



# New record of *Brachycephalus fuscolineatus* Pie, Bornschein, Firkowski, Belmonte-Lopes & Ribeiro, 2015 (Anura, Brachycephalidae) from Santa Catarina state, Brazil

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## Abstract

*Brachycephalus fuscolineatus* Pie, Bornschein, Firkowski, Belmonte-Lopes & Ribeiro, 2015 was described from Morro do Baú, Santa Catarina, Brazil. This species occurs at elevations of 640–790 m, and it is known to have an extent of occurrence of 23.63 ha. Here, we report a new record of *B. fuscolineatus* from Morro Braço da Onça, Luiz Alves, Santa Catarina. We recorded it on 21–22 December 2018 at elevations between 525 and 530 m; the new record occupied an area of 0.19 ha. We estimate 9 calling males in 36.35 m<sup>2</sup>.

## Key words

Altitudinal distribution, *Brachycephalus pernix* group, conservation, montane forest, population density.

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## Introduction

*Brachycephalus* Fitzinger, 1826 includes small diurnal anurans that inhabit the forest floor of Brazilian Atlantic Forest. Species from this genus have body lengths less than 2.5 cm and a reduced number and size of digits (e.g. Yeh 2007). Some species are brightly colored with neurotoxins in their skin (e.g. Schwartz et al. 2007). Several species are microendemic to one or a few mountain tops (Bornschein et al. 2016a), which highlights the need for conservation, especially because the number of species of the genus is underestimated. *Brachycephalus* currently includes 36 species, 22 of which have been described in the last 10 years. Among these recently described species, 15 were described from southern Brazil, which

reinforces the importance of additional studies for better estimating species richness of the genus and its conservation needs in southern mountains of Brazil.

During the last decades, our efforts were focused on the study and conservation of mountain fauna, with the description of new species (e.g. Langone et al. 2008, Maurício et al. 2014, Ribeiro et al. 2015, 2017, Bornschein et al. 2016b, Pie et al. 2018a), proposition of phylogenetic arrangements and biogeographic processes (e.g. Firkowski et al. 2016, Pulido-Santacruz et al. 2016, Maurício and Bornschein 2017, Pie et al. 2018b), and novelties of species distribution (e.g. Bornschein et al. 2012, 2016a, Teixeira et al. 2018). In this study, we present a new record for *Brachycephalus fuscolineatus* Pie, Bornschein, Firkowski, Belmonte-Lopes & Ribeiro, 2015, a

