ECOLOGICAL NICHE MODELING AND AN UPDATED GEOGRAPHICAL DISTRIBUTION MAP OF *Leptodactylus notoaktites* HEYER, 1978 (ANURA, LEPTODACTYLIDAE) WITH A NEW OCCURRENCE RECORD

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**Abstract:** *Leptodactylus notoaktites* Heyer, 1978 (Anura, Leptodactylidae) is a Neotropical frog that can be found in open areas, forest edges, and inside forest clearings in southern Brazil. In this study, we present an updated distribution map of this species for a variety of vegetation types in the Atlantic Rain Forest and report a new occurrence record in the Alto Paraná Atlantic Forests. We also performed an Ecological Niche Modeling (ENM) which combined environmental variables with occurrence records to predict environmentally suitable areas for this species. Our study was based on published data, specimens collected in the field, and specimens from Brazilian herpetological collections. The ENM predicted high environmental suitability ranging from São Paulo, Paraná, and Rio Grande do Sul states, mainly in Serra do Mar Coastal Forest areas, while the lowest values were in the states of Rio de Janeiro, Minas Gerais, Mato Grosso do Sul and some inland regions in the states of São Paulo and Paraná. Based on the distribution of *L. notoaktites*, we suggest that field efforts should be extended to inland regions of the Atlantic Rain Forest. In fact, species restricted to coastal regions of the Atlantic Rain Forest could have larger ranges than expected if data from such inland regions was available.

**Keywords:** advertisement call; anurans; Atlantic Rain Forest; environmental variables; Paraná state.

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**INTRODUCTION**

*Leptodactylus notoaktites* Heyer, 1978 (Anura, Leptodactylidae) belongs to the *L. mystaceus* species complex, which is placed in the *L. fuscus* species group (de Sá *et al.* 2014). This species is a medium-sized frog (mean snout-vent length = 47.4 mm) characterized by an upper shank and tibial barred pattern, two distinct dorsolateral folds, posterior thigh with light stripe, some individuals have white tubercles on the sole of the foot (Figure 1), and only individuals with a light mid-dorsal...
stripe have six dorsolateral folds (Heyer 1978). The species presents a white lip and black rostral range from the snout to the eardrum (Forlani et al. 2010). As for most species in the L. fuscus group, L. notoaktites lays its eggs in foam nests built in underground burrows, and as the pond floods the exotrophic tadpoles develop (Haddad & Prado 2005).

**Figure** 1. Vouchered adult male alive (a) of *Leptodactylus notoaktites* (Anura, Leptodactylidae) from the Parque Estadual Mata São Francisco, between Cornélio Procópio and Santa Mariana municipalities, state of Paraná, Brazil. Photos of the preserved specimen as follows: (b) dorsal and (c) ventral views; (d) detail of the sole of foot showing presence of white tubercles (white arrow) and (e) detail of posterior thigh with light stripe (white arrows). Photo (a) by Luis Fernando Storti; (b to e) by Guilherme de Toledo Figueiredo.

*Leptodactylus notoaktites* is found in open habitats along rivers, forest edges, and clearings inside forests. Such clearings inside forests are natural or the result of anthropogenic disturbance (Skuk & Heyer 2004). Currently, the species is distributed in the states of São Paulo, Paraná, and Santa Catarina, mainly along the coast up to 900 m a.s.l., which includes several protected areas (Skuk & Heyer 2004, de Sá et al. 2014). Despite being classified by the Red List of Endangered Species of International Union for Conservation of Nature as Least Concern and the fact that the species has a stable population trend, potential threats to this species include habitat loss due to deforestation, advance of agricultural areas, and infrastructure development for tourism, which makes additional research on the species geographical distribution necessary (Skuk & Heyer 2004).

