

A new species of *Melanophryniscus* (Anura, Bufonidae) from the Campos Gerais region of Southern Brazil

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Abstract

A new species of *Melanophryniscus* (Anura, Bufonidae) from the Campos Gerais region of Southern Brazil. A new species of *Melanophryniscus* is described from the Parque Estadual de Vila Velha, municipality of Ponta Grossa, Paraná State, Southern Brazil. The Parque Estadual de Vila Velha is located in the Campos Gerais region, an environment dominated by natural grasslands with patches of Araucaria Forest. The new species is distinguished from all congeners by its small size (12.8-14.0 mm snout-vent length in adult males) and unique color pattern of copper brown dorsum covered with small spinulose black warts; ventral surface black finely spotted with white, posterior abdomen and ventral surfaces of the forearm, hand and foot with red stains. The new species is nocturnal and breeds in the water accumulated in the leaf-axils of phytotelmata.

Keywords: Anura, Bufonidae, species description, phytotelmata, Araucaria Forest, Southern Brazil.

Resumo

Nova espécie de *Melanophryniscus* (Anura, Bufonidae) da região dos Campos Gerais, sul do Brasil. Uma nova espécie de *Melanophryniscus* é descrita do Parque Estadual de Vila Velha, município de Ponta Grossa, estado do Paraná, sul do Brasil. O Parque Estadual de Vila Velha está inserido na região dos Campos Gerais, ambiente composto por um mosaico de campos naturais com capões de Mata de Araucária. A nova espécie difere de todos os seus congêneres pelo pequeno tamanho (12.8-14.0 mm de comprimento rostro-cloacal em machos adultos) e padrão único de coloração, com a superfície dorsal do corpo marrom acobreada coberta com pequenas verrugas negras espinhentas e superfície ventral negra finamente pontuada de branco, abdômen posterior e palma da mão, pé e antebraço com manchas vermelhas. A nova espécie é noturna e se reproduz na água acumulada nas axilas das folhas de fitotelmatas.

Palavras-chave: Anura, Bufonidae, descrição de espécie, fitotelmata, Mata de Araucária, sul do Brasil.

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Introduction

The anuran genus *Melanophrynniscus* Gallardo, 1961 is considered a monophyletic group of species occurring at Northern Argentina, interandean valleys of Southern Bolivia, Southern Brazil, Paraguay, and Uruguay (Graybeal and Cannatella 1995, Frost 2007). The 21 species (Langone *et al.* 2008) in the genus are arranged into three phenetic groups proposed by Caramaschi and Cruz (2002): *M. atroluteus*, *M. cupreuscacularis*, *M. dorsalis*, *M. fulvoguttatus*, *M. klappenbachi*, *M. krauczuki*, *M. montevidensis*, *M. rubriventris*, and *M. stelzneri* (*M. stelzneri* group); *M. moreirae* and *M. sanmartini* (*M. moreirae* group); *M. cambaraensis*, *M. devincenzi*, *M. macrogranulosus*, *M. orejasimirandai*, *M. pachyrhynus*, *M. simplex*, *M. spectabilis* and *M. tumifrons* (*M. tumifrons* group). *Melanophrynniscus admirabilis* and *M. alipioi* were recently described and not assigned to species groups (Di-Bernardo *et al.* 2006, Langone *et al.* 2008). The group definitions are loose, except for *M. tumifrons* group, defined by the presence of a frontal swelling on top of snout (Baldo and Basso 2004), and a comprehensive phylogenetic analysis is required to determine the relationships among species in the genus (Di-Bernardo *et al.* 2006).

The purpose of this paper is to describe a new species of *Melanophrynniscus* from the Parque Estadual de Vila Velha, Campos Gerais region, a unique and ecologically fragile mosaic of Araucaria Forest patches and natural grasslands in Southern Brazil (Maack 2002, Ab'Saber 2003).

