



Article

A phylogeny for *Aromobates* (Anura: Dendrobatidae) with description of three new species from the Andes of Venezuela, taxonomic comments on *Aromobates saltuensis*, *A. inflexus*, and notes on the conservation status of the genus

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Abstract

Aromobates is a genus endemic to the northern Andes of Colombia and Venezuela. Limited information is available about this genus diversity, current distribution, and taxonomic characterization. Here, we describe three new species of *Aromobates* from the Andes of Venezuela. We provide the most comprehensive phylogeny of *Aromobates* to date and taxonomic notes on *Aromobates saltuensis* Rivero and *A. inflexus* Rivero. We also discuss the limitations of the current molecular and morphological diagnosis of *Aromobates*. Finally, we comment on the conservation status and research priorities for this genus.

Key words: Dendrobatidae, *Aromobates*, *Aromobates saltuensis*, *Aromobates inflexus*, phylogeny, taxonomy, conservation

Una filogenia para *Aromobates* (Anura: Dendrobatidae) con descripción de tres nuevas especies de los Andes de Venezuela, comentarios taxonómicos sobre *Aromobates saltuensis* y *A. orostoma*, y notas sobre la conservación del género

Aromobates es un género endémico de los Andes del Norte en Venezuela y Colombia, pero existe poca información sobre su diversidad genérica, distribución, y su caracterización taxonómica. Describimos tres nuevas especies de sapitos de niebla del género *Aromobates* de los Andes de Venezuela. Ofrecemos la filogenia más comprehensiva de *Aromobates* hasta la fecha y notas taxonómicas sobre *A. saltuensis* Rivero y *A. inflexus* Rivero. Discutimos las limitaciones de las diagnósis morfológicas y moleculares de *Aromobates*. Finalmente comentamos el estado de conservación y las prioridades de investigación de este género.

Palabras clave: Dendrobatidae, *Aromobates*, *Aromobates saltuensis*, *Aromobates inflexus*, filogenia, taxonomía, conservación

Introduction

The Northern Andes harbor an astonishing diversity of endemic dendrobatid frogs including those in the genera *Aromobates*, *Allobates*, and *Mannophryne*. Among them, *Aromobates* is the most enigmatic with a complex taxonomic history. When it was coined, *Aromobates* included a single species (*A. nocturnus*) recognized as the putative sister taxon of all living dendrobatids (Myers *et al.* 1991). *Aromobates nocturnus* presented several autapomorphies such as a unique defensive mercaptanlike odor and the *adductor mandibulae externus superfi-*

cialis muscle, but it also included “basal” traits such as nocturnal behavior and aquatic habits (Myers *et al.* 1991). Subsequently, La Marca (“1991” 1994) described *Nephelobates* for most of the former phenetic *Colostethus alboguttatus* group (*sensu* Rivero 1988) based on presence of fang-like teeth, and a dermal covering of the cloaca. Moreover, La Marca (“1991” 1994) considered the fang-like teeth as “derived” among dendrobatids and suggested that *Nephelobates* evolved this trait independently within Dendrobatidae. Mijares-Urrútia & La Marca (1997) expanded the definition of *Nephelobates* by including tadpole characteristics (e.g. oral disc and narial morphology), which supposedly distinguished *Nephelobates* from *Aromobates* (*A. nocturnus* only in that moment). However, Grant *et al.* (1997) on addressing the taxonomic status of *Colostethus* questioned the validity of *Nephelobates*. They pointed out that some of the synapomorphies of *Nephelobates* such as fang-like teeth were indeed present in *Aromobates*.

Only after molecular phylogenies became available, Grant *et al.* (2006) synonymized *Nephelobates* with *Aromobates* showing that *A. nocturnus* was nested within *Nephelobates*. This taxonomic action raised the total number of species of *Aromobates* from 1 to 12 by including *Nephelobates* species from most of Rivero’s *Colostethus alboguttatus* group (Rivero 1988). It was noted that most *Aromobates* have a conservative morphology, making species identification difficult when only based on external characteristics. To our knowledge, there are no morphological synapomorphies for *Aromobates* (see discussion) and this genus is arguably supported by a list of molecular synapomorphies (Grant *et al.* 2006). To this date, the search for morphological, behavioral, physiological, and genomic synapomorphies of *Aromobates* continues.

The known diversity of *Aromobates* includes 14 described species, 12 endemic to the Venezuelan Cordillera de Mérida (Barrio-Amorós 2009a; Barrio-Amorós *et al.* 2011), one (*A. saltuensis*) shared between the Cordillera de Mérida and the northernmost part of the Cordillera Oriental of Colombia, and one recently described from the Sierra de Perijá (Rojas-Runjaic *et al.* 2011). The first *Aromobates* species described was *Phyllobates alboguttatus* by Boulenger (1903) from a series of individuals collected by Mr. Briceño in Mérida (Venezuela). Seventy years later, a second species was described from Chorotal (~30 km North of Mérida) as *Colostethus meridensis* (Dole & Durant 1973). During the 1970’s, Juan A. Rivero described six species of *Aromobates* as a result of his extended work with the Instituto Venezolano de Investigaciones Científicas. Rivero’s descriptions of *Aromobates* taxa (Rivero 1976) included *Colostethus haydeae* from Páramo Zumbador in Estado Táchira, *C. leopardalis* from Páramo Mucubají in Estado Mérida, and *C. orostoma* from Boca del Monte, Estado Táchira. Rivero (1978) included the descriptions of *C. inflexus* from “El Almogral” on the road to Pregonero, *C. mayorgai* from Chorotal, Estado Mérida, and *C. saltuensis* from western Estado Táchira. However, Rivero (1984 “1982”) relegated *C. inflexus* as a synonym of *C. alboguttatus*. In 1980’s, several authors described three more species of *Aromobates* with apparently restricted distributions (i.e. known only for their type localities). These descriptions included *Colostethus durantii* from La Culata, *C. serranus* from the road to El Morro, Estado Mérida (Péfaur 1985), and *C. molinarii* from Bailadores and Las Playitas, Estado Mérida (La Marca 1985). In the 1990’s, Myers *et al.* (1991) described *Aromobates nocturnus* with an unusual high detail in terms of its morphology, tadpoles, and toxicological characterization; but they fell short in describing its vocalization. Péfaur (1993) described *Colostethus capurinensis* from Páramo El Molino, Estado Mérida. Tadpoles of several species were also described (Mijares-Urrútia & La Marca 1997). During 2000’s, a series of molecular phylogenies provided insights of large undescribed diversity within *Aromobates* (e.g., La Marca *et al.* 2002 and Grant *et al.* 2006). In 2010’s, two more species were described including *A. tokuko* from Sierra de Perijá in Estado Zulia (Rojas-Runjaic *et al.* 2011) and *A. ornatissimus* (Barrio-Amorós *et al.* 2011) from the Cordillera de Mérida in Estado Trujillo.

In an effort to estimate the diversity of *Aromobates*, we explored several parts of the Venezuelan Andes and discovered five new species, three of which are described herein. Furthermore, we provide an updated phylogeny of *Aromobates*, including notes on morphology, and natural history of two populations of *Aromobates saltuensis*. We also provide some comments on *A. inflexus* and provide some suggestion for further research on this species. Finally, we provide some insights about the conservation status of Venezuelan *Aromobates* and a discussion of the current taxonomic assessment of this genus.

Material and methods

The species diagnosis and description follow Barrio-Amorós & Santos (2009). For comparison, we examined specimens of *Aromobates alboguttatus*, *A. duranti*, *A. haydeae*, *A. leopardalis*, *A. mayorgai*, *A. meridensis*, *A. molinari*, *A. nocturnus*, *A. orostoma* and *A. serranus* housed at several institutions (see Appendix I) including CVULA (Colección de Vertebrados, Universidad de Los Andes, Mérida, Venezuela), EBRG (Museo de la Estación Biológica Rancho Grande, El Limón, Venezuela), and MHNLS (Museo de Historia Natural La Salle, Caracas, Venezuela). The information of all Colombian specimens is from the ICN (Instituto de Ciencias Naturales, Universidad Nacional de Colombia, Bogotá, Colombia) database. These data were retrieved from the Instituto de Ciencias Naturales, Facultad de Ciencias, Universidad Nacional de Colombia (<http://www.biovirtual.unal.edu.co/ICN/>).

Our assessment of species identification was confirmed by using the diagnosis provided on the relevant literature: Dole & Durant (1973) and Barrio-Amorós *et al.* (2010a) for *A. meridensis*; Rivero (1976, 1978) for *A. haydeae*, *A. leopardalis*, *A. orostoma*, *A. mayorgai* and *A. saltuensis*; La Marca (1985) for *A. molinari*; Péfaur (1985, 1993) for *A. duranti*, *A. serranus* and *A. capurinensis*; Myers *et al.* (1991) for *nocturnus*. The webbing formula follows the definition by Myers & Duellman (1982). Sex was determined by combination of the presence of testes and vocal slits (males) or oviducts (females), and direct observation of calling males. Measurements of adult frogs were taken to the nearest 0.1 mm using a digital caliper (see Table 1). The advertisement calls were recorded at approximately 0.5–1.0 m from calling animals using a portable Sony stereo cassette recorder WM-D6C and a Sennheiser ME67 directional microphone onto TDK MA90 metal bias tape cassettes. Data on the temperature were also registered from the substrate where the frogs were calling. The recordings were digitized using Adobe Audition 3.0 (Adobe Systems, CA) and encoded as Windows PCM files and formatted at 24-bit. Call variables were analyzed using Sound Analysis Software RAVEN Pro 1.4 (Charif *et al.* 2010). The spectrograms were performed using the Hamming window type, 900-sample size, and a bandwidth filter of 63.7 Hz. The dominant frequency was determined using the spectrogram slice view. Call parameters measured include (1) pulse duration, (2) interval between pulses or time between the beginning of the first pulse and the next, (3) number of pulses per second, and (4) dominant frequency.

TABLE 1. Abbreviations and definitions for measurements taken on the specimens of *Aromobates* in this study.

Measurement	Abbreviation	Definition
Snout-vent length	SVL	straight length, measured from tip of snout to vent
Shank length	SL	from outer edge of flexed knee to flexed heel
Foot length	FL	from proximal edge of inner metatarsal tubercle to tip of toe IV
Head length	HeL	from tip of snout to posterior edge of exoccipital-prootic
Head width	HW	measured across the skull at angle of jaws
Eye to naris	EN	distance from anterior edge of eye to posterior edge of naris
Eye diameter	ED	measured horizontally across eye
Tympanum diameter	TD	measured horizontally across tympanum
Disc width of Finger III	F3D	measured across widest part of disc
Disc width of Toe IV	T4D	measured across widest part of disc
Length of Finger I	F1L	measured from inner edge of thenar tubercle to tip of disc
Length of Finger II	F2L	measured from inner edge of thenar tubercle to tip of disc

F refers to finger; T refers to toe.

Phylogenetic relationships within *Aromobates* were estimated from three mitochondrial and two nuclear genes. All PCR reactions were performed using primers and conditions for Santos & Cannatella (2011). The mitochondrial segment included 12S-tRNA^{Val}-16S rRNA with ~2,400 bp (also known as H1 segment). The two nuclear gene segments were proopiomelanocortin A (POMC) and NCX1 sodium-calcium exchanger 1 (NACA) with a combined length of ~1,728 bp. Sequence validation was performed using NCBI-BLAST to rule out contamination. Additional sequences were obtained from GenBank including other dendrobatids and hyloid frogs (for accession numbers see Fig. 1). Sequence alignment was performed using SATé (Simultaneous Alignment and Tree

Estimation; Liu *et al.*, 2009). Tree estimation and nodal support were calculated as follows (1) the molecular model for each segment was determined using jModelTest (Posada 2008) and it was found to be GTR+ Γ +I for all, and (2) phylogenies were estimated using maximum likelihood (ML), maximum parsimony (MP), and Bayesian approaches using RAxML 7.0.4 (Stamatakis 2006), Paup v. 4.0 (Swofford 2000), and MrBayes v. 3.4 (Huelsenbeck & Ronquist 2001) respectively. ML analyses used 20 different random starting numbers, and only the best score phylogeny is reported; MP analyses used a heuristic approach with indel sites treated as missing, random addition of taxa, and using the branch-swapping algorithm with tree-bisection-reconnection (TBR); and Bayesian estimation was done using default settings for all priors. The Markov Chain Monte Carlo (MCMC) setup for the Bayesian analysis included four independent runs, each one with four chains of 30 million generations with a sampling rate every 1,000 generations. The convergence of the runs was determined using Tracer v. 1.4 (Rambaut & Drummond 2007). From all analyses, nodal support were determined after 200 non-parametric bootstrap replicates for both ML and MP, and posterior probabilities for Bayesian analyses.

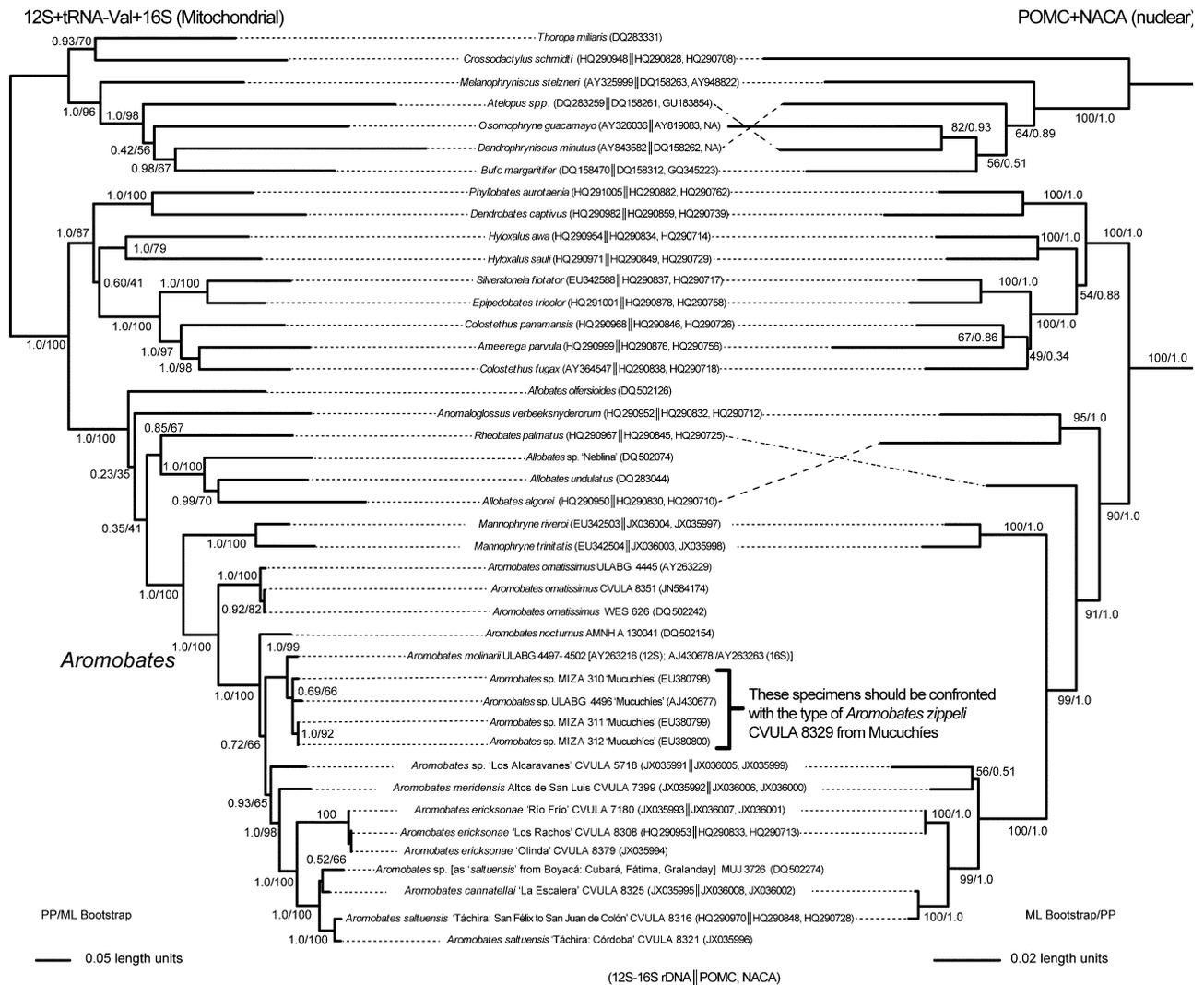


FIGURE 1. Maximum likelihood phylogenies of *Aromobates*, other dendrobatids, and closely related hyloids. The phylogenies were inferred using mitochondrial (12S rRNA -tRNA Val-16S rRNA) and nuclear (POMC and NACA) genes. Nodal support based on 200 non-parametric bootstrap replicates and Bayesian posterior probabilities. Museum voucher numbers and GenBank accession numbers are provided.

