Herpetological Conservation and Biology 11(1):90–100.
Submitted: 14 May 2015; Accepted: 22 March 2016; Published: 30 April 2016.

DISTRIBUTION, DIET, AND VOCALIZATIONS OF THE ENDANGERED COLOMBIAN TOAD *OSORNOPHRYNE PERCRASSA* (ANURA, BUFONIDAE)

***JHONATTAN VANEGAS-GUERRERO*[1,5], *GUSTAVO A. GONZALEZ-DURÁN*[2], AND *SERGIO ESCOBAR-LASSO*[3,4]**

[1]*Grupo de Herpetología de la Universidad del Quindío (GHUQ), Armenia, Quindío, Colombia*
[2]*Instituto de Ciencias Naturales, Laboratorio de Anfibios, Universidad Nacional de Colombia, Bogotá, Colombia*
[3]*Fundación R.A.N.A (Restauración de Ambientes Neotropicales Alterados), Colombia*
[4]*Programa de Postgraduación en Conservación y Manejo de Vida Silvestre, ICOMVIS, Universidad Nacional de Costa Rica, Heredia, Costa Rica*
[5]*Corresponding author, e-mail: jhonattanvanegas@gmail.com*

***Abstract.*— Herein, we describe the distribution, call, diet, and niche model of *Osornophryne percrassa*. This species has been recorded at 23 locations of Colombia, 14 of which are reported in this work. In addition, we reported the first record for the Department of Valle del Cauca, southern Colombia. Based on the points of occurrence, we estimated the potential geographic distribution of this species through the maximum entropy algorithm, and the model prediction showed a good performance. The model shows that The Cordillera Central provides the higher habitat availability for the species occurrence, with no geographical barriers that hinder the spread of the species. We analyzed the stomach contents of 14 females. We identified 73 Arthropoda prey and nine Nematoda prey. Coleopterans and isopods were numerically and volumetrically the more important prey, respectively. We found a positive and significant correlation between snout-vent length (SVL) and head width (HW). Our data indicate that the species is a generalist/opportunistic feeder with a sit-and-wait strategy for obtaining prey. We described the vocal repertoire (advertisement and courtship calls) based on 23 advertisement calls and 11 courtship calls from a male. The advertisement call consists of 8–10 Peep notes whereas the courtship call consists of 6–9 Peep notes. The advertisement calls of *O. guacamayo* were similar in temporal and structural characteristics to those of *O. percrassa*. This is the first description of the courtship calls not only for *O. percrassa* but also for the genus.**

Key Words.—advertisement call; amphibians; Central Andes; Colombia; ecological modeling; geographic distribution

INTRODUCTION

Colombia has a high diversity of amphibians (823 species; Frost 2015) and more threatened species than any other country (274 species; Acosta-Galvis 2015), but there are very few plans to conserve native herpetofauna (Velásquez et al. 2008). Because natural ecosystems are being altered rapidly, little time is available to study natural history parameters required to optimize the management plans for and to determine vulnerability of species (Urbina-Cardona 2011). Andean toads in the genus *Osornophryne* (Bufonidae) are endemic to a region north of the Andes in Colombia and Ecuador, in a small and topographically complex biogeographic area (Páez-Moscoso and Guayasamin 2012) with mountain forests and Páramo at elevations between 2,100 and 4,000 m (Páez-Moscoso and Guayasamin 2012). The taxonomy, systematics, and monophyly of this genus are well established by morphological and molecular characters (Ruiz-Carranza and Hernández-Camacho 1976; Páez-Moscoso et al. 2011; Páez-Moscoso and Guayasamin 2012). Among the 11 species currently known, six of them are listed by the International Union for Conservation of Nature (IUCN) in categories with high extinction risk. Because of their restricted geographic distribution, *Osornophryne antisana*, *O. guacamayo*, *O. percrassa*, *O. puruanta* and *O. talipes* are listed as Endangered (EN), and *Osornophryne sumacoensis*, known from a single locality, is listed as Vulnerable (VU; Bolívar and Lynch 2004). The *Osornophryne* toads exhibit inguinal amplexus, direct development, and defensive behaviors (Ruiz-Carranza and Hernández-Camacho 1976; Escobar-Lasso and González-Duran 2012); however, most aspects of the ecology and natural history of these toads are unknown (Escobar-Lasso and González-Duran 2012).

Five species of *Osornophryne* occur in Colombia, including *O. antisana*, *O. bufoniformis*, *O. guacamayo*, *O. percrassa*, and *O. talipes*. *Osornophryne percrassa* (Herveo Plump Toad; Fig. 1) is endemic to the Colombian Central Andes (Mueses-Cisneros 2003; Páez-Moscoso and Guayasamin 2012) in the Departments of Antioquia, Caldas, Quindío, and Tolima at elevations between 2,700 and 3,700 m (Loaiza-Piedrahíta et al. 2014). This species is found in leaf litter and under rocks in the Andean forests and Páramos, but has not

FIGURE 1. *Osornophryne percrassa* (A: PSO-ZC 2258; B: ICN 55682) collected in the sector of La Mina, Páramo of Chili, municipality of Sevilla, Department of Valle del Cauca (C), Cordillera Central of Colombia. (Photographs by Sergio Escobar-Lasso).

been recorded in disturbed habitats (Bolívar and Lynch 2004). Ruiz-Carranza and Hernández-Camacho (1976), Escobar-Lasso and González-Duran (2012) and Burbano-Yandi et al. (2015) discuss aspects of morphology, habitat use, and behavior.

Osornophryne percrassa is listed as Vulnerable on the Colombian Red List of amphibians because the known geographical distribution is < 5,000 m^2 and suitable habitat is fragmented and declining in both extent and quality on the Colombian Cordillera Central (Bernal and Quevedo 2004). Despite notable advances in the understanding of phylogenetic relationships and taxonomy (Páez-Moscoso and Guayasamin 2012), little is known about either the ecology or precise distribution of *O. percrassa*. Here we provide information on the distribution, diet, and vocalizations of *O. percrassa*. Additionally, we examine bioclimatic variables that potentially influence the species distribution.