Materials and Methods

The description is based on 12 specimens (11 males and one female) collected in January 2006 during a field trip to Parque Estadual de Vila Velha, municipality of Ponta Grossa, Paraná State, Southern Brazil (25°13'S, 50°01'W; Figure 1). The area is composed of

natural grassland with abundance of phytotelmata. A clutch was collected; one egg was preserved and stored in 5% formalin (MHNCI 4886 lot), the other eggs were kept in the laboratory to develop until the larval stage (MHNCI 4886 lot). Two tadpoles and two juveniles were collected in the field (MHNCI 4885 lot). Tadpoles and juveniles were preserved in 5% formalin and stored in 70% ethanol. Adults were collected and fixed in 10% formalin and stored in 70% ethanol. Specimens are deposited in the herpetological collection of the Museu de História Natural Capão da Imbuia (MHNCI), Curitiba, Paraná State, Brazil.

Considering that the specimens collected were readily identified as a new species of *Melanophrynniscus* by its small size and color pattern, species comparisons are restricted to information provided in literature (Bokermann 1967, Klappenbach 1968, Cei 1980, Langone 1994, Céspedes and Alvarez 1999, Prigioni and Langone 2000, Cruz and Caramaschi 2003, Baldo and Basso 2004, Di-Bernardo *et al.* 2006, Langone *et al.* 2008).

Morphological terminology, diagnosis, description and general characteristics follow Caramaschi and Cruz (2002) and Baldo and Basso (2004). Measurements were taken with calipers to the nearest 0.02 mm under a stereoscopic microscope and include: snout-vent length (SVL), head length (HL), head width (HW), eye diameter (ED), interocular distance (IOD), eye-nostril distance (END), internarial distance (IND), upper eyelid width (UEW), thigh length (THL), tibia length (TL), and foot length (FL).

Species Description

Melanophrynniscus vilavelhensis sp. nov.

(Figure 2)

Holotype - MHNCI 4873, adult male collected at Parque Estadual de Vila Velha (25°13'S, 50°01'W; 824 m a.s.l. - Figure 1), municipality of Ponta Grossa, Paraná State,