Results

Phylogenetic relationships within *Aromobates*

Aromobates is monophyletic and sister to *Mannophryne* (Figs. 1 and 2) as appeared in previous studies (Vences *et al.* 2003; Grant *et al.* 2006; Santos & Cannatella 2011; Santos 2012). Our analyses (MP, ML, and Bayesian) strongly support the monophyly of *Aromobates*. Similarly, *Mannophryne* and *Aromobates* are also strongly supported as sister clades and well-nested within Aromobatinae. Within *Aromobates*, two lineages were evidenced based on the mitochondrial tree: (1) *Aromobates ornatissimus* as the closest taxa to last common ancestor of *Aromobates* and *Mannophryne*, and (2) the rest and more inclusive *Aromobates* taxa. Overall, the phylogeny of the *Aromobates* is well-resolved at the species level (i.e., most nodal support >90% ML bootstrap) with the exception of *A. meridensis* and *A. sp.* ‘Los Alcaravanes’ which are weakly supported as sister species in the nuclear phylogeny.

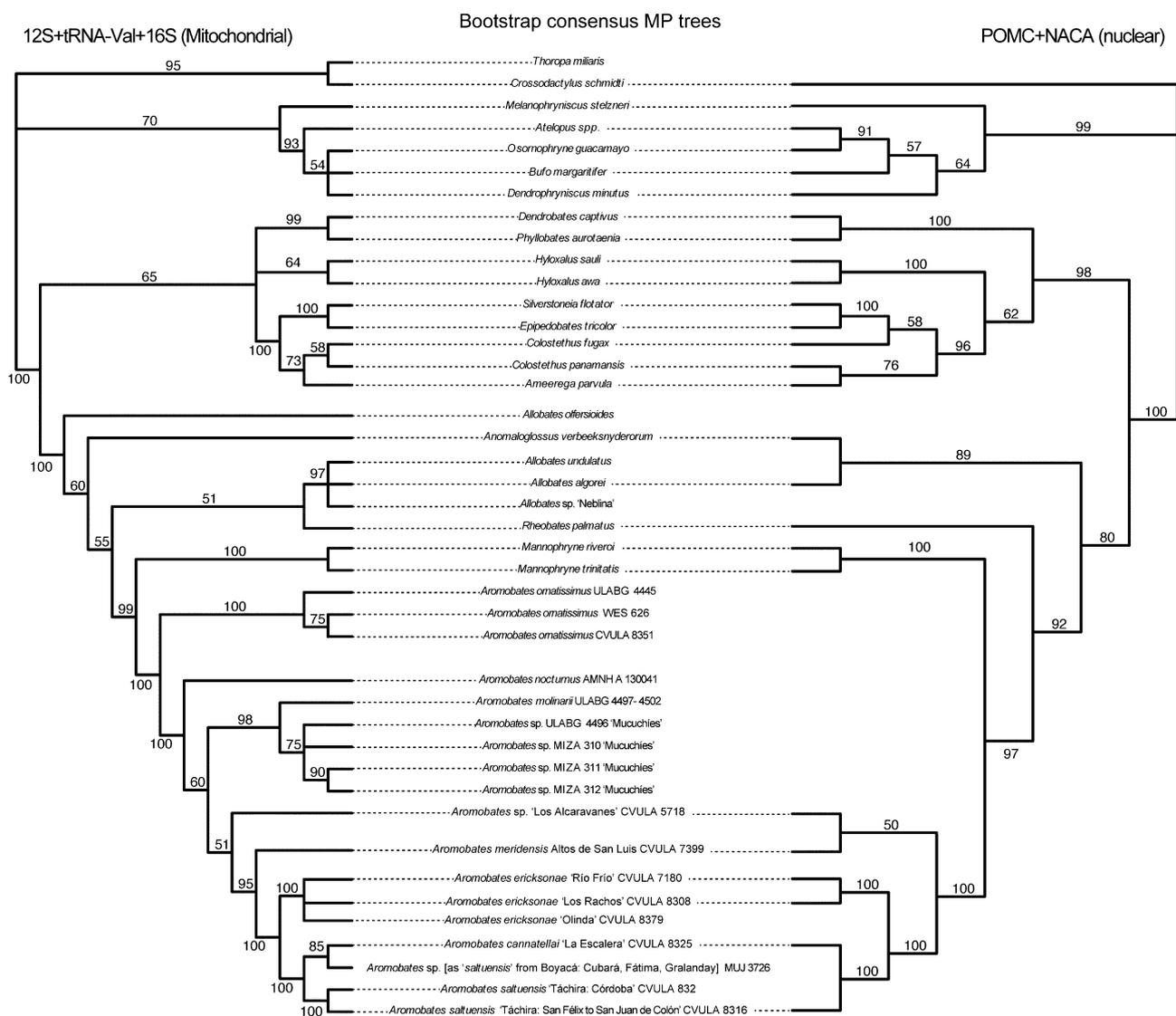


FIGURE 2. Bootstrap consensus of maximum parsimony trees of *Aromobates*, other dendrobatids, and closely related hyloids. The phylogenies were inferred using mitochondrial (12S rRNA -tRNA Val-16S rRNA) and nuclear (POMC and NACA) genes.

Our estimated phylogenies support a progression from northeast to southwest along the Cordillera de Mérida of Venezuela (Fig. 3). *Aromobates ornatissimus* and *A. nocturnus* have the north-easternmost distribution. This last species was also supported as sister taxon to the rest of *Aromobates* species which have a more central and south-

western distribution along the Cordillera de Mérida. Among the species with central distribution, we found *Aromobates molinarii*, *A. meridensis*, and two unnamed species, one from Calderas Estado Barinas (Barrio-Amorós 2010) and the other from the paramo de Mucuchíes (for discussion on its possible identity, see below). Following the south-western progression, we found *A. saltuensis* and two new species described here. All three species are distributed nearby the Táchira Depression and close to border between Venezuela and Colombia. Finally, we found a recent radiation of *Aromobates* into Northern Eastern Andes of Colombia by a possible unnamed species related to one of the new species described herein, but identified as *A. saltuensis* from Cubará, Colombia by Grant et al. (2006; for discussion see *A. saltuensis* notes).

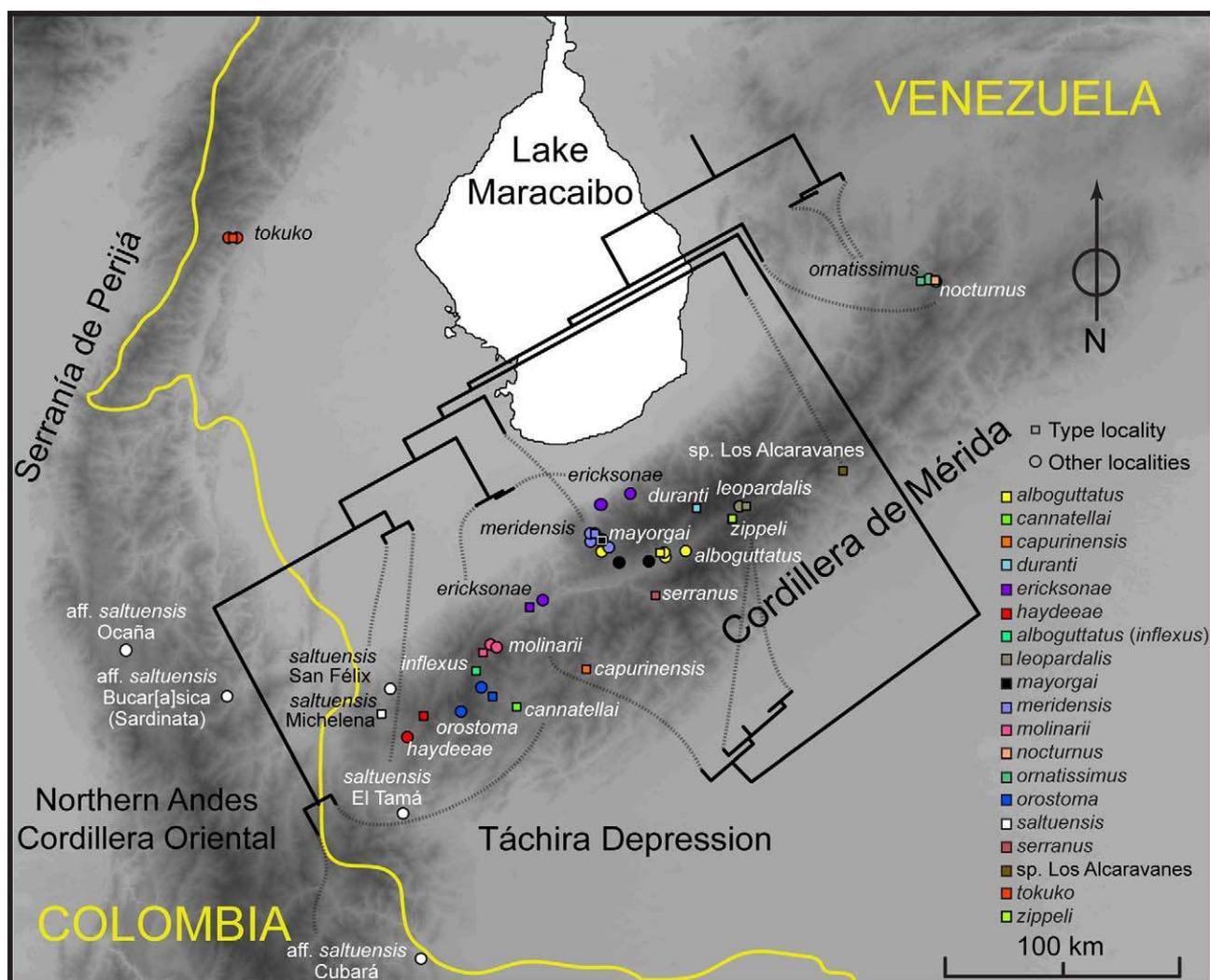


FIGURE 3. Collection localities including type localities of *Aromobates* (squares) along the Cordillera de Mérida, Serranía de Perijá, and northern Eastern Cordillera of Colombia. Several species might be sympatric and syntopic.

Species descriptions

We include these new species in the genus *Aromobates* based on their phylogenetic position (Figs 1 and 2), the list of molecular synapomorphies (Table 2), and combined morphological diagnostic characters (after Grant *et al.* 2006 and Barrio-Amorós *et al.* 2011). The putative unambiguous synapomorphies given by Grant *et al.* (2006) for the genus are molecular. However, the only molecular marker in common with their dataset is H1 (12S and 16S rDNA), but the identification of this marker's unambiguous synapomorphies *sensu* Grant *et al.* (2006) is not possible (see discussion for extended commentary).

TABLE 2. List of the apomorphic molecular characters from nuclear genes with unambiguous changes and used for diagnosis of *Aromobates*. The list includes the consistency index for each character, and the original, and transformed character states from the common ancestor of *Aromobates* and *Mannophryne* to the extant *Aromobates* species included in the analyses.

POMC (477 bp)			
Character Position	CI	Ancestor	Aromobates
159	0.167	G	A
167	0.250	C	A
183	1.000	T	C
NACA (1242 bp)			
Character Position	CI	Ancestor	Aromobates
567	0.500	T	C
579	0.333	T	C
825	1.000	T	C
873	0.333	T	C
972	0.750	A	C
1048	0.250	C	A
1180	0.500	C	T
1300	1.000	A	C
1314	1.000	T	C
1455	0.500	A	C
1482	1.000	T	C
1581	1.000	T	C
1665	0.250	T	C

***Aromobates cannatellai* sp. nov.**

Holotype. CVULA 8327, an adult male from Parque Cascada de la Escalera, at the entrance of Mesa de Pérez, Municipio Uribante, Estado Táchira, Venezuela (8.0031 N, 71.7316 W), elevation 1140 m, collected by the authors on June 6th, 2007.

Paratopotypes. Three adult females CVULA 8325-26, 8328, all with same data as the holotype; CVULA 8328 collected by the senior author on November 26th, 2005; CVULA 8325-26 collected by the authors on June 6th, 2007.

Referred specimens. CVULA 8843-44, with same data as the holotype.

Tadpoles. Unknown.

Definition: We assigned this species to *Aromobates* based on its phylogenetic position (Figs. 1 and 2), the combination of the molecular synapomorphies (Table 2), and the following characters. (1) Skin on dorsum smooth anteriorly, with small tubercles posteriorly. (2) Paired dorsal scutes present on digits. (3) Distal tubercle on FIV present. (4) FIV Length reaches half distal subarticular tubercle of FIII. (5) FI longer than FII. (6) Digital discs present. (7) Finger discs moderately expanded. (8) Finger fringes present on interior side of FII and FIII on male, absent on females. (9) Metacarpal ridge absent. (10) FIII not swollen in adult males. (11) Carpal pad absent. (12) Male excrescences on thumb absent. (13) Thenar tubercle present, small. (14) Black arm gland in adult males absent. (15) Tarsal keel to mid-tarsus, straight, the end tubercle-like. (16) Toe discs moderately expanded. (17) Toes moderately webbed. (18) Metatarsal fold absent. (19) External coloration with ill-defined to well-defined pale dorsolateral stripes; ventrolateral stripe absent; oblique lateral stripe ill-defined to well-defined, formed by a series of diffuse white spots or solid. (20) Gular-chest markings present on females with diffuse spotting, apparently absent on males. (21) Dermal collar absent. (22) Male throat coloration very dark grey; female throat coloration pale grey to dirty white. (23) Male abdomen evenly stippled, dark grey. (24) Female abdomen color pattern immaculate white to grey. (25) Iris coloration dark brown to orange, pupil ring little evident. (26) Large intestine unpig-

mented. (27) Testes white. (28) Median lingual process absent. (29) Tympanum indistinct, tympanic annulus absent. (30) Vocal sac distinct. (31) Teeth present on the maxillary arch. (32) Body size small, only one male at 22.6 mm; females ($n = 3$) 25.3–28.6 mm, mean = 27.4 ± 1.8 .

Comparisons: *Aromobates cannatellai* (characters in parentheses) differs by its smaller size from large species of the genus, such as *A. alboguttatus* (Boulenger, 1903), *A. leopardalis* (Rivero, 1978), *A. meridensis* (Dole & Durant, 1973), *A. nocturnus* (Myers, Paolillo & Daly, 1991), and *A. capurinensis* (Péfaur, 1993), all with maximum SVL > 31 mm (up to 28.6 mm). Seven other species of *Aromobates* have similar size as *Aromobates cannatellai* including *A. durantii* (Péfaur, 1985), *A. haydeae* (Rivero, 1978), *A. mayorgai* (Rivero, 1980), *A. molinarii* (La Marca, 1985), *A. orostoma* (Rivero, 1978), *A. saltuensis* (Rivero, 1980), and *A. serranus* (Péfaur, 1985). *Aromobates durantii* has its venter gray with white dots (unicolor grey or white without dots), finger and toe discs are not expanded (expanded), and it has a more extensive webbing (barely more than basal webbing). *Aromobates haydeae* has no fringes on fingers (present only on males), tarsal fold ill-defined (well-defined), dorsal color is reddish copper (dark brown), and ventral parts are orange in life (grey). *Aromobates mayorgai* has FI shorter than FII or equal (FI longer than FII), ventral parts are yellow in life (grey), and oblique lateral stripe is absent (present and formed by small whitish spots). *Aromobates molinarii* has FII and II equal in length (FI longer than FII), tarsal keel is ill-defined (straight and well-defined), and oblique lateral stripe is absent (present). *Aromobates orostoma* has a distinct tympanum (indistinct), and FI shorter than FII (FI longer than FII). *Aromobates saltuensis* has FI and FII equal in length (FI longer than FII), fringes on fingers low on FII and FIII (mainly absent), its call consists of duplets notes (single notes); TI is short, the tip never reaching the mid-subarticular tubercle of TII (TI is moderately long, and the tip reaches the mid-subarticular tubercle of TII). *Aromobates serranus* has a distinct tympanum (indistinct), FI shorter than FII (FI longer than FII), and specimens belly areas are creamy white with brown reticulation (immaculate grey to white).

