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Amphibians of Vassununga State Park, one of the last remnants of semideciduous Atlantic Forest and Cerrado in northeastern São Paulo state, Brazil

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Abstract: Although São Paulo state has one of the best known amphibian fauna in Brazil, there are still protected areas for which the species composition remains unknown. Here, we present the first species list of anuran amphibians in Vassununga State Park. This area is one of the last remnants of semideciduous Atlantic Forest and Cerrado in the northeastern region of São Paulo state, southeastern Brazil. To survey species, we visited six sites (two ponds, two streams, and two transects) in December, January, and February of 2014-2015 and 2015-2016, totalizing 18 days of field samplings at each site. We recorded 24 anuran species belonging to four families: Bufonidae (2 species), Hylidae (11 species), Leptodactylidae (10 species), and Microhylidae (1 species). Anurans consist mainly of generalist and widely distributed species. Although none of the species recorded are threatened with extinction according to the International Red List of Endangered Species (IUCN), four species have declining population and another three species have unknown population trends.

Keywords: Anurans, Biodiversity, Cerrado, Atlantic Forest, Species list, Conservation.

Anfíbios do Parque Estadual de Vassununga, um dos últimos remanescentes de Mata Atlântica Semidecidual e Cerrado no nordeste do estado de São Paulo, Brasil

Resumo: Embora o estado de São Paulo tenha uma das faunas de anfíbios mais bem conhecidos do Brasil, existem áreas protegidas cuja composição de espécies ainda é desconhecida. Aqui, nós apresentamos a primeira lista de espécies de anfíbios anuros para o Parque Estadual de Vassununga. A área é um dos últimos remanescentes de Mata Atlântica Semidecidual e Cerrado no nordeste do estado de São Paulo, sudeste do Brasil. Para o inventário das espécies nós visitamos seis locais (duas lagoas, dois córregos, e duas trilhas) em dezembro, janeiro e fevereiro de 2014-2015 e 2015-2016, totalizando 18 dias de amostragens de campo em cada local. Nós registramos 24 espécies de anuros pertencentes a quatro famílias: Bufonidae (2 espécies), Hylidae (11 espécies), Leptodactylidae (10 espécies) e Microhylidae (1 espécie). A anurofauna consistiu principalmente de espécies generalistas e amplamente distribuídas. Embora nenhuma das espécies registradas esteja ameaçada de extinção segundo a IUCN, quatro espécies estão em declínio populacional e outras três espécies têm tendência populacional desconhecida.

Palavras-chave: Anuros, Biodiversidade, Cerrado, Mata Atlântica, Lista de espécies, Conservação.

Introduction

Knowing the species composition of a region is important because it is the basic dataset for ecology, systematics, biogeography, and conservation biology (e.g., Collen et al. 2008; Da Silva et al. 2012, 2014). It is recognized that Brazil has an amazing amphibian diversity (1080 species; Segalla et al. 2016), with the São Paulo state harboring 22% of known species (Rossa-Feres et al. 2011). Although, the amphibian fauna of São Paulo state is one of the best known in the country, there are still considerable herpetological survey gaps in some regions (Rossa-Feres et al. 2011). These gaps are more evident inland (i.e., northwestern and northeastern regions of the state) than in the

coastal region of the state, where most surveys have been conducted (Rossa-Feres et al. 2011). For example, the increased number of anuran surveys in the last decade has contributed to records of new species in the inland areas of the state (Vasconcelos et al. 2006, Prado et al. 2008, Da Silva et al. 2009, 2010). Even protected areas such as the Vassununga State Park (created in 1970) still have not been subject to a complete faunal survey. This is worrisome because this protected area harbors one of the last remnants of Cerrado and semideciduous Atlantic Forest in the region, two biomes that are considered global hotspots for biodiversity conservation (Myers et al. 2000, Mittermeier et al. 2004). The Vassununga State Park is located in the northeastern region of the São Paulo state, which is considered one of the most deforested and fragmented regions

in the state (Rodrigues et al. 2008). The vegetation cover has been removed for the establishment of agricultural crops, pastures and urban areas, which reduced the vegetation to 9% of its original extent (Kronka et al. 1993). Here, we provide the first list of anuran species of the Vassununga State Park, a region considered to be a geographical gap in the inventories of species in the São Paulo state, Brazil.