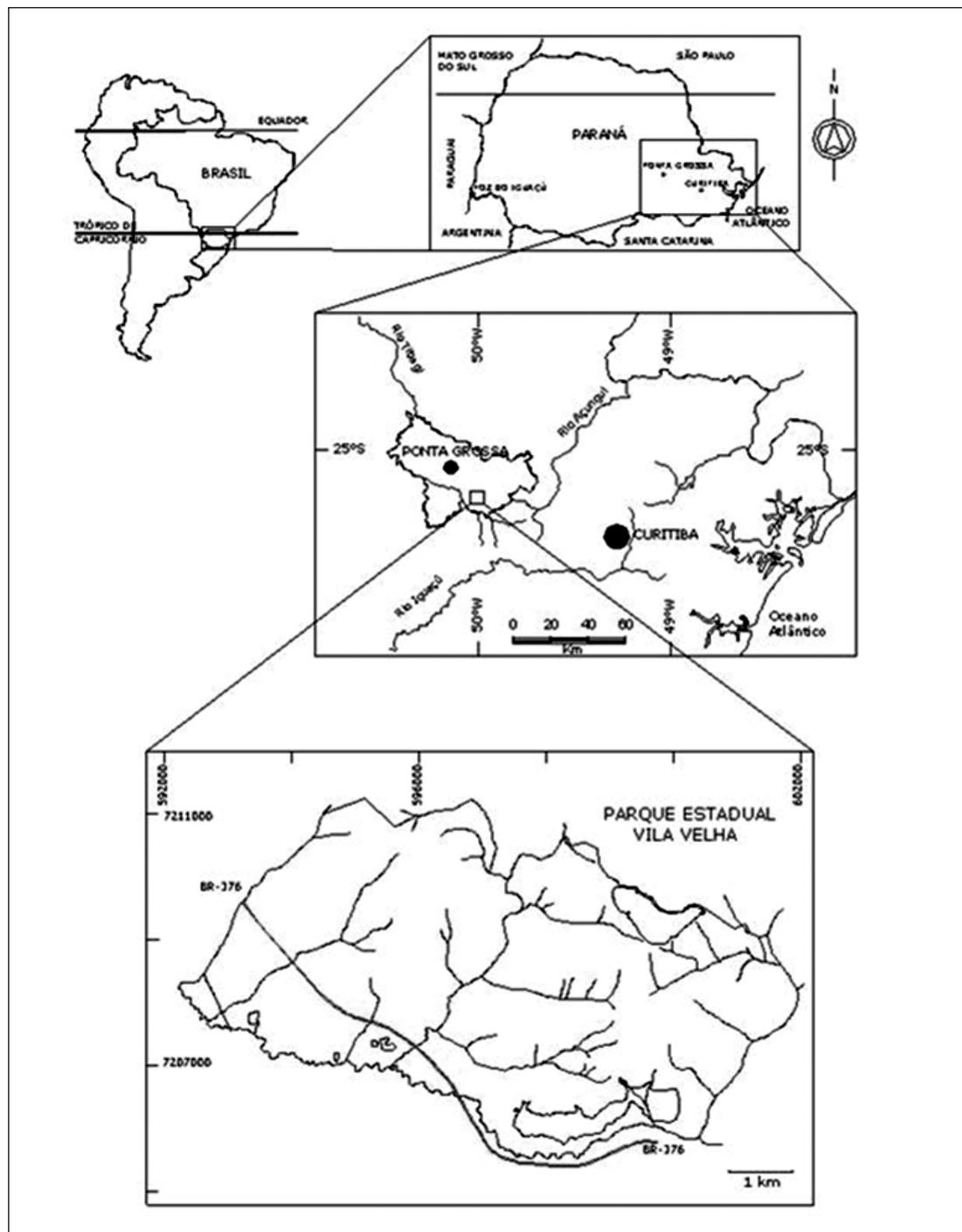


Figure 1 - Location map of the Parque Estadual de Vila Velha, Paraná State, Southern Brazil.

Brazil, on 12 January 2006 by L. B. Crivellari, M. Bornschein and G. C. Steinbach-Padilha.

Paratypes - MHNCI 4874-80, 4882-84, ten adult males; and MHNCI 4881, an adult female; collected on 21 January 2006 by L. B. Crivellari, M. Bornschein and G. C. Steinbach-Padilha.

Etymology - The new species is named in reference to the type locality, the Parque Estadual de Vila Velha, Paraná State, Southern Brazil.

Diagnosis - *Melanophrynniscus vilavelhensis* is diagnosed by a combination of the following characters: (1) small size for the genus (SVL males 12.8-14.0 mm, N=11; female 17.2 mm, N=1); (2) body elongated; (3) head longer than wide; (4) dorsum copper brown, covered with small spinulose black warts; (5) flank black, finely spotted with white posteriorly; (6) limbs black, covered with small spinulose black warts; (7) posterior region of abdomen and ventral surface of hand, forearm and foot with red stains.

Description of holotype - body elongate, limbs slender. Head slightly longer than wide (HW/HL = 0.84); HL approximately 38% of SVL. Snout short, truncated in dorsal view, rounded in lateral profile, and slightly protruding. Nostrils small, oval, laterally directed, located near the tip of the snout. Pupil horizontally elliptical. ED slightly smaller than IOD (ED/IOD = 0.9) and longer than IND (ED/IND = 1.95), UEW (ED/UEW = 1.95) and END (ED/END = 1.63). UEW and IND equal (UEW/IND = 1.0) and smaller than IOD (UEW/IOD and IND/IOD = 0.45). Tympanic membrane concealed; vocal sac subgular; tongue cylindrical, rounded; choanae small, rounded and broadly separated. Fingers short, basal webbed, relative length of fingers IV < I < II < III, finger III twice as long as finger II; finger tips not expanded; outer metacarpal tubercle prominent, large, rounded; inner metacarpal tubercle approximately one third of the outer,

less prominent, elliptic; palmar supernumerary tubercles present, rounded, small. Toes short, basal webbed; relative length of toes I < II < V < III < IV, toe IV three times as long as toe III; toe tips not expanded; metatarsal tubercles elliptic, not prominent, outer metatarsal tubercle twice the size of inner metatarsal tubercle; subarticular tubercles small and poorly developed. Skin of dorsum and limbs rugose, covered by small spinulose black warts. Ventral surface of body predominantly smooth, with small warts concentrated on arms and femoral region.

