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Short Communication

The advertisement call of *Pristimantis unistrigatus* (Anura: Strabomantidae) from the Ecuadorian Andes

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In anurans, advertisement calls are typically emitted during breeding seasons by adult males to attract females and to convey a territorial signal to conspecific males (Wells 2007, Toledo *et al*. 2014, Köhler *et al*. 2017). Vocalizations may provide species-specific information useful for taxonomic and systematic assessments because they act as a premating isolating mechanism and may thus carry a phylogenetic signal (Köhler *et al*. 2017). Moreover, studying the vocalizations of frogs and toads enhances a more comprehensive understanding about their population and distribution dynamics, allowing for the acoustic monitoring of targeted species (Teixeira *et al*. 2019).

Pristimantis unistrigatus (Günther, 1859), also known as "Cutín Quiteño", is a small frog (SVL: males = 14.9–28.8 mm; females = 22.5–38.5 mm) that occurs at high elevations (2200 and 3490 m a.s.l.) along the Andean region encompassed between southern Colombia and central Ecuador (Frost 2023). The species exhibits multiple cryptic coloration patterns throughout its range, but it typically has distinctive light brown colors with dark markings on the dorsum and a homogeneously white belly (Lynch 1981; Figure 1). Other characters such as the dorsum shagreened with scattered flat warts, venter areolate, without dorsolateral folds, tympanum partially concealed, and absence of tubercles on the heel help in its differentiation (Lynch 1981, Yánez-Muñoz *et al*. 2009, 2020, Ron *et al*. 2018). *Pristimantis unistrigatus* is most similar to *P. cajamarcensis* (Barbour and Noble, 1920) and *P. modipeplus* (Lynch, 1981), but *P. unistrigatus* is different from both of the aforementioned species (characters in parenthesis) because of the absence of prominent tubercles on the upper eyelid and on the dorsum (present), few supernumerary tubercles (many), and brown to yellow-colored posterior surfaces of the thighs (red).

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Figure 1. *Pristimantis unistrigatus* from different localities of Quito, Ecuador. Adult male (DHMECN 16826) from Mojanda (**A**). Unvouchered specimens from Verde Cocha (**B**), Tababela, Aeropuerto Internacional de Quito (**C**), and Parque La Carolina (**D**).

The species was initially included in the former *Eleutherodactylus unistrigatus* group, which was created by Lynch and Duellman (1980) to account for a great number of South American species based on phenetic similarities. *Pristimantis unistrigatus* was subsequently referred to the redefined *E. unistrigatus* group considering similarities in external adult morphology (e.g., granular venter, long fifth toe; see Lynch and Duellman 1997). Hedges *et al*. 2008 included sequences of the species from northern Ecuador and even though the *P. unistrigatus* species group (former *E. unistrigatus* group in Lynch and Duellman 1997) was demonstrated to be non-monophyletic, the authors retained the species and the species group as previously defined by Lynch and Duellman (1997). Follow-up phylogenetic studies did not include *P. unistrigatus* in any recognized species group within the genus *Pristimantis* (see Padial *et al*. 2014). More recently, García-Gómez *et al*. (2022) recognized

a redefined *P. unistrigatus* supported by molecular evidence and by the presence of nuptial pads as a morphological synapomorphy.

Pristimantis unistrigatus regularly calls during day and night in a wide variety of habitats, ranging from mesic undisturbed ecosystems (e.g., *Eucalyptus*–pasture habitat, paramo) to urbanized valleys and cities (Lynch and Duellman 1980; Lynch 1981, Yánez-Muñoz *et al*. 2009, 2020). This frog can be typically found either inside bromeliads that are close to the ground (< 3 m height) or hidden underground beneath rocks, logs, and/or construction materials (Lynch 1981, Ron *et al.* 2018). However, despite its relative abundance, information about the reproductive biology of the species remains scarce. Lynch and Duellman (1980) provided an onomatopoeic description of the call of *P. unistrigatus*, which they referred to as a "quonk" emitted in a series of 5–6 notes. Lynch (1981) later referred to the call of *P. unistrigatus* as a "hollow click" similar to the sound produced when rapping two hollow bamboo tubes together. However, the author emphasized the need to assess more acoustic data to better understand the taxonomy of the species in light of the geographic variation noted in terms of coloration, size, and vocal anatomy throughout contiguous Andean populations, and with respect to isolated populations at lower elevations (Lynch 1981). To date, information about the life history and bioacoustics of the species remains scarce as there is no formal characterization of the advertisement call of *P. unistrigatus*. Here we provide a quantitative and qualitative characterization of its advertisement call, accounting for temporal, spectral, and structural parameters.

