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New Distributional Records of the *Bothrops Bilineatus Bilineatus* (Wied 1825) (Serpentes: Viperidae) in Low Xingu River, Pará, Brazil

Emil José Hernández-Ruz, Elciomar Araújo De Oliveira

Laboratório de Zoologia, Faculdade de Ciências Biológicas, Universidade Federal do Pará, Campus de Altamira, Rua Coronel José Porfírio, Altamira – PA, Brazil.

emilhih@yahoo.com

Abstract: We register for the first time B. bilineatus bilineatus for Municipality of Senador José Porfírio, Pará, Brazil (2.568778°S, 51.848583°W, datum: WGS 84). At 10 February 2012. E. J. Hernández-Ruz et al. Collected one exemplar of this species (LZA 703). Whit this article brings another record for B. b. bilineatus, far more than 200 km from the nearest records (Santarém and Tucuruí). Due to the rarity of encounters with B. bilineatus, any record is important because this result may be due to the low number of surveys carried out in these regions, so that there are large gaps in the distribution of B. bilineatus.

Keywords: Snakes, Amazonia, Diversity, lowland rainforest

1. Introduction

The species of viperid *Bothrops bilineatus* (Wied-Neuwied 1821), is known to the Amazon region of Bolivia, Peru, Ecuador, Colombia, Venezuela, Guyana, Suriname, French Guiana and Brazil [1]. Two subspecies are currently recognized: *B. b. Bilineatus* found in Venezuela, Guyana, Suriname, French Guiana and Brazil; and *B. b. smaragdina* Hoge 1966, with records in the Amazon rainforest of Bolivia, Peru, Ecuador, Colombia, Venezuela and Brazil [1].

Bothrops b. bilineatus is found in the Brazilian states of Amazonas, Mato Grosso, Rondônia, Pará, Amapá, Maranhão, Bahia and Rio de Janeiro [2, 1], with the type locality the city of Marabá River Peruhype, Bahia. Despite the records in several Brazilian states, the encounter with this species is very rare. In eastern Pará, despite the intense collections made by Cunha and Nascimento [3] and Cunha et al. [4], few individuals were found. Campbell and Lamar [5] recognize that, due to its arboreal habit pattern of cryptic coloration and low population density in their occurrence areas, they are species of hard to record. Bernarde et al. [2] suggest new trigger points for the states of Amapá, Amazonas, Rondonia and Mato Grosso. In this note, we add new information on the geographical distribution of B. bilineatus (Figure 1), record for the Brazilian state of Pará, on the right bank of the lower Xingu River.

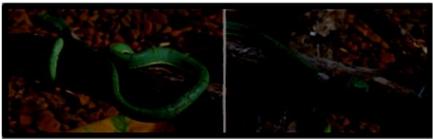


Figure 1. Bothrops bilineatus bilineatus found in the municipality of Senador José Porfírio (A eB).

2. MATERIAL AND METHODS

The specimen used in this study were fixed in 10% formalin and preserved in 70% alcohol. At 10 February 2012. E. J. Hernández-Ruz *et al.* Collected one exemplar of this species (LZA 703). Was collected on vines about 50 cm from the ground. Posterior spawning in plastic bags. The specimens were collected with the authorization for collection and transportation 32401 from SISBIO, Brazil.