Color of holotype in life - dorsum copper brown covered by black spinulose warts; interocular region black. Snout with black stains, upper and lower lip margins with small white spots. Limbs black with black spinulose warts; hind limbs with dorsal gray stains; forelimbs finely spotted with light brown proximally and light gray distally. Dorsal surface of hands black with light gray spots; fingers black with light red stains along fingers I and II, small light red spots scattered along finger III, on tip of finger IV and on interdigital membranes. Dorsal surface of foot black, with light gray spots; toes black with light red stains along fingers I and II, scattered and small spots on toes III and IV and on tip of toe V. Black stains from below eyes to axils; lateral body, between fore and hind limbs, black spotted with white, inguinal region copper brown with black stains, with a few inconspicuous white spots. Venter black with white spots and light red stains at the posterior abdomen, hand, foot and forearm; vent surrounded with a dark red stain. Iris black with fine copper spots, and bordered with silver spots.

Measurements of holotype - SVL 13.0, HL 4.9, HW 4.1, ED 1.95, IOD 2.2, END 1.2, IND 1.0, UEW 1.0, THL 5.2, TL 5.0, FL 4.8.

Color of holotype in ethanol - the red color tones on venter become grayish-white. The white and copper brown colors darken slightly,

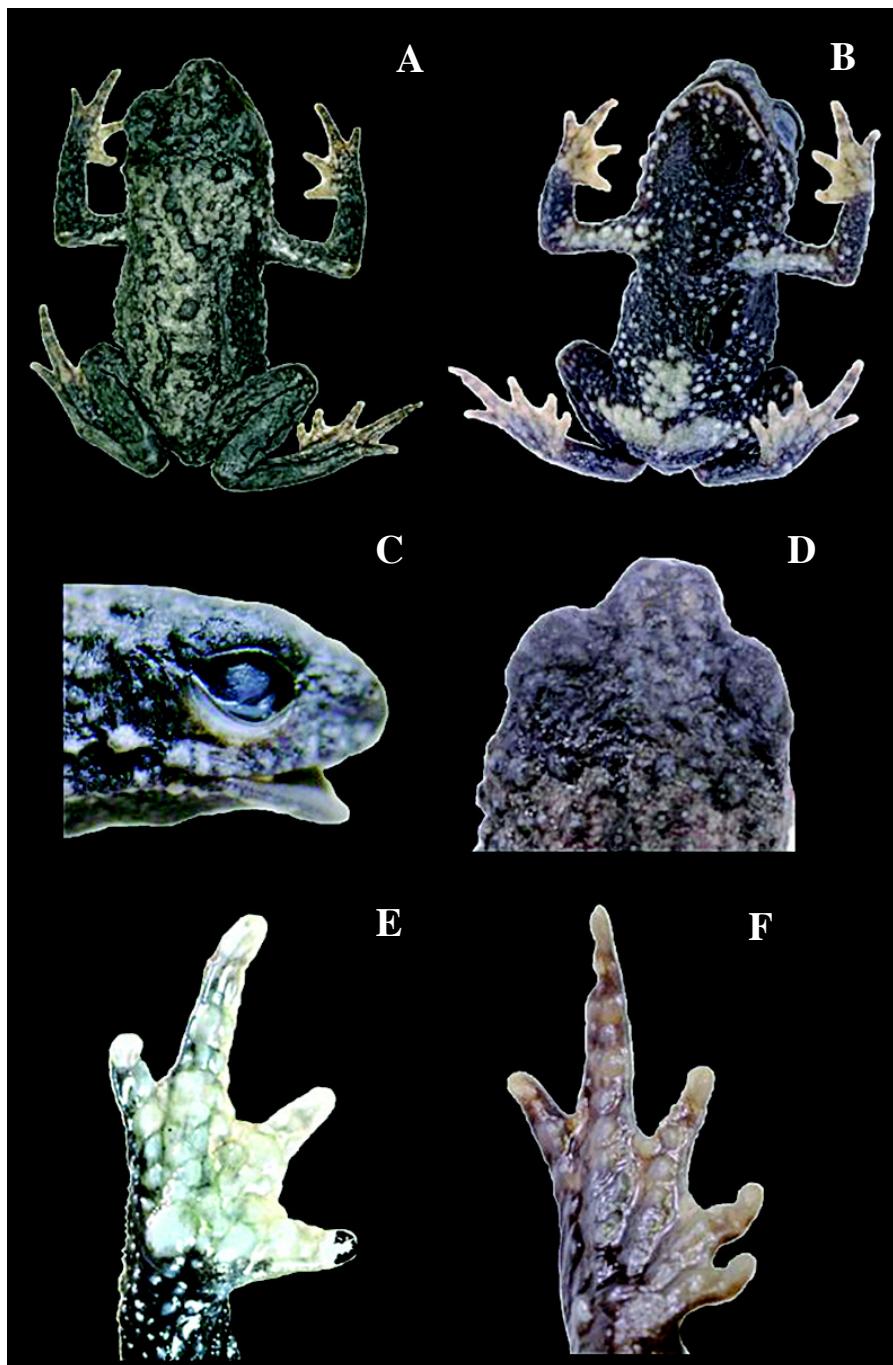


