

ORIGINAL ARTICLE

Filling gaps on the distribution of Amazonian bats, new records of four poorly sampled species: *Trinycteris nicefori* Sanborn, 1949, *Lionycteris spurrelli* Thomas, 1913, *Macrophyllum macrophyllum* (Schinz, 1821), and *Dasypterus ega* (Gervais, 1856)

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ABSTRACT

Bats are the second most diverse mammalian order worldwide. In the Neotropics, Brazil has one of the greatest bat richness, but species records are still very heterogeneously distributed, especially in Amazonia. Although this biome harbours 76% of Brazil's bat fauna, basic knowledge on the distribution of Amazonian bats is far from well known, with intensive surveys at only a few sites and many areas without any records. The reduction of those gaps will improve the understanding of the bats' distribution, with implications for management and conservation. Here we review the distribution of *Trinycteris nicefori*, *Lionycteris spurrelli*, *Macrophyllum macrophyllum*, and *Dasypterus ega* in the Brazilian Amazonia, presenting several new records for the region based both on literature review and on our own unpublished data. Our study contributes to the knowledge of the geographic distribution of these species, and the new records presented here help fill a gap in the distribution of bats in the western Brazilian Amazonia.

INTRODUCTION

Chiroptera is the second most speciose mammal order worldwide (Simmons 2005, Solari et al. 2019), with more than 1,400 recognized species (Solari et al. 2019, Simmons & Cirranello 2020). In the Neotropical region, Brazil has one of the greatest richness of bat species, with 181 species (Garbino et al. 2020).

However, the geographic distribution patterns of many bat species in the Brazilian territory still have many gaps (Bernard et al. 2011a, 2011b, Aguiar et al. 2020, Delgado-Jaramillo et al. 2020). This lack of knowledge of biogeographic patterns is known as the Wallacean deficit (Lomolino 2004, Whittaker et al. 2005, Bini et al. 2006), and it can be an impediment to effective conservation strategies and management (Meyer et al. 2005, Newbold 2010, Hortal et al. 2015).

The Wallacean deficit is even more problematic for most bat species in Amazonia (Bernard et al. 2011b), which harbour 76% of the Brazilian bat species (Delgado-Jaramillo et al. 2020). Though, basic knowledge on the distribution of Amazonian bats is far from well known, with intensive samplings conducted at only a few sites while many areas remain underexplored (Bernard et al. 2011b). Fragmented records of bat assemblages in Amazonia show that large gaps still need to be filled, including northern and southern portions of the Amazonas state, distant from the state capital (Manaus) (Bernard et al. 2011b).

The reduction of those gaps will improve the understanding of bat species' distribution and may have implications for environmental management and conservation strategies. With nearly 24% of its territory studied (Bernard et al. 2011b), Amazonia is considered one of the priority areas for bat surveys in Brazil (Bernard et al. 2011a, 2011b, Aguiar et al. 2020). Therefore, in this study,

we review the distribution of *Trinycteris nicefori* Sanborn, 1949, *Lionycteris spurrelli* Thomas, 1913, *Dasypterus ega* (Gervais, 1856), and *Macrophyllum macrophyllum* (Schinz, 1821) – species have a wide distribution, but rarely captured – in the Brazilian Amazonia, providing new records of these species for the state of Amazonas.

MATERIALS AND METHODS

Focal species

The Niceforo's big-eared bat (*T. nicefori*) is a gleaning insectivore of the Phyllostomidae family, subfamily Glyphonycterinae (Kalko et al. 1996, Simmons & Voss 1998, Williams & Genoways 2008, Baker et al. 2016). The known geographic distribution of *T. nicefori* ranges from southern Mexico, Belize, Costa Rica, Nicaragua, Panama, Guatemala, Trinidad and Tobago, Colombia, Venezuela, Guyana, French Guiana, Suriname, Peru, Ecuador, Bolivia, and Brazil (Williams & Genoways 2008, Pérez & López 2012, Rocha et al. 2013, Vásquez & Durán 2017, Lima et al. 2018). In Brazil, *Trinycteris nicefori* has been recorded in Amazonia and other biomes, such as Cerrado and the Atlantic Forest (see references in Table 1 in Supplementary Material).

