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A new species of direct-developing frog of the genus *Pristimantis* (Anura: Terrarana: Craugastoridae) from Cordillera del Cóndor, Ecuador, with comments on threats to the anuran fauna of the region

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Abstract

A new frog in the genus *Pristimantis* is described from a cloud forest on the western flanks of the Cordillera del Cóndor and eastern Andean slopes in the province of Zamora Chinchipe, southeastern of Ecuador. We inferred its phylogenetic position using DNA sequences of mitochondrial and nuclear genes. The new species is strongly supported as part of a clade that includes *P. ardalonychus*, *P. cajamarcensis, P. ceuthospilus*, *P. chalceus*, *P. minutulus*, *P. luteolateralis, P. parvillus*, *P. ockendeni*, *P. unistrigatus*, and *P. walkeri*. It can be distinguished from all other species from Cordillera del Cóndor and congeneric species by the unique combination of the following characters: (1) iris light blue with black reticulations; (2) skin of dorsum finely shagreen with scattered pustular tubercles and absence of dorsal folds; (3) tympanic membrane and tympanic annulus visible; (4) snout rounded in dorsal and lateral view; (5) upper eyelid bearing two or three enlarged subconical tubercles; (6) cranial crest absent; (7) males lacking vocal sac and slits; and (8) venter uniformly bright red, light red, salmon or orange. The new species is most closely related to *P. ardalonychus, P. cajamarcensis*, *P. ceuthospilus*, *P. ockendeni* and *P. unistrigatus*. We consider the new species to be Endangered following IUCN criteria because it has been severely affected by large scale open-pit mining in some localities. Currently, the amphibian fauna of the Cordillera del Cóndor and nearby protected areas are threatened by large–scale copper and gold mining projects with devastating effects on ~20 species, including several undescribed ones.

Key words: *Pristimantis yantzaza* **sp. nov.**, bioacoustics, conservation, systematics, taxonomy

Resumen

Describimos una nueva especie del género *Pristimantis* de los bosque nublados del flanco occidental de la Cordillera del Cóndor y las estribaciones orientales de los Andes en la provincia de Zamora Chinchipe, sureste de Ecuador. Inferimos su posición filogenética en base a secuencias de ADN de genes mitocondriales y nucleares. La nueva especie forma parte de un clado que incluye a *P. ardalonychus*, *P. cajamarcensis, P. ceuthospilus*, *P. chalceus*, *P. minutulus*, *P. luteolateralis, P. parvillus*, *P. ockendeni*, *P. unistrigatus*, y *P. walkeri*. La nueva especie se diferencia de todas las especies que habitan en la Cordillera del Cóndor y otras especies congenéricas por la combinación de los siguientes caracteres: (1) iris azul claro con reticulaciones negras; (2) piel del dorso finamente granular con tubérculos pustulares dispersos y ausencia de pliegues dorsales; (3) membrana y anillo timpánicos visibles; (4) hocico redondeado en vista dorsal y lateral; (5) párpado superior con dos o tres tubérculos subcónicos alargados; (6) crestas craneales ausentes; (7) machos con saco y hendiduras vocales ausentes; y (8) vientre uniformemente rojo, rojo claro, salmón o anaranjado. La nueva especie se encuentra cercanamente

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relacionada a *P. ardalonychus, P. cajamarcensis*, *P. ceuthospilus*, *P. ockendeni* y *P. unistrigatus*. Asignamos a la nueva especie a la categoría "En Peligro" siguiendo los criterios de la IUCN debido a que ha sido severamente afectada por la minería a gran escala en las localidades donde es conocida su presencia. Actualmente, la fauna anfibia de la Cordillera del Cóndor y áreas protegidas cercanas se encuentra amenazada por proyectos mineros de cobre y oro a gran escala, con efectos devastadores en ~20 especies de Anfibios, incluyendo varias especies aún no descritas.

Palabras clave: *Pristimantis yantzaza* **sp. nov.**, bioacústica, conservación, sistemática, taxonomía,

Introduction

The genus *Pristimantis* Jimenez de la Espada, 1871 comprises about 517 species distributed in Central and South America (Heinicke 2007; Hedges *et al.* 2008; Padial *et al*. 2014, Frost 2017). *Pristimantis* is the most diverse and morphologically variable group of anuran amphibians of the Neotropical region (Hedges *et al.* 2009), and occurs in a variety of habitats and microhabitats (Hedges *et al*. 2008). Currently, 200 species of the genus *Pristimantis* are recognized in Ecuador (Frost 2017; Ron *et al*. 2017). A large proportion of Ecuadorian species are endemic to the country (226 species, 46% of total), especially from montane localities in Amazonia slopes and Andean highlands, and the Cordilleras del Cóndor and Cutucú (Lynch 1979; Lynch & Duellman 1980, 1988; Duellman & Pramuk 1999, Ron *et al*. 2017).

Anuran species diversity in the Cordillera del Cóndor is significantly underestimated (Almendáriz *et al*. 2014). 104 species are known to occur on this region, and a high percentage of taxa (~39%) remain unnamed or have unclear taxonomy (Almendariz *et al*. 2014). Over past decade, several new species have been discovered in this region (Cisneros–Heredia & Morales–Mite 2008; Teran–Valdez & Guayasamin 2010; Almendáriz *et al*. 2012, 2014, 2017; Brito *et al.* 2014). These new species are found in historically inaccessible areas (Stotz *et al.* 1996). Additionally, the Cordillera del Condor and nearby protected areas are currently threatened by large–scale copper and gold mining projects with devastating effects on biodiversity. Given that naming species is essential for species conservation (Angulo & Icochea 2010) it is a priority to describe species in this area as soon as possible. Herein, we provide evidence for, and name and describe a new species of the genus *Pristimantis* collected during several expeditions during the past ten years to the cloud forest on the Cordillera del Condor, southeastern Ecuador.

Materials and methods

Morphology. Specimens collected were euthanized in a solution of Lidocaine 2% and fixed in 10% formaldehyde. Adult specimens were transferred to and kept in 70% ethanol. Prior to fixation, tissue samples from some specimens were preserved in 96% ethanol. Terminology and abbreviations used to describe the morphological characters follows Lynch & Duellman (1997) and Duellman & Lehr (2009): SVL (snout–vent length), distance from tip snout to posterior margin of vent; HL (head length) distance from the tip of the snout to the posterior angle of jaw articulation; HW (head width), greatest wide of head measured to the level of the jaw articulation; ED (eye diameter), distance between anterior and posterior borders of eyes; END (eye–nostril distance), distance from the posterior margin of the nostril to anterior margin of the eye; EW (eyelid width), transversal distance between anterior and posterior margin of the eyelid; IOD (inter–orbital distance), distance between left and right eyes; IND (inter–nostril distance), distance between left and right nostrils; ETD (eye–tympanum distance), distance from the posterior margin of the eye to anterior margin of the tympanum; TYD (tympanum diameter), distance between the anterior and posterior margins of the tympanic annulus; TL (tibia length), length of flexed leg from knee to heel; FL (femur length), length of flexed leg from knee to cloaca; FoL (foot length), distance from the heel to the tip of Toe IV; and HaL (hand length), distance from the elbow to the tip of Finger III. All measurements were taken using Neiko® 01407A digital caliper, accurate to the nearest 0.1 mm. Observations on the color of the frogs in life were based on field notes and color slides of specimens. Sex was determined by examination of secondary sexual characters (i.e., presence or absence of vocal slits). All examined specimens and additional comparative materials are housed in following institutions: British Museum of Natural History (BMNH), London, United Kingdom; División de Herpetología, Museo Ecuatoriano de Ciencias Naturales, Instituto Nacional de Biodiversidad (DHMECN), Quito, Ecuador; Division de Herpetología, Escuela Politécnica Nacional (MEPN), Quito, Ecuador;

Museo de Anfibios y Reptiles, Fundación Herpetológica Gustavo Orcés (FHGO), Quito, Ecuador; Museo de Zoología, Pontificia Universidad Católica del Ecuador, Quito, Ecuador (QCAZ); National Museum of Natural History (NMNH), Washington, USA; Natural History Museum, The University of Kansas (KU), Kansas, USA;

DNA extraction, amplification and sequencing. Genomic DNA was extracted from muscle or liver tissue preserved in 96% ethanol or tissue storage buffer, using standard phenol–chloroform extraction protocols (Sambrook *et al*. 1989). We used a polymerase chain reaction (PCR) to amplify DNA fragments for the mitochondrial gene 16S rRNA (739 bp) and the nuclear gene RAG-1 (601 bp) using primers listed in Goebel *et al*. (1999), Wiens *et al*. (2005), and Moen & Wiens (2009). PCR amplification was performed under standard protocols and sequenced by the Macrogen Sequencing Team (Macrogen Inc., Seoul, Korea). The newly generated DNA sequences were deposited in GenBank (Table 1).

TABLE 1. Vouchers, GenBank accession numbers and locality data for gene sequences of *Pristimantis* used in phylogenetic analyses.