Material and Methods

1. Study area

The Vassununga State Park (PEV; 21°43'05"S, 47°35'49"W; 553 m above sea level) is located in the municipality of Santa Rita do Passa Quatro, northeastern São Paulo state (Figure 1). It is a Protected Area with 2071.42 ha created on October 26, 1970. The region holds different physiognomies of two Brazilian biomes, Atlantic Forest and Cerrado, which are considered global hotspots for biodiversity conservation (Myers et al. 2000, Mittermeier et al. 2004). In the PEV, the Atlantic Forest is represented by the Seasonal Semideciduous Forest *sensu stricto* (Veloso et al. 1991, Pennington et al. 2006), which is characterized by having 20-50% of tree species that lose part or all their leaves in the winter or during the dry season. The Cerrado is represented by three formations (Oliveira-Filho & Ratter 2002): (i) "campo cerrado", which is formed by dry grassland scattered with shrubs and small trees; (ii) "cerrado *sensu stricto*", which is dominated by trees and shrubs that are often 3-8 m tall but still with a

fair amount of herbaceous vegetation among them; and (iii) "cerradão", which is an almost closed woodland with crown cover of 50% to 90% that is made up of trees, often of 8–12 m or even taller, that cast considerable shade so that the ground layer is greatly reduced. The climate of this region is characterized by two well-defined seasons during the year: hot and wet (September to April), during which approximately 85% of the annual rainfall occurs, and a pronounced dry season (May to August), with average precipitation of only 15% of the annual rainfall. The average annual precipitation is 1,427 mm (± 246.83 SD, <http://www.ciiagro.sp.gov.br/ciiagroonline/>).

2. Research Design and Field Methods

We sampled anurans in two ponds (TP1 = 21°43'9.83"S, 47°38'44.5"W; TP2 = 21°43'52.6"S, 47°35'3.6"W), two streams (S1 = 21°38'59.3"S, 47°38'23.1"W; S2 = 21°43'14.6"S, 47°35'45.4"W), and two transects (T1 = 21°36'35.3"S, 47°37'22.2"W; T2 = 21°43'29.8"S, 47°35'42.7"W; Figure 2) in December, January, and February of 2014-2015 and 2015-2016 totalizing 18 days of field samplings in each site. The surveys were concentrated in this period because most anuran species from this region has reproductive activity during the rainy season (Vasconcelos & Rossa-Feres 2005, Provete et al. 2011). We used surveys at breeding sites (Scott Jr. & Woodward 1994) and visual encounters (Crump & Scott Jr. 1994), two methods that are considered to be complementary (Crump & Scott Jr. 1994, Zimmerman 1994), to record the presence of anuran at these sites. For the surveys at breeding sites, we recorded calling males from

