Habitat use and reproductive activity of anurans from a Cerrado area in Minas Gerais state, southeastern Brazil

Bruno F. Fiorillo¹*, Renato C. Nali², and Cynthia P. A. Prado³,⁴

Abstract. The Brazilian Cerrado is one of the 25 most important biodiversity hotspots, considered the most threatened savanna on earth. Amphibians are especially vulnerable to habitat modifications due to behavioural and physiological traits. Herein, we list anuran species of a Cerrado area in the municipality of Sacramento, Minas Gerais State, Brazil, after 12 months of efforts between 2010 and 2011. Also, we provide information about habitat use and reproductive activity based on field surveys, where we registered habitat occupation and temporal distribution of the anurans at the study site. A total of 20 species belonging to seven families were recorded; two species are considered Data Deficient by the IUCN Red List of Threatened Species, two have unreported status, and one is considered Critically Endangered. Moreover, seven of the 20 species need status updating. We highlight that fragments in altered areas are crucial to maintain amphibian diversity, especially for species with restricted distributions, taxonomic uncertainties and/or that require more information on their conservation status.

Keywords. Amphibians, Neotropics, Inventory, Conservation, community

Introduction

The Cerrado is the Brazilian second largest morphoclimatic domain, covering about 2 million km² in central, western and southwestern regions of the country (Ratter et al., 1997). The vegetation cover varies in structure and composition (Furley and Ratter, 1998), with different phytophysigonomies, from semideciduous forests to grass fields, including gallery forests along the streams and rivers (Eiten, 1972; Ratter et al., 1997). This domain is considered one of the 25 most important terrestrial hotspots, encompassing areas of great biodiversity with a high level of endemism (Mittermeier et al., 2000). In the last decades, however, the Cerrado has been intensively modified, especially for agriculture practices and cattle ranching (Ratter et al., 1997; Klink and Machado, 2005). Only 20% of its original area still remains, of which less than 9% is protected (Mittermeier et al., 2000; MMA, 2018), being considered the most endangered savannah of the world (Silva and Bates, 2002).

The diversity of anuran species in the Cerrado is high, with about 209 described species, in which ca. 51% (108 species) are endemic (Valdujo et al., 2012). Although studies on species composition and anuran activity patterns in southeastern Cerrado has increased (Canelas and Bertoluci, 2007; Ribeiro-Júnior and Jaime Bertoluci, 2009; Maffei et al., 2011; Araujo et al., 2013; Pirani et al., 2013), some regions remain poorly known, hampering proper conservation practices. Thus, new studies that portray not only the species composition of a given area (check lists), but also their relationships with biotic and abiotic factors are essential for conservation and insights into community ecology.

The environmental heterogeneity of the Cerrado has been considered as an important factor in the determination of anuran diversity, since the mosaic of adjacent habitats (e.g., open vs. forested habitats, humid vs. dry environments) create a gradient of resources.
(e.g., microhabitats; Oda et al., 2009; Campos et al., 2013; Conte et al., 2013). In addition to the influence of environmental structure on the spatial distribution of anurans, the seasonal climate of the Cerrado also affects their temporal distribution, in which most species reproduce during the rainy season (Rodrigues et al., 2007; Giaretta et al., 2008; Oda et al., 2009). It is known that the different habitats may hold up anuran population in different life stages and their disconnection can lead to population declines (habitat-split, see Becker et al., 2007). Thus, basic patterns of diversity and distribution of amphibian populations need to be uncovered. Managers can then use this information to balance metapopulation considerations and habitat quality to establish effective conservation policies (Marsh and Trenham, 2001).

In this study we (i) gathered data on the species richness, temporal and spatial distribution of an anuran community from the Cerrado in southeastern Brazil, and (ii) compared our results with those from other studies conducted in nearby areas.

Materials and Methods

Study Area.—The present study was carried out in the municipality of Sacramento (19.85°S, 47.43°W, Fig. 1), southwestern Minas Gerais state, southeastern Brazil, in farms located between Peixoto dam and Serra da Canastra National Park. The landscape is composed by mountains, flat and rounded tops and steep slopes with cuestas, canyons, and latosols are the predominant soil type. The main phytophysionomies are cerradão, cerrado, interfluvial fields, gallery forests and semideciduous forest (Araujo et al., 2009). The influence of other biomes, such as Atlantic rainforest is apparently low (Romero and Martins, 2002). The climate is classified as mesothermal with a dry winter (Araujo et al., 2009) and rainy season typically occurs from September to March (Nakajima and Semir, 2001).

