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A new species of *Andinobates* (Amphibia: Anura: Dendrobatidae) from west central Panama

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Abstract

Dendrobatid frogs are among the best known anurans in the world, mainly due to their toxicity and associated bright colors. A recently described dendrobatid genus, *Andinobates*, comprises frogs distributed among the Colombian Andes and Panama. During field work in the Distrito de Donoso, Colón province, Panama, we found a poison frog that we here describe as a new species. The new species belongs to the *A. minutus* species group and is described herein as *Andinobates geminisae* **sp. nov.** This new species differs from all other members of the group by having uniformly orange smooth skin over the entire body and a distinctive male advertisement call. The new species is smaller than other colorful dendrobatids present in the area, such as *Oophaga pumilio* and *O. vicentei*. We also provide molecular phylogenetic analyses of mitochondrial DNA sequences of dendrobatids and summarize genetic distances among *Andinobates* species. *Andinobates geminisae* occurs in Caribbean versant rainforest on the westernmost edge of the known distribution of *A. minutus*, and represents the fourth species within this genus in Panama. This is vulnerable to habitat loss and excessive harvesting and requires immediate conservation plans to preserve this species with a restricted geographic range.

Key words: *Andinobates geminisae* **sp. nov.**, conservation, DNA barcoding, geographic distribution, molecular phylogenetics, Panama, poison dart frogs

Resumen

Los dendrobátidos, están entre los anuros más conocidos del mundo, debido principalmente a sus colores brillantes que cautivan la vista humana. Un nuevo género de dendrobatidos recientemente descrito, *Andinobates*, incluye ranas distribuidas entre los Andes colombianos y Panamá. Durante trabajos de campo realizados en el área del Distrito de Donoso, Provincia de Colón, Panamá, hemos encontrado una de estas joyas, la que procedemos a describir aquí como una nueva especie. Esta especie, pertenece al grupo de especies de *A. minutus* y se diferencia de los otros miembros del grupo por tener el dorso y el vientre uniformemente naranja, piel lisa y por el llamado de advertencia de los machos. Su tamaño pequeño, también diferencia a la nueva especie, de otros dendrobatidos coloridos presentes en el área, tales como: *Oophaga pumilio* y *O. vicentei*. Nosotros también proveemos aquí un análisis filogenético molecular de secuencias de ADN mitocondrial, que destaca las distancias genéticas entre las especies más cercanas. *Andinobates geminisae* **sp. nov**., se encuentra en los bosques lluviosos de la vertiente del Caribe, en el borde occidental de la distribución conocida de *A. minutus*, y representa la cuarta especie de este género en Panamá. Como otras especies del género, con distribuciones geográficas restringidas, *A. geminisae* es vulnerable a la pérdida del hábitat y a la recolección excesiva, por lo tanto este nuevo taxón requiere del inmediato desarrollo y aplicación de planes de conservación para preservar sus poblaciones.

Palabras claves: código de barras de ADN, *Andinobates geminisae* **sp. nov.**, conservación, distribución geográfica, filogenética molecular, Panamá, Ranas dardo-venenosas

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Introduction

Dart-poison frogs (Dendrobatidae) form the most diverse group of diurnal frogs in the Neotropics, comprised of diminutive animals that possess aposematic coloration and toxic skin secretions (Daly *et al.* 1987; Grant *et al.* 2006). Dendrobatids have experienced particularly high rate of primary species descriptions, especially in recent decades (Grant *et al.* 2006). Taxonomic problems among these frogs are often caused by color polymorphisms which are common within and between species (Silverstone 1975; Brown *et al.* 2011); thus molecular genetics, bioacoustics, ecological and behavioral data have been applied recently to solve many taxonomic problem within the family (Brown *et al.* 2011).

Within Dendrobatidae, frogs of the genus *Ranitomeya* sensu Grant *et al*. 2006, have been placed within different genera since the first species of the group was described over one hundred years ago (Boulenger 1884 "1883"; Bauer 1988; Brown *et al.* 2011). The great diversity within the genus is due to the rapid evolution of striking color variation over short geographic distances within species, as well as the convergent evolution of similar coloration between species, likely driven by Müllerian mimicry (Symula *et al.* 2001; Brown *et al.* 2011). After the revision by Grant *et al.* (2006) the genus *Ranitomeya* Bauer, 1986 was further divided into *Excidobates* Twomey & Brown 2008 and *Andinobates* Twomey, Brown, Amézquita, & Mejía-Vargas, 2011. Currently, 13 species are included in *Andinobates,* 3 in *Excidobates* and 16 in *Ranitomeya* (Amezquita *et al.* 2013; Frost 2013). All of these species are comprised of brightly colored and diminutive frogs.

Many species of *Ranitomeya* and *Andinobates* are restricted to small geographic areas [e.g., *A. claudiae* (Jungfer, Lötters & Jörgens 2000)*, A. cassidyhornae* Amézquita, Márquez, Medina, Mejía, Kahn, Súarez & Mazariegos 2013, and *R. summersi* Brown, Twomey, Pepper & Sánchez-Rodríguez 2008], while some others are widespread [e.g., *A. fulguritus* (Silverstone 1975), and *R. variabilis* (Shreve 1935)]. Species with small distributions are susceptible to population declines due to habitat fragmentation and illegal smuggling for the pet trade (Gorzula 1996; Pepper *et al.* 2007; Brown *et al.* 2011). Thus, these species require special conservation plans to guarantee their survival.

Three members of the *Andinobates minutus* species group (Brown *et al.* 2011) are currently known to occur in Panama: *A. claudiae*, *A. fulguritus* and *A. minutus* (Shreve 1935). Panamanian *Andinobates* are restricted to Caribbean lowland forests, and only *A. claudiae* is endemic to the region. Here we described a second endemic species of *Andinobates* restricted to a small area in the lowland forest on the Caribbean versant, Distrito de Donoso, Colón Province, Panama. Other dendrobatids that occur on the Caribbean lowland of Panama include *Oophaga pumilio* (Schmidt 1857) and *O. vicentei* (Jungfer, Weygoldt & Juraske 1996). These two species could be confused with a member of the *Andinobates* genus, as these genera live in similar habitats, are small, colorful and show a high color variation over all their distribution (Jungfer *et al.* 1996; Batista & Köhler 2008). Interestingly, despite the intensive literature about amphibians from Panama (Jaramillo *et al*. 2010), the new species had not been reported in any scientific publication (e.g. Silverstone 1975; Savage 1968; Daly et al. 1987; Brown et al. 2011).

The new species belongs to the genus *Andinobates* by having < 20 mm snout to vent length (SVL), bright coloration, ventrolateral stripe absent, head narrower than body; teeth absent; vocal slits present in males; first finger distinctly shorter than second; finger discs II and III weakly to moderately expanded; toe disc III and IV weakly expanded; toe V unexpanded; toe webbing absent; median lingual process absent. According molecular genetic evidence we assigned this new species to the *A. minutus* species group, *sensu* Brown *et al.* (2011).

Methods

Field work was carried out in the Distrito de Donoso, Colón Province, Panama, during biodiversity inventories between 2009 and 2010 (Fig. 1). The collecting area is part of the Isthmian-Atlantic moist forest (WWF *et al.* 2010), with a dominant vegetation of canopy trees (more than 20 m in height). The understory is substantially open, with some seedlings, palms, vines, and the leaf litter around 10 cm deep. The specimens reported in this paper were collected during opportunistic searches during the day.

Specimens were sacrificed with a euthanasia solution (T61), fixed in 10% formalin or in 94% ethanol, and finally stored in 70% ethanol. Prior to fixation of specimens, a fresh liver sample of the holotype was taken and stored in the tissue sample collection of the Círculo Herpetológico de Panamá (CH). Morphological nomenclature, measurements and diagnosis follow the methodology of Myers (1982) and Brown *et al.* (2006, 2011): SVL, femur length from vent to lateral surface of knee (FL), tibia length from heel to lateral surface of knee (TL), foot length from proximal edge of metatarsal tubercle to tip of toe IV (FoL), hand length from proximal edge of metacarpal tubercle to tip of longest finger (HaL), head length from most exposed corner of occipitum to tip of snout (HL), head width between tympana (HW), body width under axilla (BW), interorbital distance (IOD), horizontal tympanum diameter (TD), horizontal eye diameter (ED), length of finger I from proximal edge of median palmar tubercle to tip of finger disc (L1F), length of finger II from proximal edge of median palmar tubercle to tip of finger disc (L2F), width of third finger (3FW) at penultimate phalanx just anterior to disk; width of disk of third finger (3FD) at greatest width; width of third toe (3TW) at penultimate phalanx just anterior to disk; width of disk of third toe (3TD) at greatest width; width of fourth toe (4TW) at penultimate phalanx just anterior to disk; width of disk of fourth toe (4TD) at greatest width. Sex of specimens was determined by the presence of vocal slits in adult males. All measurements were made using digital calipers and a dissecting microscope. Measurements are given in millimeters, with mean ± standard deviation, and range in parenthesis.

Colors and color codes (the latter in parentheses) used in the color description of the holotype are those of Köhler (2012). Specimens were deposited in the Museo de Vertebrados de la Universidad de Panamá (MVUP), Museo Herpetológico de Chiriquí (MHCH) at the Universidad Autónoma de Chiriquí, and in the Círculo Herpetológico de Panamá (CH). Comparisons between similar species are based on original descriptions and on first-hand observation of museum and live specimens. Geographic coordinates and altitude were taken with a Garmin GPSmap 60CSx given in decimal degrees, recorded in WGS84 datum and rounded to the fourth decimal place. Elevations are rounded to the nearest ten meters. Figures 1 and 7 were created using ArcGIS 10 (ESRI 2009).