MATERIALS AND METHODS

***Distribution and niche modeling*.—**To clarify the distribution of *O. percrassa*, we acquired information from three sources: (1) voucher specimens deposited in scientific collections (Colección Zoológica Universidad del Tolima, Ibague, Tolima (CZUT); Instituto de Ciencias Naturales, Universidad Nacional de Colombia, Bogotá, Cundinamarca (ICN); Instituto Alexander von Humboldt, Villa de Leyva, Boyacá (IAvH); Museo de Historia Natural Universidad de Caldas, Manizales, Caldas (MHNUCa); Museo de Herpetología Universidad de Antioquia, Medellín, Antioquia (MHUA); and Colección de Zoología Universidad de Nariño, Pasto, Nariño (PSO-CZ), (2) records from Ruiz-Carranza and Hernández-Camacho (1976), Mueses-Cisneros (2003), Bernal and Quevedo (2004), Llano-Mejía et al. (2010), Escobar-Lasso and González-Duran (2012), Vanegas-Guerrero and Fernández-Roldán (2014), and Loaiza-Piedrahíta et al. (2014), and (3) field observations. We used this distributional information to generate a potential distribution model for *O. percrassa*. We used ArcGIS 10.1 (ESRI 2011) to identify a 200 km influence area of locality occurrence and construct the distribution model. We characterized the environmental niche using 19 climatic layers provided by WorldClim.org (Hijmans et al. 2005; Appendix 1) with a resolution of 30 arcseconds (about 1 km^2 = 0.0083 degrees of pixel size of the layers).

We created the potential model using the maximum entropy algorithm generated with the software Maxent 3.3.3k (http://www.cs.princeton.edu/~schapire/maxent/), which estimates the distribution probability of the species presence according to the confirmed occurrence localities and associated environmental conditions (Phillips et al. 2006). The recommended values of Maxent were used to the convergence threshold (10–5) and maximum interactions number (500). We avoided the Extrapolate and Clamping options to reduce an

TABLE 1. Call variables measured for *Osornophryne percrassa*. Variable definitions follow those of Cocroft and Ryan (1995) and Hutter *et al*. (2013).

Parameter	Characteristic described
Call/note interval	Measured time between a call/note
Call duration (s)	Measured from start of the amplitude rise away from background noise to a return to background noise
Number of notes	Total number of notes per call
Note length (ms)	Time from start to end of one note; measured for notes at the beginning, middle, and end of call
High and lower frequency (KHz)	Lower and high limit of the fundamental frequency
Dominant frequency (KHz)	Frequency of call which contains the greatest concentration of energy
Power, percent of max	Measure for harmonic frequencies, percent of power in harmonic compared to max power of call

overestimation of the model. We selected automatically the program suitable regularization values, including those to reduce overfitting. We also carried out automatically the selection of features (environmental variables or functions thereof), following default rules dependent on the number of presence records. We calculated a *P*-value across the set of Jackknife predictions using the program *P*-Value Compute made available as supplementary, which is used to test models when the localities number is little and it was taken into account the number of localities to this study (Pearson et al. 2007).

We used a Jackknife test to calculate the relative contribution of each variable in a preliminary model. Considering the results of this analysis, the variables that did not have any contribution were excluded and a new model was performed with the purpose of demonstrating the ecological requirements of the species (Phillips et al. 2006). We evaluated the predictive efficiency of the ecological niche model of *O*. *percrassa* by a Jackknife cross-validation with one of the observed localities excluded. We applied a decision threshold (based on the training localities) and we tested the ability to predict the excluded localities. Because Maxent output is ASCII format, we selected a threshold to create a binary distribution model; we selected a conservative threshold based on the lowest value foretold associated with any sampling sites observed for the model (Pearson et al. 2007).

***Diet*.—**We extracted stomachs of 14 toads (ICN 55683, 85-86, MHN-UC 686-692, and PSO-ZC 2257-2260; for collection localities see Table 2) found under tree trunks and leaf litter in pasturelands. Because of our small sample size of males, we analyzed the stomach content of only adult females. However, males were also sacrificed and preserved. For all captured toads, we measured snout-vent length (SVL), head width (MW), and head length (HL) to the nearest 0.1 mm with manual calipers.

We identified each prey item to either order or family and measured prey items for length (L) and width (W) to the nearest 0.1 mm using manual calipers. We estimated prey volume using the formula for a prolate spheroid: $V=4/3\pi$ (prey length/2) × (prey width/2)^2. The diet of *O*. *percrassa* was quantified as the number (N_i) and the volume (V_i) of each prey taxon (i). We used the Shannon-Wiener diversity index (H') to estimate dietary niche breadth. Because we could not assume normality, we used the Spearman Rank Multiple Correlation to analyze morphology and diet.

***Vocalizations*.—**We recorded both advertisement calls and courtship calls from one individual *O*. *percrassa* at 0107 h 30 September 2013 during a field trip around the municipality of Cajamarca, Department of Tolima, Colombia (4°28’27.6”N, 75°29’38.1”W; 3,323 m elevation). We considered both acoustic characteristics and social contexts of the calls to verify that advertisement calls were distinct from courtship calls. Males used courtship calls only when females were nearby.

To record calls, we positioned a unidirectional microphone (Sennheiser K6/K6P; Sennheiser Electronic GMBH & Co. Kg, Wedemark, Germany) connected to a digital recorder (Marantz PMD671; D&M Holdings, Inc., Tokyo, Japan) 20–30 cm in front of a calling male. We recorded the body size (SVL) of males with digital calipers. We did not record environmental temperature. We estimated the temporal and spectral parameters (Table 1) of the advertisement calls using RAVEN Pro 1.4 (Bioacoustics Research Program 2011). Call parameter definitions follow Cocroft and Ryan (1995) and Hutter et al. (2013). We digitized recordings at 16 bits resolution and 44.1 kHz sampling rate. We analyzed oscillograms and spectrograms with a Fast Fourier Transformation window of 256 points and Blackman algorithm. We measured low and high frequencies of the calls at 20 dB (re 20 mPA) below the peak of intensity of the dominant frequency, which is the value at which the signal energy could still be clearly distinguished from the background noise. We reported the measures as the mean ± one SD followed by the range.

RESULTS

Distribution and modeling niche*.—Osornophryne percrassa* has been reported at 23 locations, nine of which were obtained from either the literature or

TABLE 2. Locality data (sorted from north to south) from records of *Osornophryne percrassa* in Colombia. Museum acronyms as in the text. For localities, ◊ = historical locations, ■ = new records, and * = first record at Valle del Cauca.