![Figure 1. Sampling localities of the anuran surveys. (A) Topographic map with the geographic location of the municipality of Sacramento (white star), Minas Gerais state, southeastern Brazil; (B) Map showing the four sampling localities for the anuran surveys (I, II, III and IV), with their general aspects. Satellite image extracted from Google Earth (CNES/Airbus 2018).](image-url)
During the year of sampling, mean air temperatures ranged from 17.5–23.6°C and accumulated monthly rainfall from 0.2–540 mm; the mean annual air temperature was 21.5°C and the accumulated annual rainfall was 1796 mm. These data were provided by the automatic meteorological station of the municipality of Sacramento, Minas Gerais state, monitored by the Instituto Nacional de Meteorologia (INMET, 2011).

Field Activities.—Given that environmental heterogeneity has a significant influence on the composition of anuran communities (Santos et al., 2007; Silva and Rossa, 2011), we tried to sample sites with variable characteristics. We sampled four different areas (see Fig. 1; Table 1): I) gallery forest along a permanent stream, but also permanent and temporary ponds nearby (20.2725°S, 47.0733°W; 677 m a.s.l.); II) forest fragment containing a temporary stream (20.2192°S, 47.1058°W; 880 m a.s.l.); III) open area which remained flooded during the rainy season, with an adjacent forest fragment (20.2056°S, 47.1316°W; 839 m a.s.l.); IV) wet field, near an artificial temporary pond (20.2148°S, 47.1320°W; 816 m a.s.l.).

We sampled during an entire year from September 2010 to August 2011 (except for March), twice a month in the rainy season (October to February) and once a month during the dry season (April to September), mainly between 19:00h and 02:00h. We sampled at least two nights each month, totalling 105 h/man. Our sampling methods were active search for individuals at breeding sites, as well as indirect sampling guided by males’ vocalisation and occasional encounters (Heyer et al., 1994). Species reproductive period was determined based on the following field observations: a) vocalising males; b) pairs in amplexus; c) clutches and/or tadpoles. Habitat use of adults was recorded at each encounter, including type of habitat (forested vs. open area) and microhabitat (type of substrate, type of water body - temporary or permanent pond or stream). The taxonomic nomenclature used in the present study is according to Frost (2019).

Besides field observations, we hand-captured some individuals of most species as vouchers, anesthetized with lidocaine 10%, fixed in formalin 10%, and preserved in alcohol 70% (Heyer et al., 1994). Individuals were deposited at the Coleção de Anfíbios Célio F. B. Haddad (CFBH), Departamento de Zoologia, I.B., Universidade Estadual Paulista, Rio Claro, São Paulo state, Brazil (Appendix 1).

Results
We recorded 20 anuran species belonging to seven families in the municipality of Sacramento, Minas Gerais state: Bufonidae (2 species), Brachycephalidae (1), Dendrobatidae (1), Hylidae (8), Leptodactylidae (6), Microhylidae (1), Phyllomedusidae (1) (for details see Table 2; Figs. 2 and 3). Hylidae accounted for the highest number of species (40%), followed by Leptodactylidae (30%) and Bufonidae (10%); other combined families accounted for 20% of the species. According to the IUCN (International Union for Conservation of Nature) Red List of threatened species, 14 of the species sampled here were classified as Least Concern (LC), two species classified as Data Deficient (DD), one as Critically Endangered (CR), and two have

Table 1. Description of the sampled sites, municipality of Sacramento, Minas Gerais state, southeastern Brazil (see Fig. 1).

