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A Remarkable Ontogenetic Change in Color Pattern in a New Species of *Oreophryne* (Anura: Microhylidae) from Papua New Guinea

Fred Kraus[1] and Allen Allison[1]

Official Publication of the American Society of Ichthyologists and Herpetologists

A Remarkable Ontogenetic Change in Color Pattern in a New Species of *Oreophryne* (Anura: Microhylidae) from Papua New Guinea

Fred Kraus[1] and Allen Allison[1]

We describe a new species of scandent/arboreal frog from Sudest Island, Louisiade Archipelago, off the southeastern tip of New Guinea that exhibits a remarkable ontogenetic change in color pattern. Juveniles are shiny black with lemon-yellow spots; adults are uniform peach with bright blue eyes. We detail the ontogenetic changes in color-pattern elements that comprise this transformation. This ontogenetic change, uniform peach adult coloration, and blue iris all appear to be unique features within the genus *Oreophryne*, most of whose members are tan, brown, or gray. We presume the striking juvenile color pattern to serve an aposematic function, but this conjecture requires testing. The new species is restricted to a relatively small patch of cloud forest perched on the highest peak of Sudest Island. Climate change may pose a threat to the new species if changing rainfall or temperature regimes result in the loss of this forest.

AMONG frogs, ontogenetic changes in color pattern are generally of a modest nature. Hoffman and Blouin (2000) noted that such changes have been observed for at least 39 species. However, most of these instances involve darkening of animals with age or ontogenetic transitions between green and brown color states. Relatively few frogs show striking ontogenetic differences in color or pattern, and most of these examples are seen in the African genus *Hyperolius*. Many species of that genus ontogenetically lose or acquire pattern elements such as stripes, blotches, mottling, or hourglass markings, or they change colors with age, often acquiring striking color patterns (Schiotz, 1971, 1999; Hoffman and Blouin, 2000). A further example of remarkable change is seen in *Nectophryne*. In *N. afra*, juveniles are black and bear several bluish-white lines or rings, whereas adults are black or brown with dorsolateral stripes that are brown or yellow (Scheel, 1970). In *N. batesii*, juveniles are black with silver bands and stripes around the body and legs (Blackburn and Droissart, 2008), whereas adults are brown with dark blotches (Boulenger, 1913). With the exception of these African bufonid and hyperoliid genera, strikingly different color-pattern schemes between juveniles and adults are rare in frogs.

The genus *Oreophryne* is one of the largest known components of the Papuan microhylid fauna, with 33 species currently named from New Guinea and immediately adjacent islands and another ten species named from Indonesian islands to the west ($n = 8$) or from the Philippines ($n = 2$). Most species of this genus are arboreal, although species living in the alpine grasslands of New Guinea are terrestrial (Zweifel et al., 2005). The genus has long been a taxonomic morass because several valid names have until recently been erroneously placed in synonymy or improperly applied, some of the early described species remain unassociated with natural populations, and many additional species await description (Zweifel et al., 2003). Even so, among the Papuan species, more than half have been described since 2000. In the course of conducting herpetofaunal surveys in Papua New Guinea we discovered a new species of *Oreophryne* that is remarkable for its drastic ontogenetic change in color pattern, a phenomenon that is, as noted above, relatively rare in frogs. We take this opportunity to describe this species and its color ontogenesis here.

MATERIALS AND METHODS

Specimens were fixed in 10% buffered formalin and transferred to 70% ethanol for storage. All measurements were made with digital calipers or an optical micrometer to the nearest 0.1 mm, with the exception that disc widths were measured to the nearest 0.01 mm. Measurements, terminology, and abbreviations follow Zweifel (1985) and Kraus and Allison (2006): body length from snout to vent (SVL); tibia length from heel to outer surface of flexed knee (TL); horizontal diameter of eye (EY); distance from anterior corner of eye to center of naris (EN); internarial distance, between centers of external nares (IN); distance from anterior corner of eye to tip of snout (SN); head width at widest point, typically at the level of the tympana (HW); head length, from tip of snout to posterior margin of tympanum (HL); horizontal tympanum diameter (TY); width of the third finger disc (3rd F); width of the fourth toe disc (4th T). We recorded calls in the field using a Sennheiser ME66 microphone and a Sony MDSJE480 minidisc recorder. Call structure was analyzed using the computer program Avisoft-SASLab Pro, v4.51 (Avisoft Bioacoustics; http://www.avisoft.com/). All latitude and longitude coordinates use the Australian Geodetic Datum, 1966 (AGD 66).

We confirmed by dissection generic assignment of the frogs using the presence of an eleutherognathine jaw and reduction of clavicles and procoracoids (Parker, 1934). Comparisons to congeners relied on direct comparison to museum material and to information provided in original descriptions by Roux (1910), Parker (1934), Richards and Iskandar (2000), Günther et al. (2001), Günther (2003a, 2003b), Zweifel (2003), and Zweifel et al. (2003, 2005).

A sample of one adult male (BPBM 20468) and one juvenile (BPBM 20474) was used to assess mitochondrial DNA (mtDNA) variation between the two extremes of color-pattern variation to verify conspecificity. Genomic DNA was extracted from approximately 5 mg of ethanol-preserved liver tissue using a DNeasy Blood and Tissue Kit (Qiagen, Inc., Valencia, CA) following the recommended protocol. Two fragments of approximately 520 bp of the mitochondrial 16S gene and 380 bp of the cytochrome *c* oxidase subunit 1 (CO1) gene were amplified and sequenced using the primers 16S-L and 16S-H for 16S (Köhler and Günther, 2008), and LepF1 and LepRI for CO1 (Smith et al., 2008). Reactions were performed in a 50 mL solution containing 1–

[1] Department of Natural Sciences, Bishop Museum, 1525 Bernice Street, Honolulu, Hawaii 96817; E-mail: (FK) fkraus@hawaii.edu; and (AA) allison@hawaii.edu. Send reprint requests to FK.
Submitted: 19 January 2009. Accepted: 26 May 2009. Associate Editor: D. Kizirian.
 DOI: 10.1643/CH-09-015

2 ng genomic DNA, 200 mM of each dNTP, 2.5 units Taq DNA polymerase (D4545, Sigma Chemical, St. Louis, MO), 3 mM $MgCl_2$, each primer at 0.8 mM, and 1× PCR buffer. PCR cycling parameters were as published in Smith et al. (2008) for CO1 and Köhler and Günther (2008) for 16S. PCR product was gel extracted using QIAQuick Gel Extraction Kit (Qiagen, Inc.). Cycle sequencing of 100 fmol double-stranded PCR product was carried out with each primer using a CEQ DTCS Quick Start Kit (Beckman-Coulter, Fullerton, CA) following the recommended protocol, and the products sequenced on a CEQ8000 genetic analysis system. Both strands were sequenced, and sequences were aligned to minimize codon differences. All sequences are deposited in GenBank (FJ625827–30).