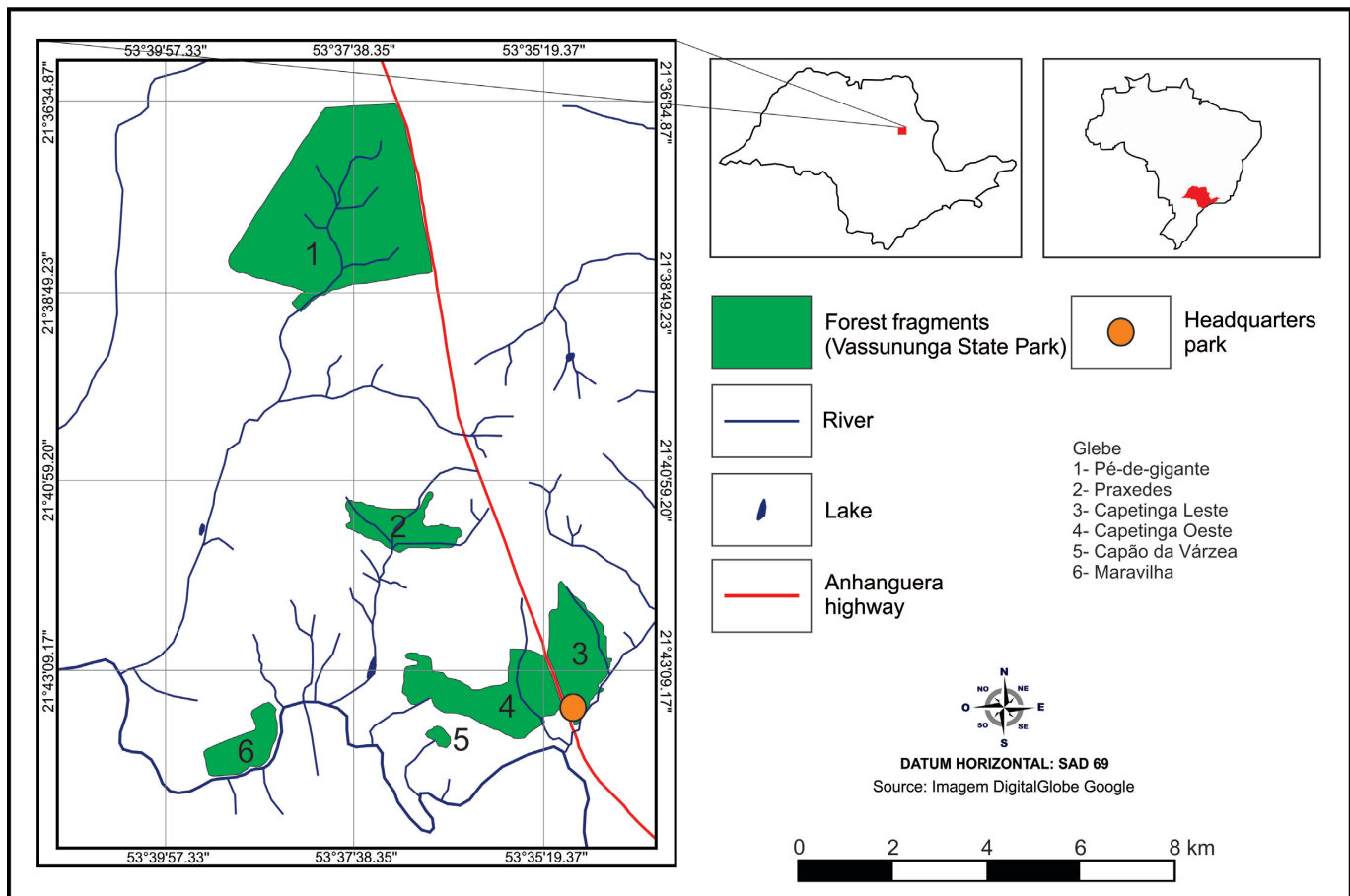


Figure 1. Maps showing the São Paulo state highlighted in Brazil (at right) and the study area highlighted in northeastern São Paulo state (at center). In detail (at left), the six isolated forest fragments (named Glebe) that comprise the total area of the Vassununga State Park.



Figure 2. Sites sampled in the Vassununga State Park: (a) temporary pond in open area (TP1; 21°43'9.83"S, 47°38'44.5"W), (b) temporary pond within gallery forest (TP2; 21°43'52.6"S, 47°35'3.6"W), (c) stream within gallery forest (S1; 21°38'59.3"S, 47°38'23.1"W), (d) stream within gallery forest (S2; 21°43'14.6"S, 47°35'45.4"W), (e) transect in the Cerradão formations (T1; 21°36'35.3"S, 47°37'22.2"W), and (f) transect in Cerradão formations (T2; 21°43'29.8"S, 47°35'42.7"W).

