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A recently extinct new species of *Dryadobates* (Anura: Aromobatidae) from South Brazil: species description and implications for the historical distribution and recent extinction history of the clade

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Abstract

We describe a new species of *Dryadobates* on the basis of a single adult male collected in 1963 in Tarumã, Curitiba, Paraná, Brazil. Although the specimen is badly desiccated and extremely fragile, it can be readily assigned to Dendrobatoidea on the basis of its posterodorsally concealed tympanum, phalangeal swelling on finger IV, and tarsal keel. The new species shares with other species of *Dryadobates* the same morphology of finger discs II, IV, and V, absence of a metatarsal fold, presence of a complete pale oblique lateral stripe, absence of a pale dorsolateral stripe, presence of basal webbing between toes III–IV (absent between all other toes), presence of pale paracloacal marks, and small size. It is diagnosed from all nominal congeners by possessing a conspicuous pale oblique lateral stripe that is discrete and well defined along its entire length (i.e., not anteriorly indistinct). The type locality lies ca. 550 km south of the nearest congener in Angra dos Reis, Rio de Janeiro, suggesting the existence of additional *Dryadobates* populations between these localities. However, despite extensive surveys in suitable habitats at multiple localities in São Paulo and Paraná over the past 50 years, no other specimens of *Dryadobates* have been collected. Further, the region of the type locality—once characterized by forests, streams, and expansive fields and wetlands—has been transformed into a highly developed residential and commercial area lacking suitable habitat for *Dryadobates*, leading us to presume this species to be extinct. With the current species, *Dryadobates* now comprises seven described species, four of which disappeared within a 15–20-year window between the mid-1960s and mid-1980s. The drivers of these recent extinctions remain unclear, underscoring the urgent need to gather data on the natural history, distribution, and population status of extant *Dryadobates* species to inform effective conservation strategies.

Key words: Amphibia, Dendrobatoidea, Neotropics, Nurse frogs, Rocket frogs, Systematics, Taxonomy

Resumo

Descrevemos uma nova espécie de *Dryadobates* com base em um único macho adulto coletado em 1963 em Tarumã, Curitiba, Paraná, Brasil. Apesar de o espécime estar severamente desidratado e extremamente frágil, pode ser prontamente atribuído a Dendrobatoidea com base em seu tímpano oculto posterodorsalmente, inchaço falângico no quarto dedo da mão e quilha tarsal. A nova espécie compartilha com outras espécies de *Dryadobates* a mesma morfologia dos discos manuais dos dedos II, IV e V, a ausência de dobra metatarsal, a presença de uma faixa oblíqualateral pálida completa, a ausência de faixa dorsolateral pálida, a presença de membrana interdigital basal entre os dedos III–IV dos pés (e ausência entre todos os demais dedos), a presença de marcas paracloacais claras e o pequeno porte. Distingue-se de todos os congêneres nominais por apresentar uma conspícua faixa lateral oblíqua pálida, discreta e bem definida ao longo de toda a sua extensão (i.e., não indistinta anteriormente). A localidade-tipo situa-se a cerca de 550 km ao sul do congênere mais próximo, Angra dos Reis, Rio de Janeiro, o

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que sugere a possível existência de populações adicionais de *Dryadobates* entre essas regiões. No entanto, apesar de levantamentos extensivos em habitats adequados em diversas localidades de São Paulo e Paraná ao longo dos últimos 50 anos, nenhum outro espécime de *Dryadobates* foi coletado. Além disso, a região da localidade-tipo—originalmente composta por florestas, riachos, campos abertos e áreas alagadas—foi transformada em uma zona residencial e comercial densamente urbanizada, desprovida de habitat adequado para *Dryadobates*, levando-nos a presumir que esta espécie esteja extinta. Com a espécie aqui descrita, *Dryadobates* passa a contar com sete espécies descritas, das quais quatro desapareceram em um intervalo de 15 a 20 anos, entre meados da década de 1960 e meados da década de 1980. As causas dessas extinções recentes permanecem desconhecidas, ressaltando a necessidade urgente de se obter dados sobre a história natural, distribuição e estado populacional das espécies ainda existentes de *Dryadobates*, a fim de embasar estratégias eficazes de conservação.

Introduction

Dryadobates Grant *et al.*, 2025 was recently described for the Atlantic Forest clade of aromobatid nurse frogs and includes *D. alagoanus* (Bokermann, 1967), *D. bokermanni* Grant *et al.*, 2025, *D. capixaba* (Bokermann, 1967), *D. carioca* (Bokermann, 1967), *D. lutzi* Grant *et al.,* 2025, and *D. olfersioides* (Lutz, 1925a). *Dryadobates* is also the only dendrobatoid taxon known to occur in the Atlantic Forest. Despite extensive fieldwork in southern Rio de Janeiro, São Paulo, and Paraná over the last 50 years (for summaries see Rossa-Feres *et al.* 2011, 2018; Vancine *et al.* 2018; Dorigo *et al.* 2018; Santos-Pereira *et al.* 2018), the southernmost confirmed locality for *Dryadobates* is Mambucaba, in the municipality of Angra dos Reis, RJ (23°1'S 44°30'W; Verdade & Rodrigues 2007; Grant *et al.* 2025). It was therefore highly unexpected to find a record in the Smithsonian National Museum of Natural History of a specimen, USNM 148487, identified as "*Phyllobates brunneus*" (Cope, 1887) (considered the senior synonym of *Eupemphix olfersioides* by Cochran 1955; see Grant *et al.* 2025) from Tarumã, Curitiba, PR, as this locality lies nearly 550 km southwest of Angra dos Reis.