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2.1. Molecular Biology

Total genomic DNA was extracted by phenol-chloroform protocol [6] and resuspended with 30 μL of TE. DNA quality was verified by agarose gel electrophoresis. Polymerase Chain Reaction (PCR) was performed to amplify a fragment of 497 bp of the 16S mitochondrial gene. The PCR to fragment of the 16S mitochondrial gene (total volume of 15 μL) contained 6.7 ddH2O μL, 1.5 uL of 25 mM MgCl2, 25.1 μL of 10 mM dNTPs (2.5mM each dNTP), 25.1 μL of 10x buffer (75 mM Tris HCl, 50 mM KCl, 20 mM (NH4)2SO4), 1.5 μL 16S A primer (5'-CGCCTGTTTACCAAAAACATCGCCT-3'), 1.5 μL 16S B primer (5'-CCGGTCTGAACTCAGATCACGT-3') [7], 0.3 μL of 1 U Taq polymerase, and 1 μL of DNA (30 - 50 ng/μL) under the following conditions: an initial temperature of 92°C (60 sec.), followed by 35 cycles of 92°C (60 sec.), 50°C (50 sec.) and 72°C (1.5 min.) and a final extension at 72°C for 7 min. The sequencing reactions were performed using BigDye terminator kit as (*Applied Biosystems*). Following the manufacturer's protocol. The primer forward 16S A (mtDNA), it was used in the sequencing reaction. The products of the sequencing reactions were precipitated with ethanol according to manufacturer's recommendations, resuspended in 10 uL of deionized formamide (ABI) and subsequently injected into the 3130xl ABI automated sequencer (*Applied Biosystems*).

3. RESULTS

The new record was made during a herpetological field collection in the municipality of Senador Jose Porfirio (Figure 2), 02°34′ 51.63 "S, 51° 56′13.47" W. Only one individual was collected, it was curled up in a vine, to about 1.30 m tall. The individual was sacrificed with overdose of chlorhydrate of lidocaine. The tissue was removed from the liver and stored in 95% alcohol, then the individual was fixed with 10% formalin and deposited (LZATM 0703) in Herpetological of Zoology Laboratory Collection, Campus Altamira, Federal University of Pará. We use characteristics presented by Campbell and Lamar [5] to identify the sample.

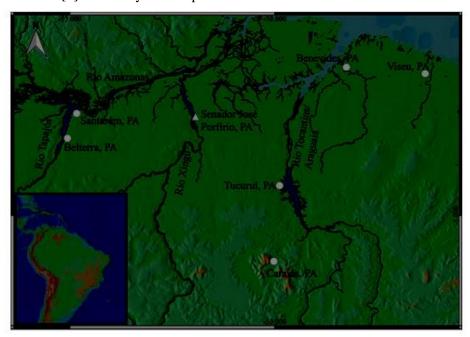


Figure2. Known distribution of Bothrops bilineatus bilineatus to the state of Pará. White circles represent the data from the literature and white triangle new record.

4. DISCUSSION

In the state of Pará, Cunha (1967) records *B. b. bilineatus* for the city of Benevides (01°34'S, 48° 14'W) and Cunha and Nascimento (1993) report to the community of Bela Vista, in the city of Viseu (01°18 'S, 46°16' W), near the Rio Gurupi and the Sierra Norte in the municipality of Carajás (05°51'S, 50°12'W) is suggesting that species is rare in eastern Para. Silva Jr and Sites (1995) record *B. b. bilineatus* for the municipality of Tucuruí, PA, (04°17'S, 49° 38'W), but does not designate subspecies. Campbell and Lamar (2004) also indicate the occurrence of *B. b. bilineatus* to Santarem, PA (2°30'S, 54°50'W) and Curuá-Una region, PA (03°06'S, 55°04'W), however the coordinates provided by the authors refer to the municipality of Belterra, far in about 90 km from Curuá-Una.

Avila-Pires et al. [8] no found *B. b. bilineatus* during inventory in the northern region of Pará. The species was also never found in two large inventories in the regions of Manaus and Presidente Figueiredo, Amazonas [9]. The same is true for several other regions of the Brazilian Amazon [10, 11, 12]. This article brings another record for *B. b. bilineatus*, far more than 200 km from the nearest records (Santarém and Tucuruí). Due to the rarity of encounters with *B. b. bilineatus*, any record is important because this result may be due to the low number of surveys carried out in these regions, so that there are large gaps in the distribution of *B. b. bilineatus*.

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