranhenses Environmental Protection Area (EPA) covers 16 municipalities in the state of Maranhão and 2,680,911.2 ha. The main biomes are the Amazon forest (38.3%) and coastal and marine environments (61.7%), comprising mangroves, estuaries, rivers, dunes, restingas, and lagoons, which have a rich diversity of marine and terrestrial species (ICMBIO 2017, SEMA 2023). The EPA's coastline is quite jagged and plays a role as a natural nursery for various fishing species and a landing and feeding site for migratory shorebirds from the Northern Hemisphere. In addition, the EPA protects biodiversity and preserves the ecosystems of this region, where endangered species such as manatees, groupers, dolphins, and sea turtles are found (Hazin 2008).

The state of Maranhão has the second-longest coastline in Brazil (640 km) (El-Robrini et al. 2006) and is located on the eastern portion of the northern coast, with a rich coastline of Amazonian influence, including the restinga vegetation of the state west coast and the Reentrâncias Maranhenses (Almeida Jr. et al. 2018). The restinga ecosystem has great floristic diversity, known for harboring plant species from various phytogeographic domains, including Caatinga, Atlantic Forest, Cerrado, and even Amazonia, covering around 79% of the Brazilian coast (Lacerda et al. 1993, Rabelo et al. 2024). However, this biodiversity is under constant threat due to the progressive anthropogenic activities, leading to the reduction or extinction of species (Soares et al. 2021).

Restingas are not uniform along the Brazilian coast and have a high geomorphological variety that has favored the establishment of very diverse plant and animal communities (Lacerda et al. 1984). Distinct types of vegetation can be recognized and are referred to as "restinga physiognomies" (Galvão et al. 2018): herbaceous and sub-shrub vegetation (or beach and dune vegetation), shrub vegetation, tree vegetation, low forest, and high forest (Azevedo et al. 2014). Erect herbaceous plants, shrubs of different heights and trees, and trees predominate in fields, shrubby vegetation, and forests, respectively (Silva & Britez 2005, Lima et al. 2018).

Several floristic, ecological, and conservation studies on the restinga flora have been carried out along the Northeast coast of Brazil (Costa et al. 2018, Rodrigues et al. 2019), particularly in Maranhão. Among the authors who have expanded the records on species richness and composition in restingas in this state, one can highlight the works by Amorim et al. (2016) in the dunes of Araçagi Beach, between the municipalities of São José de Ribamar and Paço do Lumiar; Almeida Jr. et al. (2018) in the Reentrâncias Maranhenses EPA; Serra et al. (2016) in the Merck restinga, in São José de Ribamar; Silva et al. (2016) in the São Marcos dunes, in São Luís; Lima & Almeida Jr. (2018) in the Panaquatira restinga, in São José de Ribamar; and Correia et al. (2020) in Alcântara. To maintain these studies over time is necessary to expand knowledge about the plant richness of the restinga ecosystem.

Despite legal conservation initiatives that have transformed some stretches of restinga into conservation units, there is still a lack of information on important ecological aspects of this ecosystem (Azevedo et al. 2014). For example, the relationships between plants and their respective pollinators or dispersers are very relevant for structuring communities and are also essential in the natural distribution of species and in the exchange of genetic material within and outside populations (Reis et al. 2012, Kuhlmann & Ribeiro 2016).

Knowledge about the relationships between organisms and the environment and the interactions between species allows for a more qualified contribution to the definition of new conservation areas and better conservation strategies, as well as providing data for the recovery of areas that have been intensely degraded (Azevedo et al. 2014). In this context, studies on floristic composition and vegetation structure are crucial for understanding their structure and dynamics, which are basic parameters for the management and restoration of various plant communities.

This study aimed to analyze and inventory the floristic composition of the herbaceous and shrubby vegetation in the restinga of Araoca Beach in the municipality of Guimarães, in the Reentrâncias Maranhenses EPA. We categorized the physiognomies and identified the environmental impacts in this area, contributing to conservation strategies for the restinga ecosystem.

Material and Methods

1. Study area

The study was conducted in the restinga of Araoca Beach (2° 03' 03" S; 44° 29' 51" W) located in the municipality of Guimarães, part of the Reentrâncias Maranhenses EPA (ICMBIO 2023), western coast of Maranhão (Figure 1). The relief is characterized by coastal plains shaped by marine and fluvial agents and processes that give rise to beaches, mangroves, swamps, apicum (i.e., hypersaline tidal flats), lagoons, and cliffs (Ramos 2008). Coastal tablelands with higher topographic levels than those of the Maranhão lowlands (generally not exceeding 100 m in altitude) occur in this region. The regional climate is Tropical Aw, according to the Köppen classification (Alvares et al. 2013). The average annual temperature is 27ºC and the average annual rainfall is 2055 mm (Maranhão 2019). The rainy season is between November and June and the dry season is between July and October (Alvares et al. 2013).

2. Data collection

The field expeditions were conducted biannually: in July and September 2022, and in May and October 2023, covering both the dry and rainy seasons. Each expedition lasted four days and included morning and afternoon shifts (from 8:00 AM to 5:00 PM), with the participation of three researchers.

The species in the restinga areas were recorded using the free-walking method (Filgueiras et al. 1994), which consists of traversing the entire study area, identifying, collecting, and recording plant species on field datasheets. Fertile individuals were collected, pressed, and dried in an oven at 60 ºC for 72 h following the herborization techniques recommended by Fidalgo & Bononi (1984). The specimens were deposited in the Rosa Mochel Herbarium (SLUI) at the State University of Maranhão.

Species were identified according to the dichotomous keys found in specialized literature, e.g. Souza and Lorenzi (2019) and Flora e Funga do Brasil (2023), and comparison with specimens from the SLUI herbarium and virtual herbaria from the SpeciesLink and Flora e Funga do Brasil databases. In addition, taxonomists experts in Plantaginaceae, Fabaceae and Convolvulaceae were consulted. The photos were taken using a Nikon D5300 digital camera to record the vegetative and reproductive parts of the specimens in the field. Herbarium acronyms are according to Thiers (2023). The names of the authors of the species follow Brummitt & Powell (1992). The floristic list was compiled

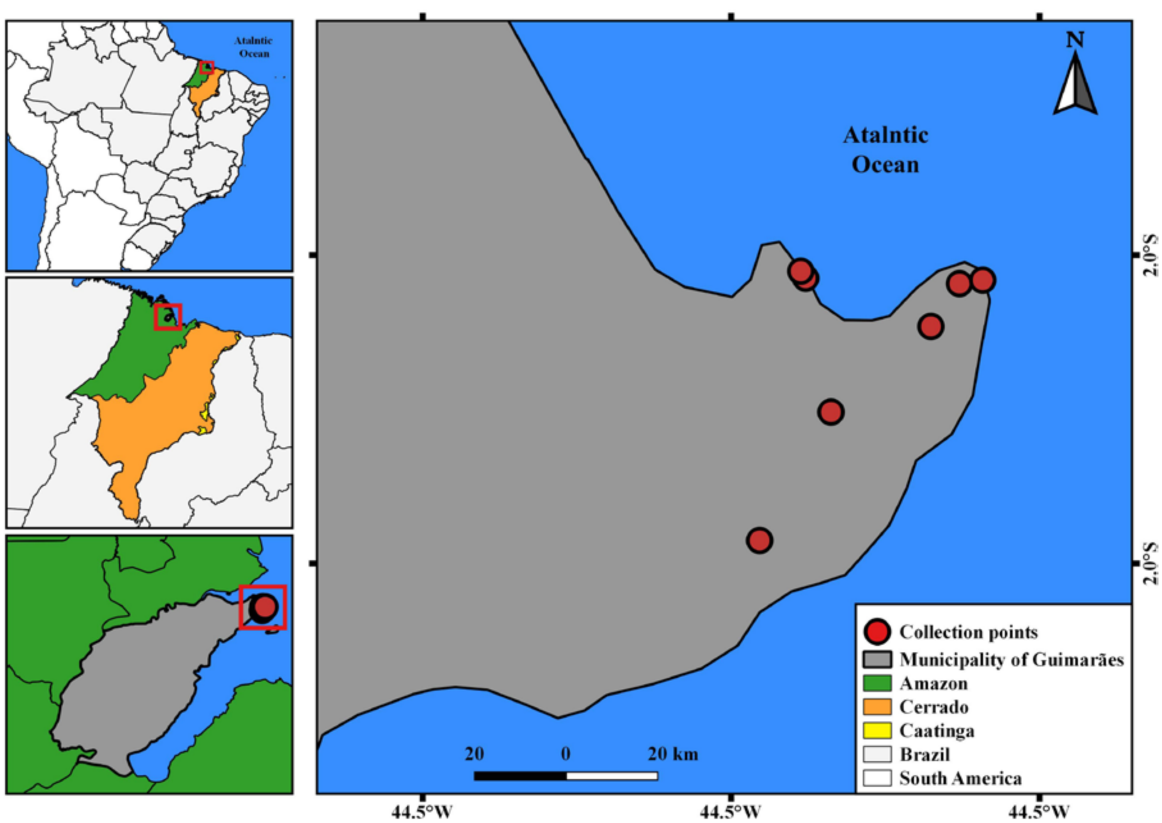

Figure 1. Map of the restinga of Araoca Beach. Red dots indicating collection locations.

according to the classification system based on APG IV (2016) and the spelling of species names and authors was confirmed by consulting the Missouri Botanical Garden database ("Tropical System", tropicos.org) and the REFLORA database (Flora e Funga do Brasil 2023).

Species were classified according to habit (Souza et al. 2013) and life form (Raunkiaer 1984). Pollination syndromes were based on Faegri & van der Pijl (1979) and van der Pijl (1972) and species were grouped into wind-pollinated species (anemophilous) and animal-pollinated species (zoophilous). The types of fruit dispersal were associated with the morphological characteristics of the diaspores of the genera described for the restinga ecosystem, which are included in the Flora e Funga do Brasil database. Species that disperse their diaspores by wind were grouped as anemochoric; species that disperse their diaspores by animals were grouped as zoochoric; and species that disperse by the plant itself were grouped as autochoric (Laurentino et al. 2016, Diniz et al. 2021).

For the classification of physiognomies, we followed the methods proposed by Silva & Britez (2005) and Nascimento-Junior (2012), considering the descriptions in Lima et al. (2017). We only considered physiognomies related to the herbaceous-shrub vegetation of the restinga, which include: 1) Field formations (characterized by herbaceous species, which can be erect, cespitose, reptant, and/or rhizomatous); and 2) Shrubby vegetation (characterized by shrub and sub-shrub species).

3. Similarity analysis

Based on the floristic list obtained in this study, a similarity analysis was conducted by comparing it with studies from restinga areas in the state of Maranhão. The following studies were selected: Lima & Almeida Jr. (2018) on Panaquatira Beach, São José de Ribamar; Correia et al. (2020) on Itatinga Beach, Alcântara; and Guterres et al. (2020) on Guia Beach, São Luís Island (Table 1). Only taxa identified to the species level were considered, while species with uncertain identifications (e.g., those marked as "cf.") and those not belonging to the stratum addressed in this study (herbaceous-shrub) were excluded from the analysis. We classified plants as herbaceous or shrubby based on their exhibited life forms during the data collection.

Subsequently, a Detrended Correspondence Analysis (DCA) was used to verify the floristic relationships of herbaceous-shrub species with the restinga areas was conducted using the Vegan package (Oksanen et al. 2023) in R software, version 4.4.2 (R Development Core Team 2024). The analysis was based on a binary matrix composed by 278 species, allowing for the visualization of similarity patterns and floristic relationships among the compared areas. The results are presented in a scatter plot, where the proximity of points indicates the floristic similarity between the analyzed areas.

Results and Discussion

We recorded 122 herbaceous and shrub species, belonging to 95 genera and 50 families, distributed in different formations and vegetations associated with the restinga of Araoca Beach (Table 2). The families with the highest species richness were Fabaceae (24), Cyperaceae (10), Rubiaceae (6), Asteraceae (5), Myrtaceae (5), Poaceae (5), Eriocaulaceae (4), Lamiaceae (4), and Plantaginaceae (4). These nine families correspond to 18% of the families sampled and accounted for 54.9% of the total species collected. The families with the highest richness were also mentioned in studies carried out on Panaquatira Beach (Lima & Almeida Jr. 2018).

Herbaceous species (59) accounted for 48.4% of the samples, with Cyperaceae (10), Poaceae (5), Eriocaulaceae (4), and Plantaginaceae (4) standing out as the most representative families for this life form. Shrubs (29) accounted for 23.8% of the plants collected and the families with the highest shrub species richness were Myrtaceae (5), Rubiaceae (5), Fabaceae (3), and Combretaceae (2). In the sub-shrub component, 21 individuals (17.2% of the species identified) were collected from seven families: Fabaceae (12), Lamiaceae (3), Turneraceae (2), Amaranthaceae (1), Euphorbiaceae (1), Malvaceae (1), and Plumbaginaceae (1).