Description of the holotype. The holotype is an adult male of 22.6 mm SVL (Fig. 4A): body slender, almost quadrangular in cross-section; dorsal skin smooth anteriorly, becoming shagreen posteriorly with scattered small tubercles, especially abundant on the sacral area; dorsal skin of thighs and shanks smooth; skin on throat and chest is smooth, finely granular on belly; head is longer than wide, HeL = 34.9% of SVL; HW = 34.0% of SVL; snout is short, ETS 44.3% of HeL, pointed in profile, inclined posteroventrally, pointed in dorsal and ventral view; nares are situated laterally to the tip of snout; narial openings are barely visible when viewing the head from the front, not visible when viewing dorsally; and evident when viewing from the ventral aspect; canthus rostralis is quadrangular, the loreal region is nearly flat; interorbital region is wider than the upper eyelid; snout little longer than ED; tympanum is indistinct, with only its anteroventral portion being barely visible, concealed posterodorsally by a supratympanic bulge formed by the superficial slip of *m. depressor mandibulae*; on the right side there is a piece of skin broken over the tympanum, and on the left side a smaller piece also broken on the bulge; the supratympanic bulge can be low in profile (right side) or very notable (left) due to preservation artifacts; tympanum is positioned closely behind eye and lower, close to the angle of jaws; teeth present on maxillary arch; vocal slits large, arched from the posterior one third of the tongue to a point anterior to the angles of jaws; vocal sac present and single; tongue is short, quadrangular, one-third free posteriorly.

Hand of moderate size (27.0% SVL); relative lengths of adpressed fingers are $\text{III} > \text{I} > \text{II} > \text{IV}$; discs of all fingers are moderately expanded, horizontally oval; FIII disc is 1.5 wider than distal end of adjacent phalanx; the base of palm has a large, triangular palmar tubercle; and on base of FI there is a smaller (approximately 1/3 of the palmar tubercle), oval thenar tubercle; one or two subarticular tubercles on fingers (one large each on FI and FII, two smaller each on FIII and FIV, the distal one of FIV distinct); and all tubercles are flat and round except that of the FI, which is oval; supernumerary tubercles absent. Fringes on fingers are low and indistinct on interior side of FII and FIII, absent of exterior side of the same fingers and on FI and FIV.

Hind limbs are of moderate length, SL = 47.7% of SVL; relative lengths of adpressed toes are $\text{IV} > \text{III} > \text{V} > \text{II} > \text{I}$; TI is moderately long, the tip reaching the mid-subarticular tubercle of TII; toe discs are moderately expanded, TIV is about 1.6 times wider than distal end of adjacent phalanx; feet are moderately webbed; formula is $\text{I2-2}\frac{1}{2}\text{II2-2}\frac{1}{4}\text{III3-4IV4}\frac{1}{2}\text{-3V}$; fringes present on all toes and extensive, flap-like, folding is present around the toes; one to three non-protuberant, small subarticular tubercles are present (one on TI and TII, two on TIII and TV, three on TIV, all oval to elongate); two metatarsal tubercles present, including a small rounded, subconical outer tubercle, and a little smaller elongated inner tarsal tubercle; on right foot there is a mid-tarsal tubercle (lacking on left foot), about same size of the outer, low, and round; a short but well-defined tarsal keel is present, thick, straight, transverse across tarsus, from proximal edge of inner metatarsal tubercle to mid-tarsus, where it looks like a tubercle; cloacal opening is located at the upper level of thighs, without an anal sheath.

Measurements of holotype (in mm): SVL: 22.6; SL: 10.8; FL: 10.0; HeL: 7.9; HW: 7.7; ETS: 3.5; EN: 1.9; ED: 3.0; TD: -; HD: 6.1; 1FiL: 3.4; 2FiL: 3.1.

Color: The specimen in life has a general very dark, almost black coloration (Fig. 4A). Dorsum is black with small brown spots on the frontal area; it has two dark brown dorsolateral stripes, from the tip of the snout through the canthus rostralis, above the eye, reaching the sacral area, becoming paler behind the eye to the sacrum. Flanks are black with the oblique lateral stripe that is constituted by a series of small but diffuse white spots. There is a series of small irregular dirty white spots on the upper lip area and on the insertion of the upper arm. Arms are dirty black, with small brown spots on the forearm. Upper surface of thighs and shank are dark brown with one black crossbar and diffuse black spotting. Tarsi and anterior side of thighs are grey to black. Hands and feet are black dorsally. Ventrally the specimen is light grey.

After preservation, the holotype is mainly black in coloration. The dorsolateral stripes are indistinct grey, and crossbars are more evident on hind limbs. Ventrally, the specimen's throat, chest, and belly are evenly stippled black under magnification, giving it a general ventral coloration that is dark grey. Under the specimen hind limbs areas, their coloration is pale grey. Specimen's palms and soles are pale dark grey. Undersides of discs are pale grey.



FIGURE 4. *Aromobates cannatellai* sp. nov. (A) Male holotype, CVULA 8327. Photo by CBA. (B) Unsexed specimen from type locality, not collected. Photo by Liz del Valle. (C) Female paratype, CVULA 8328. Photo by CBA. (D) Ventral view of female paratype CVULA 8328. Photo by CBA.

Variation. No other males are known to determine conspecific variability. Females are larger and apparently dichromatic. Morphological differences include the absence of fringes on fingers II and III. The only female photographed alive (CVULA 8328) is the darkest of the series (Fig. 4C). Dorsum brown with dorsolateral stripes are pale brown and not very distinctive. Specimens are without an evident pattern on their dorsal, arms, and hind limbs; flanks have the same dark coloration than dorsum, except ventrally and closer to their bellies, where they become white. Specimens' oblique lateral stripe is pale yellow, diffuse, and ill defined. Specimens are ventrally grey, patternless, with a profusion of melanophores under magnification (Fig. 4D). In preservative, specimens are dorsally dark grey with dorsolateral stripes ill defined, little paler than surrounding background; hind limbs are brown with ill-defined crossbars. In preservative, CVULA 8325-26 specimens have similar colorations. Their dorsa are brown and irregularly spotted, the dorsolateral stripes are pale brown to grey and well-defined, and their flanks are darker. Variation on oblique lateral stripe ranges from well-defined as in CVULA 8326 (similar to that in Fig. 4B) to a series of white spots as in CVULA 8325. All specimens have evident crossbars on their legs. The venter of all specimens is white with a diffuse grey spotting extending to their throat and chest. Palms and soles are dark grey as in CVULA 8325 to pale grey as on CVULA 8326. The anal sheath is indistinct in all examined specimens.



FIGURE 5. Habitat of *Aromobates cannatellai* **sp. nov.** at the type locality. Photo by CLBA.

Natural history. All individuals were active during the day. The habitat is evergreen Andean forest, where the species is strictly limited to slow-flowing streams. The type locality offers three different microhabitats. One is a large slab where the water runs, as a small canyon (Fig. 5), another is on the edges of a natural pool created by a waterfall, and a third one was at the entrance to a natural cave where a small stream flowed to the exterior. Males usually called from concealed spots such as leaf litter debris or under rocks. *Aromobates cannatellai* is sympatric, but was not seen syntopically, with *Allobates algorei* and *Mannophryne orellana*. Other herpetofauna associated at the type locality are *Gonatodes ligiae* and *Bothrops venezuelensis*, the latter of which could act as a potential predator.

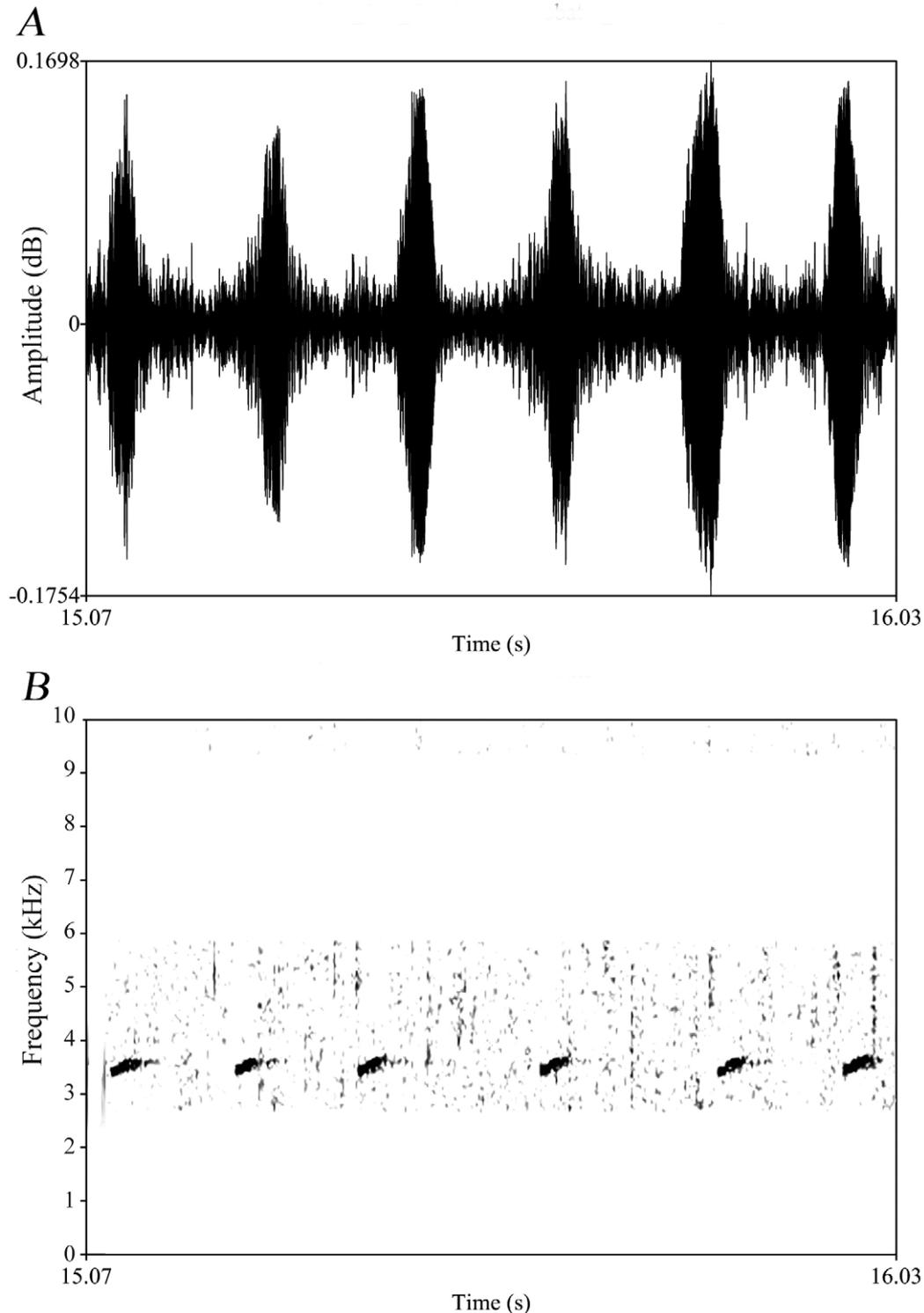


FIGURE 6. (A) Waveform and (B) spectrogram of a 1-second sequence of the call of *Aromobates cannatellai* **sp. nov.** Background noise has been strongly reduced and frequencies below 100–500 Hz were filtered from the spectrogram.

Vocalization. A single call was analyzed, recorded at 21.4 °C (substrate temperature) at dusk on June 6th, 2007 at the type locality. The vocalization is a constant trill and it lasted for several minutes. We used 25.6 sec of the recorded call for analysis, which consisted of 133 pulsed notes. The waveform of a section of the call is showed in Fig. 6A and the corresponding spectrogram in Fig. 6B. Six consecutive notes were chosen at random to determine spectral and temporal variables. The dominant frequency range is 2998.0–3236.9 Hz. Pulse rate was 5–6 pulses per second. Note and inter-note duration with their mean, SD and range in seconds were (1) note duration, 0.04 ± 0.01 (0.03–0.05) and (2) inter-note, 0.13 ± 0.01 (0.12–0.15).

Distribution. *Aromobates cannatellai* is only known from its type locality (Fig. 3).

Phylogenetic relationships. The sister taxon of *Aromobates cannatellai* is an undescribed species from Colombia identified as *A. saltuensis* by Grant *et al.* (2006) from Cubará. We have not revised the voucher specimen (MUJ 3726 from the Museo de Historia Natural, Universidad Javeriana, Bogotá, Colombia) and we are not able to rule out if *A. cannatellai* and the Colombian specimen are conspecific. We could have considered MUJ 3726 as *Aromobates* aff. *cannatellai*. However, we think that this individual affinity to *A. cannatellai* should be determined only after comparison of its voucher MUJ 3726 with the *A. cannatellai* type series. For discussion on Grant's *A. saltuensis*, see the notes on phylogenetic relationships and distribution of *A. saltuensis* in this paper.

Etymology. *Cannatellai* is a patronym for Dr. David Cannatella (University of Texas at Austin) for his extensive work on amphibian evolution, systematics, and biodiversity. Dr. Cannatella has also mentored and supported many young Latin American and US herpetologists as undergraduate students, graduate students, and postdoctoral scholars.

Aromobates ericksonae sp. nov.

Holotype. CVULA 8309, an adult male from Los Ranchos, Santa Cruz de Mora, Estado Mérida, Venezuela (8.3989 N, 71.6801 W), elevation 1193 m, collected by the authors on June 1st, 2007.

Paratopotypes. Five adult males CVULA 8299, 8308, and 8310-12; and seven adult females CVULA 8300-04, and 8313-14. All specimens with the same information as the holotype; with the exception of CVULA 8299-304 that were collected by E. Arrieta on December 2003.

Referred specimens. CVULA 8305-07, from Santa Cruz de Mora via Los Ranchos, Estado Mérida, Venezuela (8.4265 N, 71.6281 W), 937 m, collected by the authors on June 1st, 2007. CVULA 7180, from Río Frío, Municipio Córdoba, northwestern slope of the Cordillera de Mérida, Estado Mérida, Venezuela (8.8500 N, 71.2833 W), 676 m. CVULA 7344 from Cascada La Palmita, in the way from La Azulita to Santa Elena de Arenales, Estado Mérida, Venezuela (8.7333 N, 71.4333 W), 910 m; CVULA 8379-82 from Olinda, Estado Mérida, Venezuela (8.7373 N, 71.4681 W), 800 m.

Tadpoles. Unknown.

Definition: We assigned this species to *Aromobates* based on its phylogenetic position (Figs. 1 and 2), the combination of the molecular synapomorphies (Table 2), and the following characters. (1) Skin on dorsum smooth. (2) Paired dorsal scutes present on digits. (3) Distal tubercle on FIV present. (4) FIV length reaches half distal sub-articular tubercle of FIII. (5) FI longer than FII. (6) Digital discs present. (7) Finger discs barely expanded. (8) Fringes present on fingers. (9) Metacarpal ridge absent. (10) FIII not swollen in adult males. (11) Carpal pad absent. (12) Male excrescences on thumb absent. (13) Thenar tubercle present, small. (14) Black arm gland in adult males absent. (15) Tarsal keel thick, long, to mid-tarsus, straight. (16) Toe discs weakly expanded. (17) Toe webbing basal. (18) Metatarsal fold absent. (19) External coloration with pale dorsolateral stripes; ventrolateral stripe absent; oblique lateral stripe formed by a series of diffuse yellow and white spots. (20) Gular-chest markings present, from fine reticulation to diffuse spotting. (21) Dermal collar absent. (22) Male throat coloration white with profusion of melanophores to dirty white with dark brown reticulation or spotting; female throat coloration white with a profusion of melanophores to white few irregular reticulation or spotting. (23) Male abdomen color immaculate white. (24) Female abdomen color pattern immaculate white. (25) Iris coloration golden with black, fine reticulation; gold-colored pupil ring. (26) Large intestine unpigmented. (27) Enlarged white testes. (28) Median lingual process absent. (29) Tympanum indistinct, tympanic annulus absent. (30) Vocal sac distinct. (31) Teeth present on the maxillary arch. (32) Body size small, males ($n = 8$) 20.5–24.4 mm, mean = 21.9 ± 1.1 ; females ($n = 8$) 22.3–25.7 mm, mean = 24.2 ± 1.4 .