Locality	Department	Municipality	Museum voucher	Elevation	Latitude	Longitude	Reference
1◊	Antioquia	Sonsón	MHUA-A 8202	3178	5°43’32”	75º15’08”	Loaiza-Piedrahíta et al. 2014
2■	Caldas	Pensilvania	MHN-UCa 686	3406	5°23’27.26”	75°16’10.77”	New Record
3■	Caldas	Neira	MNH-UCa 659	3594	5°8’17.52”	75°19’32.35”	New Record
4■	Caldas	Neira	MHN-UCa 691	3450	5°8’15.29”	75°19’35.00”	New Record
5■	Caldas	Neira	MHN-UCa 690	3450	5°8’12.61”	75°19’34.47”	New Record
6■	Caldas	Neira	MHN-UCa 689	3450	5°7’26.40”	75°19’44.26”	New Record
7■	Caldas	Neira	MHN-UCa 688	3588	5°7’25.60”	75°19’37.15”	New Record
8■	Caldas	Neira	MHN-UCa 687	3615	5°6’49.94”	75°19’37.20”	New Record
9■	Tolima	Herveo	MHN-UCa 692	3840	5°5’7.86”	75°17’35.38”	New Record
10◊	Tolima	Herveo	ICN 319-36	3200	5°5’4”	75°17’37”	Ruiz-Carranza and Hernández-Camacho 1976
11◊	Caldas	Manizales	MHN-UCa 542	2820	5°4’59.44”	75°24’27.37”	Rojas-Morales et al. 2014
12◊	Tolima	Herveo	ICN 00540-41, 01596, 01641-42, 01922, 01924-25, 02618, 02624-25, 03178-79, 03186, 03208, 03211-12, 03507, 03515, 04738, 17642-43, 17645, 18755-57, 18759-61		5º4’45”	75°10’56”	Mueses-Cisneros 2003
13◊	Caldas	Villa María	MHN-UCa 453	2850	4°59’17.31”	75°24’24.66”	Rojas-Morales et al. 2014
14◊	Caldas	Villa María	MHN-UCa 452	2850	4°59’11.59”	75°24’23.62”	Rojas-Morales et al. 2014
15◊	Quindío	Salento	ICN 33754	2700	4°30’14”	75°33’19”	Mueses-Cisneros 2003
16■	Tolima	Cajamarca	Not collected	3512	4º28’44.2”	75º29’29.1”	New Record
17◊	Tolima	Cajamarca	ICN 10015-23	3040	4°28’12”	75°33’44”	Mueses-Cisneros 2003
18◊	Quindío	Calarcá	Not collected	3600	4°26’30,0”	75°34’35,4”	*Pers. Obs.*
19■	Quindío	Cordoba	Not collected	3409	4° 22’19.4”	75° 35’55.7”	New Record
20■	Tolima	Cajamarca	CZUT-A 167-68, 286	3620	4°15’28”	75°33’35.0”	New Record
21■	Tolima	Roncesvalle	Not collected	3483	4°04’49.9”	75°43’57.4”	New Record
22■	Tolima	Roncesvalle	ICN 55685-86	3260	3°57’30.09”	75°43’05.93”	New Record
23*	Valle del Cauca	Sevilla	ICN 55682-84, 55687-88, PSO-ZC 2257-60	3053	3°58’39.83”	75°49’38.46”	New Record

scientific collections, and 14 were new records (Table 2). We report the first records of *O. percrassa* from the Department of Valle del Cauca, Colombia (see Castro-Herrera and Vargas-Salinas 2008; Cardona-Botero et al. 2013), based on nine individuals collected 25 January 2014 at the sector of La Mina, Páramo of Chili, municipality of Sevilla (3°58’39.83”N, 75°49’38.46”W; 3,053 m; Table 2; Figs. 1–2). Thus, the current distribution of *O. percrassa* in the Cordillera Central extends from northern Department of Antioquia, in the Páramo of Sonsón (Loaiza-Piedrahíta et al. 2014), to the south, in the Departments of Tolima and Valle del Cauca, including the new record at the Páramo of Barragán and Chili. The new record extends 65 km south the original distribution range. The remaining 14 records fill geographic gaps along the Cordillera Central in the Andes of Colombia between the departments of Antioquia and Valle del Cauca.

The ecological niche model (Fig. 3) predicted a larger area of suitable habitat than was previously assumed (low omission rates). The model connects the *O. percrassa* distribution from the localities of the north at the Department of Antioquia (Páramo de Sonsón) with

TABLE 3. Prey composition of the diet of *Osornophryne percrassa*. Abbreviations are Vol = Volume in mm^3, % Vol = percentage volume, N = number prey, % N = percentage prey, and F = frequency of occurrence.

Prey	N	% N	Vol	% Vol	F
ARTHROPODA					
ARACHNIDA					
ACARI	8	9.8	6.0	0.2	5
ARANEAE	6	6.2	14.4	0.6	5
PSEUDOSCORPIONIDA	4	4.9	7.2	0.3	3
MIRIAPODA					
Diplopoda	5	6.1	23.2	0.8	3
Chilopoda	1	1.2	1.6	0.1	1
INSECTA					
COLEMBOLLA	1	1.2	16.4	0.6	1
COLEOPTERA					
Carabidae	5	6.1	313.7	11.4	3
Chrysomelidae	1	1.2	19.3	0.7	1
Curculionidae	13	15.9	178.8	6.5	8
Staphylinidae	4	4.9	7.0	0.3	4
Tenebrionidae	2	2.4	14.2	0.5	2
Undetermined	8	9.8	104.9	3.8	7
DERMAPTERA	2	2.4	173.9	6.3	2
DIPTERA	1	1.2	1.2	0.0	1
HEMIPTERA					
Tingidae	1	1.2	6.8	0.2	1
Undetermined	2	2.4	38.6	1.4	2
HYMENOPTERA	3	3.7	6.5	0.2	3
LEPIDOPTERA	1	1.2	32.5	1.2	1
ORTHOPTERA	1	1.2	76.3	2.8	1
ISOPODA	5	6.1	1,689.6	61.5	5
NEMATODA	9	11.0	16.6	0.6	8
Total	82	100	2748.5	100	14

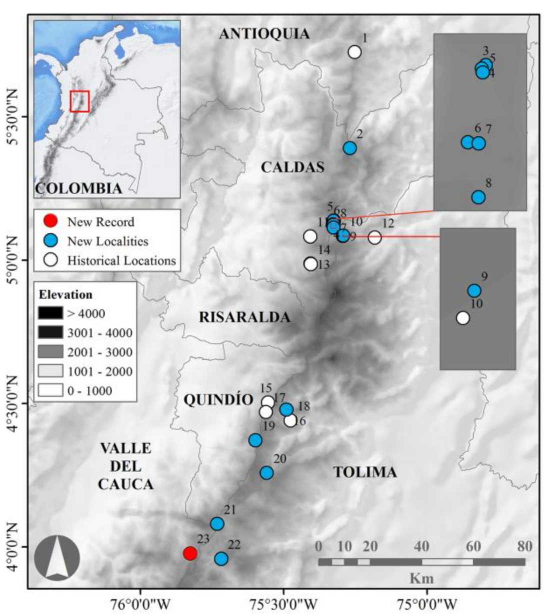

FIGURE 2. Distribution of *Osornophryne percrassa* in the Cordillera Central of Colombia. Details of the localities are summarized in Table 2.

with those in the south at the Department of Valle del Cauca. The validation test indicated a good performance by the model with an AUC value of 0.996 and a Jackknife test with high success rates using Maxent (MTP) and a statistical significance of $P < 0.001$. Likewise, Annual Mean Temperature (BIO1) was the variable with the largest predictive power (47.3%).