Lianas (4) and vines (8) accounted for 9.8% of the species sampled, grouped into seven families. The families having vines were Fabaceae (3), Passifloraceae (2), Lygodiaceae (1), Menispermaceae (1), and Vitaceae (1). The Fabaceae family also had liana species (2), followed by Araceae (1) and Euphorbiaceae (1). The survey also recorded a single palm species, *Desmoncus polyacanthos* Mart. (Arecaceae) corresponding to 0.8% of the species collected in Araoca. In the rainy season, we observed the highest number of herbaceous and shrub species blooming, while a marked decrease in blooming in the dry season (July and August).

Fabaceae were highly represented in this study (19.7% of the total species – Figure 2) as in other restinga areas in the state of Maranhão. Lima & Almeida Jr (2018) cataloged 31 species (16.3%) of Fabaceae in the Panaquatira restinga, in the municipality of São José de Ribamar, and Guterres et al. (2020) recorded 16 species of the family (20.9%) in the restinga in Guia Beach, in São Luís. In addition, Fabaceae was also predominant in the restinga of Itatinga (26 species – 17.5%), in the municipality of Alcântara (Correia et al. 2020).

Species of the Fabaceae family have a remarkable diversity of habits. Fabaceae is the family of Brazilian flora with the greatest species richness and one of the largest in the world (Lewis et al. 2005, Flora e Funga do Brasil 2023). In terms of species number, Fabaceae stands out as one of the three richest families in Brazil with approximately 3038 species (Flora e Funga do Brasil 2023).

The species *Borreria verticillata* (L.) G.Mey., *Chamaecrista diphylla* (L.) Greene, *Crotalaria retusa* L., *Turnera melochioides* Cambess., *Utricularia simulans* Pilg., were the ones with the highest number of individuals collected and were found within the entire study area. *Anacardium occidentale* L., *Byrsonima crassifolia* (L.) Kunth, *Cenostigma bracteosum* (Tul.) Gagnon & G.P.Lewis, *Chrysobalanus icaco* L, *Myrcia multiflora* (Lam.) DC., and *Myrcia splendens* (Sw.) DC. were the most common woody species.

Considering floristic studies carried out in restingas in Maranhão (Araujo et al. 2016, Lima et al. 2017, Lima & Almeida Jr 2018, Guterres et al. 2020), the families listed in our survey are the most representative in the restinga ecosystem of the state. This is due to their different habits, pollination, and dispersal strategies (Lima & Almeida Jr 2018). Pollination in the restinga is an important ecosystem service. *Byrsonima crassifolia* and *Anacardium occidentale* stand out for having flowers attractive to pollinators and contribute to less attractive plants also being potentially pollinated in their proximity (Paiva & Almeida Jr 2020).

Among the pollination syndromes, of the 122 species recorded in Araoca 100 were exclusively zoophilic and 15 were classified as anemophilous. Four species belonging to the Eriocaulaceae family had both pollination types. For zoophilic plant species, which may also have self-pollination mechanisms, only *Crotalaria retusa* (Fabaceae) stood out

Table 1. Comparative table based on species used in similarity analyses in restinga areas in the State of Maranhão, Brazil.

Site	City/State	Acronym	Number of species used	Reference
Araoca	Guimarães/Maranhão	araoca	122	Present study
Panaquatira	São José de Ribamar/Maranhão	sjriba	49	(Lima & Almeida Jr 2018)
Praia da Guia	São Luís/Maranhão	sluis	45	(Guterres et al 2020)
Itatinga	Alcântara/Maranhão	alca	54	(Correia et al. 2020)

Table 2. List of herbaceous-shrub species recorded in the floristic survey with indication of origin, habit, and pollination and dispersal syndromes, in the restinga ecosystem of Araoca Beach, Guimarães, MA. Origin: na: native; en: naturalized species. Habit: ab: shrub; ev: herbaceous; sb: subshrub; li: liana; tr: climbing plant. Pollination: zo: zoophilic; an: anemophilic; au: self-pollination. Dispersal: hi: hydrochory; zo: zoochory; au: autochory; an: anemochory. SLUI = Herbário Rosa Mochel, State University of Maranhão, São Luís.

FAMILY/Species	Origin	Habit	Pollination	Dispersal	Herbarium/Vouchers
ACANTHACEAE					
Avicennia schaueriana Stapf & Leechm. ex Moldenke	na	ab	zo	hi	SLUI 7756
Dicliptera ciliaris Juss.	na	ev	zo	au	SLUI 7835
AIZOACEAE					
Sesuvium portulacastrum (L.) L.	en	ev	zo	au	SLUI 7865
AMARANTHACEAE					
Alternanthera brasiliana (L.) Kuntze	na	sb	zo	au	SLUI 7833
Alternanthera tenella Colla	na	ev	zo	au	SLUI 7132
Blutaparon portulacoides (A.St.-Hil.) Mears	na	ev	zo	au	SLUI 6641
ANACARDIACEAE					
Anacardium occidentale L.	na	ab	zo	zo	SLUI 7831
ARACEAE					
Philodendron acutatum Schott	na	li	zo	zo	SLUI 6664
ARECACEAE					
Desmoncus polyacanthos Mart.	na	pa	zo	zo	SLUI 7509
ASTERACEAE					
Baltimora geminata (Brandegee) Stuessy	na	ev	zo	au	SLUI 7503
Elephantopus mollis Kunth	na	ev	zo	an	SLUI 7128
Emilia sonchifolia (L.) DC.	en	ev	zo	an	SLUI 7885
Sphagneticola trilobata (L.) Pruski	na	ev	zo	an	SLUI 7131
Tilesia baccata (L.) Pruski	en	ab	zo	zo	SLUI 7508
BATACEAE					
Batis maritima L.	na	ev	zo	au	SLUI 7507
HELIOTROPIACEAE					
Euploca polyphylla (Lehm.) J.I.M.Melo & Semir	na	ev	zo	au	SLUI 7830
BURMANNIACEAE					
Burmannia capitata (Walter ex J.F.Gmel.) Mart.	na	ev	zo	au	SLUI 7848
CACTACEAE					
Cereus jamacaru DC.	na	ab	zo	zo	SLUI 7873
CHRYSOBALANACEAE					
Chrysobalanus icaco L.	na	ab	zo	zo	SLUI 6649
COMBRETACEAE					
Conocarpus erectus L.	na	ab	zo	zo	SLUI 7828
Laguncularia racemosa (L.) C.F.Gaertn.	na	ab	zo	zo	SLUI 7859

FAMILY/Species	Origin	Habit	Pollination	Dispersal	Herbarium/Vouchers
COMMELINACEAE					
Murdannia nudiflora (L.) Brenan	en	ev	zo	au	SLUI 7890
CONVOLVULACEAE					
Ipomoea imperati (Vahl) Griseb.	na	ev	zo	au	SLUI 7864
CYPERACEAE					
Bulbostylis truncata (Nees) M.T.Strong	na	ev	an	au	SLUI 7866
Cyperus ligularis L.	na	ev	an	au	SLUI 7895
Cyperus obtusatus (J.Presl & C.Presl) Mattf. & Kük.	na	ev	an	au	SLUI 7867
Cyperus polystachyos Rottb.	na	ev	an	au	SLUI 7749
Cyperus sellowianus (Kunth) T.Koyama	na	ev	an	au	SLUI 7861
Eleocharis geniculata (L.) Roem. & Schult.	na	ev	an	au	SLUI 7898
Eleocharis interstincta (Vahl) Roem. & Schult.	na	ev	an	au	SLUI 7877
Fimbristylis dichotoma (L.) Vahl	na	ev	an	au	SLUI 7500
Fimbristylis cymosa R.Br.	na	ev	an	au	SLUI 6661
Scleria microcarpa Nees ex Kunth	na	ev	an	au	SLUI 7853
ERIOCAULACEAE					
Eriocaulon cinereum R.Br.	na	ev	an/zo	au	SLUI 7111
Paepalanthus subtilis Miq.	na	ev	an/zo	an	SLUI 7888
Syngonanthus gracilis (Bong.) Ruhland	na	ev	an/zo	an	SLUI 7122
Tonina fluviatilis Aubl.	na	ev	an/zo	au	SLUI 8751
EUPHORBIACEAE					
Cnidoscolus urens (L.) Arthur	na	sb	zo	zo	SLUI 7829
Dalechampia scandens L.	na	li	zo	au	SLUI 7876
Euphorbia hyssopifolia L.	na	ev	zo	au	SLUI 7098
FABACEAE					
Abrus precatorius L.	na	li	zo	au	SLUI 7471
Aeschynomene evenia C.Wright & Sauvalle	na	ev	zo	zo	SLUI 7855
Cenostigma bracteosum (Tul.) Gagnon & G.P.Lewis	na	ab	zo	au	SLUI 7102
Centrosema brasilianum (L.) Benth.	na	tr	zo	au	SLUI 7090
Chamaecrista desvauxii (Collad.) Killip	na	sb	zo	au	SLUI 7099
Chamaecrista diphylla (L.) Greene	na	sb	zo	au	SLUI 7750
Chamaecrista nictitans (L.) Moench	na	sb	zo	au	SLUI 6654
Chamaecrista tenuisepala (Benth.) H.S.Irwin & Barneby	na	sb	zo	au	SLUI 6667
Clitoria falcata Lam.	na	tr	zo	au	SLUI 7091
Clitoria simplicifolia (Kunth) Benth.	na	sb	zo	au	SLUI 7130
Crotalaria retusa L.	en	sb	zo/au	au	SLUI 7834
Desmodium barbatum (L.) Benth.	na	sb	zo	zo	SLUI 7860
Desmodium incanum (Sw.) DC.	en	sb	zo	zo	SLUI 7112
Entada polystachya (L.) DC.	na	ab	zo	au	SLUI 7759
Galactia striata (Jacq.) Urb.	na	tr	zo	au	SLUI 7872
Indigofera hirsuta L.	na	ev	zo	au	SLUI 7879
Macroptilium lathyroides (L.) Urb.	na	ev	zo	au	SLUI 7092
Macropsychanthus grandiflorus (Mart. ex Benth.) L.P.Queiroz & Snak	na	li	zo	au	SLUI 7840
Mimosa candollei R.Grether	na	ev	zo	au	SLUI 7826

FAMILY/Species	Origin	Habit	Pollination	Dispersal	Herbarium/Vouchers
Stylosanthes angustifolia Vogel	na	sb	zo	zo	SLUI 7117
Zornia brasiliensis Vogel	na	sb	zo	zo	SLUI 7843
Zornia guanipensis Pittier	na	sb	zo	zo	SLUI 7498
Zornia latifolia Sm.	na	sb	zo	zo	SLUI 7093
GENTIANACEAE					
Schultesia guianensis (Aubl.) Malme	na	ev	zo	au	SLUI 7120
HELICONIACEAE					
Heliconia psittacorum L.f.	na	ev	zo	zo	SLUI 7858
IRIDACEAE					
Cipura paludosa Aubl.	na	ev	zo	au	SLUI 7849
LAMIACEAE					
Hyptis atrorubens Poit.	na	ev	zo	au	SLUI 7838
Hyptis crenata Pohl ex Benth	na	sb	zo	au	
Marsypianthes chamaedrys (Vahl) Kuntze	na	sb	zo	au	SLUI 7891
Mesosphaerum suaveolens (L.) Kuntze	na	sb	zo	au	SLUI 7856
LECYTHIDACEAE					
Gustavia augusta L	na	ab	zo	zo	SLUI 7889
LENTIBULARIACEAE					
Utricularia fimbriata Kunth	na	ev	zo	au	SLUI 7850
Utricularia simulans Pilg.	na	ev	zo	au	SLUI 7757
Utricularia subulata L.	na	ev	zo	au	SLUI 7854
LINDERNIACEAE					
Lindernia crustacea (L.) F.Muell.	na	ev	zo	au	SLUI 7845
LYGODIACEAE					
Lygodium venustum Sw.	na	tr		au	SLUI 7753
MALPIGHIACEAE					
Byrsonima crassifolia (L.) Kunth	na	ab	zo	zo	SLUI 7496
MALVACEAE					
Pavonia cancellata (L.) Cav.	na	ev	zo	zo/au	SLUI 7474
Sida ciliaris L.	na	sb	zo	zo	SLUI 7748
MELASTOMATACEAE					
Comolia villosa (Aubl.) Triana	na	ab	zo	au	SLUI 7127
Noterophila bivalvis (Aubl.) Kriebel & M.J.R.Rocha	na	ev	zo	au	SLUI 7103
MENISPERMACEAE					
Odontocarya duckei Barneby	na	tr	zo	zo	SLUI 7505
MYRTACEAE					
Eugenia flavescens DC.	na	ab	zo	zo	SLUI 6658
Eugenia punicifolia (Kunth) DC.	na	ab	zo	zo	SLUI 7096
Myrcia guianensis (Aubl.) DC.	na	ab	zo	zo	SLUI 7104
Myrcia multiflora (Lam.) DC.	na	ab	zo	zo	SLUI 7104
Myrcia splendes (Sw.) DC.	na	ab	zo	zo	SLUI 7825
OCHNACEAE					
Ouratea hexasperma	na	ab	zo	zo	SLUI 7874
ONAGRACEAE					