The chestnut long-tongued bat (*L. spurrelli*) is a gleaning nectarivore of the Phyllostomidae family, subfamily Lonchophyllinae. It is widely distributed in the Neotropics occurring in Panama, Colombia, French Guiana, Guyana, Surinam, Peru, Venezuela, and Brazil (Handley 1967, Hall 1981, Griffiths & Gardner 2008). In Brazil, *L. spurrelli* was recorded in Amazonia, Cerrado, transitional areas of Cerrado and Caatinga, the Atlantic Forest, and Caatinga (see references in Table 3 in Supplementary Material).

The Wied's long-legged bat (*M. macrophyllum*) is a gleaning insectivore of the Phyllostomidae family, subfamily Phyllostominae, that hunts over water (Harrison & Pendleton 1974, Harrison 1975, Gardner 1977, Meyer et al. 2005). This species is distributed in Mexico, Costa Rica, Panama, Honduras, El Salvador, Nicaragua, Guatemala, Belize, and in all countries of South America (Ruschi 1952, Harrison 1975, Williams & Genoways 2008, Rodriguez & Pineda 2015, Díaz et al. 2016). In Brazil, there are records of this species in Amazonia, the Atlantic Forest, Cerrado, and Caatinga (see references in Table 3 in Supplementary Material).

For southern the yellow bat (*D. ega*) we follow the nomenclature proposed by Baird et al. (2015), who found a deep divergence between lasiurine bats using a molecular approach. Based on this, those authors proposed dividing the *Lasiurus* genus into three (*Lasiurus*, *Aeorestes* and *Dasypterus*), although debate on this taxonomic interpretation is ongoing (Ziegler et al. 2016, Novaes et al. 2018). *Dasypterus ega* is an aerial insectivore bat of the Vespertilionidae family (Kurta & Lehr 1995), subfamily Vespertilioninae. This species is widely distributed and occurs from southwestern United States, Mexico, Belize, Costa Rica, El Salvador, Guatemala, Honduras, Panama, Trinidad and Tobago, and in most countries in South America, except for Chile and French Guiana (Kurta & Lehr 1995, Gardner & Handley 2008, Baird et al. 2015, Barquez & Diaz 2016). In Brazil, specimens of *D. ega* have records in

Amazonia, Atlantic Forest, Cerrado, Pantanal, Pampas, and Caatinga (see Table 4 in Supplementary Material).

Study site and bat surveys

We captured bats in three Central Amazonian protected areas (Amanã Sustainable Development Reserve, Baixo Juruá Extractive Reserve, and Juami-Japurá Ecological Station) and also in the municipality of Tefé, in Amazonas state. The regional climate is categorized as Af, with mean annual precipitation between 2,800 to 3,100 mm, and an annual mean temperature of 26°C (Alvares et al. 2013).

Bats were captured with 12 × 3 m mist nets set at ground level and located on open trails. In each sampling night, we used 20 nets, which were opened at 5pm for eight hours and checked every 20 minutes. Additionally, we performed a single active search under a floating house (i.e., traditional houses in the region, where the underground are commonly used as roosting sites by bats) at Juami-Japurá Ecological Station. Concomitant to this surveys, in the same protected area, we also captured bats on a mist net set above water level. It was open only a single day during the same time. We also collected a specimen (*D. ega*) that was found dead by a local in his home at the Amanã Sustainable Development Reserve.

For each individual we recorded conventional morphometric data (forearm, head-body length, body length, forearm length, tibia length, ear length, and tragus length), sex and reproductive status. Each bat was marked by cutting a tuft of fur from the shoulder region to avoid measuring recaptured individuals. Each captured bat was placed in individual cotton bags. We conducted fieldwork under the research permits granted by the Chico Mendes Institute for Conservation and Biodiversity (Biodiversity Authorization and Information System n° 26.162-1 and 42.111-3). Specimens of all species were collected following the protocols defined by the American Society of Mammalogy (Sikes et al. 2011, 2016).

Voucher specimens were fixed in 4% formalin and later preserved in 70% ethanol and are currently deposited in the Mammal Collection of the Mamirauá Institute for Sustainable Development. We took measurements of the skull with digital callipers to the nearest 0.01 mm, using selected dimensions based on Velazco (2005), as follows: greatest length of skull, condyloincisive length, mastoid breadth, zygomatic breadth, braincase breadth, postorbital breadth, palatal length, maxillary tooththrow length, dentary length, mandibular tooththrow length (See Supplementary Information).

