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Tadpole and vocalizations of *Chiasmocleis hudsoni* (Anura, Microhylidae) in Central Amazonia, Brazil

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The genus *Chiasmocleis* is distributed from Panama to southern South America and contains 21 recognized species (Frost 2007). Eight of them are associated with Amazonian rainforests (Frost 2007). However, only the larvae of four species and the vocalization of three species have been described for species occurring in this region (Nelson 1973; Duellman 1978; Zimmerman & Bogart 1988; Hero 1990; Schlüter & Salas 1991; Lescure & Marty 2000; Vera Candioti 2006). The tadpole of *C. hudsoni* has not been formally described; it was mentioned briefly (diagrammatic drawings and larval color notes) in Hero's tadpole identification key from Central Amazonia (Hero 1990), as *Chiasmocleis* cf. *ventrimaculata*. In this paper we describe the tadpole and the vocalizations of *C. hudsoni* and also provide comments on the spawning sites, clutch size and breeding periods.

We collected clutches and tadpoles of *Chiasmocleis hudsoni* in streamside ponds in January 2004 and from February to March 2005, at Reserva Florestal Adolpho Ducke (RFAD) (02°55' and 03°01'S, 59°53' and 59°59'W) in Manaus, Amazonas, Brazil. Tadpoles were preserved immediately after collection and used in the description. Identification was based on newly metamorphosed individuals obtained after keeping clutches in the laboratory. Tadpole stages were defined follow Gosner (1960). Descriptive terminology and morphometric variables follow Altig & McDiarmid (1999). Measurements were taken with millimetric oculars. We recorded vocalizations of eight individuals in RFAD from February to March 2005 using a Sony TC-D5M tape recorder coupled to an external condenser microphone (TECT Model UEM-83). We used Raven 1.2 software to analyze calls (Blackman function, size 20 ms, 3 dB filter Bandwidth 80 Hz, overlap 80 ms, hop size 16.4 and DFT size 2048). Voucher specimens were deposited in the Herpetological Collection of the Instituto Nacional de Pesquisas da Amazônia (Tadpole lots: INPA-H 16958–16965).

Description of tadpoles: The description is based on nine tadpoles (INPA-H 16958) at stage 33. Body rectangular in dorsal view (Fig 1A) and oval in lateral view (Fig 1B). Body and tail 37 and 63 % of total length, respectively. Body widest than deep. Body is highest in its posterior third and wider immediately behind eyes. Snout broad and bluntly rounded in dorsal and ventral view and rounded in lateral view. Eyes located and directed laterally. Interorbital distance four times larger than maximum eye diameter. Narial openings not visible at stages 27 and 33. Spiracle single, long and wide, positioned medially and ventrally; distal border projecting over the anal tube, visible in lateral and dorsal view (Fig. 1A and B). Anal tube positioned along ventral midline, attached directly to ventral fin. Tail musculature heavy and practically equal in size to dorsal and ventral fins along the anterior third of the tail. Dorsal and ventral fins similar in height. Dorsal fin originating at tail-body junction, increasing throughout the first third of the tail, and then gradually diminishing to a tip. Ventral fin originating at the posterior ventral terminus of the body, slightly arched and, maintaining the same height throughout the proximal two-thirds of the tail. Tail pointed; flagellum absent. Mouth small (1.44±0.05 mm, 1.40–1.50, N=7) and terminal (Fig. 1C). Upper lip large, fleshy, covering the opening. Lower lip narrow, generally arched. Jaw sheaths, papillae, and tooth rows absent. Snout-vent length of metamorphosed froglets (stage 46) was 4.97±0.21 mm (4.60–5.30, N=10). Froglets were similar to adults in color patterns and body shape. Measurements of tadpoles at other developmental stages are presented in Table 1.

Variation: At stage 41, 42 and 44 the external nares are located in dorsal area and consists of a rounded whitish spot located dorsally and the narial distance is 0.65 ± 0.07 mm (0.60–0.70, N=4, stages 41 and 42).