FAMILY/Species	Origin	Habit	Pollination	Dispersal	Herbarium/Vouchers
Ludwigia hyssopifolia (G.Don) Exell	na	ev	zo	au	SLUI 784
Ludwigia leptocarpa (Nutt.) H.Hara	na	ab	zo	au	SLUI 7842
PASSIFLORACAE					
Passiflora subrotunda Mast.	na	tr	zo	zo	SLUI 7897
Passiflora foetida L.	na	tr	zo	zo	SLUI 7868
PLANTAGINACEAE					
Bacopa angulata (Benth.) Edwall	na	ev	zo	au	SLUI 7758
Bacopa aquatica Aubl.	na	ev	zo	au	SLUI 7862
Bacopa salzmannii (Benth.) Wettst. ex Edwall	na	ev	zo	au	SLUI 7094
Bacopa sessiliflora (Benth.) Edwall	na	ev	zo	au	SLUI 7841
PLUMBAGINACEAE					
Plumbago scandens L.	na	sb	zo	zo	SLUI 7881
POACEAE					
Axonopus capillaris (Lam.) Chase	na	ev	an	an	SLUI 7824
Dactyloctenium aegyptium (L.) Willd.	en	ev	an	an	SLUI 7088
Eragrostis maypurensis (Kunth) Steud.	na	ev	an	au	SLUI 7752
Spartina alterniflora Loisel.	na	ev	an	an	SLUI 7896
Sporobolus virginicus (L.) Kunth	na	ev	an	an	SLUI 7511
POLYGALACEAE					
Senega glochidata	na	ev	zo	au	SLUI 785
POLYGONACEAE					
Coccoloba latifolia Lam.	na	ab	zo	zo	SLUI 7878
PTERIDACEAE					
Ceratopteris pteridoides (Hook.) Hieron.	na	ev		au	SLUI 7871
RHIZOPHORACEAE					
Rhizophora mangle L.	na	ab	zo	zo	SLUI 7875
RUBIACEAE					
Borreria verticillata (L.) G.Mey.	na	ev	zo	au	SLUI 7836
Chomelia obtusa Cham. & Schltdl.	na	ab	zo	zo	SLUI 7472
Chomelia ribesioides Benth. ex A.Gray	na	ab	zo	au	SLUI 7863
Cordiera sessilis (Vell.) Kuntze	na	ab	zo	zo	SLUI 7478
Isertia hypoleuca Benth.	na	ab	zo	zo	SLUI 7106
Mitracarpus strigosus (Thunb.) P.L.R.Moraes, De Smedt & Hjertson	na	ev	zo	au	SLUI 7097
Tocoyena sellowiana (Cham. & Schltdl.) K.Schum.	na	ab	zo	zo	SLUI 7882
SAPINDACEAE					
Pseudima frutescens (Aubl.) Radlk.	na	ab	zo	au	SLUI 7893
SAPOTACEAE					
Manilkara bidentata (A.DC.) A.Chev.	na	ab	zo	zo	SLUI 6651
SOLANACEAE					
Solanum stramoniifolium Jacq.	na	ab	zo	zo	SLUI 7087
TURNERACEAE					
Turnera melochioides Cambess.	na	sb	zo	zo	SLUI 7754
Turnera subulata Sm.	na	sb	zo	zo	

FAMILY/Species	Origin	Habit	Pollination	Dispersal	Herbarium/Vouchers
VIOLACEAE					
Pombalia calceolaria (L.) Paula-Souza	na	ev	zo	au	SLUI 7126
VITACEAE					
Cissus erosa Rich.	na	tr	zo	zo	SLUI 7894
XYRIDACEAE					
Xyris macrocephala Vahl	na	ev	zo	au	SLUI 7501
Xyris savanensis Miq.	na	ev	zo	au	SLUI 7847

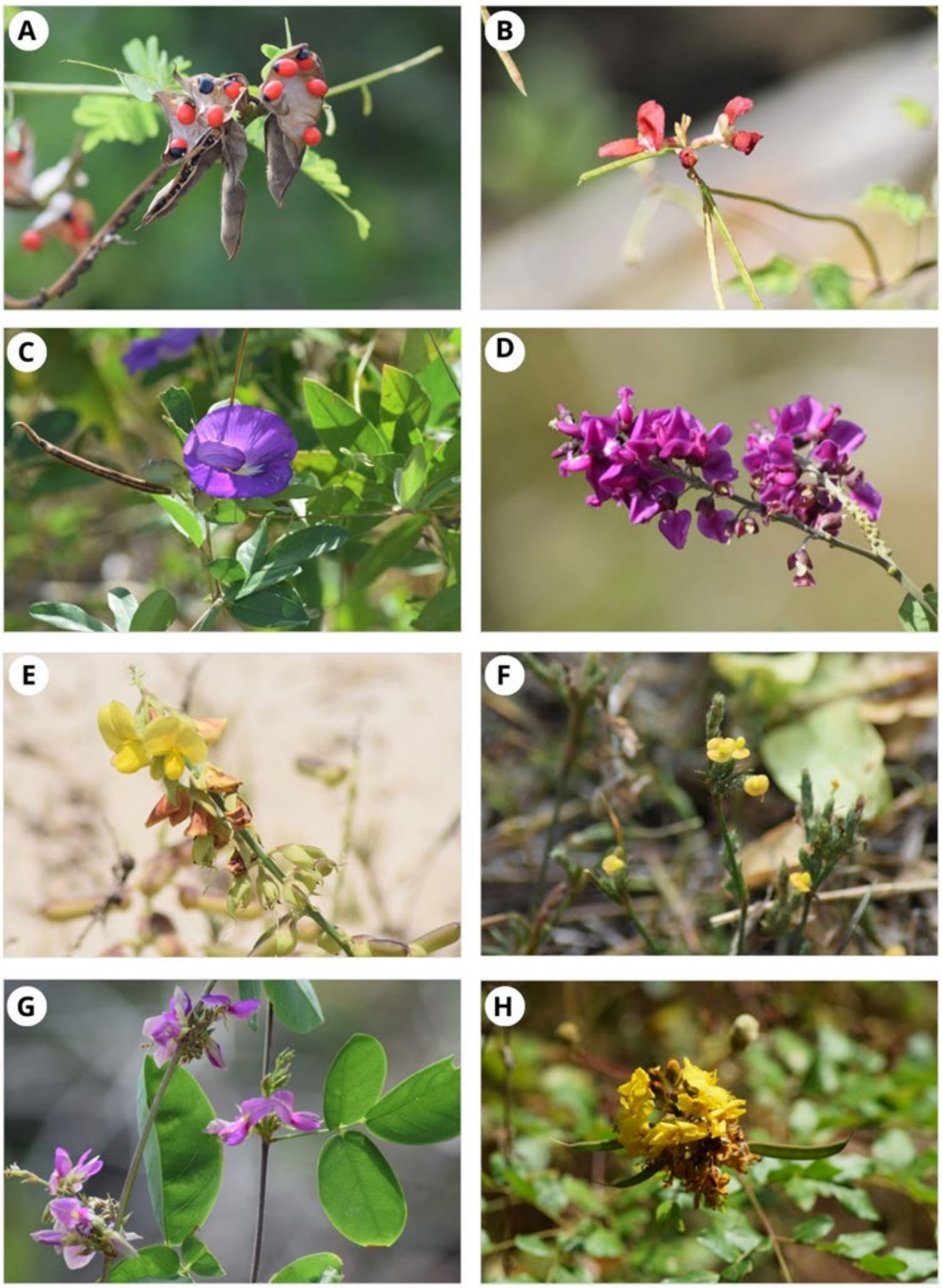

Figure 2. Some listed species of the herbaceous-shrubby vegetation of the restinga of Araoca Beach, Guimarães, MA. Fabaceae. a) *Abrus precatorius*. b) *Macroptilium lathyroides*. c) *Centrosema brasilianum*. d) *Macropsychanthus grandiflorus*. e) *Crotalaria retusa*. f) *Stylosanthes angustifolia*. g) *Galactia striata*. h) *Cenostigma bracteosum*.

in this survey. We collected two species of fern belonging to the families Lygodiaceae (*Lygodium venustum* Sw.) and Pteridaceae (*Ceratopteris pteridoides* (Hook.) Hieron.). As they belong to the group of cryptogams, they do not fit into the pollination syndromes for angiosperms.

We identified four types of dispersal syndromes: hydrocoria, considered a secondary form of dispersal, in one of the species collected (*Avicennia schaueriana* Stapf & Leechm. ex Moldenke); anemochorous in 7.4% of the individuals (9); exclusive zoochory in 33.6% of the individuals (41); and exclusive autochorous in 57.4% of the samples (70). Only *Pavonia cancellata* (L.) Cav. showed two types of dispersal, zoochory and autochorous, representing 0.8% of the total. In a study on fruit and seed dispersal in the dunes of São Marcos Beach, Pires et al. (2021) highlighted zoochory in 62.5% of the species as the main dispersal syndrome, followed by anemochorous in 26.78% and autochorous in 5.36%.

The Araoca restinga has a considerable number of native species, which account for 94.3% of the samples in this study. Fewer naturalized species were found, such as *Emilia sonchifolia* (L.) DC and *Tilesia baccata* (L.) Pruski in the dunes, *Sesuvium portulacastrum* (L.) L. and *Murdannia nudiflora* (L.) Brenan in open field; *Crotalaria retusa* in sub-shrub physiognomy, *Desmodium incanum* (Sw.) DC. and *Dactyloctenium aegyptium* (L.) Willd., both in the dunes and open field areas. These seven species account for 5.7% of the total richness.

Although the number of naturalized species collected in the study area is considered small, it indicates anthropization. However, this result cannot be used as a single parameter to assess the anthropization levels or vegetation degradation in Araoca. Future studies could cover a greater number of naturalized, exotic, or threatened species in the different phytophysiognomies of the restinga in Guimarães. Additionally, it is important to highlight that the naturalized species identified in this study are able to reproduce and maintain populations without compromising the diversity of the local flora, and do not necessarily behave as invasive species.

1. Phytophysiognomies

Araoca Beach has extensive areas of sandy strands with different restinga formations (Figure 3), varying from areas with herbaceous vegetation to trees and different anthropization levels. These formations are interspersed with areas of sediment deposition from the water table, sandy-muddy soil next to the herbaceous vegetation (further away from the seafront), flooded areas and freshwater marshes at the transition from shrub to forest vegetation, fixed dunes, and significant bands of mangrove parallel to the coast.