19 h to 24 h, while for visual encounter survey, we walked slowly inside the forest fragment (transects) or around ponds and streams for 30 min looking for individuals hidden under trunks, bromeliads, stones, branches, and leaf litter. All collected specimens were anesthetized and killed with 10% lidocaine (spray solution), fixed in 10% formaldehyde, and stored in 70% ethanol. All specimens are housed in the Coleção de Anfíbios do Departamento de Zoologia da Universidade Estadual Paulista (CFBH), municipality of Rio Claro, São Paulo state, Brazil.

3. Data Analysis

To assess the sampling efficiency of the surveys, we used a species accumulation curve (Gotelli & Colwell 2001) generated from the data of incidence of anurans during the inventory period. The total number of species recorded each day was considered as a sample, totaling 18 samples.

Considering the diverse richness estimators available, we chose to use the first-order Jackknife algorithm based on its performance when compared to other estimators (Magurran 2004, Walther & Moore 2005, Hortal et al. 2006). All analyzes were performed in R v 3.2.2 (R Core Team, 2016) using the *vegan* (Oksanen et al. 2016) and *BiodiversityR* (Kindt & Coe 2015) packages with 1,000 randomizations. Taxonomic nomenclature follows Frost (2016). The conservation status of species was obtained from the International Red List of Endangered Species (IUCN 2015).

Results and Discussion

We recorded 24 species of anuran amphibians (Table 1, Figure 3) belonging to four families: Bufonidae (2 species), Hylidae (11 species), Leptodactylidae (10 species), and Microhylidae (1 species). Although the

Table 1. Anuran species recorded at the Vassununga State Park. Sites: TP1 = temporary pond 1, TP2 = temporary pond 2, S1 = stream 1, S2 = stream 2, T1 = transect 1, T2 = transect 2. Formations: CE = “Cerradão”, GF = Gallery Forest, OA = open area. *Species with declining population according to IUCN (2015).

Family / Species	Sites	Formations
Family Bufonidae		
<i>Rhinella ornata</i> (Spix, 1824)*	T1	CE
<i>Rhinella schneideri</i> (Werner, 1894)	TP1, S1, T1	OA, GF, CE
Family Hylidae		
<i>Dendropsophus elianeae</i> (Napoli & Caramaschi, 2000)	TP1	OA
<i>Dendropsophus jimi</i> (Napoli & Caramaschi, 1999)	TP1	OA
<i>Dendropsophus minutus</i> (Peters, 1872)	TP1	OA
<i>Dendropsophus nanus</i> (Boulenger, 1889)	TP1, TP2	OA, GF
<i>Hypsiboas albopunctatus</i> (Spix, 1824)	TP1, TP2, S2	OA, GF
<i>Hypsiboas faber</i> (Wied-Neuwied, 1821)	TP1	OA
<i>Hypsiboas lundii</i> (Burmeister, 1856)*	T1, S1	CE, GF
<i>Itapotihyla langsdorffii</i> (Duméril & Bibron, 1841)*	TP2	GF
<i>Scinax fuscovarius</i> (A. Lutz, 1925)	TP1, S2	OA, GF
<i>Scinax similis</i> (Cochran, 1952)	TP1, S2	OA, GF
<i>Trachycephalus typhonius</i> (Linnaeus, 1758)	TP1, TP2	OA, GF
Family Leptodactylidae		
<i>Leptodactylus fuscus</i> (Schneider, 1799)	TP1, T2	OA, CE
<i>Leptodactylus labyrinthicus</i> (Spix, 1824)	S2	GF
<i>Leptodactylus latrans</i> (Steffen, 1815)	TP1	OA
<i>Leptodactylus mystaceus</i> (Spix, 1824)	TP2	GF
<i>Leptodactylus mystacinus</i> (Burmeister, 1861)	TP1	OA
<i>Leptodactylus podicipinus</i> (Cope, 1862)	TP1	OA
<i>Physalaemus centralis</i> Bokermann, 1962	TP1	OA
<i>Physalaemus cuvieri</i> Fitzinger, 1826	TP1	OA
<i>Physalaemus marmoratus</i> (Reinhardt & Lütken, 1862)	TP1	OA
<i>Physalaemus nattereri</i> (Steindachner, 1863)*	TP1	OA
Familia Microhylidae		
<i>Elachistocleis cesarii</i> (Miranda-Ribeiro, 1920)	TP1	OA

species accumulation curve is close to achieving asymptote, the observed species richness was smaller than the estimated species richness, indicating that more species could be recorded if we increase the sampling effort or utilize other sampling methods (e.g., pitfall traps, Figure 4).