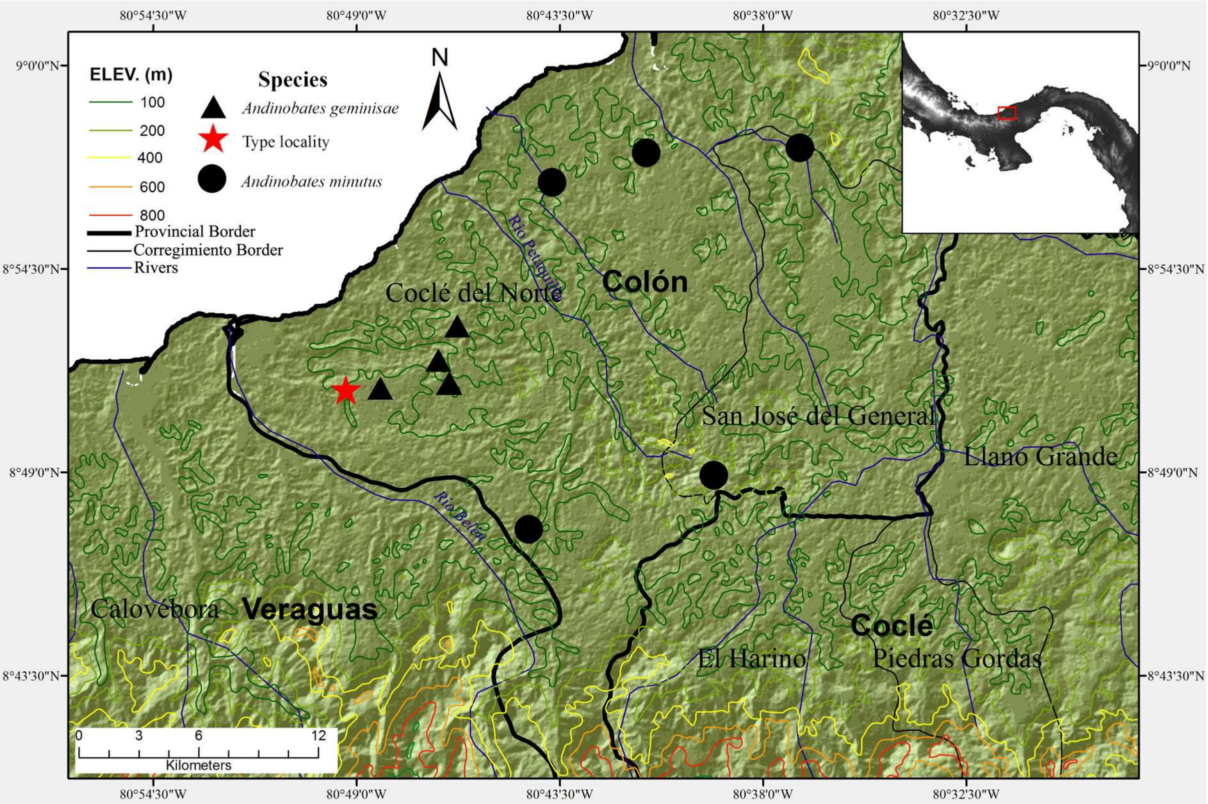

FIGURE 1. Distributional map of *Andinobates geminisae* and *A. minutus* at the Río Belén basin, Colón Province, Panama.

Molecular genetic methods. Genomic DNA was extracted from liver tissue of holotype MVUP 2428 using a BioSprint 96 (QIAGEN, Valencia, CA, USA) robotic extractor based on magnetic beads. We collected mitochondrial DNA (mtDNA) sequence data from two genes, the Folmer fragment of cytochrome oxidase I (COI-5'), also known as the animal Barcode of Life (Hebert *et al.* 2003), and a fragment of the 16S ribosomal RNA gene (16S). The COI marker was PCR-amplified using the primer pair dgHCO2198 (5'–TAA ACT TCA GGG TGA

CCA AAR AAY CA–3') and dgLCO1490 (5'–GGT CAA CAA ATC ATA AAG AYA TYG G–3') (Folmer *et al.* 1994; Meyer *et al.* 2005) and 0.25 μg/μL of bovine serum albumin. The 16S marker was amplified using 16SB-H (aka, 16Sbr-H) (5'–CCG GTC TGA ACT CAG ATC ACG T–3') and 16SA-L (aka, 16Sar-L) (5'–CGC CTG TTT ATC AAA AAC AT–3') (Kessing *et al.* 2004). For both markers, PCR contained 2.0 mM of Mg^{2+} and utilized an annealing temperature of 49ºC, with all other reaction conditions standard (Kessing *et al.* 2004). PCR products were cleaned using Exo I and SAP enzymes (Werle *et al.* 1994), with Sanger sequencing reactions run on ABI 3130 automated sequencers. Chromatograms were assembled and cleaned using Sequencher 5.0 (Gene Codes Corporation, Ann Arbor, Michigan, USA). Chromatograms along with cleaned DNA sequences and collection data for MVUP 2428 were posted at the Barcode of Life Data Systems (Ratnasingham & Hebert 2007) under Process ID number BSAMQ714-13. DNA sequence data were also deposited in GenBank under accession numbers KM212166 (COI) and KM212167 (16S).

To compare the mtDNA data from specimen MVUP 2428 with published molecular data (Table 1), we conducted *BLAST* searches (Altschul *et al.* 1990) in GenBank for sequences similar to the resulting COI and 16S gene fragments. The newly obtained data were combined with the first 100 BLAST hits and separate alignments were obtained for each gene using *Kalign* version 2.04 (Lassmann & Sonnhammer 2005) and reviewed and edited by eye in Mesquite version 2.74 (Maddison & Maddison 2009). Preliminary neighbour-joining (NJ) trees (Saitou & Nei 1987) for each gene suggested the DNA sequences were from the genus *Andinobates* (see Results). We therefore trimmed the dataset to include only sequences of *Dendrobates sensu lato* (the resulting alignments thus contained samples from the currently recognized genera *Andinobates*, *Ranitomeya*, *Oophaga* Bauer 1994, *Dendrobates* Wagler 1830, and *Excidobates* Twomey & Brown 2008) plus an outgroup sequence for each gene (*Phyllobates* Duméril & Bibron 1841, for 16S and *Ameerega* Bauer 1986 for COI, see Results), obtained from published sources (Clough & Summers 2000; Crawford *et al.* 2010; Faivovich *et al.* 2005; Grant *et al.* 2006; Santos & Cannatella 2011; Santos *et al.* 2003; Santos *et al.* 2009; Vences *et al.* 2000; Wang 2011; Wang & Shaffer 2008; Wollenberg *et al.* 2006). Since our research focused on the phylogenetic placement of specimen MVUP 2428, prior to phylogenetic analyses, we also removed redundant lineages from all genera except *Andinobates*. The resulting COI alignment contained 658 sites including 227 parsimony-informative characters, and the 16S alignment contained 559 sites and 121 parsimony-informative characters.

Prior to model-based phylogenetic inference, we used *jModeltest* version 0.1.1 (Posada 2008) and the corrected Akaike Information Criterion (AICc) (Akaike 1974) to select the best-fit model of molecular evolution for each gene separately, including outgroup sequences. The Tamura-Nei model with rate heterogeneity, TrN+G+I (Hasegawa *et al.* 1987; Tamura & Nei 1993; Yang 1994), was implemented for the COI dataset and the general time-reversible model with rate heterogeneity, GTR+G +G (Tavaré 1986), for the 16S dataset. Given the minimal overlap in taxon-sampling between the COI and 16S datasets, we inferred separate maximum likelihood (ML) molecular phylogenies (Felsenstein 1981) for each gene using the software *GARLI* version 2.0.1019 (Zwickl 2006) employing default search parameter values, including five replicate ML searches. Clade support was assessed by non-parametric bootstrapping (Felsenstein 1985) involving 200 re-sampling replicates with single ML searches per bootstrap dataset, and results summarized using the program *SumTrees* (Phylogenetic Tree Split Support Summarization) version 3.3.1. (Sukumaran & Holder 2010).

Genetic distance methods. Pairwise genetic distances among non-redundant samples of *Andinobates* were calculated separately for the COI (5 samples) and 16S (12 samples) mitochondrial gene fragments using *PAUP** version 4.0a124 for Macintosh OS X (Swofford 2000). Although *jModeltest* suggested more complex best-fit models including a rate heterogeneity parameter for these few samples, we assumed the HKY model for both genes as this model captures essential features of vertebrate mtDNA evolution while providing more conservative genetic distance estimates comparable with previous DNA barcoding studies (Collins *et al.* 2011).

Bioacoustic analyses. The advertisement call of the new species was recorded using a Digital Sony recorder (ICD-P630F) (with an internal microphone) placed roughly 1.0 m from the calling male MHCH 1571 and recorded using 16 kHz at 16-bit sampling size and the file was saved in MP3 format. The spectral and/or temporal parameters of two calls and one call interval were analyzed and the power spectra were calculated in Raven Pro 1.4 (Window: Blackman, DFT: 2048 samples, 3 dB filter bandwidth: 158 Hz; Charif *et al.* 2004). Terminology used in the advertisement call description follows Duellman & Trueb (1994) and Brown *et al.* (2011). Images of the oscillogram and the sound spectrogram were processed using Adobe Photoshop CS2; the call is available in AmphibiaWeb.org.