<table>
<thead>
<tr>
<th>Site</th>
<th>Coordinates</th>
<th>Elevation (m)</th>
<th>Vegetation</th>
<th>Water bodies</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>20.2725°S, 47.0733°W</td>
<td>677</td>
<td>Gallery forest with leaf litter, grasses, many bushes, and trees up to 15 m height</td>
<td>Permanent stream with permanent and temporary ponds nearby formed by the main water course</td>
</tr>
<tr>
<td>II</td>
<td>20.2192°S, 47.1058°W</td>
<td>880</td>
<td>Forest fragment composed mostly by short trees and bushes</td>
<td>Temporary stream that formed a flooded area, as well as flowing water around one of the margins</td>
</tr>
<tr>
<td>III</td>
<td>20.2056°S, 47.1316°W</td>
<td>839</td>
<td>Open area covered mostly by grasses and some bushes, which remained flooded during the rainy season; adjacent forest fragment similar to site I</td>
<td>Temporary ponds and stream that formed many smaller water bodies within the forest fragment</td>
</tr>
<tr>
<td>IV</td>
<td>20.2148°S, 47.1320°W</td>
<td>816</td>
<td>Cattle pasture that remained flooded during most of the year</td>
<td>Swamp and artificial temporary pond (10 x 15 m)</td>
</tr>
</tbody>
</table>
Figure 2. Some anurans found at the municipality of Sacramento, Minas Gerais state, southeastern Brazil, from the families Hylidae and Phyllomedusidae. (A) *Boana albopunctata*; (B) *Boana faber*; (C) *Boana lundii*; (D) *Bokermannohyla ibitiguara*; (E) *Dendropsophus elianeae*; (F) *Dendropsophus minutus*; (G) *Scinax fuscovarius*; (H) *Pithecopus ayeaye*. 
Figure 3. Some anurans found at the municipality of Sacramento, Minas Gerais state, southeastern Brazil, from the families Bufonidae, Brachycephalidae, Leptodactylidae and Microhylidae: (A) *Rhinella rubescens*; (B) *Rhinella diptycha*; (C) *Ischnocnema cf. penaxavantinho*; D) *Physalaemus nattereri*; (E) *Pseudopaludicola murundu*; (F) *Leptodactylus fuscus*; (G) *Leptodactylus labyrinthicus*; (H) *Elachistocleis cesarii*. 
unreported status (see Table 2; IUCN, 2017).

Most species were observed in open habitats or at the edge of forests during the rainy season (sites III and IV; Tables 1 and 3). The exceptions were _Bokermannohyla ibitiguara_, _Boana lundii_, _Ischnocnema cf. penaxavantinho_, and _Ameerega flavopicta_, which were found mostly in forest habitats, mainly associated with streams (Table 2). We also observed juveniles of _A. flavopicta_ near streams. Individuals of _Boana faber_ occurred in both open and forested environments, but we did not observe males in calling activity. We considered the species of the family Hylidae as the only truly arboreal, which were mostly found perched on branches of trees or shrubs at a height higher than one meter. We considered the remaining species as terrestrial, using the ground or rocks along streams as substrates for their activities. Most species from different families used mainly lentic water bodies, whereas _Ameerega flavopicta_, _Boana lundii_, _Bokermannohyla ibitiguara_ and _Ololygon canastrensis_ used lotic environments. Most species were active during the rainy season at the study site (Table 3).
The diversity of species found here can be considered representative of the anuran fauna of the Cerrado domain when compared with other areas that were intensively sampled (Haddad et al., 1988; Brasileiro et al., 2005; Barros, 2011; Pirani et al., 2013). The number of anuran species recorded here was slightly lower than that from the protected area of Furnas do Bom Jesus State Park (PEFBJ; 24 species; ca. 40 km away from the study area; Araujo et al. 2009), and lower than that from the Serra da Canastra National Park (PNSC; 38 species; ca. 120 km away from the study area; Haddad et al., 1988; Barros, 2011), which is a much larger conservation unit. This shows the importance of the rural area of the municipality of Sacramento in terms of anuran diversity and its representativeness within the Cerrado.

