**Nota**

Cuad. herpetol. 40 (1): 00-00 (*2026)*

# New record of leg-interweaving behavior in *Boana polytaenia* (Cope, 1870) (Anura: Hylidae): an expansion of the antipredator repertoire in Neotropical anurans

**Juliana Midori Kobayashi Cunha[1,3], Vinícius Frederico Russo[1,3], Luana Camargo Matos[1,3], Paulo Henrique Correa Dória[1], Marcelo José Sturaro[1,2]**

[1] Universidade Federal de São Paulo, Instituto de Ciências Ambientais, Químicas e Farmacêuticas (ICAQF), Departamento de Ecologia e Biologia Evolutiva, Av. Professor Artur Riedel, 275, Jardim Eldorado, Diadema, CEP 09972-270, São Paulo, Brazil

[2] Museu de Zoologia da Universidade de São Paulo (MZUSP), Av. Nazaré, 481, CEP 04263-000, São Paulo, SP, Brazil.

[3] Programa de Pós-graduação em Ecologia e Evolução, Universidade Federal de São Paulo, Instituto de Ciências Ambientais, Químicas e Farmacêuticas (ICAQF), Av. Professor Artur Riedel, 275, Jardim Eldorado, Diadema, CEP 09972-270, São Paulo, Brazil

Recibida: 01 Septiembre 2025
Revisada: 09 Diciembre 2025
Aceptada: 02 Febrero 2026
Editor Asociado: S. Quinzio

doi: 10.31017/CdH.2026.(2025-035)

ABSTRACT

Amphibians exhibit a wide range of antipredator behaviors, such as thanatosis, alarm vocalizations, and distraction displays, aimed at reducing predation risk. Leg-interweaving is a rare antipredator behavior, characterized by crossing the hind limbs over the body. Here, we report the first record of this behavior for *Boana polytaenia*, which represents the first documented occurrence within the genus. It is thought to represent a displaced behavior in an out-of-context response to stress rather than a true defense mechanism, although it is discussed to have a potential role in enhancing aposematic displays, reproductive signals, chemical defenses, physical obstruction against predation, or body pattern distortion, favoring camouflage. The inconsistent occurrence and scarce reports of leg interweaving emphasize the importance of further experimental studies to determine its functional role and ecological relevance, thereby enriching our understanding of the evolution and functional diversity of defensive strategies in anurans.

Key words: Legs-interweaving; Anuran behavior; Defensive strategies.

RESUMEN

Los anfibios exhiben una amplia gama de comportamientos antidepredadores, como la tanatosis, las vocalizaciones de alarma y las exhibiciones de distracción, destinados a reducir el riesgo de depredación. El entrelazamiento de las piernas es un comportamiento antidepredador poco común, caracterizado por el cruce de las extremidades posteriores sobre el cuerpo. Aquí, reportamos el primer registro de este comportamiento para *Boana polytaenia*, lo que representa la primera ocurrencia documentada dentro del género. Se cree que representa un comportamiento desplazado en una respuesta de estrés fuera de contexto, en lugar de un mecanismo de defensa real, aunque se discute su papel potencial en la mejora de las exhibiciones aposemáticas, las señales reproductivas, las defensas químicas, la obstrucción física contra la depredación o la distorsión del patrón corporal, favoreciendo el camuflaje. La ocurrencia inconsistente y los escasos reportes de entrelazamiento de piernas enfatizan la importancia de realizar más estudios experimentales para determinar su papel funcional y relevancia ecológica, enriqueciendo así nuestra comprensión de la evolución y la diversidad funcional de las estrategias defensivas en los anuros.

Palabras claves: Entrelazamiento de piernas; Comportamiento de anuros; Estrategias defensivas.

Author for correspondence: jmk.cunha@unifesp.br

Animal behavior encompasses the observable responses of an organism to internal or external stimuli, representing a direct product of natural selection that reflects adaptations enhancing survival and reproductive success (Tinbergen, 1963; Alcock, 2013). These responses variate from simple motor responses to complex behavior patterns, such as communication, parental care, and predator avoidance. As a central component of biology, behavior is particularly diverse in vertebrates, where it is shaped by multiple selective pressures, among which predation represents one of the most intense (Lima & Dill, 1990). Among anurans, behavioral diversity is exceptionally pronounced. Extensive research has documented intricate strategies in reproductive behavior (Wells, 2007), territorial defense (Toledo *et al.*, 2015), acoustic communication (Gerhardt & Huber, 2002), and, notably, antipredator behavior (Toledo & Haddad, 2009; Caro, 2014; Ferreira *et al.*, 2019). Given their heightened vulnerability to predators across life stages, anurans have evolved a wide array of defensive mechanisms, including morphological, physiological, and behavioral traits (Duellman & Trueb, 1994; Toledo *et al.*, 2011; Mukherjee & Heithaus 2013).