On 06 November 2016 we recorded five calling males of *P. unistrigatus* between 19:00 and 20:00 h at 14°C in the outdoor gardens of the neighborhood "La Granja", located in the city of Quito, Pichincha Province, Ecuador (00°11'12" S, 78°29'54" W, 2860 m a.s.l.). We found the frogs calling close to the ground (< 50 cm height) on bromeliads and inside the cavities of rockery walls. To record the calls, we used a digital recorder Marantz PMD 661 and a unidirectional microphone Sennheiser ME 66 positioned 1–1.5 m from the calling individuals. Call recordings were made in digital format at a sampling rate of 44.1 kHz and a 16-bit resolution. We deposited digital copies of audio files in Fonoteca Zoológica (www.fonozoo.com), Museo Nacional de Ciencias Naturales, Consejo Superior de Investigaciones Científicas, Madrid, Spain (FZ14198–14201). Voucher specimens of *P. unistrigatus* from the study site (not collected by us) are housed in the collection of amphibians from the PUCE (QCAZA 40510, 65313; see Ron *et al*. 2018).

To characterize the vocalizations, we followed a note-centered approach (Köhler *et al*. 2017) considering quantitative [minimum frequency (Hz), maximum frequency (Hz), dominant frequency (Hz), call duration (s), note duration (s), number of notes per call, call rate (calls per minute), note rate (notes per second), inter-call interval (s), inter-note interval (s)] and qualitative variables (shape of the envelope of the call, pulse structure, amplitude modulation, and frequency modulation). We analyzed oscillograms and spectrograms in Raven Pro 1.5 (Bioacoustics Research Program 2014) with the following settings: window type Hann, window size of 512 samples, 3 dB filter bandwidth of 124 Hz, 50% overlap, hop size of 256 samples, DFT size of 512 samples, and grid spacing at 86.1 Hz. Spectrograms were used to calculate the minimum frequency, maximum frequency, and dominant frequency (Hz) with the Raven parameters of 'Frequency 5%', 'Frequency 95%', and 'Peak frequency', respectively. We used oscillograms to manually obtain the number of notes, and to measure the duration (s) of calls and notes and the silence intervals in between them using the function 'Delta Time'. We estimated the note/call rate as the total number of notes/calls divided by the time from the beginning of the first note/call to the end of the last one. We generated sound figures with the seewave package v. 1.7.6 (Sueur *et al*. 2008) in

R software 4.0.2 (R Development Core Team 2020). Spectrograms were made with a Hanning window function (512 bands FFT resolution) and a relative amplitude color scale of 30 dB (red = maximum amplitude).

We analyzed 15 calls and 211 notes, and a total average of 3 ± 3.39 (range 1–9) calls per individual and 18.2 ± 6.5 (8–26) notes per call. Calling males have a single subgular, highly distensible vocal sac and emit a trill composed of 4–26 non-frequency modulated, pulsatile notes (Figure 2). The call envelope is triangular-shaped with the amplitude peak located at the beginning of the call duration (Figure 2F). Call duration ranges from 1.1–8.2 s and note duration varies from 0.006–0.02 s. The minimum frequency of the call ranges from 1981–2670 Hz and the dominant frequency varies from 2153–2928 Hz. Maximum frequency varies from 2325–2928 Hz. Call rate ranges from 0.2–1 calls/min and the silence intervals between calls vary from 18.2–77.8 s. Note repetition rate ranges from 0.08–1.05 notes/s and the silence intervals between notes vary from 0.1–0.5 s. Table 1 shows information of the call parameters of each recorded individual.