TABLE 1. DNA sequence data for the mitochondrial 16S ribosomal and cytochrome oxidase subunit I (COI) genes obtained from GenBank and used for molecular analyses of the phylogenetic position of *Andinobates geminisae* specimen MVUP 2428. See GenBank entries and original publications for further details on specimens, except the species name *Ranitomeya toraro* is based on Brown *et al.* (2011). n/a indicates data were not available or not applicable.

Genus	**species**	**voucher**	**locality**	**16S**	**COI**	**Publication**
Ameerega	*trivittata*	ICN 50437	Leticia, Amazonas, CO	n/a	DQ502929	Grant *et al.* 2006
Andinobates	*bombetes*	TNHC FS 4975	Finlandia, Barbas, Quindío, CO	EU342669	n/a	Santos *et al.* 2009
Andinobates	*claudiae*	USNM-FS 59979	Isla Popa, Bocas del Toro, PA	DQ502024	DQ502747	Grant *et al.* 2006
Andinobates	*claudiae*	USNM-FS 59980	Isla Popa, Bocas del Toro, PA	DQ283042	DQ502748	Grant *et al.* 2006
Andinobates	*claudiae*	USNM-FS 51785	Isla Colón, Bocas del Toro, PA	DQ502027	n/a	Grant *et al.* 2006
Andinobates	*claudiae*	KS 9	Isla Colón, Bocas del Toro, PA	HQ290983	n/a	Santos & Cannatella 2011
Andinobates	*claudiae*	KS 9	Isla Colón, Bocas del Toro, PA	EU342671	n/a	Santos *et al.* 2009
Andinobates	*fulguritus*	n/a	San Blas, PA	AF124116	n/a	Vences *et al.* 2000
Andinobates	*fulguritus*	MHNUC 340	Bahía Solano, Chocó, CO	DQ502106	DQ502817	Grant *et al.* 2006
Andinobates	*geminisae*	MVUP 2428	Río Caño, Coclé del Norte, Colón, Panama	KM212167	KM212166	This paper
Andinobates	*minutus*	KRL 0790	El Copé, Coclé, PA	DQ502168	DQ502870	Grant *et al.* 2006
Andinobates	*minutus*	AJC 1938	Brewster, Chepo, Panamá, PA	n/a	FJ766811	Crawford *et al*. 2010
Andinobates	aff. *minutus*	TNHC FS 4943	La Troje, Quibdó, Chocó, CO	EU342670	n/a	Santos *et al.* 2009
Andinobates	sp.	TNHC FS 4943	La Troje, Quibdó, Chocó, CO	HQ290989	n/a	Santos & Cannatella 2011
Andinobates	*opisthomelas*	GECOH 1212S	Isla del Sol, Guatape, Antioquia, CO	JQ936633	n/a	Marquez *et al.*, 2012
Andinobates	*virolinensis*	TNHC FS 4950	Virolín, Santander, CO	EU342667	n/a	Santos *et al.* 2009
Andinobates	*cassidyhornae*	GECOH 1534S	La Mesenia, Jardin, Antioquia, CO	JQ936638	n/a	Marquez *et al.*, 2012
Dendrobates	*auratus*	USNM 31318	Bocas del Toro, PA	AY843581	n/a	Faivovich *et al.* 2005
Dendrobates	*auratus*	CH 6605	El Copé, Coclé, PA	n/a	FJ766697	Crawford *et al*. 2010
Dendrobates	*azureus*	MRT 5089	Brazil	n/a	DQ502829	Grant *et al.* 2006
Dendrobates	*leucomelas*	isolate 645	captive bred	n/a	DQ502850	Grant *et al.* 2006
Dendrobates	*tinctorius*	MRT 5087	Brazil	n/a	DQ502830	Grant *et al.* 2006
Dendrobates	*truncatus*	TNHC 64417	Magdalena Valley, CO	EU342651	n/a	Santos *et al.* 2009

......continued on the next page

TABLE 1. (Continued)

Genus	species	voucher	locality	16S	COI	Publication
Dendrobates	*truncatus*	isolate 1151	captive bred	n/a	DQ502872	Grant *et al.* 2006
Excidobates	*captivus*	QCAZ 27443	Panguitza, Zamora Chinchipe, EC	EU342666	n/a	Santos *et al.* 2009
Oophaga	*arborea*	CWM 18636	Fortuna, Chiriquí, PA	DQ502036	DQ502763	Grant *et al.* 2006
Oophaga	*histrionica*	TNHC FS 4879	La Troje, Quibdó, Chocó, CO	EU342662	n/a	Santos *et al.* 2009
Oophaga	*histrionica*	IW 0845	n/a	n/a	HQ841132	Wang 2011
Oophaga	*lehmanni*	CWM 19050	Valle del Cauca, CO	DQ502034	DQ502761	Grant *et al.* 2006
Oophaga	*pumilio*	n/a	pet trade (Bastimentos morph)	DQ768799	n/a	Wollenberg *et al.* 2006
Oophaga	*pumilio*	TNHC FS 4814	Isla Colón, Bocas del Toro, PA	EU342663	n/a	Santos *et al.* 2009
Oophaga	*pumilio*	OMNH 33297	Isla de Diamante, Río San Juan, NI	n/a	DQ502784	Grant *et al.* 2006
Oophaga	*pumilio*	isolate Cn2	Isla Colón, Bocas del Toro, PA	n/a	EU934662	Wang & Shaffer 2008
Oophaga	*sylvatica*	LSUMZ 14730	Santo Domingo, EC	n/a	DQ502781	Grant *et al.* 2006
Oophaga	*vicentei*	KRL 0789	El Copé, Coclé, PA	DQ502167	DQ502869	Grant *et al.* 2006
Phyllobates	*bicolor*	TNHC 62488	n/a	AY364580	n/a	Santos *et al.* 2003
Ranitomeya	*ventrimaculata*	QCAZ 16561	Yasuni, Francisco de Orellana, EC	AY364566	n/a	Santos *et al.* 2009
Ranitomeya	*fantastica*	n/a	captive bred (Tarapoto, PE)	AF128622	n/a	Clough & Summers 2000
Ranitomeya	*imitator*	KS 13	Tarapoto, San Martin, PE	EU342672	n/a	Santos *et al.* 2009
Ranitomeya	*sirensis*	JCS '40A'	pet trade (PE)	HQ290986	n/a	Santos & Cannatella 2011
Ranitomeya	*reticulata*	TNHC 61143	PE	AY364567	n/a	Santos *et al.* 2003
Ranitomeya	*reticulata*	MJH 3754	Alpahuayo, Loreto, PE	n/a	DQ502827	Grant *et al.* 2006
Ranitomeya	*vanzolinii*	LSUMZ 13697	Porto Walter, Acre, BR	EU342674	n/a	Santos *et al.* 2009
Ranitomeya	*vanzolinii*	OMNH 36036	Porto Walter, Acre, BR	n/a	DQ502908	Grant *et al.* 2006
Ranitomeya	*variabilis*	KS 12	Tarapoto, San Martin, PE	EU342679	n/a	Santos *et al.* 2009
Ranitomeya	*ventrimaculata*	LSUMZ 15394	Rio Ituxi, Amazonas, BR	EU342675	n/a	Santos *et al.* 2009
Ranitomeya	*ventrimaculata*	LSUMZ 13770	Porto Walter, Acre, BR	EU342685	n/a	Santos *et al.* 2009
Ranitomeya	*toraro*	OMNH 37440	40 km S of Manaus, Amazonas, BR	n/a	DQ502905	Grant *et al.* 2006
Ranitomeya	*ventrimaculata*	OMNH 34091	Cuyabeno, Sucumbios, EC	n/a	DQ502791	Grant *et al.* 2006

Results

Andinobates geminisae sp. nov.

Fig. 2

Holotype. MVUP 2428 (CH 9386: original field number of Círculo Herpetológico de Panamá), an adult male (Fig. 2 A–B) collected in the headwaters of the Río Caño, Coclé del Norte, Distrito de Donoso, Colón Province, Panama, on February 21, 2011 at 09:36 AM at GPS coordinates 8.8536° N, 80.8214° W and 89 m elev. (Fig. 1) Voucher collected by Samuel Valdés and Carlos De La Cruz.

Paratypes. All specimens were collected between the Río Coclé del Norte and the Río Belén basin (http://www.hidromet.com.pa/cuencas.php; retrieved March 4, 2013), Coclé del Norte, Distrito de Donoso, Colón Province, Panama, and found in the leaf litter. Specimens MHCH 2073–MHCH 2076 were collected by M. Ponce at the Manakin trail, headwaters of the Río Caño, Coclé del Norte, Distrito de Donoso, Colón Province, Panama (8.8546° N, 80.8061° W; 82 m elev.) 7.5 km east from the Río Belén outfall on March 16, 2010. Specimens MHCH 1570–MHCH 1576 were collected from March 15 to 16, 2010 by A. Batista, at the headwaters of the Río Chiquero, Coclé del Norte, Distrito de Donoso, Colón Province, Panama (8.8567° N, 80.7752° W; from 100 to 136 m elev.). MHCH 2037 was collected 07 July 2009 by M. Ponce and A. Batista, at Palmilla (Fig. 3), headwaters or the Río Chiquero, Coclé del Norte, Distrito de Donoso, Colón Province, Panama (8.8826° N, 80.7714° W; 50 m elev.).

Etymology. The specific epithet of this beautiful new species is dedicated to Geminis Vargas, the beloved wife of Marcos Ponce, for her unconditional support of his studies of Panamanian herpetology.

