GEOGRAPHIC CALL VARIATION AND NOTES ON REPRODUCTION AND ABUNDANCE OF *AMEEREGA FLAVOPICTA* (ANURA, DENDROBATIDAE)

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INTRODUCTION

Ameerega flavopicta (=Epipedobates flavopictus) (Lutz, 1925) is a distinctive dendrobatid species which reproduces in open environments subject to broad variations in humidity and temperature (Costa, Facure & Giaretta, 2006). As presently recognized, this species presents a wide distribution in the Brazilian states of Minas Gerais, Goiás, Tocantins, Pará, and Maranhão (Haddad & Martins, 1994; Colli, Bastos & Araújo, 2002; Costa, Facure & Giaretta, 2006; Giaretta *et al.*, 008). Inter - populational differences have been reported in color pattern (Haddad & Martins, 1994), and in call (Haddad & Martins, 1994; Costa, Facure & Giaretta, 2006). Eterovick *et al.*, 2005) suggested a populational decline of this species at Serra do Cipó (MG).

OBJECTIVES

In the present study we report new data on inter - populational call differences and the existence of cephalic amplexus in the species. We also address comments on habitat and abundance of this species in a region where populational decline has been suggested.

MATERIAL AND METHODS

Field works were carried out sporadically between November 2004 and November 2007 in three municipalities in Minas Gerais state: Araguari (MG) (two sites: 18º27'S; 45º85'W; 18º27'S; 48º31'W; 600 m alt.), Perdizes (MG) (19º11'S; 47º07'W; 890 m alt.), and Santana do Riacho (MG) (19º17'S; 43º35'W; Serra do Cipó, 900 m alt.) and one municipality in Goiás state: Caldas Novas (GO) (17º46'S; 48º39'W; 906 m alt.). The two most distant localities (Santana do Riacho and Caldas Novas) were about 550 Km apart in an East–West direction. All these localities were originally covered by Cerrado (Brazilian savanna) vegetational formations. We also took records of the habitat where individuals were heard in these localities.

Calls were recorded with a Boss 864 digital recorder (44.100 Hz, 16 bit resolution) and a Sennheiser ME67 microphone. Sound analyses were performed in the Sound Ruler software (Gridi - Papp, 2004) using a FFT (Fast Fourier Transformation) length of 512. Temperature range of call records from Araguari was 29–31º C, Perdizes 20–20.9º C, from Serra do Cipó 16.5–18.2º C and from Caldas Novas 23.5–25.8º C. All males records were done during the morning in Araguari (n = 3), Perdizes (n = 2) and Serra do Cipó (n = 3); in Caldas Novas, two males were recorded during the morning and one in the afternoon.

Within a call, the minimum frequency corresponded to median frequency of first pulse; the maximal was that of the last pulse. Population differences in advertisement call parameters (minimum and maximum frequencies, call duration, call rate, pulses number, and inter - call intervals) were evaluated through a Principal Components Analysis (PCA) applied on a correlation matrix (Manly, 1986) considering mean values (n = 6 calls/individual) for each recorded male. Call parameters data were standardized prior to analysis. The PCA was performed in the Fitopac 1.5 software (Shepherd, 2005). Call parameters, as summarized by the PCI scores, were correlated with individual temperature data through Spearman rank coefficient (Zar, 1999) and to eliminate temperature effects from the ordination, we carried out a partial PCA entering temperature as covariate using the CANOCO 4.5 statistical software (Ter Braak & Smilauer, 2002). Again, call values of parameters were standardized prior to analysis (Zar, 1999).

One amplectant pair was observed (Serra do Cipó) while courting and egg - laying; the most important behaviors are reported. A voucher specimen is in the anuran collection (AAG - UFU 2526, Caldas Novas, GO) of the Museu de Biodiversidade do Cerrado, Universidade Federal de Uberlândia, MG, Brazil.

