

# Seasonality, age structure and reproduction of *Leptodactylus (Lithodytes) lineatus* (Anura, Leptodactylidae) in Rondônia state, southwestern Amazon, Brazil

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**ABSTRACT.** *Leptodactylus (Lithodytes) lineatus* (Schneider, 1799) is an Amazonian leaf litter frog considered rare or uncommon in several studies on anuran communities. Despite being a widely distributed frog in Amazonian forests, knowledge of the biology and ecology of this species is relatively scarce. This species has been reported to live in association with leaf-cutter ant nests (*Atta* spp.) during the breeding period. In this paper we present data on the seasonality of this species and some reproductive information gathered at a locality of Rondônia state, northwestern Brazil. Field work was carried out between April 2001 and March 2002, with the use of pitfall traps with drift fences as a survey method. *Leptodactylus (L.) lineatus* had a higher capture frequency in this locality compared to that of other studies carried out in other Amazonian localities, possibly because this species has secretive habits, such as calling and breeding from nests of leaf-cutting ants, and are difficult to find during visual encounter surveys. The breeding period occurs between October and March. Calling males and egg-bearing females were found between September and February and juvenile recruitment occurred mainly from the end of the rainy season to the beginning of the dry season (February to June). Males and females show sexual dimorphism in SVL, females being significantly larger than males. The number of ovarian eggs per female varies from 110 to 328 and analyses indicate that there is a significant correlation with SVL.

**KEYWORDS.** *Leptodactylus (Lithodytes) lineatus*, Amazon, breeding period, sexual dimorphism.

**RESUMO.** Sazonalidade, estrutura etária e reprodução de *Leptodactylus (Lithodytes) lineatus* (Anura, Leptodactylidae) no estado de Rondônia, Brasil. *Leptodactylus (Lithodytes) lineatus* (Schneider, 1799) é uma espécie de anuro amazônico considerado raro ou incomum em diversos estudos de comunidades de anfíbios. Embora apresente ampla distribuição na Floresta Amazônica, conhecimento sobre a sua biologia e ecologia são escassos. Esta espécie tem sido relatada por viver em associação com ninhos de formigas cortadeiras (*Atta* spp.) durante o período reprodutivo. Neste trabalho, nós apresentamos dados de sazonalidade e reprodução desta espécie em uma localidade do estado de Rondônia, noroeste do Brasil. Trabalhos de campo foram conduzidos de abril de 2001 a março de 2002, com o uso de armadilhas de queda com rede direcionadora. *Leptodactylus (L.) lineatus* teve uma alta taxa de captura nesta localidade, quando comparada com outros estudos em outras localidades na Amazônia, possivelmente devido a esta espécie possuir hábitos crípticos, como vocalizar e reproduzir dentro de ninhos de formigas, e ser dificilmente encontrada durante procura visual ativa. O período reprodutivo ocorre entre outubro e março. Machos vocalizando e fêmeas ovuladas foram encontrados entre setembro e janeiro e o recrutamento de juvenis ocorreu principalmente no final do período chuvoso. Machos e fêmeas apresentaram dimorfismo sexual quanto ao comprimento rostro-cloacal (CRC), sendo as fêmeas maiores que os machos. O número de folículos ovarianos por fêmea variou de 110 a 328 e análises indicaram que existe uma correlação significativa entre este número e o CRC.

**PALAVRAS-CHAVE.** *Leptodactylus (Lithodytes) lineatus*, Amazônia, período reprodutivo, dimorfismo sexual.

*Leptodactylus (Lithodytes) lineatus* (Schneider, 1799) is an Amazonian leaf litter frog poorly documented in community research (SCHLÜTER & REGÖS, 1981; CALDWELL & ARAÚJO, 2005; LIMA *et al.*, 2006), despite being categorized as Least Concern (GAA) due to its wide distribution in the Amazonian forests of Eastern Peru, Ecuador, Bolivia, and Brazil through southern Venezuela and to the Guianas (FROST, 2006). Knowledge of the biology and ecology of this species is relatively scarce (REGÖS & SCHLÜTER, 1984; HOOGMOED, 1986; ZIMMERMAN & RODRIGUES, 1990; LAMAR & WILD, 1995; SCHLÜTER & REGÖS, 1996).

