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Conspecific interaction of *Hylodes phyllodes* Heyer & Cocroft, 1986 (Anura; Hylodidae) in an Atlantic Forest fragment, Southeastern Brazil.

Interação conspecífica de *Hylodes phyllodes* Heyer & Cocroft, 1986 (Anura; Hylodidae) em um fragmento da Floresta Atlântica, Sudeste do Brasil.

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Abstract The species Hylodes phyllodes Heyer and Cocroft, 1986 is a diurnal anuran primarily associated to lotic environments inside the forests. It can be found throughout the Serra do Mar mountain range in the South and Southeast regions of Brazil. We conducted observations at the Ecological Reserve Rio das Pedras (RERP) in the municipality of Mangaratiba, Rio de Janeiro state, Brazil to study visual communication and vocalization of males of H. phyllodes. We used a live puppet to stimulate *H. phyllodes* behavior. A live puppet consists in a specimen of H. phyllodes tied with a fine thread via its pelvis to a fishing rod. We used a video camera to record the species behavior. We observed 20 males of *H. phyllodes* and confirmed that this species may occupy the same call site for several days, thus site fidelity. Under experimental showing some

conditions, 35% (n = 7) of the observed individuals showed immobility in front of the live puppet; 25% (n = 5) emitted a warning call; 20% (n = 4) showed physical confrontation; 10% (n = 2) emitted a sound distinct from the warning call; and 10% (n = 2) displayed an escape behavior.

Keywords: animal behavior, Atlantic Forest, live puppet, visual displays.

Resumo A espécie *Hylodes phyllodes* Heyer e Cocroft de 1986 é um anuro diurno principalmente associado a ambientes lóticos no interior das florestas. Ele pode ser encontrado em toda a Serra do Mar, nas regiões Sul e Sudeste do Brasil. Nós realizamos observações na Reserva Ecológica Rio das Pedras (RERP) no município de Mangaratiba, Estado do Rio de Janeiro, Brasil, para estudar comunicação visual e vocalização dos machos de H. phyllodes. Utilizamos uma marionete viva para estimular o comportamento de H. phyllodes. Uma marionete viva é um espécime vivo de H. phyllodes que foi amarrado com um fio fino na sua pélvis em uma extremidade e a outra extremidade foi presa a uma vara de pescar. Foi utilizada uma câmera de vídeo para registrar o comportamento da espécie. Foram observados 20 machos da espécie H. phyllodes e foi confirmado que esta espécie pode ocupar o mesmo sítio de vocalização por vários dias, mostrando assim alguma fidelidade ao sítio. Sob as condições experimentais utilizadas, 35% (n = 7) dos indivíduos observados mostraram imobilidade frente à marionete viva; 25% (n = 5) emitiu uma chamada de alerta; 20% (n = 4) mostraram confronto físico; 10% (n = 2) emitiram um som distinto do chamado de alerta e 10% (n = 2) mostraram um comportamento de fuga.

Palavras-chaves: comportamento animal, exibição visual, Floresta Atlântica, marionete viva.

Introduction

Visual and sound communications are displayed by groups of vertebrates. In anurans, various the communication is mainly based on vocalization, whereas there is limited use of visual, chemical or tactile signals (Duellman and Trueb 1994).Visual interactions are diverse and probably evolved independently under several ecological factors (Hödl and Amézquita 2001). Another accepted hypothesis is that visual signals have evolved from the ritualization of intentional movements, not previously used for direct communication, and in some cases was favored by the environmental conditions (Hödl and Amézquita 2001). Visual communication is widespread in diurnal frogs, and is probably favored by the availability of sun light (Lindquist and Hetherington, 1996). However, new data have provided evidence that nocturnal visual signaling may be more widespread than previously thought (Abrunhosa and Wogel 2004; Hartmann et al. 2005; Toledo et al. 2007; Barros and Feio 2011; Lipinski et al. 2012).

According to Haddad and Giaretta (1999), visual signaling is widespread among species of the family Hylodidae, which are usually associated with lotic ecosystems, mostly rivers and streams (Heyer *et al.* 1990). Recently, several studies have shown, through direct in situ observation, the visual and acoustic repertoire for the genus *Hylodes* (Hartmann *et al.* 2004;

Hartmann *et al.* 2006). The aim of this work was to study visual communication and vocalization of males of *Hylodes phyllodes* Heyer & Cocroft, 1986 in a private reserve in the State of Rio de Janeiro.