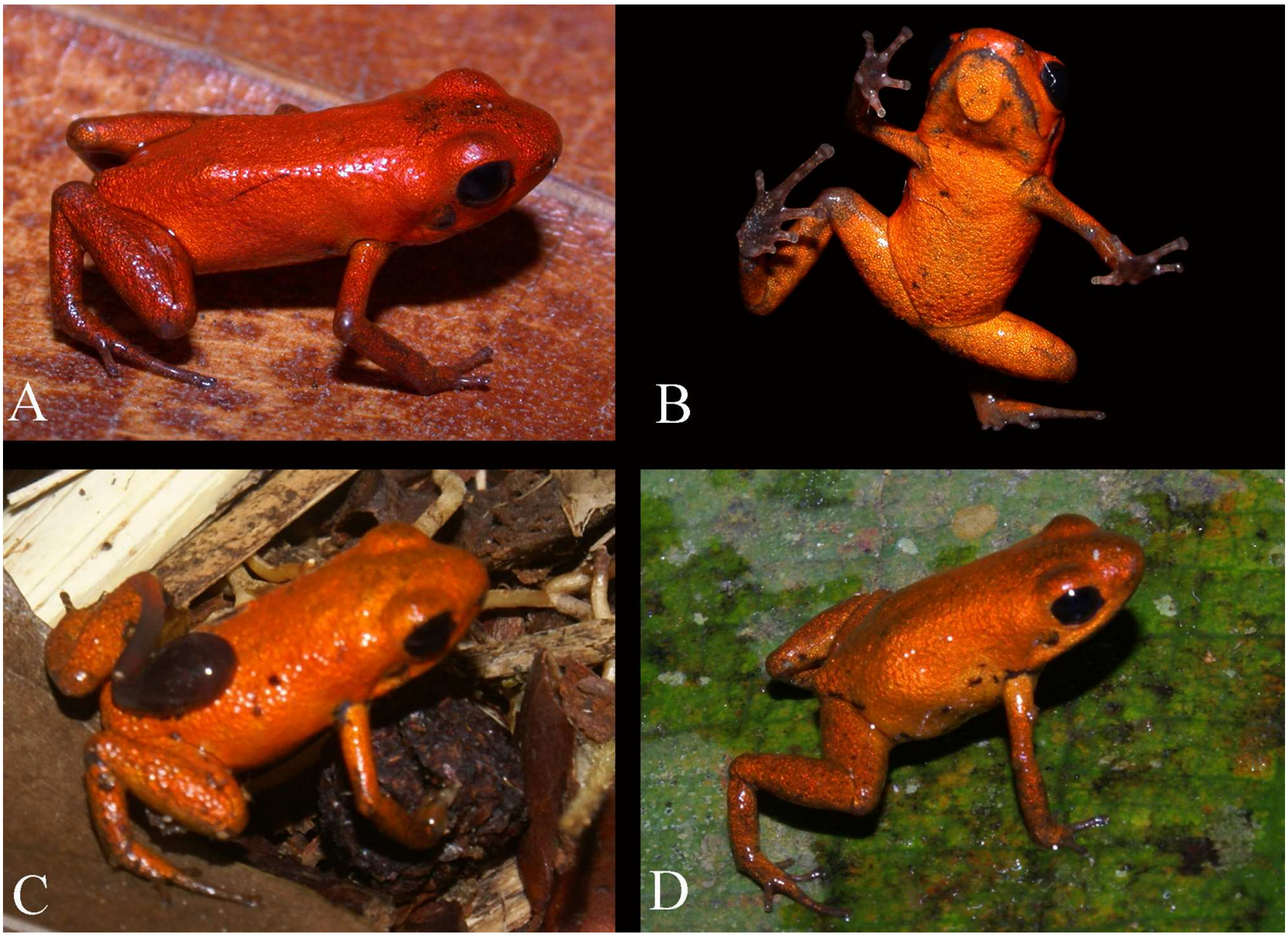

FIGURE 2. *Andinobates geminisae* **sp. nov.** A–B) Holotype in life (MVUP 2428); C) Individual of unknown sex photographed in the field carrying a tadpole (8.8567° N, 80.7752° W; 136 m elev.); D) Specimen from Palmilla, Distrito de Donoso, Colón Province, Panama (8.8825° N, 80.7714° W; 50 m elev.).

Diagnosis. *Andinobates geminisae* is characterized by the following combination of characters: (1) Tiny size, adults SVL 11.63–13.63 (Table 2); (2) Dorsal coloration conspicuous; (3) dorsum and venter uniformly orange, without stripes; (4) limbs and venter brightly colored, same as dorsum; (5) Dorsal skin smooth; (6) finger I reduced and shorter than finger II; (7) finger discs III–IV moderately expanded, disc of finger III 1.11–2.38 times wider than finger width; (8) thenar tubercle slightly elongate and low; (9) toe discs II–IV weakly expanded, disc on toe III 1.03–1.71 times wider than finger width; (10) no fringes or webbing on fingers or toes; (11) outer metatarsal tubercle small and slightly pointed, inner metatarsal tubercle rounded. (12), maxillary and premaxillary teeth absent; (13) tympanum oval, posterodorsal margin hidden.

TABLE 2. Measurements and morphological proportions for eleven adult specimens of *Andinobates geminisae* (see Methods for abbreviations).

Trait	Measurements (mm)	Trait	Proportion (%)
SVL	12.68 ± 0.87 (11.63–13.63)	IND/SVL	0.14 ± 0.01 (0.13–0.18)
HW	4.31 ± 0.20 (4.05–4.64)	TL/SVL	0.45 ± 0.03 (0.41–0.52)
HL	4.01 ± 0.18 (3.75–4.40)	HL/SVL	0.32 ± 0.02 (0.29–0.35)
IND	1.81 ± 0.21 (1.45–2.15)	HW/SVL	0.34 ± 0.02 (0.31–0.38)
IOD	1.66 ± 0.12 (1.44–1.79)	HL/HW	0.93 ± 0.04 (0.87–1.00)
ED	1.85 ± 0.15 (1.55–2.05)	ED/SVL	0.15 ± 0.01 (0.12–0.17)
SL	1.15 ± 0.23 (0.85–1.74)	TD/SVL	0.05 ± 0.01 (0.04–0.07)
TD	0.68 ± 0.09 (0.51–0.88)	SL/SVL	0.09 ± 0.01 (0.07–0.13)
TL	5.73 ± 0.36 (4.73–6.09)	HaL/SVL	0.21 ± 0.02 (0.16–0.26)
FoL	4.65 ± 0.41 (3.83–5.35)	FAL/SVL	0.26 ± 0.02 (0.23–0.30)
FAL	3.26 ± 0.22 (2.85–3.54)	FoL/SVL	0.37 ± 0.03 (0.34–0.46)
HaL	2.71 ± 0.24 (2.23–3.08)	4TD/4TW	1.37 ± 0.23 (1.11–2.00)
3FW	0.31 ± 0.06 (0.21–0.38)	3TD/3TW	1.30 ± 0.21 (1.03–1.71)
3FD	0.47 ± 0.05 (0.39–0.55)	3FD/3FW	1.57 ± 0.37 (1.11–2.38)
3TW	0.31 ± 0.05 (0.22–0.39)		
3TD	0.40 ± 0.05 (0.34–0.48)		
4TW	0.35 ± 0.04 (0.28–0.40)		
4TD	0.48 ± 0.07 (0.38–0.60)		
BW	4.61 ± 0.47 (4.11–5.81)		

Similar species. *Andinobates geminisae* **sp. nov.** can be distinguished from other species of the genus (see table 3; Fig. 5) by its predominantly uniform orange coloration through its body, smooth dorsum, and a distinctive male advertisement call (see below: vocalization description). *Andinobates geminisae* is similar in appearance to *A. opisthomelas*, but *A. opisthomelas* has granular skin texture (smooth in *A. geminisae*), limbs, throat and belly blackish (uniform orange in *A. geminisae*). *Minyobates steyermarki* (Rivero 1971) also has reddish coloration, but has the first finger as long as the second or greater (*A. geminisae* finger I shorter than finger II). *Andinobates minutus* is closely related to *A. geminisae* (Fig. 5), which reaches the western limit of the distribution of *A. minutus*, but despite their very close geographical distribution (Fig. 1, 6), the coloration of *A. geminisae* contrasts with that of *A. minutus* (see Fig. 2, 5B). Furthermore, *Andinobates minutus*, *A. claudiae* and *A. fulguritus* have higher dominant frequency in their advertisement calls than *A. geminisae* (4.8–6.4 in the three first species, versus 4.4–4.5 kHz in *A. geminisae*; see Brown *et al.* 2011; Fig. 7); *Andinobates minutus, A. claudiae* and *A. fulguritus* also have fewer pulses per note than *A. geminisae* (28–71 in the three first species, versus 87–88 in *A. geminisae*; see Brown *et al.* 2011). *Oophaga pumilio* and *O. vicentei* are larger than *A. geminisae* (11.63–13.63 mm SVL); *O. pumilio* is usually >17 mm SVL (Batista & Köhler 2008; Köhler 2011) and *O. vicentei* 19–21 mm SVL (Jungfer et al. 1996, Lötters et al, 2007). The dorsal color pattern of *O. pumilio* is usually red with blue legs, however it present a great color variation, from green, yellow, blue or red, uniform or with dark blotches (8 C–D). Two populations of uniformly red *O. pumilio* are known in Panama, those from Cerro Tebata (Fig. 8 E–F) and from Solarte Island.

