## Predation by the Pebas four-eyed opossums *Philander pebas* on the toad *Rhinella margaritifera* in the west Brazilian Amazon

## Depredación por la zarigüeya Pebas de cuatro ojos *Philander* pebas sobre el sapo *Rhinella margaritifera* en la Amazonía Brasileña occidental

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Species of *Philander* occur in tropical and subtropical forests. Some species have a diet consisting of arthropods, the species within the genus are considered opportunistic vertebrate predators. Information about the natural history, ecology, and behavior of this genus is still scarce. Here, we present the first documented case of predation on the *Rhinella margaritifera* by *Philander pebas* in the Amazonian floodplain forest. The observation documented here was made during fieldwork in a trail located in the Area de Relevante Interesse Ecológico Japi-im-Pentecoste, of Cruzeiro do Sul, Acre, Brazil. We photograph the records using camera with the help of lighting lanterns. We encountered 1 specimen of *P. pebas* moving through a tangle of lianas and palm leaves, we observed the opossum with *Rhinella margaritifera* securely held in its mouth. The toad was gripped dorsally by the widest part of its head and ventrally by part of its neck, encompassing the parotoid glands. We noticed that *R. margaritifera* in its mouth was alive and displayed a defensive behavior known as "puffing up the body". Remarkably, the opossum displayed no apparent signs of toxin effects while grasping the toad near its parotoid glands. These findings challenge conventional assumptions about the toad's defense mechanisms against predators. Our record expands the understanding of predator-prey interactions in the Amazonian the significance of further research to unravel the dynamics of these novel interactions between opossums and venomous amphibians.

Key words: Acre; Didelphidae; feeding; food web; marsupial.

Las especies de *Philander* se encuentran en bosques tropicales y subtropicales. Algunas especies tienen una dieta compuesta por artrópodos, las especies dentro de este género se consideran depredadores oportunistas de vertebrados. La información sobre la historia natural, ecología y comportamiento de este género aún es escasa. Aquí presentamos el primer caso documentado de depredación de *Rhinella margaritifera* por parte de *Philander pebas* en el bosque de llanura aluvial amazónica. La observación documentada aquí, se realizó durante trabajos de campo en el Área de Relevante Interesse Ecológico Japiim-Pentecoste, en Cruzeiro do Sul, Acre, Brasil. Fotografiamos los registros utilizando una cámara con la ayuda de linternas. Encontramos 1 ejemplar de *P. pebas* moviéndose a través de lianas y hojas de palma. Observamos a la zarigüeya con una *Rhinella margaritifera* firmemente sostenido en su boca. El sapo estaba agarrado dorsalmente por la parte más ancha de su cabeza y ventralmente por parte de su cuello, abarcando las glándulas parotoides. Notamos que *R. margaritifera* en su boca estaba viva y mostraba un comportamiento defensivo conocido como "inflar el cuerpo". Sorprendentemente, la zarigüeya no mostró signos aparentes de efectos producidos por las toxinas mientras agarraba al sapo cerca de sus glándulas parotoides. Estos hallazgos desafían las suposiciones convencionales sobre los mecanismos de defensa del sapo contra los depredadores. Nuestro registro amplía la comprensión de las interacciones depredador-presa y destaca la importancia de realizar más investigaciones para desentrañar la dinámica de estas nuevas interacciones entre zarigüeyas y anfibios venenosos.

Palabras clave: Acre; alimentación; Didelphidae; marsupial; red alimentaria.

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Species of *Philander*, commonly known as 'gray four-eyed opossums' or 'pouched four-eyed opossums,' occur in lowland tropical and subtropical forests from México to northern Argentina (<u>Voss et al. 2018</u>). The genus *Philan-der* comprises 10 scansorial predator (<u>Voss 2022</u>) species that consume a wide variety of invertebrates, small vertebrates, corn, nectar, carrion, and fruits. They predominantly inhabit primary and secondary rainforests (<u>Ceotto et al.</u>)

2009; Macedo et al. 2010; Voss et al. 2018). Although some species (e.g., Philander opossum Linnaeus, 1758) have a diet consisting of 80 % arthropods, the species within the genus are considered opportunistic vertebrate predators (Charles-Dominique et al. 1981; Leite et al. 1994). Information about the natural history, ecology, and behavior of several species of this genus is still scarce (Vieira and Moraes 2003). Regarding their feeding habits, there is some information

available about *P. frenatus* (Olfers, 1818) (<u>Cáceres 2004</u>), *P. opossum* (<u>Castro-Arellano *et al.* 2000; <u>Gómez-Martínez *et al.* 2008</u>), and *P. andersoni* Osgood, 1913 (<u>Niño-Reyes *et al.* 2020; <u>Carrasco-Quiñones 2021</u>), while for *P. pebas* <u>Voss *et al.* 2018</u>, information is scarce or absent.</u></u>