Materials and methods

Study area

This work was carried out in the Reserva Ecológica Rio das Pedras (RERP), a private protected area (Reserva Particular do Patrimônio Natural - RPPN) covering approximately 1,361 ha in the Mangaratiba municipality, Rio de Janeiro state. The RERP is a fragment of the Ombrophilous Atlantic Forest along the southern coastal portion of the state. This area is composed of secondary forest with a large number of herbaceous and epiphytes species, and trees of up to 40 m high (Mynssen and Windisch 2004). Streams and rivers are also common and form the hydrographic basin of the Rio Grande. The climate is categorized as "subquente" (Nimer 1989) with temperatures ranging from 22°C to 38°C. The significant variations of altitudinal range near the coast of Serra do Mar causes high rainfall rates during the year, with the highest averages in the months of December, January, and February (Mynssen and Windisch 2004).

Data collection

The observations were conducted every other week at a transect on the banks of the Rio das Borboletas (22° 59' 27'' S, 44° 06' 06'' W) from January to November 2004, in daytime from 08:00 am to 05:00 pm, totaling 198 man/hours (11 months X 9 hours X 2 times per month). We used the animal focal sampling method (Lehner 1979; Martin and Bateson 1986). The resident males were recorded with a digital video camera.

We induced behavioral responses of isolated resident males by tying a fine thread on pelvis of live males *H. phyllodes* to a 2 m long stick. All procedures were done carefully in order to avoid animal suffering. After the experiment, the individual was released at the site where it was caught. The tied individual was displayed in front of the other conspecific residents at a short distance, sufficient for visual contact, simulating an occasional encounter between the two individuals. After recordings, we evaluated air temperature and humidity using a digital thermo hygrometer. Two voucher specimens were fixed according to standard procedures and deposited at the Amphibian Collection of the Museu Nacional/Universidade Federal do Rio de Janeiro (MNRJ 37900 and MNRJ 37901). Authorization to carry out the

fieldwork was given by the manager of the reserve. The average temperature during the study period was 26.6 ± 1.4 °C and the average humidity was 75.7 ± 5.6 %.

Results

Behavioral responses of 20 males of *H. phyllodes* were recorded. Adult males of H. phyllodes at the RERP emitted calls at specific breeding sites, housed in small shelters formed by rocks along the banks of streams. When intruder males (tied conspecific) were placed in front of resident males, 35% (n = 7) of the resident males remained motionless; 25% (n = 5) emitted warning call, and 10% (n = 2) emitted sounds different from the warning and advertisement calls toward the tied conspecific. In 20% (n = 4) of the observations direct physical confrontations were observed between resident males and tied conspecific. This behavior varied widely in intensity, ranging from a simple touch with the limbs, attempts to displace the opponent and bites. In 10% (n = 2) of the observations we observed escape behavior by the resident male.

Discussion

The resident males of *H. phyllodes* did not display visual signaling, such as leg stretching. However, some studies under natural conditions have reported visual signaling for this species (Hartmann *et al.* 2005) and other congeneric (Haddad and Giaretta 1999; Narvaes and Rodrigues 2005). Indeed, although live puppets were well succeeded to stimulate natural behaviors, handling of them may have interfered in resident male behavioral response also.

The species *H. phyllodes* has a cryptic color pattern and a lighter color on the throat and vocal sacs (Haddad and Giaretta 1999, Hartmann *et al.* 2005). The emission of advertisement calls may be associated with the display to the non-resident male and agonistic encounters between males were described by Hartmann *et al.* (2005). Resident males in the RERP population exhibited a similar behavior. They also opened their mouths to bite the non-resident males. Attempts to displace the nonresident males with their limbs were also observed. The fact that most individuals did not display any known and reported behavior can be attributed to factors such as: (1) possibly the lower density of this population observed when compared to other populations studied, like that of Picinguaba, São Paulo, by Hartmann *et al.* (2006), thus reducing the need of visual signals; (2) the methodology used could have inhibited or even changed the behaviors or (3) the fact that the individuals usually vocalized in very specific and enclosed sites reduces the visual area of the resident males hindering this type of communication.

All individuals of *H. phyllodes* were observed calling in small shelters along the banks of streams, similar to the observations conducted by Hartmann *et al.* (2005) for several individuals in the populations of Picinguaba, São Paulo state (Brazil). Further studies on the behavior of this species in other protected areas will be important to corroborate an existence of patterns that may possibly be related to abiotic factors, biotic or even intraspecific. Human interference with wild populations is not well understood, and requires further studies to learn how much the human presence interferes with their behavior, especially regarding the visual and sonorous communication of *H. phyllodes*.

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