All anuran families found here were also recorded in the protected areas PNSC and PEFBJ (Bufonidae, Brachycephalidae and Dendrobatidae - only for PNSC, Hylidae, Leptodactylidae, Microhylidae and Phyllomedusidae). However, for instance, Centrolenidae and Hylidae were recorded for the PNSC, and Cycloramphidae and Craugastoridae were recorded in the PEFBJ (Haddad et al., 1988; Araujo et al., 2009; Barros, 2011). In the Cerrado there are only two records of the family Centrolenidae, both within riparian forests and rocky fields in Minas Gerais state: Serra do Cipó National Park (Eterovick and Sazima, 2004) and PNSC (Haddad et al., 1988). Centrolenids are generally associated with tropical forests (Wells, 2007), thus, the low abundance of this family in the Cerrado may have contributed to its absence in our study area. The family Hylodidae is composed by rheophilic species, mainly associated with the Brazilian Atlantic Forest (Lingnau et al., 2008; Pimenta et al., 2008; Verdade and Rodrigues, 2008; Laia and Rocha, 2012) and thus is rare or absent in the Cerrado domain. Other reasons may explain the absence of some species of Craugastoridae and Cycloramphidae found in PEFBJ (Barycholos ternetzi and Odontophrynus cultripes, respectively). Although Barycholos ternetzi is widely distributed throughout the Cerrado, it is associated with deciduous and semi-deciduous montane forests (Araujo et al., 2009), which differ from the environments sampled here (mainly gallery forests and open wet fields). Odontophrynus cultripes presents explosive reproduction, and was
observed in the PEFBJ only after heavy rains (Araujo et al., 2009). Such events occurred a few times throughout the year in the study area, making it difficult to register explosive species.

Most species occurred and reproduced in localities II and III, which are small forest fragments (Table 2; Fig. 1). These fragments were frequently humid and presented a great number of potential reproductive sites even during the dry season (April to August). In preserved habitats, adults can easily access suitable aquatic breeding sites and metamorphs can move into suitable terrestrial habitats within a few meters from aquatic breeding sites (Becker et al., 2009). However, in highly fragmented landscapes, water bodies and forest fragments can become disconnected by unsuitable matrix, contributing to interruption of natural life-cycle and decline of amphibian populations in disturbed regions (Becker et al., 2007). Thus, our results suggest that these small fragments, even if disturbed, might be important in maintaining the diversity and reproductive cycle of anurans in this region.

High rainfall and temperature usually have a positive effect on the reproductive activity of amphibians (Salvador and Carrascal, 1990; Donnelly and Guyer, 1994; Zina et al., 2007). In our study, most species were reproducitively active during the rainy season (Table 2), and their reproductive activity was markedly seasonal. This is expected mainly in environments that present well-delimited dry and rainy seasons, such as the study area (Dietz, 1984; Prado et al., 2005, Oda et al., 2009). Most species used temporary ponds to reproduce in the study area, so abiotic factors such as a decrease in precipitation levels and air temperature may be related to the interruption of the reproductive activity of these species during the dry season (Zina et al., 2007; Oda et al., 2009).

Some species found here show restricted distributions and/or taxonomic problems, which reinforce the relevance of our new records. *Ischnocnema penaxavantinho* is endemic to the Cerrado, previously known only from its type-locality (municipality of Uberlândia, Minas Gerais State, southeastern Brazil; Giaretta et al., 2007; Giaretta, 2008). More recently, the species was found at the Silvânia National forest (Goiás State), extending its distribution ca. 260 km northwards (Bastos et al., 2015), and in the municipalities of Presidente Olegário (ca. 230 km from the type-locality: Assis et al., 2018) and Paracatu (ca. 270 km from the type-locality; Del Prette et al., 2018), both in Minas Gerais State. This is a cryptic species and can be easily confused with the congener *I. juipoca*, from which is mostly distinguishable only through its advertisement call (Bastos et al., 2015). Our record of *Ischnocnema cf. penaxavantinho* at the study area could potentially expand its distribution in approximately 185 km to the southeast of the type-locality, but only if confirmed in future studies. That includes further sampling of this locality to collect more specimens and registering their advertisement calls.

*Pseudopaludicola murundu* was described from the municipality of Rio Claro, São Paulo state, southeastern Brazil (Toledo et al., 2010a), and its geographic distribution was recently expanded to the Espinhaço mountain range, in Minas Gerais state, after synonymization with *Pseudopaludicola serrana* (Pansonato et al., 2014). Thus, besides São Paulo state, the species is currently known from other three localities in Minas Gerais state, all above 1000 m altitude: (1) Serra do Cipó, municipality of Santana do Riacho, (2) Serra da Moeda, municipality of Brumadinho (*P. serrana* type-locality), and (3) Serra do Lenheiro, municipality of São João del Rei (Pansonato et al., 2014). Thus, we are expanding its distribution to another important mountain range in Minas Gerais state, the Serra da Canastra (ca. 250 km from Rio Claro, São Paulo state, and 300 km from São João del Rei, Minas Gerais state), at an intermediate elevation (ca. 880 m) between its type-locality and the Espinhaço mountain range.

