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RESEARCH ARTICLE

Annual and daily patterns of calling activity in male *Scinax fuscomarginatus* (Anura: Hylidae) from Central Brazil

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ABSTRACT. Bioacoustics is an effective way of recording detailed data during population surveys and monitoring. In the present study, we used an automated digital recorder (ADR) to describe the temporal variation in the calling activity of *Scinax fuscomarginatus* (Lutz, 1925) in central Brazil. We also evaluated the role of climatic variables (air temperature and precipitation) on calling activity by using a Generalized Additive Model (GAM). We conducted the recordings at five ponds in the Cerrado savanna of Rio Verde Municipality, in Goiás state between November 2013 and October 2014. The analysis of the 43.2 hours of acoustic recording showed that *S. fuscomarginatus* has a prolonged breeding pattern. The ADR provides a fine-scale description of the nocturnal calling pattern, as well as the oscillations between the rainy and dry seasons. The results of the analytical model also indicate that calling patterns were related to minimum (but not maximum) air temperatures and precipitation, which may be related to their reproductive and thermoregulatory requirements. Based on these findings, we conclude that the ADR method has potentially valuable applications for the collection of data on the natural history of anuran species, as well as supplying important insights for conservation initiatives.

KEY WORDS. Acoustic survey, anurans, automated recording, breeding period, phenology, seasonal variability, vocalization behavior.

INTRODUCTION

Acoustic communication is an important component of the reproductive behavior and social interactions of frogs. The acoustic signals emitted by these amphibians are often a prominent part of the attraction of potential mates, territorial defense, and the recognition of conspecifics (Gerhardt and Huber 2002). These characteristics favor the use of acoustic signals in surveys of anuran populations. Different techniques are used to survey and monitor anurans in the field. The most common methods are drift fences, pitfall traps, visual encounter and manual calling surveys, audio strip transects, and automated recordings (Heyer et al. 1994). The automated recording of acoustic signals (or Automated Digital Recording, ADR) has become increasingly popular in recent years for the surveying of anurans (Dorcas et al. 2009, Sugai et al. 2018). Although this technique does not allow the researcher to detect some important aspects of anuran reproduction (e.g., the presence of non-calling females and egg masses), it can provide valuable information on the presence of a particular species and detect interspecific variability in occurrence patterns (Bridges and Dorcas 2000, Schalk and Saenz 2016), as well as reducing considerably sampling effort in the field (Hsu et al. 2005, Madalozzo et al. 2017). The principal advantage of this technique is that it permits the collection of data continuously over prolonged periods, that is, a number of consecutive days, as well as the possibility of implementing automatic species identification procedures (Acevedo and Villanueva-Rivera 2006). In general, an ADR also increases the probability of detecting acoustic signals in the field.

The acoustic behavior of many Neotropical anurans varies seasonally, in particular in species that occur in highly seasonal environments (e.g., Prado et al. 2005, Kopp et al. 2010, Andrade et al. 2019, Ulloa et al. 2019). This behavioral flexibility provides important cues for the identification of the breeding season, and the link between environmental conditions, such as air temperature and precipitation, and activity patterns (Wells 2007). This is especially the case for the anurans of the Brazilian Cerrado (Colli et al. 2002). The anurans of this savanna biome are either explosive breeders, reproducing during a few days or weeks, or prolonged breeders (sensu Wells 1977), breeding more or less continuously over five or more months during the rainy season (Kopp et al. 2010). However, few studies of Cerrado anurans have used automated recording for long-term monitoring (see Guerra et al. 2020, Ramalho et al. 2020), despite its potential for the collection of high-resolution temporal data on anuran calling activity, which can be analyzed systematically in the context of climatic variables (e.g., air temperature and precipitation) to evaluate the influence of environmental conditions on vocalisation patterns.

Here, we describe the temporal calling patterns (daily and seasonal) of *Scinax fuscomarginatus* (Lutz, 1925) from data collected using an ADR. We also evaluate whether the annual pattern is related to air temperature and precipitation. *Scinax fuscomarginatus* has an ample distribution in Brazil (Brusquetti et al. 2014), occurring in open formations typical of the Cerrado and Pantanal biomes (Brasileiro et al. 2005, Toledo and Haddad 2005a, 2005b). Previous descriptions of the calling behavior of this species were based on visual encounters and manual recordings (Pombal-Jr 1997, Bernarde and Kokubum 1999, Toledo and Haddad 2005a). These studies defined the reproductive pattern as prolonged (sensu Wells 1977) with calling being recorded between sunset and midnight. However, these descriptions did not focus on the full annual cycle, which may have affected the resolution of the analysis of the anuran calling patterns (e.g., Bridges and Dorcas 2000).