Oreophryne ezra, new species

Figures 1, 2; Table 1

Holotype.—BPBM 20468 (field tag FK 9759), 24.7 mm SL, male, Papua New Guinea, Milne Bay Province, Sudest Island, W slope Mt. Rio, 11.5054347°S, 153.429945°E, 630–730 m, 23 April 2004, F. Kraus.

Paratypes.—(12 adults, 23 juveniles) BPBM 20467, W slope Mt. Rio, 11.5058876°S, 153.4294871°E, 650 m, Sudest Island, 21 April 2004; BPBM 20469–73, 20483–85, PNGM 24086–88, same data as holotype; BPBM 20474–82, W slope Mt. Rio, 11.5081521°S, 153.4308608°E, 730–800 m, Sudest Island, 23 April 2004; BPBM 20486, W slope Mt. Rio, 11.5058876°S, 153.429945°E, 680 m, 25 April 2004; BPBM 20487–91, PNGM 24089–90, W slope Mt. Rio, 11.50659°S, 153.4305°E, 730 m, 25 April 2004; BPBM 20492–95, PNGM 24091–92, same data as BPBM 20474, except collected 25 April 2004.

Diagnosis.—A moderately sized species of *Oreophryne* (male SVL = 22.3–26.1 mm, $n = 10$, female SVL = 25.4 mm, $n = 1$) distinguished from all other members of the genus in having the combination of a ligamentous connection of the procoracoid to the scapula, well-developed webbing between the toes, fifth toe longer than the third, relatively wide head (HW/SVL = 0.39–0.42), relatively long snout (EN/SVL = 0.097–0.108), smooth or finely granular dorsal skin, blue iris, red-orange flash markings in the groin and hidden surfaces of the legs, and a remarkable ontogenetic change in color pattern from black with yellow spots in juveniles to uniform light peach in adults (Fig. 1). The uniformly pale peach dorsal color of adults is only shared with a small percentage of adults of *O. inornata* (most of which have a yellower ground color and irregular brown dorsal markings). The particular form of ontogenetic color-pattern change seen in *Oreophryne ezra* is apparently unique among frogs.

Comparisons with other species.—The new species differs from *O. alticola*, *O. asplenicola*, *O. brevicrus*, *O. clamata*, *O. crucifera*, *O. flava*, *O. habbemensis*, *O. idenburgensis*, *O. kampeni*, *O. notata*, *O. pseudasplenicola*, and *O. waira* in lacking (vs. having) a procoracoid that reaches the scapula, with the procoracoid reaching the middle of the coracoid and being connected to the scapula by a ligament. It differs from *O. atrigularis*, *O. brevirostris*, *O. geminus*, *O. minuta*, *O. terrestris*, and *O. wapoga* in having (vs. lacking) webbing between the toes. It differs from *O. insulana*, *O. kapisa*, *O. sibilans*, and *O. unicolor* in having well-developed webbing between the toes that reaches the basal tubercles of T1–T5 (vs. having only basal webbing). It differs from *O. albopunctata*, *O. biroi*, *O. hypsiops*, and *O. mertoni* in having the fifth toe longer than the third (vs. both toes of equivalent size). Of the remaining species, *Oreophryne ezra* differs from *O. anthonyi* in its much smaller size (adult SVL = 22.3–26.1 mm vs. 30–47 mm in *O. anthonyi*), longer snout (EN/SVL = 0.097–0.108 vs. 0.079–0.086 in *O. anthonyi*), and chattering call (vs. a series of barks in *O. anthonyi*); from *O. brachypus* in its wider head (HW/SVL = 0.39–0.42 vs. 0.34–0.38 in *O. brachypus*), longer snout (EN/SVL = 0.097–0.108 vs. 0.077–0.089 in *O. brachypus*), and in lacking (vs. having) webbing between the fingers; from *O. geislerorum* in its wider head (HW/SVL = 0.39–0.42 vs. 0.32–0.38 in *O. geislerorum*), uniform peach color pattern (vs. brown or gray with darker stripes in *O. geislerorum*), and chattering call (vs. a short buzz in *O. geislerorum*); from *O. inornata* in its smaller size (adult SVL = 22.3–26.1 mm vs. 28–41 mm in *O. inornata*), longer snout (EN/SVL = 0.097–0.108 vs. 0.078–0.098 in *O. inornata*), blue (vs. tan) iris, and chattering call (vs. single honk in *O. inornata*); from *O. loriae* in its wider head (HW/SVL = 0.39–0.42 vs. 0.35–0.39 in *O. loriae*), smooth dorsal skin (vs. with scattered small pustules), blue (vs. tan or bronze) iris, red-orange flash markings (vs. none in *O. loriae*), and chattering call (vs. single honk in *O. loriae*); from *O. parkeri* in its wider head (HW/SVL = 0.39–0.42 vs. 0.29–0.35 in *O. parkeri*), smooth (vs. pustulose) dorsal skin, longer snout (EN/SVL = 0.097–0.108 vs. 0.071–0.087 in *O. parkeri*), and uniform peach color pattern (vs. brown or gray with numerous white flecks in *O. parkeri*); and from *O. wolterstorffi* in its wider head (HW/SVL = 0.39–0.42 vs. 0.35 in the single known specimen of *O. wolterstorffi*), longer (EN/SVL = 0.097–0.108 vs. 0.082 in *O. wolterstorffi*) and broader (IN/SVL = 0.082–0.098 vs. 0.077 in *O. wolterstorffi*) snout, and uniform peach color pattern (vs. brown spotted with gray in *O. wolterstorffi*).