*Elachistocleis cesarri* was re-described based on specimens collected in São Paulo state, and its geographic distribution was expanded to Minas Gerais and Goiás states (Toledo et al., 2010b). The species is found in open areas during the rainy season (Toledo et al., 2010b), as observed in the present study. From our collected individuals, some diagnostic characteristics were observed, such as white or yellow ventral staining marks, the posterior post-commissural glands in the corner of the mouth, and hidden tympanum (Toledo et al., 2010b). However, the taxonomic status of *E. cesarri* remains unclear, in light of new evidence that some species of the genus may be synonyms of *E. cesarri* (Loredam, 2015), which limits the recognition of several species not described in South America. Our record and collection of *E. cesarri* individuals may contribute to the unveiling of its uncertain taxonomic status. In addition, both *P. murundu* and *E. cesarri* are not accounted for on the IUCN website (IUCN, 2017).

Considering the IUCN Red List, out of the 14 species recorded here and classified as Least Concern, five need status updates (Table 2). Moreover, two species are classified as Data Deficient (*Bokermannohyla ibitiguara* and *Ololygon canastrensis*), which means
that their conservation status classification depends upon information on the populations’ status (Pimenta et al., 2005; Nali and Prado, 2012). According to Howard and Bickford (2014), Data Deficient species are more likely to suffer extinction, because it is unknown whether these species are actually threatened or not. *Pithecopus ayeaye* is classified as Critically Endangered according to the IUCN, but due to recent reports of its occurrence in different localities and its synonymization with *P. itacolomi*, the species has been removed from the threatened category in the Brazilian List of Endangered Species (Baêta et al., 2009; ICMBio, 2018). However, how its fragmented distribution impacts its genetic diversity was not considered, as this species contains at least three different Evolutionary Significant Units (Magalhães et al., 2017).

Although we found fewer species than others surrounding localities (PNSC and PEFBJ), our survey showed a relatively high richness that is representative of the anurofauna from the Cerrado domain. Furthermore, this was the first survey for the municipality of Sacramento, Minas Gerais state, southeastern Brazil. These primary data are one of the most important tools in decision-making, regarding the management of natural areas (Silveira et al., 2010). Since the study area has been impacted mainly by agricultural activities and cattle ranching, the data generated here should be considered in the decisions of future projects that may impact the local environment, as well as those that encompass the Serra da Canastra National Park. We highlight that even small, disturbed fragments are crucial to maintain the diversity and reproduction of anurans, especially of those species with restricted distribution and that require more information on their conservation and taxonomic status (Pimenta et al., 2005; Howard and Bickford, 2014; IUCN, 2017).

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**References**


biology of *Phyllomedusa azurea* Cope, 1862 and *P. sauvagii* Boulenger, 1882 (Anura) from the Cerrado, Central Brazil. Journal of Natural History 41: 1841–1851.


### Appendix 1.
Specimens collected during this study (families in bold), deposited in the Coleção de Anfíbios Célio F. B. Haddad (CFBH), Universidade Estadual Paulista, Rio Claro, São Paulo state, Brazil. Specimens are followed by their voucher numbers.

**Bufonidae:** *Rhinella rubescens* (CFBH 34381);

**Dendrobatidae:** *Ameerega flavopicta* (34347);

**Hylidae:** *Boana albopunctata* (34293, 34315, 34318, 34320, 34330, 34342, 34361), *Boana faber* (34365), *Boana lundii* (34305-34307, 34316, 34317, 34322, 34329, 34341, 34346, 34349, 34350, 34353-34355, 34364, 34375, 34380, 34388), *Bokermannohyla ibitianguara* (31743-31771), *Dendropsophus elianeae* (34324, 34325), *Dendropsophus minutus* (34300, 34301, 34326, 34332, 34340, 34348, 34351, 34362, 34374), *Scinax fuscovarius* (34295, 34296, 34303, 34331, 34338, 34339, 34352);


**Microhylidae:** *Elachistocleis cesarrii* (34294, 34297-34299, 34302, 34367, 34368); *Phyllomedusidae:* *Pithecopus ayeaye* (34311-34313).