In the context of antipredator behavioral, strategies that reduce the likelihood of detection, capture, or consumption by predators are particularly significant (Feder & Lauder, 1986). Many of these behaviors have evolved under long-term selective pressures, favoring individuals with effective responses to threat (Edmunds, 1974; Ruxton *et al.*, 2004). Documented antipredator strategies include thanatosis (feigning death), body inflation, alarm vocalizations, legs-interweaving, distraction display, among others (Toledo *et al.*, 2010; Toledo *et al.*, 2011). These behaviors often function synergistically with morphological adaptations, such as aposematic coloration or mimicry, enhancing their overall defensive efficacy (Skelhorn & Rowe, 2006). For instance, thanatosis, widely reported in amphibians, involve complete immobility to deceive predators, proving especially effective against visual predators or those that rely on movement cues (Toledo *et al.*, 2010; Ramalho *et al.*, 2019). Body inflation, a common trait in many Bufonidae species, increases the animal apparent body volume, making it harder for predator to grasp or ingest (Moroti *et al.*, 2018; Ferreira *et al.*, 2019). In contrast, legs-interweaving is a rare and less-studied behavior, documented in only few families of frogs (e.g., Bufonidae, Craugastoridae, Hylidae, Hyperolidae, and Leptodactylidae). Its function remains debated: it may serve as an active defense mechanism or represent displaced behavior, an out of-context response to stress (Velasco *et al.*, 2025).

Leg-interweaving behavioral was observed in an adult male of *Boana polytaenia* at the Instituto Alto Montana da Serra Fina, Itamonte municipality, Minas Gerais state, Brazil. The specimen was collected on April 12 2025 at 20:20 by the senior author (SISBIO Permit # 66852-7) while vocalizing 20 cm above water in grasses, in the Pinhão Assado waterfall (-22.358591, -44.797457). During photographic documentation prior to euthanasia, the individual jumped from a surface, falling approximately 10–15 cm. Immediately after impact, it exhibited leg-interweaving behavior, characterized by crossing its hind limbs over the body and exposing the orange-colored inner thigh surfaces. This posture was maintained for approximately two minutes without any additional stimulus before the animal spontaneously resumed normal movement. The euthanasia was conducted using specimen lethal dose of sodium thiopental. Subsequently, the specimen was fixed in 4% formalin solution for 24 h and, later, transferred to 70% ethanol for long-term preservation. The specimen was deposited in Coleção Teresa Cristina Sauer de Avila-Pires (CHTC 1591), Universidade Federal de São Paulo, campus Diadema, São Paulo, Brazil.

This record represents the first documented occurrence of leg-interweaving behavior in the genus *Boana*, representing the thirteenth recorded anuran species to exhibit this rare antipredator strategy (Velasco *et al.*, 2025). Beyond expanding the known taxonomic distribution of this behavior, this finding holds particular significance in understanding origins and functional diversity of defensive mechanisms in Neotropical anurans. The presence of leg-interweaving in *B. polytaenia* suggests either convergent evolution of this strategy or a potentially wider phylogenetic distribution among hylids than previously recognized.

Aposematic coloration consists of conspicuous visual patterns that typically signal cutaneous toxins in amphibians, warning predators of an individual's unpalatability or danger (Toledo *et al.*, 2010; Ferreira *et al.*, 2019). These displays commonly employ high-contrast hues (e.g. red, orange, yellow, or blue) that enhance predator recognition and memory (Toledo & Haddad, 2009; Protti, 2019). Many anurans exhibit such coloration only on normally concealed