Description of holotype.—Incision on left side. Head wide (HW/SVL = 0.40), with steep, slightly concave loreal region. Canthus rostralis rounded, straight when viewed from above. Nostrils directed laterally, much closer to tip of snout than to eyes. Internarial distance shorter than distance from naris to eye (EN/IN = 1.09, IN/SVL = 0.089, EN/SVL = 0.10). Snout truncate when viewed from the side, shallowly angulate when viewed from above. Eyes moderately large (EY/SVL = 0.13); eyelid approximately two-thirds width of interorbital distance. Tympanum indistinct and small (TY/SVL = 0.040), partly sunken in surrounding flesh. Skin smooth with network of weakly raised vermiculations; ventral surfaces granular. Supratympanic fold absent. Fingers unwebbed, bearing discs with terminal grooves; relative lengths 3>4>2>1. Finger discs approximately twice widths of penultimate phalanges. Subarticular tubercles low but distinct; inner metacarpal tubercle large, oval, well developed, but low; outer obscure. Toes webbed, webbing reaching basal subarticular tubercles on T3–T5; bearing discs with terminal grooves; relative lengths 4>5>3>2>1. Toe discs smaller than those of fingers (3^{rd}F/4^{th}T = 1.32), somewhat less than twice width of penultimate phalanges. Subarticular tubercles low but distinct; inner metatarsal tubercle large, oval; outer small and round. Hind legs of moderate length (TL/SVL = 0.43).

Dorsum tan with minute scattered black punctations, these slightly denser on eyelids and top of snout. Limbs same except that sides, forearm insertions, F1 and F2, T1–T3, and most of upper surfaces of thighs pale orange-straw; narrow strip of tan on upper surfaces of thighs. Nares

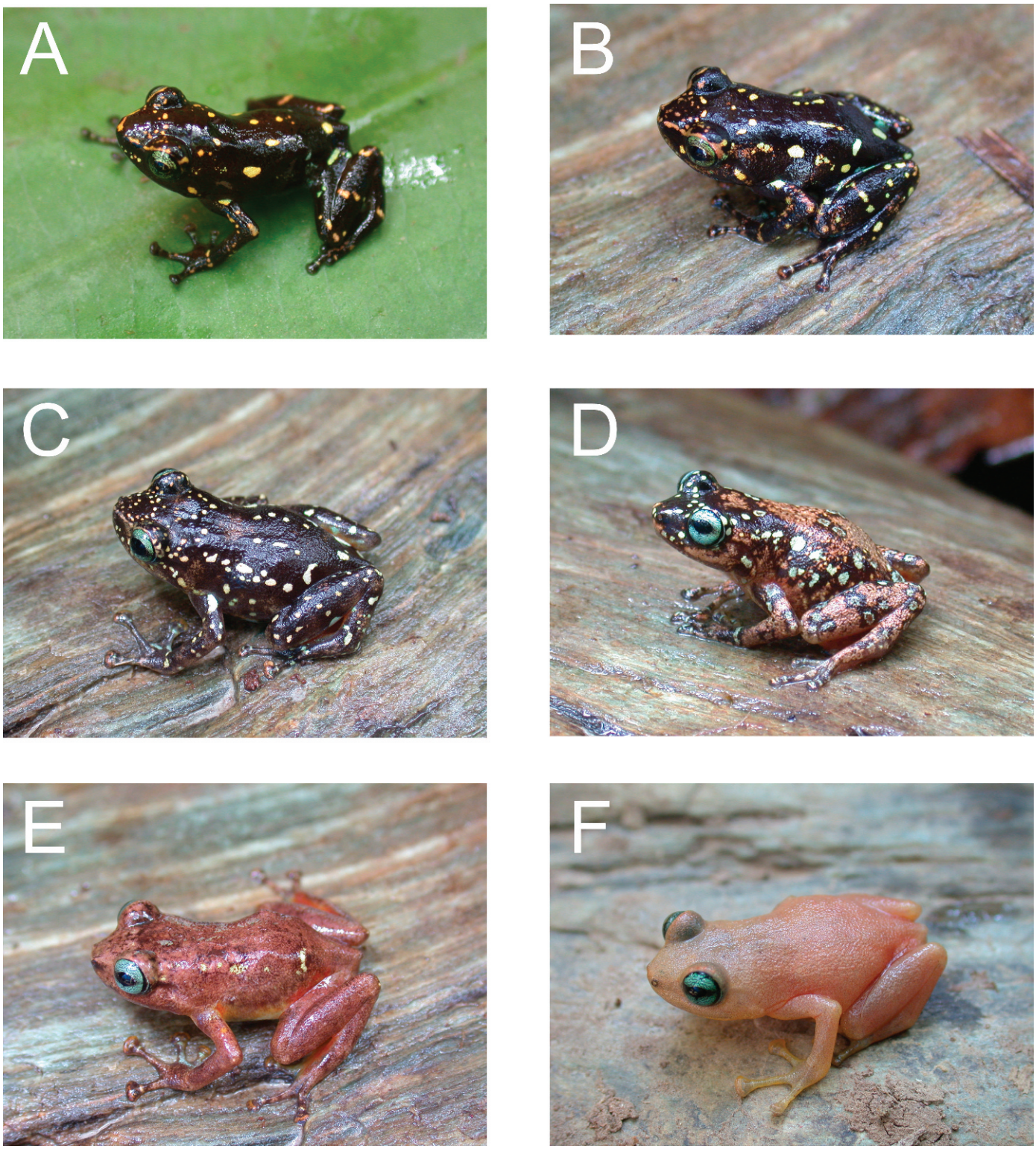

Fig. 1. Portraits showing ontogenetic transformation of dorsal color pattern in *Oreophryne ezra*. (A) BPBM 20474, 12.7 mm SVL; (B) BPBM 20494, 16.5 mm SVL; (C) BPBM 20492, 21.7 mm SVL; (D) BPBM 20486, 26.3 mm SVL; (E) BPBM 20493, 22.6 mm SVL; and (F) BPBM 20467, 24.7 mm SVL. Note progressive change in eye color from greenish bronze to caerulean; lightening, followed by loss, of black pigmentation; lightening, reduction, and loss of yellow spots; and gradual increase in peach coloration.

outlined with dark brown; upper rim of nictitating membrane dark brown. Venter very pale straw with many granules retaining faint wash of white color. Palmar and plantar surfaces pale straw lightly to moderately stippled with brown. Iris silver-white heavily stippled with black.