Coloration: In life, dorsum transparent brown or orange, venter whitish and tail transparent. In fixed specimens, dorsum transparent brown with dark melanophores, venter whitish with melanophores on anterior part and tail transparent with melanophores on dorsal fin and on the posterior third of the ventral fin.



FIGURE 1. *Chiasmocleis hudsoni* tadpole at stage 33. (A) Dorsal view; (B) lateral view (scale = 5 mm); (C) oral apparatus (scale = 1 mm). Specimen from Reserva Florestal Adolpho Ducke, Manaus, Amazonas, Brazil.

TABLE 1. Measurements (in mm) of 17 tadpoles of Chiasmocleis hudsoni in Gosner's (1960) stages 27, 33, 41, 42 and 44. Va	alues are
means \pm standard deviation; range is given in brackets.	

Characters	Stage 27	Stage 33	Stage 41	Stage 42	Stage 44
	(N = 4)	(N = 9)	(N = 2)	(N = 1)	(N = 1)
Total length	8.52±0.22 (8.20-8.70)	11.47±0.42 (11.00-12.10)	15.10-15.30	12.50	9.80
Body length	2.73±0.31 (2.30-3.00)	4.23±0.10 (4.10-4.40)	5.80-6.00	6.10	5.30
Tail length	5.80±0.12 (5.70-5.90)	7.22±0.40 (6.80–7.80)	9.30–9.30	6.40	4.50
Maximum tail height	1.45±0.13 (1.30–1.60)	2.42±0.16 (2.20-2.60)	1.80 - 1.80	1.60	0.60
Tail muscle width	0.45±0.06 (0.40–0.50)	0.67±0.05 (0.60–0.70)	1.40-1.40	1.30	0.60
Tail muscle height	0.55±0.06 (0.50–0.60)	0.79±0.06 (0.70–0.90)	1.30-1.50	1.50	1.00
Internarial distance	-	-	0.60-0.70	0.70	1.20
Interorbital distance	0.93±0.10 (0.80–1.00)	1.71±0.11 (1.60–1.90)	2.10-2.20	1.90	1.40

Vocalizations: (Mean \pm standard deviation and range). Males of *Chiasmocleis hudsoni* emitted two distinct calls: an *advertisement call* and a *sporadic call*. The advertisement call consisted of a long call (49.8 \pm 13.5 sec, 32.4–67.3, N=5) with a number of multipulsed notes produced in series (Fig. 2A). Two clearly distinct note types were observed in this species: the introductory note (Fig. 2A, a) was observed in the beginning of each vocalization but was clearly observed only in three individuals, due to the overlap in the vocalization of many individuals in the other records. These notes can be long (9.2 \pm 3.7 pulses, 6–13, N=4) with mean duration of 249.2 ms (\pm 101.9 ms, 137–349, N=4). The dominant frequency (3,795.2 \pm 837.5 Hz, 2,551–4,349, N=4) is lower than in the notes emitted after them. The pulse duration

 $(12.0\pm7.3 \text{ ms}, 3.0-31.0, \text{N}=36)$ and the interval between pulses $(17.0\pm8.0 \text{ ms}, 2.0-25.0, \text{N}=35)$ are greater than the secondary notes. The chorus notes (Fig. 2A, b) are short and consist of multipulsed notes $(5.3\pm0.6 \text{ pulses}, 5-8, \text{N}=40)$ emitted repetitively. The mean of fundamental frequency is 4,109.2 Hz (±193.9, 3,625–4,407, N=40) and the mean of dominant frequency is 4,624.2 Hz (±153.5, 4,263–4,866, N=40). Note duration varies between 71–126 ms (98.6±11.7 ms, N=40) being separated by an interval of 19–52 ms (25.7±6.6 ms, N=40); mean pulse duraton is 9.0 ms (±2.0, 5–15, N=120); mean pulse interval is 12.9 ms (±3.2, 5–20, N=170). This type of note basically forms the typical advertisement call during chorus activity. The advertisement call, therefore, is formed by long and multipulsioned introductory notes followed by sequences of short notes (chorus notes) forming repetitive series.