Although Santos-Pereira *et al.* (2018) did not examine USNM 148487 in their study of the anurans of Paraná, they understandably dismissed the record as "almost certainly erroneous." We shared their skepticism, assuming it to be a misidentified juvenile *Crossodactylus* Duméril & Bibron, 1841 or *Hylodes* Fitzinger, 1826 (Hylodidae Günther, 1858), which are broadly distributed throughout the region (Santos-Pereira *et al.* 2018) and easily confused with *Dryadobates*. However, although the specimen possesses paired dorsal digital scutes, which is consistent with Hylodidae, those structures are also present in Dendrobatoidea. The specimen is too fragile to assess the condition of the thigh and jaw musculature; however, as in all dendrobatoids (e.g., Myers & Ford 1986; Grant *et al.* 2006) the posterodorsal portion of the tympanic annulus is concealed (presumably by the m. depressor mandibulae), whereas the tympanic annulus is superficial to the m. depressor mandibulae and entirely visible externally in hylodids. Additionally, USNM 148487 possesses phalangeal swelling on finger IV, which is unique to Dendrobatoidea (Cavalcanti *et al.* 2022), and a well-defined, short, strongly curved, tubercle-like tarsal keel, whereas all hylodids possess a tarsal fringe, defined as a conspicuous dermal flap running along the entire length of the preaxial edge of the tarsus (Grant *et al.* 2006).

Having failed to refute the taxonomic identity of the specimen, we assumed the locality must have been recorded incorrectly; however, we also failed to uncover documentary evidence to support that hypothesis. The USNM catalogue records USNM 148487 as having been collected 9 January 1963 by the entomologist Doris H. Blake and batrachologist Doris M. Cochran—both experienced collectors—together with a single individual of *Physalaemus cuvieri* Fitzinger, 1826 (USNM 148137). Cochran's unpublished travelogue, deposited in the Smithsonian Institution Archives under accession number SIA 007151, documents Blake and Cochran's travels in Brazil, beginning with their flight from New York to Brasília on 3 December 1962 and ending with their flight from Foz do Iguaçú to Buenos Aires on 11 January 1963. Prior to arriving in Curitiba, Blake and Cochran were in Rio de Janeiro 5–26 December 1962, when they could have collected USNM 148487 and subsequently mislabeled the locality; however, Cochran's notes only mention live frogs in Rio de Janeiro when they visited a "lovely pond where the smith frog breeds" and observed "tiny toads and treefrogs under the grass around [the pond]" (22 December 1962) and "*Hyla circumdata* sleeping in a deep cistern […] and a very dark juv. *Eleuth*. *guentheri*" (25 December 1962). Additionally, USNM 148487 differs conspicuously from any known species from the area of Rio de Janeiro (see below), and Blake and Cochran did not visit Amazonian localities where *Allobates* Zimmermann & Zimmermann, 1988, the sister group of *Dryadobates*, could have been collected. In contrast, Cochran's notes explicitly state that they "caught two frogs" in a "big field full of anthills and cow-chewed grass clumps" on 9 January 1963 at Tarumã, Curitiba, which is consistent with the entry in the USNM catalogue. Consequently, there is no basis to reject the record as erroneous.

Below we describe the new species of *Dryadobates* from Tarumã and discuss the implications of this surprising new record for the biogeography and history of recent extinctions of the clade.

Materials and Methods

Given the fragility of USNM 148487, we inferred the sex and maturity of this specimen from the phalangeal swelling on finger IV, which is exclusive to adult males (Cavalcanti *et al.* 2022), and the well-developed left vocal slit. For comparative material, we scored males with vocal slits on both sides of the mouth and enlarged testes as adults, those with only one vocal slit and moderate sized testes as subadults, and those lacking slits on both sides as juveniles. We considered females with expanded, convoluted oviducts and enlarged oocytes to be adults, those with only weakly expanded, non- or weakly convoluted oviducts and poorly differentiated oocytes to be subadults, and those with small, undifferentiated oocytes and unexpanded, straight oviducts to be juveniles. For hand morphology, we follow Fabrezi & Alberch (1996) in considering finger I of other tetrapods to be absent in Anura and number fingers accordingly. As such, we follow Grant *et al.* (2017) and Cavalcanti *et al.* (2022) in referring to the swollen third finger of earlier literature (e.g., Grant *et al.* 2006) as swollen finger IV. We used the method of Kaplan (1997) to evaluate the relative lengths of fingers II and III, and we follow Anganoy-Criollo *et al.* (2022) in defining hyperdistal subarticular tubercles. The webbing formulation is that of Savage & Heyer (1967), whereby webbing is quantified by the number and proportion of free phalanges (see also Myers & Duellman 1982; Savage & Heyer 1997). Comparative data were taken from Grant *et al.* (2025).

We took the following measurements to the nearest 0.1 mm (to avoid pseudoprecision) using dial calipers: snout–vent length (SVL), forearm length from proximal edge of palmar tubercle to outer edged of flexed elbow (FAL), hand length from proximal edge of palmar tubercle to tip of finger IV (H); tibia length from outer edge of flexed knee to heel (TL), head width between angle of jaws (HW), head length diagonally from corner of mouth to tip of snout (HL), eye length from posterior to anterior corner (EL), eye–naris distance from anterior corner of eye to center of naris (EN), distance between centers of nares (N), snout length from anterior corner of eye to tip of snout (SL), minimum interorbital distance (IOD), diameter of tympanum (TYM). We also attempted to measure foot length from the proximal edge of the outer metatarsal tubercle to the tip of toe IV (FL), but the specimen was too fragile to straighten the digits.

We attempted to obtain historical DNA from the single known specimen. We sampled a small piece of muscle from the right shank in the USNM Herpetology Division following the guidelines of Nakamura *et al.* (2024). We then carried out all subsequent procedures up to and including PCR amplification in the dedicated cleanroom laboratory of the Department of Zoology, Institute of Biosciences, University of São Paulo following recommended procedures to avoid cross-contamination (e.g., Llamas *et al.* 2017; Fulton & Shapiro 2019; Straube *et al.* 2021). We washed tissues with 1 mL phosphate buffer saline solution to decrease potential inhibitors (e.g., formaldehyde). We extracted DNA using the proteinase K treatment (Straube *et al.* 2021), purified DNA following Dabney *et al.* (2013), and quantified the extractions using a Qubit fluorometer with high sensitivity reagents and 1 µL of DNA extract. We prepared dual-indexed single-stranded DNA libraries, with excision of uracil and abasic sites carried out by uracil-DNA glycosylase and endonuclease VIII, respectively (Gansauge *et al.* 2017). The input DNA amount was ≤ 13 ng. Finally, we performed high-throughput DNA sequencing in Illumina NovaSeq X Plus (I.5B, 100 cycles; single-end reads) at TUCF Genomics (Tufts University School of Medicine, Boston, MA), generating a total of 54.4 M reads for this sample.