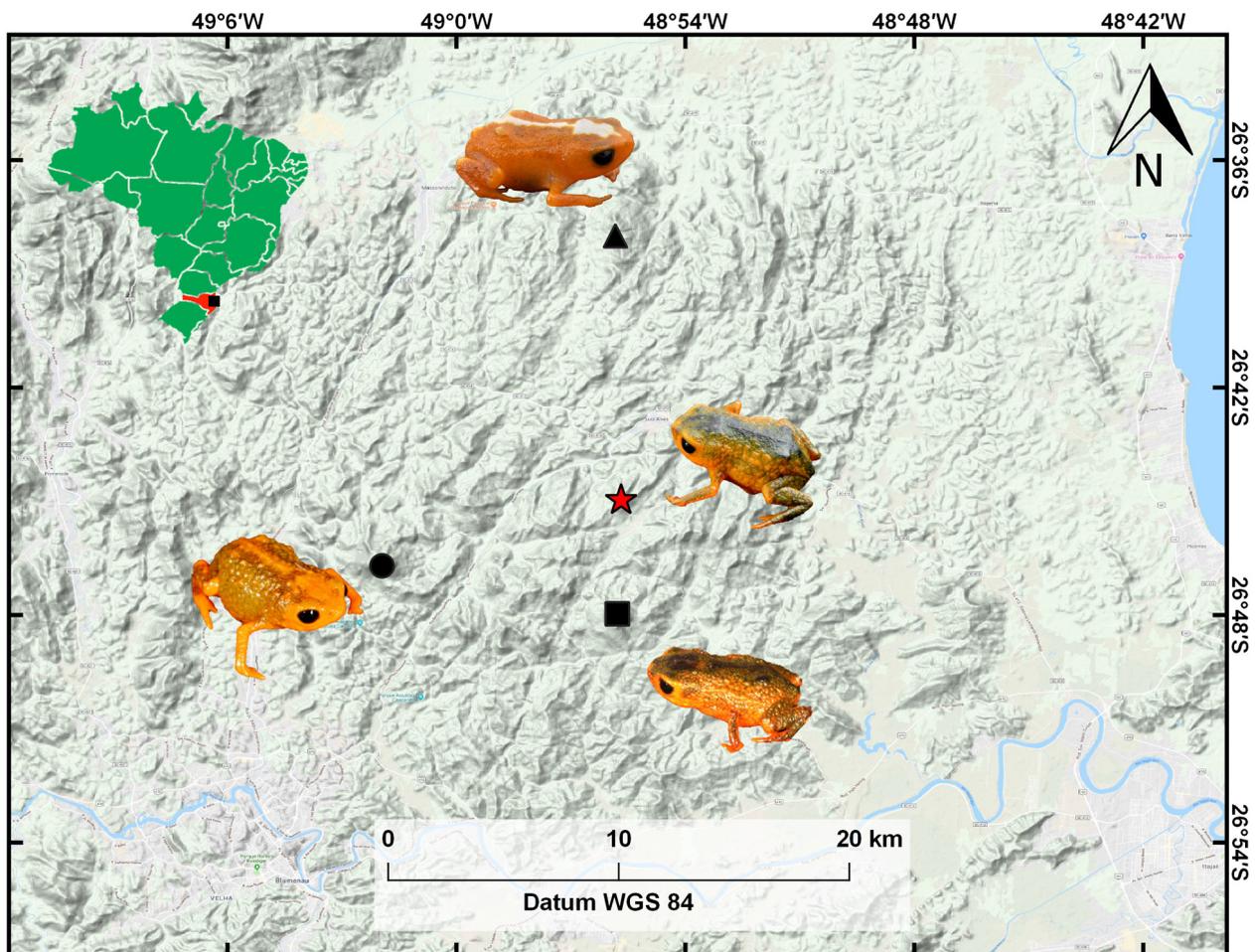
recently described species that until now was only known in its type locality (Ribeiro et al. 2015). In addition, to aid in the identification of new specimens, we reassess the color variation of the type series of *B. fuscolineatus*.

## Methods

We participated in field trips in high altitudes in order to find populations of *Brachycephalus*. We worked during the day, walking as quietly as possible along the margins of roads and trails, as well as in the interior of the forest, for acoustic identification of *Brachycephalus*. Upon detection of its particular sound, we approached to make recordings. After that, we removed the leaf litter to try to find specimens. We searched for *Brachycephalus fuscolineatus* at Morro Braço da Onça (21–22 December 2018) at 370–540 m above sea level (a.s.l.) in primary and secondary forests, *Eucalyptus* plantations, and palm (*Archontophoenix alexandrae* H. Wendl. & Drude) plantations. We collected specimens for identification and photographic documentation. ICMBio/SISBIO provided a collection permit (#55918-1). We anaesthetized and euthanized specimens using 2% chloridrate lidocaine, fixed them in 10% formalin, and stored them in 70% ethyl alcohol solution. We deposited the specimens in the Museu de História Natural Capão da Imbuia (MHNCI), Curitiba, state of Paraná.

We recorded the geographical coordinates from points where *B. fuscolineatus* was found using a Garmin etrex 10 GPS (datum WGS84). We retrieved the altitude of these points from Google Earth Pro v. 7.1.4.1529 when plotting the points, as in Bornschein et al. (2016a). We made a polygon connecting points of records of the species by the minimum convex polygon method using Google Earth, as recommended by IUCN (2012). We then estimated the area of the polygon using GEPATH v. 1.4.5. We classified the area of the polygon as “extent of occurrence” or “area of occupancy” (IUCN 2012) in accordance with the distribution pattern of calling males (Bornschein et al. 2016b), as homogeneously distributed through the area (= area of occupancy) or patchily distributed (= extent of occurrence). We treated as patchily distributed locations in which we found patches with at least 2,500 m<sup>2</sup> without records of calling males between patches with records of calling males that were just as large as or even larger.

