

SHORT COMMUNICATION

Repertoire of antipredator mechanisms in the Brazilian toad *Frostius pernambucensis* (Anura: Bufonidae)

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Anuran amphibians, along with the diversification of species, established themselves in practically all regions of the globe (except Antarctica and most of the oceanic islands (Duellman and Trueb 1994, Frost 2021)). These animals, throughout all stages of life, correspond to an important element of the food chain, serving as predators and prey for various vertebrate and invertebrate groups, both in aquatic and terrestrial environments (Duellman and Trueb 1994, Verburg *et al.* 2007). They have developed several morphological, physiological and behavioral adaptations to overcome their many predators (Duellman and Trueb 1994,

Verburg *et al.* 2007, Ferreira *et al.* 2019). These defensive strategies can be exhibited singly or in combination, depending on the factors underlying predator and prey interactions (Duellman and Trueb 1994).

Most antipredator mechanisms expressed by anurans can be observed in the species that live in the Atlantic Forest (Haddad *et al.* 2013, Ferreira *et al.* 2019), an ecoregion considered a biodiversity hotspot worldwide (*sensu* Myers *et al.* 2000). Part of this is due to the high diversity of species, which are currently known more than 600 amphibian species, of which around 80 percent are endemic to this ecoregion (Rossa-Feres *et al.* 2017). With this said, the basic aspects of the natural history of many species remains poorly understood, including defensive repertoires (Ferreira *et al.* 2019).

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Among these species, *Frostius pernambucensis* (Bokermann, 1962) (Figure 1) is a small bufonid toad endemic to the Atlantic Forest of Northeastern Brazil (states of Pernambuco, Alagoas, Sergipe and the northern region of the state of Bahia; Frost 2021). Some defensive behaviors for this species have been listed by Haddad *et al.* (2013) and Ferreira *et al.* (2019) such as mechanisms to avoid detection (camouflage) or to avoid predation (aposematism, body inflation, contraction, death feigning, counterattack, and poisonous secretions). Some of these behaviors are associated with a variety of postures in order to play dead or to break-up the animal's characteristic silhouette promoting the predator's disinterest (Toledo and Haddad 2009, Ferreira *et al.* 2019). Additionally, these movements may maximize the display of the aposematic coloring of the belly and limbs. Here, we update the repertoire of defensive behaviors of *F. pernambucensis*, including one previously unregistered mechanism for the species: leg interweaving, being the first record for a representative of the Bufonidae family.

The records were taken during two field expeditions at the Estação Ecológica de Murici (ESEC de Murici), municipality of Murici, state of Alagoas, Northeastern Brazil (09°13'09.6" S, 35°52'44.2" W; datum WGS84; 562 m a.s.l.). On 20 March 2021 at 17:40 h, we found a *F. pernambucensis* individual in a basal axil of a terrestrial bromeliad of the species *Aechmea leptantha* (Harms) Leme and J.A. Siqueira. After the individual failed to escape capture, it displayed a sequence of defensive behaviors. Initially, we realized that the individual was performing a contraction display. Briefly, the individual remaining immobile, ventral side upwards, with their fore and hind limbs contracted close to the body as to display the yellowish color of the belly (Figure 1A). When placed on a leaf, the individual performed the "leg interweaving" behavior (Figure 1B–C). This behavior consisted of an irregular movement of the hind limbs, where the individual kept their limbs crossed in an "X" shape while remaining

ventral side upwards. During the leg interweaving behavior, the individual kept its forelimbs bent, close to its body with its eyes closed, tilting its head backwards. After a few seconds, the individual returned to its original position, perched on the leaf.

The second record was taken on 27 July 2021 at 21:30 h. We found an individual vocalizing on a tree leaf 1.5 m above the ground. We manually relocated the individual from the leaf to a nearby area, to avoid immediate escape after the contact, where we stimulated it with soft touches in order to obtain a defensive response. First, the toad performed death feigning behavior while remaining immobile, with its entire body in contact with the ground. In death feigning, the anuran assumes a posture in which it remains immobile even when touched, appearing to be dead, keeping its fore and/or hind limbs loose, causing the predator to lose interest (Toledo *et al.* 2010). While in this state the individual had notably slow reflexes: for example, when we stretched its hind limb, the animal did not immediately retract it, but instead kept it outstretched for about five seconds (Figure 1E). When we touched it again, the animal returned the limb close to its body. Following this display, the individual performed body inflation behavior every time we approached it, inflating its body and tilting its head forwards (Figure 1F). This behavior occurs when the anuran fills its lungs with air, increasing in size (Toledo *et al.* 2011). This behavior can make it difficult to attack or handle the anuran, and it can also scare off a potential predator (Caro 2014, Ferreira *et al.* 2019). After taking photographic records, the animal was returned to the same location where it was found. None of the individuals were collected.