We assessed read quality using FASTQC v. 0.12.1 (Andrews 2010), trimmed Illumina adapters and < 21 bp reads using Cutadapt v. 1.16 (Martin 2011), removed PCR duplicates using Tally (Davis *et al.* 2013) and contaminant reads from human and bacterial references using FastqScreen v. 0.15.3 (Wingett & Andrews 2018), and summarized results from data preprocessing using FastQC (Andrews 2010). Finally, using the MIRA v. 4.0.2 assembler in MITObim v. 1.8 script (Hahn *et al.* 2013; parameters: mismatch = 3, k-bait = 15) and initial reference seeds from the whole mitogenome (GenBank accession number MT627202) and separate mitochondrial loci (PV199515, PV223915, PV224085) from *D. bokermanni*, as well as several more distantly related *Allobates* seeds (*A. amissibilis*, MT627204; *A. carajas*, MK060072; *A. femoralis*, KJ130660; *A. kingsburyi*, HQ290963; *A. talamancae*, HQ290974; *A. zaparo*, HQ291003), we performed baiting and iterative read mapping of the remaining 269799 filtered reads (0.5% of the total sequenced reads), none of which successfully mapped to the reference seeds. As such, we were unable to detect endogenous DNA in the sample.

Species Account

***Dryadobates erythropus* sp. nov.**

ZooBank LSID: urn:lsid:zoobank.org:pub:5B23A73E-822C-4F4F-B501-4316AD53F244
(Fig. 1; Table 1)

Holotype. USNM 148487, adult male collected at Tarumã, Curitiba, Paraná, Brazil, 9 January 1963, by Doris H. Blake and Doris M. Cochran.

Etymology. Like the specific epithets *alagoanus*, *capixaba*, and *carioca*, *erythropus* refers to inhabitants of the region of the type locality. Specifically, it is derived from the Greek *erythros* (red) and *pous* (foot), Latinized as *pus*, from the Portuguese term *pé-vermelho* (red-foot), a colloquial nickname for people in rural areas of Paraná, originating from the farmers who often worked barefoot on the characteristic red soil of the northern part of the state.

Generic placement. We place the new species in *Dryadobates* on the basis of the morphology of its digital discs (finger discs II, IV, and V unexpanded), absence of a metatarsal fold, presence of a complete pale oblique lateral stripe, absence of a pale dorsolateral stripe, presence of basal toe webbing between toes III–IV and absence between all other toes, presence of pale paracloacal marks, and small size (Grant *et al.* 2025).

Definition. A small species of *Dryadobates* (adult male SVL 13.5 mm, females unknown); testes of adult males unknown; throat of adult males immaculate; distal subarticular tubercle absent on finger V; inner metatarsal tubercle approximately same size as outer metatarsal tubercle, well separated from subarticular tubercle of toe I; toes II–III free of webbing; pale oblique lateral stripe complete, solid, discrete, well defined along entire length (i.e., not diffuse or narrow and faint anteriorly).

Diagnosis. *Dryadobates erythropus* **sp. nov.** differs from all nominal congeners in possessing a conspicuous pale oblique lateral stripe that is discrete and well defined along its entire length (anteriorly indistinct in *D. alagoanus*, *D. bokermanni*, *D. capixaba*, *D. carioca*, *D. lutzi*, and *D. olfersioides*). It further differs from all congeners except *D. olfersioides* in lacking melanophores on the throat of adult males. It differs from *D. olfersioides* in being much smaller (adult male SVL 13.5 mm in *D. erythropus* **sp. nov.**, 17.0–18.8 mm in *D. olfersioides*; Table 1). *Dryadobates erythropus* **sp. nov.** further differs from *D. capixaba*, *D. carioca*, and *D. olfersioides* in lacking basal webbing between toes II–III, *D. lutzi* in presenting the inner metatarsal tubercle approximately the same size as outer metatarsal tubercle, well separated from subarticular tubercle of toe I (inner metatarsal tubercle elongate, up to ca. 2–3× longer than outer metatarsal tubercle, almost reaching subarticular tubercle of toe I in *D. lutzi*), and *D. bokermanni* and *D. carioca* in smaller adult male SVL (14.9–17.9 mm in *D. bokermanni*; 14.6–16.1 mm in *D. carioca*; Table 1).

Measurements of holotype (mm). SVL 13.5, FAL 3.5, H 3.7, TL 6.3, HW 3.9, HL 4.0, EL 1.9, EN 1.2, N 1.9, SL 1.9, IOD 1.7, TYM 1.0.

Holotype description. Adult male 13.5 mm SVL; left vocal slit present, tissue too badly desiccated and fragile to probe for right vocal slit; testis and large intestine pigmentation undetermined. Ventral and most dorsal surfaces smooth; exposed surface of shank and posterior dorsum with low, inconspicuous granules. Postrictal and cloacal tubercles absent. Head width 29% of SVL and 98% of diagonal HL. EL 48% of HL. Snout artifactually concave (due to desiccation) in dorsal view (Fig. 1A), SL 48% of HL (presumably longer prior to desiccation). Nares slightly flared, directed posterodorsad, EN 63% of EL and 63% of SL. Loreal region weakly concave, almost vertical. Canthus rostralis well defined, sharply rounded. Tympanic ring strongly defined externally along anteroventral half of tympanum. Tympanum directed posterodorsad, TYM 53% of EL. Supratympanic bulge associated with the underlying depressor musculature present.