Species	Voucher number	GenBank Accession No.		Locality
		16S	RAG1	
P. altamazonicus	KU215460	EF493670	EF493441	Peru: Madre de Dios, Cuzco Amazonico, 15 km E Puerto Maldonado
P. ardalonychus	KU212301	EU186664	-	Peru: San Martin, Rio Cerranayacu, 76 km NW Rioja
P. cajamarcensis	KU217845	EF493663	-	Ecuador: Loja, 13 km S Yangana
P. ceuthospilus	KU212216	EF493520	-	Peru: Cajamarca, Chota, 12 km W Llama
P. chalceus	KU177638	EF493675	-	Ecuador: Carchi, Maldonado
P. croceoinguinis	KU217862	EF493665	-	Ecuador: Morona-Santiago, 53.8 km E Bella Union via Santiago
P. diadematus	KU221999	EU186668	-	Peru: Loreto, Teniente Lopez
P. imitatrix	KU215476	EF493667	-	Peru: Madre de Dios, Cuzco Amazonico, 15 km E Puerto Maldonado
P. inguinalis	ROM40164	EU186676	-	Guyana: District 7, Mount Ayanganna
P. lirellus	KU212226	EF493521	-	Peru: San Martin, Rioja, Rio Cerranayaca, 76 km NW Rioja
P. luteolateralis	KU177807	EF493517	-	Ecuador: Pichincha, Tandapi
P. marmoratus	ROM43913	EU186692	-	Guyana: District 7, Mount Ayanganna
P. minutulus	KU291677	EU186657	-	Peru: Pasco, Cacazu
P. ockendeni	KU222023	EF493519	EF493434	Peru: Loreto, 1.5 km N Teniente Lopez
P. orestes	KU218257	EF493388	-	Ecuador: Azuay, 7 km E Sigsig
P. parvillus	KU177821	EF493352	-	Ecuador: Pichincha
P. pulvinatus	KU181015	EU186723	-	Venezuela: Bolivar, km 127, El Dorado-Santa Elena de Uairen Rd
P. simonbolivari	KU218254	EF493671	-	Peru: Madre de Dios, Bosque Protector, Cashca Tororas
P. unistrigatus	KU218057	EF493387	EF493444	Ecuador: Imbabura, 35 km E Pquela
P. versicolor	KU218096	EF493389	EF493431	Ecuador: Zamora -Chinchipe, 1.7 km E Loja border
P. walkeri	KU218116	EF493518	EF493428	Ecuador: Pichincha, 5 km W La Florida
P. yantzaza	QCAZ63294	MF615248	MF615250	Ecuador: Zamora-Chinchipe, 20 km E Santa Clara Del Vergel
P. yantzaza	QCAZ63295	MF615249	MF615251	Ecuador: Zamora-Chinchipe, 20 km E Santa Clara Del Vergel

DNA sequence analyses. The sequences were edited and assembled using the program Geneious R9 (Kearse *et al*. 2012), and aligned with the closest BLAST matches downloaded from Genbank using MAFFT v. 7 (Katoh & Standley 2013) under the G-INS-i option. For taxon sampling selection we performed preliminary neighboor-

joining and maximum likelihood phylogenetic analysis including all 16S and RAG-1 sequences of *Pristimantis* available from GenBank (> 1000 terminals), using the MEGA 6 software (Tamura *et al*. 2013). The resulting trees showed us the position of the new species and based on this, we used all the available species closest to our species for more intensive searches and for calculating support. A subset of 20 *Pristimantis* ingroup terminals/species was selected. The edited alignments of both 16S and RAG-1 were concatenated to get a final single alignment, which was then used for all further phylogenetic analyses. We used PartitionFinder v. 1.1.1 (Lanfear *et al*. 2012) to select the best-fit models of sequence evolution with a Bayesian information criterion (BIC). Molecular phylogenetic relationships were inferred using Maximum Likelihood (ML) and Bayesian Inference (BI). ML analyses were conducted in GARLI v. 2.1 (Zwickl 2006) performing five independent searches with 100 replicates each, with the "genthreshfortopoterm" set to 100,000. Node support was assessed with non-parametric bootstrapping (Felsenstein 1985) with 1000 pseudoreplicates. The 50% majority rule consensus for the bootstrap trees was obtained with Geneious R9 (Kearse *et al*. 2012). Bayesian analyses were conducted with MrBayes 3.2.6 (Ronquist & Huelsenbeck 2003) performing two parallel runs with four incrementally heated Markov chains each with 14,000,000 generations, with a sampling frequency of 100. The first 25% of the trees were discarded as burn-in and the remaining ones were used to generate a 50% majority rule consensus tree, as well as to estimate the Bayesian posterior probabilities. Uncorrected *p*-genetic distances were estimated with software MEGA6 (Tamura *et al.* 2013).

Bioacoustics. Terminology and measurements used in acoustic analysis follow Duellman & Pyles (1983), Cocroft & Ryan (1995), Angulo *et al*. (2003), Diaz & Cádiz (2007), and Batallas & Brito (2016). Calls were recorded on June 2011, at Los Encuentros, Yantzaza, province of Zamora Chinchipe by Jorge Brito, from one unvouchered specimen (Digital Sound Collection DHMECN unnumbered) at a distance less than 3 m, using unidirectional microphone Sennheiser K6–C and Olympus WS–750 digital recorder. We used Adobe Audition CS6 software for noise filtration and temporal analysis (Batallas & Brito 2014). For calls diagramming and spectral analysis we used Raven 1.4 software (Charif *et al*. 2010), with a frequency resolution of 44.1 kHz and 16 "bits" using window Hann to 50 % of superposition and Fourier Fast Transformation (FFT) with 512 points of resolution. Parameters analyzed were: (1) Dominant frequency: measuring along all call and all components; (2) Calls duration: time from the start to the end of a call; (3) Call intervals: time elapsed between calls; (4) Calls/minute: calls repetition rate lapse per minute; (5) Pulses/calls: number of acoustic units in an amplitude recognized pattern; (6) Duration of pulses; (7) Intervals between pulses; (8) Pulses/second: pulse repetition rate lapse per second.

Results

Phylogeny. Alignments resulted in 739 and 610 positions for 16S and RAG-1, respectively. PartitionFinder under BIC identified two partition scheme (16S/RAG-1) with GTR+I+G as the optimal substitution model for both aligned fragments (Scheme BIC: 14020.5). The phylogenetic trees constructed by Bayesian inference and Maximum likelihood showed the same topology, with the ML tree providing higher support values at deeper nodes (Fig. 1). Both our preliminary analyses of 16S and the combined analysis of RAG and 16S placed the new species as closely related to several species from the former *Pristimantis unistrigatus* and *P. frater* species groups: *P. unistrigatus*, *P. ockendeni*, *P. ceuthospilus*, *P. cajamarcensis*, *P. ardalonychus*, and *P. minutulus* (a well-supported clade recovered by Padial *et al*. 2014), distributed in Colombia, Ecuador, and Peru. In our tree *Pristimantis yantzaza* **sp. nov.** appears to be closest to the clade formed by *P. unistrigatus* and *P. ockendeni* (Fig. 1); however, support values were low. Uncorrected *p*-genetic distances for the gene *16S* between *P. yantzaza* **sp. nov.** and its closest relatives range from 11% to 15%, *P. ardalonychus, P. cajamarcensis, and P. unistrigatus* being the closest (Table 2). These large genetic divergences, its phylogenetic position, and morphological divergence supported by unique traits provide evidence that populations herein named as *Pristimantis yantzaza* **sp. nov.** constitute a species that was unknown and unnamed until now.

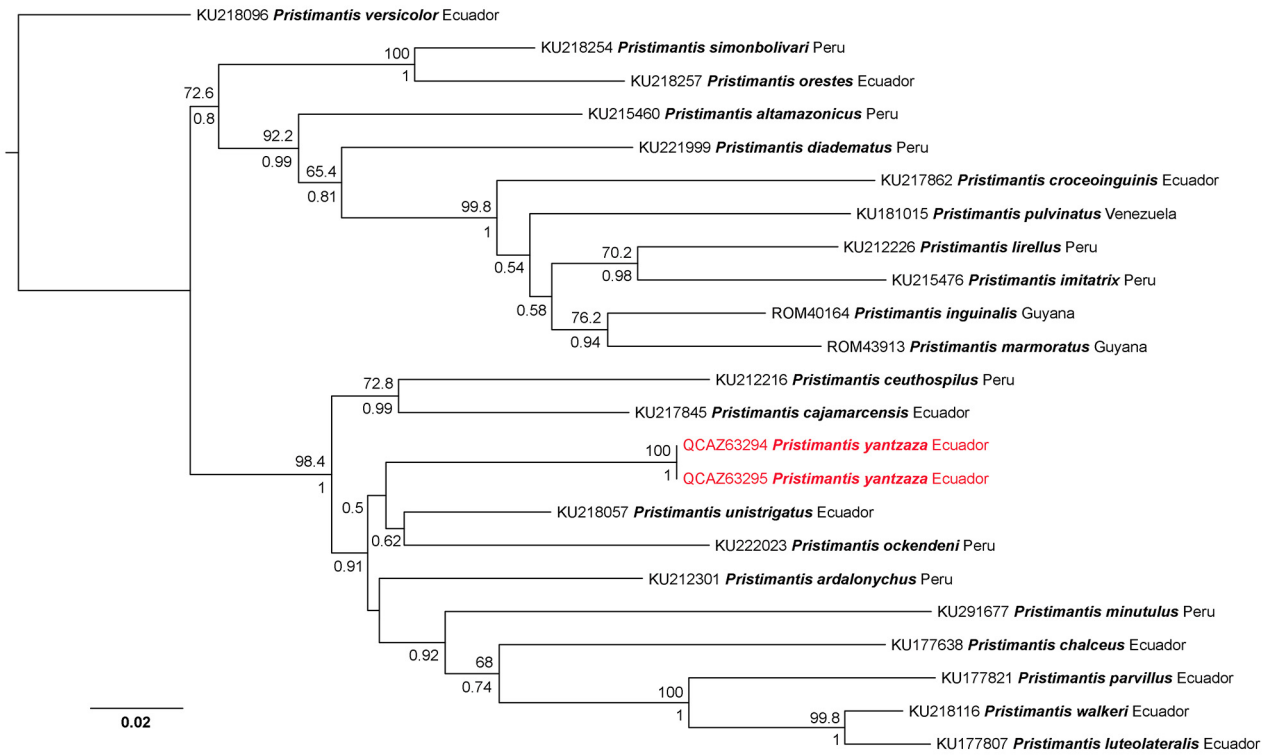

FIGURE 1. Maximum likelihood tree inferred from 3221 aligned positions of the 16S (2589 aligned bp) and RAG-1 (632 aligned bp) genes depicting the relationships of *Pristimantis yantzaza* **sp. nov.**. Bootstrap values are shown above the branches and Bayesian posterior probabilities are shown below except when they are below 50 (bootstrap) or 0.5 (posterior probability). The tree was rooted with *Pristimantis versicolor*. Museum catalog numbers and locality of origin for vouchers are listed in Table 1.

TABLE 2. Uncorrected pairwise distances (%) for a fragment (739bp) of the mitochondrial gene 16S for *Pristimantis yantzaza* **sp. nov.** and its closest relatives.