This species has been reported to live in association with leaf-cutter ant nests (*Atta* spp.) (SCHLÜTER & REGÖS, 1981) during the breeding period (LAMAR & WILD, 1995). Males call from subterranean burrows in these nests (SCHLÜTER & REGÖS, 1981; RODRÍGUEZ & DUELLMAN, 1994; LAMAR & WILD, 1995); the species produces foam nests, and tadpoles possibly complete development in ponds (RODRÍGUEZ & DUELLMAN, 1994; LAMAR & WILD, 1995;

SCHLÜTER & REGÖS, 1996). Given its coloration and the diurnal activity of juveniles, this species is considered a mimic of some aromobatids (*Allobates femoralis* (Boulenger, 1884)) and dendrobatids (*Ameerega* spp.) (LAMAR & WILD, 1995). Recently, FROST *et al.* (2006) placed *Lithodytes lineatus* and *Adenomera* Fitzinger, 1867 species in the genus *Leptodactylus* Fitzinger, 1826, and *Lithodytes* was considered a subgenus.

During a research studies conducted on anuran communities in the municipality of Espigão do Oeste, State of Rondônia (Brazil) (see BERNARDE, 2007; BERNARDE & MACEDO, 2008), we captured several individuals of *Leptodactylus (L.) lineatus*. In this paper we present data on the occurrence of this species (males, females and juveniles) over the course of a year and some reproductive information (calling activity of males, number and diameter of ovarian eggs in females). Herein, we compare the abundance of this species at our study site with that of other Amazonian localities.

## MATERIAL AND METHODS

This study was carried out at Jaburi Farm, municipality of Espigão d'Oeste (11°35' – 11°38'S; 60°41' – 60°45'W), southwest Amazon, State of Rondônia, Brazil. The region's altitudes oscillate around 280 meters. The vegetation is constituted by Amazonian Forest (mainly Open Rain Forest, see VELOSO *et al.*, 1991), where the following species are found (OLIVEIRA, 2002): Big Leafed Mahogany (*Swietenia macrophylla* (Meliaceae)), Cerejeira (*Torresia acreana* (Poaceae)), Lapacho (*Tabebuia serratifolia* (Bignoniaceae)), Rubber Tree (*Hevea brasiliensis* (Euphorbiaceae)) and Brazil Nut Tree (*Bertholletia excelsa* (Lecythidaceae)). Several areas in this region have been deforested and transformed for cattle ranching and agricultural farming. The annual mean precipitation is 2,100 mm, with a dry period normally taking place from April to September. The mean annual temperature is 26°C (climate data from the town of Espigão d'Oeste). Colonization in Rondônia State, mainly along highway BR 364, caused progressive deforestation of the Amazon rainforest during the 1980's (VANZOLINI, 1986).

Field work was carried out from April 2001 to March 2002 with the use of pitfall traps with drift fences (CAMPBELL & CHRISTMAN, 1982). In this method, plastic barrels (200 L) were buried every 10 m in a straight line and connected by a one-meter high canvas fence. Six trap arrays were built with four barrels each and a 44 m long fence. The trap arrays were positioned in three different habitats, with two trap arrays in each habitat: inside the forest, away from permanent water bodies; inside the forest ca 100 m from a river (about 4 m wide and 0.80 m deep) and in pasture areas, far from permanent water bodies. Each trap array was built at least 1,800 m away from the next one, and for each habitat both trap arrays were separated by a minimum distance of 200 m. Traps remained open all year and were monitored from 3 to 4 times a week. Other data, such as calling activity and individuals observed in the leaf litter were also occasionally recorded. The senior author became familiar with the advertisement call of this species from calling males kept captive for two nights in November 2001.

Specimens captured were killed in 10% alcohol and fixed in 10% formalin. To describe the age structure of this population, specimens were classified as adult males, adult females and juveniles. The individuals were sexed by dissection or by reproductive condition (e.g. presence of ovarian eggs in females and extended vocal sacs or hypertrophied gonads in males). The snout-vent length of the smallest female with mature ovarian eggs was used to set the lower size limit of this category; data on male adult size in the literature (RODRÍGUEZ & DUELLMAN, 1994; LIMA *et al.*, 2006) were used to set the lower size limit of the adult male category. The number and diameter of mature ovarian eggs were measured via the removal of ovaries. The maximum diameter of five eggs was obtained for each female and the mean was calculated. Snout-vent length (SVL) and egg diameter were determined to the nearest 0.1 mm with callipers. The SVL data were used to separate females without ovarian eggs from juveniles. All specimens collected are housed in the "AAG-UFU"

herpetological collection at the Museu de Biodiversidade do Cerrado, in the Universidade Federal de Uberlândia (MG), Brazil.