Measurements (in mm).—SVL = 24.7, TL = 10.6, HW = 9.8, HL = 8.0, IN = 2.2, EN = 2.4, SN = 3.4, EY = 3.3, TY = 1.0, 3^{rd}F = 1.75, 4^{th}T = 1.33.

Mensural variation for the type series is shown in Table 1. Surprisingly little ontogenetic change in morphometric variables occurs in this species, with only the relative size

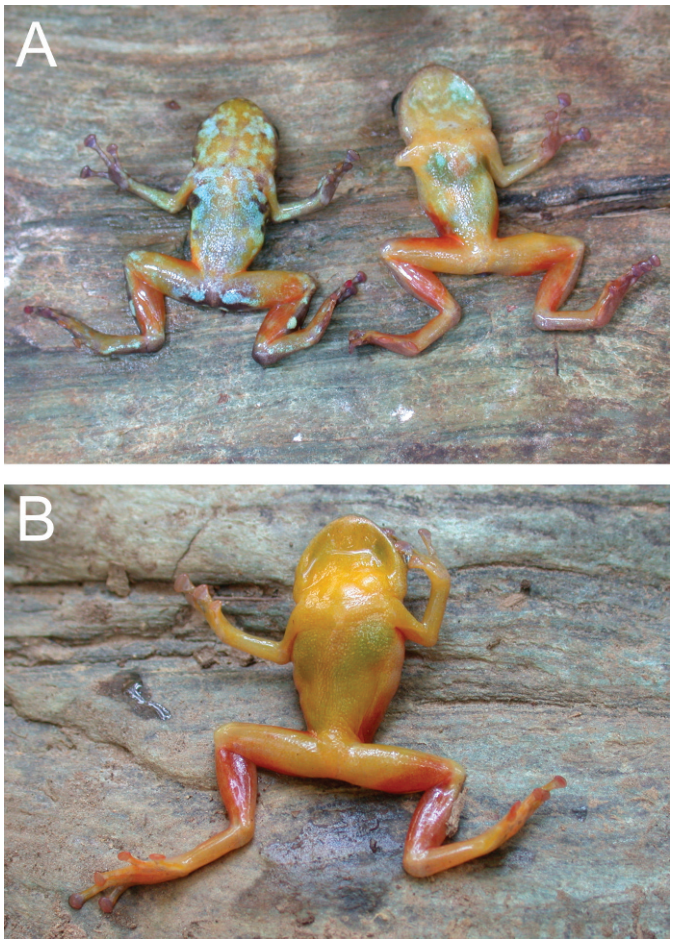

Fig. 2. Portraits showing late ontogenetic transformation of ventral color pattern in *Oreophryne ezra*. (A) Small mature male (BPBM 20492, 22.3 mm SVL, left) and immature female (BPBM 20493, 22.6 mm SVL, right); and (B) fully transformed adult male (BPBM 20467, 25.5 mm SVL). Note late loss of blue flecking and residual dark ventral coloration remaining on BPBM 20492.

of toe discs being smaller in juveniles than in adults. Otherwise, central tendencies for these values are similar between adults and juveniles, although the latter often have a wider range of values (Table 1), no doubt due to the lesser accuracy of measuring smaller frogs.

Ontogenetic change in color pattern is dramatic for this species. Field notes of a typical juvenile (BPBM 20474, Fig. 1A) in life: ''Dorsum and legs shiny black with golden-yellow spots. Venter dark gray mottled with blue. Iris metallic green.'' Another juvenile (BPBM 20475) had the venter flecked with light gray instead of blue; a third (BPBM 20482) had both the dorsum and venter black with minute light gray flecks. But the typical color pattern among juveniles consists of three elements: dorsum shiny black with bright yellow spots, venter dark gray with blue flecks, and iris metallic green (Fig. 1A). In contrast, field notes for a typical adult (Fig. 1F) in life: ''Dorsum uniform pale peach, smooth. Iris pale metallic blue. Entire ventral surface lemon yellow. Axilla, groin, fore and aft of thighs, and under shanks all red-orange.'' As animals age, the black color becomes invaded with pink or peach, first in small flecks (Fig. 1B, C) and then broadening (Fig. 1D, E) to eventually cover the entire dorsum, with no black remaining (Fig. 1F). At the same time, the yellow spots of the juveniles lighten from lemon yellow (Fig. 1A) to cream-yellow (Fig. 1C, D),

Table 1. Comparative Mensural Data for Type Series of *Oreophryne ezra*.

	Adults (n = 13)		Juveniles (n = 23)	
Character	Mean	Range	Mean	Range
SVL (mm)	24.1	22.3–26.1	10.0	6.9–16.0
TL/SVL	0.45	0.43–0.49	0.44	0.41–0.50
EN/SVL	0.104	0.097–0.108	0.103	0.096–0.110
IN/SVL	0.090	0.082–0.098	0.098	0.092–0.108
SN/SVL	0.15	0.14–0.16	0.16	0.14–0.18
TY/SVL	0.044	0.038–0.049	0.042	0.034–0.052
EY/SVL	0.14	0.13–0.16	0.15	0.14–0.17
HW/SVL	0.41	0.39–0.42	0.40	0.38–0.44
HL/SVL	0.35	0.32–0.37	0.36	0.34–0.39
3^{rd}F/SV	0.075	0.069–0.079	0.063	0.054–0.075
4^{th}T/SV	0.056	0.050–0.061	0.044	0.036–0.052
EN/IN	1.15	1.09–1.24	1.04	1.00–1.14
EN/SN	0.70	0.67–0.72	0.67	0.57–1.00
EY/SN	0.95	0.83–1.06	0.95	0.57–1.08
HL/HW	0.81	0.81–0.90	0.91	0.86–0.95
3^{rd}F/4^{th}T	1.33	1.25–1.44	1.44	1.33–1.70

then gradually disappear, although a few will often persist even after the black color has disappeared (Fig. 1E). Ventrally, the dark venter turns yellow, the blue flecks gradually disappear, and red-orange flash colors are added to the axilla, groin, and hidden surfaces of the legs (Fig. 2A), eventually resulting in adult ventral coloration of uniform bright lemon yellow with bright red-orange flash markings (Fig. 2B). Lastly, the metallic green-bronze iris of the juvenile (Fig. 1A) lightens into a bright sky-blue iris in the adult (Fig. 1D–F). This results in the typical adult coloration of a uniform peach or light pink dorsum with a bright blue eye.