In the restinga, herbaceous vegetation plays an important role in stabilizing the substrate, preventing the action of erosive agents, and protecting sediments from the action of the wind. Shrub vegetation

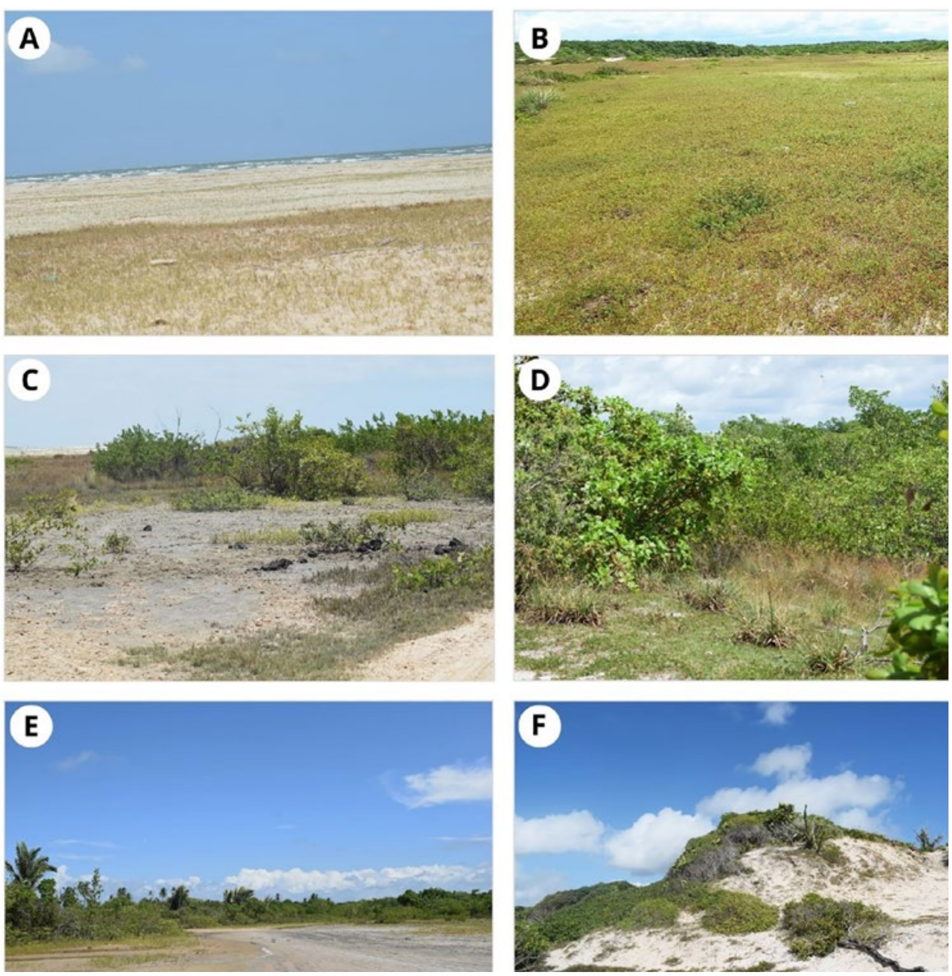

Figure 3. Phytophysiognomyes of the restinga of Araoca Beach, Guimarães, MA. a) Open, non-floodable fields in the dry period. b) Open fields that are not floodable during the rainy season. c) Open floodable fields close to mangroves, in the dry period. d) Open fruticete. e) Open floodable field, in a dry period. f) Dunes.

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favors shading, greater availability of nutrients, humidity, and moderate temperatures. These conditions provided by the shrub layer contribute to plant diversity in the restinga (Assumpção & Nascimento 2000, Carvalho et al. 2018).

In the restinga of Araoca Beach, we observed dense herbaceous-shrub vegetation, physiognomies of unflooded open field, flooded open field (with halophytes), flooded closed shrubby vegetation, unflooded open shrubby vegetation. These physiognomies were also found in the works by Correia et al. (2020) and Lima & Almeida Jr (2018) for the restingas of Itatinga (in Alcântara) and Panaquatira (in São José de Ribamar).

It is important to note that the different physiognomies in restinga areas can occur irregularly in the more delimited strips, forming a mosaic-like configuration (Machado 2016). The existence of transition zones in the restinga regions of Maranhão contributes to the vegetation not having an easily recognizable identity, resulting in a mosaic of physiognomies (Castro et al. 2012, Machado 2016).

1.1. Herbaceous physiognomy

Araoca's herbaceous vegetation is predominantly composed of halophilous, psammophilous, and stoloniferous plants, extending above the mean high tide level. The species found in this vegetation are *Alternanthera brasiliana* (L.) Kuntze, *Alternanthera tenella* Colla, *Blutaparon portulacoides* (A.St.-Hil.) Mears, *Ipomoea imperati* (Vahl) Griseb., *Sesuvium portulacastrum*, *Utricularia simulans*, and *Sporobolus virginicus* (L.) Kunth, this latter being dominant. These herbaceous species are extremely adapted to the saline soil's lack of nutrients, high temperatures, and sand mobility.

The herbaceous plants found in the open fields are under a sandy cordon along the coast, locally known as Ponta de Atins (Figure 4). This area is made up of vegetation with upright and cespitose species, and it is possible to see sparse thickets of species commonly found in mangroves, such as, *Rhizophora mangle* L., located in small dune formations, against a sandy muddy strip close to the mangrove. Still in Ponta de Atins, on Araoca Beach, there is an area of flooded open field located at the back of the mangrove line, where halophyte and cespitose species are found, such as *Batis marítima* L., *Cyperus ligularis* L., *Cyperus polystachyos* Rottb., and *Eleocharis geniculata* (L.) Roem. & Schult.

The characterization of this physiognomy, the presence of reptant species and propagules typical of mangrove species in Maranhão is close to what Lima et al. (2017) described for the Panaquatira restinga, in São José de Ribamar, and Guterres et al. (2020) for the unflooded open field of Guia Beach, on São Luís Island. Still considering the herbaceous vegetation of Araoca Beach, in unflooded formations, the herbaceous vegetation is interspersed with sub-shrub and shrub formations, where species such as *Borreria verticillata*, *Conocarpus erectus* L., *Crotalaria*

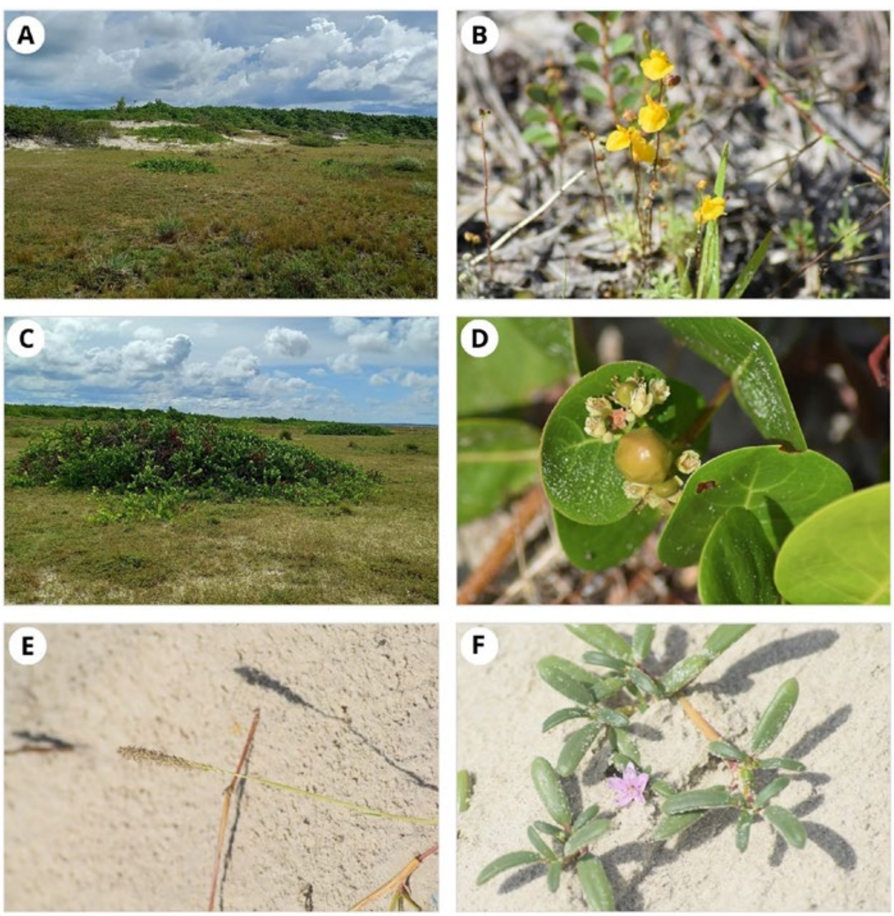

Figure 4. Restinga of Araoca Beach, Guimarães, MA. a) Ponta de Atins area, open field. b) *Utricularia simulans*, a herbaceous plant present in the open field area. c) Open field close to the mangrove area, presence of small dune formations and spaced thickets of mangrove species. d) *Chrysobalanus icaco* developing in the open field. e) S*porobolus virginicus* (Poaceae). f) *Sesuvium portulacastrum* (Aizoaceae).

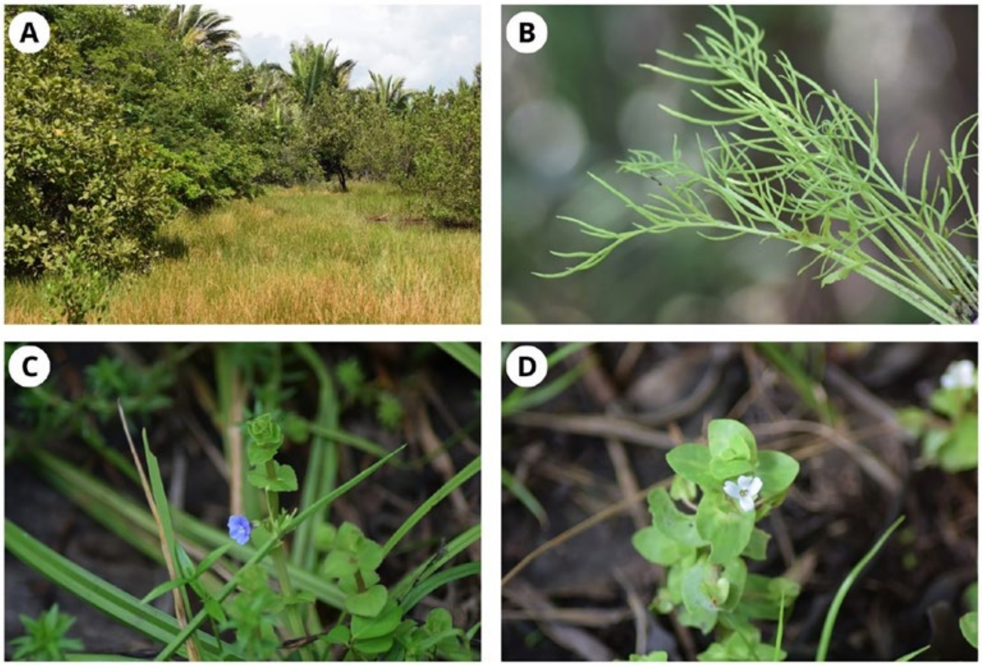

Figure 5. Restinga of Araoca Beach, Guimarães, MA. a) Phytophysiognomy of floodable closed fruticete. b) *Ceratopteris pteridoides* developing in a humid area with a closed fruticete. c) *Bacopa aubletiana* in herbaceous vegetation within a closed floodable fruticete. d) *Bacopa salzmannii* in a flooded environment with closed fruticete.

retusa, *Schultesia guianensis* (Aubl.) Malme, *Sida ciliaris* L., and *Turnera melochioides* are found.

As for the herbaceous-shrub physiognomy in the flooded closed field (Figure 5), small species predominate, such as *Axonopus capillaris* (Lam.) Chase, *Bacopa aquatica* Aubl., *Bacopa salzmannii* (Benth.) Wettst. ex Edwall, *Ceratopteris pteridoides*, *Lindernia crustacea* (L.) F.Muell., *Nepsera aquatica* (Aubl.) Naudin, *Xyris savanensis* Miq. This vegetation can be found in some stretches of marshy areas of post-beach vegetation, where there is a considerable concentration of organic matter in the soil resulting from the decomposition of leaves of shrub species such as *Anacardium occidentale*, *Cereus jamacaru* DC., and *Myrcia multiflora*, which form a favorable substrate for the establishment of these taxa.

In the Panaquatira restinga, Lima et al. (2017) categorized this set of herbaceous-shrub physiognomy in a closed flooded field as flooded closed shrubby vegetation, with *Anacardium occidentale* and *Lindernia crustacea* as one of the main shrubs and herbaceous representatives, respectively.

1.2. Shrub physiognomy

In Araoca sparse shrub thickets in different developmental stages are interspersed with open areas where herbaceous individuals are present or not, which characterizes the physiognomy of open shrubby vegetation. The vegetation is mainly composed of *Anacardium occidentale*, *Byrsonima crassifolia*, *Cenostigma bracteosum* (Tul.) Gagnon & G.P.Lewis, *Cordiera sessilis* (Vell.) Kuntze, *Chrysobalanus icaco*, *Myrcia guianensis*, *M. multiflora*, and *Solanum stramoniifolium* Jacq. Associated with this vegetation, sub-shrub species such as *Borreria verticillata*, *Mitracarpus strigosus* (Thunb.) P.L.R.Moraes, De Smedt & Hjertson, and *Crotalaria retusa* are highly abundant.

