



## Advertisement call and male morphology of the Malagasy treefrog *Boophis arcanus* from the Ranomafana region, south-eastern Madagascar

JANA C. RIEMANN<sup>1,5</sup>, SERGE H. NDRIANTSOA<sup>1,2</sup>, MIGUEL VENCES<sup>3</sup>,  
MARK-OLIVER RÖDEL<sup>4</sup> & JULIAN GLOS<sup>1</sup>

<sup>1</sup>Biocenter Grindel, University of Hamburg, Martin-Luther-King Platz 3, 20146 Hamburg, Germany

<sup>2</sup>Département de Biologie Animale, Université d'Antananarivo, Antananarivo 101, Madagascar

<sup>3</sup>Zoological Institute, Technical University of Braunschweig, Mendelssohnstr. 4, 38106 Braunschweig, Germany

<sup>4</sup>Museum für Naturkunde, Leibniz Institute for Research on Evolution and Biodiversity at the Humboldt University Berlin, Herpetology, Invalidenstr. 43, 10115 Berlin, Germany

<sup>5</sup>Corresponding author's E-mail: Jana.Riemann@uni-hamburg.de

With over 70 nominal species, treefrogs of the genus *Boophis* (family Mantellidae) represent one of the most species-rich lineages in the amphibian fauna of Madagascar (Cadle 2003). Most *Boophis* are characterized by relatively loud and conspicuous calls, but specimens can be difficult to collect because males are often perched high on trees. Therefore, many species are so far only known from a few specimens, usually males. An exception is the recently described species *Boophis arcanus*, which is only known from two female specimens and for which no further biological data are available (Glaw *et al.* 2010). The two females were found at night sitting on trees and bushes in the rainforest of Mahakajy, a private reserve close to Ranomafana National Park (RNP) in the southern central east of Madagascar. Based on recordings and specimens collected during recent survey work in RNP and nearby forest fragments, we here describe male morphology and the advertisement call of this species.

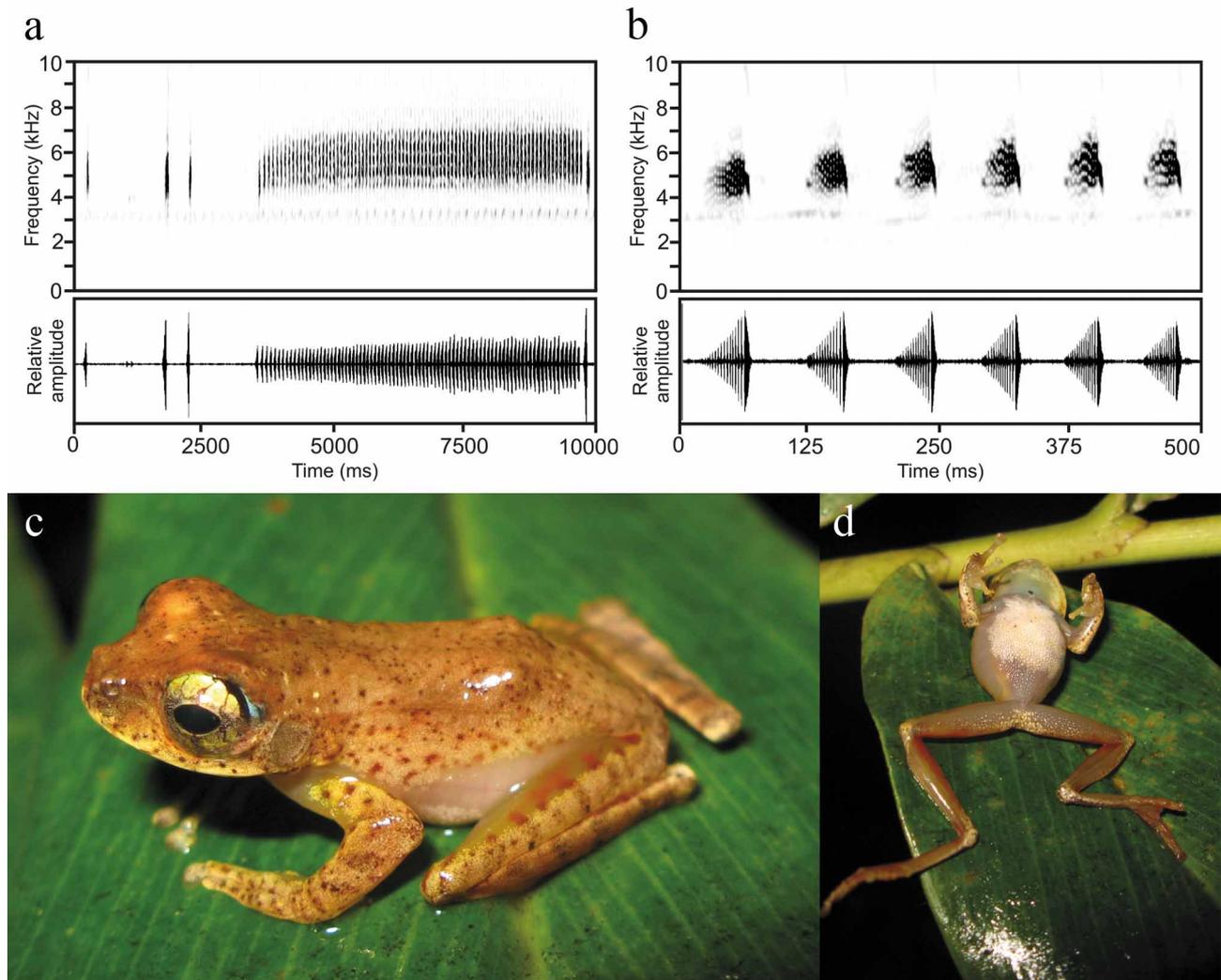
Specimens were collected during an opportunistic survey at night. Locality information was taken with a Garmin etrex VISTA HCx GPS receiver. Specimens were deposited at the Museum für Naturkunde, Berlin (ZMB) and identified based on DNA sequences of a fragment of the 16S rRNA gene. Vocalization was recorded in the field using a Roland EDIROL R-09 recorder with internal microphone (sampling frequency: 44.1 kHz, recording mode: wav 24 bit). The call was analysed with the software Cool edit Pro 2.0 (values presented as mean  $\pm$  standard deviation).