The species is frequently heard calling incessantly for long periods along suitable calling sites across urban areas (e.g., gardens of households and apartment complexes), which makes it remarkable in terms of the behavioral transformations that allow it to occur in altered areas with high levels of disturbance. Given that acoustic signals are the most common and widely used means of communication among anurans, we speculate that the ambient noise produced by highways, businesses, and people (= anthropophony) may restrict the calling activity of the species (Vélez *et al*. 2013). We encourage the study of calls across populations with different levels of habitat disturbance as this could lead to accurate assessments of transformations and variation associated with the bioacoustics of urban adapted species.

Table 1. Measurements of acoustic parameters of the vocalization of *Pristimantis unistrigatus*. Minimum frequency (Min*F*), maximum frequency (Max*F*), dominant frequency (D*F*), call duration (CD), note duration (ND), note rate (NR), and internote interval (IN). Values are given as mean ± standard deviation (minimum–maximum) [number of measurements taken / number of calls]. *SVL of individuals 2 and 4 is 27.3 mm and 28 mm, respectively.

Individual	Min*F* (Hz)	Max*F* (Hz)	D*F* (Hz)	CD (s)	ND (s)	NR (notes/s)	IN (s)
1	2161 ± 26.5 (2153–2239) [20/1]	2403 ± 26.5 (2325–2411) [20/1]	2286 ± 43.9 (2239–2325) [20/1]	6.1 [1/1]	0.01 ± 0.002 (0.009–0.02) [20/1]	3.2 [20/1]	0.3 ± 0.006 (0.3–0.3) [10/1]
2*	2328 ± 97.4 (2153–2411) [33/2]	2756 ± 43.1 (2670–2842) [33/2]	2633 ± 71.5 (2325–2670) [33/2]	4.4 ± 1.3 (3.5-5.3) [2/2]	0.01 ± 0.002 (0.007–0.02) [33/2]	3.7 [33/2]	0.3 ± 0.02 (0.2–0.3) [10/2]
3	2396 ± 95.9 (1981–2497) [45/2]	2794 ± 118.3 (2497–2928) [45/2]	2545 ± 72.4 (2153–2584) [45/2]	7.1 ± 1.5 (6-8.2) [2/2]	0.01 ± 0.001 (0.009–0.02) [45/2]	3.1 [45/2]	0.3 ± 0.03 (0.3–0.4) [10/2]
4*	2499 ± 120.6 (2067–2584) [105/9]	2781 ± 102.6 (2497–2928) [105/9]	2630 ± 103.4 (2325–2756) [105/9]	4.1 ± 1.7 (1.1-6.8) [9/9]	0.01 ± 0.002 (0.006–0.02) [105/9]	2.9 ± 2.9 (2.5-3.7) [105/9]	0.3 ± 0.09 (0.1–0.4) [10/9]
5	2605 ± 39.8 (2583–2670) [8/1]	2917 ± 30.4 (2842–2928) [8/1]	2756 ± 46 (2670–2842) [8/1]	2.9 [1/1]	0.01 ± 0.001 (0.01–0.02) [8/1]	2.8 [8/1]	0.4 ± 0.06 (0.3–0.5) [7/1]

