Defensive behavior of *Ololygon carnevallii*

**CARAMASCHI & KISTEUMACHER, 1989**

(Anura: Hylidae)

Abwehrverhalten von *Ololygon carnevallii* CARAMASCHI & KISTEUMACHER, 1989

(Anura: Hylidae)

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**ABSTRACT**

High predatory pressure exerted on anurans is likely to be a factor in the development of the large defensive repertoire presented by these animals. *Ololygon carnevallii* CARAMASCHI & KISTEUMACHER, 1989, is a medium sized species of the genus; it occurs in the biomes of Mata Atlântica and its transition to Cerrado, in Minas Gerais State, Brazil. Based on field observations, the authors describe various defensive behaviors of this species. Field observations took place from August 2015 to July 2016, in two fragments of the Atlantic Forest in the municipalities of Mariléria and Mesquita. *Ololygon carnevallii* exhibited behaviors of active escape, hiding, mouth-gapping, thanatosis (death-feigning), cloacal discharge, puffing up the body and crouching down, which probably increases its chance of survival.

**KEY WORDS**

Amphibia. Anura: Hylidae, Scinaxinae; *Ololygon carnevallii, Scinax carnevalli*; antipredator behavior, defensive behavior, ethology, natural history, Atlantic Forest, Minas Gerais, Brazil

**INTRODUCTION**

Anurans are prey for numerous vertebrate and invertebrate species (DUELLMAN & TRUEB 1986; TOLEDO 2005; TOLEDO et al. 2007). This predatory pressure may be responsible for the development of the large defensive repertoire that anurans exhibit (MARTINS et al. 1993; TOLEDO et al. 2007; TOLEDO et al. 2011). Among anuran defensive behaviors, active escape and immobility are common (MARCHISIN & ANDERSON 1978; TOLEDO et al. 2011). In addition, anurans display diverse and complex defensive repertoires (WILLIAMS et al. 2000; TOLEDO et al. 2011; GALLY et al. 2014) from isolated behaviors to a combination of behaviors, called synergistic behavior. Such multiple tactics developed to hinder or disrupt the predator’s action more efficiently (MARCHISIN & ANDERSON 1978; TOLEDO et al. 2011).

Within the genus, *Ololygon carnevallii* CARAMASCHI & KISTEUMACHER, 1989, is a medium sized species (snout-vent-length 21–25 mm in males; 30–32 mm in females), that is arboreal and nocturnal (CARAMASCHI & KISTEUMACHER 1989). It occurs in open and forested areas in the biome Mata
During a herpetological study carried out in two fragments of the Atlantic Forest located in the municipalities of Marliéria (19.724509° S; 42.700646° W; datum = WGS84) and Mesquita (19.258769° S; 42.553933° W), in the state of Minas Gerais, Brazil, the authors observed several types of defensive behavior in 106 individuals of *O. carnevallii*. Field work took place in the dark (18:00 – 22:00 h) during August 2015 and July 2016, and included sporadic collecting of herpetological specimens. Behaviors were documented by photographs and films (the latter causing poor image quality of Figs. 1A, 2) using a Sony HX 300 SLR camera, TOLEDO et al. (2011) were followed to classify the behaviors displayed by the individuals. Voucher specimens (License: Instituto Chico Mendes de Conservação da Biodiversidade, Brasília – ICMBio 52251-1) were euthanized according to governmental regulations (Conselho Federal de Biologia – CFBio N° 148/2012; Regulamenta os procedimentos de captura, contenção, marcação e coleta de animais vertebrados previstos nos Artigos, 4º, 5º, 6º e 8º da Resolução CFBio nº 301/2012) by immersion in an aqueous solution of benzocaine hydrochloride 250 mg/l or intraperitoneal injection of liquid lidocaine. Specimens were fixed in 10% formalin solution and stored in ethanol 70%, in the amphibian collection at the Museu de Zoologia João Moojen da Universidade Federal de Viçosa (MZUFV), Minas Gerais, Brazil.

RESULTS

During field observations, *Oologygon carnevallii* exhibited seven types of defense behavior: active escape, hiding, mouth-gapping, thanatosis (death-feigning), cloacal discharge, puffing up the body and crouching down. When the observer approached, the most common defensive strategies were active escape and hiding.

Of the 106 individuals found, two males exhibited mouth-gapping behavior (Fig. 1A). The first observation of this behavior was in the field at 20:00 h on June 30, 2016 (air temperature 11.1 °C and humidity 95%) near a stream in the Municipality of Marliéria. The specimen was perched on a bush 2.35 m above the ground. Once illuminated, the frog immediately began displaying the mouth-gapping behavior with 13 repetitions within 74 seconds. Each mouth-gapping event (duration 2 to 12 sec) consisted of three phases: mouth closed, mouth totally open and mouth partially opened. During the 13 repetitions, this cycle occurred in intervals of 4 to 6 seconds. After the 13 repetitions the frog finalized its display by closing its mouth completely. Throughout the observation the individual remained at the same perch location.

The second observation of this behavior occurred during the daytime in the laboratory with an individual from the Municipality of Mesquita. The frog was inside a plastic collecting bag, which contained moist vegetation. When the observer approached, the male began to display the mouth-gapping behavior (each mouth-gapping lasting approximately 1 sec), with eight repetitions for 48 seconds. During all the repetitions the animal opened its mouth completely and then closed it, during intervals ranging from 4 to 6 seconds between the acts.
Approximately 20% of the individuals handled exhibited the behavior of thanatosis. After capture, the specimens kept their ventral side up, eyes open, with the front and hind limbs of one side, closer to the body than the limbs of the opposite side (Fig. 1B).