The Ranomafana region comprises mid-altitude (500–1300 m a.s.l.) rainforest with a mean annual rainfall of 1700–4300 mm (Wright & Andriamihaja 2003). Most of the remaining forest habitat is located inside RNP (43,500 ha). The area around the park consists of a fragmented landscape with forest patches that are embedded in a matrix of cultivated fields (rice, bananas) and secondary vegetation; narrow gallery forests often grow along small streams. Five males of *Boophis arcanus* were observed on June 1<sup>st</sup> 2010, between 19:30–20:30 h along a small stream running through rice fields and degraded land bordered by some riparian forest in Beremby (21°14.426' S, 047°31.558' E, app. 620 m a.s.l.), near RNP. All five males were found on five different neighbouring trees sitting on leaves in 1.5 m height. We collected two voucher specimens of which one individual (ZMB 77315) was observed calling.

The single analysed call of *B. arcanus* (ZMB 77315) contains two different note types (Fig. 1a, and b). The call starts with a series of three pulsed notes of type 1, emitted in irregular intervals (interval between first and second note: 1437 ms, interval between second and third note: 374 ms). After an interval of 1262 ms, a series of 85 pulsed notes of note type 2 follows, separated by only very short intervals of 35–72 ms ( $41.4 \pm 11.1$  ms,  $n = 10$ ) at the beginning and only 24–32 ms ( $26.7 \pm 2.3$  ms,  $n = 10$ ) at the end of the series. Another single note of note type 1 follows after an interval of 81 ms ending the call. Note type 1 shows the following characters: note duration, 61–86 ms ( $70 \pm 11.9$  ms,  $n = 4$ ); number of pulses, 11–23 ( $15.3 \pm 5.3$ ,  $n = 4$ ); dominant frequency range, ca 4000–6000 Hz; maximum call energy, 4900 Hz. Characters of note type 2: note duration, 37–42 ms ( $39.4s \pm 1.8$  ms,  $n = 10$ ) at the beginning and 43–55 ms ( $49.3 \pm 3.1$  ms,  $n = 10$ ) at the end; number of pulses, 11–14 ( $12.2 \pm 1.0$ ,  $n = 10$ ) at the beginning and 18–25 ( $21.5 \pm 1.8$ ) at the end; note repetition rate, 13.7 notes/s; dominant frequency range, 4600–7000 Hz; and four energy maxima, 4800 Hz, 5300 Hz, 5800 Hz, 6500 Hz.