Diet.—We examined stomach contents of 14 female *O. percrassa* (mean SVL = 33.55 ± 4.33; range 23.7–38.8 mm). We found prey in each stomach and isopods were volumetrically the most important prey, followed by ground coleopterans (Carabidae) with other prey items found in smaller amounts (Table 3). Coleopterans were numerically the most important prey; the weevils (Curculionidae) were numerically the most important family (Table 3). Coleopterans were the most common prey because they were found in the 85% of individuals and represented 23.2% of volume. Other prey types as isopods were less common in the diet but because of their large size, are the most important by volume (Table 3). For the individuals examined, the mean number of prey items consumed was 5.4 ± 8.03. The dietary niche

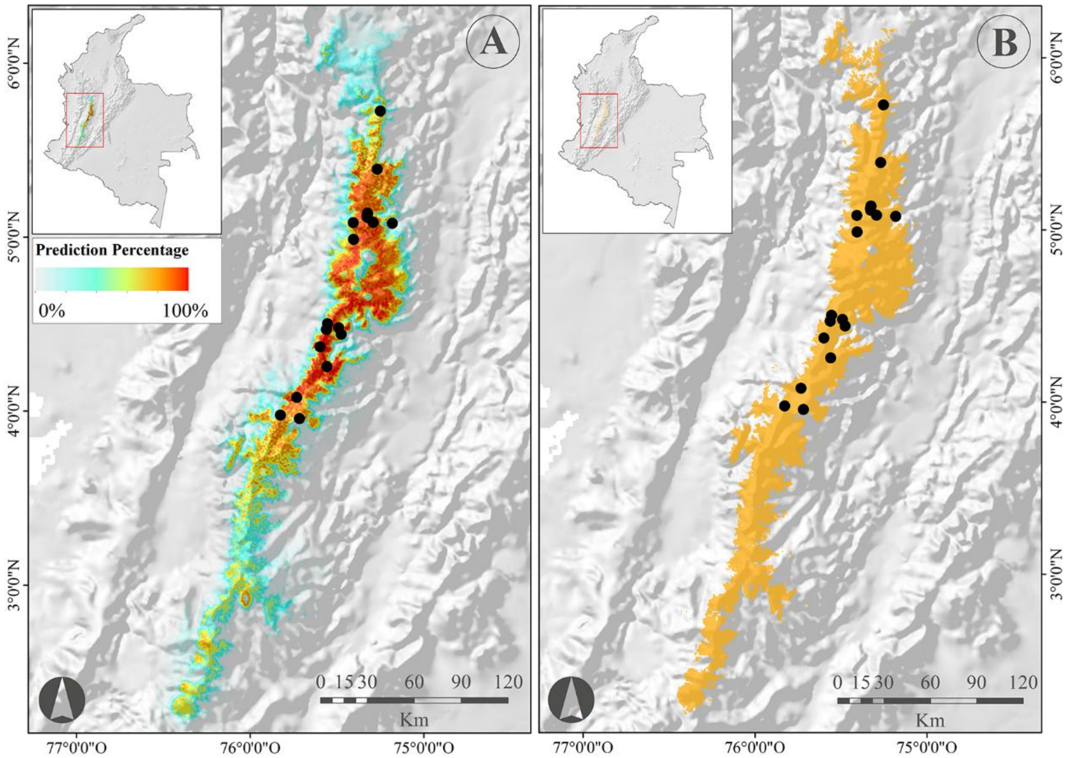

FIGURE 3. Modeled ecological niche of *Osornophryne percrassa* using Maxent. (A) Continuous ecological model. (B) Binary model applying a Minimum training presence. Black circles indicate presence records.

breadth (H') was 2.09.

The metrics SVL and HW, and SVL and HL were positively and significantly correlated (Table 4). We also found a significant correlation between HL and HW (Table 4). We did not find a significant correlation between any of the prey variables (number of prey, volume of prey, prey width, prey length) with any of the variables of the predator size (SVL, HL and HW; Table 4).

***Vocalizations*.—**Based on 23 advertisement calls and 11 courtship calls recorded from one male, the advertisement call of *O*. *percrassa* is composed of 8 to 10 Peep notes, but the calls have seven to nine grouped notes and one single endnote (Fig. 4). Mean note duration was 3.768 ± 1.045 ms (range, 0.2–6 ms) and notes were separated by 28.85 ± 5.57 ms (range, 20–39 ms) silent intervals. Mean call duration was 0.293 ± 0.011 s (range, 0.27–0.31 s) and, as expected, call duration increased with the number of notes per call. The mean silent interval between calls was 1.77 ± 0.26 s (range, 1.33–2.3 s). The mean dominant frequency of the call was 2.552 ± 0.326 kHz (range, 2.018–3.121 kHz), the mean low frequency was 1.085 ± 0.088 kHz (0.923–1.333 kHz) and the mean high frequency was 12.533 ± 0.449 kHz (11.769–13.397 kHz).

The courtship call of *O*. *percrassa* consists of six to nine Peep notes (Fig. 5). Mean note duration was 159.01 ± 29.10 ms (range, 106–214 ms) and consecutive notes were separated by 158.13 ± 23.26 ms (range, 121–200 ms) silent intervals. Mean call duration was 2.233 ± 0.260 s (range, 1.899–2.801 s) and, as expected, call duration increased with the number of notes per call. The mean duration of silent intervals between calls was 7.796 ± 2.201 s (range, 5–10.97 s). The mean dominant frequency of the call was 2.4 ± 0.139 kHz (range, 2.12–2.589 kHz), the mean low frequency was 1.315 ± 0.254 kHz (range, 0.773–1.518 kHz) and the mean high frequency was 11.617 ± 1.359 kHz (range, 7.818–12.959 kHz).