Mitochondrial DNA confirms that the two specimens examined (one small and black-and-yellow, one large and uniform-peach color) were invariant across both their 16S (521 bp) and COI (378 bp) sequences. Further, all black-and-yellow forms were clearly non-reproductive (juveniles), and only large specimens that were uniform peach or peach with a few yellow spots were reproductively mature.

Call.—Animals called from dusk into night perched on vegetation or trunks of small trees and produced a rapid, chattering call. Males called approximately every 30–120 s. We recorded 13 calls produced by three individuals (Table 2). The call consists of about 30 pulsed notes (range 29–32) and averages 2.07 s in duration (range 1.94–2.26). This results in a mean repetition rate of 14.8 notes/s (range 14.2–15.7). The duration of the first note is four times longer that that of subsequent notes (Fig. 3A, B), with a mean of 0.20 s (range 0.16–0.25). This note has an average of 74 pulses (range 62–88) and a mean pulse rate of 376 pulses/s (range 258–476). The subsequent notes have a mean duration of 0.04 s (range 0.03–0.04). Each of these notes generally has 3–5 pulses. The mean interval between the first and second note was 0.04 s (range 0.02–0.06) and was about the same as the intervals between subsequent notes (Fig. 4A). This resulted in a repetition rate of 16.1 notes/s (range 15.6–16.7) if the first note was excluded. The call has a dominant frequency of 2470 Hz (range 2040–2670) but with the energy spread over a fairly wide band from ~1000–3000 Hz (Fig. 4B). There is no apparent frequency modulation (Fig. 4C).

Table 2. Call Characteristics of *Oreophryne ezra*. All calls were recorded in the vicinity of the type locality on 23 April 2004. BBBM 20468 (holotype) was recorded at 1900 h. The uncaptured individual was recorded at 1930 h and BPBM 20469 was recorded at 2015 h. Air temperature was 20°C for all three recordings. Parts of call C from BPBM 20468 could not be measured accurately because of interference from a calling insect.

Specimen	Call no.	Total notes	Call duration (s)	Repetition rate (notes/s)	Duration first note (s)	Duration first internote (s)	Total pulses first note	Pulse rate first note (pulses/s)	Mean duration notes 2 to last (s)	Repetition rate notes 2 to last (notes/s)	Dominant frequency (Hz)
BPBM 20468	A	29	1.94	15.0	0.1567	0.0567	71	453.1	0.0404	16.3	2580
	B	30	2.07	14.8	0.1809	0.0471	81	447.8	0.0399	16.2	2580
	C	29	1.97	14.5	0.1980	0.0480	70	353.5	NA	16.0	2670
	D	30	2.04	14.8	0.1716	0.0533	78	454.5	0.0400	16.1	2580
	E	30	2.02	14.9	0.1820	0.0468	82	450.5	0.0392	16.3	2580
	F	30	2.04	14.8	0.1904	0.0408	88	462.2	0.0320	16.2	2510
Uncaptured	A	32	2.15	14.8	0.1806	0.0264	86	476.2	0.0366	15.8	2430
	B	30	2.15	15.7	0.2344	0.0294	64	273.0	0.0400	15.7	2280
	C	32	2.26	15.6	0.1945	0.0209	81	416.5	0.0437	15.6	2490
	D	29	2.07	14.2	0.2354	0.0315	76	322.9	0.0420	15.9	2040
BPBM 20469	A	29	1.99	14.6	0.2469	0.0518	64	259.2	0.0375	16.7	2450
	B	30	2.05	14.6	0.2405	0.0518	62	257.8	0.0369	16.6	2450
	C	31	2.11	14.8	0.2413	0.0524	63	261.1	0.0442	16.7	2450

Etymology.—The name is a noun in apposition and is named for the first author's wife.

Range.—Known only from the upper elevations of Mt. Rio, 630–800 m, Sudest Island, Milne Bay Province, Papua New Guinea (Fig. 5). This is the only peak on Sudest of such an elevation; the species was not found or heard below 630 m, and its range is almost certainly restricted to this rather small area.

Ecological notes.—Animals were common. Adults were typically found at night while calling perched on the upper surfaces of leaves 2 m or more above the ground. One adult was found by day in a resting position on a dry leaf on the ground. Juveniles were found conspicuously perched on leaves and stems of plants 1–2 m above ground in the mid- to late-afternoon. One juvenile was found active on the tip of a pandanus leaf 2 m above ground soon after nightfall. When conspicuously perched in the daytime, juveniles were not easily disturbed.

One juvenile and one adult specimen were examined for stomach contents (the larger series of types was spared further dissection). The stomach of the juvenile contained ten ants of the genus *Crematogaster*; that of the adult contained 30 formicine ants of an undetermined genus.

Oreophryne ezra appears limited to a small patch of cloud forest that occurs on the upper elevations of Mt. Rio at 630–800 m. Vegetation of this forest was briefly described by Brass (1959:52–53).

DISCUSSION

The new species brings to 34 the number of *Oreophryne* species described from the Papuan region. It is the only member of the genus to have such bright and distinctive dorsal coloration. Most species in this genus are brown, gray, or greenish-brown dorsally, presenting an overall drab impression. Not only do adults of *O. ezra* contrast with this general pattern in being pale pink or peach with bright blue eyes, but the juveniles are even more strikingly divergent from what we presume to be the ancestral pigmentation pattern for the genus. Indeed, the striking coloration, conspicuous perching of the juveniles on vegetation during daylight hours, and apparent lack of well-developed escape behavior easily lead one to hypothesize that their vivid color pattern serves an aposematic function. It is difficult to imagine what other explanation would account for these observations, and significant associations between bright coloration and toxicity have been demonstrated for other frogs (Summers and Clough, 2001; Santos et al., 2003). Should this hypothesis prove true, it would be an unusual instance of aposematism in frogs because of its ontogenetic loss. However, confirmation of the hypothesis awaits testing to determine whether juveniles and/or adults contain skin toxins and, if so, experimental determination of their effect on predator behavior.