However, these populations are larger (> 17 mm SVL; Batista & Köhler 2008) than *A. geminisae*. *Oophaga vicentei* is mainly a tree-dwelling species (*A. geminisae* is terrestrial). The dorsal color of *O. vicentei* near the type locality of *A. geminisae* varies from brown, reddish, yellow or blue, all with dark blotches or a reticulated pattern, not uniform, and the venter is sky blue (Fig. 8 A–B). The call duration in *O. pumilio* is 0.04–0.07 s (Myers & Daly 1976), and the call duration of *O. vicentei* is 0.14–0.20 s (MHCH 2983, a male recorded at Donoso, Colón, Panama, 8.82246° N, -80.70504° W, 294 m elev., 22 July 2011, 07:30, 23 °C), while the call duration of *A. geminisae* is much longer (1.60 s).

FIGURE 3. Habitat (forest to the right) of *Andinobates geminisae* sp. nov. at Palmilla, Distrito de Donoso, Colón Province, Panama (8.8826° N, 80.7714° W; 50 m. elev.) Photo taken on July 07, 2009).

Measurements of the holotype in mm. (See Table 2) SVL 13.63, HW 4.19, HL 4.21, IND 1.84, IOD 1.49, ED 1.80, EYE-NAR 1.03, TYMP 0.77, TL 5.86, FOOT 5.08, FAL 3.32, HaL 2.86, 3FW 0.38, 3FD 0.48, 3TW 0.38, 3TD 0.45, 4TW 0.38, 4TD 0.50, BW 4.40, LIIF 1.25 LIF 0.85, IND/SVL 0.13, TL/SVL 0.43, HL/SVL 0.31, HW/SVL 0.31, HL/HW 1.00, ED/SVL 0.13, TD/SVL 0.06, SL/SVL 0.08, HAL/SVL 0.21, FL/SVL 0.37, FAL/SVL0.24, BW/SVL 0.32, 4TD/4TW 1.32, 3TD/3TW 1.18, 3FD/3FW 1.26, LIIF / LIF 1.47.

Description of the holotype (Fig. 2A–B). An adult male with slender body; smooth dorsal skin, venter coarsely areolate; eye length longer than snout; ratio SL/ED 57%; tympanum of moderate size, ratio TD/ED 43%; tympanum distinguishable, annulus tympanicus oval and partially visible through skin, posterodorsal margin hidden, positioned closely behind orbit and upper jaw; head equally wide as long (ratio HL/HW: 1.00); head width 31% of SVL; snout nearly rounded from above and rounded in profile; nares situated near tip of snout and laterally directed, visible ventrally and in front view, barely visible dorsally; internares distance 1.84 mm, 44% of head width; canthus rostralis rounded, loreal region flat; paired vocal slits present and each is located adjacent to body of the mandible between the tongue and the jaw articulation; tongue elongate, median lingual process absent; hands moderate in size, 21% of SVL; relative lengths of adpressed fingers I<II<IV<III; finger IV slightly longer than II, finger II reaching the disk on finger IV when adpressed; finger III disk 1.26 times wider than distal end of adjacent phalanx; palmar tubercle rounded, slightly elevated, and slightly larger than thenar tubercle; thenar tubercle low and elongate; subarticular tubercles rounded and globular; no supernumerary tubercles; palmar and plantar accessory tubercles absent; no nuptial pads; no fringes on fingers; hind limbs of moderate lengths, TL 43% of SVL; relative lengths of adpressed toes I<II<III<V<IV; when adpressed, tip of toe I reaches to the proximal tubercle of

toe II; disc of toe IV weakly expanded, 1.32 times wider than distal end of adjacent phalanx; no fringes on toes; one to three nonprotuberant subarticular tubercles present (one on toes I and II, two on toes III and V, three on toes IV, subarticular tubercles of toe IV weakly defined); inner metatarsal tubercle elongated; outer metatarsal tubercles small rounded and conical and smaller than inner; plantar tubercles small and rounded; tarsal ridge absent; toe disks weakly expanded; hands and feet without webbing.

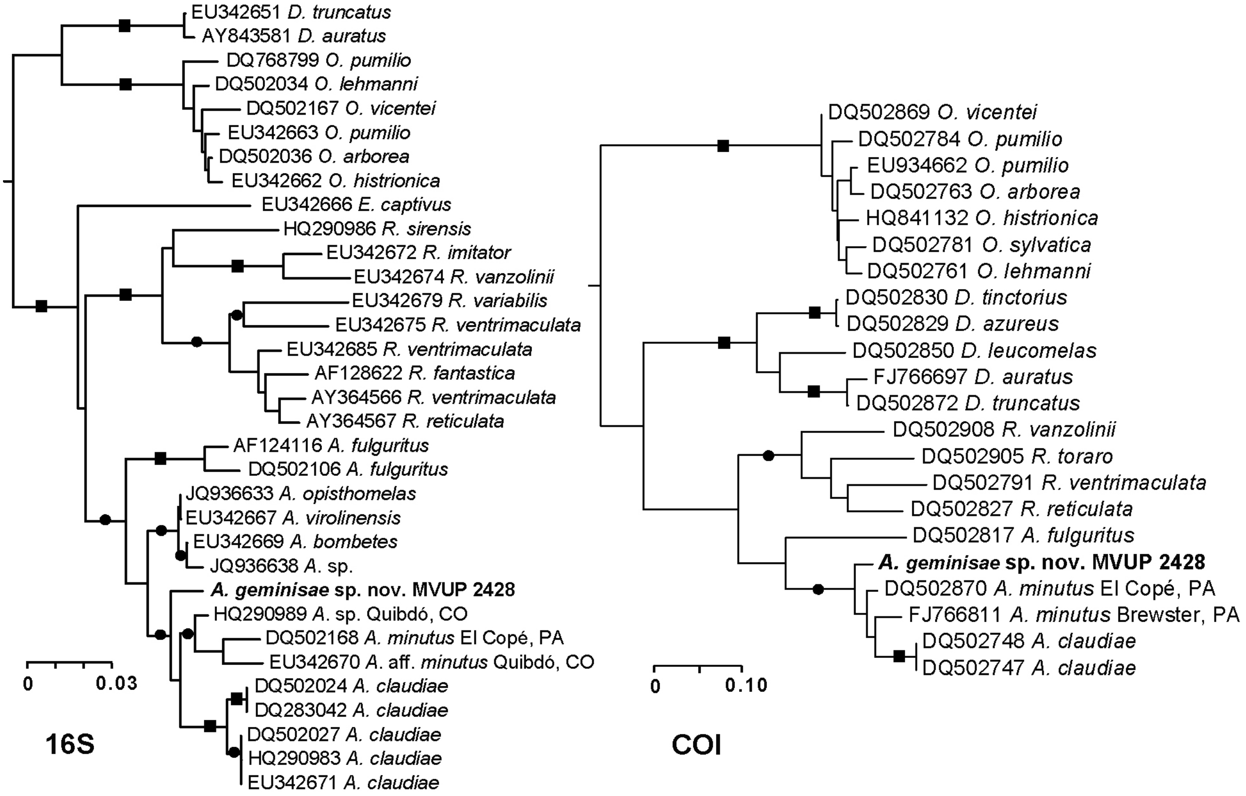

FIGURE 4. Maximum likelihood (ML) trees inferred independently for the 16S ribosomal (left) and COI protein-coding (right) mitochondrial genes. Scale bars indicate model-based patristic distances estimated from ML analysis (see text for details). Bootstrap support ≥ 90% is indicated by squares on the corresponding branch, with support ≥70% but <90% indicated by circles. Not shown are outgroups *Phyllobates bicolor* (AY364580) for 16S and *Ameerega trivittata* (DQ502929) for COI. Specimen details are provided in Table 1.

Variation. Eleven adult specimens collected including the holotype and one juvenile; SVL: 12.68 ± 0.87 (11.63–13.63); males: 12.70 ± 0.74 (11.63–13.63; n: 8); females: 13.25 ± 0.05 (13.20–13.29; n: 3); one juvenile: 10.80. Females slightly bigger than males.

Color in life (Fig. 2). Coloration of most specimens fit with the color description of the holotypes, as follows: dorsum uniform Chrome Orange (74), interorbital area slightly suffused with dark pigmentation; lower tympanum area, junctions of elbow and knee Fawn Color (258); fingers and toes Glaucous (272), suffused with Spectrum Orange (9); plantar and palmar areas Prout's Brown (47); subarticular tubercles Pale Neutral Gray (296); junction between axila and body Fawn Color (258); throat and belly Spectrum Orange (9); lips of lower jaw Chestnut (30); iris black.

Color in preservative. Dorsum posterior to supraorbital region and anterior to the lumbar region Light Neutral Gray (298); front of head and limbs Russet (44); ventral surfaces Hazel (26) with the glandular spots Lavender (202). In preservative specimens MHCH 1991 and MHCH 1993 have little dark spots on throat and belly. Specimens MHCH 1991, MHCH 1571, MHCH 1573 and MHCH 1575 have dark pigmentation on throat.

Tadpole. Known only from photo (Fig. 2C, not collected). One individual (undetermined sex) carried on back one dark tadpole, observed on 16 March 2010 at 13:50 hrs.