Figure 2 - *Melanophrynniscus vilavelhensis* sp. nov., holotype, MHNCI 4873, 13.0 mm snout-vent length, an adult male in preservative; (A) dorsal view, (B) ventral view, (C) lateral profile of head, (D) dorsal view of head, (E) ventral view of right hand, (F) ventral view of right foot. Photographs by Mendonça Jr.

Table 1 - Measurements (in mm) of *Melanophrynniscus vilavelhensis* sp. nov. paratotypes. Mean ± SD (range).

	MALES (N=10)	FEMALE (N=1)
SVL	13.42 ± 0.5 (12.8-14.0)	17.2
HL	4.89 ± 0.32 (4.2-5.5)	5.5
HW	4.37 ± 0.22 (4.1-4.7)	5.3
ED	1.88 ± 0.11 (1.7-2.0)	2.2
IOD	2.54 ± 0.43 (2.0-3.0)	3.2
END	1.7 ± 0.18 (1.4-1.9)	1.5
IND	1.18 ± 0.18 (1.0-1.5)	1.3
UEW	1.17 ± 0.08 (1.0-1.3)	1.5
THL	4.87 ± 0.4 (4.1-5.5)	6.0
TL	4.32 ± 0.29 (4.0-5.0)	5.0
FL	4.65 ± 0.42 (4.0-5.0)	6.3
BW	5.35 ± 0.49 (4.8-6.1)	8.1

but remain clearly visible. The black color remains unchanged.

Variation - individuals of *Melanophrynniscus vilavelhensis* sp. nov. show variation in size (Table 1) and some differences related to the distribution and color of spots, and the presence of stripes and stains on the head and body. It

includes the presence of a narrow copper brown occipital stripe (present in MHNCI 4874, 4877-78 and 4883), black stains on dorsum (present in MHNCI 4874, 4877-79), fine spots on forelimbs near the hands pale yellow (present in MHNCI 4874-75 and 4880), dorsum black with copper brown longitudinal stripes (present in MHNCI 4882 and 4884), copper brown stains

Table 2 - Snout-vent length of the species of *Melanophrynniscus stelzneri* group, *M. moreirae* group, *M. admirabilis*, and *M. alipioi*.