DISCUSSION

***Distribution and modeling niche*.—**Loaiza-Piedrahíta et al. (2014) suggest that the conservation status of *O. percrassa* should be re-evaluated. Our data substantiates that suggestion. The discovery of new localities, increase in known distribution, confirmation of geographical connectivity between different localities, and availability of potentially suitable habitat as determined by our prey consumed was 5.4 ± 8.03. The dietary niche all indicate that the plight of *O. percrassa*

TABLE 4. The Spearman Rank Multiple Correlation upper diagonal are correlation coefficients and lower diagonal are corresponding *P* values. Abbreviations are N = Number of prey, V = volume prey, PW = prey width, PL = prey length, SLV = snout vent length, HW = head width, and HL = head length.

	SVL	HW	HL	N	PW	PL	V
SVL	*****	0.932	0.587	0.180	0.312	0.097	0.152
HW	< 0.001	*****	0.626	0.198	0.194	0.149	0.007
HL	0.027	0.017	*****	0.584	0.218	0.198	0.225
N	0.538	0.498	0.028	*****	0.184	0.513	0.268
PW	0.278	0.506	0.454	0.529	*****	0.304	-0.442
PL	0.742	0.611	0.497	0.061	0.291	*****	-0.133
V	0.604	0.982	0.440	0.354	0.114	0.650	*****

is not as dire as previously thought and a listing of Vulnerable might be more appropriate than a listing of Endangered.

Habitat loss and fragmentation from agricultural activities should also be evaluated (Vanegas-Guerrero and Fernández-Roldán 2014) because *O. percrassa* is restricted to high altitudes in ecosystems such as Paramos and cloud forests on the Cordillera Central. Furthermore, aspects of its natural history, including semifossorial habit, inability to jump quickly, direct development, and need of sites with high humidity for oviposition (Ruíz-Carranza and Hernández-Camacho 1976, Mueses-Cisneros 2003) also jeopardize the species. Furthermore, during a 20 year period (1985 to 2005) there has been 6.8% habitat loss in Páramos coverage, as a result of anthropogenic activities (Sarmiento et al. 2013), as well as an increase of 1° C in temperature per decade in Páramos and 0.3–0.6° C in areas of subpáramo and high Andean forest increasing precipitation (Urbina-Cardona 2011). Thus, these modifications may directly affect the population ecology of the species and could, in the long term, result in local extinctions (Urbina-Cardona 2011; Botello et al. 2015).

The niche model substantiates the restriction of *O. percrassa* to the Cordillera Central, because during the lower Tertiary, Colombian mountains had reached 500 m elevation (Urbina-Cardona 2011). The Cordillera Central was the first mountain range (Upper Cretaceous) of Columbia, rising to high elevations 18–22 million years ago (Miocene), forming open vegetation areas such as ridges, summits, and peatlands, that are known as Protopáramos (Cortés-Duque and Sarmiento 2013). During the formation of the mountain chains, the glacial cycles generated contractions and expansions of the populations, causing diversification and extinction of the lineages (Van der Hammen et al. 1973; Navas 2006).

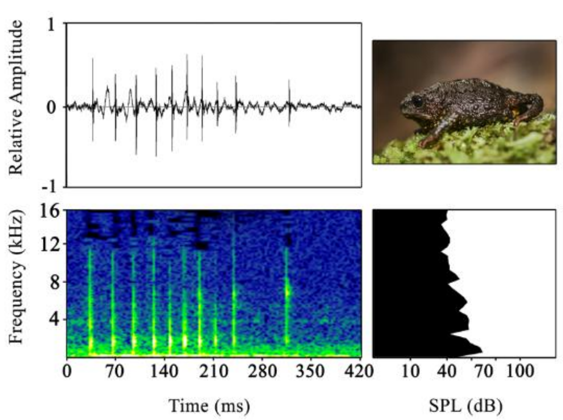

FIGURE 4. Graphic representations of an advertisement call of *Osornophryne percrassa*. (Photographed by Jhonattan Vanegas). Upper: An oscillogram showing 10 notes. Notice how the last note is separated from the rest of the notes. Below: spectrogram of the advertisement call (left) and power spectrum (right) of the call.

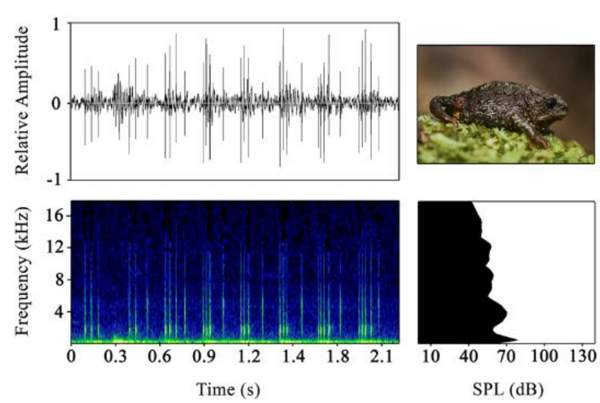

FIGURE 5. Graphic representations of a courtship call of *Osornophryne percrassa*. (Photographed by Jhonattan Vanegas). Upper: An oscillogram showing eight notes. Below: spectrogram of the courtship call (left) and power spectrum (right) of the call.

The relatively short but very active geological history of the Andean region suggests that speciation in the genus *Osornophryne* must have been fast and more recent, apparently directed by Quaternary climatic fluctuations (Yánez-Muñoz et al. 2010), with temperature a major determining factor on the distribution and occupation of new habitats (Navas 2003).

Aspects aforementioned may explain what the ecological niche model estimated was suitable habitat to meet the ecological and physiological requirements of *O. percrassa* on the Cordillera Central. This chain connects the distribution of the species from north to south on the Central Andes of Colombia. *Osornophryne percrassa* occupies a wide extension in Páramos and high Andean forest, where greater physiological barriers, in climatic terms, do not exist. The model also showed suitable habitat on the Cordillera Occidental, but because of the Páramo in the Cordillera Occidental has never been connected to the Cordillera Central, there is a series of isolated areas that could prevent the spread of the species (Lynch et al. 1997). We expect that the model shown here can help focus investigations on the critical areas of higher probability of occupancy of the species.