Vocalization. We recorded one male on 15 March 2010 in the Distrito de Donoso, Colón Province, Panama (8.8546° N, 80.8061° W) at 11:35 hrs with air temperature of 25.9ºC. The calls consisted of a buzz-like call similar to other members of the *A. minutus* species group (Brown *et al.* 2011). Two calls were recorded (Fig. 7); details of the first call as follows (with information on the second call provided in parentheses): calls consisted of 87 (88) pulses, call duration of 1.60 s (1.52 s), with an interval between the two calls of 59.4 s, with a total call rate of approximately two calls per minute. The rise time was 5.64% (8.14%) of the total call duration. The peak frequency band ranged from 4.12 to 4.74 kHz (4.03–4.80 kHz), with the dominant frequency at 4.52 kHz (4.4 kHz). Each pulse length was 7.8 ms (7.7 ms), and the pulse interval on average was 10 ms (9.5 ms). The call began with one or two pulses spaced by a pause of 42.0 ms (52.0 ms) from subsequent pulses, after which the pulse intervals remained relatively constant at 8.40 ± 1.6 ms. Before the final pulse there was an interval of silence lasting 152.0 ms (88.0 ms) followed by a click that terminated the call.

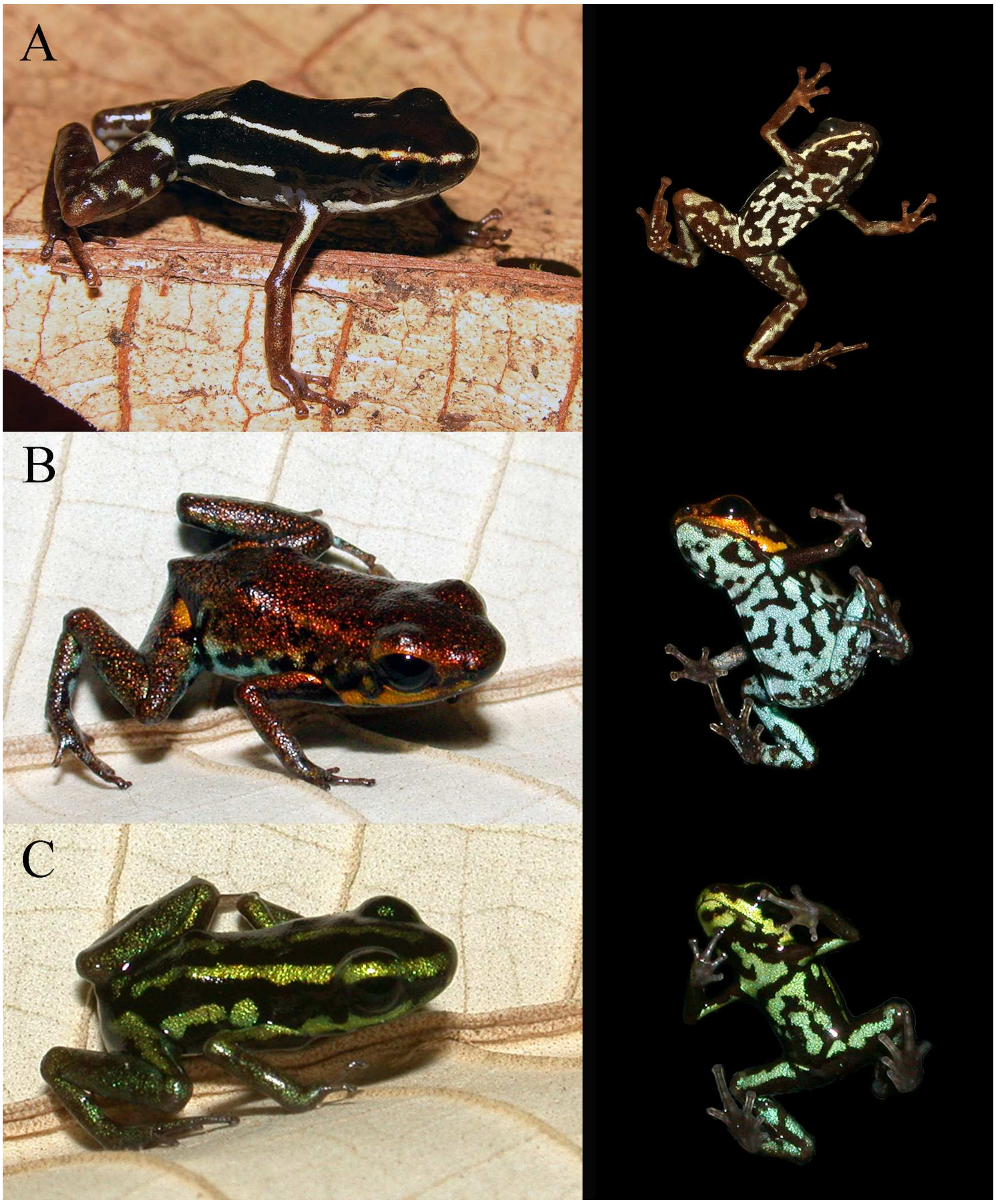

FIGURE 5. Photographic example of the other three species of *Andinobates* found in Panama, with dorsal (left) and ventral (right) views. A) *A. claudiae* (CH 5983 Isla Bastimento, Bocas del Toro Province, Panama); B) *A. minutus* (CH 6050 Cerro Brewster, Panamá Province, Panama); C) *A. fulguritus* (CH 6049 Cerro Brewster, Panamá Province, Panama).

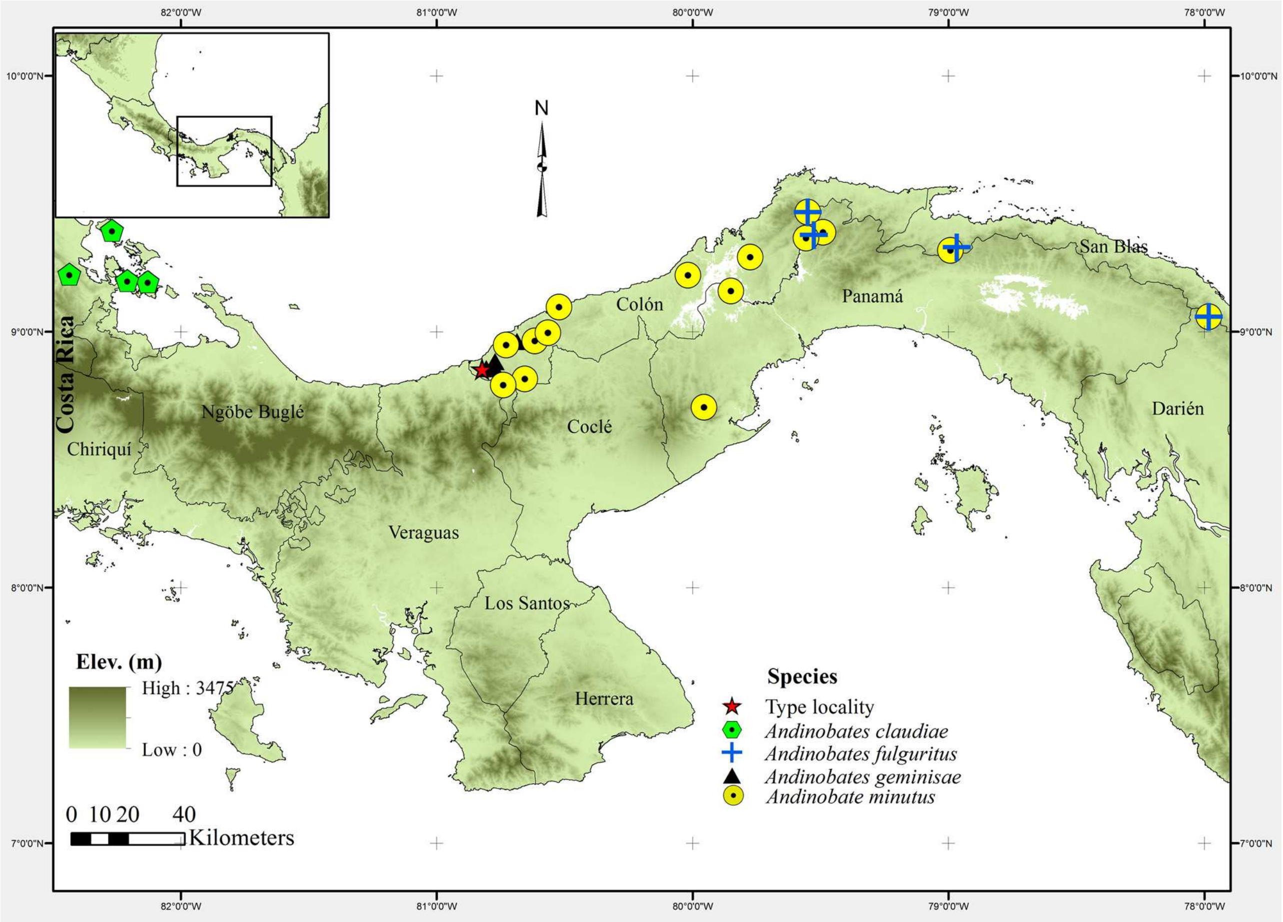

FIGURE 6. Map of the Republic of Panama showing the distribution of species of *Andinobates*.

