Journal of Herpetology, Vol. 53, No. 2, 131–143, 2019

A New, Morphologically Cryptic Species of *Adenomera* Closely Related to *Adenomera araucaria* from the Atlantic Forest of Southern Brazil (Anura, Leptodactylidae)

THIAGO R. DE CARVALHO,[1,2] CARLA S. CASSINI,[3] PEDRO P. G. TAUCCE,[1] AND CÉLIO F. B. HADDAD[1]

[1]*Laboratório de Herpetologia, Departamento de Zoologia e Centro de Aquicultura, Universidade Estadual Paulista, Rio Claro, São Paulo, Brazil*
[3]*Laboratório de Herpetologia Tropical, Departamento de Ciências Biológicas, Universidade Estadual de Santa Cruz, Ilhéus, Bahia, Brazil*

ABSTRACT.—**We describe a new species of *Adenomera* from the Brazilian Atlantic Forest corresponding to one of the candidate species indicated in the published phylogeny of the genus ("sp. R"). The new species differs from all 18 congeners by the combination of small size, absence of toe discs, and the advertisement call composed of a single nonpulsed note with pronounced frequency upsweep. From the closest related and morphologically cryptic *Adenomera araucaria*, the new species is distinguished only by its nonpulsed advertisement call (multipulsed call in *A. araucaria*). The new species is distributed in the Serra do Tabuleiro mountain range and the Santa Catarina Island, as well as localities in between them in the eastern portion of the state Santa Catarina. This is the second *Adenomera* species endemic to the Atlantic Forest of eastern Santa Catarina in southern Brazil, sympatric to *Adenomera engelsi* and allopatric to the sibling *A. araucaria*.**

RESUMO.—**Descrevemos uma nova espécie de *Adenomera* da Mata Atlântica brasileira, que corresponde a uma das espécies candidatas indicadas na filogenia publicada para o gênero ("sp. R"). A nova espécie se distingue de todos os 18 congêneres pelo pequeno tamanho corporal, ausência de discos nos artelhos e canto de anúncio composto por um tipo único de nota não pulsada com modulação ascendente bem marcada. Em relação à espécie irmã e morfologicamente críptica *A. araucaria*, a nova espécie se diferencia apenas pelo canto não pulsado (canto multi-pulsionado em *A. araucaria*). A espécie está distribuída pela formação da Serra do Tabuleiro e na Ilha de Santa Catarina, bem como em localidades intermediárias entre elas no leste de Santa Catarina. Esta é a segunda espécie de *Adenomera* endêmica da Mata Atlântica do leste de Santa Catarina, na região sul do Brasil, ocorrendo em simpatria com *Adenomera engelsi* e em alopatria com a espécie irmã *A. araucaria*.**

The frog genus *Adenomera* is distributed throughout South America east of the Andes and comprises 18 named species, in addition to 20 molecular-based lineages (i.e., candidate species) of Fouquet et al. (2014). In this phylogenetic study, members of the *Adenomera marmorata* clade were indicated as endemic to the Brazilian Atlantic Forest. The distribution of nominal species in this clade reflects two different regions in the domain: *A. marmorata* and *Adenomera ajurauna* are distributed in southeastern Brazil, whereas *Adenomera araucaria*, *Adenomera bokermanni*, *Adenomera engelsi*, and *Adenomera nana* across southern Brazil. *Adenomera araucaria* has the southernmost range of the genus, extending from the extreme south of Brazil (Rio Grande do Sul), across Santa Catarina (Fouquet et al., 2014), to south-central Paraná (Conte et al., 2010).

Here, we describe a new species of *Adenomera* from the Atlantic Forest of eastern Santa Catarina (southern Brazil), morphologically cryptic and closely related to *A. araucaria*, based on morphological, acoustic, and molecular evidence. We assign the new species to one of the confirmed candidate species (*Adenomera* sp. R) of Fouquet et al. (2014) based on mtDNA sequences from type specimens. We provide additional data on coloration and vocalizations for *A. araucaria* and comment on the distribution patterns of *Adenomera* in southern Brazil.

MATERIALS AND METHODS

Study Area and Institutional Acronyms.—We focused field collections in four localities of the Brazilian state of Santa Catarina, as well as at the type locality of *A. araucaria*: (1) Morro das Pedras trail, at Serra do Tabuleiro State Park (27.821120°S, 48.903400°W; 1,063 m above sea level [a.s.l.]; datum WGS84), in São Bonifácio; (2) Faxinal do Beppe, at Serra do Itajaí National Park (27.111856°S, 49.211756°W; 659 m a.s.l.), in Indaial; (3) São Bento do Sul (26.323219°S, 49.307291°W; 646 m a.s.l.); (4) Jaraguá do Sul (26.514888°S, 49.061255°W; 544 m a.s.l.); (5) São Francisco de Paula (29.432780°S, 50.252310°W; 944 m a.s.l.), in the state of Rio Grande do Sul. Type specimens were deposited in the following Brazilian collections: CFBH (Coleção Célio F. B. Haddad, UNESP/Rio Claro, São Paulo), MCP (Museu de Ciências de Tecnologia, PUCRS/Porto Alegre, Rio Grande do Sul), and UFMG (Universidade Federal de Minas Gerais, Belo Horizonte, Minas Gerais). Other specimens examined are given in Appendix 1. Institutional abbreviations followed Sabaj (2016). For those not included there, abbreviations are AAG-UFU (Universidade Federal de Uberlândia, Minas Gerais), LHUFCG (Laboratório de Herpetologia da Universidade Federal de Campinas Grande, in Patos, Paraíba, Brazil).

Morphology.—We measured 11 morphometric traits with the use of an ocular micrometer (0.1-mm scale) fitted to a stereomicroscope (except snout–vent length [SVL], measured with digital calipers) previously calibrated with a standard ruler and double-checked with calipers. We measured the following traits: SVL, thigh length (THL), tibia length (TL), foot length (FL), hand length (HAL), head length (HL), head width (HW), eye diameter (ED), tympanum diameter (TD), eye–nostril distance (EN), and internarial distance (IND). Morphometric definitions and terminology are essentially according to Watters et al. (2016), except HAL, HL, and HW, which are defined as: HAL, the distance between the base of inner metacarpal tubercle to the tip of Finger III; HL, the distance between the tip of the snout and the midpoint (center) of the tympanum; and HW, the distance between the midpoints of tympani. Snout-shape terminology followed Heyer et al. (1990). Toe tip development was given according to Heyer (1973).

[2]Corresponding author. E-mail: thiago_decarvalho@yahoo.com.br
DOI: 10.1670/18-172

Recordings and Acoustic Analysis.—We recorded frog calls in the field with digital recorders (Marantz PMD 660, 661, 670; sampling rates: 44.1 or 48.0 kHz; sample size: 16 bits) and Sennheiser ME66/K6 or ME67/K6 unidirectional microphones. Recordings were stored as monochannel WAVE files. Sound recordings were deposited in the acoustic repositories of CFBH and AAG-UFU collections; we list information on the files in Appendix 2. We assigned some *Adenomera* populations to either *A. araucaria* or *A.* aff. *araucaria* based on acoustic-only evidence in the cases where there was not an associated call voucher: one recording housed at Fonoteca Neotropical Jacques Vielliard (FNJV; Appendix 2) and original recordings published in Kwet (2007), all recorded by this author (AK recordings; Appendix 2). We analyzed calls using an interface built between Soundruler (Gridi-Papp, 2007) and Matlab (Matlab, 2004). Acoustic traits were quantified through automated analysis, for which we developed settings in the software to recognize and delimit the acoustic units in the time domain (notes, pulses, intervals, and pulse rate) and in the frequency domain (fundamental and dominant frequencies, and frequency modulation). Grand means (and corresponding standard deviations) for "pulse duration" were obtained from mean values of pulse duration for each multipulsed call analyzed. We set these spectrogram parameters for *A. araucaria* and *A.* aff. *araucaria*: Fast Fourier transform (FFT) size = 1,024 samples, FFT overlap = 90%, window type = Hanning, contrast = 70%. Parameters for automated recognition were (in sample sizes): *A. araucaria*: detection (smoothing = 500, resolution = 1), delineation (smooth factor = 1, smoothing = 15, and resolution = 1), critical amplitude ratio = 0.8; *A.* aff. *araucaria*: detection (smoothing = 500, resolution = 1), delineation (smooth factor = 1, smoothing = 13, and resolution = 1), and critical amplitude ratio = −1 (disabled); we list acoustic definitions and terminology in Appendix 3. We manually quantified note rate per minute in Audacity v.2.1.1 (Audacity Team, 2017). We applied a 500-Hz high-pass filter to sound files in Soundruler prior to conducting the acoustic analysis to reduce background noise caused by wind and/or rain. We produced sound figures with the use of seewave v.2.1.0 (Sueur et al., 2008) and tuneR v.1.3.2 (Ligges et al., 2017), in R v.3.5.0 (R Core Team, 2018). Spectrogram settings were: window Hanning, FFT size = 256 samples, and FFT overlap = 90%; the level of frequency components were indicated by its darkness.

Molecular Data Acquisition and Analysis.—We extracted whole cellular DNA from 100% ethanol liver or muscle tissue using the Qiagen DNeasy® kit (Qiagen, Inc., Hilden, Germany) following manufacturer's protocols or a standard ammonium precipitation method (Lyra et al., 2017). Taxon sampling of molecular analysis included all named and confirmed candidate species in the *A. marmorata* clade plus one species from each one of the five remaining *Adenomera* clades (Fouquet et al., 2014; Appendix 4). We used *Lithodytes lineatus*, *Hydrolaetare caparu*, and *Leptodactylus rhodomystax* as outgroups. We chose a fragment of the *cytochrome c oxidase* subunit I (COI) to perform our analyses and the primers dgLCO1490 (GGTCAACAAATCATAAAGAYATYGG; Meyer, 2003), dgHCO2198 (TAAACTTCAGGGTGACCAAARAAYCA; Meyer, 2003), T3–AnF1 (ATTAACCCTCACTAAAGACHAAY-CAYAAAGAYATYGG; Lyra et al., 2017), and T7–AnR1 (AA-TACGACTCACTATAGCCRAARAATCARAADARRTGTTG; Lyra et al., 2017) to perform amplification of the selected fragment. DNA amplification and purification followed Lyra et al. (2017). We sent PCR products to Macrogen Inc. (Seoul, South Korea), where they were sequenced with a BigDye Terminator Cycle Sequencing Kit (v.3.0, Applied Biosystems, Foster City, California, USA) in an ABI 3730 automated DNA sequencer (Applied Biosystems).