Diet.—The only report published on the diet of *O. percrassa* is that of Ruiz-Carranza and Hernández-Camacho (1976), who mention weevils (Curculionidae) as prey. Our data show that *O. percrassa* feeds on a variety of arthropods, mainly insects, similar to many species of anurans (Parmelee 1999). An arthropod diet based primarily on insects is not unusual for a terrestrial frog such as *O. percrassa* because of the abundance of insects that occur in terrestrial habitats (Parmelee 1999; Triplehorn and Triplehorn 2005). Furthermore, the types of arthropods in the diet can provide information on the habitat preference of the predator. For example, isopods, carabids, staphylinids, and tenebrionids in the diet suggest that *O. percrassa* inhabits leaf litter and other secretive habitats. The diet has been analyzed for several other Osornophryne species, including *O. antisana*, *O. guacamayo*, *O. puruanta*, *O. sumacoensis* (Gluesenkamp 1995; Gluesenkamp and Acosta 2001; Gluesenkamp and Guayasamin 2008). Arthropods form the bulk of the diet in each of these species. In particular, isopods, coleopterans, hymenopterans, hemipterans, salpugids, araneida, and apocrita are commonly ingested by these species. Coleopterans and hymenoptera are particularly abundant in the diets of *Osornophryne* species (Gluesenkamp 1995; Gluesenkamp and Acosta 2001; Gluesenkamp and Guayasamin 2008). Based on volume, isopods form an important component of the diet in *O. percrassa* and possibly other species; unfortunately, there is no information about volume occupied in the stomach by each kind of prey for other species (Gluesenkamp 1995; Gluesenkamp and Acosta 2001; Gluesenkamp and Guayasamin 2008). Gluesenkamp and Acosta (2001) suggest that ants, coleopterans, and crickets could be a source of precursor alkaloids used in the production of secretions from poison glands. However, the toxins present in this species are probably tetradotoxins as in *Atelopus*. If so, then their toxicity is not dependent on diet (Kim 1975; Pavelka et al 1977; Mebs et al. 1995; Hanifin 2010).

The numeric and volumetric dominance of coleopterans reported here is not different from the diet of other bufonids with secretive behavior, such as *Amazophrynella*, *Melanophryniscus* and *Rhinella* (Toft 1981; Parmelee 1999; Bonansea and Vaira 2007; Santana and Juncá 2007; Sabagh and Rocha 2012). The rare occurrence of ants in the diet of *O. percrassa*, unlike other bufonids, may be associated with the poor diversity of ants in the highlands (Fernández and Sharkey 2006). The limited consumption of other prey types also might be a result of the limited availability (Rodrigues et al. 2004).

Parmelee (1999) suggested that body size and head width in frogs determines the maximum size of prey that can be consumed. Thus, the ability of gaping in relation to the size of the frog is a limiting factor in the selection of prey (Toft 1981). In this study, SVL, HW, and HL did not determine the number and volume of prey consumed, suggesting that the prey size is not influencing the type of prey ingested by *O. percrassa*. However, this hypothesis should be reassessed with a larger sample, including both sexes, and surveying in different localities. The wide variety of prey types and sizes found in *O. percrassa* indicates that the species is a generalist/opportunistic feeder with a sit-and-wait strategy for obtaining prey, like others bufonids species (Taigen et al. 1982; Strüssmann et al. 1984; Parmelee 1999; Bonansea and Vaira 2007).

Vocalization.—Our understanding of the ecology and evolution of anuran communication is far from complete (Toledo et al. 2015). For example, recent evidence has revealed ultrasonic (Arch et al. 2008) and seismic communication in arboreal and aquatic species (Forti and Encarnação 2012). All species of *Osornophryne* toads lack hearing structures (Páez-Moscoso et al. 2011; Páez-Moscoso and Guayasamin 2012). The use of acoustic communication, such as courtship calls in *Osornophryne* toads, is surprising. Possibly these toads also rely on other mechanisms of communication, such as ultrasonic, seismic, or visual cues.

Despite important advances in understanding phylogenetic relationships among the species of *Osornophryne* (Páez-Moscoso et al. 2011; Páez-Moscoso and Guayasamin 2012), many uncertainties persist regarding phylogenetic affinities and taxonomic limits of the species (Páez-Moscoso and Guayasamin 2012). Advertisement calls are a useful character for species delimitation (Grant et al. 2006). Descriptions of the advertisement calls of other *Osornophryne* species should improve our understanding of the relationships among those species. It is important to highlight that the advertisement call has been described in *O. percrassa* by us and for *O. guacamayo* by Gluesenkamp and Acosta (2001). The advertisement call of *O. percrassa* is similar to that of *O. guacamayo* (Gluesenkamp and Acosta 2001); however, little more can be stated at this time because the description of the call of *O. guacamayo* (Gluesenkamp and Acosta 2001) and the advertisement and courtship calls of *O. percrassa* are each based on recordings of one individual. We are the first to describe the courtship call of any *Osornophyrne* toad. However, as with the advertisement call, the description of the courtship call is based on one individual, and therefore, a larger sample size is needed to improve descriptions of the courtship call in *O. percrassa*.

*Acknowledgments.***—**We thank Margarita M. López-García for help in identifying stomach contents, Fernando Vargas-Salinas, Julián A. Velásco, Mariane Targino and Paola M. Triviño for valuable comments that improved the manuscript, Fernando Castellanos, Jonathan Castellanos, Cristiam Fernandez and Susana López for field support, and Hector Fabio Arias and Viviana Andrea Ramírez for access to the Museo de Historia Natural de la Universidad de Caldas. In addition, we are grateful to John D. Lynch for comments on the manuscript and for access to the laboratory of amphibians at the Instituto de Ciencias Naturales (ICN), Universidad Nacional of Colombia, Wolfgang Buitrago for providing valuable information on the distribution of the species in Quindío Department, and Jaider Manuel Peña Cerpa for providing valuable information on the records of the species deposited in the zoological collection of the Universidad del Tolima.

LITERATURE CITED

Acosta Galvis, A.R. 2015. Lista de los Anfibios de Colombia: Reference online V.05.2015.0 (Fecha de acceso). Available at at http://www.batrachia.com. (Accessed 7 February 2016).

Arch, V.S., T.U. Grafe, and P.M. Narins. 2008. Ultrasonic signaling by a Bornean frog. Biology Letters 4:19–22.

Bernal, M.H., and A. Quevedo. 2004. Sapito de páramo. *Osornophryne percrassa*. Pp. 356−359 *In* Libro Rojo de los Anfibios de Colombia. Rueda-Almonacid, J.V., J.D. Lynch, and A. Amézquita (Eds). Conservación Internacional Colombiana, Instituto de Ciencias Naturales, Universidad Nacional, Ministerio del Medio Ambiente, Bogotá, Colombia.

Bioacoustics Research Program. 2011. Raven Pro: Interactive sound analysis software (Version 1.4). The Cornell Lab of Ornithology, Ithaca, New York, USA.

Bolívar, W., and J.D. Lynch. 2004. IUCN Red List of Threatened Species. *Osornophryne percrassa*. Version 2013.2. Electronic database at http://www.iucnredlist.org.