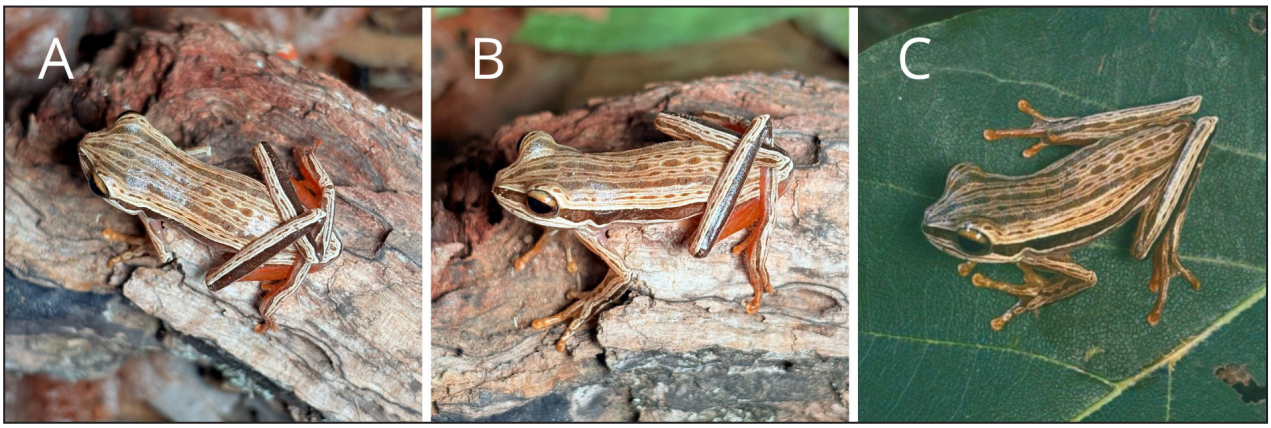


**Figure 1.** Male *Boana polytaenia* exhibiting leg-intertwining behavior. Dorsal and lateral views of the behavior observed on a wooden substrate, highlighting the display of bright orange inner coloration on the hind limbs (A–B). Same individual at rest, with hind limbs in a natural position and inner coloration not visible (C).

body regions (e.g., posterior thighs, belly, or limb surfaces), revealing them through specific defensive postures like body elevation or leg-interweaving (Mallet & Joron, 1999; Toledo & Haddad, 2005). In *B. polytaenia*, the bright orange inner thighs and tibiae remain hidden at rest but become highly visible during leg-interweaving behavior (Fig. 1A–B). This dynamic display likely enhances aposematic signaling while minimizing continuous exposure to predators (Rößler *et al.*, 2019; Barnett *et al.*, 2023). However, conspicuous coloration may also increase detection risk, creating an evolutionary trade-off between warning efficacy and predator attraction (Hall *et al.*, 2013; Rößler *et al.*, 2019). This paradox explains why many species combine aposematic with cryptic resting posture, a strategy that optimizes the balance between concealment and warning (Toledo & Haddad, 2009; Ferreira *et al.*, 2019; Pedroso-Santos *et al.*, 2022).

Bright coloration on the internal regions of anuran hind limbs has been interpreted as serving (1) a distraction mechanism during escape behaviors (e.g. rapid jumping), in which sudden flashes of color may disorient predators (Toledo *et al.*, 2010; Ramalho *et al.*, 2019), and (2) a potential visual signal in reproductive contexts. Alternatively, these color patterns may specifically enhance the efficacy of leg-interweaving behavior. When the limbs are crossed over the body in this distinctive posture, colors exposure create maximal visual contrast, potentially amplifying the aposematic signal or startling effects (Toledo *et al.*, 2011).

Beyond its potential role in enhancing aposematic displays and reproductive signal, leg-interweaving behavior may serve additional, yet understudied, defensive function. One proposed mechanism involves physical obstruction: by crossing its limbs over the body, the frog increases its effective volume and interference with predator jaws mechanisms, especially for predator that employ rapid gape-limited attacks (Wells, 2007; Ruxton *et al.*, 2019). This strategy parallels other anurans defenses, including thanatosis (death feigning) and body inflation, which similarly complicate the prey handling. (Toledo *et al.*, 2011; Gally *et al.*, 2014). A second hypothesis suggests disruption of predator search images through visual pattern distortion (Toledo & Haddad, 2009; Ramos *et al.*, 2021). Visual predators often rely on prey silhouettes for recognition (Wells, 2007; Zlotnik *et al.*, 2018), and the anomalous posture by leg-interweaving may break the frog's characteristic outline. In species like *B. polytaenia*, which exhibits longitudinal dorsal stripes, this effect could be intensified, as the behavior disrupts these linear patterns into discontinuous segments (Velasco *et al.*, 2025), potentially delaying predator identification. A third, chemically mediated function may involve the release of defensive skin secretions (Dreher *et al.*, 2015). Although undocumented in *B. polytaenia*, Velasco *et al.* (2025) demonstrated that some anurans release bioactive peptides through the skin during stress responses. Limb movements during interweaving may mechanically stimulate dermal glands, facilitating secretion release while simultaneously exposing warning coloration, thus forming a synergistic defensive strategy (Amézquita *et al.*, 2017; Velasco *et al.*, 2025). These peptides, stored in granular glands, function as both as antipredator

and antimicrobial agents, offering protection during vulnerable defensive postures.