Data from Serra do Cipó is of particular interest because it is close (60 Km, NE) to that of the type locality (Belo Horizonte) and from a site where a populational decline of *A. flavopicta* has been inferred (Eterovick *et al.*, 005). At Serra do Cipó we recorded and counted calling males along a 500 m transect.

RESULTS AND DISCUSSION

In Araguari (MG), males were found calling along erosive trenches in a pasture (cattle farming) area and along road cut banks of the BR 050 highway; both sites in steep terrain draining to the Paranaíba River. In Perdizes (MG), males were found calling along the border of deep (< 2.5 m deep, 0.5–1.0 m wide) erosive trenches in two sites, 1) at the Estação Ambiental Galheiro (a private (CEMIG) reserve with 2800 ha) and 2) along the MG 452 highway. At the Estação Ambiental Galheiro, males called from an area covered by scattered low (< 1.5 m) shrubs and grass - like vegetation. Close the highways, males were heard calling along banks of the road cut (> 10 m from the asphalt - paved line) in areas covered by sparse tall (< 2 m) grass tufts.

At Serra do Cipó (MG) we found ca. 30 males calling along a 500 m transect parallel to a second order creek in disturbed (cattle and horse farming) rocky fields and two other males beside human settlements. All males called from distances larger than 50 m of the water. Another individual was found hidden under rocks beside a creek in a recently (< 3 months) burnt area.

In Caldas Novas (GO) were found males calling in two sites, 1) at the preserved area Parque Estadual da Serra de Caldas Novas (PESCAN) and 2) along the GO 139 highway. At the PESCAN, males called mainly from rocky areas covered by scattered low shrubs and grass - like vegetation. Close the highways, many males (ca. 50) were heard calling along banks of the road cut (> 10 m from the asphalt - paved line) in areas covered by sparse tall (< 2 m) grass tufts.

The call of all populations was a single pulsed note with ascending modulation from the beginning to the end. The calls recorded at Araguari (MG) (n = 3 males) had 7–9 pulses. The first pulses had minimum frequencies between 3457–3981 Hz and maximum frequencies between 3788–4285 Hz. The call duration averaged 0.11 s and inter - call intervals, 0.31 s. Call rate was 123–146/min. Calls from Perdizes (MG) (n = 2 males) were composed by 6–9 pulses. Minimum frequencies were between 3181–3954 Hz and maximum frequencies between 3374–4340 Hz. The mean duration of the call was of 0.17 s and inter - call intervals 0.43 s. Call rate was 94–109/min. The calls from Serra do Cipó (MG) (n = 3 males) had 6–10 pulses. The minimum frequencies were between 2878–3457 Hz and maximum frequencies between 3292–3705 Hz. The call duration averaged 0.15 s and inter - call intervals, 0.46 s. Call rate was 78–129/min. Calls from Caldas Novas (GO) (n = 3 males) were composed of a single note with 6–8 pulses. The first pulses presented minimum frequencies varying between 3319–4119 Hz and the last pulses with maximum frequencies between 3650–4367 Hz. The mean call duration was 0.11 s, and inter - call intervals averaged 0.26 s. Call rate was 140–175/min.

Among frogs, intraspecific variation in advertisement call parameters have been reported to widely distributed populations (sometimes taking the form of gradients or clines) and to highly isolated populations. Geographical call variation can be found in parameters such as call duration, pulse number, frequency range, and emission rate (Smith, Osborne & Hunter, 2003; Bernal, Guarnizo & Luddecke, 2005), such as we reported here for *A. flavopicta* .