Bonansea, M.I., and M. Vaira. 2007. Geographic variation of the diet of *Melanophryniscus rubriventris* (Anura: Bufonidae) in northwestern Argentina. Journal of Herpetology 41:231−236.

Botello, F., S. Sarkar, and V. Sánchez-Cordero. 2015. Impact of habitat loss on distributions of terrestrial vertebrates in a high-biodiversity region in Mexico. Biological Conservation 184:59−65.

Burbano-Yandi, C.E., W. Bolívar-G., D.A. Velásquez-Trujillo, A.M. González-Colorado and O. Ospina-Herrera. 2015. Plan de conservación de la especie endémica de colombia sapito de páramo *Osornophryne percrassa*, en el departamento de Caldas. Corpocaldas, Manizales, Colombia.

Cardona-Botero, V.E., R.A. Viáfra-Vega, A. Valencia-Zuleta, A. Echeverry-Bocanegr, O.D. Hernández-Córdoba, A.F. Jaramillo-Martínez, R. Galvis-Cruz, J.A. Gutiérrez, and F. Castro-Herrera. 2013. Diversidad de la herpetofauna en el Valle del Cauca (Colombia): un enfoque basado en la distribución por regiones, altura y zonas de vida. Biota Colombiana 14:156−233.

Castro-Herrera, F., and F. Vargas-Salinas. 2008. Anfibios y reptiles en el departamento del Valle del Cauca, Colombia. Biota Colombiana 9:251−277.

Cocroft, R.B., and M.J. Ryan. 1995. Patterns of advertisement call evolution in toads and chorus frogs. Animal Behavior 49:283−303.

Cortés-Duque, J., and C. Sarmiento (Eds.). 2013. Visión Socio Económica de los Páramos y Alta Montaña Colombiana: Memorias del Proceso de Definición de Criterios para la Delimitación de Páramos. Instituto de Investigación de recursos Biológicos Alexander von Humboldt. Bogotá D.C., Colombia.

Escobar-Lasso, S., and G.A. González-Duran. 2012. Strategies employed by three Neotropical frogs (Amphibia: Anura) to avoid predation. Herpetology Notes 5:79−84.

ESRI. 2011. ArcGIS Desktop: Release 10.1. ESRI. Redlands, California, USA.

Fernández, F., and M.J. Sharkey (Eds.). 2006. Introducción a los Hymenoptera de la region Neotropical. Universidad Nacional de Colombia, Bogotá D.C., Colombia.

Forti, L., and L. Encarnação. 2012. Water-wave production in the Neotropical frogs *Physalaemus albonotatus* and *Pseudopaludicola mystacalis*: a seismic signal? Salamandra 48:181–184.

Frost, D.R. 2015. Amphibian Species of the World: An Online Reference. Version 6.0. Available at http://research.amnh.org/herpetology/amphibia/index.html. (Accessed 2 February 2016).

Grant, T., D.R. Frost, J.P. Caldwell, R. Gagliardo, C.F.B. Haddad, P. Kok, D.B. Means, B.P. Noonan, W.E. Schargel, and W. Wheeler. 2006. Phylogenetic systematics of dart-poison frogs and their relatives (Amphibia: Athesphatanura: Dendrobatidae). Bulletin of the American Museum of Natural History 299:1−262.

Gluesenkamp, A.G. 1995. A new species of *Osornophryne* (Anura: Bufonidae) from Volcán Sumaco, Ecuador, with notes on other members of the genus. Herpetologica 51:268–279.

Gluesenkamp, A., and N. Acosta. 2001. Sexual dimorphism in *Osornophryne guacamayo* with notes on natural history and reproduction in the species. Journal of Herpetology 35:148–151.

Gluesenkamp, A.G., and J.M. Guayasamin. 2008. A new species of *Osornophryne* (Anura: Bufonidae) from the Andean highlands of northern Ecuador. Zootaxa 1828:18–28.

Hanifin, C.T. 2010. The chemical and evolutionary ecology of tetrodotoxin (TTX) toxicity in terrestrial vertebrates. Marine Drugs 8:577–593.

Hijmans, R.J., S.E. Cameron, J.L. Parra, P. Jones, and A. Jarvis. 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25:1965−1978.

Hutter, C.R., S. Escobar-Lasso, J.A. Rojas-Morales, P.D.A. Gutiérrez-Cárdenas, H. Imba, and J.M. Guayasamin. 2013. The territoriality, vocalizations and aggressive interactions of the Red-spotted Glassfrog, *Nymphargus grandisonae*, Cochran and Goin, 1970 (Anura: Centrolenidae). Journal of Natural History 47:47−48.

Kim, Y.H., G.B., Brown, and F.A. Mosher. 1975. Tetrodotoxin: occurrence in atelopid frogs of Costa Rica. Science 189:151–152.

Llano-Mejía, J., A.M. Cortés-Gómez, and F. Castro-Herrera. 2010. Lista de anfibios y reptiles del departamento del Tolima, Colombia. Biota Colombiana 11:89−106.

Loaiza-Piedrahíta, J.D., S. Cuartas-Villa, I. Pérez, C.M. Marín-Castaño, and J.M. Daza. 2014. First record of *Osornophryne percrassa* Ruíz-Carranza & Hernández-Camacho, 1976 (Anura, Bufonidae) from the Department of Antioquia, Colombia. Herpetology Notes 7:673−674.

Lynch, J.D., P.M. Ruiz-Carranza, and M.C. Ardila-Robayo. 1997. Biogeographic patterns of Colombian frogs and toads. Revista de la Academia Colombiana de Ciencias 21:237−248.

Mebs, D., M. Yotsu-Yamashita, T. Yasumoto, S. Lötters, and A. Schlüter. 1995. Further report of the occurrence of tetrodotoxin in *Atelopus* species (family: Bufonidae). Toxicon 33:246–249.

Mueses-Cisneros, J.J. 2003. El género *Osornophryne* (Amphibia: Bufonidae) en Colombia. Caldasia 25:419−427.

Navas, C.A. 2003. Herpetological diversity along Andean elevational gradients: links with physiological ecology and evolutionary physiology. Comparative Biochemistry and Physiology, Part A 133:469−485.

Navas, C.A. 2006. Patterns of distribution of anurans in high Andean tropical elevations: insights from integrating biogeography and evolutionary physiology. Integrative and Comparative Biology 46:82−91.

Páez-Moscoso, D.J., and J.M. Guayasamin. 2012. Species limits in the Andean toad genus *Osornophryne* (Bufonidae). Molecular Phylogenetics and Evolution 65:805−822.