Spearman's test (ZAR, 1984) was used to look for possible correlations between capture frequency and rainfall and between SVL of the egg-bearing females and number of ovarian eggs. A Student's t-test was used to compare the SVL of males and females. The computational analysis was performed in JMP 5.0.1 to PC (SAS Institute).

## RESULTS AND DISCUSSION

A total of 112 individuals of *Leptodactylus* (*L. lineatus*) were captured in pitfall traps. Of these, 101 were analysed, belonging to the following sex and size classes: 46 juveniles, 32 females and 23 males (Tab. I; Fig. 1).

Less individuals (n=38) were captured during the six driest months (April to September; rainfall = 456mm) in relation to the six rainiest months (n=74; October to March; rainfall = 1,850 mm). A smaller number of individuals was captured between June and August 2001, which were the driest months (Fig. 1). However, no positive correlation was found between the capture frequency and rainfall patterns ( $r_s = 0.3184$ ;  $p = 0.3213$ ;  $n = 12$ ). Adults were more frequent during the months of September to December (Fig. 1) and the egg-bearing females were captured only from September to February. Calling activity of the males was observed from November to January in forested areas. During the whole study period, only in two occasions, a male (not founded) was heard calling from inside a nest of the leaf-cutting ant (*Atta* sp.) in open areas. The recruitment period of juveniles occurred mainly between February and March (see Figs. 1, 2).

The majority of individuals (n=84) was captured in the forest near a river environment; another 28 individuals were captured in a forest distant from permanent water bodies. No individuals were captured with pitfall traps in pasture areas.

The SVL of juveniles ranged from 19.6 to 35.85 mm (Fig. 2), in males, from 34.9 to 47.0 mm, and in females it ranged from 38.1 to 52.0 (Tab. II). Females are significantly larger (mean SVL=44.76 mm;  $t = 6.49$ ;  $DF = 21$ ;  $P < 0.0001$ ) than males (mean SVL 38.51 mm).

Females larger than 38.1 mm had mature ovarian eggs (Tab. III). The egg number, estimated from ovarian

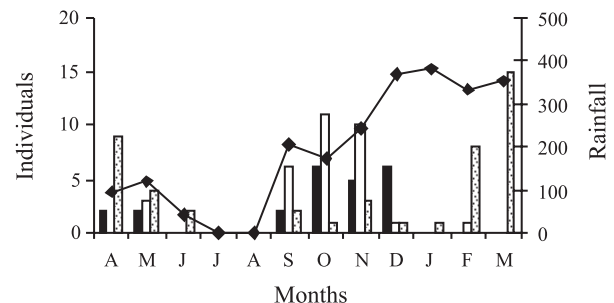


Figure 1. Frequency of males (black bars), females (white bars), juveniles (grey bars) of *Leptodactylus* (*L. lineatus*) (Schneider, 1799) and rainfall (line) from April 2001 to March 2002 in Espigão do Oeste, Rondônia, Brazil. Left axis shows number of captured individuals, right axis shows rainfall (mm), x-axis depicts months. N = 101.

Table I. Capture frequency of males, females and juveniles of *Leptodactylus (L.) lineatus* (Schneider, 1799) and rainfall (mm) from April 2001 to March 2002 in Espigão do Oeste, Rondônia, Brazil.

Months	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar
Rainfall	94	117	41	0	0	204	174	240	370	382	333	351
Males	2	2	0	0	0	2	6	5	6	0	0	0
Females	0	3	0	0	0	6	11	10	1	0	1	0
Juveniles	9	4	2	0	0	2	1	3	1	1	8	15
Total individuals	11	9	2	0	0	10	18	18	8	1	9	15

Table II. Sizes (SVL) of individual juveniles (n = 46), males (n = 23) and females (n = 32) of *Leptodactylus (L.) lineatus* (Schneider, 1799) at Espigão do Oeste, Rondônia, Brazil (measurements given in mm; SVL, mean snout-vent length; Range; SVL range; SD, Standard deviation).

	SVL	RANGE	SD
Juveniles	24.98	19.6 – 35.85	4.07
Males	38.51	34.9 – 47	2.72
Females	44.76	38.1 – 52	2.94

Table III. Snout-vent length (SVL) of egg-bearing females, number of mature ovarian eggs/female and mean egg diameter (n = 5 eggs per female) of *Leptodactylus (L.) lineatus* (Schneider, 1799) from municipality of Espigão do Oeste, Rondônia State (Brazil) (all measurements in mm).