	Taxa	1	2	3	4	5	6	7	8	9	10	11
1	*Pristimantis ceuthospilus* KU212216											
2	*Pristimantis cajamarcensis* KU217845	10.6										
3	*Pristimantis yantzaza* QCAZ63294	13.2	12.1									
4	*Pristimantis yantzaza* QCAZ63295	13.2	12.1	0.0								
5	*Pristimantis unistrigatus* KU218057	11.6	11.2	11.0	11.0							
6	*Pristimantis ockendeni* KU222023	13.9	15.2	14.4	14.4	11.4						
7	*Pristimantis ardalonychus* KU212301	11.4	12.3	12.1	12.1	10.8	13.4					
8	*Pristimantis minutulus* KU291677	14.8	13.3	14.3	14.3	14.0	16.3	13.4				
9	*Pristimantis chalceus* KU177638	15.9	14.8	14.9	14.9	13.1	14.9	14.0	13.0			
10	*Pristimantis parvillus* KU177821	17.5	14.9	15.1	15.1	13.6	15.6	14.4	16.0	14.4		
11	*Pristimantis walkeri* KU218116	16.5	15.1	15.5	15.5	15.4	17.9	14.8	16.1	13.9	10.3	
12	*Pristimantis luteolateralis* KU177807	17.0	15.7	14.3	14.3	15.3	17.7	15.2	16.3	13.5	10.4	3.8

***Pristimantis yantzaza* sp. nov.**
Suggested English name: Yantzaza´s Rainfrog
Suggested Spanish name: Cutín de Yantzaza
(Figs. 2A–B)

Holotype. FHGO 9962 (Figs. 2–3) an adult female from La Zarza, Los Encuentros, Yantzaza, (3°46'36" S, 78°29'47" W, 1478 m elevation), Zamora Chinchipe province, Republic of Ecuador, collected on 12 October 2014 by M. R. Dueñas and D. Hurtado.

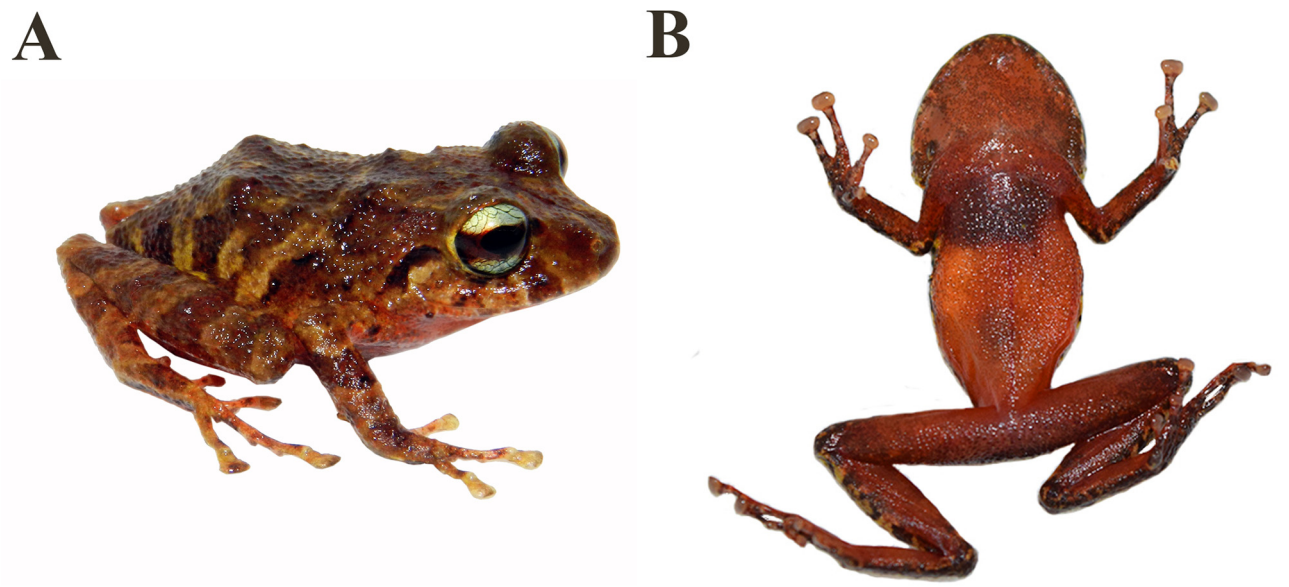

FIGURE 2. Holotype of *Pristimantis yantzaza* **sp. nov.** in life, FHGO 9962, adult female, SVL 24.4 mm: A) dorsal view, and B) ventral view.

Paratopotypes (*n* =6). DHMECN, 11696, 11697, an adult male and an adult female collected on 12 December 2012 by M. R. Dueñas; FHGO 9963–65 an adult male and two adult females with the same collection data as the holotype. FHGO 8346, an adult female, collected on 25 February 2011 by A. Barahona and H. Cadena.

Paratypes (*n* = 26). DHMECN, 11693 an adult female and DHMECN 11700, 11702 two juvenile males from Colibrí, Los Encuentros (3°45'47" S, 78°30'13" W, 1680 m), Zamora Chinchipe province, collected on 12 December 2015 by M. R. Dueñas; FHGO 8346, 9963, two adult females from Colibrí, Relaves, Yantzaza (3°45'42" S, 78°30'16" W, 1390 m), Zamora Chinchipe province collected on 25 February 2011 by A. Barahona and H. Cadena. DHMECN, 8504, an adult female, (3°34'31" S, 78°26'10" W, 1346 m), collected on 2 December; DHMECN 10157, an adult female, (3°31'55" S, 78°23'43" W, 926 m), collected on 6 December 2011; DHMECN 11147, an adult female (3°34'28" S, 78°26'04" W, 1253 m), collected on 23 June 2013; DHMECN 11149, 11153–54, 11159, three adult males and an adult female (3°35'07" S, 78°26'22" W, 1590 m), collected on June 2013, all from Concesión Mirador, Tundayme, Pangui, Zamora Chinchipe province collected by R. M. Betancourt, D. Chungandro and M. Alcoser; FHGO 8884, 8886, two adult female from Las Peñas, Relaves, Yantzaza (3°46'25" S, 78°29'43" W, 1480 m), Zamora Chinchipe province, collected on 4 November 2012 by J. H. Valencia; QCAZ 63294, 63295, two adult females from 20 km E Santa Clara del Vergel, Palanda (4°36'41" S, 78°54'12" W, 2030 m), Zamora Chinchipe Province, Ecuador, collected on 24 March 2016 by P. Székely. Ten adult males, QCAZ 65056 (4°35'54" S, 78°51'34" W, 1644 m), QCAZ 65063 (4°36'14" S, 78°49'42" W, 2112 m), QCAZ 65096, 65098–99, 65102, (4°36'6" S, 78°49'51" W, 2033 m), QCAZ 65110 (4°36'1" S, 78°49'57" W, 2014 m), QCAZ 65112 (4°36'2" W, 78°49'56" W, 2016 m), QCAZ 65143 (4°35'57" S, 78°50'4" W, 1981 m), QCAZ 65147 (4°35'40" S, 78°50'25" W, 1832 m), collected on 20–27 August 2016, all from Reserva Biológica Cerro Plateado, La Canela, Palanda, Zamora Chinchipe province, by D. Almeida, E. Nursirquia, F. Ayala, J. Pinto, A. Achig, and M. Bustos.

Referred material (*n* = 12). DHMECN, 11693 (3°45'03" S, 78°30'40" W, 1620 m) an adult female and DHMECN 11700 (3°45'47" S, 78°30'13" W, 1680 m) a juvenile male, both collected on 12 December 2012 from Colibrí, Los Encuentros, Zamora Chinchipe province by M. R. Dueñas. DHMECN 11150, 11159, two adult females (3°34'33" S, 78°26'43" W, 1250 m), DHMECN 11157, 11160 an adult male and an adult female (3°34'47" S, 78°26'22" W, 1361 m), all collected on June 2013 from Concesión Mirador, Tundayme, Pangui, Zamora Chinchipe province collected by R. M. Betancourt and D. Chungandro. Seven adult males QCAZ 65057 (4°35'40"

S, 78°51’14” W, 1619 m), QCAZ 65095 (4°36’6” S, 78°49’51” W, 2033 m), QCAZ 65103 (4°36’11” S, 78°49’40” W, 1996 m), QCAZ 65119 (4°36’10” S, 78°49’48” W, 2073 m), QCAZ 65146 (4°35’57” S, 78°50’6” W, 1980 m), QCAZ 65148, 65150 (4°35’38” W, 78°50’31 W, 1803 m), collected on 20–27 August 2016, all from Reserva Biológica Cerro Plateado, La Canela, Palanda, Zamora Chinchipe province, by D. Almeida, E. Nursirquia, F. Ayala, J. Pinto, A. Achig, and M. Bustos.

Comparisons with other species. Coloration in this section refers to live specimens unless otherwise noted. *Pristimantis yantzaza* **sp. nov.** is readily distinguished from other congeneric species in Cordillera del Condor by its bright reddish, salmon or orange ventral coloration and iris cream bluish to light blue with dark reticulations and a wide dark red mid-horizontal band. Ventral coloration is shared with *P. paquishae* (Brito *et al.* 2014) and *Pristimantis proserpens* (Lynch 1979), however *P. paquishae* has vocals slits and short and robust digits, while *P. proserpens* lacks discoidal folds and has longer digits. In Cordillera del Condor there are fifteen other species of *Pristimantis* (Almendariz *et al*. 2014) that could be related and or confused with *P. yantzaza* (characters of the new species given in parenthesis). *Pristimantis croceoinguinis* (Lynch 1968) and *P. ventrimarmoratus* (Boulenger 1912) lack tympanic annulus and tympanic membranes (annulus and membrane tympanic present); *P. trachyblepharis* (Boulenger 1918) and *P. spinosus* (Lynch 1979) have tympanum concealed beneath skin, length 30–40% eye length (annulus and membrane tympanic distinct, 40% eye length), *P. prolatus* (Lynch & Duellman 1980), *P. proserpens* (Lynch 1979), and *P. versicolor* (Lynch 1979) lack discoidal fold (discoidal fold present); *Pristimantis altamazonicus* (Barbour & Dunn 1921), *P. serendipitus* (Duellman & Pramuk 1999), and *P. exoristus* (Duellman & Pramuk 1999) have vocal slits in males (vocal slits absent); *P. diadematus* (Jimenez de la Espada 1875) has a tarsal fold (tarsal folds absent); *P. muscosus* (Duellman & Pramuk 1999) has reddish iris (iris cream bluish to light blue); *Pristimantis minimus* Terán–Valdez & Guayasamin 2010, is a minute frog, SVL 12.2 mm in males and 9.5–13.7 mm in females (SVL 17.6–22.0 in males, 21.6–34.0 in females); *P. rhodostichus* (Duellman & Pramuk 1999), has fingers bearing distinct lateral fringes (fingers bearing narrow lateral fringes), has low cranial crests (cranial crests absent). *Pristimantis tungurahua* Reyes–Püig *et al*. 2010, distributed on eastern Andean slopes, also have reddish venter coloration; however *P. tungurahua* has dorsolateral folds, subacuminate snout in dorsal view and acuminate in lateral view, and heel with prominent calcar. Additionally, *P. yantzaza* can be easily distinguished from all the other phylogenetically close species, *P. ardalonychus* (Duellman & Pramuk 1999), *P. cajamarcensis* (Barbour & Noble 1920), *P. ceuthospilus* (Duellman & Wild 1993), *P. minutulus* Duellman & Hedges 2007, *P. ockendeni* (Boulenger 1912), and *P. unistrigatus* (Günther 1859), by its characteristic reddish, salmon or orange ventral coloration.