MATERIAL AND METHODS

We conducted fieldwork at five permanent ponds in the municipality of Rio Verde (17°47’52”S; 50°55’40”W), southwestern Goiás, central Brazil. The principal types of vegetation at the sampling site were shrubby grassland (campo sujo) and shrubby grassland with trees (campo cerrado), which are typical of the Cerrado biome. The surrounding area presents a diversity of land uses, such as pasture and soybean plantations. The local climate is Aw (tropical wet savanna) in Köppen’s classification system, with annual precipitation of 1600–1900 mm and mean annual air temperatures of 22–24 °C (Alvares et al. 2014). The rainy season occurs typically between October and March. Minimum and maximum air temperatures (°C), and precipitation (mm) were obtained from a weather station 15 km from the study site.

We recorded the calling activity of *S. fuscomarginatus* males between November 2013 and October 2014, by collecting acoustic data automatically on three consecutive days per month. We installed an automated digital recorder (ADR), as described by Madalozzo et al. (2017), at each pond. The recorders were Sony ICD-PX312 (48 kbps and MP3 format).

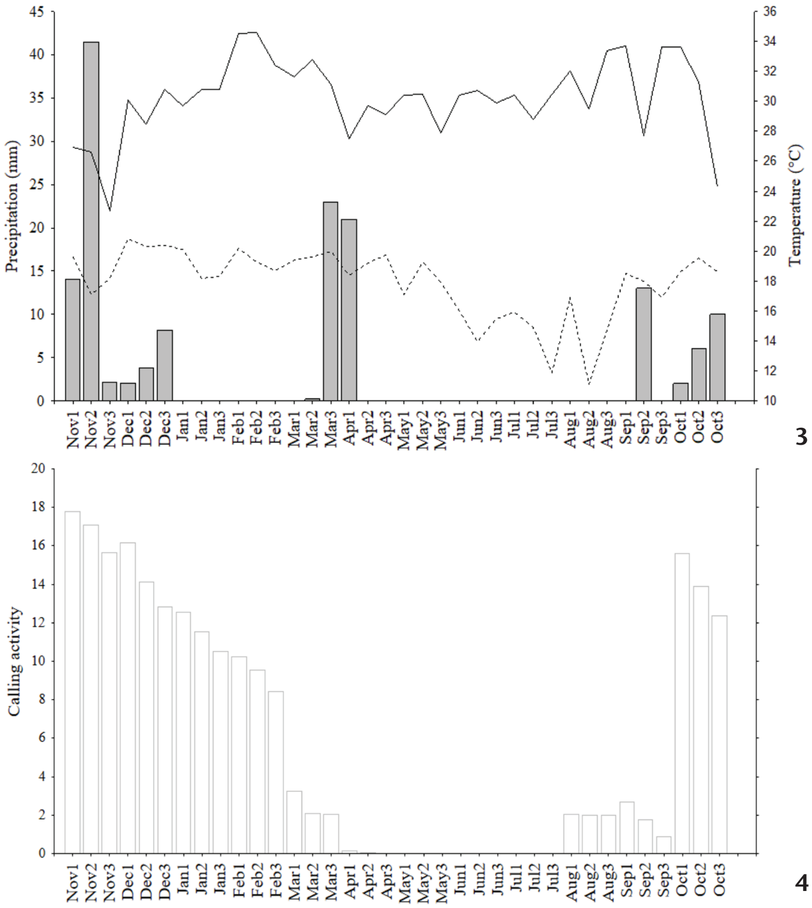
We estimated the maximum detection distance (Llusia et al. 2011) to be approximately 50 m. At each pond, we installed a recorder that remained active for 72 hours, that is, during three consecutive days and nights. At the end of this period, we removed the recorder and downloaded the data, and only installed the apparatus again the following month. With three days per month at each pond, total sampling effort was 864 hours (51,840 minutes). The recorders were installed in trees or shrubs at the margin of each pond, at approximately 50 cm above the ground, and protected by a plastic casing (Figs 1, 2). We used Audacity (Audacity Team 2019) to extract the nocturnal portion of the recordings (sensu Madalozzo et al. 2017). We then estimated the number of advertisement calls (calling activity) by listening to six-minute samples selected randomly from each hour of recording, obtained between 6:00 pm to 6:00 am, and counting the number of calls emitted during each sample. The calls of syntopic anuran species could be distinguished easily from the advertisement calls of *S. fuscomarginatus* in the recordings. This sampling procedure resulted in a total of 43.2 hours (2,592 minutes) of analyzed recordings. The call terminology followed Toledo and Haddad (2005a). The raw data were deposited at the Sound Files of Neotropical Anurans Collection (CASAN: Coleção de Arquivos Sonoros de Anuros Neotropicais) of the Federal Institute of Goiás in Rio Verde.