This observation represents the first documented case of leg-interweaving behavior in the genus *Boana*, significantly expanding our knowledge of this poorly documented antipredator strategy in anurans. The behavior's close association with sudden display of otherwise vibrant coloration reinforces its function as a complex defensive mechanism. The context-dependent nature of this color exposure - occurring exclusively during leg-interweaving - suggests an adaptive specialization for maximizing visual deterrence during predator encounters. The coordinated presentation of postural modification, dynamic color display, and potential chemical secretion release indicates an integrated defensive response. This finding highlights important questions requiring experimental investigation, particularly regarding the behavior's effectiveness against different predator types, its associated biochemical components, and the potential evolutionary trade-offs involved.

With only twelve previously documented species exhibiting this behavior, this record of *B. polytaenia* provides valuable comparative data for understanding the evolution and functional diversity of defensive strategies in anurans. The scarcity of field observations of leg-interweaving emphasizes the need for further research to clarify its adaptive significance and ecological context. This discovery not only adds to the known taxonomic distribution of this behavior but also enhances our understanding of the sophisticated antipredator adaptations employed by anurans under predation pressure.

## Acknowledgments

We thank the Instituto Alto Montana da Serra Fina, especially Vinícius Coruja, for their continued hospitality and for providing the space for fieldwork. We are also thankful to João Victor Gomes, Letícia Palomino, Jean de Carvalho, Yago Amador, Gabriela Takeuchi, Patrícia de Castro and Helena Slama for the assistance in the field. JMKC were financially supported by the Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP, process # 2025/03442-1). VFR were financially supported by the Fundo Nacional de Desenvolvimento da Educação - Programa de Educação Tutorial (PET), and the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, process # 143395/2023–8). LCM acknowledges fellowship support from CAPES (Coordenação de Aperfeiçoamento de Pessoal de Nível Superior; Grant # 88887.939777/2024–00). MJS and the study were financially supported by the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, process # 434362/2018–2).

## Literature cited

Alcock, J. 2013. Animal Behavior: An Evolutionary Approach (10th ed.). Sinauer Associates. Sunderland, MA, USA.

Amézquita, A.; Ramos, O.; González, M.C.; Rodríguez, C.; Medina, I.; Simões, P.I. & Lima, A.P. 2017. Conspicuousness, color resemblance, and toxicity in geographically diverging mimicry: the pan-Amazonian frog *Allobates femoralis*. *Evolution* 71: 1039-1050.

Barnett, J.B.; Yeager, J.; McEwen, B.L.; Kinley, I.; Anderson, H.M. & Guevara, J. 2023. Size-dependent colouration balances conspicuous aposematism and camouflage. *Journal of Evolutionary Biology* 36: 1010-1019.

Caro, T. 2014. Antipredator deception in terrestrial vertebrates. *Current Zoology* 60: 16-25.

Dreher, C.E.; Cummings, M.E. & Pröhl, H. 2015. An analysis of predator selection to affect aposematic coloration in a poison frog species. *PLoS ONE* 10: e0130571

Duellman, W.E. & Trueb, L. 1994. Biology of Amphibians. Johns Hopkins University Press. Baltimore, Maryland, USA.

Edmunds, M. 1974. Defence in animals: a survey of anti-predator defences. Longman. New York, USA.

Feder, M.E. & Lauder, G.V. 1986. Predator-prey relationships: perspectives and approaches from the study of lower vertebrates. University of Chicago Press. Chicago, USA.

Ferreira, R.B.; Lourenço-de-Moraes, R.; Zocca, C.; Duca, C.; Beard, K.H. & Brodie, E.D. 2019. Antipredator mechanisms of post-metamorphic anurans: a global database and classification system. *Behavioral Ecology and Sociobiology* 73: 1-21.

Ferreira, R.B.; Martins, I.A. & Giaretta, A.A. 2019. Death-feigning and leg-twisting in *Boana faber* (Anura: Hylidae): uncommon anti-predator behaviors in frogs. *Phyllomedusa* 18: 111-114.

Gally, M.; Zina, J.; Mira-Mendes, C.V. & Solé, M. 2014. Legs-interweaving: an unusual defense behaviour of anurans displayed by Agalychnis aspera (Peters, 1983). *Herpetology Notes* 7: 623-625.

Gerhardt, H.C. & Huber, F. 2002. Acoustic Communication in Insects and Anurans: Common Problems and Diverse Solutions. University of Chicago Press. Chicago, USA.