We estimated calling males density of *B. fuscolineatus* as Bornschein et al. (2016b), Ribeiro et al. (2017), and Pie et al. (2018a). We walked slowly in the forest for about 0.5 h, listening for individuals. In one place where we heard several males close to each other, we delimited the outer perimeter of a polygon by fixing a white line



**Figure 1.** Geographic distribution of *Brachycephalus fuscolineatus*, showing the new record (red star) and previous records (black square) from the literature (Ribeiro et al. 2015), in Santa Catarina, southern Brazil. Type localities of *B. boticario* (black dot; Ribeiro et al. 2015) and *B. mirissimus* (black triangle; Pie et al. 2018a) are also indicated.

**Table 1.** Estimated density of *Brachycephalus* spp.

Species	Locality, state	Altitude (m a.s.l.)	Sampled area (m <sup>2</sup> )	Density		Source
				One individual (independent of sex) per area (m <sup>2</sup> )	One male per area (m <sup>2</sup> )	
<b><i>B. didactylus</i> group</b>						
<i>B. didactylus</i> (Izecksohn, 1971)	Fazenda Santa Bárbara, Parque Estadual dos Três Picos, municipaliy Cachoeiras de Macacu, Rio de Janeiro	500–850	625	25.0	—	Almeida-Santos et al. 2011
	Fazenda Santa Bárbara, Parque Estadual dos Três Picos, municipaliy Cachoeiras de Macacu, Rio de Janeiro	500–800	625	25.0	—	Siqueira et al. 2009
	Monumento Natural Serra das Torres, municipality of Atílio Vivacqua, Espírito Santo	600–900	5,280	52.9	—	Oliveira et al. 2013
	Reserva Ecológica de Guapiaçu, municipality of Cachoeiras de Macacu, Rio de Janeiro	300–520	1,250	83.3	—	Siqueira et al. 2014
	Reserva Ecológica Rio das Pedras, municipality of Mangaratiba, Rio de Janeiro	60–750	750	83.3	—	Almeida-Santos et al. 2011
	Reserva Rio das Pedras, municipality of Mangaratiba, Rio de Janeiro	20–750	750	83.3	—	Rocha et al. 2013
	Vila Dois Rios, Parque Estadual da Ilha Grande, Ilha Grande, municipality of Angra dos Reis, Rio de Janeiro	220–230	640	320	—	Rocha et al. 2000
		220–230	640	160	—	
		220–230	640	91.4	—	
		200–300	2,560	20	—	
<i>B. didactylus</i>	Vila Dois Rios, Parque Estadual da Ilha Grande, Ilha Grande, municipality of Angra dos Reis, Rio de Janeiro	220–230	1,536	192.3	—	Rocha et al. 2001
		220–230	180	5.6	—	
	Vila Dois Rios, Parque Estadual da Ilha Grande, Ilha Grande, municipality of Angra dos Reis, Rio de Janeiro	220–240	468	5.1	—	Van-Sluis et al. 2007
<i>B. hermogenesi</i> (Giaretta & Sawaya, 1998)	Reserva Particular do Patrimônio Natural Salto Morato, municipality of Guaraquecaba, Paraná	200–300	2,560	100	—	Santos-Pereira et al. 2011
<b><i>B. ephippium</i> group</b>						
<i>B. ephippium</i>	Serra do Japi, municipality of Jundiá, São Paulo	1,000	1,344	1,344	—	Giaretta et al. 1997
<i>B. pitanga</i>	Núcleo Santa Virgínea, Parque Estadual da Serra do Mar, municipality of São Luiz do Paraitinga, São Paulo	?	?	0.8	—	Oliveira 2013
		?	?	0.8	—	
		?	?	0.7	—	
<b><i>B. pernix</i> group</b>						
<i>B. albolineatus</i>	Morro Boa Vista, on the border between the municipalities of Jaraguá do Sul and Massaranduba, Santa Catarina	815–835	?	—	3–4	Bornschein et al. 2016b
		790	?	—	100	
<i>B. curupira</i>	Serra do Salto, Malhada District, municipality of São José dos Pinhais, Paraná	1,160	?	—	2–3	Ribeiro et al. 2017
		1,130	?	—	6–7	
<i>B. fuscolineatus</i>	Morro Braço da Onça, municipality of Luiz Alves, Santa Catarina	530	36.35	—	4.0	This study
<i>B. mirissimus</i>	Morro Santo Anjo, municipality of Massaranduba, Santa Catarina	535	202	—	14.5	Pie et al. 2018a