We calculated the mean calling rate per day based on the total 72-minute sample collected per day of monitoring. The statistical analyses described below used these values calculated for all the 36 days (n = 36) of acoustic monitoring. We used Generalized Additive Models, or GAMs (Hastie and Tibshirani 1990, Wood 2017) to examine whether air temperature (minimum or maximum) and precipitation affected calling rates (the response variable - the mean number of advertisement calls per minute per day). The principal advantage of this approach is the use of the basis spline functions (adaptive smoothers), which enables the depiction of both linear and nonlinear relationships. This approach can incorporate putative nonlinear relationships between climatic variables and calling rates due to the seasonal variation in the climatic data. We tested the different GAMs in a forward stepwise manner by inserting climatic variables as predictors and including days as a random effect (Wood 2017). We used the Akaike Information Criterion (AIC) to select the best-fit model, considering a 5% significance level. The analyses were run in the MGCV package (Wood 2011, 2017) of the R platform (R Core Team 2019).

RESULTS

The calling activity of *S. fuscomarginatus* peaked during two periods, that is, between November 2013 and March 2014, and August-October 2014 (Figs 3, 4). The males called at the end of the dry season (August-September 2014) and at the onset of the rainy season (October 2014). The highest calling rates were recorded during the early night (6:00–8:00 pm) and decreased gradually until 6:00 am (Fig. 5). Males emitted advertisement

Figures 1–2. (1) Study site in Rio Verde, Goiás, Brazil; (2) setup of the ADR, Sony model ICD-PX312.



Figures 3–4. (3) Monthly precipitation (gray bars), minimum air temperature (dashed line), and maximum air temperature (solid line) recorded between November 2013 and October 2014 in the municipality of Rio Verde, Goiás state, Brazil. (4) The mean number of advertisement calls (calling activity) recorded per minute (white bars) per day at all the study ponds.

calls antiphonally, i.e., avoiding acoustic overlap by synchronizing their calls with those of the other males in the same area. As the chorus noise decreased over the course of the night, the number of males calling antiphonally decreased.

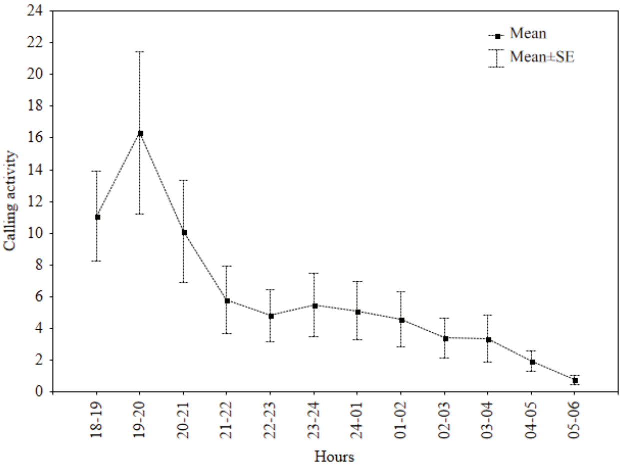
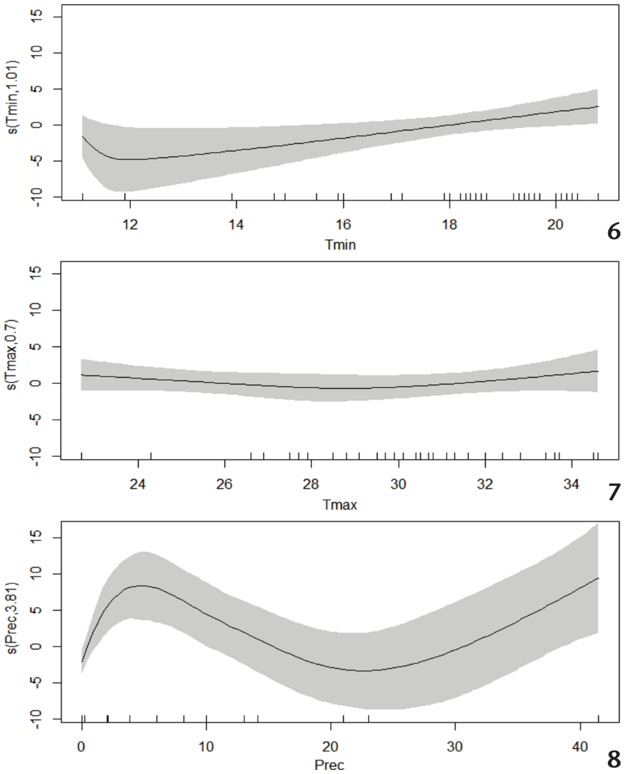


Figure 5. Nocturnal variation in the calling activity (number of advertisement calls per minute) of the *Scinax fuscomarginatus* population in the municipality of Rio Verde, Goiás state, Brazil. The mean and standard error (SE) were estimated based on the recordings from all the study ponds (2013–2014).