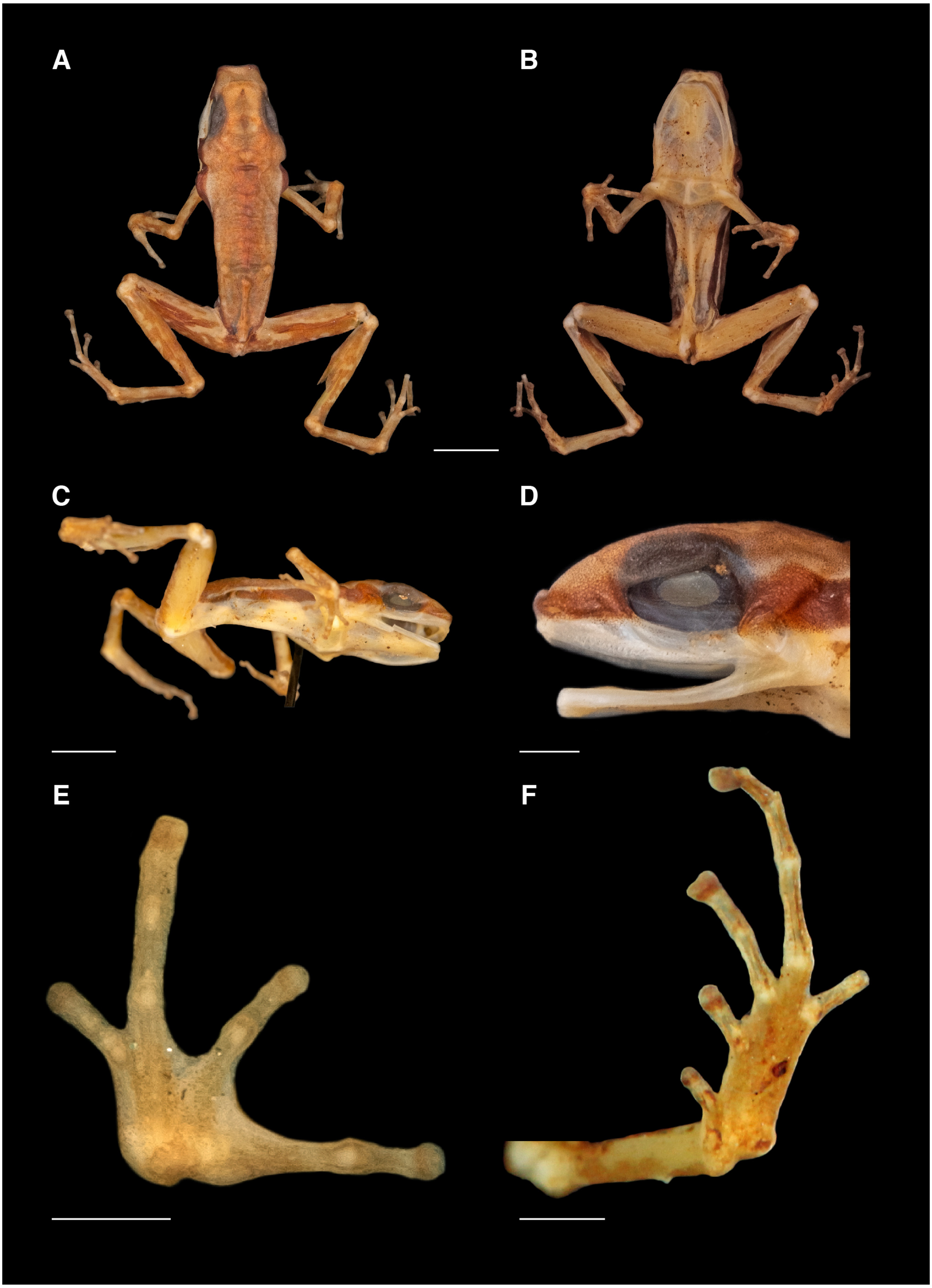

FIGURE 1. The highly desiccated adult male holotype of *Dryadobates erythropus* **sp. nov.** (USNM 148487), showing (A) dorsal, (B) ventral, and (C) lateral (right) views of body, (D) lateral (left) view of head, (E) palmar view of right hand (note phalangeal swelling of finger IV, sensu Cavalcanti *et al.* 2022), and (F) plantar view of left foot. Scale bars: A, B, C: 3 mm; D, E, F: 1 mm.

TABLE 1. Snout–vent lengths (mm) of the nominal species of *Dryadobates*. Data for all species except *D. alagoanus* were taken exclusively from type and topotype material.

Species	Sex	*N*	Minimum–maximum	$\bar{x}$ ± SE
D. alagoanus	M	13	13.2–16.3	14.6 ± 0.3
	F	5	16.0–17.0	16.5 ± 0.2
D. bokermanni	M	30	14.9–17.9	16.3 ± 0.1
	F	29	16.4–18.9	17.6 ± 0.1
D. capixaba	M	5	13.1–14.7	13.8 ± 0.3
	F	—	—	—
D. carioca	M	9	14.6–16.1	15.5 ± 0.2
	F	10	15.8–17.8	17.0 ± 0.2
***D. erythropus* sp. nov.**	M	1	13.5	
	F	—	—	—
D. lutzi	M	19	13.1–15.8	15.2 ± 0.2
	F	22	14.8–17.8	16.4 ± 0.2
D. olfersioides	M	3	17.0–18.8	17.8 ± 0.5
	F	—	—	—

Hand length 27% of SVL, 1.06 times FAL. Relative finger lengths IV > II > III > V (Fig. 1E). Finger II 1.1 times longer than finger III. Fingers too fragile to appress, finger III estimated to reach middle of distal subarticular tubercle of finger IV, finger V estimated to reach just past distal edge of basal subarticular tubercle of finger IV. Phalangeal swelling on finger IV of adult males present, dorsal and preaxial only, conspicuous, absent elsewhere on hand. Hand tubercles well defined, protuberant. Fingers II, III, and V each with a single subarticular tubercle; finger IV with two subarticular tubercles; hyperdistal subarticular tubercles absent. Thenar tubercle elliptical, palmar tubercle round. Fringes absent. Metacarpal fold absent. Discs unexpanded (state 0 of Grant *et al.* 2006), all bearing paired dorsal scutes. Carpal pad and black arm gland absent.