In the shrubby restinga, stretches with denser, closed vegetation can be found between sub-shrubby vegetation and sparse herbaceous vegetation. This shrubby vegetation (Figure 6) is located in a post-beach area with ecological characteristics that indicate flooding during rainy periods. Isolated, narrow strips of mangrove shrub species (*Avicennia schaueriana*, *Conocarpus erectus*, and *Rhizophora mangle*), as well as species associated with mangrove-restinga transition zones (*Chrysobalanus icaco*), are interspersed with dense herbaceous bands composed primarily of *Eleocharis interstincta* (Vahl) Roem. & Schult. In the same locality, this physiognomy gives way to less dense and more sparse bushes arranged in a strip of bare soil forming trails and cattle grazing areas modified by the constant anthropic action.

The physiognomy of unflooded open shrubby vegetation was also described for the restinga of Itatinga Beach, in the municipality of Alcântara. Correia et al. (2020) characterized the shrubby vegetation of Itatinga by spaced plants, mainly *Byrsonima crassifolia*, and areas of bare soil or with herbaceous vegetation such as *Crotalaria retusa* and *Cyperus ligularis*, as well as species of the genera *Zornia* and *Desmodium* (Fabaceae). Lima et al. (2017) described this same physiognomy for the Panaquatira restinga, which has *Byrsonima crassifolia* among the shrubs and the genera *Zornia* and *Desmodium* among the herbaceous. Shrub restingas can serve as ecological corridors for species in ecotonal environments and provide refuge and nursery areas (Serra et al. 2016, Costanza et al. 2017, Paiva & Almeida Jr et al. 2020).

1.3. Herbaceous-shrub physiognomy on the dune areas

The area of fixed dunes in the restinga of Araoca Beach (Figure 7) has a very diverse floristic composition compared to the herbaceous and shrubby vegetation found in the open fields of the Ponta de Atins

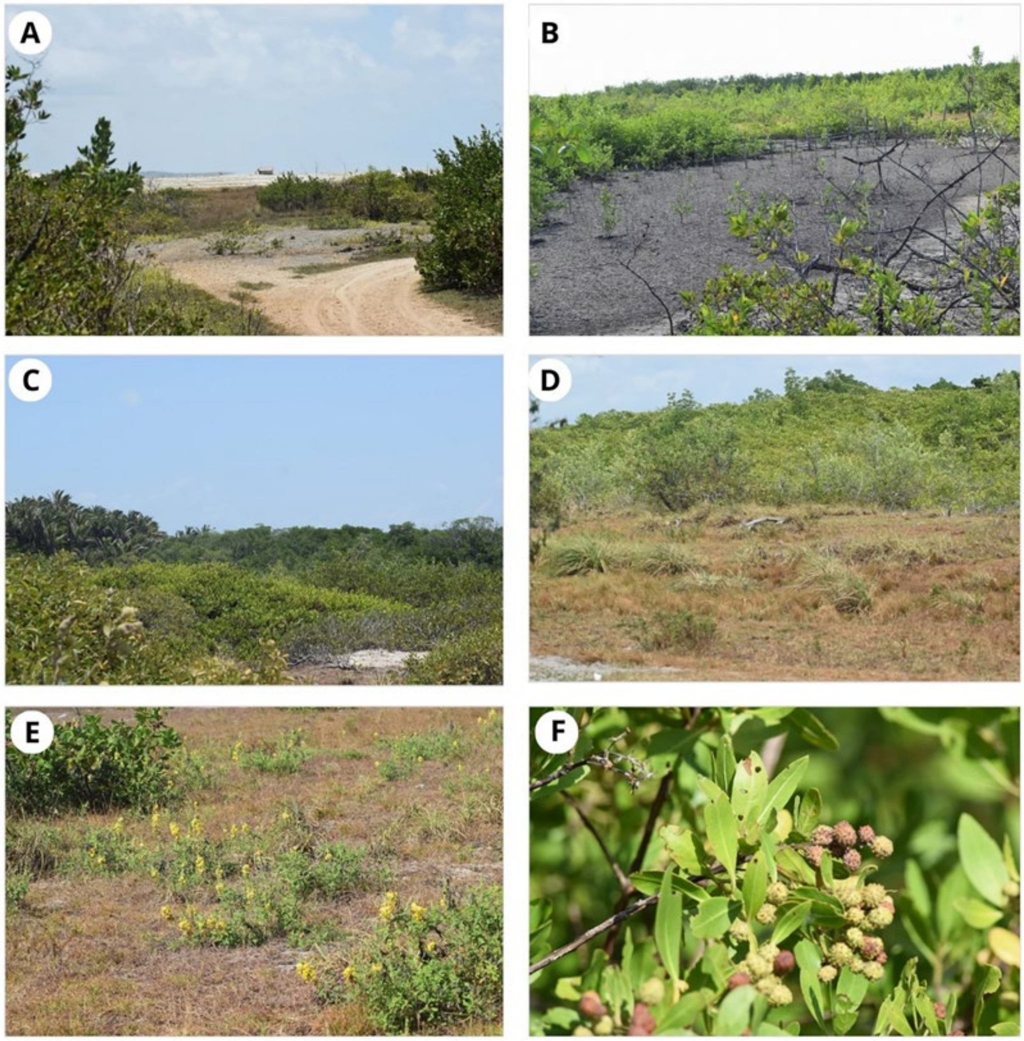

Figure 6. Restinga of Araoca Beach, Guimarães, MA. a) Phytophysiognomy of open fruit, with the presence of bare sandy soil. b) Fruticete composed of mangrove species, with a floodable area. c) Closed fruticete. d) Subshrub thickets of *Crotalaria retusa*. e) *Conocarpus erectus* (Combretaceae).

area. Shrubs dominate the dune physiognomy, with dense vegetation cover in the higher parts characterizing the closed shrubby vegetation.

The dunes have an herbaceous-shrubby vegetation rich in shrubs, herbaceous vines, lianas (woody vines), epiphytes, and halophilous and psammophilous herbs, extremely adapted to high insolation, nutrient scarcity, salinity, and sand mobility. The shrub vegetation is mostly made up of species from the Myrtaceae family (*Eugenia flavescens* DC., *E. punicifolia* (Kunth) DC., *Myrcia guianensis*, *M. multiflora*, *M. splendens* (Sw.) DC.), Rubiaceae (C*ordiera sessilis*, *Tocoyena sellowiana* (Cham. & Schltdl.) K.Schum., *Isertia hypoleuca* Benth.), Anacardiaceae (*Anacardium occidentale*), Chrysobalanaceae (*Chrysobalanus icaco*), Malpighiaceae (*Byrsonima crassifolia*), Sapindaceae (*Pseudima frutescens* (Aubl.) Radlk.), and Sapotaceae (*Manilkara bidentata* (A.DC.) A.Chev.).

Among the vines and lianas established in the dunes, we highlight species from the Passifloraceae (*Passiflora foetida* L., *P. subrotunda* Mast.), Fabaceae (*Abrus precatorius* L., *Centrosema brasilianum* (L.) Benth., *Clitoria falcata* Lam., *C. simplicifolia* (Kunth) Benth., *Entada polystachya* (L.) DC., *Galactia striata* (Jacq.) Urb.), Menispermaceae (*Odontocarya duckei* Barneby), and Vitaceae (*Cissus erosa* Rich.). Among the epiphytes, we found monilophytes such as *Lygodium venustum* (Lygodiaceae) and a species of Araceae (*Philodendron acutatum* Schott) in a denser area difficult to access.

Herbaceous plants from the Fabaceae family were found in a higher number, such as *Chamaecrista desvauxii* (Collad.) Killip, *C. diphylla*, C. *nictitans* (L.) Moench, *C. tenuisepala* (Benth.) H.S.Irwin & Barneby, *Stylosanthes angustifolia* Vogel, *Zornia brasiliensis* Vogel, *Z. guanipensis* Pittier, and *Z. latifolia* Sm. Herbaceous plants from the families Convolvulaceae (*Ipomoea imperati*), Turneraceae (*Turnera melochioides*), Boraginaceae (*Euploca polyphylla* (Lehm.) J.I.M.Melo & Semir), Eriocaulaceae (*Eriocaulon cinereum* R.Br.), and Malvaceae (*Sida ciliaris*) were also found in the lower dune areas. Among the families with the highest specific richness in this environment, Fabaceae and Rubiaceae also stood out in the study of the structural characterization and conservation status of the dunes of São Marcos Beach, in São Luís (Araujo et al. 2016).

Besides to the above-mentioned species, we also registered in Aroca Cactaceae (*Cereus jamacaru*) and Plumbaginaceae (the toxic

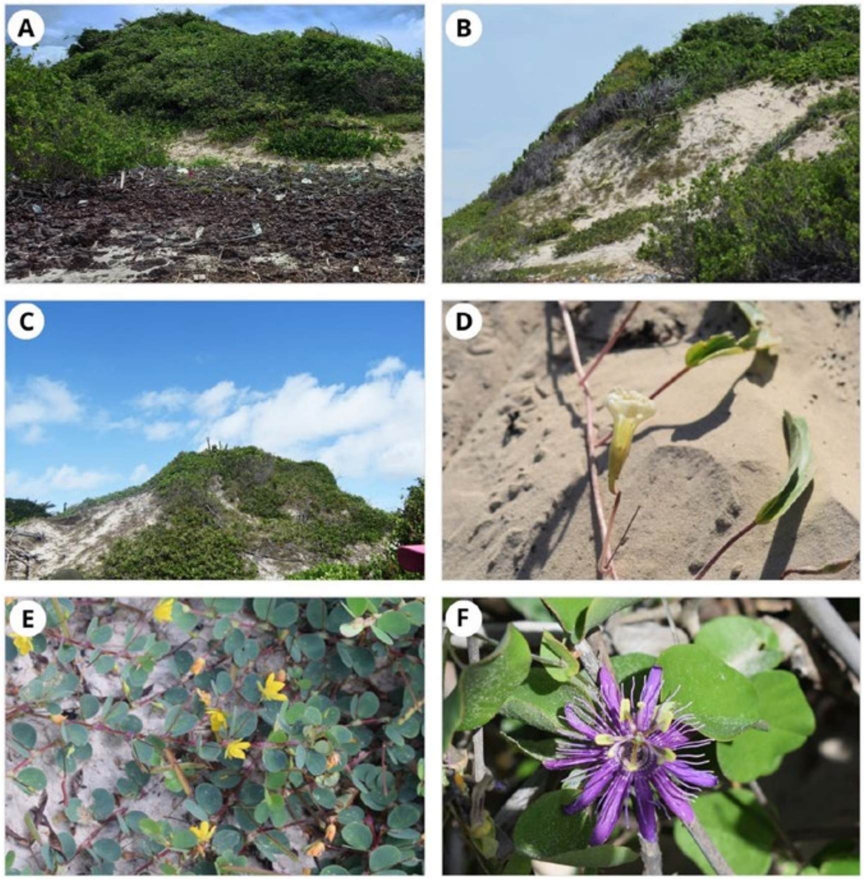

Figure 7. Restinga of Araoca Beach, Guimarães, MA. a) Dune formation in the Atins Area with rocky elements forming barriers in the lowest portion. b–c) Herbaceous-shrub phytophysiognomy of fixed dunes. d) *Ipomoea imperati* in dune areas. e) *Chamaecrista diphylla* in the lower strip of the dunes. f) *Passiflora subrotunda* in dune area.

and ornamental plant *Plumbago scandens* L.), this latter spread in the dunes through human action. We also identified a large amount of plastic, glass, and rubber waste (including packaging, bottles, and bags) in the dunes and surrounding areas. As dunes act as sediment stocks on the beach strip in erosive episodes (Amorim et al. 2023), this scenario represents a threat to the sedimentary dynamics. Paiva & Almeida Jr (2020) reinforced the role of dunes as a natural protection for the adjacent coastal zone.

The floristic survey of the dunes of São Marcos Beach carried out by Silva et al. (2016) listed the predominance of herbaceous species influenced by abiotic variables. Most of these herbaceous species have rhizome and stolon structures that play an important role in storing water and essential nutrients and reproduce asexually often via clonal reproduction, which facilitates their rapid growth and favors their effective occupation of the dune substrate (Seeliger 1998, Silva et al. 2016).

2. Floristic similarity

Based on our analysis, the floristic similarity between Araoca Beach and Guia Beach was 36.8%, with 45 species shared between the two areas: 21 herbaceous species in 13 families, 11 shrub species in 10 families, 9 sub-shrubs, 3 climbers, and 1 liana (Guterres et al. 2020). Fabaceae, Asteraceae, Lamiaceae, and Rubiaceae were the most common families in both areas. When comparing Panaquatira restinga with Araoca, the similarity was 40.1%, with 49 species (31 herbaceous) distributed in 25 families. *Utricularia simulans* should be highlighted, as it is one of the most widely distributed herbaceous plants in the Araoca restinga which was also present in Panaquatira.