Etymology. The specific epithet *yantzaza* refers to canton of Yantzaza, one of the eight cantons of the province of Zamora Chinchipe, where the new species is known to occur. In addition, Yantzaza is an Ecuadorian Shuar word that means “firefly”. Yantzaza was traditionally called “Firefly Valley” or “Valle de Yantzaza” by indigenous Shuar people. The epithet is a noun in apposition.

Diagnosis. We assign this species to the genus *Pristimantis* on the basis of its phylogenetic position. The new species also shows a general morphological similarity with many other members of the genus (including characteristic T–shaped terminal phalanges, toes without membranes, and Toe V longer than Toe III). *Pristimantis yantzaza* **sp. nov.** is characterized by: (1) skin of dorsum finely shagreen with scattered pustular tubercles, dorsal folds absent; skin of belly coarsely areolate, discoidal folds weak; thoracic fold present; (2) tympanic membrane and tympanic annulus evident, round; horizontal diameter of tympanic annulus 45–50% of eye diameter and separated from the eye by a distance of approximately one third of the length of the eye; (3) snout short, subacuminate in dorsal view, rounded in profile, *canthus rostralis* distinct, rounded in dorsal view and slightly sharp in lateral profile, loreal region slightly concave to flat; nostrils slightly protuberant, directed dorsolaterally; area between nostril slightly convex; (4) upper eyelid bearing several small and flat tubercles and two or three enlarged and sub–conical supraocular tubercles, approximately 60% of eye diameter; cranial crests absent; (5) choanae triangular not concealed by palatal shelf of maxillary arch; dentigerous processes of vomers small and ovoid; slightly smaller than choanae, separated medially by distance lower than width of odontophore; each odontophore has 5 to 7 teeth; (6) males lacking vocal sac and slits; (7) Finger I shorter than Finger II, all fingers bearing subtruncate disks, approximately three times the width of the digit proximal to the pad; (8) fingers bearing narrow lateral fringes; (9) small ulnar tubercles present; (10) inner edge of tarsus bearing indistinct tubercles, heel bearing one large tubercle and several smaller tubercles; tarsal fold absent; (11) inner metatarsal tubercle ovoid and slightly elevated, about four times the diameter of the outer tubercle; outer metatarsal tubercle small, sub-conical;

supernumerary plantar tubercles present; (12) toes bearing narrow lateral fringes; Toe IV much longer than Toe III; discs broadly expanded, elliptical, about same size as those on fingers; (13) dorsum yellowish tan to dark brown, mottled with darker brown or with cream vertebral and dorsolateral stripes; concealed surface of thigh reddish, salmon or orange with brown bars; venter and groin uniformly bright reddish or orange, throat bearing minutes spots; (14) SVL in adult males 17.6–22.1 mm (n = 6), females 23.2–34.0 mm (n = 16).

Description of holotype. Adult female (Figs. 3A–D) with, 24.4 mm SVL; head slightly wider than body; head barely wider than long; head length 41.9% of SVL. *Canthus rostralis* sharp, loreal region slightly concave to flat, sloping abruptly; nostrils slightly protuberant, directed laterally; eye-to-nostril distance 47.7% of eye diameter; lips not flared; upper eyelid bearing several small and flat tubercles and two enlarged and sub-conical supraocular tubercles; snout rounded in dorsal and lateral view; tympanic membrane and tympanic annulus evident, round, its upper and posterodorsal part obscured by rounded supratympanic fold; supratympanic fold thick and slightly warty; several low postrictal tubercles situated posteroventrally to tympanic annulus; interorbital distance greater than the width of upper eyelid; choanae small, rounded, not concealed by palatal shelf of maxillary arch; dentigerous processes of vomer small and oblique with four teeth; tongue longer than wide, bilobate, posterior 1/3 not adherent to floor of mouth.

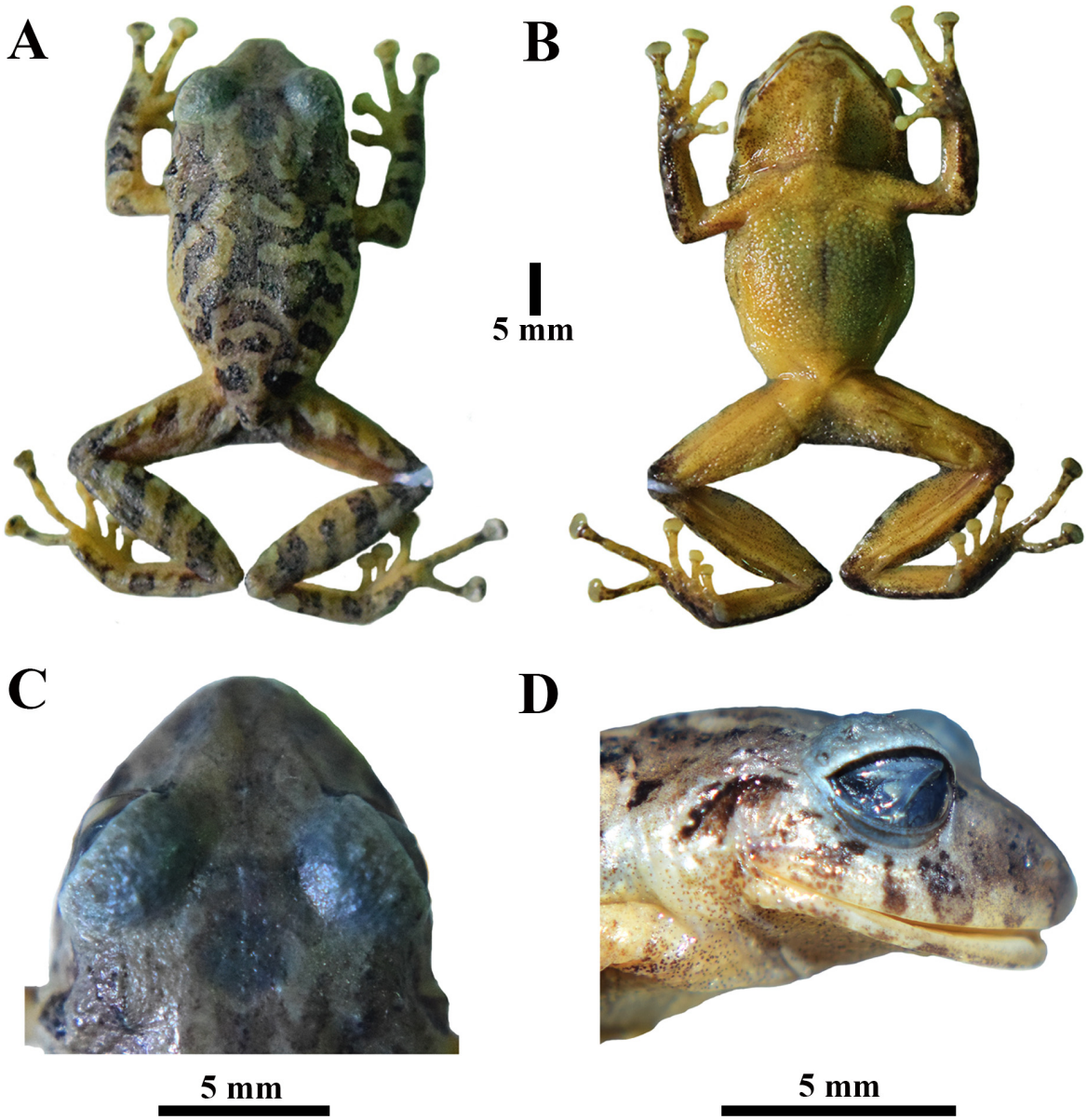

FIGURE 3. Holotype of *Pristimantis yantzaza* **sp. nov.** (FHGO 9962, adult female, SVL 24.4 mm) in preservative: A) dorsal view, B) ventral view, C) head, dorsal view, and D) head, lateral view.

Skin of dorsum finely shagreen; dorsolateral folds absent; skin of throat shagreen; thin, low mid dorsal fold starting at tip of snout and ending at cloaca (trait more visible in life); low interocular fold present (trait more visible in life); ventral and lateral skin coarsely areolate; discoidal folds weak; thoracic fold present (trait more visible in life); small ulnar tubercles present without forming a distinct fold; thenar tubercle oval equal (right hand) or barely smaller (left hand) than divided palmar tubercle; supernumerary palmar tubercles present; subarticular tubercles prominent and rounded, including the most distal tubercle; fingers bearing narrow lateral fringes, all fingers bearing pads well defined by circumferential grooves; Finger I shorter than Finger II; tips of digits subtruncated; inner digit of hand bearing barely narrower disc than outer digits (Fig. 4A); all fingers bearing pads on digital tips; tips of Finger IV reaches beyond distal subarticular tubercle on Finger III; tip of Finger II reaches distal subarticular tubercle of Finger III; knee bearing low tubercles.