Approximately 30% of individuals approached in the field exhibited crouching down behavior (Fig. 1C), with their snouts contacting the substrate, the front limbs below the head and the hind limbs below the belly. Some specimens kept their eyes half-open, while others closed them completely. During capture and manipulation in the field, about 70% of individuals discharged a liquid from the cloaca. The authors were unable to discern a particular odor coming from the liquid discharge; it was not sticky and quickly dried on the hands. In some cases, the animals inflated the body after this behavior.

**DISCUSSION**

Mouth-gaping behavior has been described for 18 species of anurans (MYERS 1966; DUELLMANN & TRUEB 1986; FORMS & POBLETE 1996; TOLEDO et al. 2011; FERREIRA et al. 2013). However, there are no records for any species of the subfamily Scinaxinae (type genus: *Scinax* WAGLER, 1830), which *O. carnevallii* belongs to (DUELLMANN et al. 2016). Individuals of *O. carnevallii* that exhibited this behavior remained in the same position in which they were perched.

Species from other families, such as *Hemiphractus fasciatus* PETERS, 1862, and *Haddadus binotatus* SPIX, 1824, arched their body backward (lightly or vigorously) during the mouth-gaping behavior (MYERS 1966; TOLEDO et al. 2011). This behavior is more conspicuous if the color of the buccal cavity or tongue is different from the body coloration. This may be the case in *O. carnevallii*, that showed different tones of slightly bluish coloration on the body surface and the oral cavity during mouth-gaping.

According to TOLEDO et al. (2011), the species known to exhibit mouth-gaping also emit defensive vocalizations except *Cycloramphus acangatan* VERDADE & RODRIGUES, 2003, *Brachycephalus* spp. and
Proceratophrys boiei (Wied-Neuwied, 1824). However, these two behaviors cannot be linked as a sequence of events. The same authors, also stated that mouth-gapping can precede an agonistic call or a bite. The male individuals of *O. carnevallii* did not emit defensive vocalizations or any other associated behavior, they soundlessly opened and closed their mouths. The difference in the number of acts (13 vs. 8) displayed by the specimens on the two occasions may be due to difference in the threat scenarios.

Toledo & Haddad (2009) found a positive relationship between snout-vent-length of anurans and the duration of their agonistic call, and a negative relationship between the body size and the dominant frequency of the call. If we consider that the size of the lungs is related to the air volume capacity and sound power, smaller anurans emit a shorter call with less intensity and may not be able to scare off most predators (Barrío 1963; Toledo & Haddad 2009). *Ololygon carnevallii* is a small species, thus, its lineage may have lost the ability to emit agonistic calls associated with the mouth-gaping behavior due to the ineffectiveness of this acoustic signal.

Thanatosis was reported for several anurans of the subfamily Scinaxinae, e.g., by Toledo (2004) for *Scinax fuscovarius*, (Lutz, 1925), by Rodrigues & Rodrigues (2007) for *Scinax fuscomarginatus* (Lutz, 1925), and by Toledo (2010) for *Scinax alter* (Lutz, 1973), and *Ololygon catharinae* (Boulenger, 1888). Varying degrees of stress triggered different antipredator behavioral responses in *Gastrotheca recava* Teixeira, Vechio, Recoder, Carvalho, Strangas, Damasceno, Sena & Rodrigues, 2012, and *Gastrotheca megacephala* Izeksohn, Carvalho-e-Silva & Peixoto, 2009, with thanatosis observed when stress levels increased (Lourengo de Moraes et al. 2016). This behavior has proven to be successful against different predators, mainly birds and snakes (Brodie 1977; Marchisin & Anderson 1978), since remaining still or pretending to be dead can reduce the aggression intensity of the predator (Brodie 1977). Manipulation is threatening to anurans since it simulates the act of being eaten. Thanatosis can prevent injuries during predation events from animals that are later regurgitated due to toxic secretions in their integument or can distract predators that are guided or stimulated by the prey’s movement (Bortheiro et al. 2014; Toledo et al. 2011). *Ololygon carnevallii* performed thanatosis only when handled and turned ventral side up, suggesting that thanatosis may be a last resort for this species as soon as active escape is not seen as a productive option.

Crouching behavior has been recorded for several anuran species (e.g., Marchisin & Anderson 1978; Williams et al. 2000; Menin & Rodrigues 2007). This behavior can be considered a type of immobility (Toledo et al. 2011), which lowers predation risk by reducing the perception of predators who use movement of the prey to locate it or whose hunting activity is triggered by the movements of the prey.
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(Marchisín & Anderson 1978; Heinzen & Hammond 1997). In addition, *O. carnevalii* has a cryptic coloration that resembles moss (Figs. 1C, 2) making visualization difficult, especially when crouching down.

Among Scinaxinae, only *S. fuscovariegatus* is known to utilize cloacal discharge as a recognized defensive strategy (Toledo et al. 2011). This behavior which is common in anurans (e.g., Kokubum 2002; Toledo et al. 2005; Carvalho-Júnior 2005) can avoid subjugation, since the cloacal content can strike the predator, or leave the anuran lighter, allowing the animal to escape faster (Toledo et al. 2011).

*Ololygon carnevalii* uses different defensive strategies when threatened, which, along with its coloration and environment, increase its chance of survival. The diversity of defensive behaviors presented by this species demonstrates that it developed mechanisms to evaluate which defensive strategies are most promising to use according to the particular situation.

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