SPECIES	SVL (mm)	Reference
<i>M. vilavelhensis</i> sp. nov.	12.8-17.2	present study
<i>M. admirabilis</i>	29.5-40.3	Di-Bernardo <i>et al.</i> 2006
<i>M. alipioi</i>	19.4-25.64	Langone <i>et al.</i> 2008
<i>M. atroluteus</i>	18.0-25.0	Langone 1994
<i>M. cupreuscacularis</i>	25.8	Céspedes and Alvarez 1999
<i>M. dorsalis</i>	20.5-24.8	Cruz and Caramaschi 2003
<i>M. fulvoguttatus</i>	23.1-28.0	Cruz and Caramaschi 2003
<i>M. klappenbachi</i>	19.8-31.9	Prigione and Langone 2000
<i>M. krauczuki</i>	18.4-24.2	Baldo and Basso 2004
<i>M. montevidensis</i>	19.0-28.0	Langone 1994
<i>M. rubriventris</i>	38.0-45.0	Cei 1980
<i>M. stelzneri</i>	25.0-30.0	Cei 1980
<i>M. moreirae</i>	23.0-30.0	Bokermann 1967
<i>M. sanmartini</i>	24.5	Klappenbach 1968

on hind limbs (present in MHNCI 4876), dark red stain around the vent with a fine white line (present in MHNCI 4876), and presence of a row of white spots on lateral head from the tympanum to the nostril (present in MHNCI 4877, 4879-80).

Juveniles present dorsal surface of body and limbs black, covered with small silver and white spots; snout as dorsum but without white spots; head laterally black. Foot light brown; flank dark brown. Venter light brown spotted with silver and white, except abdomen, which is black.

Female larger than males (Table 1) and its color pattern differs from that of the males by the following features: a narrow strip formed by pale red spots extends from the nostrils, around the upper lip, to the axils; the skin around urostyle is black. On venter, the light red stain on the abdomen is much larger than in males. Warts in the dorsum are less prominent than in males.

Species comparisons - *Melanophrynniscus vilavelhensis* sp. nov. is promptly distinguished from species of *M. tumifrons* group by the absence of the frontal swelling on top of the snout. From the other congeners, the new species can be promptly distinguished by the smaller size (Table 2) and coloration pattern. Other differences are (characters of *M. vilavelhensis* are in parenthesis): *Melanophrynniscus admirabilis* presents body stout (slender body), snout truncated in lateral view (snout rounded), dorsum pale green with yellow glands (dorsum copper brown); *Melanophrynniscus alipioi* presents ventral surface dark brown, with large, irregular shaped and uniformly distributed bright red spots on each side of pectoral area, extending slightly onto the ventral surface of the arms (venter black with white spots and light red stains on the posterior abdomen, hand, foot, and forearm); *Melanophrynniscus atroluteus* presents snout rounded in dorsal view (snout truncated), black color on dorsum generally without yellow stains and forelimbs with a yellow line on posterior

border (dorsum copper brown); *Melanophrynniscus cupreuscacularis* presents body robust (body slender), smooth skin, dorsum black with copper colored scapular region (dorsum copper brown covered with small spinulose black warts); *Melanophrynniscus dorsalis* presents body robust (body slender) and a fragmented but clear brown stripe on dorsum midline from head to urostyle region (stripe absent), pale yellow blotches on the gular region, chest, belly, and ventral surfaces of forearms and thighs (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm); *Melanophrynniscus fulvoguttatus* presents dorsal surface with numerous and irregular pale yellow spots (dorsum copper brown), a series of pale yellow spots on the border of mandible and ventral surface with numerous irregular pale yellow spots with a remarkably large longitudinal blotch on thighs (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm); *Melanophrynniscus klappenbachi* presents robust body (body slender), dorsum black with large yellow stain irregularly displayed (dorsum copper brown), with the same kind of stains on ventral surface and a large red stain with black patterns on the abdomen (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm); *Melanophrynniscus krauczuki* presents stout body (body slender), dorsum uniformly dark brown (dorsum copper brown) and dark brown ventral surface with irregular orange spots (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm); *Melanophrynniscus montevidensis* presents dorsum black with intense yellow spots (dorsum copper brown); *Melanophrynniscus rubriventris* presents snout truncated in lateral profile (snout rounded in lateral profile), dorsum dark or blackish with two large, converging, triangular yellow spots on the neck and shoulder (dorsum copper brown), two lateral elongated spots beyond the groin, several yellow spots on the back, ventral