Páez-Moscoso, D.J., J.M. Guayasamin, and M. Yánez-Muñoz. 2011. A new species of Andean toad (Bufonidae, *Osornophryne*) discovered using molecular and morphological data, with a taxonomic key for the genus. ZooKeys 108:73−97.

Parmelee, J.R. 1999. Trophic ecology of a tropical anuran assemblage. Scientific Papers Natural History Museum the University of Kansas 11:1−59.

Pavelka, L.A., Y.H. Kim, and H.S. Mosher. 1977. Tetrodotoxin and tetrodotoxin-like compounds from the eggs of the Costa Rican frog, *Atelopus chiriquiensis*. Toxicon 15:135–139.

Pearson, R.P., C.J. Raxworthy, M. Nakamura, and A.T. Peterson. 2007. Predicting species distributions from small numbers of occurrence records: a test case using cryptic geckos in Madagascar. Journal of Biogeography 34:102−117.

Phillips, S.J., R.P. Anderson, and R.E. Schapire. 2006. Maximum entropy modelling of species geographic distributions. Ecological Modelling 190:231−259.

Rodrigues, D.J., M. Uetanabaro, and C.P. Prado. 2004. Seasonal and ontogenetic variation in diet composition of *Leptodactylus podicipinus* (Anura, Leptodactylidae) in the southern Pantanal, Brazil. Revista Española de Herpetología 18:19−28.

Rojas-Morales, J.A., H.F. Arias-Monsalve, and G.A. González-Durán. 2014. Anfibios y reptiles de la región centro-sur del departamento de Caldas, Colombia. Biota Colombiana 15:73−93.

Ruiz-Carranza, P.M., and J.I. Hernández-Camacho. 1976. *Osornophryne*, género nuevo de anfibios bufónidos de Colombia y Ecuador. Caldasia 6:93−148.

Sabagh, L.T., and C.F.D. Rocha. 2012. Diet of the toad *Rhinella icterica* (Anura: Bufonidae) from Atlantic Forest Highlands of southeastern Brazil. Biota Neotropica 12:258−262.

Santana, A.S., and F.A. Juncá. 2007. Diet of *Physalaemus* cf. *cicada* (Leptodactylidae) and *Bufo granulosus* (Bufonidae) in a semideciduous forest. Brazilian Journal of Biology 67:125−131.

Sarmiento, C., C. Cadena, M. Sarmiento, J. Zapata, and O. León. 2013. Aportes a la Conservación Estratégica de los Páramos de Colombia: Actualización de la Cartografía de los Complejos de Páramo a Escala 1:100.000. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt. Bogotá D.C., Colombia.

Strüssmann, C., M.B.R. do Vale, M.H. Meneghini, and W.E. Magnusson. 1984. Diet and foraging mode of *Bufo marinus* and *Leptodactylus ocellatus*. Journal of Herpetology 18:138−146.

Taigen, T.L., S.B. Emerson, and F.H. Pough. 1982. Ecological correlates of anuran exercise physiology. Oecologia 52:49−56.

Toft, C.A. 1981. Feeding ecology of Panamanian litter anurans: patterns in diet and foraging mode. Journal of Herpetology 15:139−144.

Toledo, L. F., I.A. Martins, D.P. Bruschi, M.A. Passos, C. Alexandre, and C.F.B. Haddad. 2015. The anuran calling repertoire in the light of social context. Acta Etológica 18:87–99.

Triplehorn, C.A., and N.F. Triplehorn. 2005. Borror's Introducción to the Study of Insects. 7th Edition. Thomson Brooks/Cole, Belmont, California, USA.

Urbina-Cardona, J.N. 2011. Gradientes andinos en la diversidad y patrones de endemismo en anfibios y reptiles de Colombia: posibles respuestas al cambio climático. Revista Facultad de Ciencias Básicas 7:74−91.

Van Der Hammen, T., J.H. Verner, and H. Van Dommelen. 1973. Palynological record of the upheaval of the Northern Andes: a study of the Pliocene and lower Quaternary of the Colombian Eastern Cordillera and the early evolution of its High-Andean biota. Review of Palaeobotany and Palynology 16:1−122.

Velásquez, E., B.F. Castro, W. Bolívar-G, and M.I. Herrera. 2008. Infección por el hongo quitrido *Batrachochytrium dendrobatidis* en anuros de la Cordillera Occidental de Colombia. Herpetotrópicos 4:65−70.

Vanegas-Guerrero, J., and J.D. Fernández-Roldán. 2014. *Osornophryne percrassa* Ruiz-Carranza y Hernández-Camacho 1976. Catálogo de Anfibios y Reptiles de Colombia 2:21−24.

Yánez-Muñoz, M.H., M. Altamirano-Benavides, D.F. Cisneros-Heredia, and A.G. Gleusenkamp. 2010. Nueva especie de sapo andino del género *Osornophryne* (Amphibia: Bufonidae) del norte de Ecuador, con notas sobre la diversidad del género en Colombia. Avances 2:B46−B53.

JHONATTAN VANEGAS-GUERRERO is a professional Biologist from Universidad del Quindío (Colombia). He is member of the herpetology group of the Universidad del Quindío. His research interests include taxonomy, thermal ecology, ecomorphology, and biogeography of Neotropical amphibians and reptiles. (Photographed by Jhonattan Vanegas).

GUSTAVO A. GONZALEZ-DURAN is a Biologist from Universidad de Caldas (Colombia), and is currently working on his Master's thesis at the Universidad Nacional de Colombia in Bogota. His research as a biologist is related to morphology, systematics, and conservation of amphibians and reptiles in the Andean region of Colombia, with emphasis on the evolution and taxonomy of Brachycephaloids and the behavior of snakes. (Photographed by Gustavo A. Gonzalez).

SERGIO ESCOBAR-LASSO is a professional Biologist from University of Caldas (Colombia) and currently he is working on his Master's degree in Conservation and Wildlife Management at the International Institute for Conservation and Wildlife Management (ICOMVIS) of the National University of Costa Rica. During his career as a biologist, he has had the opportunity to implement research projects related to conservation and wildlife management (see scientific publications in https://independent. academia.edu/SergioEscobar). His research interests include biogeography, ecology, and natural history of terrestrial vertebrates. He is the founder and president of the R.A.N.A foundation (Restauración de Ambientes Neotropicales Alterados), an organization dedicated to generate scientific knowledge in areas of ecology, zoology, and conservation biology. Furthermore, the foundation performs actions for conservation, ecological restoration, and defense of Neotropical ecosystems. (Photographed by Margarita Gil-Fernández).