We performed sequence alignment with the use of the MUSCLE algorithm (Edgar, 2004) in MEGA 7 (Kumar et al., 2016) under default parameters. We conducted the search for the best-fitting nuclear model and best partition scheme with PartitionFinder v.2.1.1 (Lanfear et al., 2017) with the use of the corrected Akaike information criterion (AICc; Hurvich and Tsai, 1989) and considering the first, second, and third positions of the codon as separate partitions a priori. The resulting best partition scheme consisted of three partitions, one for each position of the codon. The best-fitting nucleotide substitution model for the first and third positions of the codon was general time-reversible (GTR; Tavaré, 1986) with a proportion of invariant sites and a gamma distribution of rates across sites, and F81 for the second position (Felsenstein, 1981) with a proportion of invariant sites.

We reconstructed phylogenetic trees using both Bayesian inference and maximum-likelihood optimality criteria. For the Bayesian analysis we used two independent runs of 1.0×10^7 generations, starting with random trees and four Markov chains (one cold), sampled every 1,000 generations in MrBayes v.3.2.6 (Ronquist et al., 2012), discarding 25% of generations and trees as burn-in. We used the standard deviation of split frequencies (<0.01) and estimated sample size (>200) to assess run convergence. We conducted maximum-likelihood analysis using RAxML v.8.2.10 (Stamatakis, 2014), searching the most likely tree 100 times and with 1,000 nonparametric bootstrap replicates to assess support. Although we used the three positions of the codon as separate partitions, we used the GTR + Γ + I model for all of them, because RAxML does not support different models across partitions.

We computed uncorrected pairwise distances in R v.3.5.0 (R Core Team, 2018) using the packages 'ape' v.3.4 (Paradis et al., 2004) and 'spider' v.1.3-0 (Brown et al., 2012). To reduce the effect of the alignment on the genetic distances, we deleted the sites with gaps in a pairwise way (using *pairwise.deletion=T* in the *dist.dna* command).

Results

Adenomera ***kweti*** sp. nov.
(Figs. 2A,B, 3, 5, Table 1)

Adenomera araucaria (Kwet, 2007: in part; Kwet et al., 2009:99, specimens on the left of fig. 4A,B,G,H; Conte et al., 2010:203, fig. 1H); *Adenomera* sp. R (Fouquet et al., 2014).

Holotype.—CFBH 43184 (field number JPCM 547), adult male (Fig. 2A–B), from Morro das Pedras trail (27.82112°S, 48.90340°W; 1,063 m a.s.l.), Serra do Tabuleiro State Park, municipality of São Bonifácio, state of Santa Catarina, southern Brazil, on 16 November 2017, collected by Thiago R. de Carvalho, Juliane P. Monteiro, and Marcus Thadeu T. Santos.

Paratypes.—CFBH 43183, 43185–43190 (field nos. JPCM 546, 548–553, respectively), adult males, all with the same collection data as the holotype; UFMG 7066, adult female, from Gruta São José (27.830639°S, 48.966528°W; 771 m a.s.l.), at Serra do Tabuleiro State Park, municipality of São Bonifácio, state of Santa Catarina, southern Brazil, on 17 November 2010, collected by Paulo C. A. Garcia, Julia Thompson, Pedro P. G. Taucce, Paulo D. P. Pinheiro, Marcus Thadeu T. Santos, Felipe Natali, Bárbara Zaidan, Bruno H. B. Fehlberg, Manoela W. Cardoso, Ana Carolina C. Lourenço, and Délio Baêta; UFMG 7080, 7111, adult

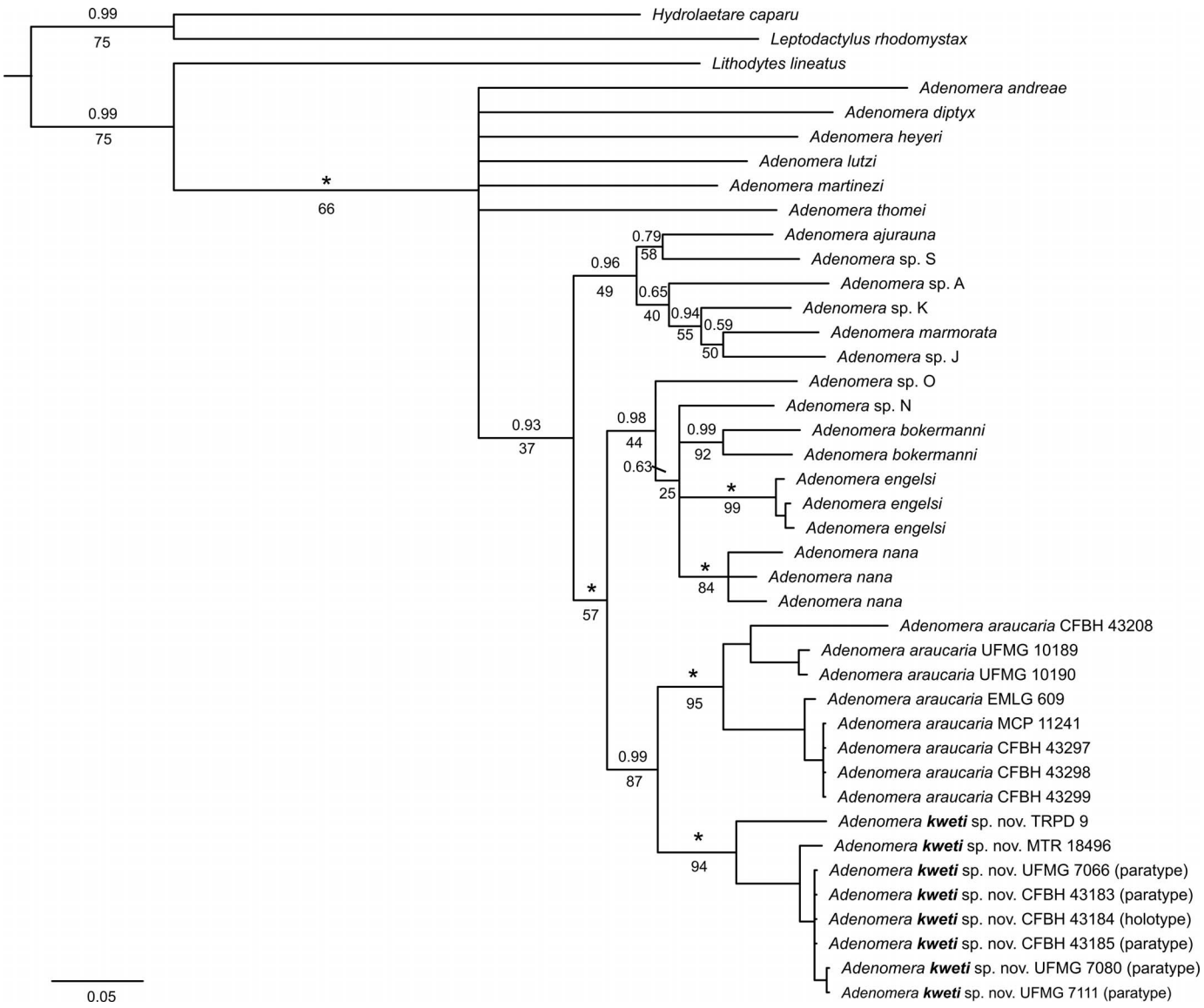

FIG. 1. The 50% majority rule consensus tree from Bayesian inference of partial mitochondrial *cytochrome c oxidase* subunit I gene (COI) showing *Adenomera kweti* and its relationships within *Adenomera*. Numbers above branches indicate posterior probabilities and numbers below branches indicate maximum likelihood nonparametric bootstrap values. Within-species support values are not shown. Asterisks (*) indicate full support.

males, from Poção trail (27.610502°S, 48.506645°W; 71 m a.s.l.), on Santa Catarina Island, in Florianópolis, state of Santa Catarina, southern Brazil, on 18 November 2010, collected by Paulo C. A. Garcia, Julia Thompson, Pedro P. G. Taucce, Paulo D. P. Pinheiro, Marcus Thadeu T. Santos, Felipe Natali, Bárbara Zaidan, Bruno H. B. Fehlberg, Manoela W. Cardoso, Ana Carolina C. Lourenço, and Délio Baêta; MCP 8212, adult male, from the municipality of Santo Amaro da Imperatriz, state of Santa Catarina, southern Brazil, on 15 December 2004, collected by Marcos Di-Bernardo, Axel Kwet, and A. F. Cordeiro; MCP 8285–8286, adult males, from Santa Catarina Island, in Florianópolis, state of Santa Catarina, southern Brazil, on 11 December 2004, collected by Axel Kwet and Anne Zillikens.

Nontype Material.—Two specimens from the state of Santa Catarina (southern Brazil): CFBH 22775, adult female from Governador Celso Ramos; MCP 1340, adult male from Santa Catarina Island.

Suggested Vernacular Name.—Kwet's nest-building frog.

Phylogenetic Relationships and Genetic Distance.—Both Bayesian inference and maximum-likelihood analyses yielded similar topologies (Fig. 1). *Adenomera kweti* was recovered as a member of the *A. marmorata* clade and the sister species of *A. araucaria* (Fig. 1). We recovered the holotype (CFBH 43184) and two paratypes (CFBH 43183, 43185) of *A. kweti* nested in a highly supported clade (1.0 of posterior probability and 94% of bootstrap) of the confirmed candidate species *Adenomera* sp. R of Fouquet et al. (2014), the sister group of the clade corresponding to nominal *A. araucaria*; this relationship also highly supported (0.99 of posterior probability and 84% of bootstrap). Genetic distances within and between species of the *A. marmorata* clade are given in Table 1.