Ecological Niche Modeling (ENMs) are important tools when evaluating species range, as they provide robust predictions of distributions or suitable environmental regions for species. Given their strong dependence on environmental variables (Duellman & Trueb 1994), ENMs seem particularly valid for anurans. ENMs use environmental variable associations and known species occurrences regions to generate models that define abiotic conditions where survival and reproduction are possible for the populations (Guisan & Thuiller 2005). Furthermore, these models can be used to define cryptic species (Raxworthy et al. 2007), predict species with potential invasion success (Peterson 2003, Peterson et al. 2006), maintain rare or endangered species (Engler et al. 2004), determine impacts of climate changes (Wiens et al. 2009), determine priority conservation areas (Chen 2009), and model the spread of crop pests (Venette et al. 2010).

Currently, there are several methods to generate ENMs from a set of environmental variables such as BIOCLIM (Nix 1986), MARS (Friedman 1991), DOMAIN (Carpenter et al. 1993), GARP (Elith et al. 2006), and Maxent (Phillips et al. 2006). However, in some particular situations uncertainties in model predictions can arise (Araújo & New 2007). Most of such limitations are due to the fact that ENMs use only presence and/or absence data, which can create a narrower true distribution than expected (Sinclair et al. 2010, Vasconcelos et al. 2012). If ENMs could include information on biotic interactions or restrictions then dispersal models would become more realistic. To avoid this limitation, it is possible to establish a priori criteria to build the ENMs. Priori criteria include: (I) potentially or abiotically suitable occurrence areas of an organism in a
future climate change scenario, and (II) areas with suitable environmental conditions for organisms with little geographical distribution information (Vasconcelos & Nascimento 2016, Vasconcelos et al. 2017).

Herein, we provide an updated distribution map and new occurrence record of *L. notoaktites* with comments on its distribution across several types of vegetation in the Atlantic Rain Forest, based on published data, a specimen collected by the authors, and specimens housed in Brazilian herpetological collections. With these information, we generated a map that indicates areas with potentially suitable environmental conditions for *L. notoaktites* using an ENM approach.

**MATERIAL AND METHODS**

**Species data collection**

A single calling male of *L. notoaktites* (MZUEL-1575; snout-vent length = 49.85 mm) was collected on October 23, 2011 at the Parque Estadual Mata São Francisco (PEMSF), a forest remnant of 865 ha located between the municipalities of Cornélio Procópio and Santa Mariana, state of Paraná, Brazil. To confirm its identity, we recorded the specimen advertisement call (air temperature 26°C) at night in an underground burrow at the forest’s edge. Calls were recorded using a SONY ICD PX-820 digital recorder coupled with a Yoga HT-81 microphone and analyzed with a sampling frequency of 44 kHz and sample size of 16 bits. We analyzed the calls with Raven Pro 1.5 for Macintosh (Cornell 2014) and constructed audiospectrograms in R software using the package “seewave” (Sueur et al. 2008; R Development Core Team 2014) with the following parameters: FFT window width = 256, Frame = 100, Overlap = 75, and flat top filter. Terminology used in the call description follows Köhler et al. (2017). For morphological and acoustic comparisons with all calls from the *L. mystaceus* species complex (Table 1), we used data from Heyer et al. (1996) and de Sá et al. (2014). Four advertisement calls of the collected male in this study were deposited at FNJV (Fonoteca Neotropical "Jacques Vielliard", Universidade Estadual de Londrina, Londrina, Paraná) sound collection (FNJV-33018 to 33021). Another individual was analyzed at the Museu de Zoologia of Universidade Estadual de Londrina (MZUEL-1333), collected by M. Z. de Lima on April 15, 2009 at Parque Ecológico Klabin, in the municipality of Telêmaco Borba and mentioned in the unpublished thesis of Machado (2004).