Tibia length 47% of SVL; FL not measured due to fragility of toe IV. Relative lengths of toes IV > III > V > II > I (Fig. 1F). Foot tubercles well defined, protuberant. Inner metatarsal tubercle approximately same size as outer metatarsal tubercle, well separated from subarticular tubercle of toe I. Toes I and II with one subarticular tubercle each, toes III and V with two, toe IV with three. Webbing absent between toes I–II, II–III, and IV–V, rudimentary webbing present between toes III–IV, giving formula III 3–4 IV. Fringes absent. Metatarsal fold absent. Tarsal keel well defined, short, strongly curved, tubercle-like, not extending from metatarsal tubercle, lying one-third of tarsal length from inner metatarsal tubercle. Discs bearing paired dorsal scutes; discs badly desiccated, but discs I and V estimated to be unexpanded (state 0 of Grant *et al.* 2006), disc II weakly expanded (state 1), discs III and IV moderately expanded (state 2).

Color in preservative (Fig. 1). Dorsum tan with dark brown stippling and blotches. Dorsolateral stripe absent. Flank uniformly dark brown, bisected by conspicuous cream oblique lateral stripe, extending from groin to posterior edge of eyelid (i.e., not extending along canthus rostralis), well defined along entire length, not becoming diffuse or narrow and faint anteriorly. Pale ventrolateral stripe not discernible (i.e., iridophores absent and not delimited ventrally by brown stippling) but inferred to have been present in life based on well-defined ventral edge of dark brown flank coloration and occurrence above arm insertion, ventral half of tympanum, and around snout (i.e., continuous with pale stripe along upper lip); surface below pale upper lip stripe with sparse melanophores. Otic region same color as flank, bordered above by pale oblique lateral stripe and below by pale ventrolateral stripe; tympanum blackish brown dorsally, cream (from ventrolateral stripe) ventrally. Dark color of flank and otic region continuing from anterior corner of eye along loreal region above pale upper lip, through nares to tip of snout. Throat, chest, and belly immaculate. Lower lip immaculate. Ventral surfaces of arm pale yellow, lacking melanophores; anterior and posterior surfaces of upper arm with longitudinal brown stripe; dorsal surfaces with pale brown stippling, even on upper arm, forming irregular pale spotting on forearm. Dorsal surfaces of outer fingers stippled pale brown, inner fingers with less stippling. Palmar surfaces pale yellow with faint brown stippling. Exposed surfaces of thigh, shank, and foot with irregular pale brown stippling. Anterior surface of thigh dark brown; posterior surface of thigh dark brown with miniscule pale spots posteroventrally; pale paracloacal mark well defined. Ventral surface of thigh and concealed surfaces of shank and foot pale yellow. Plantar surfaces pale yellow with brown stippling.

Color in life. Unknown.

Advertisement call. Unknown.

Tadpoles. Unknown.

Geographic distribution. The only known specimen of *Dryadobates erythropus* **sp. nov.** was collected in 1963 in Tarumã, Curitiba, PR (Fig. 2).

Conservation status. Currently, the type locality of Tarumã is a highly developed, mixed residential and commercial neighborhood in the city of Curitiba that lacks appropriate habitat for *Dryadobates* spp. Although many other localities in Paraná have been sampled over the past six decades (summarized by Santos-Pereira *et al.* 2018), there are no additional records of this or any other dendrobatoid frog in the state. As such, we presume this species to be extinct (IUCN 2012).

Remarks. The single known specimen of *Dryadobates erythropus* **sp. nov.** is badly desiccated and extremely brittle. Soil and grains of sand are adhered to the integument. The mandible is broken on the right side, left finger IV is broken (but still attached), the right knee is barely attached to the shank, right toe IV is broken (but still attached), the distal portion of left toe V is missing, and the left leg is partially detached from the pelvic girdle. Given the fragility of the specimen, we did not attempt to examine testis or large intestine pigmentation or straighten left toe IV for measurement.