The best-fitting model retained all the predictor variables, albeit with different trends (Table 1, Figs 6–8). The minimum air temperature had an increasing linear effect on the phenology of the calling activity, although the maximum air temperature did not explain the variability in the model significantly. By contrast, precipitation had a significant nonlinear effect on calling rates.

DISCUSSION

We used ADR to describe calling activity in *S. fuscomarginatus* in full detail. This species presented a prolonged breeding pattern (Wells 1977) with calling activity being recorded throughout the night, and during two breeding seasons per year. The peak in calling recorded during the early night in the present study is consistent with the findings of previous studies of this species in the Cerrado and Pantanal biomes (Bernarde and



Figures 6–8. Results of the Generalized Additive Model for the relationship between calling activity and (6) minimum and (7) maximum air temperatures and (8) precipitation. The inner tick marks on the x axes represent the raw values of the climatic variables. The shaded area indicates the 95% confidence interval. (Tmin) Minimum air temperature, (Tmax) maximum air temperature, (Prec) precipitation.

Table 1. Results of the Generalized Additive Model (n = 36) for calling activity (CA, mean number of advertisement calls per minute) of *Scinax fuscomarginatus*. (Tmin) Minimum air temperature (°C), (Tmax) Maximum air temperature (°C), (Prec) Precipitation (mm), (DE) Deviance explained, (EDF) Effective Degrees of Freedom.

Model structure	Terms						
	R^2	AIC	DE (%)		EDF	F	p
CA~ s(Tmin)+s(Tmax)+s(Prec)	0.60	211.17	66.6	Tmin	1.007	0.581	0.011
				Tmax	0.698	0.168	0.114
				Prec	3.808	3.904	0.000049
CA~ s(Tmin)+s(Tmax)	0.40	224.91	47.3	Tmin	2.265	5.411	0.004
				Tmax	1.891	4.703	0.016
CA~ s(Tmin)+s(Prec)	0.58	213.23	64.3	Tmin	1.380	3.413	0.039
				Prec	3.975	6.030	0.0005
CA~ s(Tmax)+s(Prec)	0.54	216.33	61.6	Tmax	1.779	1.765	0.166
				Prec	3.818	8.153	0.00007

Kokubum 1999, Prado et al. 2005, Toledo and Haddad 2005a, Kopp et al. 2010), although it does expand the known range of night-time calling behavior in *S. fuscomarginatus*. The phenology of the calling behavior of *S. fuscomarginatus* was also shown to be influenced by climatic variables (minimum air temperature and rainfall).

The ADR system can remain operational in the field for an extended period of time, increasing the probability of detecting calls. This technique can thus provide a more detailed description of calling patterns than other procedures that do not provide a continuous or permanent sample (Bridges and Dorcas 2000, Dorcas et al. 2009). Given this, ADR can be used to complement other, less permanent anuran survey procedures (Madalozzo et al. 2017) or estimate the probability that a species will vocalize during a given day or year (Saenz et al. 2006). The attributes of the ADR can improve the quality of the data at finer temporal scales for the more reliable sampling and analysis of the calling patterns of widely-distributed anuran species, such as *S. fuscomarginatus*.

The more refined data obtained using the ADR may also permit the prediction of the occurrence of *S. fuscomarginatus* under specific certain environmental conditions, i.e., temperature and rainfall. This species occurs at temporary ponds, and chorusing behavior usually occurs during the rainy season (Toledo and Haddad 2005a, Kopp et al. 2010). The non-linear trend found here in relation to precipitation indicates that the calling phenology of *S. fuscomarginatus* is related to the seasonal variation in rainfall. A similar pattern is found in many other anuran species (Prado et al. 2005, Kopp et al. 2010). Our findings indicate that *S. fuscomarginatus* becomes more active with increasing minimum air temperature, which may be related to the regulation of body temperature (Wells 2007). When the air temperature becomes too high, on the other hand, anurans tend to avoid evaporative water loss (Wells 2007). This relationship is important for the prediction of the presence of amphibians over the course of the year (e.g., Steelman and Dorcas 2010). It is important to note, however, that other environmental variables not measured in this study (e.g., relative humidity, barometric pressure, and wind speed) may also influence calling patterns in this species (e.g., Pombal-Jr 1997).

Overall, the findings of the present study have provided important insights into the phenology of calling patterns in *S. fuscomarginatus*. The procedures adopted here also appear to have good potential for the collection of data on the natural history of other frog species, providing maximum sampling coverage in the field at a relatively low cost.

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