In the comparison with the Itatinga restinga, 54 species were listed in common with Araoca, which represents a percentage similarity of 44.2%. This result was expected, considering the geographical proximity between the municipalities of Guimarães and Alcântara. Correia et al. (2020) listed 22 herbaceous species, 19 shrubs, 7 sub-shrubs, 4 vines, and 2 lianas in Araoca Beach within the physiognomies of unflooded open field, unflooded open shrubby vegetation, and unflooded closed shrubby vegetation.

The DCA analysis demonstrated the segregation of plant communities on Araoca beach in relation to other restinga areas on the DCA 1 axis (Eigenvalue= 0.4963). On the DCA 2 axis (Eigenvalue= 0.3588), Araoca beach has greater floristic proximity to Itatinga and Panaquatira beaches (Figure 8), which is in line with the similarity percentages observed.

Besides, climatic conditions and the biomes may also be significant factors influencing the levels of floristic similarity among the compared restinga areas (Rabelo et al. 2024). The Maranhão Amazon, located in a region characterized by a humid climate with little to no water deficiency (Martins & Oliveira 2011), contributes to the colonization

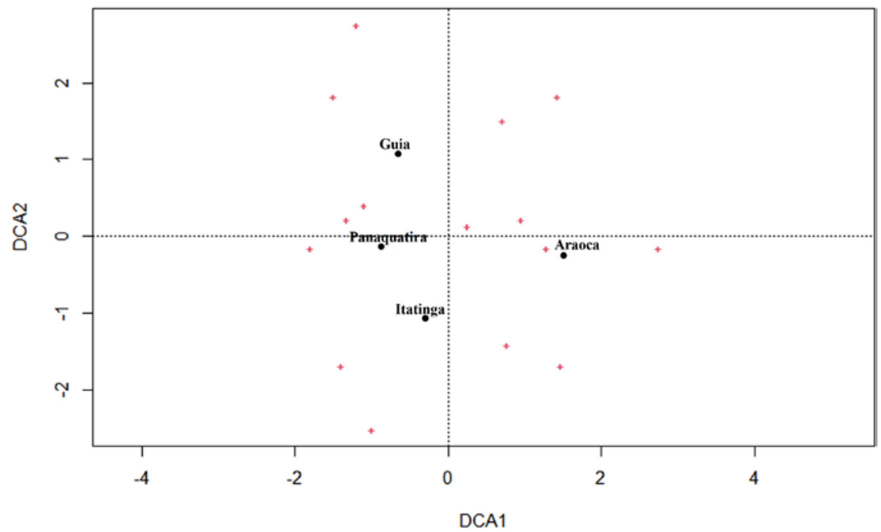

Figure 8. The results of the DCA analysis showed the following lengths of the two main axes: DCA1 = 2.5055 (eigenvalue = 0.4963) and DCA2 = 2.1326 (eigenvalue = 0.3588).

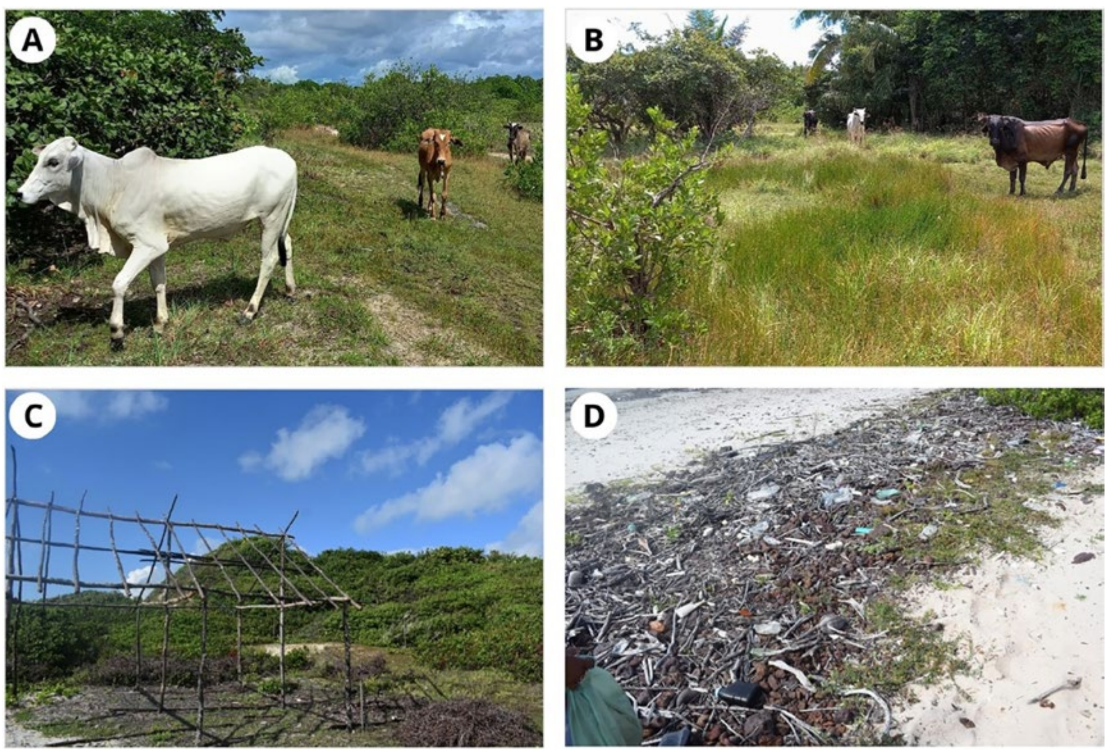

Figure 9. Records of environmental impacts on the restinga of Araoca Beach, Guimarães, MA. a–b) Cattle grazing in herbaceous and shrub fields. c) Burning in an area near the dunes for the construction of huts. d) Solid waste deposited on the lower strip of the dunes, with the growth of herbaceous species.

of the Maranhão coastal restingas by species from the Amazon biome (Serra et al. 2016, Lima & Almeida Jr. 2018).

3. Environmental impacts and restinga conservation

The resolutions of the National Environment Council (CONAMA) (No. 07/1996, No. 303/2002, and No. 417/2009) and the New Forest Code (Federal Law No. 12,651 of May 25, 2012) legitimize restingas as Permanent Protection Areas. Restingas provide ecological services that contribute to maintaining biodiversity (Darold & Irigaray 2018). Even so, the restinga is considered a fragile ecosystem, constantly threatened and degraded mainly due to unsustainable extractivism. Among the activities contributing to its degradation are illegal practices for sand extraction, expansion of agriculture, invasion of exotic species, and real estate speculation.

The different plant formations of the Araoca restinga are biologically rich, with important species in this ecosystem. The typical physiognomies are still in a good state of conservation. However, the preservation of these environments is under threat due to the expansion of human activity. Fruit species play an important role in the permanence of the local fauna, especially insects, birds, and some mammals that have different patterns of habitat use.

Another significant anthropogenic impact identified in the dunes and surrounding areas is the accumulation of plastic, glass, and rubber waste, including packaging, bottles, and bags. This pollution poses a threat to the sediment dynamics of dune vegetation. During erosive events, dunes act as sediment reserves for beach areas (Amorim et al. 2023). Furthermore, Paiva & Almeida Jr. (2020) highlight the critical role of dunes as natural barriers that protect adjacent coastal zones.

In the study conducted by Guterres et al. (2019), signs of anthropogenic disturbance were identified related to the deposition of solid waste along the restinga vegetation near the urban perimeter of Caúra Beach, in São José de Ribamar. On the other hand, the dunes of Araoca Beach are located on the sandy ridge of Ponta dos Atins, a region parallel to the coast and far from urbanized areas. Although not close to urban centers, Ponta dos Atins is frequented by tourists and fishers, which contributes to the presence of debris across the open field. Most of the waste found on the slopes of the dunes is transported by tides, originating from areas affected by beach tourism and commercial activities.

Some areas of the original vegetation of the Araoca restinga have been devastated by the expansion of tourism-related urbanization, such as the construction of bars and chalets. Some areas of the original restinga vegetation in Araoca have been devastated to accommodate urban expansion related to tourism. The construction of bars and chalets serves as strong evidence of human interference in the Araoca Beach area. In the Atins region, fishing activities carried out by residents of the municipality of Guimarães, along with waste brought in by the tides, contribute to the accumulation of garbage in the area. The formation of trails indicates the removal of shrub species, while cattle grazing in the region leads to herbivory and trampling of key herbaceous-shrub species vital to the ecosystem. Additionally, the significant amount of litter left near the dunes highlights how tourism contributes to pollution, altering the dynamics and compromising the balance of this critical ecosystem (Figure 9).

Conclusion

Our floristic survey inventoried 122 herbaceous and shrub species in the restinga of Araoca Beach, enabling the characterization of the different phytophysiognomies present in the studied area, which were previously little known. Furthermore, the results highlight the need to conduct studies aimed at maintaining the plant diversity of the Reentrâncias Maranhenses EPA, directly contributing to its protection.

This is evident when considering that the Araoca Beach ecosystem is vulnerable to potential loss of plant biodiversity due to anthropogenic pressures, such as urbanization associated with tourism, improper waste disposal near the dunes, and the formation of trails and grazing areas for livestock. These activities compromise the dynamics and balance of the vegetation on Araoca Beach.

In light of this scenario, it is imperative to implement conservation measures and promote awareness to protect this unique ecosystem and its species. Actions such as waste control, proper tourism management, and the prevention of unplanned urban expansion are essential to ensure the survival and preservation of the rich biodiversity of Araoca Beach.

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Conflicts of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

Ethics

This work was approved by the Biodiversity Authorization and Information System (SISBIO) of the Chico Mendes Institute for Biodiversity Conservation (ICMBio) under the number 84049-1.

Data Availability

The data collected and generated during this study are available in https://doi.org/10.48331/scielodata.WGB8VS.

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Short Communication

The first record of an Asian mangrove, *Heritiera fomes* Banks (Malvaceae: Sterculioideae), occurring outside of cultivation in Ecuador and the Americas

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Abstract: The first record of an Asian mangrove, *Heritiera fomes* Banks (Malvaceae: Sterculioideae), occurring outside of cultivation in Ecuador and the Americas is documented. The species was discovered in 2022 on Santay Island in the Gulf of Guayaquil, Ecuador, where it is naturalized and produces flowers and fruits at least twice per year. Populations were located on both the shore and in inland areas on the northern part of the island, which is directly influenced by fresh water from the Babahoyo and Daule rivers. *Heritiera fomes* co-occurs with native species in both mangrove swamp and dry forest. As many as 0.45 individuals/m^2 were observed. How the species was introduced into Ecuador is unknown. However, given the similarity of the environmental conditions of Santay Island to those of the native range of *H. fomes*, its invasive potential in Ecuador is of concern and discussed.
Keywords: *Exotic species; Invasive; Santay Island*.

El primer registro de un mangle asiático, *Heritiera fomes* Banks (Malvaceae: Sterculioideae), existiendo fuera del cultivo de Ecuador y las Américas

Resumen: El primer registro de un manglar asiático, *Heritiera fomes* Banks (Malvaceae: Sterculioideae), el cual ocurre fuera del cultivo en Ecuador y las Américas. La especie fue encontrada en 2022 en la Isla Santay en el Golfo de Guayaquil, Ecuador, donde parece estar naturalizada y produce flores y frutos al menos dos veces al año. Se descubrieron varias poblaciones en la parte norte de la isla, que está directamente influenciada por el agua dulce de los ríos Babahoyo y Daule. *Heritiera fomes* se encuentra tanto en la costa como áreas de la isla donde crece junto a especies nativas de manglar y bosque seco. Se observaron hasta 0.45 individuos/m^2. Se desconoce cómo se introdujo la especie. Sin embargo, dadas la similitud de las condiciones ambientales de la Isla Santay a las del hábitat de *H. fomes,* su potencial de invasión en Ecuador es motivo de preocupación y se discute.
Palavras-chave: *Especies exóticas; Invasoras; Isla Santay*.

Introduction

Introduced species are a global problem due to their impacts on biodiversity conservation (Fei et al. 2014). Some studies suggest that they are the second leading cause of biodiversity loss after habitat fragmentation (Mosquera et al. 2022). Biological invasions occur when a species is introduced from a biogeographically isolated region by human activities, establishes a self-sustaining population, and expands its range (Richardson & Pyšek 2006; Blackburn et al. 2011).