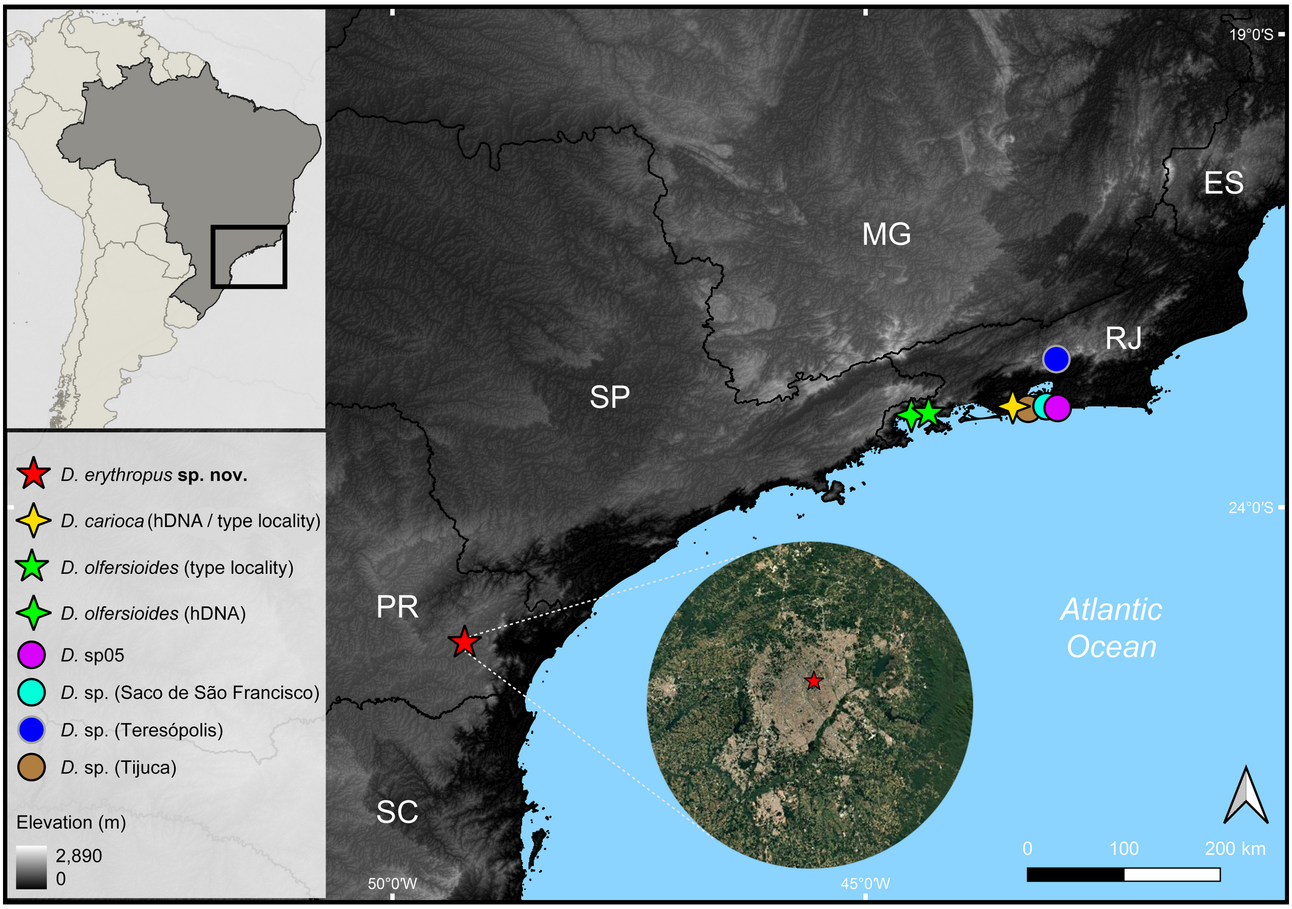

FIGURE 2. Distribution of the southernmost species of *Dryadobates* in the Atlantic Forest of southeastern Brazil, including *D. erythropus* **sp. nov.** and named and unnamed species. The expanded satellite image shows the type locality of Tarumã (star) within the urban area of present-day Curitiba.

Discussion

Our failure to detect endogenous DNA in the *Dryadobates erythropus* **sp. nov.** sample was unfortunate but not surprising. Although details are not provided in Cochran's notes, the overall condition and the soil and grains of sand adhered to the integument suggest the specimen might have died in the collecting bag some time prior to fixation, which would have resulted in extensive DNA degradation even before the damage accrued over the following six decades. Although it is possible that hybridization capture enrichment using specific mitochondrial baits could yield endogenous DNA, we are skeptical due to the complete absence of target DNA despite the large number of shotgun reads (54.4 M).

The only known locality of *Dryadobates erythropus* **sp. nov.** in Tarumã, Curitiba, PR, is nearly 550 km southwest of the nearest congener, *D. olfersioides,* in Angra dos Reis, RJ (Fig. 2). Although many anuran species and clades are continuously distributed in suitable habitat between these localities (Santos-Pereira *et al.* 2018), no specimens of *Dryadobates* have been collected. A similar, large distributional gap is shared with the *Boana claresignata* group. The two nominal species of the group, *B. claresignata* (Lutz & Lutz, 1939) and *B. clepsydra* (Lutz, 1925b), occur in the Serra da Bocaina (division between São Paulo and Rio de Janeiro states) and Serra dos Órgãos (Rio de Janeiro), while an unidentified species of the group occurs ca. 45 km east of Curitiba and 470 km south of Serra da Bocaina in Morretes, PA (Lutz 1973; Lyra *et al.* 2020).

In addition to sharing a similarly disjunct distribution, these clades share the extreme scarcity and apparent extinction of their southernmost species. As noted above, *Dryadobates erythropus* **sp. nov.** is known exclusively from the holotype collected in 1963, and the unidentified species of the *Boana claresignata* group is known from a single lot of tadpoles (MNRJ 68427) collected in 1950 (Lutz 1973; Lyra *et al.* 2020). The fact that these distantly related taxa share a similar geographic gap and apparently extinct southernmost species suggests both could have been affected by the same event(s) or process(es).

Habitat loss is the most immediately obvious potential explanation for the extinction of *Dryadobates erythropus* **sp. nov.** at the type locality. In 1963, the type locality of Tarumã (named in reference to the native tree *Vitex montevidensis*) was rural (Cochran described being taken from Curitiba to Tarumã), characterized by trees, streams, and extensive fields and wetlands. Cochran's notes describe a small, tree-lined river reaching the road and a path along which she and Blake walked, striking branches and plants to collect insects. The precise locality of *D. erythropus* **sp. nov.** was described as "a big field full of anthills and cow-chewed grass clumps." Currently, Tarumã is a highly developed, mixed residential and commercial neighborhood in the city of Curitiba that lacks appropriate habitat for *Dryadobates* spp.

Habitat loss and fragmentation is likely related to disappearances at other localities as well. The region between the southernmost records and their nearest congeners bridges the limit between the Araucária and Serra do Mar biogeographical sub-regions of the Atlantic Forest biome (Silva & Casteletti 2003), both of which have been reduced to only 12.6% and 32.2% of their original extent, respectively, with 83.4% of the surveyed Atlantic Forest fragments being smaller than 50 ha (Ribeiro *et al.* 2009). In addition to the direct effects of habitat loss, such extensive habitat fragmentation can prevent species dispersion and/or migration, resulting in decreased breeding and genetic diversity, as well as decreased resilience to extreme climatic events and increased vulnerability to infectious diseases (McCallum & Dobson 2002; Becker *et al.* 2007, 2023; Keyghobadi 2007; Oliver *et al.* 2013, 2015; Belasen *et al.* 2022; Covarrubias *et al.* 2022).