All data based on previously published studies (see Table 1) were obtained by searching in the database of “Scientific Eletronic Library Online” (Scielo; www.scielo.org) and “Google Scholar” (www.scholar.google.com) on March 2016 with the following search terms: “*Leptodactylus notoaktites*” and “*Leptodactylus notoaktites* distribution”. We collected data from the SpeciesLink Database (Cria 2016) and analyzed specimens housed in the following Brazilian herpetological collections: CFBH (Coleção Célio Fernando Baptista Haddad, Universidade Estadual Paulista, Rio Claro, São Paulo); DZSJRP-Amphibia-adults (Coleção de Anfíbios DZSJRP, Universidade Estadual Paulista, São José do Rio Preto, São Paulo); MCP-Anfíbios (Coleção de Anfíbios, Pontifícia Universidade Católica do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul); SinBiotia (Sistema de Informação do programa Biotia /FAPESP, Fundação de Amparo à Pesquisa do Estado de São Paulo, Campinas, São Paulo), ZUEC-AMP(Coleção de Anfíbios do Museu de Zoologia da UNICAMP, Campinas, São Paulo), UFMG-AMP (Coleção de Anfíbios do Centro de Coleções Taxonômicas da Universidade Federal de Minas Gerais, Belo Horizonte, Minas Gerais); FNJV (Fonoteca Neotropical "Jacques Vielliard", Universidade Estadual de Campinas, Campinas, São Paulo); and MZUEL (Coleção de Herpetofauna do Museu de Zoologia da Universidade Estadual de Londrina, Londrina, Paraná). We only considered records those identified with the term "*Leptodactylus notoaktites*". Records containing taxonomic inaccuracies (e.g., cf., aff. and gr.) were not considered. Vegetation types in the distribution map of *L. notoaktites* followed the ecoregions of Olson et al. (2001).

**Environmental data collection**

We used variables associated with the biological conditions necessary for the occurrence of the taxon: (i) 19 bioclimatic variables available at WorldClim (Fick & Hijmans 2017), which are already well documented in the literature as important bioclimatic variables for amphibian...
distributions (e.g., Duellman & Trueb 1994); (ii) altitude, slope and aspect. Altitude is highly correlated with temperature and humidity (Hoersch et al. 2002) and with solar radiation that plays an important role on moisture for habitat selection by amphibians (Wyman 1988). Slope and aspect were related to a proxy for the amount of solar radiation on the ground surface (Blank & Blaustein 2012); (iii) vegetation type (biome characteristics), which is related to species habitat (Toledo & Batista 2012); and (iv) percent of tree cover (MODIS), which affects amphibian species composition and distribution (Figueiredo et al., unpublished data; Skelly et al. 1999). Bioclimatic variables were obtained from WorldClim (WorldClim database version 2.0, http://www.worldclim.org/) and were interpolated to 30 arc-sec resolution (Fick & Hijmans 2017) with WGS84 projection; altitude, slope, aspect, and vegetation type from National Aeronautics and Space Administration - NASA (http://www2.jpl.nasa.gov/srtm/; Amaral et al. 2013); and percent of tree cover from Global Landcover Facility (http://glcf.umd.edu/data/; Amaral et al. 2013).

Model building and evaluations
We used the 35 records (see Table 1) to generate the ENM based on the machine-learning modeling method Maxent (maximum entropy algorithm; Phillips et al. 2006), which estimates areas with potentially suitable environmental conditions for *L. notoaktites*. In this approach the niche suitability ranges from zero to one; the closer to one, the greater environmental suitability for the species. To avoid over-prediction and low specificity for species distribution (i.e., Amazonian Rainforest), we cropped the bioclimatic layers to span from latitude -18 to -35 and longitude -38 to -58 (values in decimal degrees). To avoid model over-parameterization, we removed multicollinearity variables \((r > 0.8)\) and variables with low contribution to the model \((< 1\%)\) using a Jackknife test of variable importance determined by their biological relevance for *L. notoaktites*. We used this approach because the test excluded one variable at a time when running the model (Baldwin 2009) and, therefore, provided information about the activity of each variable in the model, showing how important each variable was in explaining the species distribution and how much unique information each variable contained. This approach can also present highly correlated variables, thereby allowing us to determine if the rate of contribution values are constrained due to these multicollinear variables (Phillips 2017). Nine of the 19 original bioclimatic variables, vegetation type, and percent of tree cover were used in the final model: Bio2 (Mean Diurnal Range); Bio4 (Temperature Seasonality); Bio5 (Max temperature warmest month); Bio9 (Mean temperature driest quarter); Bio14 (Precipitation of Driest Month); Bio15 (Precipitation Seasonality); Bio17 (Precipitation driest quarter); Bio18 (Precipitation of Warmest Quarter) and Bio19 (Precipitation coldest quarter). The evaluation and performance of species distribution model was tested using the threshold-independent receiver operation characteristic (ROC) analysis. The area under the ROC curve (AUC) ranged from 0 to 1. After tenfold cross validation we used the model test of the AUC scores (Elith et al. 2006, Phillips et al. 2006). AUC values greater than 0.9 are considered very good; AUC between 0.7 and 0.9 are good; and AUC values less than 0.7 are uninformative (Swets 1988). All the analyses were performed in the R platform vs. 3.3.2 (R Core Team 2014), MaxEnt software v. 3.4.1 (Phillips et al. 2006) and implemented in ArcGIS 10.5 for desktop (Esri 2016).