Hall, J.R.; Cuthill, I.C.; Baddeley, R.; Shohey, A.J. & Scott-Samuel, N.E. 2013. Camouflage, detection and identification of moving targets. *Proceedings of the Royal Society of B, Biological Sciences* 280: 20130064.

Lima, S.L. & Dill, L.M. 1990. Behavioral decisions made under the risk of predation: a review and prospectus. *Canadian Journal of Zoology* 68: 619-640.

Mallet, J. & Joron, M. 1999. Evolução da diversidade em cores de alerta e mimetismo: polimorfismos, equilíbrio variável e especiação. *Revisão anual de ecologia e sistemática* 30: 201-233.

Moroti, M.T.; Sestito, G.; Pedrozo, M.; Machado, I.F.; Santana, D.J. 2018. Defensive behaviours in *Bokermannohyla luctuosa*

(Pombal and Haddad, 1993) (Anura, Hylidae). *Herpetology Notes* 11: 233-237.

Mukherjee, S. & Heithaus, M.R. 2013. Dangerous prey and daring predators: a review. *Biological reviews* 88: 550-563.

Pedroso-Santos, F.; Sanches, P.R. & Costa-Campos, C.E. 2022. Defensive behaviours and colour patterns displayed by frogs in two rainforest areas of the northern Brazilian Amazon, with comments on mimicry patterns. *Herpetology Notes* 15: 153-164.

Protti Sánchez, F. 2019. Aposematismo en la rana *Phyllobates vittatus* (Anura: Dendrobatidae): integrando defensas, señales y riesgo de depredación. Universidad de Costa Rica.

Ramalho, W.P.; Guerra, V.; Ferraz, D.; Machado, I.F. & Vieira, L.J.S. 2019. Observations on death-feigning behaviour and colouration patterns as anti-predator mechanisms in Amazonian anurans. *Herpetology Notes* 12: 269-272.

Ramos, M.E.B.; Souza, U.F.; Dubeux, M.J.M. & Mott, T. 2021. Repertoire of antipredator mechanisms in the Brazilian toad *Frostius pernambucensis* (Anura: Bufonidae). *Phyllomedusa: Journal of Herpetology* 20: 209-213.

Rößler, D.C.; Lötters, S.; Mappes, J.; Valkonen, J.K.; Menin, M.; Lima, A.P. & Pröhl, H. 2019. Sole coloration as an unusual aposematic signal in a neotropical toad. *Scientific Reports* 9: 1128.

Ruxton, G.D.; Sherratt, T.N. & Speed, M.P. 2004. Avoiding Attack: The Evolutionary Ecology of Crypsis, Warning Signals and Mimicry. Oxford University Press. Oxford, UK.

Skelhorn, J. & Rowe, C. 2006. Avian predators taste–reject aposematic prey on the basis of their chemical defence. *Biology Letters* 2: 348-350.

Tinbergen, N. 1963. On aims and methods of ethology. *Zeitschrift für tierpsychologie* 20: 410-433.

Toledo, L.F. & Haddad, C.F.B. 2009. Colors and some morphological traits as defensive mechanisms in anurans. *International Journal of Zoology* 2009: 1-12.

Toledo, L.F.; Ribeiro, R.S. & Haddad, C.F.B. 2015. The anuran calling repertoire in the light of social context. *Acta Ethologica* 18: 87-99.

Toledo, L.F.; Sazima, I. & Haddad, C.F.B. 2010. Is it all death feigning? Case in anurans. *Journal of Natural History* 44: 1979-1988.

Toledo, L.F.; Sazima, I. & Haddad, C.F.B. 2011. Behavioural defences of anurans: an overview. *Ethology Ecology & Evolution* 23: 1-25.

Toledo, L.F. & Haddad, C.F.B. 2005. Reproductive biology of *Scinax fuscomarginatus* (Anura, Hylidae) in south-eastern Brazil. *Journal of Natural History* 39: 3029-3037.

Velasco, I.G.; De Oliveira, J.V.A.; Da Silva Maia, N.V.N.; Silva, S.N.; De Oliveira Braga, J.F. & Simões, P.I. 2025. Limbs interweaving for Neotropical anurans. *Cuadernos de Herpetología* 39: 41-45.

Wells, K.D. 2007. The Ecology and Behavior of Amphibians. University of Chicago Press. Chicago, Illinois, USA.

Zlotnik, S.; Darnell, G.M. & Bernal, X.E. 2018. Anuran predators overcome visual illusion: dazzle coloration does not protect moving prey. *Animal Cognition* 21: 729-733.