Hind limbs moderately long; tibia long 56.5% of SVL; foot long 46.1% of SVL; Toe V much longer than Toe III; tip of Toe III not reaching the distal edge of the penultimate subarticular tubercle on Toe IV; tip of Toe V extending beyond the distal edge of distal subarticular tubercle on Toe IV (Fig. 4B); disks of inner toes slightly smaller than outer fingers; tips of digits subtruncated; webbing absent; subarticular tubercles on toes distinct, projecting and ovoid; supernumerary plantar tubercles at base of toes present; inner metatarsal tubercle ovoid and elevated, almost half the length of Toe I; lateral fringes absent; outer metatarsal tubercle small, sub–conical; tarsus without fold but small sub–conical tubercles along outer border, indistinct on inner border; heel bearing one larger, rounded tubercle and several smaller tubercles; cloacal sheath and tubercles present, cloacal opening directed posteroventrally at level of thighs.

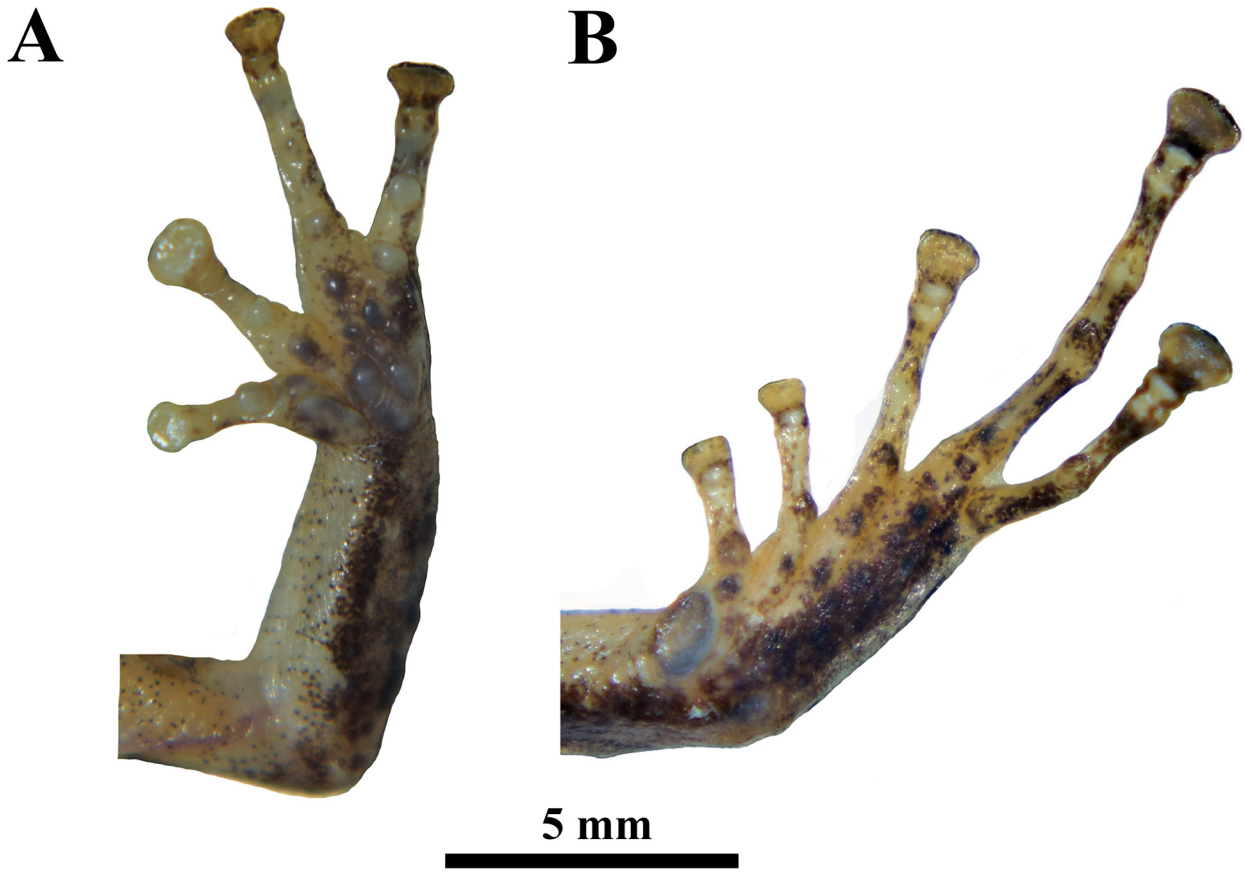

FIGURE 4. Hand and feet of the holotype of *Pristimantis yantzaza* **sp. nov.** in preservative FHGO 58044, adult female: A) palmar view of hand, B) plantar view of foot.

Coloration in life. Dorsum is yellowish–orange with brown-reddish markings; suprascapular region bearing W markings; dark brown interorbital bar; brown cloacal region. Dorsal surfaces of limbs yellowish-orange with dark brown bars. Dorsum of hands and feet with brown blotches, more abundant on outer fingers and toes; fingers I and II pale reddish–brown. Flanks bright yellowish with reddish-brown markings. Belly, axial region and groin

bright reddish lacking spots or blotches, reddish throat finely mottled with minute brown spots. The concealed surfaces of limbs are reddish-brown finely mottled with pale brown. Ventral surfaces of tarsus dark brown. Palmar surface reddish densely mottled with brown, subarticular, palmar and thenar tubercles reddish; dark brown bars originating below the eyes and extending towards the lips. Iris bright creamy with a reddish median, horizontal streak and faint black reticulations (Figs. 2A–B).

Coloration in preservative. Dorsum is pale brown with dark brown markings; dark brown interorbital bar; brown cloacal region. Dorsal surfaces of limbs pale brown with dark brown bars. Dorsum of hands and feet pale brown with dark brown blotches on outer fingers and toes. Flanks pale brown with dark brown markings. Belly, axial region and groin pale orange lacking spots or blotches, pale brown throat finely mottled. The concealed surfaces of limbs are brown finely mottled with dark brown. Ventral surface of tarsus dark brown. Palmar surface pale brown densely mottled with dark brown, subarticular, palmar and thenar tubercles dark brown; dark brown bars originating below the eyes and extending towards the lips (Fig. 3A–D).

Measurements of holotype (in mm). SVL 24.4; HL 10.3; HW 10.7; ED 4.4; END 2.1; EW 2.6; IOD 3.2; IND 1.9; ETD 1.2; TYD 1.5; TL 13.7; FL 11.2; FoL 10.8; HaL 7.3.

TABLE 3. Measurements (in mm) and proportions of adult males and females of *Pristimantis yantzaza* **sp. nov.** (range, average ± SD).

	Males *n*=17	Females *n*=16
SVL	17.6–22.1 (19.9 ± 1.6)	21.6–3.4 (29.4 ± 4.3)
HW	6.9–8.6 (7.4 ± 0.6)	8.4–14.7 (12.0 ± 1.8)
HL	7.8–9.0 (8.1 ± 0.5)	9.8– 14.9 (12.1 ± 1.6)
IOD	2.1–2.8 (2.4 ± 0.2)	2.7–4.4 (3.6 ± 0.5)
IND	1.5–2.1 (1.7 ± 0.3)	1.7–3.0 (2.4 ± 0.3)
EW	1.6–2.3 (1.8 ± 0.3)	1.7–3.4 (2.7 ± 0.5)
ED	2.3–3.8 (3.1 ± 0.6)	3.1–5.0 (3.9 ± 0.6)
EN	2.4–3.0 (2.5 ± 0.2)	2.8–4.7 (3.7 ± 0.6)
TD	0.8–2.0 (1.3 ± 0.5)	1.0–2.3 (1.4 ± 0.4)
FL	7.7–14.9 (9.8 ± 2.6)	10.4–15.2 (13.3 ± 1.6)
TL	9.5–12.2 (10.9 ± 0.9)	10.8–17.7 (15.1 ± 2.2)
FoL	7.7–9.8 (8.8 ± 0.8)	10.1–14.1 (12.6 ± 1.3)
HaL	5.9–6.4 (6.1 ± 0.2)	6.5–9.9 (8.6 ± 1.0)
HW/SVL	0.3–04 (0.4 ± 0.0)	0.4–05 (0.4 ± 0.0)
HL/SVL	0–4–0.4 (0.4–0.0)	0.4–05 (0.4 ± 0.0)
HL/HW	1.1–1.2 (1.1 ± 0.0)	0.9–1.2 (1.0 ± 0.1)
EN/HL	0.3–0.3 (0.3 ± 0.0)	0.3–0.3 (0.3 ± 0.0)
ED/HL	0.3–0.5 (0.4 ± 0.1)	0.3–0.5 (0.3 ± 0.1)
EW/IOD	0.7–0.9 (0.8 ± 0.1)	0.5–1.0 (0.8 ± 0.2)
EN/ED	0.7–1.0 (0.8 ± 0.1)	0.6–1.3 (1.0 ± 0.2)
TD/ED	0.2–0.9 (0.4 ± 0.3)	0.2–0.6 (0.4 ± 0.3)
FL/SVL	0.4–0.8 (0.5 ± 0.1)	0.4–0.5 (0.5 ± 0.0)
FoL/SVL	0.4–0.5 (0.4 ± 0.0)	0.4–0.5 (0.4 ± 0.0)

Variation. Morphometric variation in adult males and females is described in Table 3. Color variation in life is shown in Figure 5. Broad dark brown middorsal with narrow cream middorsal lines, and dorsolateral stripes extending from lever of eyelids to posterior end of body (DHMECN 11702, FHGO 8884, 9963, QCAZ 65056, 65063, 65112, 65147). A cream vertebral stripe is present in the holotype and several additional specimens (DHMECN 11702, FHGO 8346, QCAZ 65063, 65098, 64143, 65147). Brown markings on dorsum reduced to the

scapular region in a specimen (QCAZ 65096). W-shaped scapular mark on dorsum barely differentiated in the holotype and in some additional specimens (DHMECN 11702, 11693, FHGO 9962, 9965) and strongly defined in a specimen (FHGO 9964). Sacral chevron marks in four specimens (DHMECN 11693, 11697, FHGO 8346, QCAZ 65056, 65063, 65147). Supratympanic stripes barely differentiated in a specimen (QCAZ 65096). Interocular bar is strongly differentiated in a specimen (QCAZ 65110, 65096). Bar lips barely differentiated in two specimens (QCAZ 65096, 65099). Canthal stripe absent in a specimen (QCAZ 65096). A specimen (FHGO 9965) with minute pale brown spots on belly and throat. The row of tarsal tubercles is undistinguished in the preserved specimen FHGO 8264.