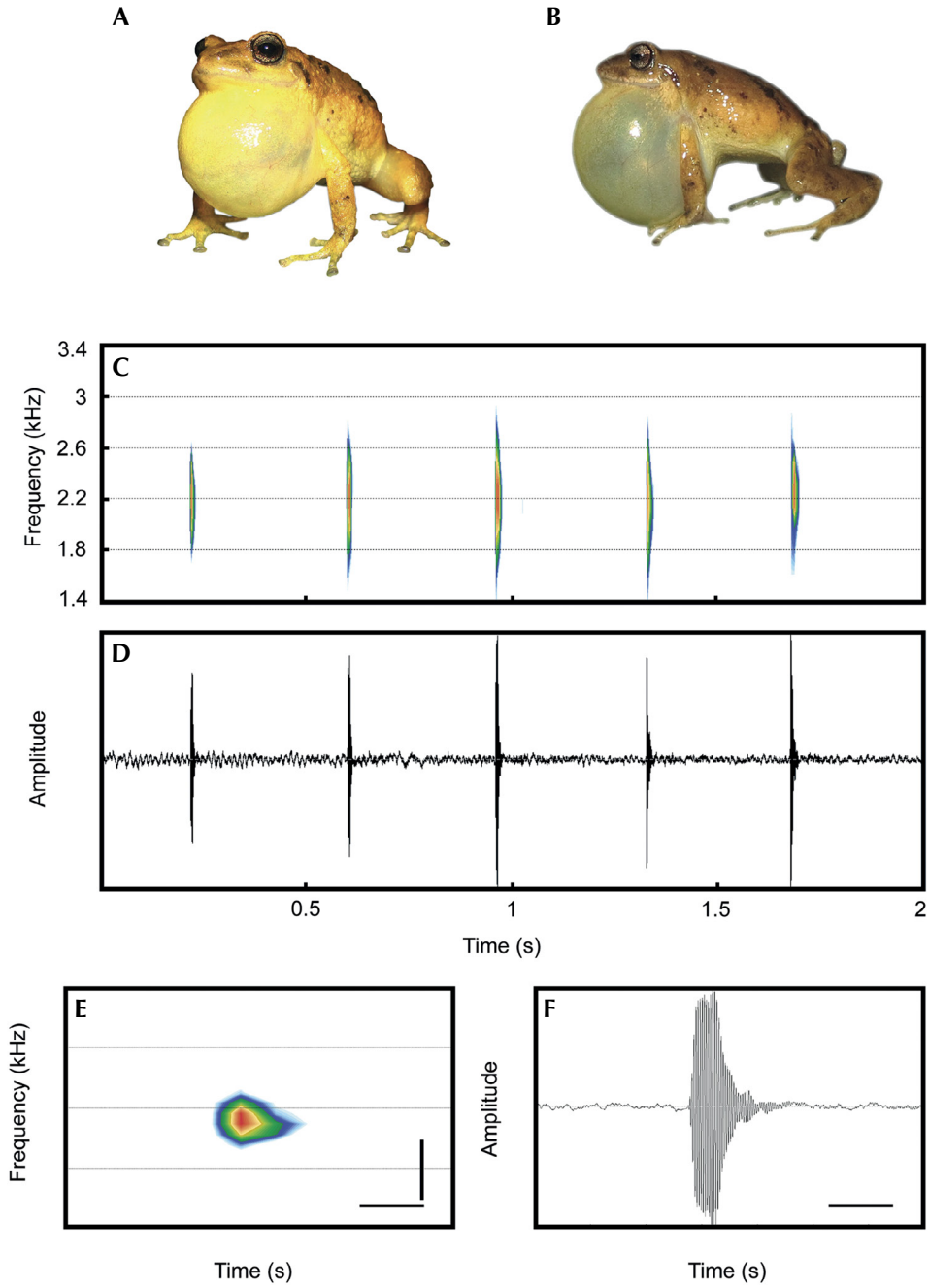

Figure 2. Advertisement call of *Pristimantis unistrigatus* from Quito, Ecuador. Adult calling male of *P. unistrigatus* showing its single, subgular highly distensible vocal sac (**A–B**). Spectrogram (**C**) and oscillogram (**D**) of the call. Detail of the spectrogram (**E**) and oscillogram (**F**) of a note. Horizontal scale bars have 0.02 s; vertical scale bars have 0.5 kHz.

This work is the first formal contribution on the vocalizations and calls of *Pristimantis unistrigatus,* despite being a species known for 150 years and one of the most common lineages in the Andes of Ecuador and Colombia. This situation is similar to most species of neotropical anurans (Rivera-Correa *et al*. 2021), of which no information is available on their bioacoustics or only anecdotal onomatopoeic information is available. We hope that this contribution will call the attention of other herpetological colleagues to generate and fill gaps in bioacoustic information on other common species of anurans in the Ecuadorian Andes.

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