on the vegetation and listened to the individuals inside it for 3 h, placing short pieces of white strands in vegetation over the litter where we heard males. The effort was made by 2 people, that moved inside the polygon very slowly to confirm the position of the singers and to place the strands. We classified the vegetation of the region according to the Brazilian Vegetation Classification System (Veloso et al. 1991).

## Results

**New records.** Brazil: Santa Catarina, municipality of Luiz Alves: Morro Braço da Onça (26°44'58" S, 048°55'41" W;

525–530 m a.s.l.) (Figs 1, 2), 22 December, 2018, coll. by Marcos R. Bornschein, Luiz F. Ribeiro, Larissa Teixeira, and Júlia Grigolato (MHNCI 10850–1; males; Fig. 3).

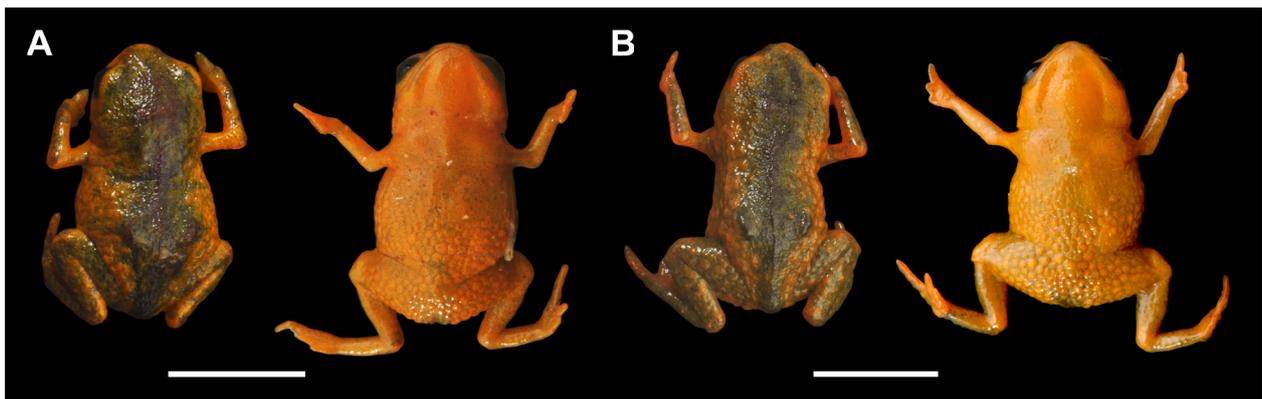
We found *Brachycephalus fuscolineatus* on 21–22 December 2018 in the leaf litter of primary forest (Floresta Ombrófila Densa Montana), about 15 m from the forest edge at 525–530 m a.s.l. Our estimation of the area of the polygon in which we heard males is only 0.19 ha of area of occupancy. We estimated 9 calling males within an area of 36.35 m<sup>2</sup> (= 1 male for every 4.0 m<sup>2</sup>; Table 1). The collected specimens were found inside curled leaves; one of specimens was on a large (ca 30 cm) leaf of *Bathysa australis* (A.St.-Hil.) K.Schum.