FIGURE 2. Spectrograms (above) and oscillograms (below) of (A) advertisement calls: a = introductory note; b = chorus note (recorded on 22 February 2005; air temperature 27.5 °C; 19:45 h), and (B) sporadic call (recorded on 06 March 2005; air temperature 29.7 °C; 17:15 h) of*Chiasmocleis hudsoni*. Specimens from Reserva Florestal Adolpho Ducke, Manaus, Amazonas, Brazil.

The sporadic call was formed by 10.1 notes (\pm 4.8, 1–21, N=16) with mean dominant frequency (4,513.8 \pm 134.0 Hz, 4,005–4,694, N=46) similar to that of the secondary notes of advertisement call. However the sporadic call had a higher number of pulses per note (10.1 \pm 3.9, 6–20, N=46) and a lower interval between pulses (5.6 \pm 2.6 ms, 2–13, N=73). The number of pulses diminishes to the end of the call (Fig. 2B). This vocalization was emitted by males during the beginning of the calling activity, generally before sunset.

Reproductive biology notes: Breeding individuals, clutches and tadpoles of *C. hudsoni* were observed in temporary isolated ponds and in streamside ponds, both in forests. Adults were found during rainy season, mainly from February to May. Males were observed calling above or within the leaf litter around temporary ponds, in decomposing branches and dead trunks, but never directly on the water surface. Vocalizations were concentrated in nocturnal periods; occasionally calling males were heard during daytime after heavy rainfall. The size of the ponds was variable from 40 to 100 m². Clutches were deposited between roots in the soil, on fallen tree trunks near ponds. Mean clutch size was 351 eggs (N=5; range 269–412). The initial stage of development of tadpoles occurs inside the eggs out of water. The tadpoles hatched at Gosner's (1960) stages 25/26 in the laboratory (N= 5 clutches) and stages 26/27 in the field (N= 1 clutch), and reached the pond after the flooding by rains. In the ponds, the tadpoles remain motionless in a diagonal position (head up) under the surface (app. 500 individuals in six ponds).

The characteristics of *C. hudsoni* tadpoles are similar to those described by Hero (1990), with one exception: the spiracle is not parasagital, but long and wide, with distal border projecting over the anal tube, visible in lateral and dorsal view. Tadpoles of *C. hudsoni* differs from other species of *Chiasmocleis* by smaller total length (11.5 mm in *C. hudsoni*). The coloration of *C. hudsoni* tadpoles (body transparent brown or orange, venter whitish and tail transparent) is also different from that *C. anatipes* (olive-tan above and yellowish white below; Duellman 1978), *C. panamensis* (reddish brown dorsum and caudal musculature; Vera Candioti 2006) and *C. shudikarensis* (tranparent body and orange tail with black margins; Hero 1990). It differs from *C. ventrimaculata* by absence of a pair of curved, bracket-shaped, and light-cream lines between the eyes (Schlüter & Salas 1991). The dominant frequency of the advertisement calls of *C. hudsoni* (4.2–4.8 kHz) is lower than calls of *C. panamensis* (4.8–5.5 kHz; Nelson 1973), *C. ventrimaculata* from Peru (5.1–6.9 kHz; Schlüter & Salas 1991), and *C. shudikarensis* from Brazil (5.5–7.5 kHz; Zimmerman & Bogart 1988), and greater than *C. ventrimaculata* from Colombia (3.3–3.7 kHz; Nelson 1973), and *C. shudikarensis* from French Guiana (3.3–3.7 kHz; Lescure & Marty 2000).

The initial stages of development of tadpoles of *C. hudsoni* occur out of water. This oviposition strategy allows a higher chance of survival for the individuals, because they are larger once they enter the ponds and they remain less time in the aquatic phase, thus decreasing contact with predators (*e.g.* tadpoles of *Leptodactylus knudseni* and *Ceratophrys cornuta* are common in isolated ponds and *L. rhodomystax* Boulenger and fishes in streamside ponds; DJR, unpubl.).

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