RESULTS
A total of 35 records were compiled: field collection (1); SpeciesLink database records (17) and literature records (17). Based on these records we found the distribution of *L. notoaktites* in three states of Brazil (Table 1 and Figure 2): São Paulo (16 records), followed by Paraná (12), and Santa Catarina (7). Most of the records are found in the coastal range of these states. Regarding the vegetation type where the records are located we listed six types (Figure 2): Alto Paraná Atlantic Forests (7); Araucaria Moist Forest (7); Grasslands (5); Serra do Mar Coastal Forests (14); and Southern Atlantic Mangroves (2).

The advertisement call of the collected specimen of *L. notoaktites* was composed of a single, harmonic, unpulsed note, with duration of

0.074 – 0.091 s (0.080 ± 0.005 s; Table 2 and Figure 3). The call had ascending frequency modulation, with a dominant frequency of 689 – 1205 Hz (1029 ± 214 Hz; Table 2 and Figure 3). Hence, the advertisement call analyzed confirms the identity of the specimen collected in the PEMSF as *L. notoaktites*. AUC test for the replicate runs of the ENMs generated for *L. notoaktites* (Figure 4) was 0.99 ± 0.004, indicating a very good model. The environmental suitability was explained primarily by Temperature Seasonality (bio4; 34.3%), Precipitation of Warmest Quarter (bio18; 23.8%) and Precipitation of Driest Month (bio14; 13.8%). Altitude, slope, and aspect were discarded due to their poor contribution to the model (< 1%). The highest environmental suitability in the model (> 0.5; represented by the water blue and blue colors in Figure 4) ranged from São Paulo, Paraná, Santa Catarina, and Rio Grande do Sul states, whereas the most probable areas of occurrence were along the coastal range of São Paulo, Paraná, and Santa Catarina states. The lowest values (< 0.3; green color in Figure 4) were in the states of Rio de Janeiro, Minas Gerais, Mato Grosso do Sul, and some inland regions of São Paulo and Paraná states.
Table 1. Known localities for *Leptodactylus notoaktites* (Anura, Leptodactylidae) in southern and southeastern Brazil. Id = Identification number included in Figure 2. States: SP= São Paulo; PR = Paraná; SC= Santa Catarina. Source type: F = Field; LT = Literature; SL = Records from SpeciesLink database.

<table>
<thead>
<tr>
<th>Id</th>
<th>Municipalities (Locality)</th>
<th>State</th>
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<th>Longitude</th>
<th>Source Type</th>
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<td>F</td>
<td>Present study (MZUEL 1575)</td>
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<td>Telêmaco Borba (Fazenda Monte Alegre)</td>
<td>PR</td>
<td>24.2857°S</td>
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<td>LT</td>
<td>Machado (2004); (MZUEL 1333)</td>
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<td>Blumenau (Parque das Nascentes)</td>
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<td>Present study (PUCRS-MCP 1482)</td>
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<td>Conte &amp; Rossa-Feres (2006)</td>
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Table 1. Continued on next page...
Table 1. ...continued

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<td>Crivellari et al. (2014)</td>
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Figure 3. Audiospectrogram and oscilogram of the advertisement call of *Leptodactylus notoaktites* (Anura, Leptodactylidae) recorded from a single male at Parque Estadual Mata São Francisco, Paraná state, Brazil. (air temperature 26°C; snout-vent length = 49.83 mm).
DISCUSSION