Indeed, chytridiomycosis, a disease caused by the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*), is known to be the proximate cause of declines around the world (Scheele *et al.* 2019) and could be related to the disappearance of these and additional species in the region. Carvalho *et al.* (2017) sampled more than 30,000 tadpoles collected throughout Brazil between 1930 and 2015 and found a strong correlation between the incidence of *Bd* and historical amphibian populations decline, especially in southeastern Brazil between 1979 and 1987. Their database (http://dx.doi.org/10.5061/dryad.4t53n) includes 1701 tadpoles of 12 families collected between Rio de Janeiro (ca. 22°S, 41°30′W) and eastern Paraná (ca. 26°S, 50°W), among which 515 (30.3%) were *Bd*-positive. The high incidence of *Bd* and the amphibian population declines between 1979 and 1987 suggest *Bd* could have contributed to the disappearance of *D. erythropus* **sp. nov.** and the unidentified species of the *Boana claresignata* group.

Finally, Twomey *et al*. (2025) found that global amphibian declines over the last 40 years are strongly associated with exposure to extreme weather events, and Ferrante *et al.* (2025) identified local climate change and extreme weather events as the primary drivers of amphibian declines in Brazil (although they did not include rate or extent of habitat loss in their analysis). Similarly, Heyer *et al.* (1988) considered a heavy frost in 1979 to be the most likely cause of the drastic changes they observed in the anuran community at Estação Ecológica de Boracéia (Salesópolis, SP), and a similar event occurred in Paraná in 1975, when the lowest temperatures of the 20th century were recorded for Curitiba, resulting in massive impacts on agriculture (e.g., Une 1982). Although the thermal tolerance of most Atlantic Forest amphibians is unknown (but see Carilo Filho *et al.* 2021), such severe conditions could have significant effects. Moreover, factors such as habitat loss and fragmentation, chytridiomycosis, climate change, and extreme weather events are expected to act synergistically to promote biodiversity loss (e.g., Laurance & Useche 2009; Ferrante *et al.*, 2025; Twomey *et al*. 2025).

In addition to establishing the presence of *Dryadobates* significantly farther south in the Atlantic Forest than was previously known, *D. erythropus* **sp. nov.** is the fourth species of this clade to have become extinct in the last 60 years—all over a 15–20-year period from the mid-1960s to the mid-1980s. The only known specimen of *D. erythropus* **sp. nov.** was collected in 1963. Similarly, Izecksohn & Carvalho-e-Silva (2001: 34) reported that *D. carioca* had not been observed since it was described in 1967, *D. capixaba* has not been observed or collected since 1975, and the last confirmed record of *D. olfersioides* dates to 1981 (Grant *et al.* 2025). Additionally, specimens of the unidentified populations of *Dryadobates* at Tijuca National Park in the city of Rio de Janeiro, RJ, and Santa Teresa, ES were last collected in 1979 and 1988, respectively (Toledo *et al.* 2023). In summary, *Dryadobates* now comprises seven formally described species, four of which (57%) are presumed extinct, with another six species awaiting description (Grant *et al.* 2025) and several more extinct populations awaiting identification. Given that the drivers of these recent extinctions are unclear, it is imperative that information be collected on the natural history, distribution, and local abundance of extant populations of *Dryadobates* to identify and mitigate threats to their survival.

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Appendix: Material examined

Dryadobates alagoanus (*N* = 45). BRAZIL: **Alagoas**: Mangabeiras, MZUSP 73707 (holotype), MZUSP 73823–73830, 78191–78197 (paratypes); Mata da Salvia, Rio Largo, MHN-UFAL 355; Mata do Catolé, Maceió, MHN-UFAL 2684, 4193, MZUSP 142680; Fazenda Santa Isabel, Água Quente, Campo Alegre, MHN-UFAL 5754; Fazenda Serra D'Água II, Matriz de Camaragibe, MHN-UFAL 7323–7324; Parque Municipal ed Maceió, Bairo ed Bebedouro, Maceió, MHN-UFAL 11779, MHN-UFAL 15794; Fazendo Brejo, Teotônio Vilela, Maceió, MHN-UFAL 13270; **Bahia**: Pindobaçu, 10°39′1.69″S, 40°22′25.82″W, MZUSP 24675–24677; **Pernambuco:** Timbaúba,

7°30′18″S, 35°19′04″W, CHUFPE 1102–1103; Sirinhaém, 8°35′27″S, 35°06′57″W, CHUFPE 1817–1823, PMSN 1214; **Sergipe**: Serra de Itabaiana, MZUSP 114587, 157758, 157765, 157769–157770, LABEV 1089 (MTR 37387).

Dryadobates bokermanni (*N* = 84). BRAZIL: **Bahia**: Universidade Estadual de Santa Cruz Campus Professor Soane Nazaré de Andrade, Ilhéus, 14°47′45″S, 39°10′19″W, 20 m, CFBH 11976, 32064, 32104, MZUESC 20087, MZUSP 16084749; Ilhéus, CFBH 4470, MZUESC 6836–6837, 7286, 7342, 7433, 18764; Aritaguá, Ilhéus, 14°42′26.657″S, 39°8′4.257″W, UFBA 10695; Centro de Pesquisas do Cacau (CEPEC), Ilhéus, MZUSP 9787293873; Comissão Executiva do Plano da Lavoura Cacaueira (CEPLAC), Ilhéus, MNRJ 23767–23770, MZUSP 81133, 93872–93877, 93879–93913; Ipiaú, 14°08′17″S, 39°44′03″W, 135 m, MBML 6579; Itacaré, 14°21′33″S, 39°00′24W″, MTR 16435; Itacaré, MZUESC 9158, 9196; Trilha da Mata, Fazenda Bonfim, Uruçuca, CFBH 32418; Fazenda Bonfim, Uruçuca, 14°36′38″S, 39°21′25″W, CFBH 34040; Parque Estadual Serra do Conduru, Uruçuca, MZUESC 17918, 18405; Parque Estadual Serra do Conduru, Uruçuca, 14°28′48.58″S, 39°6′21.17″W, MZUSP 160547–160548, 160580–160585; Fazenda Provisão, Uruçuca, MZUESC 14458, CFBH 34062; Uruçuca, MZUSP 26916, 132187.