Definition.—*Adenomera kweti* is characterized by the following combination of features: (1) small size (adult male SVL 15.4–19.3 mm), (2) toe tips unexpanded or slightly swollen (character state B–C of Heyer, 1973), (3) yellow belly, (4) nonpulsed advertisement call, (5) single-note advertisement call, (6) call dominant frequency contained in the second harmonic; (7) call with

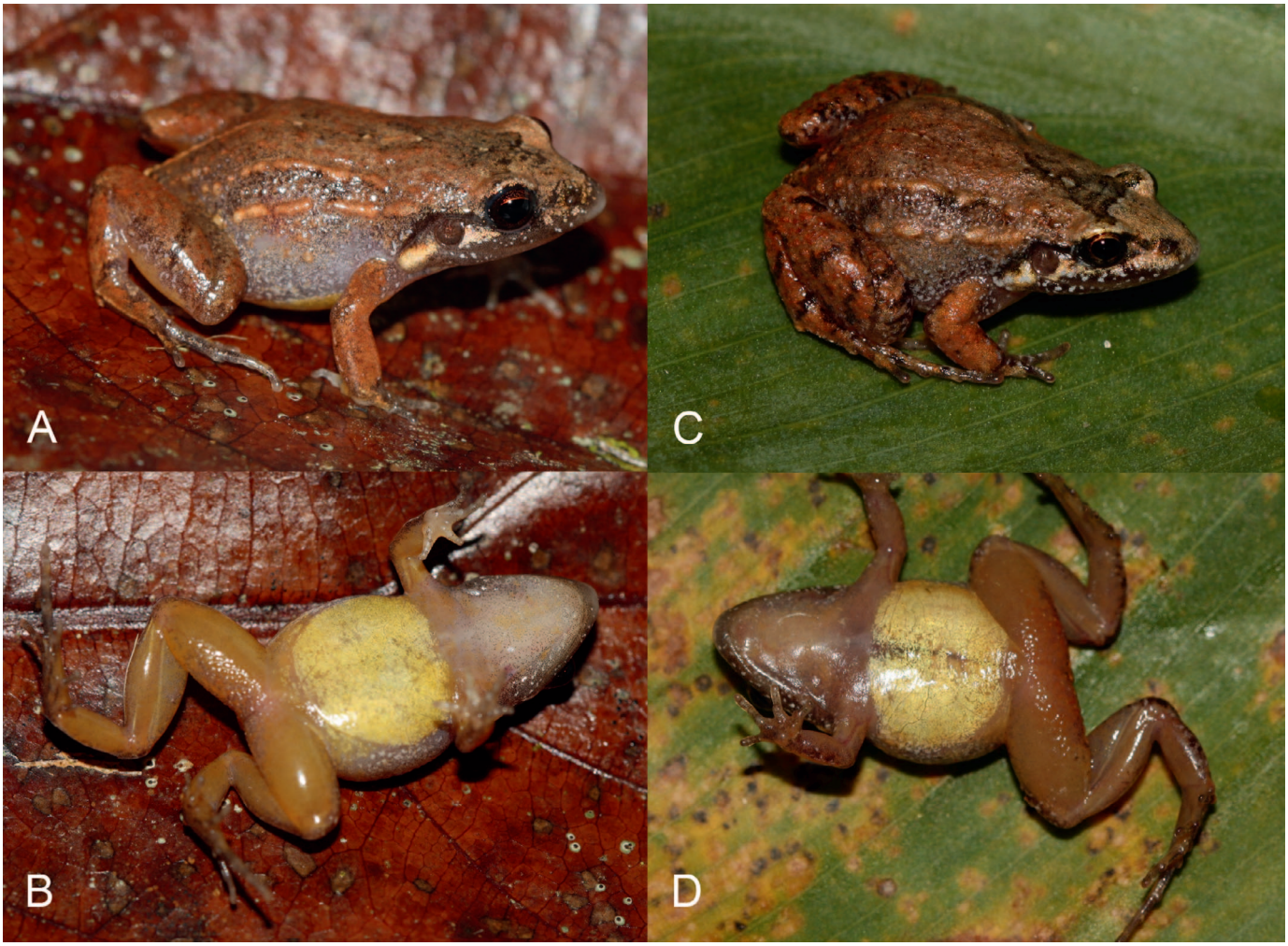

FIG. 2. Live adult males of (A,B) *Adenomera kweti* (Serra do Tabuleiro State Park, eastern Santa Catarina) and (C,D) *Adenomera araucaria* (São Francisco de Paula, Rio Grande do Sul) from their respective type localities in southern Brazil. (A,B) Holotype: CFBH 43184 (SVL 17.0 mm); (C,D) topotype: CFBH 43928 (SVL 17.8 mm).

pronounced frequency upsweep, (8) 10 transformations in the COI mitochondrial gene: A → G in position 195; C → T in positions 249, 537, 636, and 663; T → C in positions 313, 459, and 505; T → A in position 546; and C → A in position 627 (positions are relative to the partial mitochondrial genome of *Leptodactylus melanonotus*, GenBank accession number JX564873).

Diagnosis.—*Adenomera kweti* (SVL 15.4–19.3 mm; Table 2) is distinguished from almost all congeners by having small-sized adult males, except the Atlantic Forest species *A. ajurauna* (SVL 17.2–20.0 mm; Berneck et al., 2008), *A. araucaria* (SVL 17.1–18.8 mm; Kwet and Angulo, 2002), and *A. nana* (SVL 16.3–19.4 mm; Kwet, 2007). The presence of toe tips unexpanded or slightly swollen in *A. kweti* (character states B–C) distinguishes the new species from species having toe discs (*A. ajurauna*, *Adenomera andreae*, *A. marmorata*, *A. nana*, and *Adenomera simonstuarti*; Angulo et al., 2003; Kwet, 2007; Berneck et al., 2008; Angulo and Icochea, 2010; Appendix 1). The yellow belly of *A. kweti* distinguishes this species (Fig. 2B) from all congeners but *Adenomera heyeri* and *Adenomera lutzi* (Boistel et al., 2006; Kok et al., 2007), even though other *Adenomera* species from southern Brazil (e.g., *A. bokermanni* and *A. nana*) may also have yellowish tints in their bellies (T. R. de Carvalho, pers. obs.). Although its closest relative *A. araucaria* was characterized as white-bellied (Kwet and Angulo, 2002), we recently collected topotypical specimens with yellow bellies (Fig. 2D). This renders *A. kweti* and *A. araucaria* morphologically cryptic species. Nevertheless, these sister species are distinguished from each other based on their distinct vocalizations and phylogeny (see below; Figs. 1, 4).

The advertisement call of *A. kweti* (Fig. 4A–C; Table 3) is distinguished from those of congeners having pulsed notes (nonpulsed note in *A. kweti*; Table 4). From congeners also having nonpulsed notes, *A. kweti* differs by having the dominant frequency contained in the second harmonic (contained within the first harmonic in the calls of *A. ajurauna* and *A. marmorata*; Table 4). The advertisement call of the new species is emitted as single notes (multinote calls in *Adenomera cotuba*; Carvalho and Giaretta, 2013a). The pronounced frequency upsweep along the note of *A. kweti* distinguishes this species from *A. bokermanni*, *A. engelsi*, and *A. nana*, which do not have any modulation or less often subtle modulations in the initial or final portions of their notes (Kwet, 2007; Kwet et al., 2009). In addition to the nonpulsed note of *A. kweti* (Fig. 4B,C) distinguishing this species from the closest related *A. araucaria* (pulsed note; Fig. 4E,F), the new species is further distinguished by shorter note duration: *A. araucaria* (102–277 msec); *A. kweti* (60–84 msec).

Description of the Holotype.—Morphometric measurements of the holotype are given in Table 2. Adult male. Snout subelliptical

TABLE 1. Uncorrected pairwise genetic distances of partial *Cytochrome c oxidase* subunit I (COI) within and between Atlantic Forest species of *Adenomera* (*A. marmorata* clade). We ignored sites with missing data (pairwise.deletion = TRUE). Within-species distances are in bold font. Data are shown as min–max (mean).

	A. araucaria	*A. kweti*	*A. engelsi*	*A. nana*	*A. ajurauna*	*A. marmorata*	*A. bokermanni*
A. araucaria	**0.8–9.8 (5.4, *n* = 8)**						
A. kweti	9.8–12.1 (10.7)	**0.0–7.3 (2.3, *n* = 8)**					
A. engelsi	12.1–14.9 (12.8)	10.5–11.6 (10.9)	**0.5–1.0 (0.8, *n* = 3)**				
A. nana	10.6–15.6 (12.1)	9.6–13.0 (11.9)	6.7–7.9 (7.4)	**3.6–4.6 (4.0, *n* = 3)**			
A. ajurauna	12.4–15.7 (13.8)	12.3–14.9 (13.4)	11.2–12.3 (11.7)	10.4–12.6 (11.5)	**0.0–6.7 (5.6, *n* = 5)**		
A. marmorata	12.0–16.3 (13.9)	12.0–14.6 (13.3)	12.9–15.4 (13.9)	10.9–15.1 (13.2)	8.7–13.4 (11.0)	**0.0–7.5 (4.6, *n* = 13)**	
A. bokermanni	10.6–16.7 (12.9)	11.1–14.4 (13.1)	8.7–9.4 (9.1)	7.9–10.7 (9.3)	11.6–13.8 (12.7)	11.7–16.5 (13.7)	**6.5 (*n* = 2)**

from above (Fig. 3A), acuminate in profile (Fig. 3C). Nostrils closer to the snout tip than to the eyes; fleshy ridge on the tip of the snout; *canthus rostralis* rounded; loreal region concave; supratympanic fold from the posterior corner of the eye to the base of the arm; postcommissural gland ovoid; vocal sac subgular with a fold from jaw extending to arm; vocal slits present; vomerine teeth in two nearly straight rows medial and posterior to choanae and almost parallel to sagittal plane. Tongue elongated, free approximately in the posterior fourth. Relative finger lengths IV $<$ I $\simeq$ II $<$ III; fingers without ridges or fringes; finger tips rounded, not expanded or flattened; inner and outer metacarpal tubercles nearly rounded (Fig. 3D). Subarticular tubercles rounded; a few, large supernumerary tubercles. Antebrachial tubercle absent. Dorsum and limbs mostly smooth, warts concentrated on flank and inguinal region. Dorsolateral fold interrupted, extending posteriorly from scapular region to posterior third of the body length. Dorsal fold flattened, short and interrupted, extending longitudinally in the middle of the body. Lateral fold extending from supratympanic fold to groin, as a granular row posteriorly. Ventral surfaces smooth (Fig. 3B), granular between underside of thigh and cloacal region. Posterior surface of thigh granular, possessing conspicuous, nearly rounded paracloacal gland. Relative toe lengths I $<$ II $<$ V $<$ III $<$ IV; vestigial webbing at the base of toes II–III and toes IV–V (under magnification); toe tips unexpanded (character state B). Inner metatarsal tubercle ovoid, outer metatarsal tubercle subconical; inner tubercle twice the maximum diameter of outer tubercle (Fig. 3E). Tarsus smooth. Tarsal fold from inner metatarsal tubercle extending halfway of the tarsus length. Subarticular tubercles rounded or ovoid; a few supernumerary tubercles barely noticeable.