Female SVL	Number of mature ovarian eggs/female	Mean egg diameter (n = 5 eggs by female)
38.10	195	1.6
40.25	110	1.80
40.40	267	1.7
42.60	237	1.6
42.95	201	1.55
43.10	275	1.65
43.30	294	1.52
44.20	169	1.74
45.90	273	1.88
46.00	220	1.92
46.50	218	1.65
46.55	281	1.76
47.30	267	1.78
47.30	307	1.70
48.00	216	1.63
48.55	274	1.73
52.00	328	1.78

eggs, varied from 110 to 328 (mean= 243; SD = 55.08; n = 17) and it was positively correlated to female size (R=0.5172; p=0.0335; n = 17). Mature ovarian eggs measured 1.5 to 1.92 mm (mean=1.7; SD=0.11; n = 17 females).

*Leptodactylus (L.) lineatus* was the fourth most abundant anuran species captured in pitfall traps (BERNARDE & MACEDO, 2008). By comparison, during a snake community study, only four individuals (three juveniles amid the leaf litter in the forest and one adult under a fallen tree trunk in pasture areas) were observed in 960 hours of visual encounter surveys (BERNARDE & ABE, 2006). Our abundance data for *L. (L.) lineatus* are greater than those from other studies conducted elsewhere in the Amazon (Tab. IV).

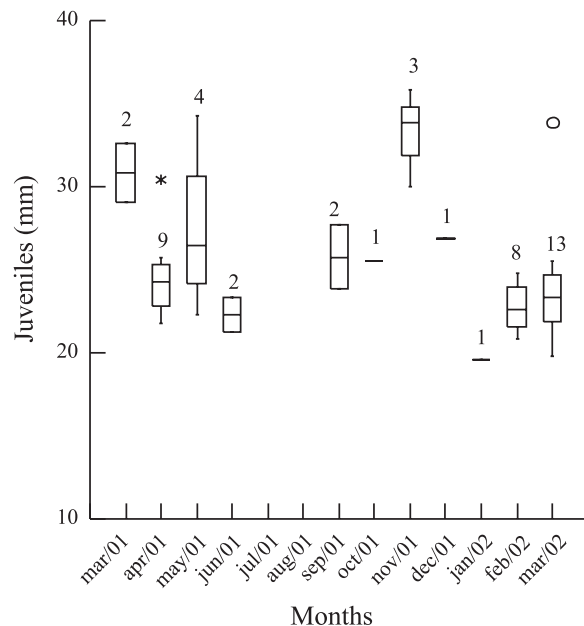


Figure 2. Seasonal variation in the abundance and size (SVL, in mm) of juvenile *Leptodactylus (L.) lineatus* (n= 46) trapped at Espigão do Oeste, Rondônia (Brazil) from April 2001 to March 2002. No individuals were trapped in July/August 2001. The number above each plot represents the sample sizes. Plots show minimum, maximum, median and quartiles. The asterisk and empty circle represent outliers.

The capture frequency of *L. (L.) lineatus* was higher at Espigão do Oeste (RO) than in many other Amazonian localities (see table IV for comparisons) possibly due to the use of pitfall traps to assess anuran community diversity in our study. The species' secretive habits, such as calling from and breeding within nests of the leaf cutting ant (SCHLÜTER & REGÖS, 1981; LAMAR & WILD, 1995), make it difficult to detect individuals during visual encounter surveys at all times of day. The lower observed abundance of this frog in anuran communities may be a result of the biases of the most common methods employed to inventory communities (visual encounter surveys, plots), which are not necessarily designed to detect this species. Without the use of pitfall traps, we would have only recorded a total of four individuals. Pitfall traps are an efficient technique to detect some species that may not be detected by other methods very often (CECHIN & MARTINS, 2000). The results of our study indicate that this is the case for the leaf litter frog *L. (L.) lineatus*.

Only one individual was found in a pasture habitat by occasional survey. Despite having been found in pasture habitats (TOCHER, 1998), *L. (L.) lineatus* appears

Table IV. Relative abundance of *Leptodactylus (L.) lineatus* published in studies on herpetofauna communities carried out in different Amazonian localities. Abundance = Number of individuals detected or assessments on relative abundance by the respective authors. VES = Visual Encounter Surveys.