Dryadobates capixaba (*N* = 5). BRAZIL: **Espírito Santo**: Lagoa do Macuco, Refúgio Sooretama, Linhares, MZUSP 76628 (holotype), 76629 (paratype), 73752 (paratype); Linhares, MNRJ 40420–40421, MZUSP 93871.

Dryadobates carioca (*N* = 21). BRAZIL: **Rio de Janeiro**: Represa Rio Grande, [Jacarepaguá, Parque Estadual da Pedra Branca,] Rio de Janeiro [previously Guanabara], MZUSP 76653 (holotype), 73714 (paratype), 73753 (paratype), 93651–93665, 93671, 100776, 100778.

Dryadobates lutzi (*N* = 56). BRAZIL: **Bahia**: Reserva Biológica do Una, Municipio de Una, 15°10′47.70″S, 39°5′56.39″W, MZUSP 132188–132206; Reserva Particular do Patrimônio Natural (RPPN) Estação Veracel/ Veracruz, Porto Seguro, CFBH 6411, MNRJ 28953–28955, 37941, MZUSP 126361–126366; Reserva Particular de Patrimônio Natural Estação Veracel/Veracruz, Porto Seguro, 16°23′11.47″S, 39°10′10.31″W, MZUSP 160549; Mascote, 15°34′25.43″S, 39°18′9.17″W, 129 m, MZUESC 20074; Estação Experimental Gregório Bondar, Comissão Executiva do Plano Lavoura Cacaueira (CEPLAC), Barrolândia, Santa Cruz Cabrália, 16°6′12.85″S, 39°12′25.24″W, MZUSP 160544–160546, MTR 34097; Fazenda Unacau, São José da Vitória, 15°9′0.00″S, 39°18′00.00″W, MZUSP 63495, MRT 6031; Serra do Teimoso, Jussari, 15°09′13.09″S, 39°31′41.21W″, MRT 5824; Serra Bonita, Camacan, 15°22′49.08″S, 39°33′0.36″W, MZUSP 149668–149669; Fazenda São Caetano, Monte Alegre, Guaratinga, 16°24′3.24″S, 39°59′2.40″W, MZUSP 160592–160602; Rio de Una, 15°16′8.47″S, 39°7′37.91″W, MZUSP 63719–63721; Fazenda Boa Sorte, Itapebi, 15°53′25.30″S 39°31′45.55″W, MZUSP 160543.

Dryadobates olfersioides (*N* = 8). BRAZIL: **Rio de Janeiro**: Angra dos Reis, AL-MN [MNRJ] 583, 783 (paralectotypes), USNM 96539 (paralectotype), 96540 (lectotype), MNRJ 5091, 5094; Mambucaba, Angra dos Reis, MZUSP 75655; Praia Grande, Mangaratiba MNRJ 33047.

***Dryadobates* sp01** (*N* = 9). BRAZIL: **Espírito Santo**: Balneário Costa Bela, Serra, 25 m, MBML 73847385, 93599365.

***Dryadobates* sp03** (*N* = 1). BRAZIL: **Bahia**: Near Estação Ecológica Estadual Wenceslau Guimarães, Fazenda Serra Verde, Nova Esperança, Wenceslau Guimarães, 13°33′42″S 39°41′04″W, 544 m, MZUSP 160591.

***Dryadobates* sp10** (*N* = 7). BRAZIL: **Bahia**: Near Estação Ecológica Estadual Wenceslau Guimarães, Nova Esperança, Fazenda Serra Verde, Wenceslau Guimarães, 13°36′12″S, 39°42′04″W, 297 m, MZUSP 160604–160605; Reserva Ecológica da Michelin, Igrapiúna, 13°49′6.69″S, 39°12′11.13″W, ID 901, 930; Península de Maraú, Maraú, 13°56′48.97″S, 38°56′45.57″W, MZUESC 20264; Península de Maraú, Maraú, 13°57′37.80″S, 38°56′47.20″W, MZUESC 20265; Ituberá, MZUESC 9268.

***Dryadobates* sp11** (*N* = 2). BRAZIL: **Espírito Santo**: Anchieta, 20°43′2.13″S, 40°44′15.49″W, HF 320, 324.

***Dryadobates* sp12** (*N* = 2). BRAZIL: **Bahia**: Mulungu do Morro, 11°59′6.69″S, 41°31′40.29″W, 1015 m, MZUSP 137995–137996.

***Dryadobates* sp.** (*N* = 1). BRAZIL: **Espírito Santo**: Baixo Guandu, MZUSP 35674.

***Dryadobates* sp.** (*N* = 2). BRAZIL: **Espírito Santo**: Santa Teresa, MZUSP 53559–53560.

***Dryadobates* sp.** (heterospecific paralectotypes of *D. olfersioides*; *N* = 8). BRAZIL: **Rio de Janeiro**: Saco [= Sacco] São Francisco, Niterói, 22°55′54″S 43°05′16″W, AL-MN 545, 991, MNRJ 92543–92547, USNM 96412.

***Dryadobates* sp.** (*N* = 15). BRAZIL: **Rio de Janeiro**: Parque Nacional da Tijuca, Rio de Janeiro, AMNH 72445–72447, MZUSP 94201, 94211, 94227, 94242, 94285, 94292, 94304, UMMZ 127922 (3 specimens), UMMZ 217618 (cleared & stained), KU 93161 (cleared & stained).