Color of Holotype in Preservative.—The snout tip lacks coloration (coincident with the fleshy ridge). Snout darker than the rest of body dorsally and laterally (Fig. 3). Dorsal surfaces light brown with faded dark brown specks on interorbital, scapular, and lumbar regions, and accompanying the dorsolateral folds; hindlimbs with transverse dark brown stripes. Sacral stripe cream colored. Postcommissural gland cream colored, speckled with dark melanophores concentrated on upper and lower margins. Tympanum brown specked with melanophores, inner rim of the tympanic annulus brown (darker than the specks over the tympanic membrane). Supratympanic fold is marked with a dark brown stripe. Melanophores concentrated on the outer margin of the ventral surface of forelimbs and throat, especially laterally. Belly and hindlimbs cream colored, melanophores scattered irregularly. Posterior surface of thigh covered with melanophores on a cream-colored background; paracloacal gland whitish cream.

Color of Holotype in Life.—The coloration in life does not differ much from the color described in preservative, just more intense in hues of brown and gray (Fig. 2A,B). Postcommissural gland, sacral stripe, and horizontal stripe between the posterior corner of the eye and the anterior margin of the tympanum cream-colored. Iris copper. Throat, chest, and flank in contact with the belly, translucent and covered with melanophores and white freckles. Belly coloration in two shades of yellow. Ventral surface of the limbs orange–yellow.

Variation.—Morphometric variation is provided in Table 2. The development of supernumerary tubercles on hand and foot and tarsal fold may be indistinct or conspicuous. Tubercles on posterior surface of tarsus and vestigial webbing between toes may be present. Toe tips vary from unexpanded or slightly swollen (character states B–C), especially in Toes III–IV. The

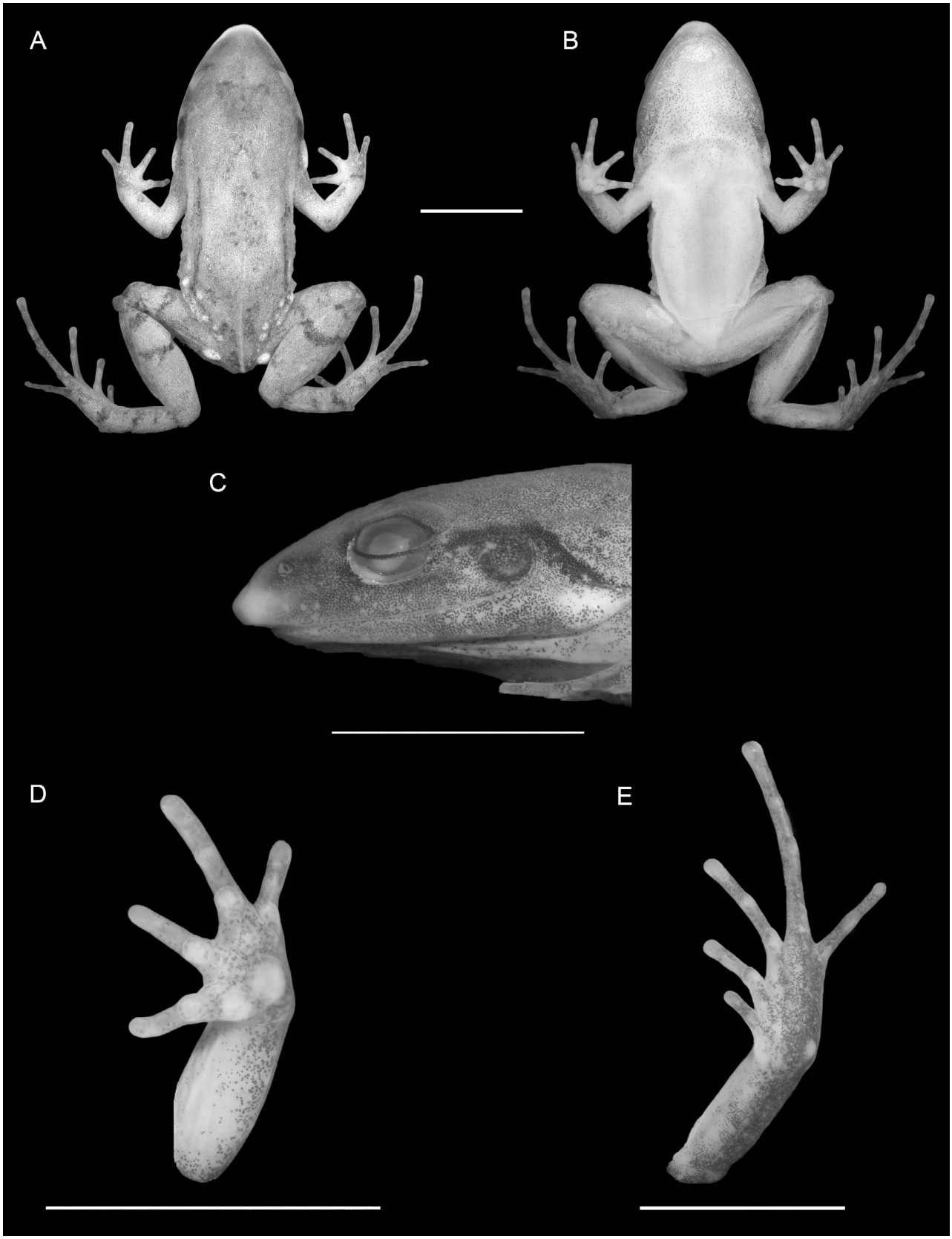

FIG. 3. Preserved holotype of *Adenomera kweti* (adult male, CFBH 43184; SVL 17.0 mm). (A,B) dorsal and ventral body, (C) lateral head, (D) palm of hand, (E) sole of foot. Scale bars = 5 mm.

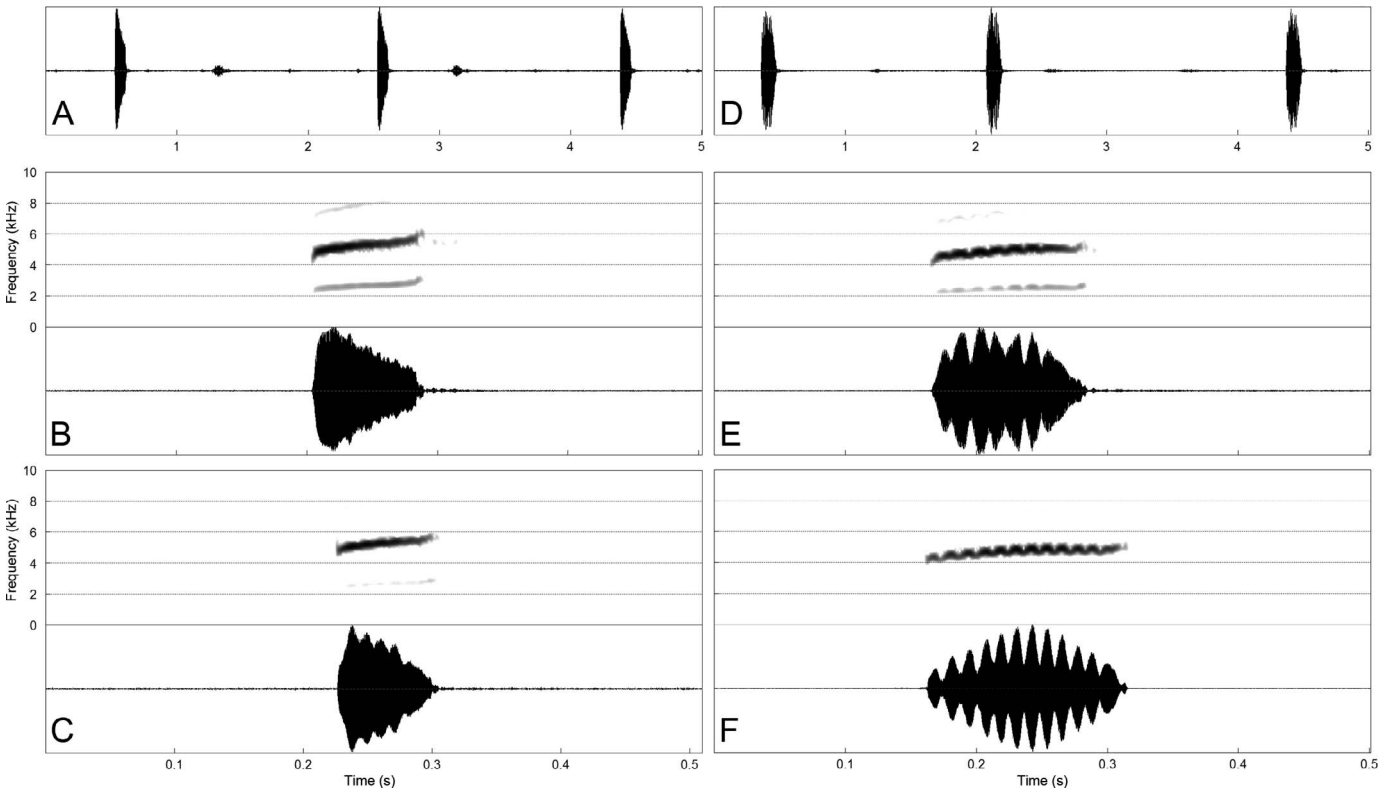

FIG. 4. Advertisement calls of *Adenomera kweti* (A–C) and *Adenomera araucaria* (D–F) from southern Brazil. (A) Nonpulsed notes of the holotype CFBH 43184, recorded from Serra do Tabuleiro State Park. (B) The third nonpulsed note in A. (C) Nonpulsed note of the male UFMG 7080, recorded from the Santa Catarina Island. (D) Multipulsed notes of the male topotype CFBH 43927, recorded from São Francisco de Paula. (E) The first multipulsed note (seven pulses) in (D). (F) Thirteen-pulse note of an unvouchered male from São Bento do Sul. Time-domain sections (A,D; ca. 5 sec) and spectrograms/oscillograms (B,C,E,F; ca. 0.5 sec) were produced approximately on the same time scale. Information on sound recordings is given in Appendix 2.

female UFMG 7066 does not have paracloacal gland and has a more rounded snout in dorsal and lateral views. There is substantial variation in life colors of the dorsal surfaces, varying from light brown, orange shades merged with brown, and gray (Fig. 5). The presence, distribution, and shape of spots and blotches, including the interorbital blotch, is highly variable. In life, specimens may be almost immaculate (Fig. 5A,D), covered with blotches and transverse bars on the dorsal surface of hindlimbs (Fig. 5C), or have the color morphotype of dorsolateral and sacral stripes (Fig. 5B). Dorsolateral fold may be conspicuous or flattened and skin texture glandular or mostly smooth. Dorsolateral stripe present in two paratypes (CFBH 43187, MCP 8285). Sacral stripe is absent in six paratypes (CFBH 43183, 43186, 43188; MCP 8212; UFMG 7080, 7111).