**Figure 2.** Top of the Morro Braço da Onça showing the slope in which we recorded *Brachycephalus fuscolineatus*, at least 15 m inland from the forest edge. In the left side, a part of the structures of a radio tower, opened areas, and *Eucalyptus* sp. can be observed. Urbanized areas can also be seen along a road, at lower altitudes.

from the collected specimens. We compared these color characteristics among the collected specimens (see lists of these specimens in Ribeiro et al. 2015, 2017, Bornschein et al. 2016b, Pie et al. 2018a and Teixeira et al. 2018).

The original description of *B. fuscolineatus* (Ribeiro et al. 2015) does not report green in the dorsal stripe of this species, only brown. However, there was green coloration on the margin of the brown dorsal stripe of the holotype (Fig. 4) and paratypes (Fig. 5) of this species, similarly to the specimen of Morro Braço da Onça MHNCI 10850 (Fig. 3A). Actually, at least 2 paratypes (DZUP 160 and 404) show very little brown in a predominantly green dorsal stripe (Fig. 5C, D). On the contrary, the specimen collected in this study (MHNCI 10851; Fig. 3B) presents a dorsal stripe exclusively green, without traces of brown. The original description of *B. fuscolineatus* also does not report yellow coloration on the body, only orange. Nonetheless, some paratypes were indeed yellow color with an orange wash (Fig. 5), as was our specimens from Morro Braço da Onça.