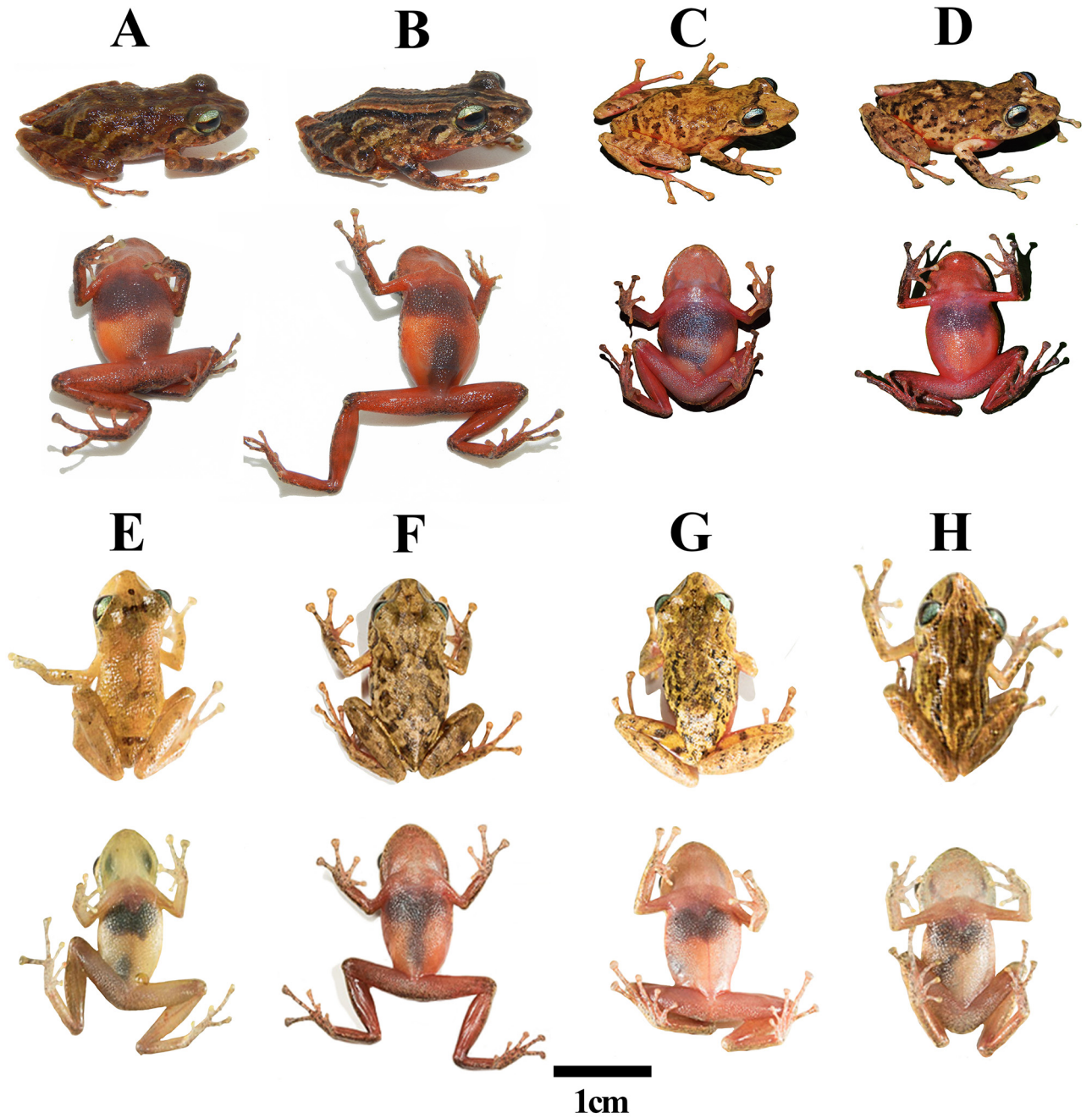

FIGURE 5. Live specimens of *Pristimantis yantzaza* **sp. nov.** showing variation in dorsal and ventral coloration. From left to right, first and second rows: FHGO 9965, FHGO 9963, QCAZ 47544, QCAZ 47545 (females), third and fourth rows: QCAZ 65095, QCAZ 65098, QCAZ 65099, QCAZ 65112 (males). All specimens are shown in the same scale.

Advertisement call. The advertisement call (mean ± SD is provided for all acoustic parameters analyzed) is characterized by a modulated frequency. The calls are a cadential series of metallic sounds, which could be

confused with cricket sounds. Dominant frequency = 3.62 ± 0.10 kHz. Call duration = 222.50 ± 35.64 ms, with intervals = 5393.30 ± 1828.60 ms, Call per minute = 12.27 ± 4.48 calls/minute. Calls are multipulsed by mean = 4.58 ± 0.67 pulses with duration = 17.23 ± 3.24 ms. Calls intervals = 38.80 ± 7.51 ms, producing a mean = 17.93 ± 2.57 pulses/second (Table 4). Pulses have a slight increase in frequency of 0.02–0.08 kHz between pulses, having 0.12–0.21 kHz between the first and last pulses. Choruses are not orderly structured, males close to each other call antiphonally (Fig. 6).

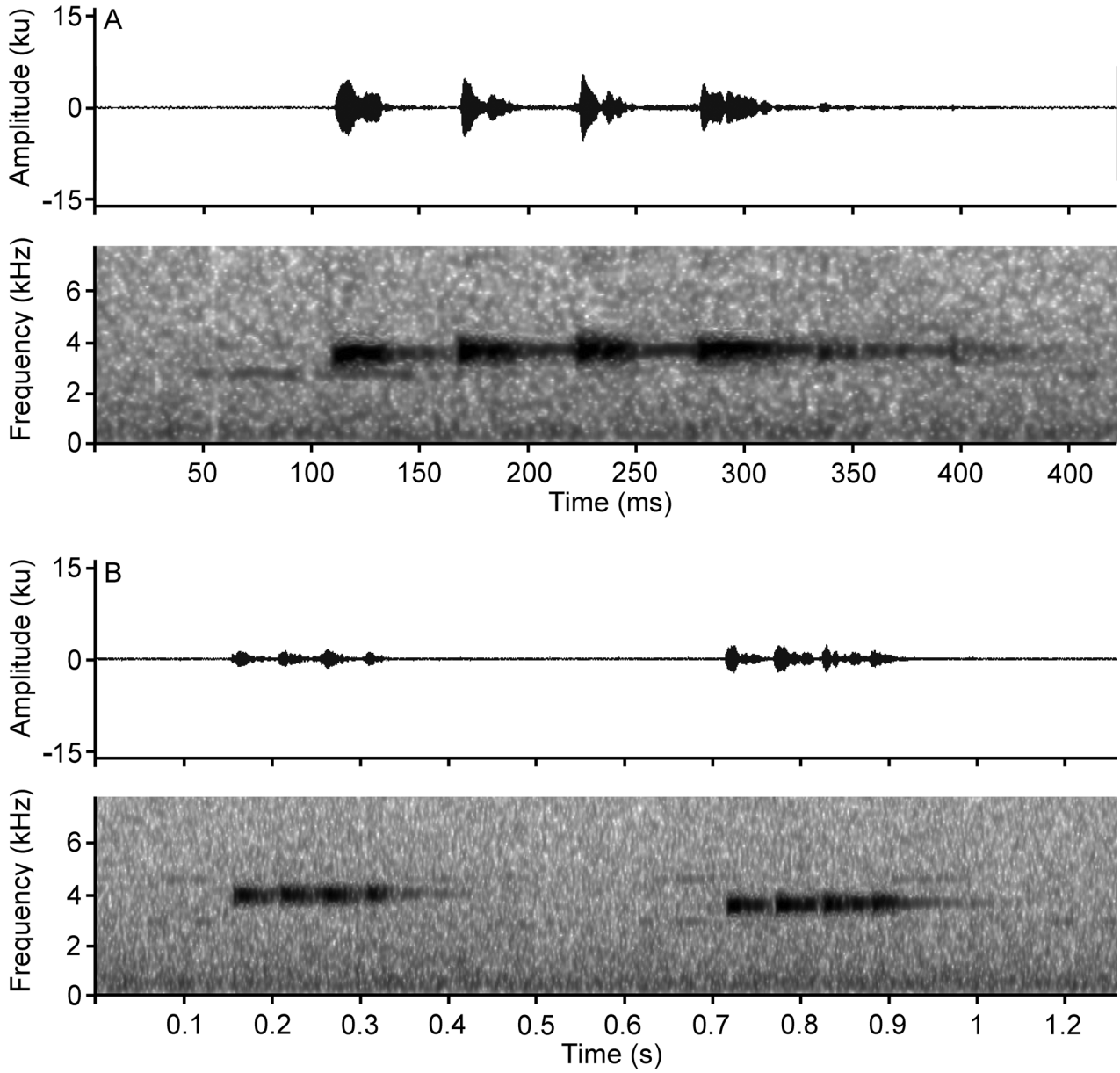

FIGURE 6. Oscillogram and spectrogram of the advertisement call of *Pristimantis yantzaza* **sp. nov.** (unvouchered specimen).

Distribution and natural history. Known only from five localities in the province of Zamora Chinchipe, all in the Cordillera del Condor (La Zarza, Colibrí, Mirador, and Las Peñas), (Cerro Plateado) (Fig. 7). The type locality is in evergreen montane forest between 1200–1800 m elevation (based on Ministerio del Ambiente de Ecuador 2012, ecosystem classification system; Fig. 8A–B). The following ecological information was obtained at the type locality. *Pristimantis yantzaza* is nocturnal, males were observed calling partially concealed among leaves. A few males were heard calling very sporadically from other small vegetation islands by day or night, but could not be located. Juveniles were observed on low vegetation or moss, adults on upper leaves of bushes 0.30 to 5.00 m above ground (Fig. 8C–D). Two amplectant pairs were collected on a bromeliad (*Guzmania* sp.). Sympatric congeners include *Pristimantis incomptus*, *P. prolatus*, *P. proserpens*, and *P. serendipitus*.