Comparisons. *Aromobates ericksonae* (characters in parenthesis) differs by its smaller size from large species of the genus, such as *A. alboguttatus*, *A. leopardalis*, *A. meridensis*, *A. nocturnus*, and *A. capurinensis*, all with maximum SVL > 31 mm (up to 25.5 mm). The eight other species of *Aromobates* (*A. cannatellai* sp. nov., *A. durantii*, *A. haydeeeae*, *A. mayorgai*, *A. molinariii*, *A. orostoma*, *A. saltuensis*, and *A. serranus*) are similar in size to *Aromobates ericksonae*. *Aromobates cannatellai* is somewhat larger, with females up to 28.6 mm (25.7) in SVL, finger discs moderately expanded (barely expanded), toe discs moderately expanded (weakly expanded), and more extensive toe webbing (basal). *Aromobates durantii* is a larger frog, up to 30.7 mm (up to 25.7 mm), with its venter gray with white dots (unicolor white without dots), and toe discs not expanded (expanded). *Aromobates haydeeeae* has no fringes on fingers (present), tarsal fold ill-defined (well defined), dorsal color reddish copper (dark to light brown), and ventral parts orange (white). *Aromobates mayorgai* has dorsal skin smooth with a few tubercles posteriorly (smooth without tubercles in preservative), FI shorter than FII or equal (FI longer than FII), ventral parts yellow in females (white), dark in breeding males with a suffusion of small white spots (only the throat is dark, belly whitish), and oblique lateral stripe absent (formed by small whitish spots). *Aromobates molinariii* is the geographically closest species but it is a larger frog with females up to 30.3 mm (up to 25.7 mm), it has FII and II equal in length (FI longer than FII), fringes absent on fingers (present), tarsal keel ill-defined (straight and well-defined), toes moderately webbed (basally webbed), and oblique lateral stripe absent (formed by diffuse whitish spots). *Aromobates orostoma* has a distinct tympanum (indistinct), FI shorter than FII (FI longer than FII), and no fringes on fingers (present). *Aromobates saltuensis* has FI and FII equal in length (FI longer than FII), dorsal skin smooth with a few tubercles posteriorly (smooth without tubercles in preservative), and supratympanic bulge absent (ill-defined but present). *Aromobates serranus* has a distinct tympanum (indistinct), dorsum brown with blotches or reticulum (usually uniform, but not reticulated), and belly parts creamy white with brown reticulation (immaculate white).

Description of the holotype. The holotype is an adult male of 21.7 mm (Fig 7A): body slender, quadrangular in cross-section; dorsal skin, including dorsal surfaces of hind limbs, and smooth in preservative; ventral skin is smooth except on belly sides, where it is granular; its head is longer than wide, HeL = 37.7% of SVL; HW = 33.6% SVL; its snout is subacuminate in profile, inclined posteroventrally, rounded in dorsal and ventral view; its nares are situated laterally to the tip of snout; its narial openings are barely visible when viewing the head from the front, they are not visible from the dorsal side, and visible when viewing from the ventral aspect; its canthus rostralis is sloping, the loreal region is flat; interorbital region is little wider than the upper eyelid; snout longer than ED; tympanum is indistinct, only barely visible on its anteroventral portion, and concealed posterodorsally by a low supratympanic bulge formed by the superficial slip of *m. depressor mandibulae*; tympanum is positioned closely behind eye and lower, close to the angle of jaws; teeth present on maxillary arch; vocal slits large, arched from mid-level of tongue to anterior to the angles of jaws; tongue is short, oval, and half-free posteriorly.

Hand has a moderate size (27.6% SVL); relative lengths of adpressed fingers are III > I > II > IV; discs of all fingers are slightly expanded, horizontally oval; FIII disc is barely wider than distal end of adjacent phalanx; the base of palm has a large, rounded palmar tubercle; and on base of FI there is a smaller (approximately 1/3 of the palmar tubercle), oval thenar tubercle; one or two subarticular tubercles on fingers (one each on FI and FII, two each on FIII and FIV, the distal one of FIV inconspicuous); and all tubercles are flat and round or oval; without supernumerary tubercles. Fringes on fingers are low and indistinct on FI and FIII of right hand (well preserved), and very notable on all fingers of left hand except FI (somewhat dehydrated). However, we think that these differences are obviously due to preservation and consider them of somehow doubtful validity for taxonomy of *Aromobates*.

Hind limbs are of moderate length, SL = 51.1% of SVL; relative lengths of adpressed toes are IV > III > V > II > I; TI is moderately long, the tip reaching the mid-subarticular tubercle of TII; toe discs are moderately expanded, TIV about 1.8 times wider than distal end of adjacent phalanx; feet are basally webbed; fringes on toes are present but low; one to three are non-protuberant and small subarticular tubercles are present (one on TI and TII, two on TIII and TV, three on TIV, proximal one almost indistinct); two metatarsal tubercles are present, including a small oval outer, and a similar in size elongated inner tarsal tubercle; on right foot there is a mid-tarsal tubercle (lacking on left foot), this tubercle is double size of the two others; tarsal keel is well-defined, thick, straight, and transverse across tarsus, located from the proximal edge of inner metatarsal tubercle to mid-tarsus; cloacal opening is located at upper level of thighs, with a short tube flap or anal sheath.

Measurements of holotype (in mm). SVL: 21.7; SL: 11.1; FL: 10.8; HeL: 8.2; HW: 7.3; ETS: 3.5; EN: 2.2; ED: 3.1; TD: -; HD: 6.0; 1FiL: 3.5; 2FiL: 3.0.

Color. The specimen in life had its dorsum patternless dark brown except for the interorbital area where it was paler irregular with diffuse markings (Fig. 7A). Two golden cream dorsolateral stripes are present from the tip of snout through the canthus rostralis and over the eyelid to the sacral area. Flanks are blackish brown immediately inferior to the dorsolateral stripes, including the loreal region; flanks below the dark brown area present a pale grey coloration with a diffuse to whitish spotting. The oblique lateral stripe is evidenced by diffuse spots ranging from yellow at the inguinal zone to dirty white for the remaining of the stripe. The length of the oblique lateral stripe begins at the inguinal zone and ends at mid flank. The upper lips are dark grey with striking white irregular large spots. Arms are greyish brown, with a pale cream area on the upper insertion of the arm, and they have a diffuse dark brown longitudinal stripe on the anterior side of the arm. Fingers are uniformly grey. Hind limbs are pale brown in their dorsal side and ornamented with darker crossbars.

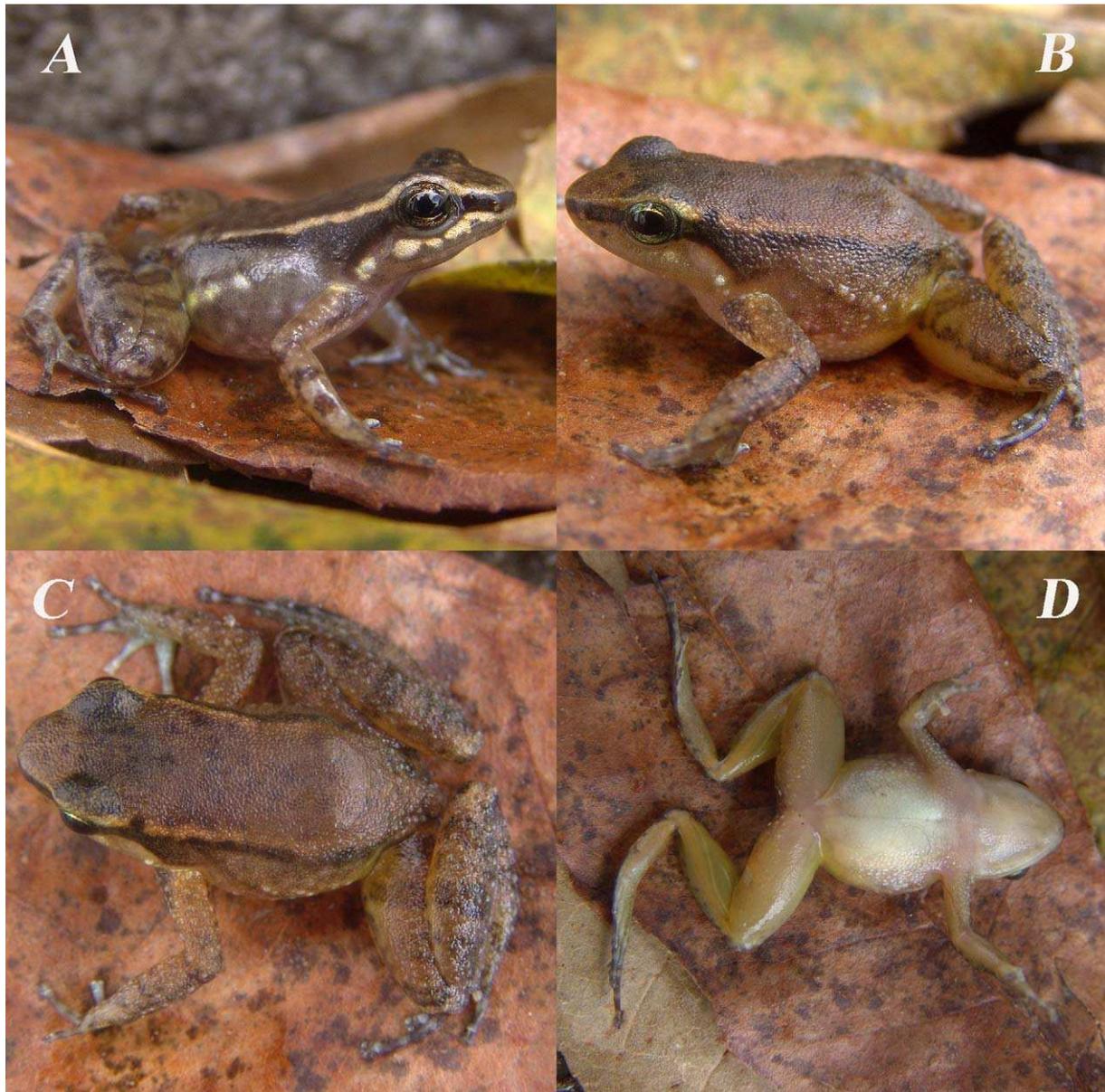


FIGURE 7. *Aromobates ericksonae* sp. nov. (A) Male holotype, CVULA 8309. (B) Female paratopotype, CVULA 8310. (C) Dorsal view of CVULA 8310, showing skin texture. (D) Ventral view of CVULA 8310. Photos by CBA.

After preservation, the holotype presents some evident coloration changes. The dorsolateral stripes became strikingly white and the crossbars are clearly visible on both arms and hind limbs. On the ventral side, the specimen presents a dark brown reticulation on throat and chest while the belly remained white. Palms and plants are pale grey in coloration.

Variation. Preserved specimens appear to have smooth skin, while living animals have shagreen to granular dorsal and lateral skin (Figure 7C). Some color variation is evidenced on the granules of the thighs and some of them are white in coloration. The dorsal pattern observed in the holotype is similar as in three other males (CVULA 8299, 8311-12) though the pale dorsolateral stripes are much more evident on the holotype. Variation was observed and some males are paler (orange brown in life) dorsally, with less marked crossbars on the limbs (CVULA 8306, 8308, 8310 and 8305). On CVULA 8299, 8308 and 8311, the dorsolateral stripes are ill-defined. Females are slightly larger in SVL than males, with 6 of 8 specimens having a dark brown dorsal background with only one specimen (CVULA 8303) having well-defined dorsolateral stripes. All specimens have a oblique lateral stripe ranging from more or less defined to a ill-defined by a series of white spots (Fig 7A). A vocal sac is evident on the male CVULA 8299. Ventrally, the coloration of this species is sexually dimorphic. Breeding males usually have dark throats but a more whitish belly with some variation. For example, the holotype and CVULA 8299, 8311, and 8312 have their chins, throats, and chests strongly to diffusely reticulated, CVULA 8306 has a diffusely spotted throat, and CVULA 8305, 8308 and 8310 have dirty white coloration (with only a profusion of melano-phores on the chin under magnification). Females also present some variation in their chin and throat coloration ranging from uniformly dirty white (CVULA 8313-14) or little spotted (CVULA 8300) to evidently irregular markings without reticulation as in CVULA 8303-04, 8301-02.

In life, the dorsal coloration can be dark brown to orange brown, and without pattern to one consisting of small dark brown spots with paravertebral distribution. The dorsolateral stripes can be white, pale yellow to orange (Fig. 7B). Ventrally, all specimens photographed are pale grey on the throat, immaculate white on the belly; the inguinal area, the thighs and shanks are pale yellow (Fig. 7D). The anal sheath is evident in the holotype and CVULA 8308, 8305, 8310, and 8299; but it is not evident in the males CVULA 8306, 8311-12. The anal sheath is also present in females except on CVULA 8301 and 8314. The anal sheath was found to be variable and most likely caused by preservation artifacts (Grant *et al.*, 2006).

Distribution and natural history. We found four populations of *Aromobates ericksonae* (Fig. 3). The topotypic population is located on a deep creek outside the small village of Los Ranchos, with males calling from the leaf litter, and females in close proximity. However, the frogs were never seen directly in the creek, which suggest that this species is riparian. All individuals were active during the day. Several small tadpoles were seen but not captured in small pools in the creek. One juvenile of *Scinax manriquei* Barrio-Amorós, Chacon & Orellana, 2004, was also seen in the same location.

Another close population of *Aromobates ericksonae* was discovered in a different habitat in the locality of Santa Cruz de Mora via Los Ranchos. This collection site is an open stream within a deciduous forest, where other species of dendrobatid are present (*Mannophryne collaris*, Barrio-Amorós *et al.* 2010b). The three individuals of *A. ericksonae* collected there were found on the forest floor or close to tree roots along the banks of a stream. Remarkably, these *A. ericksonae* were not as agile as *Mannophryne* and much easier to collect. Along the stream, several large individuals of fishing spiders (Sparassidae) were seen syntopically with both *A. ericksonae* and *M. collaris*. CBA has seen events of predation by these spiders on *Mannophryne* and they might also prey on *Aromobates*.

A third population of *Aromobates ericksonae* is present at the lower mountain forest along the road from La Azulita to Santa Elena de Arenales, where males can be heard from forests along the road from La Palmita to Olinda in rainy days. The fourth population of *A. ericksonae* is located around Río Frío and its based on a single specimen. At this locality, the individual was found in a secondary forest close to a mountain stream flowing to Río Frío and sympatric with *Mannophryne urticans* (Barrio-Amorós *et al.* 2010b).

Vocalization. A single call was analyzed from a recording collected at 22.3 °C (substrate temperature) and dusk on June 1st, 2007 at the type locality. It is a constant trill lasting for several minutes (Fig. 8A shows the waveform and 8A' the spectrogram). Seven consecutive notes were chosen at random from the spectrogram to determine spectral and temporal variables. The dominant frequency range is 3730.0 – 4314.0 Hz. Pulse rate was about one pulse per second. Note and inter-note duration with their mean, SD and range in seconds are indicated as follows (1) note duration, 0.20 ± 0.005 (0.20–0.21) and (2) inter-note, 1.08 ± 0.045 (1.05–1.16). Both, the waveform and the spectrogram are shown in Fig. 8B and B'.

Phylogenetic relationships. The sister clade of *Aromobates ericksonae* is the *saltuensis* group composed by *A. cannatellai* and *A. saltuensis*. The distribution of *A. ericksonae* suggests that the last common ancestor radiated during rise of the modern Cordillera de Mérida during the Pliocene.

Etymology. *Ericksonae* is a patronym for Ronna Erickson, a research engineer and astronomer at the University of Massachusetts Amherst USA. She has greatly supported the field research of the senior author. The name of this species is used in genitive feminine.

Remarks. CVULA 5572 presents an abnormality on the right foot. Specifically, the TIV is shorter than normal, lacking the distal phalanx, but having otherwise an apparently normal disc. Abnormalities on limbs and digits have been observed in many species of Venezuelan dendrobatids (e.g. *Mannophryne cordilleriana* and *M. riveroi*, see Barrio-Amorós *et al.* 2010b, 2010c).