Contraction and death feigning behaviors are relatively well-documented in bufonid species, mainly from the genus *Rhinella* and may be associated with aposematic coloration, such in *Melanophryniscus* and *Atelopus* genus (Toledo *et al.* 2010, Ferreira *et al.* 2019), as observed in *Frostius pernambucensis*. Both



Figure 1. Defensive behaviors registered for *Frostius pernambucensis*. Individual registered in the first expedition, presenting contracting (A), legs interweaving or limbs interweave (B–C), and undoing the latter behavior (D). Individual registered in the second expedition presenting death feigning behavior (E) and body inflation behavior (F). Photos: MJMD (A–D) and Marcio Campelo (E–F).

behaviors can be displayed after an approach of a potential predator or after being handled (Toledo *et al.* 2010). However, contraction is mainly associated with toxic species and can be displayed even after subjugation by a predator (Toledo *et al.* 2010). Once swallowed, the contracted anuran protects its vital body parts and produces noxious secretions into the predator's digestive tract, inducing its regurgitation (Sazima 1974, Toledo *et al.* 2010). In *F. pernambucensis*, the handling of the animal induced the display of the contraction behavior. The individual may have interpreted it as being swallowed and contracted its limbs to protect the vital parts. As for death feigning, this behavior was induced by small touches on the toad and may have been performed as an attempt to make the predator lose interest.

With respect to leg interweaving, as far as we know, there has been no mention in the literature of this behavior for *F. pernambucensis* or any other representative of Bufonidae. This defense mechanism is poorly-documented (Ferreira *et al.* 2019) and is known to occur in only six anuran families: Craugastoridae (1 species); Hylidae (3); Hyperoliidae (1); Leptodactylidae (1); Phyllomedusidae (1); and Ranidae (1) (Channing and Howell 2003, Gally *et al.* 2014, Lourenço-de-Moraes *et al.* 2014, Ferreira *et al.* 2019, Rojas-Padilla *et al.* 2019, Souza *et al.* 2020). Distinguishing it from other anuran species, who keep their hind limbs intertwined dorsally and the body turned dorsally, *F. pernambucensis* maintained its body in a ventral position during the leg interweaving display, displaying the yellowish color of its belly. This behavior, associated with aposematic coloration on the ventral region of the body, has only previously been shown in the African hyperoliid tree frog *Hylambates keithae* (Schjøtz, 1975) (Channing and Howell 2003). According to Toledo *et al.* (2011), leg interweaving can occur synergistically with other defense mechanisms such as the display of disruptive and/or aposematic color patterns, or toxic substance secretions. In *F. pernambucensis*,

this behavior occurred simultaneously with the contraction of anterior limbs and synergistically exposing its aposematic colors. The presence of toxic secretions was not observed, although this mechanism has been described for the species (Ferreira *et al.* 2019).

Aposematic coloration can aid in predator avoidance, but the flashy color can sometimes have an adverse effect, increasing an individual's detectability in the environment (Hall *et al.* 2013, Ferreira *et al.* 2019, Röbller *et al.* 2019). A way to compensate for this is to have two strategies: camouflage and aposematism, the chosen strategy is dependent on body position (Barnett *et al.* 2017) as in *F. pernambucensis*. In these cases, the conspicuous feature is exposed when the animal moves (Channing and Howell 2003, Hall *et al.* 2013, Röbller *et al.* 2019), such as with the exposed aposematic coloration on the hands and feet of *Atelopus spumarius* Cope, 1871 (Röbller *et al.* 2019) and can be observed, similarly, in *F. pernambucensis*. We question whether the exposure of these colors in *F. pernambucensis* also has the same effect of increasing its detectability for some of its visually-oriented predators while moving, a behavior that exposes these parts, or if these animals benefit from this exposure. Thus, we show that more studies are needed to better investigate and elucidate these issues inherent to the natural history of this species.

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