Since Vizotto (1967), 37 anuran species have been recorded in the northwestern region of the São Paulo state (Provete et al. 2011). In this study, we recorded approximately 65% of all species that occur in the region. Only *Hypsiboas albopunctatus* had already been recorded at the study site (Toledo et al. 2007). The species richness observed in this study was similar to those found both in areas with predominance of Cerrado formations such as Estação Ecológica Assis (23 species; Ribeiro-Júnior & Bertoluci 2009), E. E. Itirapina (28 species; Brasileiro et al. 2005), E. E. Jataí (21 species; Prado et al. 2009), and E. E. Santa Bárbara (33 species; Araujo et al. 2013), and areas with a predominance of seasonal semideciduous forest such as Floresta Estadual Edmundo Navarro (21 species; Toledo et al. 2003), E. E. Caetetus (24 species; Bertoluci et al. 2007; 34 species; Brassaloti et al. 2010), and Parque Estadual Morro do Diabo (28 species; Santos et al. 2009). Although none of the recorded species is currently threatened with extinction, three species (*Dendropsophus elianeae*, *Physalaemus centralis*, and *P. marmoratus*) have unknown population trends, and four species (*Rhinella ornata*, *Hypsiboas lundii*, *Itapotihyla langsdorffii*, and *Physalaemus nattereri*) have declining population (IUCN 2015). Furthermore, Vasconcelos & Doro (2016) showed that habitat loss has negative impacts in the geographic ranges of *R. ornata*, *H. lundii*, and *I. langsdorffii*. This fact becomes even more worrying considering that northeastern region of the São Paulo state has one of highest deforestation and fragmentation rates in the state (Rodrigues et al. 2008).

Approximately 50% of the species (*Dendropsophus elianeae*, *D. jimi*, *D. minutus*, *Hypsiboas faber*, *Leptodactylus latrans*, *L. mystacinus*, *L. podicipinus*, *Physalaemus centralis*, *P. cuvieri*, *P. marmoratus*, *P. nattereri*, and *Elachistocleis cesarii*) occurred exclusively in ponds in open areas, while 10 species (*Rhinella schneideri*, *Dendropsophus nanus*, *Hypsiboas albopunctatus*, *H. lundii*, *Itapotihyla langsdorffii*, *Scinax fuscovarius*, *S. similis*, *Trachycephalus typhonius*, *Leptodactylus labyrinthicus*, and *L. mystaceus*) occurred in streams and ponds. *Hypsiboas lundii* was the only species found vocalizing exclusively on streams inside gallery forest (Table 1). The large number of anuran species associated with water bodies in Cerrado and semideciduous forest fragments during the reproductive period has been demonstrated in other studies (Brasileiro et al. 2005, Bertoluci et al. 2007, Araujo et al. 2009, Santos et al. 2009, Araujo & Almeida-Santos 2011, 2013, Valdujo et al. 2012). According to Santos et al. (2009), anuran communities from Semideciduous Atlantic Forest and Cerrado environments are more similar to each other than they are to those of Ombrophilous Forest environments. This pattern of similarity in anuran communities can be interpreted as a result of the seasonally dry climate that most likely limits the occurrence of anuran species typical of the Ombrophilous Atlantic Forest in Semideciduous Atlantic Forest and Cerrado areas (Santos et al. 2009, Da Silva et al. 2012).