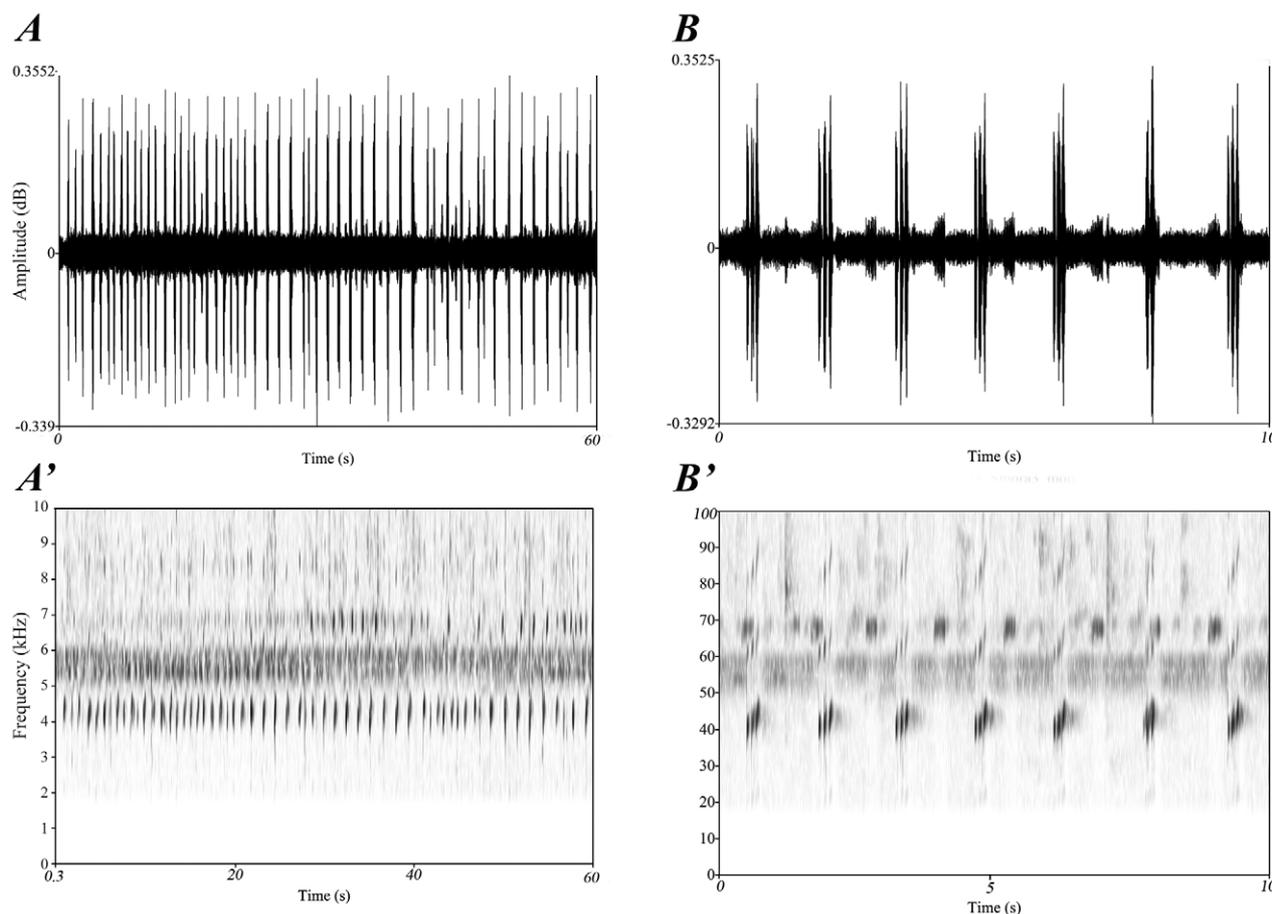


FIGURE 8. (A) Waveform and (A') spectrogram of a 60 seconds sequence of the trill call of *Aromobates ericksonae* **sp. nov.** Indistinct harmonics can be seen at around 6700 Hz. (B) Waveform and (B') spectrogram of a 10 seconds sequence of the trill call of *Aromobates ericksonae* **sp. nov.**, showing seven notes; harmonics can be seen at around 6700 Hz. Background noise from running water has been reduced and frequencies below 100–500 Hz were filtered from the spectrograms.

Aromobates zippeli **sp. nov.**

Holotype. CVULA 8329, an adult male from the immediate surroundings of Mucuchíes, Estado Mérida, Venezuela, 8.75 N, 70.8833 W, elevation 2970 m, collected by Willians Abreu on December 28th, 2004.

Paratopotypes. Four adult males, CVULA 8331-33, 8336, and seven adult females, CVULA 8330, 8334-35, 8337-40, all with the same data as the holotype. One subadult female, CVULA 8341, and a juvenile, CVULA 8342, collected by W. Abreu on April 28th, 2010, at the type locality.

Tadpoles. Unknown.

Definition: We assigned this species to *Aromobates* based on the following characters. (1) Skin on dorsum smooth to shagreen. (2) Paired dorsal scutes present on digits. (3) Distal tubercle on FIV present. (4) FIV length surpasses half distal subarticular tubercle of FIII. (5) FI slightly longer than II. (6) Digital discs present. (7) Finger discs barely expanded. (8) Finger fringes present. (9) Metacarpal ridge absent. (10) FIII not swollen in adult males. (11)

Carpal pad absent. (12) Male excrescences on thumb absent. (13) Thenar tubercle present, small. (14) Black arm gland in adult males absent. (15) Tarsal keel indistinct. (16) Toe discs weakly expanded. (17) Toe webbing formula III $\frac{1}{2}$ -3III $\frac{2}{2}$ -3IV. (18) Metatarsal fold absent. (19) External coloration with pale dorsolateral stripes; ventrolateral stripe absent; oblique lateral stripe formed by a series of diffuse bluish white spots. (20) Gular-chest markings absent, evenly stippled. (21) Dermal collar absent. (22) Male throat coloration dirty white to grey with profusion of melanophores, evenly stippled; female throat coloration white with scattered melanophores. (23) Male abdomen color white with scattered melanophores and indistinct bluish white small spots. (24) Female abdomen color pattern white with few scattered melanophores. (25) Iris coloration bronze with fine black reticulation, pale golden pupil ring. (26) Large intestine unpigmented. (27) White testes. (28) Median lingual process absent. (29) Tympanum indistinct, tympanic annulus absent. (30) Vocal sac distinct. (31) Teeth present on the maxillary arch. (32) Body size small, males ($n = 5$) 19.0–21.5 mm, mean = 20.6 \pm 1.0; females ($n = 7$) 20.0–25.0 mm, mean = 23.5 \pm 1.6.

Comparisons. *Aromobates zippeli* (characters in parenthesis) differs by its smaller size from large species of the genus, such as *A. alboguttatus*, *A. leopardalis*, *A. meridensis*, *A. nocturnus*, and *A. capurinensis*, all with maximum SVL > 31 mm (up to 25.5 mm). However, it is worthy to compare *A. zippeli* with *A. alboguttatus* and *A. leopardalis*, both larger species but very close geographically; *A. zippeli* could occur sympatrically with *A. leopardalis*. *Aromobates alboguttatus* is a larger species, up to 31.5 mm (females up to 25 mm), with a low but distinct tarsal keel (indistinct to absent), fringes on fingers evident (absent to low), basal webbing (I-III $\frac{1}{2}$ -3III $\frac{2}{2}$ -3IV-V), and this species has reticulated throat (evenly stippled). *Aromobates leopardalis* is much larger, females reaching 35.3 mm (25 mm), with no dorsolateral stripes, and not defined oblique lateral stripe (both present and distinct), extensive flap-like fringes on toes (present but not extensive), and FI shorter than FII (FI longer than FII). Nine other species of *Aromobates* (*A. cannatellai* **sp. nov.**, *A. durantii*, *A. ericksonae* **sp. nov.**, *A. haydeae*, *A. mayorgai*, *A. molinariii*, *A. orostoma*, *A. saltuensis*, and *A. serranus*) are similar in size to *Aromobates zippeli*. *Aromobates cannatellai* is somewhat larger, with females up to 28.6 mm (25.0), finger discs moderately expanded (barely expanded), toe discs moderately expanded (weakly expanded), this species has a distinct tarsal keel (indistinct to absent) and dorsal surfaces lack small white spots (present). *Aromobates durantii* is a little larger frog, up to 30.7 mm (up to 25.0 mm), this species has its venter grey with conspicuous white dots (grey to white), tarsal keel short but distinct (indistinct to absent). *Aromobates ericksonae* has a thick tarsal keel (indistinct to absent), toe webbing basal (moderately webbed I-III $\frac{1}{2}$ -3III $\frac{2}{2}$ -3IV-V), and this species surfaces are never covered by small white spots (present). *Aromobates haydeae* has no fringes on fingers (present), dorsal color reddish copper without white spots (dark to light brown with white spots), and ventral parts orange (dirty yellow). *Aromobates mayorgai* has dorsal skin smooth with a few tubercles posteriorly (smooth to shagreen without tubercles), FI shorter than FII or equal (FI longer than FII), oblique lateral stripe absent (formed by small whitish spots), and tarsal keel prominent (indistinct to absent). *Aromobates molinariii* is a larger frog, females up to 30.3 mm (up to 25.0 mm), it has FI and II equal in length (FI longer than FII), fringes absent on fingers (present), disc on FIII twice as wide as the adjacent phalanx (1.3 wider), and oblique lateral stripe absent (formed by small whitish spots). *Aromobates orostoma* has a distinct tympanum (indistinct), FI shorter than FII (FI longer than FII), and no fringes on fingers (present). *Aromobates saltuensis* has no fringes on fingers (evident), tarsal keel well-defined (indistinct to absent), and a short anal sheath (absent). *Aromobates serranus* has a distinct tympanum (indistinct), FI shorter than FII (FI longer than FII), tarsal keel present (indistinct to absent), dorsum reticulated (spotted but not reticulated), and belly parts creamy white with brown reticulation (yellow to white, no reticulation).

Description of the holotype. The holotype is an adult male of 22.0 mm (SVL): body robust, rounded in cross-section; dorsal skin, including dorsal surfaces of hind limbs, shagreen in preservative; throat and chest skin smooth, belly, finely granular; head as long as wide, HeL = 34.5% of SVL; HW = 34.5% SVL; snout is rounded in profile, rounded in dorsal and ventral view; nares are situated laterally to the tip of snout; narial openings are barely visible when viewing the head from the front, not visible when viewing dorsally; and barely seen when viewing from a ventral aspect; canthus rostralis is straight, rounded; the loreal region is flat; interorbital region is little narrower than the upper eyelid; snout is longer than ED; tympanum is indistinct, only barely visible on its anteroventral portion; supratympanic bulge is indistinct; tympanum is positioned closely behind eye and lower, close to the angle of jaws; teeth are present on maxillary arch; vocal slits are large, arched from mid-level of tongue to anterior to the angles of jaws; tongue is large, rounded, and one-third free posteriorly.

Hand of moderate size (25% SVL); relative lengths of adpressed fingers are III > I > II > IV; discs of all fingers are slightly expanded, horizontally oval; FIII is barely wider than distal end of adjacent phalanx; the base of palm

has a large, rounded palmar tubercle; and on base of FI there is an indistinct, smaller (approximately 1/3 of the palmar tubercle), oval thenar tubercle; one or two subarticular tubercles on fingers (one each on FI and FII, two each on FIII and FIV, the distal one of FIV inconspicuous); and all tubercles are flat and round or oval; without supernumerary tubercles. Fringes are present on all fingers and quite distinct.

Hind limbs quite short, $SL = 45\% SVL$; relative lengths of adpressed toes are $IV > III > V > II > I$; TI is moderately long, the tip reaching the mid-subarticular tubercle of TII; toe discs are barely expanded, TIV about 1.2 times wider than distal end of adjacent phalanx; feet are basally webbed, formula is $I-II1\frac{1}{2}-3III2\frac{1}{2}-3IV-V$; fringes on toes are present and low; one to three non-protuberant small subarticular tubercles are present (one on TI and TII, two on TIII and TV, three on TIV, proximal one almost indistinct); two metatarsal tubercles are present, including a small round outer, and a similar in size elongated inner tarsal tubercle; tarsal keel is indistinct; cloacal opening is located at the upper level of thighs; and the anal sheath is absent.

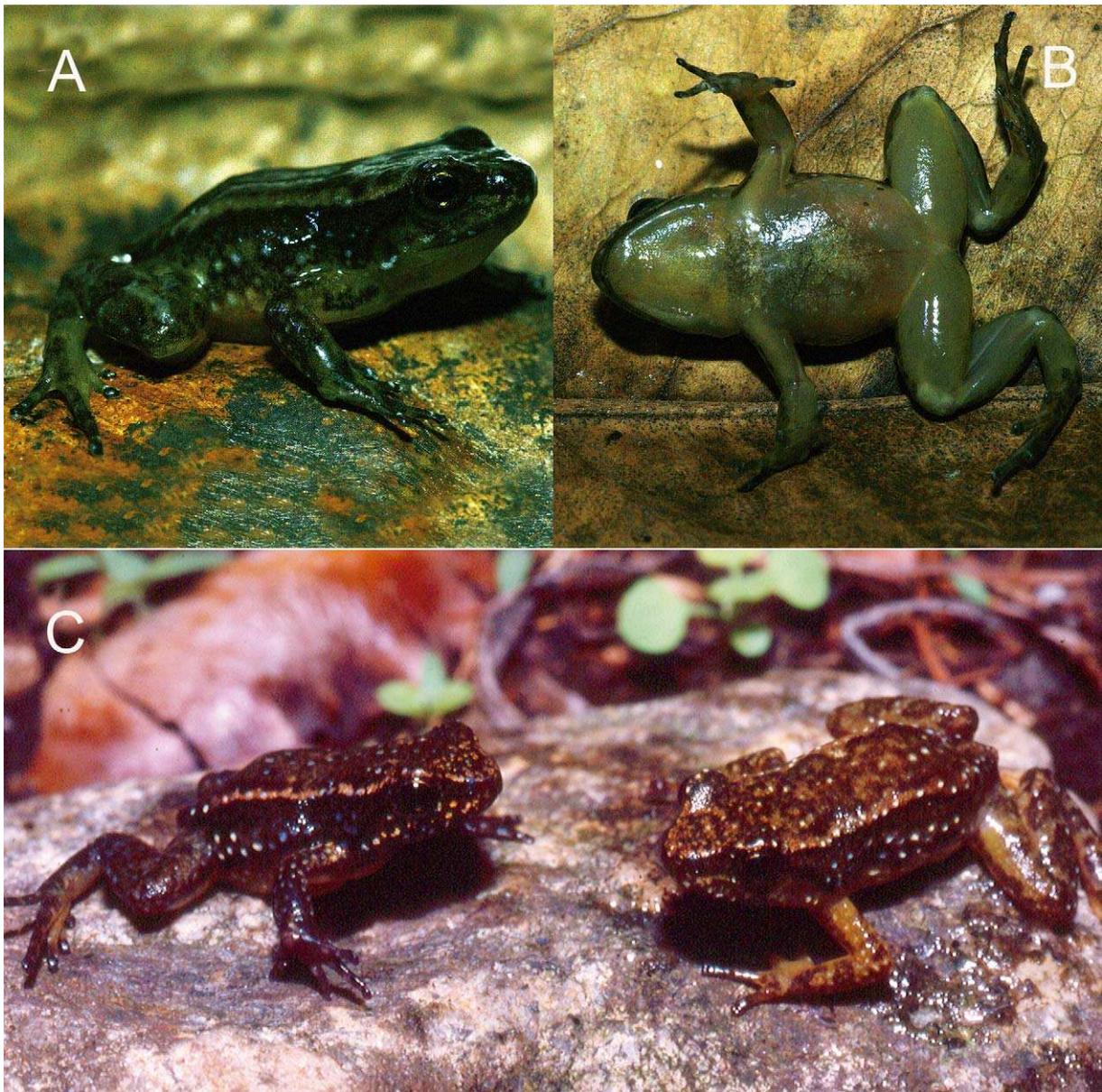


FIGURE 9. *Aromobates zippeli* sp. nov. (A) Lateral view of subadult paratopotype female CVULA 8341. (B) Ventral view of same specimen in A. (C) two uncollected specimens, showing variation in pattern. Photos by CBA.

Measurements of holotype (in mm). SVL: 22.0; SL: 9.9; FL: 8.8; HeL: 7.6; HW: 7.6; ETS: 3.2; EN: 1.8; ED: 2.6; TD: 1.2; HD: 5.5; 1FiL: 3.9; 2FiL: 3.8.

Color of the series. After preservation, *Aromobates zippeli* specimens have their dorsum very dark brown, with a little evident vertebral stripe made by small pale white spots, starting from the tip of snout to the sacrum. The two dorsolateral stripes are pale blue in life (appearing white in preservative) with small spots. Specimen flanks are black with an irregular oblique lateral stripe also made by small dirty white spots; some white spots are on the lowest part of the flanks, close to the paler ventral area. Arms are dark brown with fine irregular pale brown spotting. Hind limbs are dark brown without definite crossbars; thighs are paler than shanks, all with irregular pale brown spotting. Ventrally, throat and chest grey are evenly spotted with a profusion of melanophores (only seen under magnification); belly is dirty white, with scattered melanophores, and a few indistinct (only possible to distinguish under microscope) bluish white spots. Palms and soles are dark brown.