Regarding the geographic distribution of *L. notoaktites* (see Figure 2), our record in the PEMSF, Paraná state, considerably expanded the distribution of the species to the north of the state, and represents a new record for the Alto Paraná Atlantic Forests, an Atlantic Rain Forest vegetation type. The Atlantic Rain Forest was one of the largest rainforests of the Americas with high environmental heterogeneity (Ribeiro et al. 2009), being considered a hotspot for conservation (Myers et al. 2000). The PEMSF is located approximately 258 km in a straight-line from the type locality of the species in the municipality of Iporanga, São Paulo state, which consists of Serra do Mar Coastal Forests, and is about 140 km in a straight-line from the nearest record in the Telêmaco Borba municipality, located in the Araucaria Moist Forest (Machado 2004).

Other species similar to *L. notoaktites*, which include species from the *L. mystaceus* complex (*L. cupreus, L. didymus, L. elenae, L. mystaceus, and L. spixi*), have distinct geographical distributions. *Leptodactylus mystaceus*, which occurs in Amazonia, and Minas Gerais and São Paulo states in mesic enclaves of northeastern Brazil, as well as northern portions of the Atlantic Rain Forests of Brazil (de Sá et al. 2014), is the only species of the *L. mystaceus* complex that occurs in the nearby region of our new record of *L. notoaktites* in the PEMS (see Affonso et al. 2011). Nevertheless, when comparing the morphology of both species it is clear that only individuals of *L. notoaktites* with a light mid-dorsal stripe also have a pair of dorsal folds, while *L. mystaceus* lacks both a pair of dorsal folds and a light mid-dorsal stripe (de Sá et al. 2014). Besides, there are several acoustic differences between the species. For instance, the advertisement call of *L. mystaceus* presents no harmonic structure, while the advertisement call of *L. notoaktites* is harmonically structured (de Sá et al. 2014, present study). Therefore, bioacoustic studies can be useful for understanding ecological processes under climate change (Møller 2010) or anthropic actions in fragile systems (deforestation and fragmentation of habitats) (Tucker et al. 2014), fostering new perspectives in ecology and conservation fields described as Ecoacoustics (Sueur & Farina 2015).

Table 2. Acoustic parameter comparisons of the advertisement call for the *Leptodactylus mystaceus* (Anura, Leptodactylidae) species complex. *Species with frequency modulations in the beginning/end of call.

<table>
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<tr>
<th>Species</th>
<th>Dominant Frequency (kHz)</th>
<th>Call Duration (s)</th>
<th>Pulses</th>
<th>Call rate</th>
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<tbody>
<tr>
<td><em>L. notoaktites</em></td>
<td>689–1,205 Hz/1,550–1,722 Hz*</td>
<td>0.074–0.091 s</td>
<td>No</td>
<td>0.6/s</td>
<td>Yes</td>
<td>Present study</td>
</tr>
<tr>
<td><em>L. cupreus</em></td>
<td>2,800–3,058 Hz</td>
<td>0.16 s</td>
<td>No</td>
<td>–</td>
<td>Yes</td>
<td>Caramaschi et al. (2008)</td>
</tr>
<tr>
<td><em>L. didymus</em></td>
<td>510–1,510 Hz</td>
<td>0.09–0.32 s</td>
<td>Yes</td>
<td>1.4–3.1/s</td>
<td>Yes</td>
<td>Heyer et al. (1996), Köhler &amp; Lötters (2002)</td>
</tr>
<tr>
<td><em>L. elenae</em></td>
<td>700–870 Hz/1,370–1,500 Hz*</td>
<td>–</td>
<td>No</td>
<td>64–120/min</td>
<td>Yes/No</td>
<td>Barrio (1965), Heyer &amp; Heyer (2002)</td>
</tr>
<tr>
<td><em>L. mystaceus</em></td>
<td>700–1,400 Hz</td>
<td>0.2 s</td>
<td>Yes</td>
<td>1.8/s</td>
<td>No</td>
<td>Heyer (1978), Heyer et al. (1996)</td>
</tr>
<tr>
<td><em>L. spixi</em></td>
<td>1,500–1,722/1,981–2,067 Hz*</td>
<td>0.12 ± 10 s</td>
<td>No</td>
<td>80–97/min</td>
<td>Yes</td>
<td>Bilate et al. (2006)</td>
</tr>
</tbody>
</table>