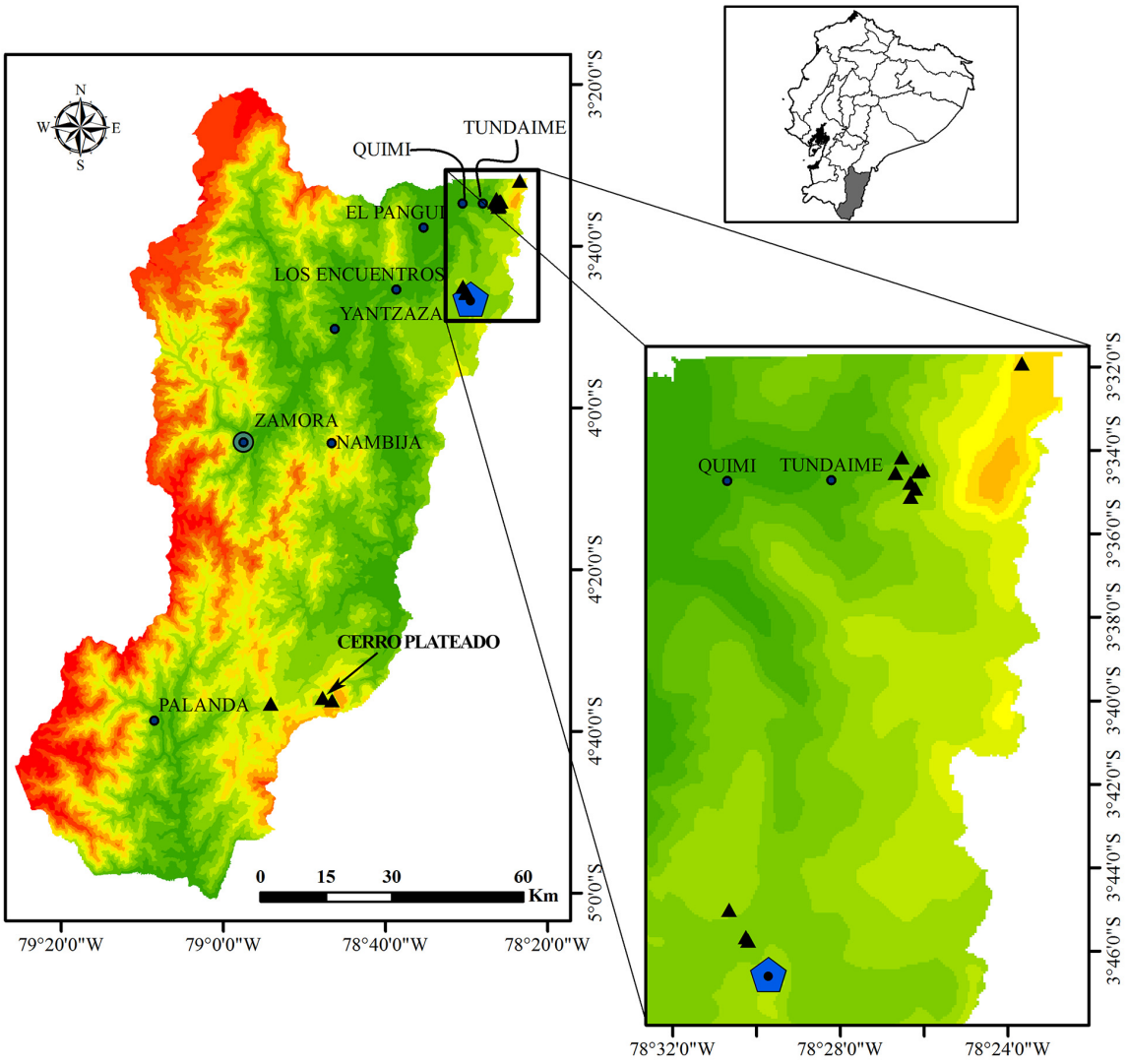

FIGURE 7. Distribution of *Pristimantis yantzaza* **sp. nov.** (triangles) in eastern Ecuador. The blue pentagon indicates the type locality.

TABLE 4. Summary of the acoustic analysis of the calls of *Pristimantis yantzaza* **sp. nov.** Sample size (N) corresponds to the calls/pulses. Parameters abbreviations are: kHz = kilohertz, ms = milliseconds, SD = Standard deviation.

Parameters	Range (mean ± SD)	Sample (*n*)
Dominant frequency (kHz)	3.44–3.81 (3.62 ± 0.10)	11/52
Call duration (ms)	147–288 (222.50 ± 35.64)	11
Call intervals (ms)	2805–8631 (5393.30 ± 1828.60)	11
Calls/minute	6.95–21.41 (12.27 ± 4.48)	11
Pulses per calls	4–6 (4.58 ± 0.67)	11/52
Pulses duration (ms)	13–26 (17.23 ± 3.24)	11/52
Pulses intervals (ms)	24–58 (38.80 ± 7.51)	11/52
Pulses/seconds	12.98–25 (17.93 ± 2.57)	11/52

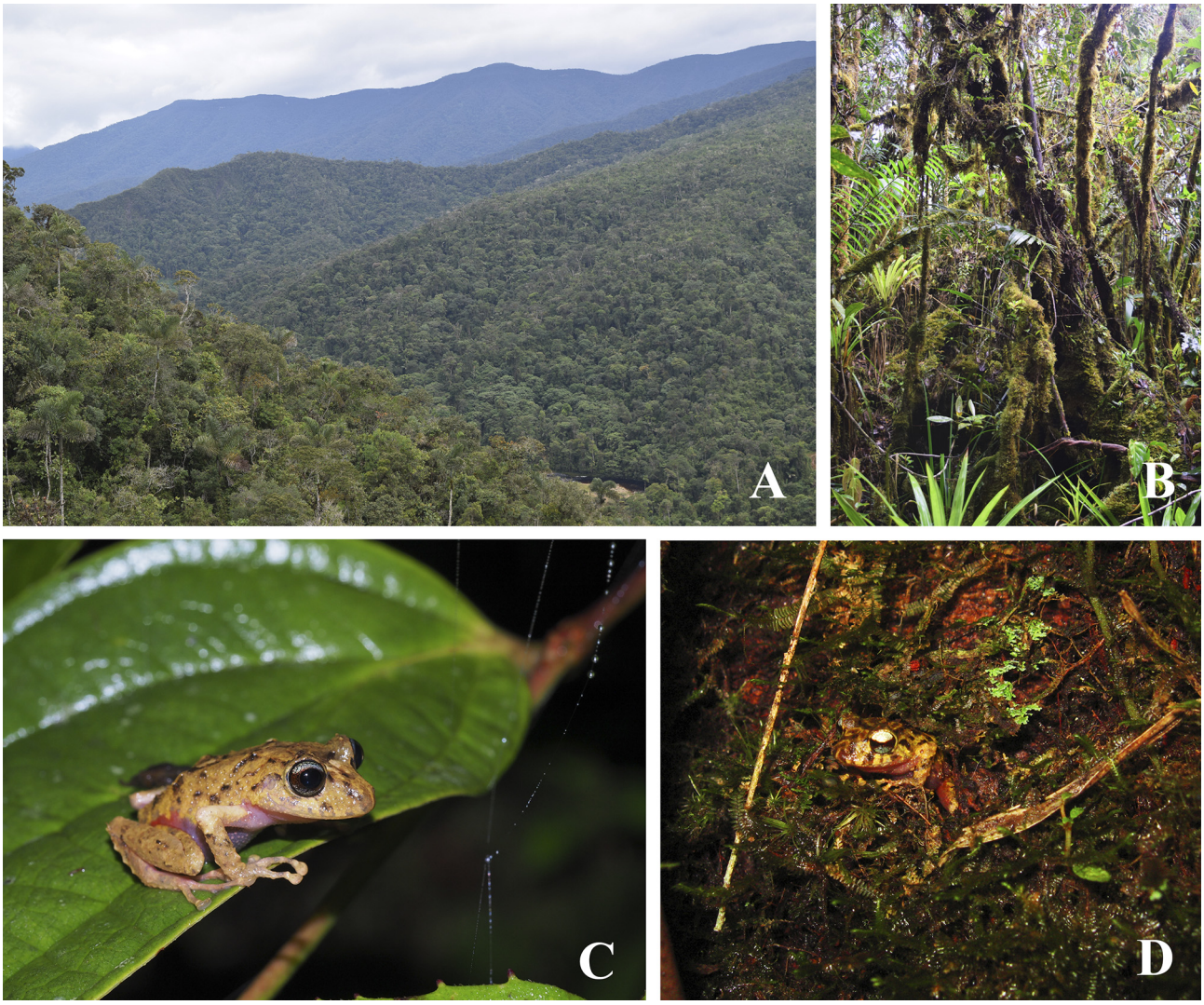

FIGURE 8. A) Panoramic view of the habitat of *Pristimantis yantzaza* **sp. nov.**, Cordillera del Condor; near Los Encuentros, Zamora Chinchipe; B) habitat at the type locality; C) an adult female perching on leaves at Palanda; and D) an adult female camouflaged on moss at Tundayme.

Conservation status. We consider this species to be Endangered following A4B1a,b(iii) IUCN criteria because: (i) its extent of occurrence is estimated to be less than 22 km^2 (i.e., total area of continuous montane forest in the Cordillera del Condor and Andes); (ii) it has been detected in five localities in the province of Zamora Chinchipe, one of which was severely affected by a large scale open-pit mining; and (iii) its habitat is severely fragmented, isolated from other such habitats and declining in extent and quality due to deforestation.

Discussion

Evolutionary relationships. Molecular evidence show that the new species is part of the genus *Pristimantis*. Its unique ventral coloration readily differentiates it from closely related species. Our phylogenetic analyses recovered the main subdivisions of the genus obtained by Padial *et al.* (2014), showing that *P. yantzaza* is related to *P. unistrigatus, P. ockendeni, P. ardalonychus, P. cajamarcensis* and *P. ceuthospilus*. All these species are part of a clade distributed in the Andes from southern Colombia and Ecuador to northern and central Peru. The phylogenetic relationships inferred from the mitochondrial and nuclear DNA matrix suggest that *P. yantzaza* closest relatives are *P. unistrigatus* and *P. ockendeni*. However, the low support for some nodes leading to *P. yantzaza* indicates that the relationships of this species may change with additional sequence data and species sampling.