Coloration in life specimens (based on living, not collected specimens and CVULA 8341) is as follows. The dorsum can be olive (Fig. 9A), light to dark brown or even black, with an irregular pattern. Alternatively, the dorsum can have irregular spotting of dark brown over a light brown background or it can be brown over black (Fig. 9B). Flanks are dark brown to black, always darker than the dorsum. The two light dorsolateral stripes can be pale yellow, golden, or olive; but are always paler than the dorsum and flanks. The ventrolateral stripe is absent. The oblique lateral stripe can be short, just in the inguinal area, or long, extending above the upper arms. This stripe is always formed by small bright white spots, than can be pale blue at midbody or above the upper arms. Arms and hind limbs have also a pale background with irregular dark spotting or crossbars. The most striking chromatic character of *Aromobates zippeli* is that any part of its dorsal body can be covered by small rounded bluish to white spots, especially in the flanks, dorsum (even over the dorsolateral stripes), upper lips and loreal region. Ventrally (Fig. 9B) a subadult female (CVULA 8341) was found to be patternless with a dirty yellow to dirty white background; a series of very diffuse small pale white spots were distinguished under magnification.

Variation. Dorsal skin can be smooth to shagreen without tubercles, both in living and preserved specimens. Fringes on fingers and toes are present on all specimens. Tarsal keel in this species is indistinct in almost all specimens, but it is completely absent in CVULA 8338 and 8340. Only CVULA 8335 and 8337 have what appears to be an anal sheath; this data agrees with Grant *et al.* (2006), which prevented that this characteristic can be variable. CVULA 8339 presented an extra toe on the left foot. The extra toe rises from the TIII towards its superior side, being of the same length and having a small disc and fringes. This case of complete polydactylia is the second reported in Venezuelan dendrobatids, after a case of partial polydactylia on *Mannophryne riveroi* (Barrio-Amorós *et al.* 2010c). CVULA 8334 has the last section of FII and III missing, showing the phalanges exposed.

The color pattern variation was also present (Fig. 9C). One individual (CVULA 8340) has irregular dark blotches on dorsum; while the series CVULA 8337-38 are patternless dorsally and their dorsolateral stripes are thin but not made with spots. Crossbars are also variable from distinct (CVULA 8332, 8335) to indistinct or ill-defined. Ventrally, males are usually darker than females. Male CVULA 8333 is the darkest in coloration, CVULA 8331-32 are similar to the holotype, and CVULA 8336 is the ventrally paler in coloration. This last specimen appears to be white at first glance with some melanophores on the throat under magnification. Females of *Aromobates zippeli* have paler ventral surfaces, mostly whitish with few scattered melanophores on the throat.

Natural history. *Aromobates zippeli* is a subpáramo dweller, known only from the type locality at 2970 m and restricted to only one creek. The individuals of this species were found under rocks and between aquatic and terrestrial vegetation along the creek. Tadpoles are unknown.

Vocalization. Unknown.

Phylogenetic relationships. Unknown. However, Manzanilla *et al.* (2009) provided molecular data from individuals identified as *Aromobates* sp. (MIZA 310-312) collected along the road from Mérida city to Barinitas, near the locality of Mucuchíes. We did not review those specimens and, consequently, we cannot rule out that they may be conspecific with *A. zippeli*.

Distribution. *Aromobates zippeli* is known from a small area of probably less than 5 km² NE of the town of Mucuchíes. During our search, we were able to find individuals only in a single creek. This general area has been profusely searched for amphibians for many years (especially, by professors and students of the Universidad de los Andes) as supported by plenty of specimens collected and housed in the CVULA collections. Based on the current information, we regard this species as a restricted endemic taxa to Mucuchíes (Fig. 3). *Aromobates leopardalis* is sympatric and it might be syntopic with *A. zippeli*. However, *A. leopardalis* is much larger and likely to be restricted to a more open páramo habitat.

Etymology. *Zippeli* is a patronym for Kevin C. Zippel, due to his passion and endless work in spreading the alarming message of the global decline of frogs, through the Amphibian Ark (Amphibian Ark: <http://www.amphibianark.org/>).

Notes on *Aromobates saltuensis* (Rivero, 1978)

Taxonomic history. *Aromobates saltuensis* was described originally as *Colostethus saltuensis* by Rivero (1978) from a single unsexed adult UPRM 5147. This specimen was collected by J.A. Rivero and A.E. Esteves on June 3rd, 1971 along the road between La Fría and Michelena, Estado Táchira, Venezuela, at an elevation of 830 m. The approximate coordinates of the type locality are 7.9676 N and 72.2612 W with an uncertainty of at least 5 km. Grant *et al.* (2006) provided information of two putative populations of *C. saltuensis* in northern Colombia and transferred this species to *Aromobates*. However, the information about the specimens identified by Grant *et al.* (2006) as *A. saltuensis* has some limitations (see distribution and phylogenetic relationships below). To this date, the distribution, conservation, and natural history of *A. saltuensis* are little known.

Referred specimens. In 2007, we collected several specimens and we referred them as *A. saltuensis*. CVULA 8315–20, from the road between San Félix and San Juan de Colón, Estado Táchira, Venezuela, 8.0736 N, 72.2293 W, 751 m; CVULA 8321–24 from Rio Negro (Parque Nacional ‘El Tamá’), Estado Táchira, Venezuela, 7.5787 N, 72.1790 W, elevation 482 m.

Diagnosis. (1) Skin on dorsum smooth to finely granular. (2) Paired dorsal scutes present on digits. (3) Distal tubercle on FIV present. (4) FIV length reaches half distal subarticular tubercle of finger III. (5) FI longer than II. (6) Digital discs present. (7) Finger discs weakly expanded. (8) Finger fringes mainly absent. (9) Metacarpal ridge absent. (10) FIII not swollen in adult males. (11) Carpal pad absent. (12) Male excrescences on thumb absent. (13) Thenar tubercle present, small. (14) Black arm gland in adult males absent. (15) Tarsal keel thick, long, to mid-tarsus, straight, ending tubercle-like. (16) Toe discs moderately expanded. (17) Toe webbing reduced. (18) Metatarsal fold absent. (19) External coloration with pale dorsolateral stripes; ventrolateral stripe absent; oblique lateral stripe formed by a series of diffuse yellow and white spots. (20) Gular-chest markings present but indistinct on males, absent on females. (21) Dermal collar absent. (22) Male throat coloration grey with profusion of melanophores; female throat coloration dirty white with a profusion of melanophores. (23) Male abdomen color immaculate white. (24) Female abdomen color dirty white. (25) Iris coloration bronze with black reticulation; pupil ring bronze. (26) Large intestine unpigmented. (27) Testes white. (28) Median lingual process absent. (29) Tympanum distinct in its inferior half, tympanic annulus absent. (30) Vocal sac distinct. (31) Teeth present on the maxillary arch. (32) Size small, males ($n = 3$) 22–23.1 mm, mean 22.6 ± 0.6 ; females ($n = 3$) 22.9–26.9, mean 25.5 ± 2.3 .

Variation in the referred series. Rivero (1978) described variation within *A. saltuensis* by confronting the type specimen against a subadult (UPRM 5593) also collected from the type locality. However, the description provided in Rivero (1978) does not allow us to unambiguously evaluate the variation among our referred series. Therefore, we provide a description of the variation only based on our referred series. Males have a SVL range of 22–23.1 mm, and females SVL range of 22.9–26.9 based on three adult specimens of each sex. Body is slender, round to quadrangular in cross-section. Dorsal skin, including dorsal surfaces of hind limbs is smooth, shagreen to finely granular, without tubercles on the posterior part of the body in preservative. Ventral skin is smooth. Head is longer than wide, HeL = 35.8–37.9 % SVL; HW = 32.8–36.3 % SVL. On males, snout is truncated to pointed in profile. On females, snout is rounded to slightly pointed. Nares are situated laterally to the tip of snout; narial openings are barely visible when viewing the head from the front, not visible when viewing dorsally; and they are visible when viewing from a ventral perspective. Canthus rostralis is angular, the loreal region is almost flat; interorbital region is little wider than the upper eyelid. Snout is longer than ED. Only half of the tympanum is distinct and it is visible at its anteroventral portion but concealed posterodorsally by a low supratympanic bulge formed by the superficial slip of *m. depressor mandibulae*. The tympanum is positioned closely behind eye and close to the angle of jaws. Teeth are present on maxillary arch. Vocal slits are short to long, arched from mid-level of tongue to the anterior angle of the jaws. The tongue is short, oval to triangular; it is half free posteriorly.

Hand is of moderate size (25–28.2 % SVL); relative lengths of adpressed fingers are III > I > II > IV; discs of all fingers are weakly expanded, horizontally oval to round; FIII is barely wider than distal end of adjacent phalanx, at its maximum point is 1.3 wider; the base of palm has a large, palmar tubercle, rounded, quadrangular to heart-shaped; and on the base of FI there is a smaller (approximately 1/3 of the palmar tubercle), oval to elongate thenar

tubercle; one or two subarticular tubercles on fingers (one each on FI and FII, two each on FIII and FIV); and all tubercles are flat to round or oval; without supernumerary tubercles. Fringes on fingers are mainly absent, but on some males and females can be present, they are low and indistinct on FII and FIII.

Hind limbs are of moderate length, $SL = 47.5\text{--}52.9\%$ SVL; relative lengths of adpressed toes are $IV > III > V > II > I$; TI is short, the tip never reaching the mid-subarticular tubercle of TII; toe discs are round to oval, moderately expanded, TIV is about 1.8 times wider than distal end of adjacent phalanx; feet is moderately webbed; formula is $I2\text{--}2\frac{1}{2}II2\text{--}3\frac{1}{2}III3\text{--}4IV$ and basal between TIV and TV; fringes on all toes are notable, short but flap-like with a folding around the toes; one to three non-protuberant, subarticular tubercles are present but small (one on TI and TII, two on TIII and TV, three on TIV, proximal one almost indistinct); two metatarsal tubercles are present, including a small rounded outer, and a slightly larger oval inner tarsal tubercle; mid-tarsal tubercles are seldom seen; tarsal keel is well-defined, thick, straight, transverse across tarsus, from proximal edge of inner metatarsal tubercle to mid-tarsus, with an ending tubercle-like structure; cloacal opening is located at the upper level of thighs, with short tube flap or anal sheath.

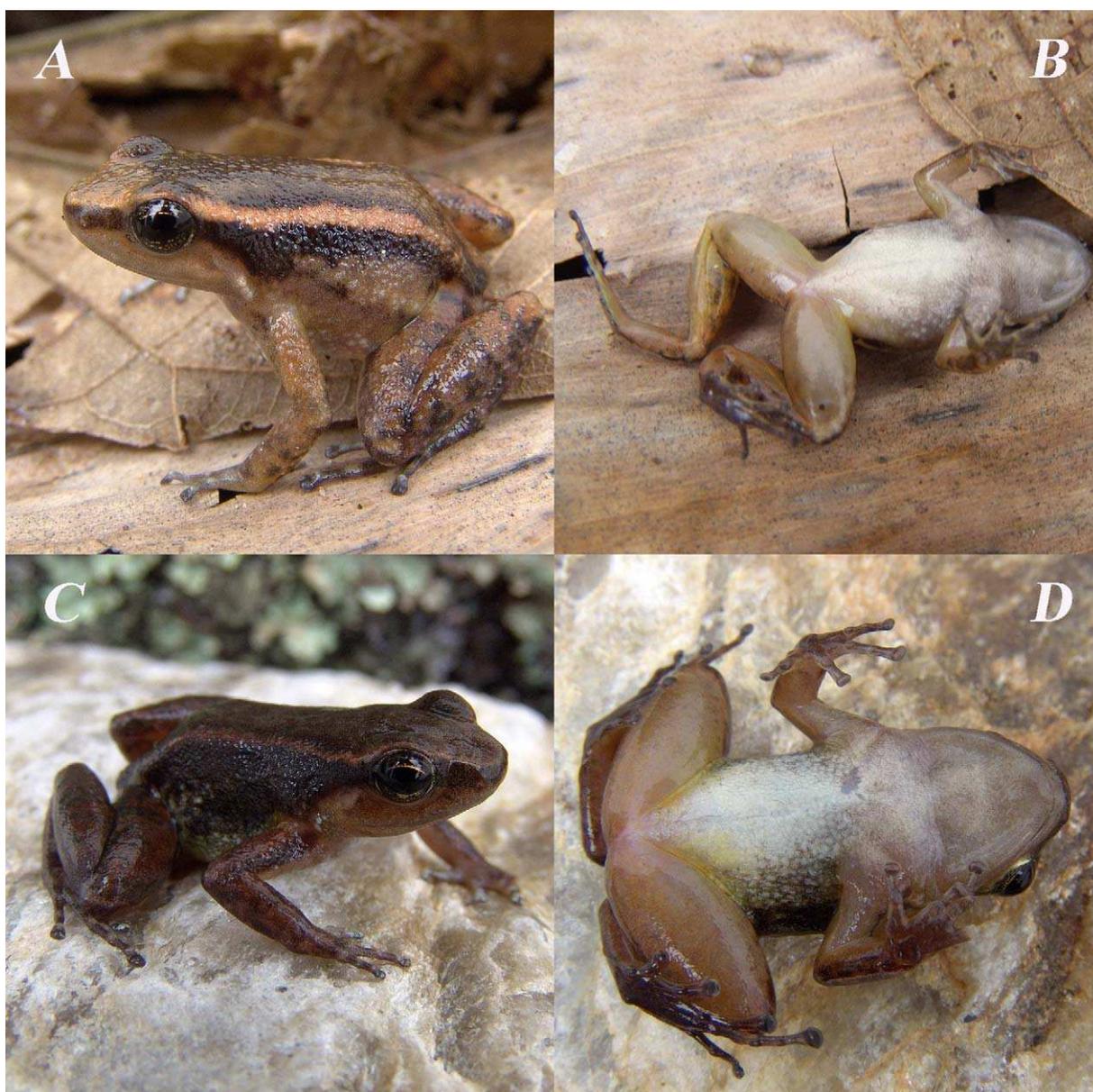


FIGURE 10. *Aromobates saltuensis* from the road from San Felix to San Juan de Colón, Estado Táchira, near type locality. (A) Adult male CVULA 8317. (B) Ventral view of the same specimen. (C) Adult female CVULA 8320. (D) Ventral view of the same specimen. Photos by CBA.

Comparison with the original description. When compared with the original description (Rivero 1978), we found a few additional details that should be noted. First, Rivero (1978) does not note a supratympanic bulge, but this characteristic varies depending on the preservation method. Second, Rivero also mention the presence of basal webbing between FII-III-IV, which is absent on the referred specimens examined (another character that can vary in preserved specimens). All the remaining characteristics described for the holotype are among the variation seen in the new specimens collected.