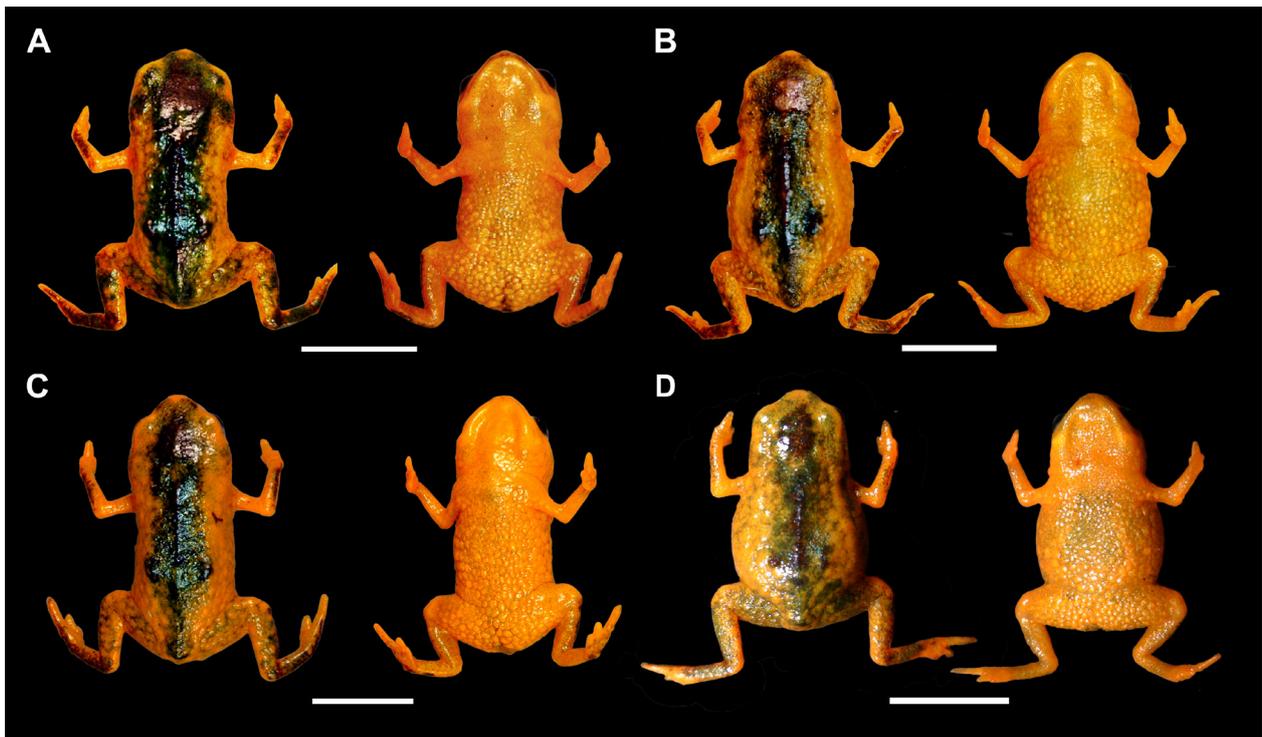


**Figure 3.** Males of *Brachycephalus fuscolineatus* collected at Morro Braço da Onça, municipality of Luiz Alves, Santa Catarina, southern Brazil. Live adults are shown in dorsal (left) and ventral (right) views. **A.** MHNCI 10850. **B.** MHNCI 10851. Abbreviation: MHNCI = Museu de História Natural Capão da Imbuia, Curitiba, Paraná. Scale bar = 5 mm.

**Identification.** The specimens collected are members of *Brachycephalus pernix* group by presenting a bufoniform body shape and linea masculina (Pie et al. 2018a). Within this group, *B. fuscolineatus* can be distinguished from *B. ferruginus* Alves, Ribeiro, Haddad & Reis, 2006, *B. izecksohni* Ribeiro, Alves, Haddad & Reis, 2005, *B. leopardus* Ribeiro, Firkowski & Pie, 2015, *B. mariaeterezae* Bornschein, Morato, Firkowski, Ribeiro and Pie, 2015, *B. mirissimus* Pie, Ribeiro, Confetti, Nadaline & Bornschein, 2018, *B. pombali* Alves, Ribeiro, Haddad & Reis, 2006, and *B. tridactylus* Garey, Lima, Hartmann & Haddad, 2012 by having a broad greenish dorsal stripe, instead of entirely yellow or orange dorsal parts, or at least with a narrow blue stripe (*B. mariaeterezae*), white stripe (*B. mirissimus*), or orange dots (*B. ferruginus*) on dorsal parts. The remaining species of the *B. pernix* group have different pattern of body coloration and are very distinctive



**Figure 4.** Holotype of *Brachycephalus fuscolineatus* DZUP 159 a few minutes after being killed and fixed in formalin, in dorsal (left) and ventral (right) view. Abbreviation: DZUP = Coleção Herpetológica do Departamento de Zoologia, Universidade Federal do Paraná, Curitiba, Paraná. Scale bar = 5 mm.



**Figure 5.** Paratypes of *Brachycephalus fuscolineatus*, all alive adults, in dorsal (left) and ventral (right) views. A = DZUP 402; B = DZUP 401; C = DZUP 404; D = DZUP 160. Abbreviation: DZUP = Coleção Herpetológica do Departamento de Zoologia, Universidade Federal do Paraná, Curitiba, Paraná. Scale bar = 5 mm.

## Discussion

The new locality is 5.4 km north of the type locality of *Brachycephalus fuscolineatus* (Fig. 1). It is also 10.8 km east/northeast from the type locality of *B. boticario* (Ribeiro et al. 2015) and 13.4 km south of the type locality of *B. mirissimus* (Pie et al. 2018a; Fig. 1). The altitudinal range of occurrence of *B. fuscolineatus* is also extended with this new record. At the type locality, *B. fuscolineatus* was recorded at 640–790 m a.s.l. (Ribeiro et al. 2015), but we found it at 525–530 m a.s.l. in Morro Braço da Onça. Bornschein et al. (2016a) estimated an extent of occurrence in 23.63 ha, that is now extended to 23.82 ha.