Morphological measurements of two preserved males were taken by MV using a caliper (all in mm; specimen ZMB 77315 (field and tissue number JG\_400), with ZMB 77316 (field and tissue number JG\_401) in parentheses; GenBank accession numbers JQ413974 – JQ413975): snout-vent length 24.3 (24.8); head width 9.1 (9.2); head length 8.7 (9.1); horizontal tympanum diameter 2.0 (1.8); horizontal eye diameter 3.6 (3.9); eye-nostril distance 1.9 (2.0); nostril-snout tip

distance 1.9 (2.0); nostril-nostril distance 2.7 (2.5); hand length 7.2 (8.1); forelimb length 15.2 (15.3); hindlimb length 37.6 (42.0); foot length including tarsus 15.6 (17.5); foot length 8.3 (9.5); tibia length 13.3 (13.4). Additional morphological characters (based on specimen ZMB 77316): tibiotarsal articulation reaches nostril when limbs are adpressed along body; vomerine teeth present; posteromedial to choanae; poorly visible whitish nuptial pads; webbing formula between toes 1(0), 2i(0.5), 2e(0), 3i(1), 3e(0), 4i(1), 4e(1), 5(0.25). Other characters are as in female holotype (Glaw *et al.* 2010). Snout-vent lengths of three additional males measured in the field were 24.5, 26.6, and 25.6 mm.



**FIGURE 1.** Sonograms and oscillograms of the advertisement call (a, b), and colour in life (c, d) of *Boophis arcanus* males from Beremby, Ranomafana region, Madagascar. Graph a) shows the entire call sequence, with three notes of note type 1, a series of type 2 notes, and a final note of type 1, recorded from specimen ZMB 77315. Graph b) represents the first six notes of the note series (of type 2 notes) and shows the pulsed structure of these notes. Photographs show dorsolateral (c) and ventral (d) view of male specimen ZMB 77316 (SVL= 24.8 mm).

Colour in life of the collected specimens (Fig. 1c, and d) was very similar to that of the female type specimens (Glaw *et al.* 2010): light brown dorsum with irregular dark spotting and an unpigmented area in the inguinal region; iris yellowish with vertical brown reticulations especially in its upper half; iris periphery turquoise whitish; chest and belly whitish, concealed dorsal portions of shanks and thighs and ventral surface of feet with a reddish shade.

In the multi-gene analysis of Wollenberg *et al.* (2011), *B. arcanus* resulted with high support as part of a *Boophis* subclade also containing *B. feonnyala*, *B. miniatus*, *B. picturatus*, and *B. piperatus*, and sister to a clade containing the latter three species. All of these species are characterized by calls consisting mainly of note series with the following temporal characteristics (Vences *et al.* 2006; Glaw *et al.* 2001, 2010): *B. feonnyala*, note duration 64–205 ms, note repetition rate 5.3 notes per second; *B. miniatus*, 38–85 ms, 11–12/s; *B. picturatus*, 47–131 ms, ca. 2.5/s; *B. piperatus*, 135–283 ms, 3.1–3.4/s. Based on these variables, the call of *B. arcanus* (37–55 ms; 13.7/s in series of note type 2) is most

similar to that of *B. miniatus* and has a distinctly shorter note duration and a faster note repetition rate than the other species. The male specimens of *B. miniatus* also resemble those of *B. arcanus* as examined herein by having a relatively large snout-vent length (23.1–26.6 mm) and large relative tympanum size (about 50% of eye diameter). However, *B. miniatus* does not constitute the sister species of *B. arcanus* (Wollenberg *et al.* 2011) and differs by a high pairwise difference in mitochondrial genes (7.2% in the 16S rRNA gene; Genbank accession number AY848639), confirming the status of *B. arcanus* as separate species and indicating that the similarities among these two taxa might be plesiomorphic.

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## References

- Cadle J.E. (2003) *Boophis*. In: Goodman, S.M. & Benstead, J.P. (Eds.), *The Natural History of Madagascar*. University of Chicago Press, Chicago, pp. 916–919.
- Glaw, F., Köhler, J., de la Riva, I., Vieites, D.R. & Vences, M. (2010) Integrative taxonomy of Malagasy treefrogs: combination of molecular genetics, bioacoustics and comparative morphology reveals twelve additional species of *Boophis*. *Zootaxa*, 2383, 1–82.
- Glaw, F., Vences, M., Andreone, F. & Vallan, D. (2001) Revision of the *Boophis majori* group (Amphibia: Mantellidae) from Madagascar, with descriptions of five new species. *Zoological Journal of the Linnean Society*, 133, 495–529.
- Vences, M., Glaw, F. & Márquez, R. (2006) *The Calls of the Frogs of Madagascar*. 3 Audio CDs and booklet, Alosa-Fonozoo, Barcelona, 44 pp.
- Wollenberg, K.C., Vieites, D.R., Glaw, F. & Vences, M. (2011) Speciation in little: the role of range and body size in the diversification of Malagasy mantellid frogs. *BMC Evolutionary Biology*, 11, article 217.
- Wright, P.C. & Andriamihaja, B. (2003) The conservation value of long-term research: a case study from the Parc National de Ranomafana. In: Goodman, S.M. & Benstead, J.P. (Eds.), *The Natural History of Madagascar*. University of Chicago Press, Chicago, pp. 1485–1488.