The species *Philander pebas* (Pebas four-eyed opossum) was recently described and named (2018), being recorded in eastern Ecuador, eastern Perú, and Amazonian Brazil. It coexists sympatrically in its distribution areas with *P. ander*soni (black four-eyed opossum), P. canus Osgood, 1913 (common four-eyed opossum), and P. mcilhennyi Gardner and Patton, 1972 (McIlhenny's four-eyed opossum). This Amazonian species can be promptly distinguished from all other Philander species by its dental morphology, size, and pigmentation. Its dorsal pelage is uniformly grayish, occasionally darker along the mid-dorsal region than on the flanks but lacks a distinct blackish stripe along the dorsum. The fur on the crown is grizzled-grayish, often quite dark but seemingly never jet black. The ventral fur is mostly gray-based, frequently appearing in cream or buff tones in the inguinal region. The scaly portion of the tail shows a white area not exceeding one-quarter of its length (Voss et al. 2018). Philander pebas primarily inhabits floodplain forest habitats, and it is predominantly found in riparian formations seasonally flooded by white-water rivers, known as "várzea" forests. It also occupies secondary growth areas and swamps with permanently waterlogged soils. In contrast, sympatric congeners typically inhabit upland (unflooded) primary forests (Voss et al. 2018, 2019).

The Rhinella margaritifera (Laurenti, 1768) (South American common toad) species group has one of the most complex histories in the systematics of Neotropical anurans, as a result 17 recognized species for the group (Dos Santos et al. <u>2015</u>). *Rhinella margaritifera* is a large bufonid with a wide distribution throughout the Amazon basin, encompassing Colombia, Venezuela, Perú, Bolivia, Brazil, and Guyana. This terrestrial species exhibits both diurnal and nocturnal activity and inhabits the leaf litter of both terra firme and várzea forests (Toft and Duellman 1979). It features a pointed snout, well-developed cranial crests, a prominent bony protrusion at the jaw articulation, a row of conical tubercles continuing along the flank and conspicuous parotoid glands (Fouguet et al. 2007; Ávila et al. 2010; Pereyra et al. 2021). Parotoids are multiglandular structures involved in chemical defense of toad. Secretions of the granular cutaneous parotoids may be very toxic to animals (Toledo and Jared 1995; Clark 1997). In bufonids they contain steroids such as bufogenines and bufotoxins that, when in contact with the buccal mucosa of many vertebrates, have cardioacceloratory properties increasing the strength of the heart beat and decreasing heart rate (Habermehl 1981).

Here, we present the first documented case of predation on the South American common toad by Pebas four-eyed opossum (*P. pebas*) in the Amazonian floodplain forest of Brazil. In addition to contributing to the understanding of *P. pebas* dietary habits and habitat preferences. The observation documented here was made on February 13, 2023, at 20:35 hr, in the rainy season, during fieldwork along a trail through floodplain forest in the Área de Relevante Interesse Ecológico Japiim-Pentecoste, coordinates are 07° 37' 20" S, 72° 47' 69" W, Municipality of Cruzeiro do Sul, Acre State, Brazil (Figure 1). The várzea forest in this region experiences seasonal flooding. Vegetation is characterized by a closed canopy of trees reaching an average height of 20 m and emergent trees up to 35 m. The forest floor is marked by the abundant Poaceae, Arecaceae, and small trees, with relatively fewer herbaceous plants such as Heliconiaceae, Marantaceae, and Costaceae (Bernarde *et al.* 2013). We obtained photographic records using a Nikon 7500 camera with lens 300 mm and the help of Headlamps with 500 lumens.

We encountered 1 specimen of P. pebas moving through a tangle of lianas and palm leaves at an approximate distance of 5 m. It continued climbing the lianas until it stopped and observed us from a height of approximately 4 m. At that moment, we observed the opossum with a R. margaritifera, securely held in its mouth (Figure 2). Pebas foureyed opossum gripped the toad dorsally by the widest part of its head and ventrally by part of its neck, encompassing the parotoid glands. As we approached and started taking photos, the animal remained motionless. We noticed that R. margaritifera in its mouth was alive, displayed an occasional slow movement of its hind legs and defensive behavior known as "puffing up the body", this behaviour consists of filling the lungs with air, and thus enlarging the frog's size (Toledo et al. 2011), being able intimidate a potential predator or avoid subjugation after being captured. After 10 min of observation, the opossum resumed its movement, climbing the lianas and disappeared into the forest canopy.