Call Description of A. kweti.—Based on recordings of five males (Appendix 2). Sample sizes for each acoustic trait and descriptive statistics (means ± SD) are given in Table 3. The advertisement call (Fig. 4A–C) consists of a single nonpulsed note emitted at a rate of 24–54 calls per minute. Note duration lasts 60–84 msec. The rise time is 9–67% of the notes' length. The fundamental frequency

TABLE 2. Measurements (in mm) for the type series of *A. kweti* and *A. araucaria* (type specimens and topotypes, and specimens from other localities in the state of Santa Catarina; see Appendix 1 for localities). Values are given as $X \pm$ SD (range).

	Adenomera kweti			*Adenomera araucaria*[b]	
Trait[a]	Holotype (CFBH 43184)	Male paratypes ($n = 12$)	Female paratype (UFMG 7066)	Type specimens and topotypes ($n = 10$ adult males)	Santa Catarina ($n = 3$ adult males)
SVL	17.0	17.0 ± 1.2 (15.4–19.3)	16.1	18.3 ± 0.6 (17.4–18.9)	18.4 ± 0.8 (17.8–19.3)
HL	5.9	5.8 ± 0.4 (5.2–6.4)	5.5	5.9 ± 0.2 (5.5–6.0)	6.4 ± 0.4 (6.0–6.7)
HW	6.3	6.4 ± 0.5 (5.7–7.4)	6.0	6.7 ± 0.5 (6.1–7.7)	6.8 ± 0.6 (6.3–7.5)
ED	1.5	1.6 ± 0.1 (1.4–1.8)	1.6	1.6 ± 0.2 (1.4–1.8)	1.6 ± 0.1 (1.5–1.7)
TD	0.9	0.9 ± 0.1 (0.8–1.0)	0.9	0.9 ± 0.1 (0.7–1.0)	1.0
END	1.2	1.3 ± 0.2 (1.1–1.8)	1.6	1.5 ± 0.1 (1.3–1.6)	1.4 ± 0.1 (1.3–1.5)
IND	1.6	1.6 ± 0.1 (1.4–1.8)	1.4	1.7 ± 0.2 (1.4–1.9)	1.7 ± 0.2 (1.5–1.9)
HAL	3.6	3.9 ± 0.3 (3.5–4.4)	3.9	3.9 ± 0.3 (3.5–4.5)	4.2 ± 0.4 (3.9–4.6)
TL	7.4	7.7 ± 0.5 (6.7–8.6)	7.1	7.9 ± 0.4 (7.4–8.5)	7.7 ± 0.5 (7.3–8.3)
SL	7.5	7.8 ± 0.9 (6.6–9.3)	8.0	7.9 ± 0.3 (7.3–8.3)	7.8 ± 0.6 (7.4–8.4)
FL	8.0	8.7 ± 0.8 (7.1–9.9)	9.0	9.0 ± 0.3 (8.5–9.5)	9.3 ± 0.6 (8.8–10.0)

[a] SVL = snout-vent length; HL = head length; HW = head width; ED = eye diameter; TD = tympanum diameter; END = eye-nostril distance; IND = internarial distance; HAL = hand length; TL = thigh length; SL = shank length; FL = foot length.
[b] Type specimens: MCP 2421, 3208, 3345, 3346, 3463, 3672, 3676; topotypes: CFBH 43927–43929; Santa Catarina: CFBH 39262, UFMG 10189–90.

TABLE 3. Advertisement call traits of *A. kweti* from the type locality (Serra do Tabuleiro State Park) and from the Santa Catarina Island, both localities in eastern Santa Catarina state, and comparative call traits of *A. araucaria* from the type locality (São Francisco de Paula, state of Rio Grande do Sul) and state of Santa Catarina (São Bento do Sul and Jaraguá do Sul). All localities in southern Brazil. Values are given as X ± SD (min–max). NA = not applicable, given *A. kweti*'s call is nonpulsed.

Traits	*A. kweti*		*A. araucaria*	
	Type locality (n = 4 males)	Santa Catarina Island (n = 1 male)	Type locality (n = 3 males)	Santa Catarina (n = 10 males)
Note duration (msec)	75.0 ± 6.0 (60–84), n = 120	75.9 ± 2.4 (70–80), n = 35	111.8 ± 4.1 (102–119), n = 32	167.5 ± 34.4 (126–277), n = 229
Note rate/min	33.1 ± 8.2 (24–42), n = 4	53.8, n = 1	29.5 ± 1.6 (28–30), n = 3	32.8 ± 8.1 (23–46), n = 10
Note rise time (%)	37.6 ± 14.7 (9–67), n = 120	18.4 ± 8.0 (11–34), n = 35	41.8 ± 3.2 (31–66), n = 32	48.5 ± 13.6 (12–65), n = 229
Pulses per note	NA	NA	4.7 ± 0.2 (4–6), n = 32	10.6 ± 2.9 (4–18), n = 229
Pulse duration (msec)	NA	NA	24.4 ± 1.7 (18–30), n = 150	16.6 ± 6.2 (11–37), n = 2281
Pulse rate/s	NA	NA	63.0 ± 3.8 (42–91), n = 32	67.1 ± 13.0 (32–88), n = 229
Fundamental frequency (Hz)	2,560.7 ± 95.1 (2,355–2,707), n = 120	2,544.3 ± 30.0 (2,509–2,595), n = 35	2,421.6 ± 32.2 (2,358–2,478), n = 32	2,262.8 ± 85.5 (2,101–2,419), n = 229
Dominant frequency (Hz)	5,157.5 ± 184.4 (4,758–5,414), n = 120	5,134.1 ± 55.8 (5,060–5,276), n = 35	4,835.1 ± 48.5 (4,673–4,931), n = 32	4,590.4 ± 199.4 (4,113–4,931), n = 229
Frequency modulation (Hz)	744.6 ± 28.1 (563–891), n = 120	563.6 ± 80.5 (431–775), n = 35	541.8 ± 31.9 (388–732), n = 32	766.7 ± 157.4 (388–1,206), n = 229

(first harmonic) ranges from 2,355 to 2,707 Hz; the dominant frequency (second harmonic) from 4,758 to 5,414 Hz. Notes have a pronounced frequency upsweep that ranges from 431 to 891 Hz.

Call redescription of A. araucaria.—Based on recordings of 13 males, including three topotypes (Appendix 2). Sample sizes for each acoustic trait and descriptive statistics (means + SD) are given in Table 3. The advertisement call (Fig. 4D–F) consists of a single multipulsed note emitted at a rate of 23–46 calls per minute. Note duration lasts 102–277 msec. The rise time is 12–66% of the notes' length. Advertisement notes are formed by 4–18 partly fused pulses, emitted at a rate of 32–91 pulses per second. Pulse duration lasts 11–37 msec. The fundamental frequency (first harmonic) ranges from 2,101 to 2,478 Hz; the dominant frequency (second harmonic) from 4,113 to 4,931 Hz. Notes have a pronounced frequency upsweep that ranges from 388 to 1,206 Hz.

Etymology.—The specific epithet is a homage to Axel Kwet in recognition of his invaluable contribution to the taxonomy of *Adenomera* in southern Brazil.

Distribution.—*Adenomera kweti* occurs across mid- and high-elevation Atlantic Forest localities in eastern Santa Catarina (southern Brazil), including the Serra do Tabuleiro mountain range, the Santa Catarina Island, and additional localities in the region (Fig. 6). Acoustic data for additional *Adenomera* populations in southern Brazil provided by Kwet (2007) confirmed the distribution range of *A. kweti* in eastern Santa Catarina.

Natural History.—At the type locality, males of the species were in calling activity during daytime (mid-afternoon) from a clearing on the trail covered with bamboos. All males were calling from under a thick vegetation layer made up of bamboo decaying leaves on the forest floor. At the end of that day, males exposed themselves on the vegetation while and shortly after a brief rain shower. Calling activity extended into early night. *Adenomera engelsi* occurs sympatrically with *A. kweti* in eastern Santa Catarina, but we did not observe those species occurring syntopically at the type locality; however, these species were observed calling syntopically and synchronically by the end of the day in other study areas (Rancho Queimado and Santa Catarina Island). *Adenomera engelsi* was observed vocalizing at the forest edge, but also used open areas (e.g., garden areas), and its calling activity was more associated with the night period. *Adenomera kweti*, on the other hand, appears to vocalize more often during daytime and tends to be more associated with forest areas, even though calling males were concentrated in forest clearings at the type locality.

Remarks.—We detected weak amplitude modulations in some calls of *Adenomera kweti* (Fig. 4C) that could be associated with the peculiar partly fused pulses among members of *Adenomera*. The temporal envelope of pulsed-call species is accompanied by sinusoidal frequency modulations in the corresponding frequency structure of their harmonics (Fig. 4E,F). In contrast, the harmonic structure in the calls of *A. kweti* mentioned earlier exhibits only linear frequency modulation along the notes (Fig. 4B,C). Therefore, we attribute the weak amplitude modulations to extrinsic factors that might have somehow affected sound propagation, such as interfering structures in the environment, given that males of many *Adenomera* species may call from underneath leaf litter or partially burrowed from inside underground chambers. Extrinsic factors related to sounds are summarized elsewhere (Ryan, 1988).

Males of *A. araucaria* were calling in the background of other species recorded from Morro do Baú (Ilhota, Santa Catarina) by a colleague (T. R. de Carvalho, pers. obs.). Those background calls were assigned to *A. araucaria* based on the major acoustic characteristics of the species' call: pulsed call with pronounced

TABLE 4. Acoustic information on *Adenomera* with special reference to advertisement call (=note) duration, pulses (presence/absence), and peak frequencies in the first two harmonics. The new species is in bold. Abbreviations: N = nonpulsed; H0 = first harmonic; H1 = second harmonic.