Although several authors have shown that the anuran species of the Cerrado and semideciduous forest fragments are mostly associated with open areas (e.g., Brasileiro et al. 2005, Araujo et al. 2009, Santos et al. 2009, Araujo & Almeida-Santos 2011, 2013, Valdujo et al. 2012), here we recorded 50% of species (12 species) occurring in forest areas. It should be emphasized that forests are important mesic environments, being used by adults and juveniles as refuge sites and for foraging, hibernation, and



Figure 3. Anuran species recorded in the Vassununga State Park, northeastern São Paulo state, Brazil. In brackets, for each voucher specimen we provide the acronym of the Coleção de Anfíbios do Departamento de Zoologia da Universidade Estadual Paulista, municipality of Rio Claro (CFBH) followed by the registration number and the snout-vent length (SVL). a = *Rhinella ornata* (CFBH 40455; SVL 38.18 mm), b = *R. schneideri* (CFBH 38871; SVL 124.39 mm), c = *Dendropsophus elianae*, d = *D. jimi* (CFBH 40456; SVL 24.03 mm), e = *D. minutus*, f = *D. nanus* (CFBH 38872; SVL 19.07 mm), g = *Hypsiboas albopunctatus* (CFBH 38856; SVL 52.74 mm), h = *H. faber*, i = *H. lundii* (CFBH 38870; SVL 64.8 mm), j = *Itapotihyla langsdorffii*, k = *Scinax fuscovarius* (CFBH 38862; SVL 39.23 mm), l = *S. similis* (CFBH 40455; SVL 38.18 mm), m = *Trachycephalus typhonius* (CFBH 38866; SVL 31.87 mm), n = *Leptodactylus fuscus* (CFBH 38869; SVL 33.98 mm), o = *L. labyrinthicus*, p = *L. latrans*, q = *L. mystaceus*, r = *L. mystacinus* (CFBH 38859; SVL 55.95 mm), s = *L. podicipinus*, t = *Physalaemus centralis* (CFBH 40457; SVL 33.68 mm), u = *P. cuvieri* (CFBH 38912; SVL 28.71 mm), v = *P. marmoratus* (CFBH 38865; SVL 37.05 mm), w = *P. nattereri* (CFBH 38867; SVL 41.48 mm), x = *Elachistocleis cesarii*. Photos: Ronildo Alves Benício.

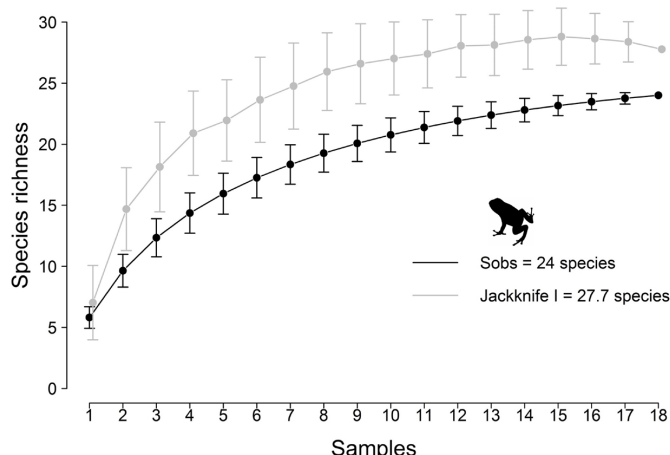


Figure 4. Species accumulation curve (black line) and Jackknife algorithm first order (gray line) representing the anuran species richness observed and estimated, respectively, based on 18 samples from December 2014 to February 2016. The dots show the mean generated by 1000 randomizations and the vertical bars indicate the standard deviation.

migration, especially during dry periods (Da Silva & Rossa-Feres 2007). This study area is one of the most deforested and fragmented in the state, and because of the lack of biological knowledge, it is classified as a priority for fauna and flora inventories (Rodrigues et al. 2008, Rossa-Feres et al. 2011). In this sense, the species list of anurans for Vassununga State Park represents an important step in increasing the knowledge about the distribution of diversity of São Paulo state.

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Author Contributions

RAB and FRS contributed in the acquisition, analysis, data interpretation and writing of the work.

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