Figure 1. Location of record of predation (red point) on the South American common toad by Pebas four-eyed opossum in Acre State, Brazil.

Negreiros Almeida et al.

Despite the knowledge gap concerning the feeding habits of most Philander species, it is known that fruits and invertebrates are important components of the diet for most species, with occasional records of vertebrates such as rodents, lizards, birds, and snakes (Leite et al. 1994; Santori et al. 1997; Carvalho et al. 1999). In fact, some studies have suggested that the genus Philander is one of the most carnivorous among neotropical marsupials (Santori et al. 1997; Vieira and Moraes 2003). Enders (1935) reported that P. opossum preferred meat to fruits in captivity. These marsupials are known to be scansorial and highly opportunistic; however, as far as we know, there are few records of hunting and consumption of amphibians by a neotropical marsupial (Julien-Laferriere and Atramentowicz 1990; Castro-Arellano et al. 2000). Philander pebas is frequently observed near the ground and in forest understory, feeding on invertebrates and fruits. This foraging method favors opportunistic encounters with diurnal terrestrial anurans (e.g., Rhinella gr. margaritifera), which at night climb low vegetation and remain inactive (Toft and Duellman 1979). In this floodplain forest, R. margaritifera is one of the most abundant amphibian species, occupying a variety of substrates. It is also the most frequently encountered species during nocturnal surveys in the Área de Relevante Interesse Ecológico Japiim-Pentecoste, resting on vegetation at heights of up to 2.10 m (Miranda et al. 2015; Fonseca et al. 2021).

Due to the abundance of R. margaritifera and its occurrence across various substrates, it is possible that the predation of this species by the marsupial P. pebas is not as opportunistic as other records of vertebrate prey in the diet of the Philander genus. Individuals of R. margaritifera climb to low vegetation up to above the ground at night where they remain inactive, being an abundant specie during the rainy season (Moravec et al. 2014), which would facilitate the encounter by *P. bebas*. Bufonids produce highly potent toxins in their skin, especially concentrated in the parotoid glands, they repel and may even kill predators (Sinhorin et al. 2020). However, unlike venomous animals, these amphibians lack the apparatus to inject the toxins. The toads, inflate their lungs and also assume special postures to present the parotoids to the source of danger (Jared et al. 2014; Regis-Alves et al. 2017). Intoxication occurs when the aggressor bites the toad, but to spray their venom only in response to physical pressure, making contact with the toxins through the oral mucosa (e.g., Toledo and Jared 1995). Nevertheless, as described in this record, P. pebas was able to bite and hold the dorsal region of the toad, where the large parotoid glands are located and added the defensive behavior (puffing up the body), it seems evident that the toad depends on the parotoid for its defense. Even so, this anti-predator mechanism appears to be ineffective against individuals of P. pebas. In the case of a bite, after venom ingestion, the potential predator could show visible reactions, e.g., intense salivation and excitation, paralysis, trembling and convulsions, often leading to death (Sakate and Lucas de Oliveira 2000; Sonne et al. 2008).



**Figure 2.** Register of *Philander pebas* with the frog *Rhinella margaritifera* held in its mouth at the Área de Relevante Interesse Ecológico Japiim Pentecoste, municipality of Cruzeiro do Sul, Acre, Brazil. Image available at <a href="mailto:rafaelbio2011@gmail.com">rafaelbio2011@gmail.com</a>.

Thus, apparently during our observation, the toxins expelled by the toad were ineffective in deterring a didelphid marsupial. This observation strongly suggests that *P. pebas* have a tolerance for parotid gland secretions from *R. margaritifera*, allowing the opossums to prey on these amphibians. Furthermore, some venom-resistant mammals, many of which are ophiophagous, also show resistance to ingested animal toxins (Voss and Jansa 2012). Therefore, given its opportunistic feeding behavior, occasional predator, and apparent ability to tolerate highly potent toxins, encountering one of the most common and abundant toads during the rainy season, presents an excellent feeding opportunity. This combination of traits allows species of *Philander* to take advantage of a wide range of food sources, potentially contributing to its success as a species.

Although this is a singular observation of predation, we present the first evidence of relatively toxic toad being preyed upon by *P. pebas*, suggesting the need for additional studies on the predatory behavior of these marsupials and how they can adapt to consume a variety of prey. Thus, such information is valuable in expanding our knowledge about the potential breadth of food items for didelphids, specifically regarding the consumption of poisonous animals. It increases our understanding of these species in their inhabited areas, including how they interact with other species and raises questions about the extent of immunity these marsupials have against other species.

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