Species	Note duration (msec)	Pulses/note	H0 frequency (kHz)	H1 frequency (kHz)	Reference
A. ajurauna	130–190	N	3.72–5.43	–	Berneck et al. (2008)
A. andreae	45–86	4	2.32–2.69	4.56–5.49	Boistel et al. (2006)
A. araucaria	102–277	4–18	2.10–2.48	4.11–4.93	Present study
A. bokermanni[a]	99–152	N	1.79–1.83	3.40–3.57	Kwet (2007)
A. coca	110–145	10–15	1.69–1.91	3.45–3.75	Angulo and Reichle (2008)
A. cotuba	69–191	8–14	1.73–1.83	3.33–3.80	Carvalho and Giaretta (2013a)
A. diptyx[b]	56–88	Pulsed	2.18–2.28	4.20–4.50	Márquez et al. (1995)
A. engelsi	96–163	N	ca. 2.00	3.46–4.29	Kwet et al. (2009)
A. heyeri	137–185	9.5	1.82–1.88	3.57–3.84	Boistel et al. (2006)
A. hylaedactyla	35–62	4–6	1.95–2.21	3.96–4.48	Angulo et al. (2003)
A. juikitam	148–202	16–21	1.88–2.11	3.70–4.17	Carvalho and Giaretta (2013a)
A. kweti	**60–84**	**N**	**2.36–2.71**	**4.76–5.41**	**Present study**
A. lutzi	41–61	N	1.64–1.81	3.27–3.62	Kok et al. (2007)
A. marmorata	100	N	4.50–5.60	–	Straughan and Heyer (1976)
A. martinezi	63–151	15–21	1.88–2.06	3.38–4.13	Carvalho and Giaretta (2013b)
A. nana	67–122	N	2.30–2.70	4.62–5.44	Kwet (2007)
A. saci	90–241	N	1.69–2.25	3.38–4.41	Carvalho and Giaretta (2013b)
A. simonstuarti	57–71	3–4	1.81–2.03	3.71–4.05	Angulo and Icochea (2010)
A. thomei	120–210	10–21	2.15–2.81	4.57–5.56	Almeida and Angulo (2006)

[a] Based on the call of *Adenomera* sp. 2 from Joinville.
[b] Reported as *A. andreae*.

frequency upsweep. Therefore, we assigned the series examined from Morro do Baú (MCP 1346–52) previously reported as *A.* cf. *araucaria* by Kwet (2007) to *A. araucaria*. Specimen MCP 1340 examined from the Santa Catarina Island is fairly preserved (especially body shape and toe tips). Even so, the only two *Adenomera* species known to date on the island are *A. kweti* and *A. engelsi*. The specimen certainly is not *A. engelsi*; therefore, we assigned it to *A. kweti*. Specimen CFBH 22175 examined from Governador Celso Ramos is within the distribution range of *A. kweti*. We tentatively assigned it to *A. kweti* while awaiting acoustic or molecular information for this population.

FIG. 5. Color variation in the type series of *Adenomera kweti*. (A–D) Paratypes: CFBH 43183 (SVL 17.8 mm), CFBH 43187 (SVL 15.4 mm), CFBH 43188 (SVL 16.5 mm), and CFBH 43190 (SVL 15.7 mm), respectively.

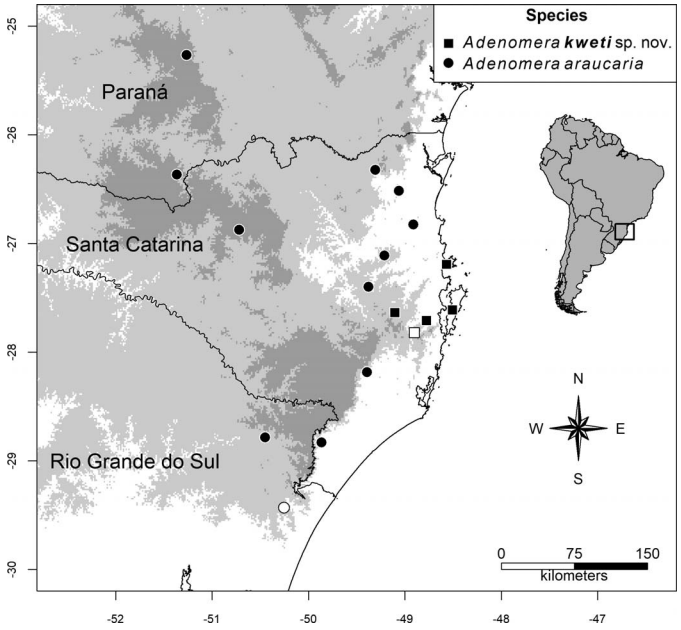

FIG. 6. Geographic distribution of *Adenomera kweti* and *Adenomera araucaria* across the three states in southern Brazil. Colors correspond to elevation quotas: white (up to 500 m), light gray (above 500 m), dark gray (above 1,000 m). White symbols indicate type localities.

DISCUSSION

Adenomera kweti and *A. engelsi* occur sympatrically in eastern Santa Catarina state and allopatric to the other three *Adenomera* species distributed in southern Brazil. There is shift in species composition toward the region of the Serra do Tabuleiro mountain range and the Santa Catarina Island. *Adenomera nana* occurs from south-central Paraná to central Santa Catarina (Conte et al., 2010; Fouquet et al., 2014); *A. araucaria* is the most widespread in southern Brazil, occurring in the southernmost Brazilian state (Rio Grande do Sul) and northward across mainland Santa Catarina to south-central Paraná, but not into the eastern portion of Santa Catarina (Conte et al., 2010; Fouquet et al., 2014); *A. bokermanni* is restricted to a small coastal area in Paraná and northern Santa Catarina (Fouquet et al., 2014). Therefore, *A. kweti* and *A. engelsi* are endemic to eastern Santa Catarina, a region covered by the Atlantic Forest in southern Brazil.

The closest related *A. kweti* and *A. araucaria* have distinct advertisement calls (*A. kweti*: nonpulsed and shorter in duration; *A. araucaria*: pulsed and longer in duration). We are intrigued that allopatric and sibling species of the genus *Adenomera* have evolved different ways of vocalizing by the increment or loss of amplitude modulations, i.e., pulses, which reflects distinct temporal envelopes in their calls. A similar pattern was already reported for *Adenomera martinezi* and *Adenomera saci*, which also are allopatric sibling species and differ from each other based mainly on the temporal envelope of their calls (nonpulsed call in *A. saci*, pulsed call in *A. martinezi*; Carvalho and Giaretta, 2013b). Unlike *A. araucaria* and *A. kweti*, which are also distinguished by distinct duration of their calls, *A. saci* and *A. martinezi* have similar call durations. Given the high taxonomic value of call features in species diagnosis in *Adenomera*, investigating differentiation patterns of advertisement calls in sibling species of *Adenomera* might be a promising area of research; it could help our understanding of how acoustic characteristics evolve among members of the genus or pairs of species in sympatric/allopatric conditions.

In general, members of the *A. marmorata* clade in southern Brazil have two major advertisement call patterns: (i) the call of *A. araucaria* and *A. kweti* have a pronounced frequency upsweep, even though these species' calls are easily distinguished by distinct temporal envelopes (pulsed vs. nonpulsed) and call durations, whereas (ii) the remaining species (*A. bokermanni*, *A. engelsi*, and *A. nana*) do not have any frequency shift in their calls (despite subtle modulations in the initial and final portions of their calls in some cases; Kwet, 2007; Kwet et al., 2009). Acoustic data are informative for the accurate assessment of the taxonomic identity of all five species of *Adenomera* distributed in southern Brazil. We highlight that there are two other genetic lineages in the *A. marmorata* clade from southern Brazil indicated by Fouquet et al. (2014), namely, *Adenomera* sp. N and *Adenomera* sp. O, that still require evaluation of their taxonomic status, including the acquisition of acoustic information for these candidate species from southern Brazil.

Acknowledgments.—We received financial support through research grants from São Paulo Research Foundation (FAPESP 2013/50741-7 and 2014/50342-8; CFBH) and from National Council for Scientific and Technological Development (CNPq 302518/2013-4). The taxonomic review of *Adenomera* species was part of CSC's and TRC's Ph.D. theses. In that period, they were supported by FAPESP with doctoral fellowships (2010/515606-8 and 2012/15763-7, respectively). TRC is currently supported with a postdoctoral fellowship from the same funding agency (2017/08489-0). The collecting permits were granted by Instituto Chico Mendes (ICMBio/SISBIO 30059-10; 4023-1) and Fundação do Meio Ambiente do estado de Santa Catarina (FATMA 18/2017 GERUC/DPEC). We thank field companions during fieldwork: Fabiana Dallacorte, Javier Toso, Juliane P. Monteiro, Marcelo Freire, and Marcus Thadeu T. Santos; Mariana L. Lyra for her help with DNA sequencing protocols; Ariovaldo A. Giaretta for research support by providing recording equipment during field expeditions. We are grateful to curators and collection managers for granting access to specimens under their care: Nadya Pupin and Danilo B. Delgado (CFBH), Luis F. Toledo and Karina Rabelo (ZUEC), Taran Grant and Aline S. Benetti (MZUSP), Gláucia F. Pontes (MCP), Paulo C. A. Garcia (UFMG), Ariovaldo A. Giaretta (AAG-UFU), Fernanda P. Werneck and Ariane Silva (INPA-H), Ana L. C. Prudente and Fabricio Sarmento (MPEG), Marcelo N. C. Kokubum (LHUFCG). We thank Simone Dena and Luis Felipe Toledo for granting us access to the *Adenomera* sound recording housed at the Fonoteca Neotropical Jacques Vielliard (FNJV); and Axel Kwet for making available his original recordings of *Adenomera* species from southern Brazil. We thank the anonymous reviewers for their careful reading of our manuscript.

LITERATURE CITED

ALMEIDA, A. P., AND A. ANGULO. 2006. A new species of *Leptodactylus* (Anura: Leptodactylidae) from the state of Espírito Santo, with remarks on the systematics of associated populations. Zootaxa 1334: 1–25.

ANGULO, A., R. B. COCROFT, AND S. REICHLE. 2003. Species identity in the genus *Adenomera* (Anura: Leptodactylidae) in southeastern Peru. Herpetologica 59:490–504.