Oreophryne ezra is apparently confined to a single small patch of cloud forest from 630–800 m perched on the top of Mt. Rio. Judging from topographical maps, this forest comprises less than 1 km^2 in total extent. Two very small peaks that rise to greater than 600 m elevation occur just to the east of Mt. Rio, and they too might possibly contain the frog, but each contains only a few hectares in the appropriate elevational range. The small areal extent of this cloud forest makes the frog highly vulnerable to any

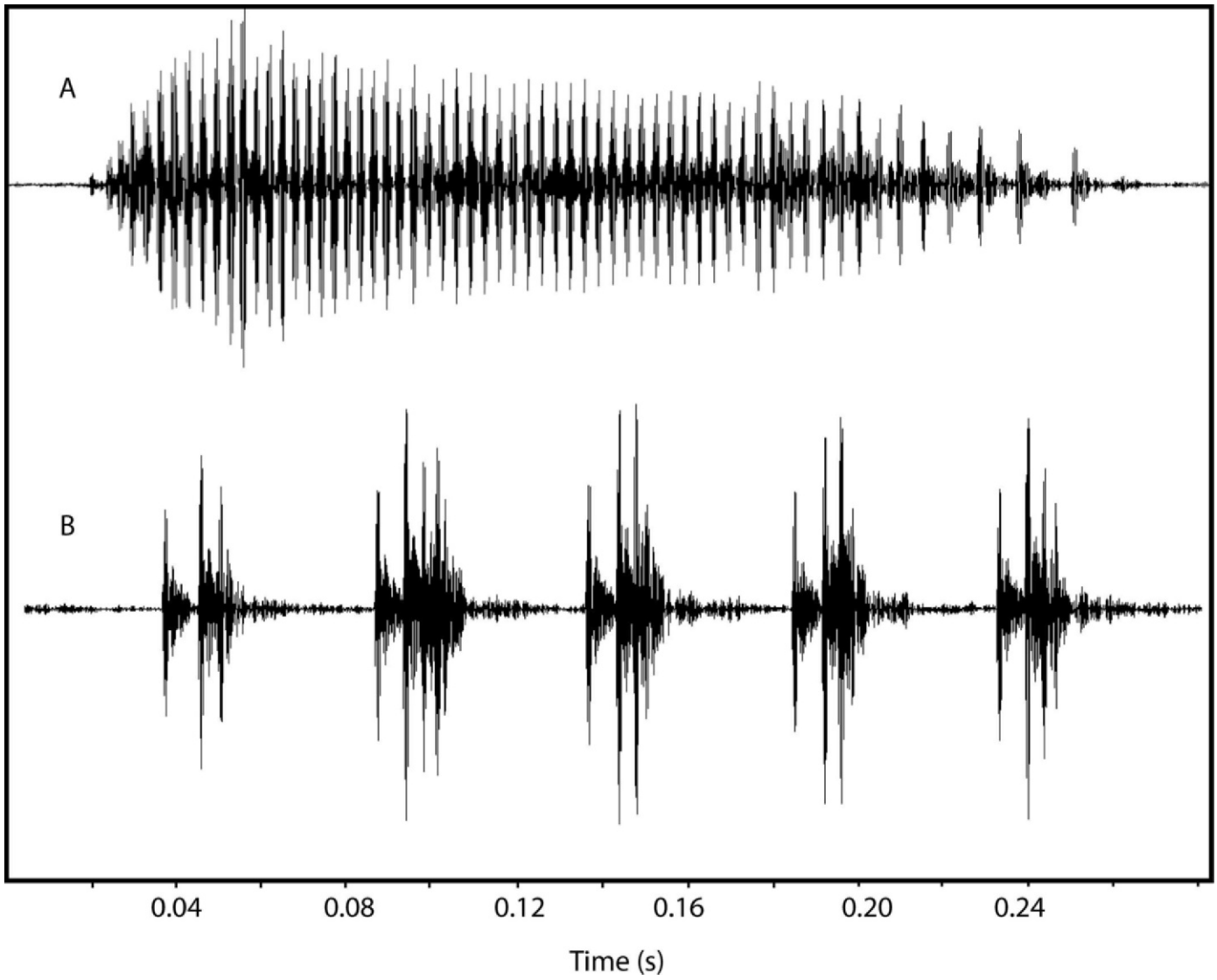

Fig. 3. The pulsed structure of call notes of *Oreophryne ezra* (BPBM 20469, paratype) recorded at the at the type locality on 23 April 2004 at 2015 h, air temperature 20°C. (A) First note; and (B) five subsequent notes.

destruction of its habitat. Direct human conversion of this forest patch appears extremely unlikely in the foreseeable future: population density on Sudest Island is low, population is concentrated in small villages along the coast, most of the island is forested, a pre-existing trail did not access the top of Mt. Rio, and inhabitants rarely venture toward the peak because they consider it sacred. However, the threat posed to the habitat of *O. ezra* by climate change would appear more significant. Predictions of habitat shifts at such a localized scale cannot be made with confidence at present, but broader trends toward climatic warming (and possible attendant reductions in rainfall or cloud cover) could be sufficient to push the 200 m of cloud forest off the top of Mt. Rio. Should climatic patterns in the Louisiade Archipelago change to such an extent, *O. ezra* will be one of several endemic species potentially threatened with extinction due to loss of its habitat.

MATERIAL EXAMINED

Oreophryne anthonyi: BMNH 1947.2.12.38–39, syntypes, Papua New Guinea, Central Province, Mt. Victoria.

Oreophryne biroi: BPBM 14509–11, Papua New Guinea, West Sepik Province, Bewani Mts., Trefas Village; BPBM 22786, Papua New Guinea, West Sepik Province, Torricelli Mts., S slope Mt. Sapau; BPBM 23481–85, Papua New Guinea, East Sepik Province, Hunstein Mts., E slope Mt. Hunstein.

Oreophryne brachypus: BPBM 22509–36, Papua New Guinea, East New Britain Province, 2–12 km NNW Marmar.