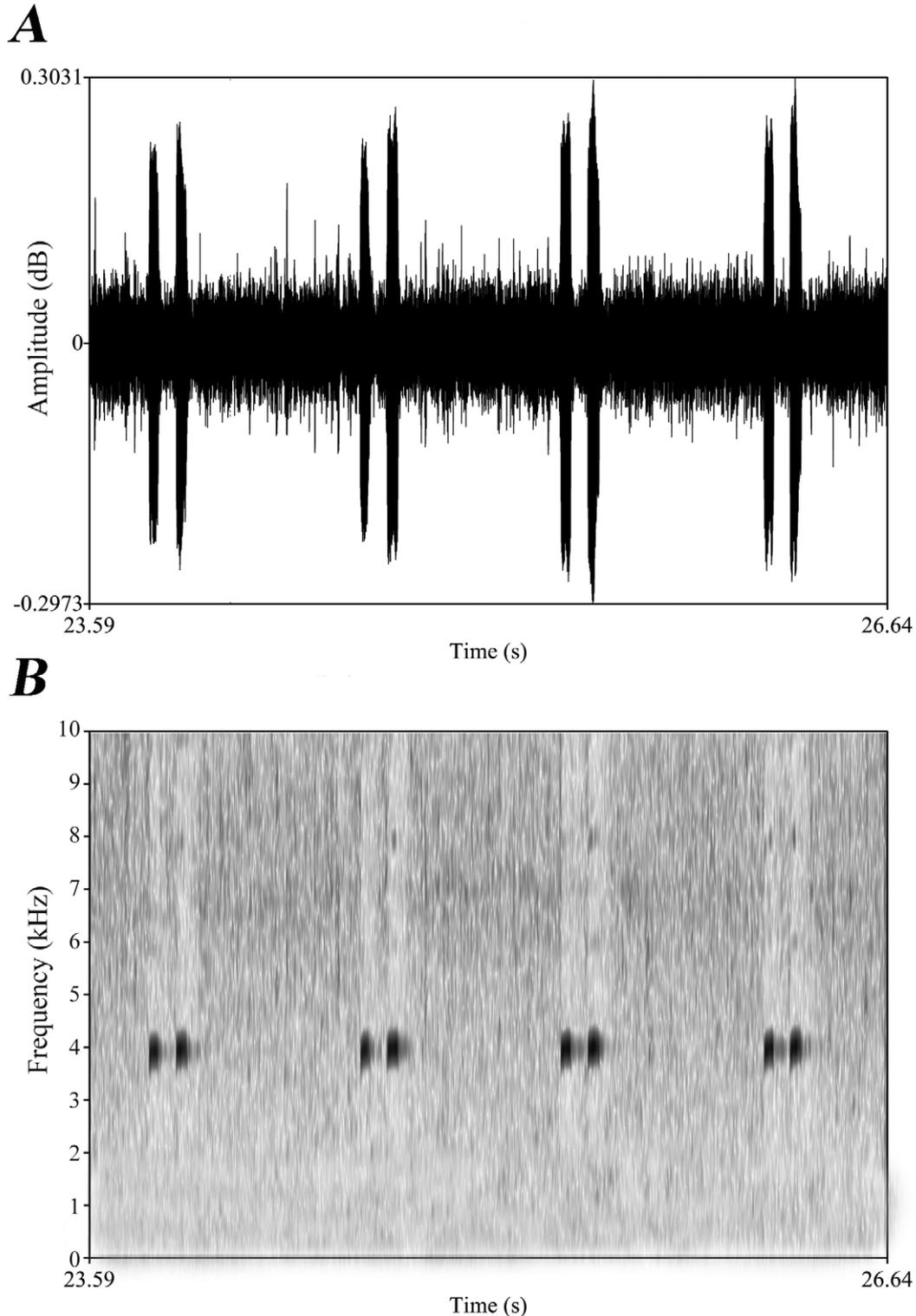


FIGURE 11. (A) Waveform and (B) spectrogram of a 2.6 seconds sequence of the call of *Aromobates saltuensis* **sp. nov.**, showing four duplets. Indistinct harmonics can be seen at around 8000 Hz. Background noise from running water has been reduced and frequencies below 100–500 Hz were filtered from the spectrogram.

Color. The referred specimens in life have dorsa with pale to dark brown coloration, patternless or presenting irregular spotting; the dorsolateral stripes can be very distinct (Fig. 10A) to ill-defined (Fig. 10C), pale orange to dark brown. The flanks can have a narrow dark brown longitudinal band below the dorsolateral stripe or they can be completely dark brown to the ventral area. The oblique lateral stripe can be solid, diffuse, or almost absent (consisting in a few, irregular and indistinct dirty white spots in the inguinal zone, even not forming a real stripe). Upper lips are patternless. Arms and hind limbs can be pale orange or brown to dark brown, with darker well-defined to ill-defined crossbars. Ventrally, males are pale grey on the throat and chest, white on the belly, with some opaque yellow on the hind limbs (Fig. 10B). Females are plain grey on the throat, dirty white on the belly (Fig. 10D). Iris is bronze with black reticulation; iris ring is also bronze.

Preserved specimens have a minimal dichromatism slightly apparent ventrally on males, as they have darker throats (pale to dark grey), consisting of a profusion of melanophores under magnification. Females are all white (with a few melanophores under magnification). The rest of the body coloration is not dichromatic in both sexes, varying in being paler or darker, with well or ill-defined crossbars on hind limbs.

Natural history. We found all *Aromobates saltuensis* associated to a slow flowing creeks surrounded by pastures (type locality), primary, and secondary forests (Rio Frio, Tamá). Several males were calling from exposed and concealed sites in the creek, such as on the top of rocks or behind small waterfalls. Some females were seen moving along the creek as well. All individuals were active during daytime.

Vocalization. The analyzed call is a section of a trill that lasted several minutes. The call was recorded at 22.5°C (water temperature) at early afternoon on the June 9th, 2007 near the type locality (see referred specimens). The analyzed section of the call has four duplets of notes (Fig. 11), lasting 2 sec. One to two duplets of notes are produced per second. The dominant frequency of the first pulse is slightly lower than the second. The first pulse has a dominant frequency range of 3437.0–3862.7 Hz. The second pulse has a dominant frequency range of 3511.0–3862.0 Hz. Four consecutive duplets were chosen at random from the spectrogram to determine temporal variables. Duplet duration 0.15 ± 0.01 (0.14–0.16) seconds, and inter-duplet interval 0.46 ± 0.17 (0.34–0.65) seconds. Note and inter-note duration with their mean, SD and range in seconds are indicated as follows (1) note duration, first note 0.04 ± 0.01 (0.03–0.04); second note 0.05 ± 0.01 (0.04–0.05); and (2) inter-note, 0.07 ± 0.01 (0.06–0.07). Both, the waveform is shown by Fig. 11A and the spectrogram by Fig. 11B.

Distribution. *Aromobates saltuensis* is known from the southwestern extreme of the Cordillera de Mérida and northeastern Cordillera Oriental, and has been reported in northeastern Colombia by Grant *et al.* (2006) and Anganoy-Criollo (2012) from Departamentos of Norte de Santander, Boyacá and Cesar, respectively (Fig. 3). However, *A. saltuensis* is likely to be a Venezuelan endemic and the Colombian specimens might represent other species and closer to *A. cannatellai* **sp. nov.** as supported by our phylogeny (Figs. 1 and 2, see next section). For this reason, we revised the specimen information associated to *A. saltuensis* specimens provided by Grant *et al.* (2006) and we found it to be problematic. The information provided of the specimens ICN 42512-16 and 33587 is inaccurate as they are from two distinct and geographically distant localities on the western slopes of the Northern Andes of Colombia. Moreover, it is not justified why all specimens are assigned to a single population from Bacar[a]sica (the name is misspelled as ‘Bucarsica’ in the original paper, Grant *et al.* 2006: 242). First, the locality information for the specimens ICN 42512-16 corresponds to Corregimiento Pueblo Nuevo, Sitio El Reposo, 4.8 km on the road of Ocaña to Pueblo Nuevo (8.2304 N, 73.3890 W at ~1560 m). This locality is 61 km WNW of the Bacar[a]sica site provided by Grant *et al.* (2006). Second, the data provided for the specimen ICN 33587 corresponds only to the municipality and the following information is more accurate: 25.3 km by road from Sardinata at 1040 m, municipio Bacarasica, Norte de Santander. The approximate coordinates of ICN 33587 are 8.1326 N and 72.8960 W with an uncertainty of at least 3 km.

Grant *et al.* (2006) provided information of a third population that most likely is based on molecular data (no information is provided about specimen morphological identification). The voucher specimen is MUJ 3726 (Museo de Historia Natural, Universidad Javeriana, Bogotá, Colombia) from Cubará, Fátima, Quebrada Gralanday in Boyacá on the eastern slopes of the Northern Andes of Colombia, which is closer in our analysis to *A. cannatellai* than to *A. saltuensis*, although both species are nested in a “*saltuensis*” clade. The approximate coordinates for the MUJ 3726 specimen are 6.8790 N and 72.1879 W. The distance from the Cubará population to the two other northern populations in Norte de Santander is 160–200 km in straight line. Moreover, all localities of the *A. saltuensis* specimens from Grant *et al.* (2006) are at least 61 kms of distance from the approximate type locality of *A. saltuensis* provided by Rivero’s description near Michelena (7.9676 N and 72.2612 W) on the Mérida Andes. Specifically,

Sardinata is at least 61 km W, Pueblo Nuevo is at least 128 km WNW, and Cubará is at least 122 km S from Michelena. Anganoy-Criollo (2012) reported further extension in the distribution of *A. cf. saltuensis* from two new localities in the southernmost edge and western versant of the Sierra de Perijá at ~133 km northwest from Michelena type locality. These two new localities are very close within the Departamento de Cesar, namely, El Gitano (near Rio de Oro at approximately 8.3166 N, 73.4167 W) and San Cayetano (near Gonzales at approximately 8.3931 N, 73.3844 W). La Marca (2005), on the other hand, has suggested that *Aromobates* species most likely show restricted distributions with no more than 60–70 km². Therefore, it is unclear to us if all these specimens correspond to a single species or even less if they are conspecific with *A. saltuensis* or even with *A. tokuko* (see phylogenetic relationships below). Thus, we consider the presence of *A. saltuensis* in Colombia as highly unlikely, but closely related allies might be present.

Our referred specimens are less than 13 km from the type locality near Michelena. The original description *A. saltuensis* (Rivero 1978) indicates a collection site at an elevation of 830 m on the road from La Fria (elevation 119 m) to Michelena (elevation 1230 m) in Estado Táchira (Venezuela); this road is 25 km long. The locality of the CVULA 8315–20 specimens are from the same road described by Rivero (1978), and about 12.5 km from the georeferenced type locality. In consequence, we consider the CVULA 8315–20 series to be topotypic. The series CVULA 8321–24 from ‘Rio Frío, Tamá’ is about 44 km from type locality. Therefore, according to our data, we consider *Aromobates saltuensis* to be endemic to Venezuela. However, we suggest a careful comparison of all Grant *et al.* (2006) and our specimens with the type UPRM 5147 (University of Puerto Rico) to further confirm our insights.

Phylogenetic relationships. We found *Aromobates saltuensis* to be well-nested in *Aromobates* (Fig. 1). This suggests a relative recent extension and radiation of the *saltuensis* clade into Eastern Cordillera of Colombia. Grant *et al.* (2006) provided nuclear and mitochondrial sequences from a specimen identified as *A. saltuensis* with the voucher number MUJ 3726 from Cubará (Colombia: Boyacá). However, it is unclear if these authors actually identified the specimen as *A. saltuensis* as discussed on previous section. We found MUJ 3726 to be closely related to *A. cannatellai* based on the 12S-16S rRNA segment. At the moment, we suggest recognizing MUJ 3726 as voucher from a different species and we restrict *A. saltuensis* only to the Venezuelan localities.

The taxonomic status of *Aromobates inflexus* (Rivero, 1978)

Colostethus inflexus has a problematic taxonomic history. It was described by Rivero (1978), who collected two specimens that seemed different to him among many “*alboguttatus*” from “El Almogral” near Boca de Monte on the crossroad between La Grita-Bailadores and Pregonero roads (Venezuela, Estado Táchira) at 3075 m. Initially, Rivero considered that enough evidence supported *C. inflexus* as a valid species, but later he considered that all specimens from El Almogral were indeed of *Aromobates alboguttatus* and later Rivero (1984) synonymized *Colostethus inflexus* with *A. alboguttatus*. However, as is currently known, *Aromobates alboguttatus* is endemic to northern Mérida city (Rivero 1961; Barrio-Amorós 1998, 2004; La Marca 2005), at about 100 km NNE from the *C. inflexus* type locality. All species of *Aromobates* show rather restricted distributions with no more than 60–70 km² (La Marca 2005). Based on this precarious argument, we preliminary suggest that specimens assigned to *C. inflexus* must be different from *A. alboguttatus* and a proper comparison should be made. Rivero (1976) also described *Colostethus orostoma* from Boca del Monte, a locality close to the type locality of *C. inflexus*. Additionally, we reviewed a series of specimens (CVULA 3528-29) identified as *A. orostoma* and they do not fit the ventral color pattern described by Rivero (1976). We suggest a more thorough comparison of the type material along with new collections to resolve the status of *C. inflexus* and *A. orostoma*.

Aromobates vocalizations

Only the call of five species of *Aromobates* are known to date, those of *Aromobates meridensis* (Barrio-Amorós *et al.* 2010a), *A. ornatissimus* (Barrio-Amorós *et al.* 2011), *A. cannatellai*, *A. ericksonae* and *A. saltuensis* (this work). Table 3 resumes the basic parameters of each species vocalization.

TABLE 3. Basic parameters in *Aromobates* vocalizations.

Species	Temperature	Pulse rate (per second)	Dominant frequency (Hz)	Note length/call length (sec)
<i>A. cannatellai</i>	21.4 °C	5–6	2998–3237	0.04/CT ¹
<i>A. ericksonae</i>	22.3 °C	0.8	3730–4314	0.2/CT
<i>A. meridensis</i>	19.0 °C	9.4	2400–2950	0.05/2.4
<i>A. ornatissimus</i>	19.0 °C	0.3	3057–3120	0.10/–
<i>A. saltuensis</i>	22.5 °C	1–2 ²	3437–3863	0.15 ³ /CT

¹CT: continuous trill; ²notes in duplets; ³duplet instead of single pulsed note.

The current state of *Aromobates* species conservation

Aromobates species have suffered a significant decline in the last 20 years. Searches for *A. nocturnus*, *A. alboguttatus*, *A. duranti*, *A. haydeae*, *A. leopardalis*, and *A. serranus* during the past decade have failed to produce additional finds (Barrio-Amorós 2001). The last species seen were *A. mayorgai*, *A. molinari*, and *A. meridensis* (Barrio-Amorós *et al.* 2010a). In 2011, five new species (i.e., *A. totuko*, *A. ornatissimus*, and the three species described herein) and *A. saltuensis* are recognized as extant. *Aromobates* species decline is apparently related to altitude as most extant species occur below an elevation of 1000 m (e.g., *A. cannatellai*, *A. ericksonae* and *A. saltuensis*) and relatively abundant in their type localities (except *A. ornatissimus* that occurs at 2350 and was still abundant in March 2010 at its type locality and surroundings). Here, we provide a summary of the current conservation status of *Aromobates* based on our own observations and a new category list for all Venezuelan *Aromobates* (Table 4).

TABLE 4. List of endangered *Aromobates* species mentioned by selected different sources. DD= Data Deficient; LC= Least Concern; NT= Near Threatened; VU= Vulnerable; EN= Endangered; CR= Critically Endangered. Criteria follows the IUCN parameters (Stuart *et al.* 2008). The double category in Rodríguez & Rojas Suárez (2008) refers to the Regional / Global categories in the Red List. The *A. meridensis* account we follow in this work is that after Barrio-Amorós *et al.* (2010).

	Rodríguez & Rojas Suárez (2008)	Stuart <i>et al.</i> (2008)	this work
<i>Aromobates alboguttatus</i>	NT / EN B1ab(iii,v)+2ab(iii,v)	EN B1ab(iii,v)+2ab(iii,v)	CR A2abc+B1ab(ii,iv)
<i>A. cannatellai</i>	-	-	CR B2a(iii)
<i>A. capurinensis</i>	DD / DD		DD
<i>A. duranti</i>	DD / EN B1ab(iii)+2ab(iii)	EN B1ab(iii)+2ab(iii)	EN B1ab(iii)+2ab(iii)
<i>A. ericksonae</i>	-	-	EN B2a(ii,iii)
<i>A. haydeae</i>	DD / EN B1ab(iii,v)+2ab(iii,v)	EN B1ab(iii,v)+2ab(iii,v)	EN B1ab(iii,v)+2ab(iii,v)
<i>A. leopardalis</i>	DD / CR A2ace; B2ab(v)	CR A2ace; B2ab(v)	CR A2ace; B2ab(v)
<i>A. mayorgai</i>	DD / EN B1ab(iii)+2ab(iii)	EN B1ab(iii)+2ab(iii)	CR A2ace; B1ab(iii,v)
<i>A. meridensis</i>	NT / CR B2ab(iii)	CR B2ab(iii)	CR A2ace; B1ab(iii,v)
<i>A. molinari</i>	DD / EN B1ab(ii,iii)+2ab(ii,iii)	EN B1ab(ii,iii)+2ab(ii,iii)	EN B1ab(ii,iii)+2ab(ii,iii)
<i>A. nocturnus</i>	CR A2a; B2ab(v)/CR A2a; B2ab(v)	CR A2a; B2ab(v)	CR A2a; B2ab(v)
<i>A. orostoma</i>	DD / EN B1ab(ii,iii)+2ab(ii,iii)		EN B1ab(ii,iii)+2ab(ii,iii)
<i>A. saltuensis</i>	DD / EN B1ab(iii)	EN B1ab(iii)	EN B2a(ii,iii)
<i>A. serranus</i>	DD / EN B1ab(ii,iii)+2ab(ii,iii)		EN B1ab(ii,iii)+2ab(ii,iii)
<i>A. zippeli</i>	-	-	CR A2abc+B1ab(ii,iii,iv)

Aromobates alboguttatus was a species frequently encountered around Mérida city, at least until the 1990s. The senior author, along with parties of students of the Universidad de los Andes and other interested people, searched for it many times since 1995, with no success. Piñero & La Marca (1996) were the last to perform a natural history research on this species, using 149 specimens for a dietary study.