ANGULO, A., AND J. ICOCHEA. 2010. Cryptic species complexes, widespread species and conservation: lessons from Amazonian frogs of the *Leptodactylus marmoratus* group (Anura: Leptodactylidae). Systematics and Biodiversity 8:357–370.

ANGULO, A., AND S. REICHLE. 2008. Acoustic signals, species diagnosis, and species concepts: the case of a new cryptic species of *Leptodactylus* (Amphibia, Anura, Leptodactylidae) from the Chapare Region, Bolivia. Zoological Journal of the Linnean Society 152:59–77.

AUDACITY TEAM. 2017. Audacity(R): Free Audio Editor and Recorder, version 2.2.1. Available at https://www.audacityteam.org. Archived by WebCite at http://www.webcitation.org/732btrNLV on 9 October 2018.

BERNECK, B. V. M., C. O. R. COSTA, AND P. C. A. GARCIA. 2008. A new species of *Leptodactylus* (Anura: Leptodactylidae) from the Atlantic Forest of São Paulo State, Brazil. Zootaxa 1795:46–56.

BOISTEL, R., G. C. MASSARY, AND A. ANGULO. 2006. Description of a new species of the genus *Adenomera* (Amphibia, Anura, Leptodactylidae) from French Guyana. Acta Herpetologica 1:1–14.

BROWN, S. D. J., R. A. COLLINS, S. BOYER, M. C. LEFORT, J. MALUMBRES-OLARTE, C. VINK, AND R. H CRUICKSHANK. 2012. Spider: An R package for the analysis of species identity and evolution, with particular reference to DNA barcoding. Molecular Ecology Resources 12:562–565.

CARVALHO, T. R., AND A. A. GIARETTA. 2013a. Bioacoustics reveals two new syntopic species of *Adenomera* Steindachner (Anura: Leptodactylidae: Leptodactylinae) in the Cerrado of central Brazil. Zootaxa 3731:533–551.

CARVALHO, T. R., AND A. A. GIARETTA. 2013b. Taxonomic circumscription of *Adenomera martinezi* (Bokermann, 1956) (Anura: Leptodactylidae: Leptodactylinae) with the recognition of a new cryptic taxon though a bioacoustic approach. Zootaxa 3701:207–237.

CONTE, C. E., F. NOMURA, R. A. MACHADO, A. KWET, R. LINGNAU, AND D. C. ROSSA-FERES. 2010. Novos registros na distribuição geográfica de anuros na floresta com araucária e considerações sobre suas vocalizações. Biota Neotropica 10:201–224.

EDGAR, R. C. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. Nucleic Acids Research 32:1792–1797.

FELSENSTEIN, J. 1981. Evolutionary trees from DNA sequences: a maximum likelihood approach. Journal of Molecular Evolution 17: 368–376.

FOUQUET, A., C. S. CASSINI, C. F. B. HADDAD, N. PECH, AND M. T. RODRIGUES. 2014. Species delimitation, patterns of diversification and historical biogeography of the Neotropical frog genus *Adenomera* (Anura, Leptodactylidae). Journal of Biogeography 41:855–870.

GRIDI-PAPP, M. 2007. Soundruler: acoustic analysis for research and teaching. Available at http://soundruler.sourceforge.net. Archived by WebCite at http://www.webcitation.org/732cA62V3 on 9 October 2018.

HEYER, W. R. 1973. Systematics of the marmoratus group of the frog genus *Leptodactylus* (Amphibia, Leptodactylidae). Contributions in Science, Natural History Museum 251:1–50.

HEYER, W. R., A. S. RAND, C. A. G. CRUZ, O. L. PEIXOTO, AND C. E. NELSON. 1990. Frogs of Boracéia. Arquivos de Zoologia 31:231–410.

HURVICH, C., AND C. TSAI. 1989. Regression and time series model selection in small samples. Biometrika 76:297–307.

KOK, P. J. R., M. N. C. KOKUBUM, R. D. MACCULLOCH, AND M. LATHROP. 2007. Morphological variation in *Leptodactylus lutzi* (Anura, Leptodactylidae) with description of its advertisement call and notes on its courtship behavior. Phyllomedusa 6:45–60.

KUMAR, S., G. STECHER, AND K. TAMURA. 2016. MEGA7: Molecular Evolutionary Genetics Analysis version 7.0 for bigger datasets. Molecular Biology and Evolution 33:1870–1874.

KWET, A. 2007. Bioacoustic variation in the genus *Adenomera* in southern Brazil, with revalidation of *Leptodactylus nanus* Müller, 1922 (Anura, Leptodactylidae). Mitteilungen aus dem Museum für Naturkunde in Berlin, Zoologische Reihe Supplement 83:56–68.

KWET, A., AND A. ANGULO. 2002. A new species of *Adenomera* (Anura, Leptodactylidae) from the Araucaria forest of Rio Grande do Sul (Brazil), with comments on the systematic status of southern populations of the genus. Alytes 20:28–43.

KWET, A., J. STEINER, AND A. ZILLIKENS. 2009. A new species of *Adenomera* (Amphibia: Anura: Leptodactylidae) from the Atlantic rain forest in Santa Catarina, southern Brazil. Studies on Neotropical Fauna and Environment 44:93–107.

LANFEAR, R., P. B. FRANDSEN, A. M. WRIGHT, T. SENFELD, AND B. CALCOTT. 2017. PartitionFinder 2: new methods for selecting partitioned models of evolution for molecular and morphological phylogenetic analyses. Molecular Biology and Evolution 34:772–773.

LIGGES U., S. KREY, O. MERSMANN, AND S. SCHNACKENBERG. 2017. tuneR: analysis of music and speech. Available at https://CRAN.R-project.org/package=tuneR. Archived by WebCite at http://www.webcitation.org/74jswKLGU on 17 December 2018.

LYRA, M. L., C. F. B. HADDAD, AND A. M. L. AZEREDO-ESPIN. 2017. Meeting the challenge of DNA barcoding Neotropical amphibians: polymerase chain reaction optimization and new COI primers. Molecular Ecology Resources 17:966–980.

MÁRQUEZ, R., I. DE LA RIVA, AND J. BOSCH. 1995. Advertisement calls of Bolivian Leptodactylidae (Amphibia, Anura). Journal of Zoology 237:313–336.

MATLAB. 2004. The language of technical computing, version 6.5.2. The MathWorks Inc., USA.

MEYER, C. P. 2003. Molecular systematics of cowries (Gastropoda: Cypraeidae) and diversification patterns in the tropics. Biological Journal of the Linnean Society 79:401–459.

PARADIS, E., J. CLAUDE, AND K. STRIMMER. 2004. APE: analyses of phylogenetics and evolution in R language. Bioinformatics 20:289–290.

R CORE TEAM. 2018. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. Available at http://www.r-project.org. Archived by WebCite at http://www.webcitation.org/732cRpqcw on 9 October 2018.

RONQUIST F., M. TESLENKO, P. VAN DER MARK, D. L. AYRES, A. DARLING, S. HÖHNA, B. LARGET, L. LIU, M. A. SUCHARD, AND J. P. HUELSENBECK. 2012. MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61:539–542.

RYAN, M. J. 1988. Constraints and patterns in the evolution of anuran acoustic communication. Pp. 637–677 in B. Fritzsch (Ed.), The Evolution of the Amphibian Auditory System. John Wiley & Sons, Inc., USA.

SABAJ, M. H. 2016. Standard symbolic codes for institutional resource collections in herpetology and ichthyology: an online reference (v6.5). Available at http://www.asih.org/sites/default/files/documents/symbolic_codes_for_collections_v6.5_2016.pdf. Archived by WebCite at http://www.webcitation.org/74ilEB3Nr on 16 December 2018.

STAMATAKIS, A. 2014. RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics 30:1312–1313.

STRAUGHAN, I. R., AND W. R. HEYER. 1976. A functional analysis of the mating calls of the Neotropical frog genera of the *Leptodactylus* complex (Amphibia, Leptodactylidae). Papéis Avulsos de Zoologia, São Paulo 29:221–245.

SUEUR, J., T. AUBIN, AND C. SIMONIS. 2008. Seewave: a free modular tool for sound analysis and synthesis. Bioacoustics 18:213–226.

TAVARÉ, S. 1986. Some probabilistic and statistical problems in the analysis of DNA sequences. Lectures on Mathematics in the Life Sciences 17:57–86.

WATTERS, J. L., S. T. CUMMINGS, R. L. FLANAGAN, AND C. D. SILER. 2016. Review of morphometric measurements used in anuran species descriptions and recommendations for a standardized approach. Zootaxa 4072:477–495.

Accepted: 24 February 2019.
Published online: 15 May 2019.

ZooBank IDs: urn:lsid:zoobank.org:act:D98C77EB-BA80-41E9-B9C5-3319E2FF09B8; urn:lsid:zoobank.org:pub:ED5C76D4-1FB0-4F28-AF97-7194FF951910

APPENDIX 1. OTHER SPECIMENS EXAMINED

Adenomera ajurauna—BRAZIL: SÃO PAULO: Santo André: Paranapiacaba (AAG-UFU 5024; MCP 13115).

Adenomera andreae—BRAZIL: AMAPÁ: Serra do Navio (AAG-UFU 5994, 6006–07, CFBH 43259, 43265); AMAZONAS: Manaus (INPA-H 34045, 34048, 34073–74, 34076, 34081–82, 34084–86, 34090; ZUEC 3937, 3969, 3973–74, 7799); PARÁ: Belém (AAG-UFU 2797–98); Nova Timboteua (AAG-UFU 2788–94).

Adenomera araucaria—BRAZIL: RIO GRANDE DO SUL: Bom Jesus (paratypes: MCP 3345–46); São Francisco de Paula (holotype: MCP 2421; paratypes: MCP 1794, 1849, 3208–09, 3463, 3672, 3676–77; topotypes: CFBH 43927–29, MCP 9896, 11199, 11241, 12055); SANTA CATARINA: Ilhota: Morro do Baú (MCP 1346–52); Indaial: PARNA da Serra do Itajaí (CFBH 43208); Jaraguá do Sul (CFBH 39262); Lebon Régis (MCP 9647, 9672); São Bento do Sul (UFMG 10189–90).

Adenomera bokermanni—BRAZIL: SANTA CATARINA: Garuva (CFBH 43152, 43154); Itapoá (LHUFCG 181–86; MCP 12783–84).