Distribution and natural history (Figs. 1, 3 & 6). *Andinobates geminisae* is known only from the Río Belén basin (http://www.hidromet.com.pa/cuencas.php; retrieved March 4, 2013; Fig. 1 and 6). Most *A. geminisae* were found in nearly pristine forest with occasional perturbations by people looking to establish settlements (Fig. 3). All individuals were found on ridges of small hills, suggesting that *A. geminisae* prefers upland drier areas over flat, moister low-lying areas. Most frogs were found exposed on top of the leaf litter, nearby tree roots or near the entrance of cavities under rocks or logs. The bright coloration of *A. geminisae* contrasts sharply with the forest floor, allowing the investigator to see the frogs easily. In an area of 40 m^2 (8.8567° N, 80.7752° W; 136 m. elev.) we observed approximately 15 individuals in 45 minutes. Calling males of *A. geminisae* were heard from 08:00 to 14:00, suggesting that this species calls actively throughout the day during the breeding season. Potential areas for reproduction used by this species may include phytotelmata found at the type locality, e.g., water-filled tree hollows plant axial water (e.g. in *Heliconia* spp.), and dry leaves filled with rain water, though no tadpoles were found. Dendrobatid species sympatric with *A. geminisae* included *Colostethus pratti*, *Dendrobates auratus*, *Oophaga vicentei*, *Phyllobates lugubris,* and *Silverstoneia flotator*.

Conservation status. Despite the fact that most of the terrestrial habitat in the Donoso region is comprised of primary forest, during recent decades the area has been impacted by deforestation for pasture and agriculture land by non-indigenous colonists (see Fig. 3), as well as migration of indigenous peoples from the Comarca Ngöbe Buglé to establish new settlements. More recently, multinational mining companies have begun surveying the region with intentions to build a copper mine. Therefore, the development and implementation of conservation action plans should be a high priority to assure the survival of *A. geminisae*.

Genetic distance results (Tables 4–5, Fig. 4). Assuming HKY distances at the COI marker, *A. geminisae* (MVUP 2428) showed 5.6% divergence (Table 4) from *A. minutus* (DQ502870, KRL 0790, Parque Nacional General de División Omar Torrijos Herrera, Coclé Province, Panama), 8.5% from a second *A. minutus* sample

(FJ766811, CH 9887, Cerro Brewster, Distrito de Panamá, Panama Province, Panama), 8.6% divergence from *A. claudiae* (DQ502747, USNM-FS 59979, Isla Popa, Bocas del Toro Province, Panama) and 18.7% from *A. fulguritus* (DQ502817, MHNUC 340, Bahía Solano, Chocó, Colombia). (Adding a G-rate heterogeneity parameter with values estimated by ML, these distances would be 8.4%, 15.5%, 15.9% and 73.5%, respectively.) For the 16S data, *A. geminisae* (MVUP 2428) was most similar to *A.* sp. (HQ290989, TNHC FS 4943, Quibdó, Chocó, Colombia) and *A. minutus* (DQ502168, KRL 0790, Parque Nacional General de División Omar Torrijos Herrera, Coclé Province, Panama) with 2.5% HKY distances from either sample. Relative to other Colombian congeners, the new species showed 2.7% divergence from the two genetically identical *A. opisthomelas* (JQ936633) and *A. virolinensis* samples (EU342667), and 3.1% divergence to *A. bombetes* (EU342669). At 16S, *A. geminisae* showed 3.4% divergence from *A. claudiae* (DQ283042 and DQ502027), and 3.6% divergence from Colombian samples *A. cassidyhornae* (JQ936638, GECOH 1534S, Antioquia, Colombia) and *A.* aff. *minutus* (EU342670, TNHC FS 4943, Quibdó, Chocó, Colombia). Finally, *A. fulguritus* showed 4.9% (AF124116) to 5.5% (DQ502106) divergence from the new species.

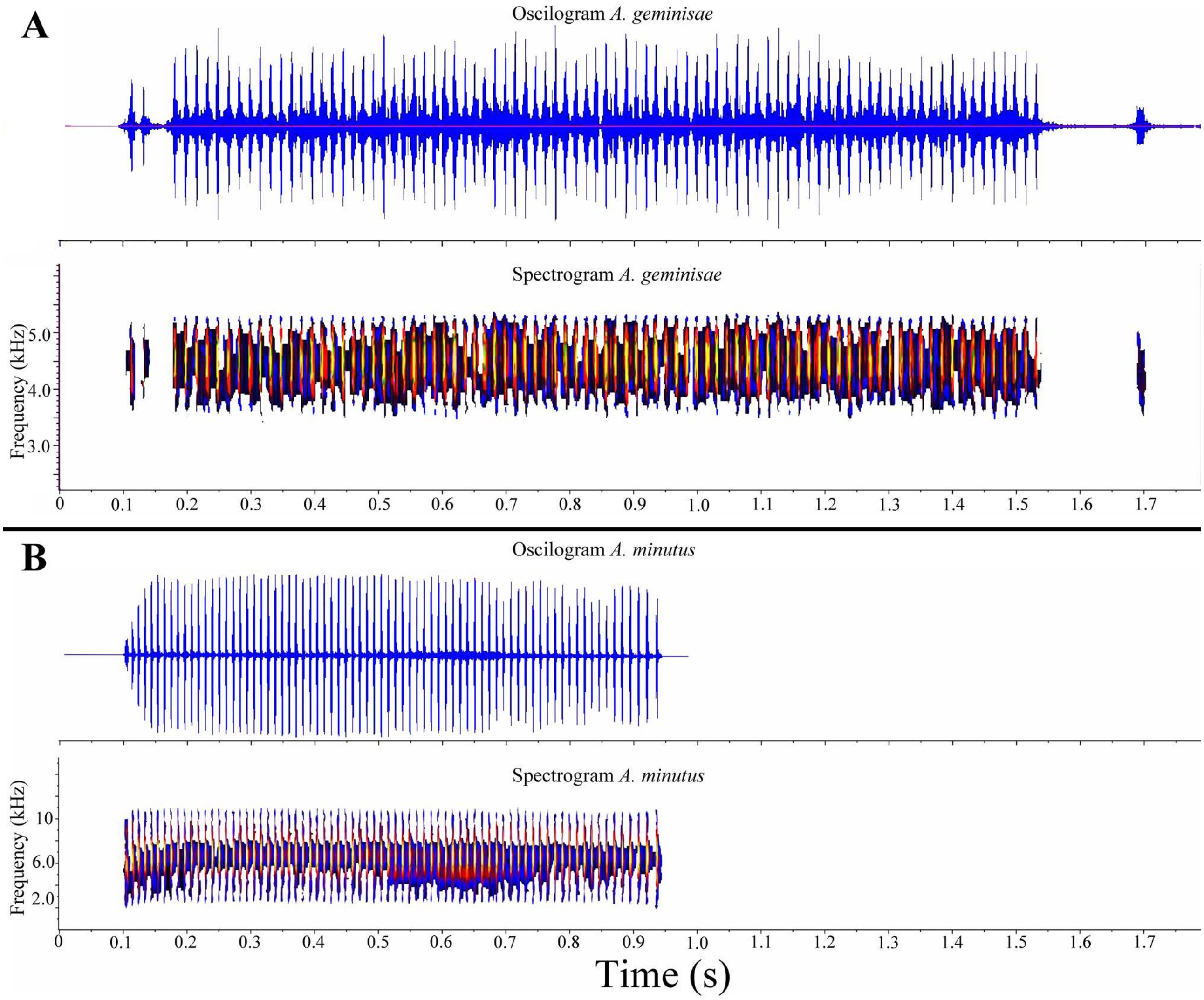

FIGURE 7. Spectrogram (above) and oscillogram (below) of the advertisement call of (A) *Andinobates geminisae* **sp. nov.** (note the clicks at the beginning and end of call) and (B) *A. minutus* (for comparison) recorded at the Manakin trail in the Distrito de Donoso, Colón Province, Panama (8.8546° N, 80.8061° W).

TABLE 3. Main diagnostic characters and character states for members of the *Andinobates minutus* species group. Data for *A. claudiae*, *A. fulguritus*, and *A. minutus* were taken from original descriptions.

Traits	***A. claudiae***	***A. fulguritus***	***A. minutus***	***A. geminisae***
SVL males	13.1–13.4	13.50 –16.50	13.00 –15.00	11.63–13.63
SVL females	13.2	14.00 –16.50	12.00 –15.50	13.20–13.29
all	13.1 –13.4	13.50–16.50	12.00–15.50	10.80–13.63
dorsal coloration	black or brown with gold or white stripes	black with gold, yellow, or yellow-green	black or brown with orange, gold or yellow stripes	uniform Orange
ventral coloration	black with white speckled	black, marbled with green.	blue or white spots or marbling	uniform Orange
Dorsal pattern	dorsal and dorsolateral stripes	lateral and median stripes	complete dorsolateral and incomplete lateral stripes	uniform
Ventral pattern	speckled	spots or marbling	spots or marbling	uniform, rarely with black patches
Call Description	buzz	short buzz	tonal buzz	tonal buzz
Dominant frequency	5.80–6.10	4.80–5.10	5.4–6.40	4.40–4.50
Pulse per note	55–65	28–52	20–71	87–88
Note length	0.19–0.34	0.85–1.03	0.20–1.10	1.52–1.60
Inter-note interval	18–24	3.1–5.9	1.20–3.20	59.4

TABLE 4. Estimates of evolutionary divergence among 16S mtDNA gene sequences of the *Andinobates* spp. used in the phylogenetic analysis. Numbers represent the uncorrected p-distances.