The estimated density of *B. fuscolineatus* of 1 male every 4.0 m<sup>2</sup> is lower than that estimated for *B. curupira* Ribeiro, Blackburn, Stanley, Pie & Bornschein, 2017 (1 male each 2–3 m<sup>2</sup>) and is similar to the estimate for *B. albolineatus* Bornschein, Ribeiro, Blackburn, Stanley & Pie, 2016 (1 male each 3–4 m<sup>2</sup>) but greater than that obtained for *B. mirissimus* (1 male each 14.5 m<sup>2</sup>; Table 1). All these estimates counted calling males of species of the *B. pernix* group (Table 1). In Table 1, we compiled other estimates of densities for *Brachycephalus* (data for both males and females). There are estimates of densities as high as 1 individual per less than 1 m<sup>2</sup> for *B. pitanga* Alves, Sawaya, Reis & Haddad, 2009, as well as reduced densities such as 1 individual in 1,344 m<sup>2</sup> for *B. ephippium* (Spix, 1824), both of the *B. ephippium* group (Table 1). Due to the differences in methods, we consider the proportion of 1 female for each male singer sampled,

reducing by half the estimated area for individuals of *B. albolineatus*, *B. curupira*, *B. fuscolineatus*, and *B. mirissimus*, regardless of sex.

The fauna and flora from Morro Braço da Onça presents a mixture of montane and low-altitude elements. Other species recorded at the locality, apart from *B. fuscolineatus*, are typically montane birds (*Attila phoenicurus* Pelzeln, 1868, *Carpornis cucullata* (Swainson, 1821), and *Scytalopus speluncae* (Ménétrières, 1835); taxonomy according to Mauricio et al. 2010). Species that are indicative of lowland or submontane forests include the plants *Attalea dubia* (Mart.) Burret, *Bathysa australis*, *Cecropia glaziovii* Sneath., *Euterpe edulis* Mart., *Geonoma gamiova* Barb.Rodr., and *Guadua angustifolia* Kunth and the bird *Phylloscartes kronei* Willis & Oniki, 1992.

The restricted area of occurrence we estimated for *B. fuscolineatus* at Morro Braço da Onça was surprising. This locality is highly degraded by the presence of roads, the edge effects of openings in the forest along the edges of roads (which let in sunlight and lower the humidity), palm and *Eucalyptus* plantations, and the use of herbicides. We do not know if deforestation has reduced the area of occurrence of the species at this locality, although deforestation reduces moisture at the edge of forest (Laurance et al. 2002) and affects the distribution of species. In Morro Braço da Onça, we only recorded *B. fuscolineatus* 15 m away from the forest edges. We also recorded this species only at 525–530 m a.s.l., despite also searching at 350 m a.s.l. and above. *Brachycephalus fuscolineatus* is a member of the *B. pernix* group (Ribeiro et al. 2015), a montane group with

few records at lower altitudes (Bornschein et al. 2016a). Additionally, the Morro Braço da Onça is in a region that was predicted as having low climatic suitability for the occurrence of *Brachycephalus* (Pie et al. 2013). We do not rule out the possibility that Morro Braço da Onça represents a relict distribution, probably highly reduced by climate change and deforestation.

Although the forests at the type locality of *B. fuscolineatus* are more intact and the species there has a greater extent of occurrence (Bornschein et al. 2016a), its presence in fragments such as those reported here may be important for monitoring population trends. In the long term, population decline and changes in vegetation, might be first noticed in smaller fragments than in larger ones.

The precarious state of conservation of eastern mountains of Santa Catarina, including Morro Braço da Onça is worrisome. The mountains of this region are strategic places for the installation of cellphone towers, which can result in the suppression of vegetation along roads, construction of power plants and common housing, accumulation of construction and tourist waste on mountain slopes, introduction of invasive alien plants, and selective cutting of trees, among other impacts. The mountains are also excellent areas for the expansion of *Eucalyptus* and *Pinus* plantations and for the cultivation of the palm *Archontophoenix alexandrae*. There are 4 species of *Brachycephalus* endemic of only 1 or 2 mountains in eastern Santa Catarina state (*B. albolineatus*, *B. boticario*, *B. fuscolineatus*, and *B. mirissimus*), and many threats occur in this area. It is therefore important to search the region for new occurrences of these species and to find areas of priority for conservation and species management.

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## Authors' Contributions

MRB, LFR, and LT participated on field work and data collection; MRB and LFR conducted the analysis; MRB wrote the text; LFR and MRB took photographs.

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