The first two components of the PCA accounted for 66.8% and 19.7% of total variance in advertisement calls, respectively. The plot of Axis I and II showed a separation between the populations from Perdizes (MG) and Serra do Cipó (MG) from those of Caldas Novas (GO) and Araguari (MG) along the first component. The main call parameters that contributed to the first component were call duration (auto vector = 0.45) on the positive side and call rate (number of calls per minute) (–0.44) and minimum (–0.43) and maximum (–0.41) frequencies on the negative side. The second component represent basically pulses number (auto vector = –0.54) on the negative side and inter - call intervals (0.54) on the positive side. Summarizing, the advertisement calls of Caldas Novas and Araguari populations differed from those of Perdizes and Serra do Cipó in both temporal and spectral (frequency) features. Individuals from Araguari and Caldas Novas had shorter calls with higher minimum and maximum frequencies and were emitted in a higher rate. Intra - populational variation in pulses number was greater than that among populations.

Based on a larger sample, here we can reinforce our previous suggestions (Costa, Facure & Giaretta, 2006) that the advertisement calls of Western populations (Caldas Novas, GO) present shorter notes and smaller call intervals than those from East (e.g. Serra do Cipó, MG); localities at the extremes of our sample data. The negative correlation we found between temperature and PCI scores (rs = - 0.682; n = 11; P < 0.05) indicates that, in general, in hotter localities one can find higher call rates and shorter calls, a relationship well documented in the literature (Narins *et al.*, 007). We found also that localities do not mixed up in a temperature x PCI plot, indicating that the geographical component of the call differences variation was not just an artifact generated by the differences in temperature. At present, the Paranaíba River (about 200 m wide) is a major geographic barrier and historically can be the responsible for the isolation of the populations of *A. flavopicta* of Caldas Novas (GO) and Eastern populations (Araguari, Perdizes and Serra do Cipó - MG).

In December 2005 (7:00h), we followed a courting pair at Serra do Cipó. Shortly (5 min) after being found in a narrow rocky crevice, the couple entered amplexus in the point the rock contacted earth (20 cm deep). Amplectic position varied along the observation time (45 min) from axillary, mid - body, to cephalic (with the dorsal surface of the male's first fingers contacting the corner of the female mouth), the last being the most frequent.

In the last 15 years one of us (AAG) visited three times (1990, 1997, and 2005) the population of Santana do Riacho in South Serra do Cipó, at margins the MG 10 highway.

In the last trip (2007), we observed an expressive number (ca. 30) of calling males and even found a reproducing pair. This area was altered by human activities (human settlements, pastures for cattle and horse farming) in our first trips and since then the situation get worse (more human visitation, larger amount of improperly disposed domestic garbage, and MG 10 asphalt pavement). Besides this increased disturbances, in all our visits there was an expressive number of calling males there.

Experts (colleagues and AAG) officially reviewed the List of Endangered Fauna of the state of Minas Gerais (Biodiversitas, unpublished) and, using IUCN definitions (see Azevedo - Ramos *et al.*, 004), regarded *A. flavopicta* as a species of "Least Concern".

The decline of other populations of *A. flavopicta* at South Serra do Cipó (19º16'-19º17' S, 43º34'-43º35' W) was reported by Eterovick *et al.*, 2005) and these authors suggested anthropogenic factors (cattle farming, spread of human settlements, and fires) as plausible causes of this supposed decline. We (Costa, Facure & Giaretta, 2006; present study) have pointed to *A. flavopicta* as a species quite resistant to stressing (dry/hot) habitats and to human implemented modifications (e.g. cattle farming, natural vegetation removal, fire, and vehicle traffic). If in reality some local populations are declining at Serra do Cipó, other causes (Blaustein *et al.*, 994; Davidson, Shaffer & Jennings, 2001) than direct/local anthropogenic effects should be considered.

CONCLUSION

Based on a larger sample, we reinforce our previous suggestions that the advertisement calls of Western populations (Caldas Novas, GO) present shorter notes and smaller call intervals than those from East (Araguari, Perdizes and Serra do Cipó-MG).

We found that amplectic position is variable (axillary, mid - body, and cephalic) but recognize the cephalic amplexus as the main type to the species.

We have pointed to *A. flavopicta* as a species quite resistant to stressing (dry/hot) habitats and to human implemented modifications (e.g. cattle farming, natural vegetation removal, fire, and vehicle traffic).

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