Aromobates leopardalis is a large species known from the páramo de Mucubají, where it was formerly abundant, as many jars of specimens are housed at the CVULA evidence. In August 1995 the senior author, accompanied by Amelia Diaz de Pascual, saw the last tadpoles and smelled the peculiar and deep skunk-like aroma of this species (possibly similar to that in *A. nocturnus*). Since then, and despite of many searches in the area by different teams of herpetologists and students, no further specimens have been found. In 2005, a thorough search for *Atelopus mucubajiensis*, a microsympatric species, was made (Barrio-Amorós 2009b); but no individuals of *A. leopardalis* were found.

Aromobates mayorgai was searched along with *A. meridensis* and *Atelopus carbonerensis* during 2000–2008 by the senior author and collaborators. *Aromobates mayorgai* was never found and it is considered extinct from its type locality, Chorotal, Mérida, at 1800 m, and we suspect a severe decline could extirpate most if not all of *A. mayorgai* populations.

Aromobates meridensis is a critically endangered frog, as the last census came up with an extremely worrying number: only 56–129 adults remained in 2006 at a site close to the type locality, where it is now considered extinct (Barrio-Amorós *et al.* 2010a). Moreover, life specimens of this species were infected with *Batrachochytrium dendrobatidis*, which has a worrisome prognosis (M. Lampo, pers. com.; Barrio-Amorós 2009c).

Aromobates nocturnus is the largest known dendrobatid, attaining an SVL of up to 64 mm. This species is also probably extinct, since searches for individuals of this species at its type locality since 1991 have been unsuccessful (Barrio-Amorós 2001). During a recent expedition by the senior author, no new specimens were found (Barrio-Amorós *et al.* 2011). However, a new species was discovered close to its type locality (*Aromobates ornatissimus*) which might be syntopic and abundant at its restricted distribution area. This opens the possibility that *A. nocturnus* might still exist in this area.

Aromobates zippeli can be considered as one of the most endangered extant *Aromobates*, as its habitat is completely surrounded by agricultural fields where agrochemicals are frequently employed. We urge for an immediate census of this species and the identification of more populations for an urgent protection and monitoring plan. Even with no more data at hand, we suspect that the species is currently at high risk of extinction. We propose that the species be listed under category CR A2abc+B1ab(ii,iii,iv) of the IUCN criteria (see Stuart *et al.* 2008) (Table 4).

Discussion

In terms of the *Aromobates* phylogenetic history, this genus is positioned near the ‘crown’ of all extant species of Dendrobatidae with an estimated divergence time in the late Oligocene (Santos *et al.* 2009). Given *Aromobates* age (i.e., ~35 MYA), the restricted distribution of *Aromobates* is remarkable. Several factors might have contributed to *Aromobates* limited distribution including the complex orogeny of the northern Andes and the development of the Orinoco Basin (Díaz de Gamero 1996). The rise of the Andes and marine incursions from the Caribbean at the beginning of the Miocene might have contributed to *Aromobates* isolation starting in the Miocene-Pliocene boundary (Manzanilla *et al.* 2009; Santos *et al.* 2009). Historical biogeography of dendrobatids also supports this complex scenario and suggests an *Aromobates* recent radiation at 16.960 ± 3.664 MYA (Santos *et al.* 2009). Therefore, *Aromobates* might have originated on the shores of a paleo-Orinoco river system that ended with the formation of the current Maracaibo Lake system (Hoorn *et al.* 1995).

We described three new species of *Aromobates* as well as confirmed that *A. saltuensis* is present near its type locality in Venezuela. We suggest that the *Aromobates* populations known from Colombia are at least one undescribed species and all them are members of the *saltuensis* clade, but are not *saltuensis sensu stricto*. Our three descriptions increase the known diversity of *Aromobates* to 17 species.

The most recent taxonomic and phylogenetic reviews of *Aromobates* were provided by Grant *et al.* (2006) and Barrio-Amorós *et al.* (2011). The only unambiguously optimized synapomorphies for this genus are molecular (Grant *et al.* 2006: page 160). However, several problems emerge with the diagnosis of *Aromobates* using Grant *et al.* (2006) criteria. First, the number species of *Aromobates* with molecular data used by them are only 5 of the 14 previously known species. Second, the number of sequences used to define molecular synapomorphies in *Aromobates* by Grant *et al.* (2006) is uneven and ranged from 1 to 9 genes. Specifically, *Aromobates molinari* and *A. cf. zippeli* included only a small 16S rDNA ~550 bp fragment gathered from the GeneBank, *Nephelobates sp.* WES 626 (i.e., *Aromobates ornatissimus*, see Barrio-Amorós *et al.* 2011) and *Aromobates* aff. *saltuensis* have 8 genes;

and *A. nocturnus* have 9 genes (~6,000 bp). Third, most of the 61 synapomorphies suggested by Grant *et al.* (2006: 257) are possible autapomorphies or synapomorphies of only one or two species of *Aromobates*. Specifically, the distribution of synapomorphies are as follows, 1 from the 28S gene (present in *Aromobates ornatissimus* and *A. nocturnus*), 8 from cytochrome *b* gene (present in *A. nocturnus*, *A. ornatissimus*, and *A. aff. saltuensis*), 47 from the H1 segment (supposedly present in all 5 species, but this is not possible to determine as 2 species include only the short 16S rDNA fragment), 1 from the RAG1 gene (present in *A. nocturnus* and *A. aff. saltuensis*), and 4 from the SIA gene (present in *A. nocturnus* and *A. ornatissimus*). Fourth, from the 9 genes sequenced in *Aromobates*, only 6 synapomorphies (i.e., 6/6,000 or ~0.10% of all sites) were found in nuclear data while the remaining 55 (i.e., 55/6,000 or ~0.92% of all sites) came from the mitochondrial data. These observations suggest extremely low phylogenetic information in their nuclear markers and an overwhelming contribution by mitochondrial genes.

Within the suggested mitochondrial synapomorphies, 47 are present in stem-and-loop RNA genes (i.e., H1: 12S, tRNA-Val, and 16S) and they represent 72.3% (i.e., 47/65) of all synapomorphies for *Aromobates*. Grant *et al.* (2006) apparently divided H1 segment into 16 smaller fragments derived from an initial Clustal X (Thompson *et al.*, 1997) alignment and provided a list of their unambiguously optimized synapomorphies within each fragment with a positional number. We tried without success to identify, which fragment corresponds to which section of the H1 and the actual lengths of each of the 16 fragments are unknown. The reason is that the Grant *et al.* 2006 supplementary material includes two files (1) an unaligned FASTA formatted sequences file 'dendro_mt.poy' with a '#' indicating the limiting regions between fragments and (2) the actual POY alignment file 'Grant_et al2006_IA.ss' which defines their homologous positions but it does not have such fragment identifiers. This data formatting renders the identification of H1 synapomorphies provided in Grant *et al.* Appendix 8 rather useless in its actual format. For us, this lack of clarity also reflects a well-known fact that mitochondrial ribosomal genes are very difficult to align (Schloss 2010) and alignments are strongly dependent on the software and the starting parameters employed (Morrison & Ellis 1997). In spite of these, Grant *et al.* (2006) used the POY alignment of the H1 segment to determine molecular synapomorphies for *Aromobates* and justified its validity by intuition and "strategic" use of the algorithms of POY (Grant *et al.* 2006: page 48). However, several simulation studies have showed that POY based alignments are strongly dependent on starting parameters, inferior to ML approaches (Ogden & Rosenberg 2007), and less accurate to inferred phylogenies (Strugnell & Nishiguchi 2007). For all these reasons, it is nearly an impossible task to determine from the Grant *et al.* (2006) H1 POY alignment if their diagnostic synapomorphies are truly homologous. Finally, the H1 segment has not been thoroughly explored in dendrobatids for several of the common problems with mitochondrial markers (e.g., introgression and incomplete lineage sorting). These problems might affect taxonomic conclusions drawn from mtDNA gene trees (Funk & Omland 2003). In consequence, we suggest that Grant *et al.* (2006) molecular synapomorphies based on H1 should not be used for taxonomic characterization of *Aromobates* or other any genera within Dendrobatidae (i.e., Dendrobatidae + Aromobatidae sensu Grant *et al.* 2006).

In the present study, we could also have ventured in providing more diagnostic molecular synapomorphies based on H1 segment (i.e., 12S and 16S rDNA mitochondrial genes) which was used to construct our phylogeny of *Aromobates*. However, we feel that it is a disservice to the community to perpetuate or even extend the use H1 alignments as source of synapomorphies without a clear idea of homology. In contrast, we do provide some molecular diagnostic characters based on two nuclear markers derived from unambiguous alignments of exons (Table 2). More molecular data from *Aromobates* are needed to fully characterize the molecular diversity of this lineage.

Within the morphological characters that diagnose *Aromobates*, we found several additional problems with Grant *et al.* (2006) criteria: pages 160–161 and further referred in Lötters *et al.* (2007): page 49. First, we found that some morphological characters are rather ambiguous and uninformative. For example, character 2 of Grant *et al.* (2006) indicates that pale oblique lateral stripe could be either present or absent, character 6 indicates that toe webbing could be from basal to extensive, and character 8 indicates that finger I could be either shorter than, equal to, or longer than finger II. These characters include all possible variations found in Dendrobatidae (i.e., Dendrobatidae + Aromobatidae sensu Grant *et al.*, 2006) and are uninformative at the genus level. Second, some diagnostic characters become uninformative as their states are evaluated across *Aromobates*. For example, character 3 of Grant *et al.* (2006) indicates that a pale dorsolateral stripe is present; however, we found that this character state is not always present and at least four species lack this character state (*A. leopardalis*, *A. meridensis*, *A. nocturnus* and *A. ornatissimus*). Other example is character 7 on the morphology of the third finger of adult males indicated as not swollen. However, Barrio-Amorós *et al.* (2010a) stated that *A. meridensis* has FIII swollen in males during repro-

ductive periods, a characteristic that might be present on other congenics. Third, Grant *et al.* (2006) does not discuss some very relevant autapomorphies of the type species, *Aromobates nocturnus*. For example, the noxious mercaptan-like odor or the presence of *adductor mandibulae externus superficialis* muscle present in *A. nocturnus*. However, we report here that *Aromobates leopardalis* might also have noxious mercaptan-like odor when handled, suggesting that this might be a diagnostic character in some *Aromobates*. Similarly, *adductor mandibulae externus superficialis* was included in the Lötters *et al.* (2007) diagnosis of *Aromobates* and might be a useful diagnostic character. Fourth, Grant *et al.* (2006) included a character present in a single species and this species was not evaluated for any other character (i.e., molecular or morphological). Specifically, character 15 indicates that the chromosome number for *Aromobates* is $2n = 24$ based only on the karyotype of *A. leopardalis*. We do not know more reports on chromosome number in *Aromobates* and this character should be considered with caution.

Phylogenetic studies have provided an excellent tool to identify new species and phylogenetic relationships within *Aromobates* and Dendrobatidae in a larger extent. However, we reiterate that the search for morphological, behavioral, physiological, and genomic synapomorphies that define *Aromobates* is far from complete. We clarify that at the core of our criticisms are not to restrain and hinder taxonomic advance, but proper generic diagnosis. We think is not enough to find well-supported clades to coin generic names. As represented here in *Aromobates*, molecular phylogenies support its monophyly, but we still much ignorant of what defines *Aromobates*. The only unambiguous synapomorphies of this clade are molecular, and among them we still are unsure which ones are truly homologous with the exception of a handful derived from nuclear markers. We hope future revisions of dendrobatid taxonomy will deliver those “unambiguously optimized synapomorphies” which will make generic definitions stable and trustworthy.

We found *Aromobates* to be monophyletic and sister to *Mannophryne* as in previous studies (Vences *et al.* 2003; Grant *et al.* 2006; Santos & Cannatella 2011; Santos 2012). Within *Aromobates*, two lineages were evidenced including (1) *Aromobates ornatissimus* with a distinctive larger branch length (i.e., number substitutions accumulated over time) and (2) a more inclusive *Aromobates* radiation with a southwestern distribution. Our phylogenetic tree supports a progression of the dendrobatid radiation of *Aromobates* and *Mannophryne* from northeast to southwest of the Cordillera de Mérida along the Venezuelan Andes. Therefore, our phylogenies suggest a biogeographic pattern for *Aromobates* that parallels that of *Mannophryne*. Both lineages seemed to have followed a pattern of dispersal and diversification along the rise of Cordillera de Mérida and formation of the modern Orinoco Basin (Manzanilla *et al.* 2009; Díaz de Gamero 1996). Furthermore, the phylogenetic position of the *Aromobates* unnamed species from Colombia suggests a relative recent (i.e., Pliocene) radiation of this genus into the northernmost extreme of the Cordillera Oriental of Colombia.

Aromobates is mainly a Venezuelan endemic genus with three populations of at least one unknown species in northeastern Colombia. Within its distribution, *Aromobates* is mainly endemic to the Cordillera de Mérida, from where the vast majority of species is known (15 of 17 species). However, members of this genus occur at the northeasternmost edge of the Northern Andes of Colombia (i.e., Cordillera Oriental) where it enters Venezuela (*Aromobates saltuensis*), and at the eastern versant of the Serranía de Perijá (*A. tokuko*; Rojas-Runjaic *et al.* 2011). For us is not surprising that new species such as *A. tokuko* are being discovered and many more could exist on the western slopes of the Perijá. Only describing amphibian diversity will provide enough leverage for promoting conservation initiatives in such megadiverse regions as the Venezuelan and Colombian Andes

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APPENDIX I. Specimens examined.

- Aromobates alboguttatus* (11).—VENEZUELA: Estado Mérida: Monte Zerpa, 2200 m, CVULA 1171, 1236–1238; La Mucuy, Parque Nacional Sierra Nevada, MHNLS 7089–7095.
- Aromobates durantei* (1).—VENEZUELA: Estado Mérida: Sierra de la Culata, CVULA 0845.
- Aromobates haydeae* (5).—VENEZUELA: Estado Táchira: 15 km SW Zumbador, CVULA 0910-0911, 0917; Páramo Zumbador, Mesa del Aura CVULA 1067–1068.
- Aromobates leopardalis* (7).—VENEZUELA: Estado Mérida: Páramo de Mucubají, 3400 m, CVULA 5890, 5892, 3083–3084; Mucubají, Paratypes MHNLS 15093–15095 (ex-UPRM 5227, 5340 and 5232 respectively).
- Aromobates mayorgai* (4).—VENEZUELA: Estado Mérida: La Carbonera CVULA 0281; El Chorotal (El Sinal), carretera Mérida a La Azulita, Paratypes MHNLS 15119–15121 (ex-UPRM 5340, 5590 and 5591 respectively).
- Aromobates meridensis* (18).—VENEZUELA: Estado Mérida: Vía Mérida-La Azulita, 2000 m (CVULA 1448, 1491, 1670); La Empalizada, El Chorotal (CVULA 2167); El Chorotal (CVULA 2328–2389, 2335); Carretera 7 km N San Eusebio, El Chorotal, Distrito A. Bello (CVULA 4767–4772); La Empalizada (CVULA 5056, 5060-5063).
- Aromobates molinari* (2).—VENEZUELA: Estado Mérida: Cascada de Bailadores (CVULA 1873–1874).
- Aromobates nocturnus*. VENEZUELA: Estado Trujillo: Agua de Obispos (EBRG 2219-28) (paratypes).
- Aromobates "orostoma"* (3).—VENEZUELA: Estado Mérida: Km 590 Páramo Batallón y La Negra (CVULA 3529); Estado Táchira: Páramo La Negra, vía Pregonero, 2500 m (CVULA 3528-29).
- Aromobates saltuensis* (9).—VENEZUELA: Estado Táchira, road between San Félix and San Juan de Colón (CVULA 8315–20). Rio Negro, Parque Nacional 'El Tamá' (CVULA 8321–24).
- Aromobates serranus* (5).—VENEZUELA: Estado Mérida: Vía El Morro (CVULA 3376–3381).