surface red, immaculate (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm); *Melanophryniscus stelzneri* presents stout body (body slender), dorsum black or dark with yellow spots on arms and shoulders (dorsum copper brown), small yellow spots on flank (flank black, finely spotted with white posteriorly) and ventral surface black with bright red spots on belly and femur (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm); *Melanophryniscus moreirae* presents dark dorsum (dorsum copper brown), red stains on flank (flank black, finely spotted with white posteriorly); *Melanophryniscus sanmartini* presents dorsum dark brown, light stain over the head extending laterally over the shoulder blade until the base of the arm (dorsum copper brown), with ochre stains in the gular region and in venter, limited posteriorly by a transversal red-orange band with black patterns (venter black with white spots and light red stains on posterior abdomen, hand, foot, and forearm).

Distribution - the new species is known only from the type locality, a natural grassland area with patches of Araucaria Forest, located at Parque Estadual de Vila Velha, Campos Gerais region, municipality of Ponta Grossa, Paraná State, Southern Brazil.

Habitat and natural history - *Melanophryniscus vilavelhensis* sp. nov. is a nocturnal toad. The habitat of the species consists of a marshy area located in a natural field ($25^{\circ}14'82''S$, $50^{\circ}00'24''W$, 824m a.s.l.). *Melanophryniscus vilavelhensis* sp. nov. was found on two occasions. Nine individuals were found between 08:00 pm and 01:00 am on 12-13 January 2006. Two tadpoles, two juveniles and three adults were collected on this occasion from the water accumulated in leaf-axils of *Eringium* sp. (Apiaceae). In 21 January 2006, 15 individuals were heard calling after rain in the water accumulated in the leaf-axils of

terrestrial *Eringium* sp. and *Eriocaulum* sp. (Eriocaulaceae) phytotelmata from 08:45 pm to 12:00 pm. A clutch containing eight eggs was found and collected floating in the water accumulated in the leaf-axils of *Eriocaulum* sp.

Other anuran species found in the study site were *Physalaemus gracilis*, *Dendropsophus minutus* and *Hypsiboas albopunctatus*.

Discussion

Melanophryniscus vilavelhensis sp. nov. is the second species in the genus to use phytotelm as breeding sites. The reproductive mode was recently described by Langone *et al.* (2008) to *Melanophryniscus alipioi*, considered a ‘phytotelm breeding species’, classified as Mode 6 according to Haddad and Prado (2005).

The nocturnal habitats, unusual breeding site, morphology, distinctive color pattern and the diminutive size of *Melanophryniscus vilavelhensis* sp. nov differ dramatically from other members of the genus. The breeding microhabitat and nocturnal behavior of *Melanophryniscus vilavelhensis* sp. nov. are uncommon among members of the genus, that are mostly diurnal and its reproduction normally occurs in ponds or in lotic environments (Bokermann 1967, Braun 1978, Braun and Braun 1979, Céspedes and Alvarez 1999, Baldo and Basso 2004, Di-Bernardo *et al.* 2006, Cairo *et al.* 2008). The use of leaf-axils is not unique among bufonids, and is also present in *Dendrophryniscus brevipollicatus*, which lives and breeds in epiphytic bromeliads (Carvalho 1949).

The conservation status of *Melanophryniscus vilavelhensis* sp. nov. should probably be considered Data Deficient until further information on its distribution and habitat requirements permit a more thorough assessment. Reproductive modes can promote restricted habitat selection dependent on the availability of sites for reproduction (Bertoluci and Rodrigues 2002, Moraes *et al.* 2007) and, associated to this limitation, it is possible that the new species is dependent on the unique

mosaic of grassland and Araucaria Forests of the Campos Gerais region. The discovery of *Melanophrynniscus vilavelhensis* sp. nov. adds to the already high conservation value of these unique habitats.

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