Most of the records of *L. notoaktites* are along the coastal region of Brazil, mainly in the Serra do Mar Coastal Forests (e.g., Reserva Natural Salto Morato, municipality of Guaraqueçaba, Paraná state; Parque Estadual Intervales, municipalities of Ribeirão Grande, Guapiara, Sete Barras, Eldorado, and Iporanga, São Paulo state) and Araucaria Moist Forest (e.g., São José dos Pinhais, municipality of Piraí do Sul, Paraná state) (Table 1 and Figure 2). The other records are in the Alto Paraná Atlantic Forest (e.g., Parque Estadual Mata São Francisco, municipality of Rio Claro, Paraná.
state) and Grasslands (in the phytogeographic unit of Campos Gerais, municipalities of Ponta Grossa, Carambeí, Castro, Paraná state), where there are ecotone with Grasslands and Araucaria Moist Forest (Table 1 and Figure 2). Some individuals were recorded in Southern Atlantic Mangroves (Estação Ecológica da Juréia – Itatins, municipality of Iguape; Parque Estadual Ilha do Cardoso, municipality of Cananéia; and Reserva Natural Rio da Cachoeira, municipality of Antonina, São Paulo state). This species also occurs in ecotones with Alto Paraná Atlantic Forests and Cerrado domain vegetation (e.g., municipality of Corumbataí, São Paulo state) and perhaps may even occur in the Cerrado, since the model predicted suitable environments there (see Figure 4).

Figure 4. Predicted distribution of *Leptodactylus notoakitites* (Anura, Leptodactylidae) based on environmental suitability. Occurrence localities can be found in Table 1 and Figure 2.

Records of *L. notoakitites* outside of its occurrence region predicted by ENM may be the result of the lack of field efforts in some regions. This gap in records could limit more robust predictions by the model. This may occur because the geographical range of any species is based on several complex interactions between intrinsic factors of the species (e.g., life history, dispersal abilities, environmental requirements) and extrinsic factors (e.g., variations in space and time), which limit the known distributions (Brown *et al.* 1996). However, many other factors could be affecting their distribution, such as species colonizing regions outside of their known occurrence and breeding in areas that are significantly different from their former pristine breeding habitats as result of deforestation (Rubbo & Kiesecker 2005). Another possibility is that the species has historically occurred in these environments (in another age) and may now be isolated in refugia (Reside *et al.* 2014). Refugia are habitats where populations of species can retreat to, persist in, and even expand their geographical range over ecological and evolutionary time scales.
of millennia (Keppel et al. 2012).

Herein we show that the distribution of \textit{L. notoaktites} is not restricted to the Brazilian Coast and that it can also occur in inland forests of the Atlantic Rain Forest. Nevertheless, this species still does not have a large distribution (\textit{e.g.}, \textit{L. mystaceus}), which reinforces the need to maintain suitable habitats for this species for conservation and management actions. For biological conservation, ENMs have been mainly applied to discover biodiversity, study species invasion, conservation efforts, and climate changes effects (Rangel & Loyola 2012). Thus, studies that model species distributions along ecosystems, especially for those species with restricted distribution, can serve as a tool for decision-making and indicate priority sites to be preserved. These regions must have suitable conditions for the survival/persistence of rare and/or endangered species or even species with restricted distribution.

Furthermore, we highlight that the gaps in field efforts in certain regions, as in the northern Paraná state, should be considered to estimate the range of \textit{L. notoaktites}. As found for this species, it is possible that several anuran species with known ranges restricted to the Serra do Mar Coastal Forests and Southern Atlantic Mangroves of Brazil could actually have larger distributions. In fact, the anurans diversity in Alto Paraná Atlantic Forests and Araucaria Moist Forests may be higher than expected.

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