Locality	Abundance	Method and sampling effort	Literature
Espigão do Oeste, RO (Brazil)	116	Pitfall (365 days)	This study
	Abundant	VES (960 person-hours)	
Along highway BR 364, RO (Brazil)	1	VES (80 days)	VANZOLINI (1986)
Rio Xingu, PA (Brazil)	3	VES (54 days)	CALDWELL & ARAÚJO (2005)
	Uncommon		
Manaus, AM (Brazil)	Rare	Not mentioned	LIMA <i>et al.</i> (2006)
Caracará, RR (Brazil)	3	VES (60 days)	CALDWELL & VITT (1999)
	Rare		
Loreto (Peru)	13	VES (93 days) Pitfall	DUELLMAN & MENDELSON (1995)
Cusco Amazónico (Peru)	2	VES (887 person days)	DUELLMAN & SALAS (1991)
	Rare		DUELLMAN (1995)
Balta, Ucayali (Peru)	1	VES (420 days) Pitfall	DUELLMAN & THOMAS (1996)
Cocha Cashu (Peru)	4	80 plots (5 x 5 m)	RODRÍGUEZ (1992)

to be mostly a forest (primary and secondary forest) dweller (AICHINGER, 1991; RODRÍGUEZ & DUELLMAN, 1994; CALDWELL & ARAÚJO, 2005). In the municipality of Espigão d'Oeste, the capture frequency for this species was greater with proximity to the river, possibly because it provided the most appropriate environment for breeding, especially for tadpole development (see RODRÍGUEZ & DUELLMAN, 1994; LAMAR & WILD, 1995).

Egg-bearing females and calling males were found in the rainy season (September to February), and juvenile recruitment occurred mainly during the end of rainy season to beginning of the dry season (February to June). Although some species of anurans can breed throughout the year (DUELLMAN, 1978), in unstable environments (e.g. that have dry spells) the reproduction of some species is limited to the rainy season (AICHINGER, 1987; MOREIRA & LIMA, 1991). The seasonal recruitment pattern observed in *L. (L.) lineatus* is similar to that observed in other species that depend on standing water for larval development in Central Amazon (MOREIRA & LIMA, 1991).

Our results for size ranges in *L. lineatus* (males 34.9–47 mm, females 38.1–52 mm, juveniles 19.6–35.8 mm) compare with those in the existing literature as follows: male 45 mm, female 56 mm (DUELLMAN, 1978); males 33.7 (average, n = 3) (RODRÍGUEZ, 1992); males 33–45 mm, females 44–56 mm (RODRÍGUEZ & DUELLMAN, 1994); males 33.1–37.5 mm, females 37.7–47.2 mm, juveniles 14.9–15.6 mm (DUELLMAN & MENDELSON, 1995); female 38.9 mm (DUELLMAN & THOMAS, 1996); juveniles 17.6–20.8 (CALDWELL & VITT, 1999); males 33–43 mm, females 48–56 mm (LIMA *et al.*, 2006). The locality with the smallest individuals (of all sexes and age structures) was Loreto, Peru (DUELLMAN & MENDELSON, 1995). The largest females were reported in Manaus, Brazil (LIMA *et al.*, 2006); Santa Cecília, Ecuador (DUELLMAN, 1978), and Iquitos, Peru (RODRÍGUEZ & DUELLMAN, 1994). The largest males were reported in Espigão do Oeste, present study. As in other species of the genus *Leptodactylus* (MARTINS, 1988; GIARETTA & KOKUBUM, 2004), size-related secondary sexual dimorphism was also found in *L. (L.) lineatus*, with females being significantly larger than males.

The ovarian egg numbers recorded for females of

this species are 195 and 230 (DUELLMAN, 1978) and an average of 250 (LIMA *et al.*, 2006). In this study we recorded a mean of 243 eggs, with ranges from 110 to 328 eggs (n = 17). There was a significant correlation between number of ovarian eggs and female size, a pattern which is also observed in other species of the genus *Leptodactylus* (MARTINS, 1988; PRADO *et al.*, 2000; GIARETTA & KOKUBUM, 2004).

*Leptodactylus lineatus* is one of the most abundant leaf litter frogs in the forests of Espigão do Oeste, Rondônia, Southwestern Amazon (Brazil). The number of individuals detected in other studies may be a reflection of the methods used (visual encounter surveys and plots), and not necessarily a lower density. The secretive habits of this frog (males call within nests of leaf cutting ants) do not facilitate visual detection.

Breeding occurs during the rainy season. Calling males and egg-bearing females were recorded from September to February, and juvenile recruitment occurs mainly from the end of rainy season to the beginning of the dry season (February to June).

Males and females show size-related secondary sexual dimorphism, with females being significantly larger than males.

The number of ovarian eggs per female varies from 110 to 328 and there is a significant correlation between number of eggs and body size.

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