Literature search

In addition, we also searched for these species records in scientific databases SpeciesLink (<http://www.splink.org.br>), VertNet (<http://www.vertnet.org>), and the Global Biodiversity Information Facility (<http://www.gbif.org>). We also reviewed known records for the four focal species in Brazil by searching the scientific literature in Web of Science and Google Scholar using keywords *T. nicefori* (*Micronycteris nicefori*), *L. spurrelli*, *M. macrophyllum*, and *D. ega* (*Lasiurus ega*).

In order to filter the records in terms of quality and reliability, we only considered as valid those records that had geographic coordinates and vouchers deposited in a zoological collection. For the species that are not easily identifiable (i.e. *L. spurrelli* and *T. nicefori*), we considered those records when there was at least one bat expert in the list of authors of the scientific publication reporting the identification of the voucher. The same parameters apply to the species *M. macrophyllum* and *D. ega* in terms of coordinates and vouchers. However, because these species are easy to identify, we assumed that all of these species identifications were correct.

For *T. nicefori* and *L. spurrelli* we also confirmed the identification through molecular, as we already had genetic sequences for these species, which were part of another study. Muscle tissue of *T. nicefori* and *L. spurrelli* were amplified through PCRs and sent to Sanger sequencing for the mitochondrial cytochrome c oxidase subunit 1 gene (COI) using the primers LCO1490 (5'GGTCAACAAATCATAAAGATATTGG3') and HCO2198 (5'TAAACTTCAGGGTGACCAAAAAATCA3') (Folmer et al. 1994). Our protocols for DNA extraction, amplification and sequencing followed Carnieli et al. (2016). The sequences were aligned and edited in BIOEDIT 7.2.5 (Hall 1999) and were compared to previously published sequences on BOLD Systems v4 platform at <http://barcodinglife.org> to confirm the identification of the specimens information deposited in GenBank.

Diagnostic characters of the species

Bats were identified according to Gardner & Handley (2008) and Díaz et al. (2016). Nomenclature followed Simmons (2005) and to Gardner & Handley (2008) including modifications proposed by Baird et al. (2015).

Based on external characters, *T. nicefori* may be confused with individuals of the genus *Carollia* Gray, 1838. However, *T. nicefori* does not show central papilla-like protuberances in the lower lip, such as in *Carollia* (Charles-Dominique et al. 2001). *Trinycteris nicefori* includes small-sized specimens with body weight of 7–11 g, an adult head-body length of 51–58 mm, forearm length of 35–41 mm, and greatest length of skull 20.7–22.0 mm (Simmons & Voss 1998, Williams & Genoways 2008). A skin band on the top of the head connecting the ears is absent (Williams & Genoways 2008). Four banded dorsal pelage (a pale basal band is narrow and inconspicuous), and a pale median dorsal stripe is usually evident on the lower back (Williams & Genoways 2008). There are two different pelage colour phases described by Sanborn (1949): the grey phase, with grey-brown dorsal pelage, and the red phase, where the pelage is bright orange-brown, contrasting sharply with the dark brown wing membranes (Simmons & Voss 1998, Williams & Genoways 2008).

Lionycteris spurrelli is a small bat, with a forearm length between 33–37.5 mm, an adult head-body length of 53–60 mm, and a weight ranging from 7 to 11 g (Woodman & Timm 2006, Griffiths & Gardner 2008). This species has a dark brown pelage, and a pale basal band is not present on all hairs; thus, the pelage appears to be basally darker than

at the tips (Woodman & Timm 2006, Griffiths & Gardner 2008). The ventral pelage is lighter than the dorsal one, and the medial portions of the uropatagium are conspicuously furred (Woodman & Timm 2006, Griffiths & Gardner 2008). *L. spurrelli* has a narrow muzzle, elongated, the rostrum is shorter than the braincase, and the palatal margin of the mesopterygoid fossa is comparatively deep and U-shaped (Griffiths & Gardner 2008).