		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**
1	CH 9386 *A. geminisae*															
2	DQ502168 *A. minutus* isolate 1149	0.02														
3	HQ290989 *A.* sp. Quibdó	0.02	0.03													
4	JQ936633 *A. opisthomelas* isolate GECOH 1212S	0.03	0.04	0.03												
5	EU342670 *A.* aff *minutus* JCS 2008 TNHC FS 4943	0.03	0.02	0.01	0.04											
6	HQ290983 *A. claudiae*	0.03	0.04	0.02	0.03	0.03										
7	DQ502027 *A. claudiae* isolate330	0.03	0.04	0.02	0.03	0.03	0.00									
8	DQ502024 *A. claudiae* isolate323	0.03	0.04	0.03	0.03	0.04	0.01	0.01								
9	DQ283042 *A. claudiae* USNM FS59980	0.03	0.04	0.03	0.03	0.04	0.01	0.01	0.00							
10	EU342667 *A. virolinensis* TNHC FS 4950	0.03	0.04	0.03	0.00	0.04	0.03	0.03	0.03	0.03						
11	EU342671 *A. claudiae* KS9	0.03	0.04	0.02	0.03	0.03	0.00	0.00	0.01	0.01	0.03					
12	EU342669 *A. bombetes* TNHC FS 4975	0.03	0.04	0.03	0.00	0.04	0.03	0.03	0.03	0.03	0.00	0.03				
13	JQ936638 *A.* sp. RM 2012 isolate GECOH 1534S	0.03	0.04	0.03	0.01	0.05	0.03	0.03	0.04	0.04	0.01	0.03	0.01			
14	AF124116 *A. fulguritus*	0.05	0.06	0.05	0.04	0.06	0.05	0.05	0.06	0.06	0.04	0.05	0.04	0.04		
15	DQ502106 *A. fulguritus* isolate 499	0.05	0.06	0.05	0.04	0.06	0.06	0.06	0.06	0.06	0.04	0.06	0.05	0.05	0.02	

TABLE 5. Estimates of evolutionary divergence among COI mtDNA gene sequences of the *Andinobates* spp. used in the phylogenetic analysis. Numbers represent the uncorrected p-distances.

		1	2	3	4	5	6
1	CH 9386 *A. geminisae*						
2	DQ502870 *A. minutus* isolate 1149	0.05					
3	FJ766811 *A. minutus* AJC 1938	0.08	0.05				
4	DQ502748 *A. claudiae* isolate 324	0.08	0.07	0.08			
5	DQ502747 *A. claudiae* isolate 323	0.08	0.07	0.08	0.00		
6	DQ502817 *A. fulguritus* isolate 499	0.16	0.15	0.16	0.17	0.17	

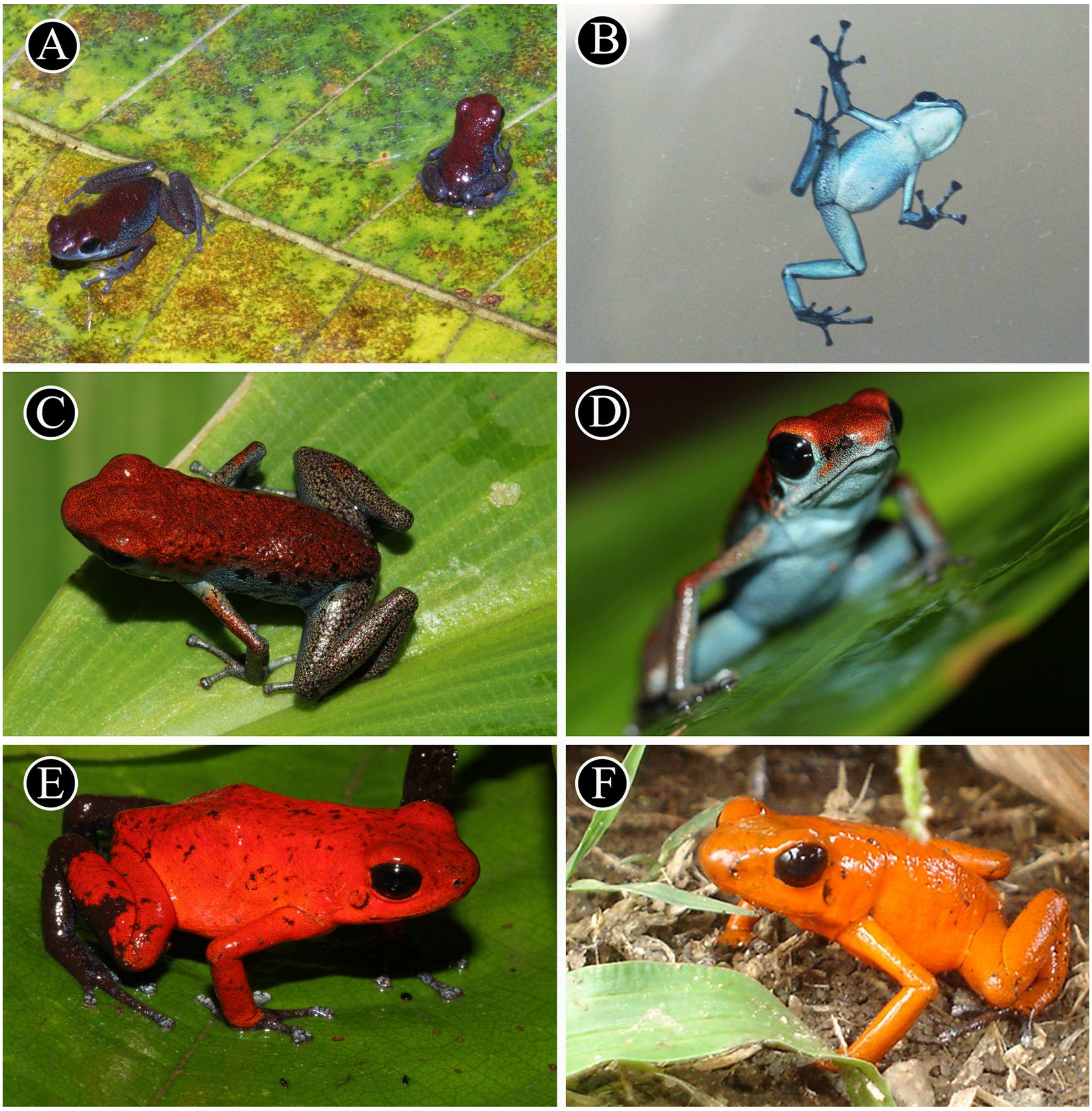

FIGURE 8. Other dendrobatid frogs, similar to *Andinobates geminisae* present in the Caribbean lowlands of Panama. A-B) *Oophaga vicentei*, from Donoso, Colón province, near type locality of *A. geminisae* (8.81823 °N, 80.69956° W); C-D) *Oophaga pumilio*, from Kusapín, Bocas del Toro province, Panama; E-F) *O. pumilio* from Las Delicias, Bocas del Toro province, Panama.

Discussion

According to the molecular phylogenetic results, we include *A. geminisae* in the *A. minutus* species group. However, the new species shows some marked differences with those diagnostic characters for the group proposed by Brown *et al.* (2011). The lack of stripes on dorsum and the pale marbling on venter, along with the bright coloration are discordant traits relative to the current definition of the *A. minutus* species group. Polymorphism within and rapid divergence among species of *Ranitomeya* and *Andinobates* in coloration is common (Brown *et al.* 2011), often caused by mimicry or local adaptation (Symula *et al.* 2001). Therefore, the marked differences presented here between *A. geminisae* and other species of the *A. minutus* group should not be surprising. Additional traits such as larvae morphology and call properties appear to be more conserved within the genus (Brown *et al.* 2011).

To identify possible cryptic species of Neotropical frogs from genetic distance data, thresholds of 10% at COI and 3% at 16S have been suggested (Fouquet *et al.*, 2007), whereas *A. geminisae* (MVUP 2428) shows at COI 6–8% divergence and at 16S 2.5% divergence from *A. minutus* Table 4–5). Many dendrobatid lineages, however, show even smaller interspecific genetic distances (Roberts *et al.*, 2006), e.g., among species of *Oophaga* and *Dendrobates* (Fig. 4). The phylogenetic results statistically place the new species firmly in the *Andinobates minutus* species group (Fig. 4), while the above genetic distances suggest that *A. geminisae* could be considered a candidate species (Padial *et al.* 2010). We therefore used morphological and bioacoustic data to diagnose and differentiate the new species from all congeners within the species group and within the geographic region.

The call of *A. geminisae* showed remarkable differences compared to the other species of *Andinobates* from Panama. The presence of one or two spaced pulses ("clicks") at the beginning or at the end of the call have been reported previously in *A. minutus* (Myers & Daly 1976, Fig. 16B), however the specimen recorded is from an indeterminate locality (either Colombia or Panama), and the call is >5.4 kHz, thus unlikely to be conspecific with *A. geminisae.* It is not known if the spaced pulses found in *A. geminisae* (Fig. 7) have some influence on behavior, but in many anuran species the calling male may modify his call, and usually these modifications play a role in communication. e.g., in the mating call, the territorial call, the distress call, the warning call or the release call (Littlejohn 1977; Narins *et al*. 2000).

Amphibians are in constant threat by many factors that have been discussed at length elsewhere (Lips *et al.* 2006; Wake & Vredenburg 2008; Hof *et al.* 2011). For the geographically restricted *A. geminisae* as for other Caribbean lowland endemics in Panama (e.g., *A. claudiae,* and *Craugastor evanesco* Ryan *et al.* 2010) habitat reduction remains the main issue that could threaten population stability and persistence over the short term. Integrative studies on this new species are urgently required, due to the lack of information on this newly discovered species. Data on population ecology, bioacoustic behavior, breeding biology and parental care are much needed for *A. geminisae.*

Acknowledgement

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