Oreophryne geislerorum: BPBM 5285–86, Papua New Guinea, Morobe Province, Mindik; BPBM 18510–12, Papua New Guinea, Morobe Province, NW slope Mt. Shungol; BPBM 31473, 31877, Papua New Guinea, Morobe Province, Bowutu Mts., Kamiali Wildlife Management Area.

Oreophryne hypsiops: BPBM 1171, paratype, Papua New Guinea, East Sepik Province, Dreikiker; BPBM 1179, paratype, Papua New Guinea, East Sepik Province, Wewak.

Oreophryne idenburgensis: AMNH 49663, holotype, AMNH 49666, paratype, Indonesia, West Papua Province, Idenburg River, 18 km SW Bernhard Camp.

Oreophryne inornata: AMNH 57000, holotype, AMNH 56731, 56912, 56984, 57262, 57353, paratypes, Papua New Guinea, Milne Bay Province, Goodenough Island, E slope Oya Madau; BPBM 16217–41, 16243, 16245, Papua New Guinea,

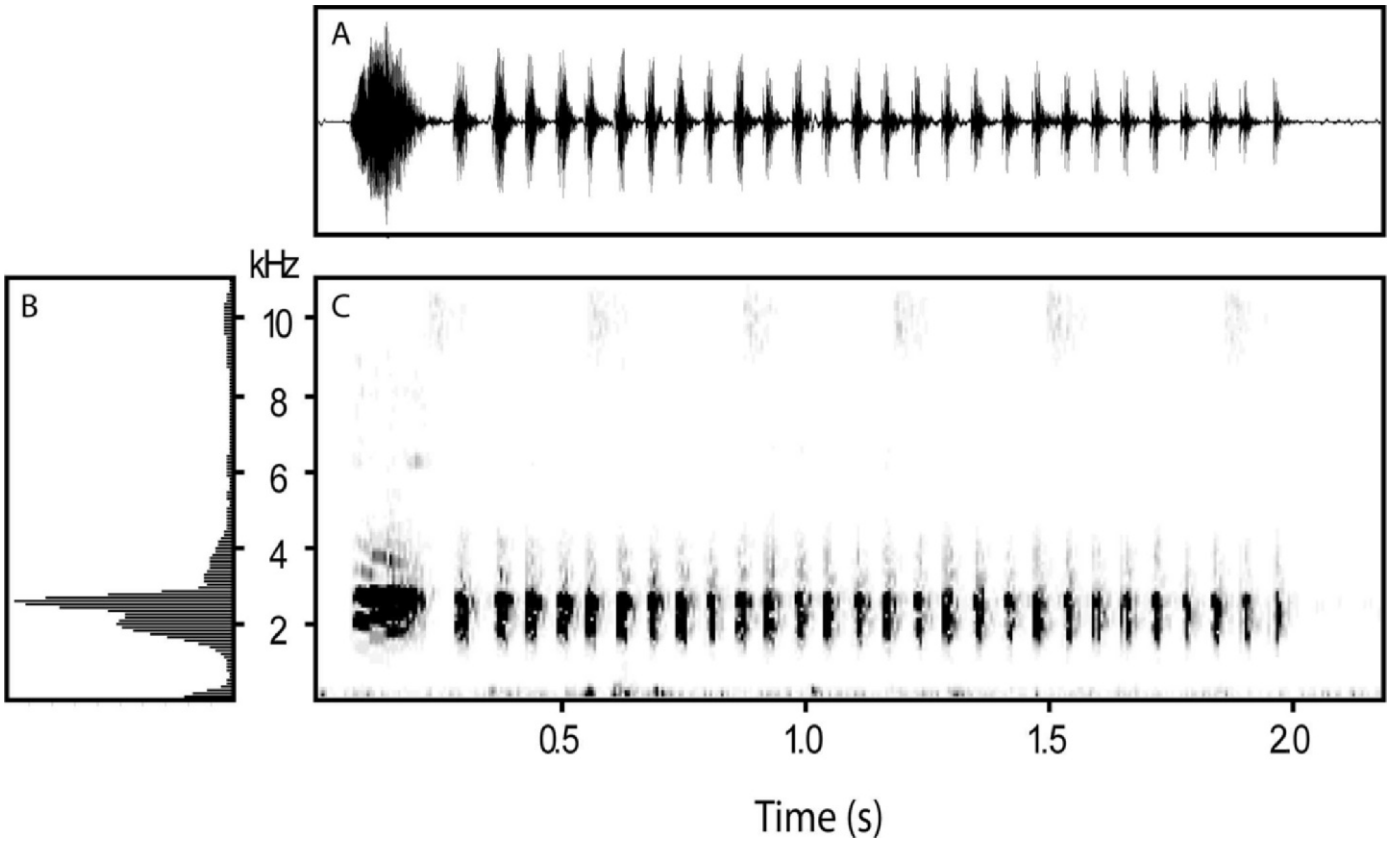

Fig. 4. Spectrogram of call A (Table 2) of *Oreophryne ezra* (BPBM 20468, holotype) recorded at the at the type locality on 23 April 2004 at 1900 h, air temperature 20°C. (A) Waveform; (B) power spectrum; and (C) spectrogram.

Milne Bay Province, Fergusson Island, E slope Oya Tabu; BPBM 16249–56, Papua New Guinea, Milne Bay Province, Fergusson Island, S slope Oya Waka.

Oreophryne insulana: AMNH 57267, holotype, AMNH 56915, 57265–66, paratypes, Papua New Guinea, Milne Bay Province, Goodenough Island, E slope Oya Madau; BPBM 16119, 16546–51, Papua New Guinea, Milne Bay Province, Fergusson Island, E slope Oya Tabu.

Oreophryne kampeni: BMNH 1947.2.12.14, holotype, BMNH 1947.2.12.43–44, paratypes, Papua New Guinea, Central Province, Moroka.

Oreophryne loriae: BMNH 1947.2.12.41–42, paralectotypes, BPBM 22537–39, UPNG 2184, 4155, Papua New Guinea, Central Province, Moroka; UPNG 1683, 2611, 4934, Papua New Guinea, Milne Bay Province, Alotau; BPBM 16552–55, Papua New Guinea, Milne Bay Province, Normanby Island.