Macrophyllum macrophyllum is considered the smallest phyllostomine, with a head-body length of 40–53 mm, a forearm length of 32.9–40 mm, a greatest length of skull ranging from 16–18 mm, and a body mass from 7 to 11 g (Harrison 1975, Williams & Genoways 2008). This species can be easily diagnosed by the presence of longitudinal rows of dermal projections in the ventral surface of the uropatagium (Harrison 1975).

Dasypterus ega is a small bat, with an adult head-body length between 106–132 mm, a forearm length ranging from 40,5 to 55,1 mm, a weight that oscillates from 13 to 20 g, with a colouration ranging from yellowish-buff to reddish-olive. The dorsal face of the uropatagium is covered with hair up to its half (Acosta & Lara 1950, Handley 1960, Barquez et al. 1993, Kurta & Lehr 1995, Miranda et al. 2011, Díaz et al. 2016). The skull is characterized by a sagittal crest, a single upper premolar, well-developed tympanic bullae, trifold lower incisors, and arranged almost perpendicularly to the maxillae (Handley 1960, Baker et al. 1971, Kurta & Lehr 1995).

RESULTS

Trinycteris nicefori

We found 53 records of this species (Table 1 in Supplementary Material; Fig. 2), although in the last publication on the records of this species in Brazil there were only 28 records (Silva et al. 2020a). Our search found almost twice as many records for the Amazon, especially for the state of Amazonas, when compared to this publication. With our three additional records, there are 56 occurrence records in Brazil for this species (Fig. 2).

An adult female of *T. nicefori* (IDSM03661) was captured on 04 May 2011, in a terra firme primary forest of the rural area of Tefé (-3.444444S; -64.698214W) (Fig. 1; Table 1 in Supplementary Material). In this location, together with the specimen of *T. nicefori*, the following species were recorded: *Dermanura cinerea* Gervais, 1856, *Artibeus lituratus* (Olfers, 1818), *Carollia benkeithi* (Solari & Baker, 2006) *Carollia perspicillata* (Linnaeus, 1758), *Mesophylla macconnelli* (Thomas, 1901), *Phyllostomus hastatus* (Pallas, 1767), *Platyrrhinus fusciventris* Velazco et al. 2010, *Rhinophylla pumilio* W. Peters, 1865, *Tonatia maresi* Williams et al., 1995, *Uroderma bilobatum* Peters, 1866, and *Vampyriscus bidens* (Dobson, 1878). Another adult female of *T. nicefori* (IDSM01367) was captured in a terra firme primary forest of the Juami-Japurá Ecological Estation (-1.692917S; -67.916167W) on 02 June 2017. In this location, together with the specimen of *T. nicefori*, the following species were captured: *Micronycteris microtis* Miller, 1898, *Micronycteris hirsuta* (W. Peters, 1869), and *T. maresi*. A third adult male *T.*

nicefori (IDSM01497) was captured on 18 July 2018 in a terra firme primary forest, in the Baixo Juruá Extractive Reserve (-3.822861S; -66.080111W). The COI gene barcoding sequences from IDSM01497 specimen supported the species-level identifications (GenBank accession MT900627). Together with the specimens of *T. nicefori*, the following species were recorded: *C. perspicillata*, *Saccopteryx bilineata* (Temminck, 1838), *Artibeus planirostris* (Spix, 1823), and *Phyllostomus elongatus* (É. Geoffroy, 1810).



Fig. 1 - Specimen of *T. nicefori* collected in Tefé, state of Amazonas. Ventral (A), and dorsal (B) views, (C) lateral view of skull, (D) protruding upper incisors, (E) lateral view of the mandible. Scale bar: 20 mm.