Adenomera cotuba—BRAZIL: GOIÁS: Teresina de Goiás (holotype: AAG-UFU 1400; paratypes: AAG-UFU 0808, 1397–99, 1401–04).

Adenomera diptyx—BRAZIL: MATO GROSSO: Cáceres (AAG-UFU 5366); Santo Antônio de Leverger: São Vicente (AAG-UFU 1435–1438).

Adenomera engelsi—BRAZIL: SANTA CATARINA: Anitápolis (CFBH 20226–27); Florianópolis (holotype: MCP 6415; paratypes: MCP 6379, 6439–40, 7704–05, 8255–56, 8266–67; topotype: MCP 6344); Rancho Queimado (CFBH 13590–91); Santo Amaro da Imperatriz (AAG-UFU 3569–70; CFBH 43213–15); São Bonifácio (CFBH 43182).

Adenomera heyeri—BRAZIL: PARÁ: Oriximiná: ESEC-Grão-Pará (MPEG 30099–101).

Adenomera hylaedactyla—BRAZIL: ACRE: Cruzeiro do Sul (AAG-UFU 5907–11); Feijó (AAG-UFU 5895–97); AMAZONAS: Manaus (INPA-H 22410–13, 26606–09); São Gabriel da Cachoeira (AAG-UFU 3859–66); RORAIMA: Cantá (AAG-UFU 5540–43).

Adenomera juikitam—BRAZIL: GOIÁS: Teresina de Goiás (holotype: AAG-UFU 1406; paratypes: AAG-UFU 0807, 1405).

Adenomera lutzi—BRAZIL: RORAIMA: Upper Maú River (INPA-H 6247); GUYANA: Potaro-Siparuni (MZUSP 150799–804).

Adenomera marmorata—BRAZIL: RIO DE JANEIRO: Tijuca (MZUSP 83374–76, 84079–81, 84083).

Adenomera martinezi—BRAZIL: PARÁ: Novo Progresso: Cachimbo (holotype: MZUSP 73695; allotype: MZUSP 73684; topotypes: AAG-UFU 1515–25).

Adenomera nana—BRAZIL: SANTA CATARINA: Jaraguá do Sul (MCP 8149–50); Joinville (MCP 8633); São Bento do Sul (MCP 8751–55).

Adenomera saci—BRAZIL: GOIÁS: Alto Paraíso de Goiás (holotype: AAG-UFU 1339; paratypes: AAG-UFU 0108–09, 0762–63; ZUEC 3287).

Adenomera thomei—BRAZIL: ESPÍRITO SANTO: Linhares: Povoação (AAG-UFU 6185–86).

Appendix 2. Information on Sound Recordings

Adenomera kweti (type locality).—Morro das Pedras trail, Serra do Tabuleiro State Park, São Bonifácio, state of Santa Catarina, southern Brazil:

(1) Unvouchered recording
Sound file—TRC16b; recorded on 16 November 2017, at 1604 h, air: 22.9°C.
(2) Voucher: male, CFBH 43183 (paratype)
Sound file—TRC17b; recorded on 16 November 2017, at 1649 h, air: 19.6°C.
(3) Voucher: male, CFBH 43184 (holotype)
Sound file—TRC18; recorded on 16 November 2017, at 1656 h, air: 19.6°C.
(4) Voucher: male, CFBH 43185 (paratype)
Sound file—TRC19; recorded on 16 November 2017, at 1709 h, air: 19.6°C.

Adenomera kweti (other localities).—Poção, Córrego Grande, Florianópolis (Santa Catarina Island), southern Brazil:

(1) Voucher: male, UFMG 7088
Sound file—PPGT15; recorded on 18 November 2010. Time of recording and temperature unspecified.

Adenomera araucaria (type locality).—São Francisco de Paula, state of Rio Grande do Sul, southern Brazil:

(1) Unvouchered recording
Sound file—TRC144b; recorded on 12 October 2018, at 1734 h, air: 19.6°C.
(2) Voucher: male, CFBH 43927
Sound file—TRC146b; recorded on 12 October 2018, at 1739 h, air: 19.6°C.
(3) Unvouchered recording
Sound file—TRC150; recorded on 12 October 2018, at 1804 h, air: 19.6°C.

Adenomera araucaria (other localities).—Recording 1 was obtained from Jaraguá do Sul; 2–10 from São Bento do Sul. Both localities in the state of Santa Catarina, southern Brazil:

(1) Voucher: male, CFBH 39262
Sound file—PPGT16; recorded on 11 November 2014. Time of recording and temperature unspecified.
(2) Unvouchered recording
Sound file—PPGT17; recorded on 13 December 2011. Time of recording and temperature unspecified.
(3) Unvouchered recording
Sound file—PPGT18; recorded on 13 December 2011. Time of recording and temperature unspecified.
(4) Voucher: male, UFMG 10191
Sound file—PPGT19; recorded on 13 December 2011. Time of recording and temperature unspecified.
(5) Unvouchered recording
Sound file—PPGT20; recorded on 13 December 2011. Time of recording and temperature unspecified.
(6) Unvouchered recording
Sound file—PPGT21; recorded on 13 December 2011. Time of recording and temperature unspecified.
(7) Voucher: male, UFMG 10192
Sound file—PPGT22; recorded on 13 December 2011. Time of recording and temperature unspecified.
(8) Unvouchered recording
Sound file—PPGT23; recorded on 13 December 2011. Time of recording and temperature unspecified.
(9) Unvouchered recording
Sound file—PPGT24; recorded on 13 December 2011. Time of recording and temperature unspecified.
(10) Unvouchered recording
Sound file—PPGT25; recorded on 13 December 2011. Time of recording and temperature unspecified.

Acoustic-based species assignments were also based on the following sound recordings from the state of Santa Catarina in southern Brazil (1–3: *A. araucaria*; 4 and 5: *A. kweti*):

(1) Unvouchered recording
Sound file—FNJV 32401; recorded by Karoline Ceron, from Parque Estadual da Serra Furada, in Orleans, on 12 September 2014, at 1710 h.
(2) Unvouchered recording
Sound file—AK22B12; recorded from Blumenau, on 12 December 2004, at 1540 h, air: 21.0°C.
(3) Unvouchered recording
Sound file—AK27A06; recorded from Lebon Régis, on 18 January 2006, at 1730 h, air: 23.7°C.
(4) Voucher: male, MCP 8212
Sound file—AK22B23/AK22B25; recorded from Santo Amaro da Imperatriz, on 15 December 2004, at 1700 h, air: 21.0°C.
(5) Unvouchered recording
Sound file—AK21A07; recorded from Taquaras, in Rancho Queimado, on 26 November 2004, at 1645 h, air: 25.5°C.

APPENDIX 3. Acoustic definitions and terminology.

Acoustic traits	Brief description
Time domain	
Call traits	
Note duration (sec)	From initial 10% amplitude to final 10% of note amplitude
Note rise time (%)	Point of maximum amplitude relative to note duration
Note rate (notes/minute)	Note number minus 1, divided by the duration between the onset of first and last notes
Pulse traits	
Pulse duration (msec)	From initial 10% to final 10% of pulse amplitude
Pulse rate (pulses/sec)	Pulse number minus 1, divided by duration of peak-to-peak from first to last pulse of the note
Frequency domain	
Call traits	
Fundamental frequency (Hz)	Peak frequency of the first harmonic
Dominant frequency (Hz)	Frequency containing the greatest energy in the note
Linear frequency modulation (Hz)	Difference between the peak frequency from final 10% to initial 10% of note amplitude

APPENDIX 4. Terminals and accession numbers of sequences included in the phylogenetic and genetic distance analyses.

Species	GenBank ID	Phylogenetic matrix	Genetic distance matrix
Hydrolaetare caparu	KC603988	Yes	No
Lithodytes lineatus	KC604003	Yes	No
Leptodactylus rhodomystax	KC603993	Yes	No
Adenomera ajurauna	KF674770	Yes	Yes
A. ajurauna	KF674772	No	Yes
A. ajurauna	KF674771	No	Yes
A. ajurauna	KF674773	No	Yes
A. ajurauna	KF674774	No	Yes
Adenomera andreae	KF674556	Yes	No
Adenomera araucaria	KF674754	Yes	Yes
A. araucaria	KF674755	Yes	Yes
A. araucaria	MK458586	Yes	Yes
A. araucaria	MK458587	Yes	Yes
A. araucaria	MK458591	Yes	Yes
A. araucaria	MK458592	Yes	Yes
A. araucaria	MK458593	Yes	Yes
A. araucaria	MK458594	Yes	Yes
Adenomera bokermanni	KF674806	Yes	Yes
A. bokermanni	KF674807	Yes	Yes
Adenomera diptyx	KF674600	Yes	No
Adenomera engelsi	KF674764	Yes	Yes
A. engelsi	KF674765	Yes	Yes
A. engelsi	KF674766	Yes	Yes
Adenomera heyeri	KF674700	Yes	No
Adenomera kweti	MK458583	Yes	Yes
A. kweti	MK458584	Yes	Yes
A. kweti	MK458585	Yes	Yes
A. kweti	MK458588	Yes	Yes
A. kweti	MK458589	Yes	Yes
A. kweti	MK458590	Yes	Yes
A. kweti	KF674756	Yes	Yes
A. kweti	KF674757	Yes	Yes
Adenomera lutzi	KF674685	Yes	No
Adenomera marmorata	KF674783	No	Yes
A. marmorata	KF674784	No	Yes
A. marmorata	KF674785	No	Yes
A. marmorata	KF674782	No	Yes
A. marmorata	KF674786	No	Yes
A. marmorata	KF674778	Yes	Yes
A. marmorata	KF674777	No	Yes
A. marmorata	KF674781	No	Yes
A. marmorata	KF674779	No	Yes
A. marmorata	KF674775	No	Yes
A. marmorata	KF674776	No	Yes
A. marmorata	KF674780	No	Yes
A. marmorata	KF674787	No	Yes
Adenomera martinezi	KF674697	Yes	No
Adenomera nana	KF674767	Yes	Yes
A. nana	KF674768	Yes	Yes
A. nana	KF674769	Yes	Yes
Adenomera thomei	KF674793	Yes	No
Adenomera sp. A	KF674803	Yes	No
Adenomera sp. J	KF674751	Yes	No
Adenomera sp. K	KF674802	Yes	No
Adenomera sp. N	KF674788	Yes	No
Adenomera sp. O	KF674789	Yes	No
Adenomera sp. S	KF674762	Yes	No