Species on the clade of *P. yantzaza* and relatives were assigned by Hedges *et al*. (2008) to the *Pristimantis unistrigatus* group (except *P. ockendeni* which was considered part of the *P. frater* group). The *P. unistrigatus* group was rendered the most diverse group of the genus, with almost 200 species distributed from the Amazon Basin to the high Andes in northeastern South America. However, the group was rejected by Padial *et al*. (2014), who found it to be polyphyletic. Therefore, we refrain to assign *P. yantzaza* to any species group until an exhaustive taxonomic revision of *Pristimantis* redefines species groups based on well supported clades.

Threats to the anuran amphibian fauna at the type localities. The Cordillera del Cóndor is a mountain range in eastern Ecuador that is largely isolated from the main Andes. The area is characterized by an exceptional soil (i.e., composed of sand with few nutrients) and black and white water streams (Guayasamin *et al*. 2011). These features have a notable influence on biodiversity and endemism (Schulenberg & Awbrey 1997; Freile & Santander 2005; Clark *et al*. 2010; Guayasamin *et al.* 2011; Almendariz *et al.* 2014). However, the Cordillera del Cóndor forms part of the most threatened ecoregion in the world (Myers *et al*. 2000). Guayasamin *et al*. (2011), indicate that the most direct threats to the area's biodiversity are: (1) expansion of agriculture and cattle ranching, (2) logging, (3) large and small scale mining, and (4) the introduction of diseases to the site, and (5) the effects of climate change.

Based on our field data, large-scale mining is the major threat for biodiversity conservations in this area. This activity, especially open-pit mining, has caused intensive logging, destroying locally several important ecosystems, habitats and microhabitats of anuran amphibian populations (Valencia and Dueñas, *unpublished data*). At localities where *P. yantzaza* occurs, Pangui and Yantzaza, there are two private concessions mining large amounts of cooper and gold. The cantons of Pangui include the mining localities of Condor Mirador, Wawayme, Eneretsa, Escombrera, Las Maravillas, San Marcos, Tundayme, Vivero, Quimi and Valle del Quimi; while Yantzaza includes Los Encuentros an extensive area of cloud forest. Natural areas in both cantons are severely threatened and could be destroyed in a few years.

In June 2007, one of us (JHV) sampled the amphibian fauna in six localities at Pangui including the mining area. Vegetation at the sites consisted of secondary forest and artificial open areas. The surveys showed high diversity of amphibian fauna (27 species, search efford 160 person/hours) within a reduced area (~1000 m^2). Eight years later, between 2014 and 2015, JHV and MDT visited three of the six localities sampled in 2007 and found that all vegetation had been destroyed, and soil and waters were polluted.

To mitigate the environmental impacts, the mining company is implementing an amphibian and reptile rescue program at Pangui and Yantzaza. The program consists on releasing individuals rescued from area impacted by mining. Several rescued individuals are released on previously selected areas. The rescue program apparently occurs before, during, and after the logging activities. At Pangui, some of the areas where rescued animals were released will be severely impacted by mining in the future. Consequently, a new rescue and release program will be necessary for the rescued amphibian and reptiles. Also, irreversible and negative impact has been observed on the water bodies (rivers, streams, marshes, and seasonal ponds), due to the increase in suspended solids during mining operations. Additionally, rivers and streams are used as deposit of sediments, affecting the quality of the water (Fig. 9).

Mining activities have affected the survival of amphibian populations, of the genera *Allobates*, *Centrolene*, *Hyalinobatrachium*, *Hyloxalus*, *Hypsiboas*, *Leptodactylus*, *Noblella, Nymphargus*, *Pristimantis*, *Ranitomeya, Rulyrana*, and *Teratohyla* (Valencia and Dueñas*, unpublished data*) (Table 5). Populations declined because of the accelerated and extensive environmental impacts, especially complete destruction of the natural vegetation. Unfortunately, the impacted area is expanding and more populations of rare species will be affected. For example, during fieldwork in April 2016, JHV visited Tudayme and found a small population of *Callimedusa equatoriana*, a species listed as Endangered (EN) under IUCN criteria (Coloma & Ron 2004), in this area that will undergo mining operations.

A minimum of eight previously unknown species of anurans have been discovered during the last eight years from this area (e.g., Cisneros–Heredia & Morales–Mite 2007; Almendáriz *et al.* 2014). Nevertheless, surveys suggest that the herpetofauna of Cordillera del Cóndor is incompletely known and many species wait to be described (Almendariz *et al*. 2014). Amphibian populations will disappear in the near future, some perhaps even before they can be found, named, and scientifically described unless serious conservation actions take place.

TABLE 5. Date of the last observations for species of amphibians found at some localities near Tundayme (Cordillera del Cóndor) where native vegetation has been severely or totally destroyed, and water bodies strongly polluted.

Taxa	Locality records	Last seen date
AROMOBATIDAE		
Allobates kingsburyi	San Marcos	August 2014
DENDROBATIDAE		
Excidobates condor	San Marcos	August 2009
Hyloxalus italoi	Mina	August 2014
CENTROLENIDAE		
Centrolene audax	Enerentsa	August 2014
Centrolene durrellorum	Mina	August 2014
Hyalinobatrachium pellucidum	Mina, Borde Mina	May 2013
Nymphargus cochranae	Borde de Mina	August 2014
Rulyrana mcdiarmidi	Mina, Eneretsa	August 2014
Teratohyla amelie	Mina	August 2014
CRAUGASTORIDAE		
Pristimantis delius	Mina	August 2014
Pristimantis metabates	Enerentsa	May 2010
Pristimantis acuminatus	Mina, Eneretsa	August 2014
Pristimantis katoptroides	Mina	July 2011
Pristimantis rhodostichus	Eneretsa	August 2014
Pristimantis serendipitus	Mina, Ampliación Escombrera, Enerentsa, Cóndor Mirador	August 2014
HEMIPHRACTIDAE		
Hemiphractus bubalus	Contrafuerte Wawayme	August 2014
HYLIDAE		
Boana boans	Mina	August 2014
Boana cinerascens	San Marcos	August 2014
Boana fasciata	Ampliación Escombrera, Enerentsa, San Marcos	August 2014
LEPTODACTYLIDAE		
Leptodactylus knudseni	San Marcos	August 2014

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FIGURE 9. Intensive habitat destruction and fragmentation are important threats for amphibian populations in Cordillera del Condor, sector of Tundayme, Zamora Chinchipe: A) panoramic view of extensive deforestation at Tundayme; B) fragmented forest due to new roads opened near to Mirador; C) wetlands destroyed at San Marcos; D) small streams with deposit of sediments in Eneretsa.

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APPENDIX I. Specimens examined.

Pristimantis altamazonicus: ECUADOR: Morona Santiago, Makuma, Centro Shuar Shimpim (FHGO 1958, 4818, 4830, 4834).

Pristimantis croceoinguinis: ECUADOR: Sucumbios, Santa Cecilia (KU 110789, holotype photo); Morona Santiago, Taisha, Makuma, Centro Shuar Makuma (FHGO 5120, 5184).

Pristimantis diadematus: ECUADOR: Napo, Río Aguarico (KU 123381, holotype photo); Zamora Chinchipe, El Pangui, Tundayme, (FHGO 7813, 7842, 7853), Zamora Chinchipe, El Pangui, Tundayme, La Mina (FHGO 7814, 7867); Zamora Chinchipe, El Pangui, Tundayme, Cara de Indio (FHGO 7859).

Pristimantis exoristus: ECUADOR: Morona Santiago, Unsuants, Cordillera del Cutucú (FHGO 10249).

Pristimantis paquishae: ECUADOR: Zamora Chinchipe, Yantzaza, Los Encuentros, Concesión Emperador (FHGO 11494–97); Zamora Chinchipe, Paquisha Alto (MEPN 15337, holotype photo)

Pristimantis prolatus: ECUADOR: Napo, 2 km SSO Rio Reventador (KU 166008, holotype photo); Zamora Chinchipe, El Pangui, Tundayme, Vivero (FHGO 11414–18). Ecuador, Zamora Chinchipe, Yantzaza, Las Peñas (FHGO 8864–77); Zamora Chinchipe, Yantzaza, Los Encuentros (FHGO 8898, 8913, 8918); Zamora Chinchipe, San Carlos, Río Nambija (FHGO 9003).

Pristimantis proserpens: ECUADOR: Morona Santiago, Rancho Sapote and Suro (NMNH 198484, holotype photo); Zamora Chinchipe, Curintza (FHGO 2666); Zamora Chinchipe, Romerillos Alto (FHGO 2797).

Pristimantis rhodostichus: ECUADOR: Zamora Chinchipe, El Pangui, Tundayme, La Mina (FHGO 7861, 7877, 7880); Zamora Chinchipe, La Pituca, cuenca del Río Curintza, (FHGO 2307–09).

Pristimantis serendipitus: ECUADOR: Zamora Chinchipe, El Pangui, Río Wawayme, (FHGO 7856, 7860, 7887); Zamora Chinchipe, El Pangui, Tundayme, Escombrera Norte (FHGO 7907); Zamora Chinchipe, La Pituca, cuenca del Río Curintza, FHGO (1762, 1790).

Pristimantis spinosus: ECUADOR: Morona Santiago, Sapote, descanso de San Rafael (NMNH 199891, holotype photo); Zamora Chinchipe, Espínola, Amaluza, Angashcola, (FHGO 7349–51).

Pristimantis trachyblepharis: ECUADOR: Pastaza, Mera (KU 120190, holotype photo); Zamora Chinchipe, El Pangui, San Marcos, (FHGO 7844); Zamora Chinchipe, El Pangui, San Cruz (FHGO 7827).

Pristimantis ventrimarmoratus: ECUADOR: Morona Santiago, Alshi Zuña, 9 de Octubre (FHGO 9177–9178); Napo, Cascada San Rafael (FHGO 1951–52, 1963). PERU: Junín, Chanchamayo (BMNH 1947.2.15.73, holotype photo).

Pristimantis versicolor: ECUADOR: Zamora Chinchipe, La Pituca, cuenca del Río Curintza, (FHGO 1740–41, 1750, 1767, 1795, 1830); Zamora Chinchipe, 15 km al este de Loja (KU 119866, holotype photo).