Oreophryne parkeri: BPBM 22781–85, Papua New Guinea, West Sepik Province, Torricelli Mts., S slope Mt. Sapau; BPBM 23475–76, Papua New Guinea, West Sepik Province, Bewani Mts., Trefas Village; BPBM 23477–80, Papua New Guinea, East Sepik Province, Hunstein Mts., E slope Mt. Hunstein.

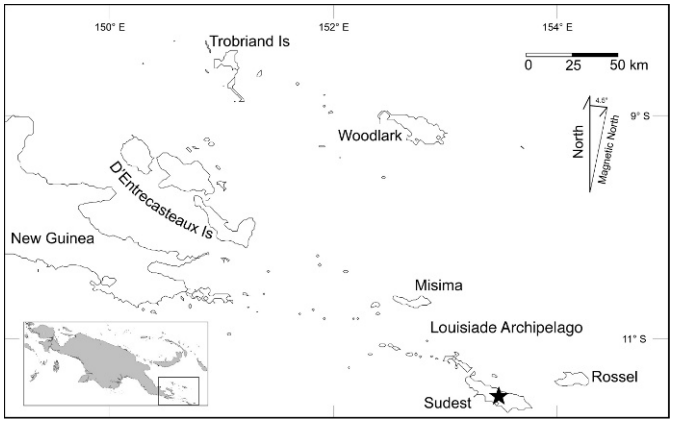

Fig. 5. Map of southeastern New Guinea and adjacent islands, showing type locality of *Oreophryne ezra* on Sudest Island.

ACKNOWLEDGMENTS

We thank P. Lagalaga, F. Malesa, M. Samson, G. Shea, J. Slapcinsky, and J. Volo for field assistance; L. Doni, N. Doni, C. Graham, D. Mitchell, and Conservation International for logistical assistance; S. James for kindly providing the DNA sequences; E. Sarnat for identifying stomach contents; P. Imada and C. Kishinami for specimen curation; B. Evans for preparing the figures and map; the PNG National Museum and Art Gallery for providing in-country collaborative assistance; and the PNG Department of Environment and Conservation, PNG National Research Institute, and Milne Bay Provincial Government for permission to work in Milne Bay Province. This research was supported by National Science Foundation grants DEB-0103794 and DEB-0743890. This is contribution 2009-006 to the Pacific Biological Survey.

LITERATURE CITED

Blackburn, D. C., and V. Droissart. 2008. Natural history notes: *Nectophryne batesii*: juvenile coloration. Herpetological Review 39:208–209.

Boulenger, G. A. 1913. On the presence of two closely allied species of toads of the genus *Nectophryne* in Cameroon. Annals and Magazine of Natural History, series 8 12:70–72.

Brass, L. J. 1959. Results of the Archbold Expeditions. No. 79. Summary of the Fifth Archbold Expedition to New Guinea (1956–1957). Bulletin of the American Museum of Natural History 118:5–69.

Günther, R. 2003a. Three new species of the genus *Oreophryne* from western Papua, Indonesia. Spixiana 26:175–191.

Günther, R. 2003b. Further new species of the genus *Oreophryne* (Amphibia, Anura, Microhylidae) from western New Guinea. Zoologische Abhandlungen, Dresden 53:65–85.

Günther, R., S. J. Richards, and D. Iskandar. 2001. Two new species of the genus *Oreophryne* from Irian Jaya, Indonesia. Spixiana 24:257–274.

Hoffman, E. A., and M. S. Blouin. 2000. A review of colour and pattern polymorphisms in anurans. Biological Journal of the Linnean Society 70:633–665.

Köhler, F., and R. Günther. 2008. The radiation of microhylid frogs (Amphibia: Anura) on New Guinea: a mitochondrial phylogeny reveals parallel evolution of morphological and life history traits and disproves the current morphology-based classification. Molecular Phylogenetics and Evolution 47:353–365.

Kraus, F., and A. Allison. 2006. Three new species of *Cophixalus* (Anura: Microhylidae) from southeastern New Guinea. Herpetologica 62:202–220.

Parker, H. W. 1934. A Monograph of the Frogs of the Family Microhylidae. British Museum (Natural History), London.

Richards, S., and D. Iskandar. 2000. A new minute *Oreophryne* (Anura: Microhylidae) from the mountains of Irian Jaya, Indonesia. Raffles Bulletin of Zoology 48:257–262.

Roux, J. 1910. Reptilien und Amphibien der Aru- und Kei-Inseln. Abhandlungen der Senckenbergischen Naturforschenden Gesellschaft 33:211–247.

Santos, J. C., L. A. Coloma, and D. C. Cannatella. 2003. Multiple, recurring origins of aposematism and diet specialization in poison frogs. Proceedings of the National Academy of Sciences of the United States of America 100:12792–12797.

Scheel, J. J. 1970. Notes on the biology of the African tree-toad, *Nectophryne afra* Buchholz & Peters, 1875, (Bufonidae, Anura) from Fernando Poo. Revue de Zoologie et de Botanique Africaines 81:225–236.

Schiotz, A. 1971. The superspecies *Hyperolius viridiflavus* (Anura). Videnskabelige Meddelelser fra Dansk Naturhistorisk Forening 134:21–76.

Schiotz, A. 1999. Treefrogs of Africa. Edition Chimaira. Frankfurt am Main, Germany.

Smith, M. A., N. A. Poyarkov, and P. D. Herbert. 2008. CO1 DNA barcoding amphibians: take the chance, meet the challenge. Molecular Ecology Resources 8:235–246.

Summers, K., and M. E. Clough. 2001. The evolution of coloration and toxicity in the poison frog family (Dendrobatidae). Proceedings of the National Academy of Sciences of the United States of America 98:6227–6232.

Zweifel, R. G. 1985. Australian frogs of the family Microhylidae. Bulletin of the American Museum of Natural History 182:265–388.

Zweifel, R. G. 2003. A new species of microhylid frog, genus *Oreophryne*, from Papua New Guinea. American Museum Novitates 3419:1–8.

Zweifel, R. G., H. G. Cogger, and S. J. Richards. 2005. Systematics of microhylid frogs, genus *Oreophryne*, living at high elevations in New Guinea. American Museum Novitates 3495:1–25.

Zweifel, R. G., J. I. Menzies, and D. Price. 2003. Systematics of microhylid frogs, genus *Oreophryne*, from the North Coast Region of New Guinea. American Museum Novitates 3415:1–31.