Mangroves, which form a coastal intertidal ecosystem present in the tropics and subtropics, are not exempt from invasions (Krauss & Ball 2013). Until recently the inherent salinity, anoxia, and wave conditions impacting mangrove ecosystems were thought to prevent invasion by alien species (Biswas et al. 2018). However, multiple reports of invasive species are known now for mangroves, and they cause reduced natural regeneration, increased sedimentation, and reduced biodiversity (Fei et al. 2014; Fazlioglu & Chen 2020). Most of these introductions have been accidental (>80%); many fewer have been intentional (~18%) and these latter often are from a desire to take advantage of the benefits of mangroves (e.g., soil stabilization and coastal protection) (Chowdhury et al. 2016; Biswas et al. 2018).

Mangrove species also can be invasive (Fazlioglu & Chen 2020). The impact of exotic mangrove species on mangrove ecosystems varies according to the site. For example, *Sonneratia apetala* Banks on Hainan Island, China, improved microenvironmental conditions and favored the recruitment of native mangroves (Xin et al. 2013). In contrast, *Laguncularia racemosa* (L.) C.F.Gaertn. on the same island displaced the native mangrove *Rhizophora apiculata* Blume (Fazlioglu & Chen 2020). On Molokai Island in Hawaii, *R. mangle* L. and *Conocarpus erectus* L. replaced exotic species established more than a century ago, generating a completely new plant formation on the island and possibly altering ecological processes (Allen 1998).

It has been suggested that tidal dynamics create and vacate multiple microhabitats of low salinity and waves, increasing susceptibility to invasion by invasive non-halophyte and facultative halophyte species (Krauss & Ball 2013; Sarker et al. 2019). In Ecuador, this susceptibility could be exacerbated when considering the high anthropic pressure observed in mangroves; pressure created by aquaculture and urbanization (Morocho et al. 2022).

Santay Island is a delta located at the mouths of the Babahoyo and Daule rivers and receives a flow of 1600 $m^3 s^{-1}$ of fresh water, exhibiting salinity levels of up to 4.2 ppm (Arreaga Vargas 2000; Villegas et al. 2021). These low salinity conditions have favored the invasion in the vicinity of the island of non-halophyte alien species such as *Eichhornia crassipes* (Mart.) Solms, as well as native dry forest woody taxa such as *Entada polystachya* (L.) DC., *Mimosa pigra* L., and *Erythrina fusca* Lour. (personal observation).

Recently, an exotic mangrove, *Heritiera fomes* Banks, was discovered in the northern part of Santay Island, an area directly influenced by the Babahoyo and Daule rivers. In its native range in Asia, *H. fomes* although extensive in its distribution is considered a threatened species due to progressive declines in populations caused by expansion of the agricultural frontier and exploitation of its wood (Kathiresan et al. 2010; Chowdhury et al. 2016; Khan et al. 2020). In the world's most extensive mangrove formation, the Sundarbans, *H. fomes* dominates hypo- and meso-saline habitats forming monospecific stands and clearly prefers upstream sites (i.e., freshwater-dominated river systems) (Iftekhar & Saenger 2008; Sarker et al. 2016). Given the similarity of the environmental conditions of its native habitat to that of Santay Island, in terms of salinity and tidal patterns as well as traits related to invasive species (high fecundity, high survivability of seeds), *H. fomes* potentially represents a threat to the native flora of the island and its surroundings. Given these concerns, the objective of this research was to describe the current distribution of *H. fomes* on Santay Island in order to provide baseline ecological data.

Materials and Methods

1. Study area

Santay Island is a 2,215 ha delta located at the mouth of the Guayas River (2°13'26"S, 79°51'22"W) (Figure 1). It is bounded on the west by the city of Guayaquil and on the east by Durán and is impacted directly by industrial and agricultural wastewater from both these municipalities (Deknock et al. 2019). Until 1980, before the island was expropriated by the Ecuadorian government, it was an area of intensive livestock and agricultural activity (Zambrano Moreira et al. 2019). At present, these activities have been replaced by tourism that was facilitated by the construction of bridges to connect the island with both Guayaquil and Durán.

At least three plant formations can be distinguished in the area: (a) mangroves (i.e., *Avicennia germinans* (L.) L., *Conocarpus erectus*, *Laguncularia racemosa*, and *Rhizophora* spp.), with *Annona glabra* L., and wetland grasslands that include the fern *Tectaria fernandensis* C.Chr., which dominate the shores and frequently flooded areas; (b) shrubby elements of dry forests (e.g., *Erythrina fusca*, *Entada polystachya*, and *Mimosa pigra*) in inland areas with less flooding frequency; and (c) in the transitions between these formations, where meadows dominated by exotic and native species (e.g., *Hymenachne* sp., *Paspalum* sp., *Cyperus* sp., and *Uniola pittieri* Hack.) are observed, interspersed with patches of emergent palm trees (*Roystonea oleracea* (Jacq.) O.F.Cook and *R. regia* (Kunth) O.F.Cook) that grow to 40 m tall.

2. Discovery and identification

In early 2022, park rangers from the Isla Santay National Recreation Area noticed the presence of an unfamiliar plant similar to a mangrove in the northern part of the island (2°11'48"S, 79°51'8"W). Given their initial observation and taking advantage of field work conducted as part of a larger project focused on mangroves of Ecuador, fertile samples of the plant were collected to determine its taxonomic identity. Voucher specimens were made from June 2022 to January 2023, which corresponds to the dry season in this area.

These vouchers were deposited in the National Herbarium of Ecuador (QCNE) in Quito where this unknown plant tentatively was identified as an unknown genus and species of Malvaceae (Sterculioideae). Photographs were sent to one of us (LD) who confirmed that the family and subfamily were correct and who recognized the plant as a species of *Heritiera* Aiton, which is not native to the Americas. Once specimens were provided, the unknown "mangrove" was identified as *H. fomes* (Figure 2). Although the authorship of this species name often is attributed to Francis Buchanan-Hamilton (abbreviated as "Buch.-Ham."), the preface to Symes (1800), where the name was first published, states that "Sir Joseph Banks selected and described the plants." Therefore, the correct author citation is Banks.

Around the locality where *Heritiera fomes* was first found, land surveys were conducted in a radius of 1 km to determine the distribution of the species on the island. Simultaneously, flooded areas near and around the island were surveilled by boat. In both cases, all observed occurrences of the species were georeferenced. In this process, two more populations of *H. fomes* were located to the southwest of the original site discovered by park rangers, at 100 and 300 m distance respectively from the shore (Figure 1).

3. Abundance, density, and habitat

To estimate population density, 10 × 10 m plots were installed in each of the three identified populations. All individuals present in each plot were censused. Individuals with a basal diameter less than 1 cm measured at 5 cm above the ground were considered seedlings

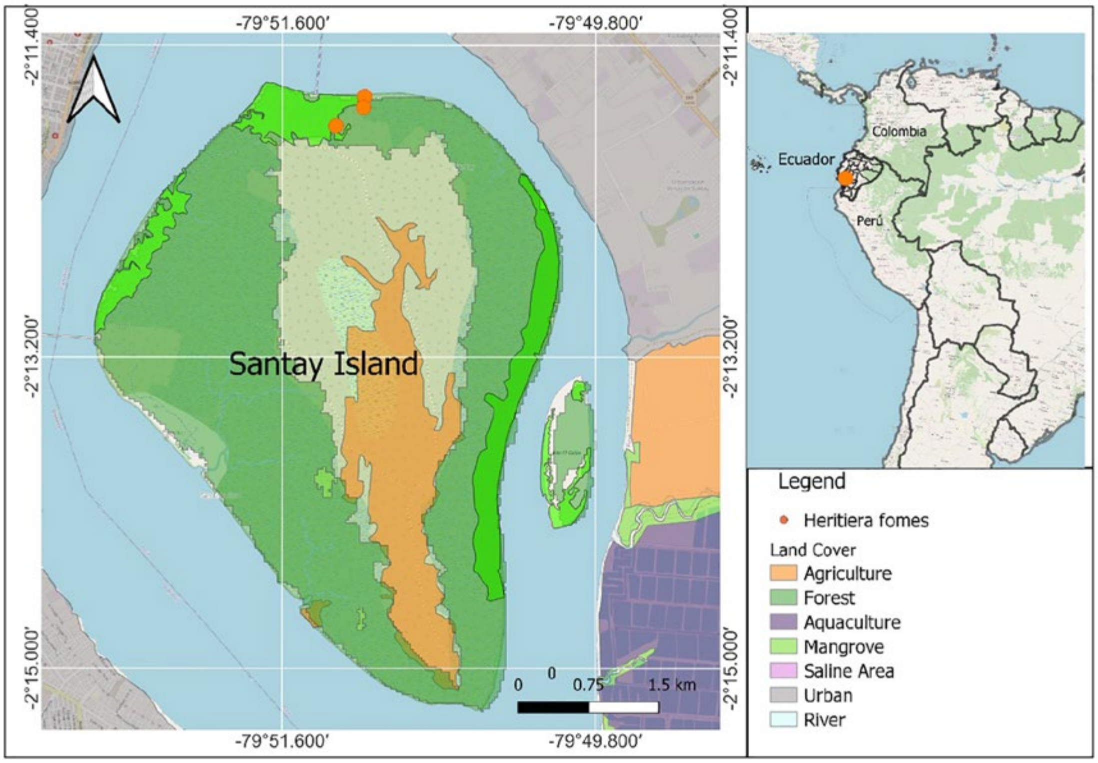

Figure 1. Map of Santay Island, Ecuador showing the areas where *Heritiera fomes* occurs. (Layers of land use and political boundaries were obtained from Open Street Map, Ecosystem Classification System of Continental Ecuador, and the Military Geographic Institute; see Google Street Contributors 2024; Instituto Geográfico Militar 2023; Ministerio del Ambiente del Ecuador 2012).

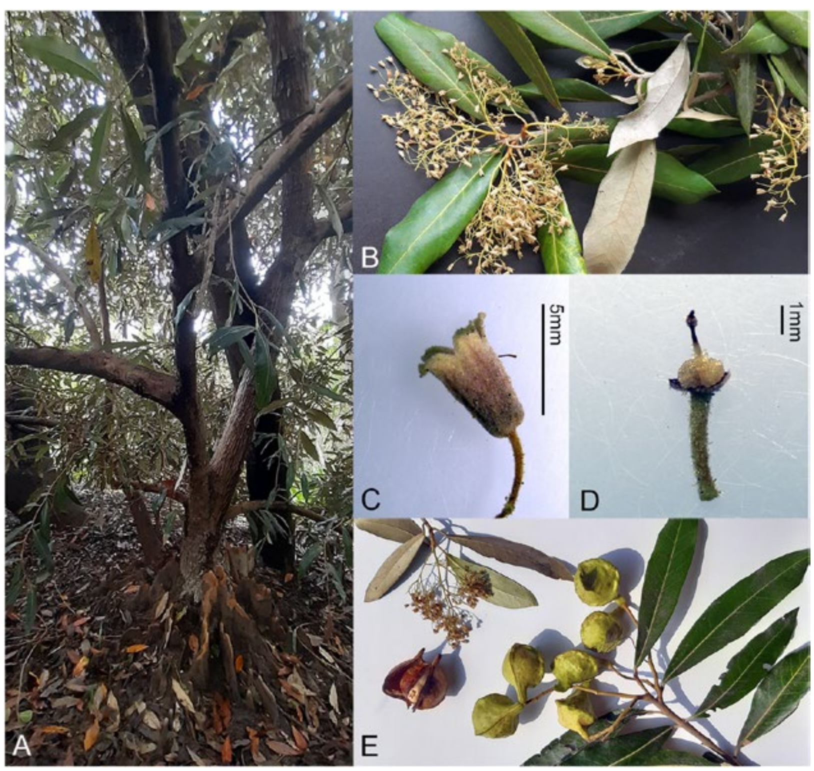

Figure 2. *Heritiera fomes* on Santay Island, Ecuador. – A. Habit of mature tree (note plank buttresses at base). – B. Inflorescence and leaves. – C. Flower. – D. Staminate flower with calyx removed (note the androgynophore and subapical ring of anthers). – E. Inflorescence (upper left), infructescence (right) with green fruit, and a single detached older fruit (lower left) (note the irregular knobby surface and transverse ridges on the green fruit, which are characteristic of *H. fomes*).

(Chen et al. 2010; Wahyuningtyas et al. 2022), individuals with a diameter >1 cm and with woody stems branched from the ground were considered shrubs, and individuals with woody stems not branched above the ground were considered trees. The habit and diameter of all individuals were recorded, and height was estimated; diametric tapes were used to measure the diameter and an aerial pruner to estimate the height. For trees and shrubs, the diameter was measured at 1.3 m above the ground (DBH) and in seedlings diameter was measured at 5 cm above the ground (Wahyuningtyas et al. 2022). In addition, associated plant taxa, habitat, water level, and canopy cover were recorded for each site. The data were digitized using Libreoffice version 7.3.2.2. The processing, analysis, and mapping of the data were conducted in the statistical programming software R version 4.1.3 using the tidyverse, sf, and ggspatial packages (R Core Team 2022).

Results

In total, 14 trees, 16 shrubs, and 51 seedlings of *Heritiera fomes* were recorded in the three plots. *Heritiera fomes* was observed with flowers in June, July, November, and December, and with fruits in December and January. The abundance, between adults and seedlings, was 64 individuals on the shore, nine individuals from 9 to 100 m distance from the shore, and eight individuals at 300 m from the shore. The density of seedlings was highest on the shore with up to 0.45 seedlings/m^2 observed. Seedling density decreased as the distance from the shore increased, registering 0.05 seedlings/m^2 at 300 m from the shore. Additionally, seedlings were found at a distance of between 0.3 to 4 m from a parent plant.

The average height of the trees was 8.79 m with some individuals reaching more than 13 m (Table 1). Notably, branching patterns varied according to habitat with the number of branching forks being higher in areas with high frequency of flooding. Shrubs measured an average of 2.68 m tall and did not exceed 5 m, while seedlings reached an average of 0.42 m in height. With respect to diameter, trees had the greatest range of variation, from 5 to 23 cm DBH, but in general they averaged 13.26 cm DBH. Shrubs had an average diameter of 4.38 cm, although several individuals were found with diameters less than 3 cm.

Regarding habitat, *Heritiera fomes* had an abundance of 72 individuals in areas with high frequency of flooding, while in areas with less frequent flooding, where soil compaction and the frequency of dry forest taxa are higher, abundance dropped to nine individuals. In general, shrubs but not trees and seedlings tend to be present in habitats with higher canopy cover, although in all three habitats individuals are likely to be found both in areas without canopy cover and in areas with up to 90% cover (Table 2). Seedlings tend to be more frequent in habitats with low water levels, while trees and shrubs can be present both in dry areas as well as flooded and swampy areas.

Within *Heritiera fomes* populations different taxa associated with each life form were observed. Seedlings were mostly associated with *Avicennia germinans*, *Crinum kunthianum* M.Roem., and *Rhizophora racemosa* G.Mey. (Table 3). Shrubs occurred mainly in areas where *Laguncularia racemosa*, *Sphagneticola trilobata* (L.) Pruski, and *C. kunthianum* were present. Trees were more abundant in areas where *A. germinans* and *Roystonea oleracea* occurred.

Table 1. Measures of central tendency and dispersion of the variables DBH (cm) and Height (m) for each life form recorded for *Heritiera fomes* on Santay Island.

Life form	n	Min	Max	Mean	SD
DBH					
seedling	51	0.5	0.7	0.56	0.08
shrub	16	2.0	9.5	4.38	2.27
tree	14	5.0	23.0	13.26	6.92
Height					
seedling	51	0.4	0.5	0.42	0.04
shrub	16	1.0	5.0	2.68	1.35
tree	14	6.0	15.0	8.79	3.24

Table 2. Measures of central tendency and dispersion of the variables Canopy cover (%) and Water level (cm) for each life form recorded for *Heritiera fomes* on Santay Island.

Life form	n	Min	Max	Mean	SD
Canopy cover					
seedling	51	12	90	46.16	21.23
shrub	16	0	90	71.38	30.91
tree	14	0	80	43.00	26.76
Water level					
seedling	51	5	100	9.31	18.57
shrub	16	5	100	49.38	35.44
tree	14	5	100	51.43	44.22

Table 3. Identity of taxa and number of individuals found close to occurrences of *Heritiera fomes* for each life form on Santay Island.

Species	Seedling	Shrub	Tree
Albizia multiflora (Kunth) Barneby & J.W.Grimes	0	5	0
Avicennia germinans (L.) L.	43	4	11
Crinum kunthianum M.Roem.	32	6	3
Entada polystachya (L.) DC.	2	1	1
Erythrina fusca Lour.	0	1	2
Laguncularia racemosa (L.) C.F.Gaertn.	6	9	1
Passovia pedunculata (Jacq.) Kuijt	2	1	1
Rhizophora mangle L.	0	0	1
Rhizophora racemosa G.Mey.	32	3	5
Roystonea oleracea (Jacq.) O.F.Cook	11	5	6
Sphagneticola trilobata (L.) Pruski	0	6	1
Tectaria fernandensis C.Chr.	0	2	1

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Discussion

The native range of *Heritiera fomes* is discontinuous. It is found in the Bay of Bengal where it occurs extensively near the mouth of the Ganges and Brahmaputra rivers in India and Bangladesh and in the Andaman Sea where it occurs at the mouth of the Irrawaddy (type locality) and Great Tenasserrim rivers in Myanmar and further south in peninsular Thailand (Kostermans 1959a, 1959b; Spalding et al. 1997; Malick 1991; Phengklai 2001; Rahman et al. 2012). Reports of the natural occurrence of this species in the Andaman Islands are suspect and probably represent misidentifications of *H. littoralis* Aiton (Phengklai 2001; POWO 2024).

In its native habitat, *Heritiera fomes* is a tree 15–25 m tall and 1.5–28 cm in DBH and is considered endangered (Kathiresan et al. 2010). Historically, larger trees of up to 2 m in circumference existed but they have mostly been harvested impacting the species in its natural distribution (Troup 1921; Hossain & Nizam 2002; Kathiresan et al. 2010). Trunks have prominent plank buttresses and after ca. three years the trees produce pneumatophores. Fruits on trees in India and Bangladesh measure 3.8–5.1 × 2.5–3.8 cm (Hossain & Nizam 2002). The species is found on the land side of mangrove forests (Spalding et al. 1997) because it does not tolerate regular flooding and prefers slightly to moderately saline areas (Hossain & Nizam 2002; Mandal et al. 2021). In the dense forest of *H. fomes*, the understory is practically absent (Hossain & Nizam 2002).

How *Heritiera fomes* arrived in Ecuador is unknown. Long distance dispersal seems unlikely. Drift from the Bay of Bengal and the Andaman Sea in the Indian Ocean would not find its way easily to the Pacific Ocean and once there, it would have to enter the South Pacific Subtropical Gyre, be carried north along the coast of South America in the Peru Current, and then make its way through the Gulf of Guayaquil to the northern end of Santay Island. A more likely explanation for the introduction of *H. fomes* is that its fruit inadvertently was transported from Asia to Ecuador by cargo ship. There is active ship commerce between these two areas and an incidental introduction is a possibility since the fruit of *H. fomes* has the ability to float. Venegas Vinueza (2010) documented exports and imports between India and Ecuador from 2005 to 2009 and the principal mode of transport was maritime with the main port of entry Guayaquil. In the last 25 years there has been an increase in exports from Ecuador to Bangladesh (OEC 2024). As of 2011, trade began to grow between Ecuador and Asian countries such as Myanmar and Thailand, among others (Coba 2019). Nonetheless, we cannot rule out an intentional introduction of *H. fomes* by ship's crew since the species has been used for medicinal purposes to treat stomach and skin conditions and to treat diabetes (Mahmud et al. 2014).

There is a single historical record of *Heritiera fomes* cultivated in the Americas. In the late 18th and early 19th centuries the species was cultivated in Trinidad, an island in the Caribbean. The sole collection known, *W.E. Broadway 796*, is vouchered by leaves and loose fruit collected in 1907 from a tree planted ca. twenty years earlier. Vouchers are deposited in the Université Claude Bernard Lyon 1 (leaves: LY [barcodes LY0151635, LY0151635, & LY0151637]; see also https://www.gbif.org/occurrence/3427793841) and in the Muséum national d'Histoire Naturelle, Paris (fruit: P [barcode P02443774]; see also https://www.gbif.org/occurrence/3436227327). Another collection of *Heritiera* made in the Botanic Gardens, Trinidad (*W.E. Broadway s.n.*, US [barcode 03383553]) although distributed as *H. fomes* was misidentified and is, in fact, *H. littoralis* (personal observation).

1. When did Heritiera fomes become established on Santay?

When *Heritiera fomes* became established on Santay Island is unknown. The size of the trees and the number of plants suggest it has been present for ca. 15–20 years. Flowering and fruiting are abundant, plants establish and grow easily, and although the species has only been found at three sites, they are all places where the environmental conditions are more similar to the native habitat of the species than elsewhere on the island. Notably, the northern part of the island is rich in sediments deposited by the Babahoyo and Daule rivers.

2. Invasive potential of Heritiera fomes

In native hyposaline habitats, the DBH of *Heritiera fomes* increases between 1.08 and 1.3 mm yr-1, reaching up to 25 m, which signifies it is a fast-growing species (Iftekhar & Saenger 2008; Chowdhury et al. 2016). *Heritiera fomes* flowers between March and August, and bears fruit between April and September. As much as 40% of the litter produced by the species corresponds to reproductive organs, which suggests that *H. fomes* allocates a large part of its biomass to reproduction (Azad et al. 2020; Mariam & Alamgir 2022). The germination of its seeds and the survival of its seedlings increases up to 96% and 25%, respectively, in hyposaline environments showing its preference for this type of habitat (Sarker et al. 2016; Mariam & Alamgir 2022). *Heritiera fomes* exhibits high SLA (Specific Leaf Area) and LI (Leaf Index) phenotypic plasticity under different light and salinity conditions; traits related to a broad ecological niche and shade tolerance (He et al. 2018; Khan et al. 2020). In fact, on Santay Island, *H. fomes* has established itself near species such as *Roystonea oleracea* and *Samanea saman* (Jacq.) Merr., which are species that compact the soil and produce constant shade, and also in completely uncovered habitats, thus showing a high range of tolerance to light and to competition with other species.

High growth rates, high fecundity, high phenotypic plasticity, hydrochory, and a persistent seed bank are linked to inherent traits of invasive species in wetlands (Zedler & Kercher 2004; Pyšek & Richardson 2007; Blackburn et al. 2011). In fact, it has been suggested that high SLA values and growth rates could be predictors of invasive species (Grotkopp & Rejmánek 2007). In this context, it is logical to suggest that mangroves located upstream with high freshwater flow are more susceptible to plant invasions; in fact, most of the invasive plants reported in the world are non-halophytes (Biswas et al. 2018).

In light of the traits exhibited by *Heritiera fomes* in its native habitat, it is prudent to consider its invasive potential on Santay Island. Environmental conditions (e.g., salinity < 2 dS m^{-1} and low waves) are similar to its native habitat in Sundarbans (Sarker et al. 2019). Nonetheless, it is ironic that in its native range, *H. fomes* is now considered to be an endangered species primarily due to overexploitation of its timber, which raises a paradox regarding its occurrence on Santay Island. This paradox can be explained by considering conditions on the island. Freed from biotic (e.g., competitors, predators, and diseases), abiotic (e.g., high salinity and waves), and anthropogenic pressures, *H. fomes* could improve its survivability on the island. This scenario has been observed in

Laguncularia racemosa in China, which, being free from salt stress and having a wide phenotypic plasticity has been able to displace native species (Gu et al. 2019; Fazlioglu & Chen 2020).

Invasive species are one of the greatest threats to biodiversity in the world. The report of new records of exotic species is important to prevent and mitigate the effects of a potential invasive species and prepare future actions to favor native biodiversity. The impact of *Heritiera fomes* on the native flora and fauna in Ecuador is uncertain and will require longitudinal studies to assess its impact on biodiversity. *Heritiera fomes* possesses several traits that qualify it as an invasive species. The environmental conditions in the Guayas River are conducive to its adaptability as they are like those of its native habitat. Although *H. fomes* is found currently only on Santay Island, favorable conditions exist throughout the Gulf of Guayaquil and suggest that further efforts are needed to locate and, if discovered, monitor the species in other areas. By doing so, we can better understand and manage its potential impact on the region's ecosystem.

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Author Contributions

Natalia Molina-Moreira: conceptualization; methodology; writing-original draft; writing-review and editing.

Roberto R. Román: conceptualization; methodology; writing-original draft; writing-review and editing.

Laurence J. Dorr: conceptualization; writing-original draft; writing-review and editing.

Efraín Freire: writing-original draft.

Álvaro Torres-Domínguez: methodology.

Conflicts of Interest

The authors declare that they have no conflicts of interest related to the publication of this manuscript.

Ethics

This study did not involve human beings and/or clinical trials that should be approved by an Institutional Committee.

Data Availability

Voucher specimens for the presence of *Heritiera fomes* in Ecuador were deposited in the National Herbarium of Ecuador (QCNE) in Quito and the U.S. National Herbarium (US) in Washington, DC. The primary data analyzed during the current study are reported in the main text as Tables 1–3. The authors confirm that all data necessary for reproducing the study findings are available in the designated dataset.

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