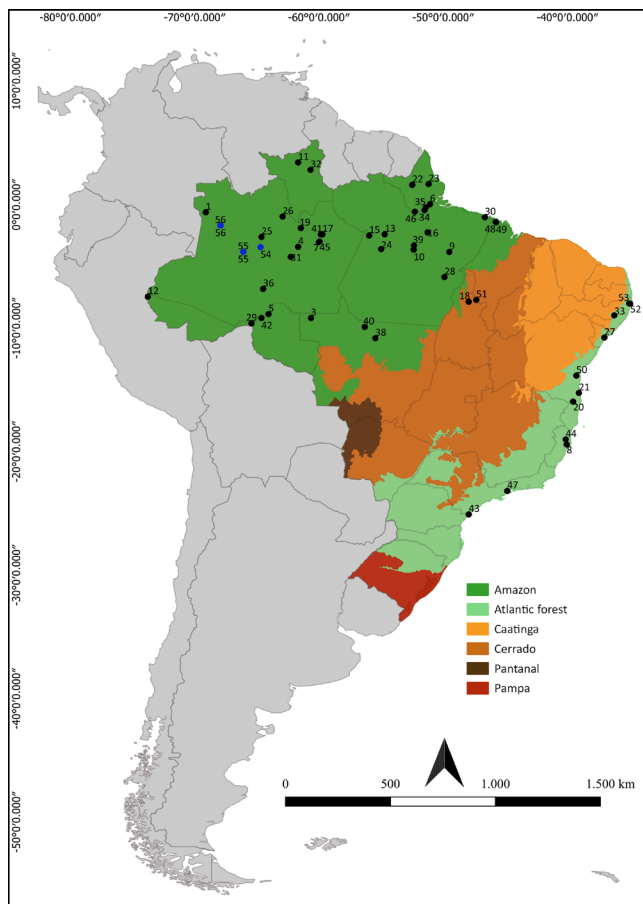


Fig. 2 - Geographic distribution of *T. nicefori* in Brazil. Numbers indicate localities are listed in Table 1 in Supplementary Material. The black circles represent known records, the blue circles represent new records.

The specimen captured in Tefé presented the typical dorsal stripe. However, the other captured specimens did not present it, suggesting a substantial interindividual variation (Starrett 1976, Simmons & Voss 1998). All specimens also showed only the “grey phase” (Sanborn 1949, Starrett 1976).

Lionycteris spurrelli

We found 32 records of this species (Table 2 in Supplementary Material; Fig. 4). With our new record, we have compiled 33 records of occurrence in Brazil for this species.

In the Baixo Juruá Extractive Reserve (-3.758694S; -66.068306W), one adult female of *L. spurrelli* (IDSM01519) was captured on 23 July 2018 in a terra firme secondary forest (Fig. 3). The COI gene barcoding sequences from this specimen supported the species-level identifications (GenBank accession MT900626). The other species recorded were: *A. lituratus*, *A. planirostris*, *A. obscurus* (Schinz, 1821), *C. benkeithi*, *C. perspicillata*, *Carollia brevicauda* (Schinz, 1821), *Chrotopterus auritus* (Peters, 1856), *Rhinophylla fischeriae* Carter, 1966, and *V. bidens*.

Macrophyllum macrophyllum

We found 52 records of this species (Table 3 in Supplementary Material; Fig. 6). However, in a recent publication on the distribution of this species, only 20 records were included, of which seven were for the Amazon (Feijó et al. 2015). Although there are other records in the literature for *M. macrophyllum* (Mok & Lacey 1980, Marques 1985b, Reis & Peracchi 1987, Barnett et al. 2006, Moratelli et al. 2010) there were only six records for Amazonia in the most recent compilation (Feijó et al. 2015). With our two additional records and our search, we have compiled 54 occurrence records in Brazil for this species (Table 3 in Supplementary Material, Fig. 6).



Fig. 3 - Skull and mandible of *Lionycteris spurrelli* specimen captured at Tefé: (A) lateral view of skull, (B) protruding upper incisors (C) dorsal view of the mandible. Scale bar: 20 mm.

Three adult males of *M. macrophyllum* (IDSM 01347-01349) (Fig. 5) were collected at Juami-Japurá Ecological Station (-1.773306S; -67.59975W) on 31 May 2017. At the same locality, we also recorded *P. elongatus* (É. Geoffroy, 1810). The specimens were roosting under a floating house

and were captured in an active search during the daytime. On 13 June 2017, two adult males of *M. macrophyllum* (IDSM 01443-01444) were collected in a mist net set above water next to the floating house. In the same location, we have also recorded: *Molossus molossus* (Pallas, 1766), *Noctilio leporinus* (Linnaeus, 1758), and *Myotis* sp. Another adult male (IDSM02768) was found dead on the campus of Instituto Mamirauá in Tefé (-3.353703S; -64.733094W) on 14 July 2009.

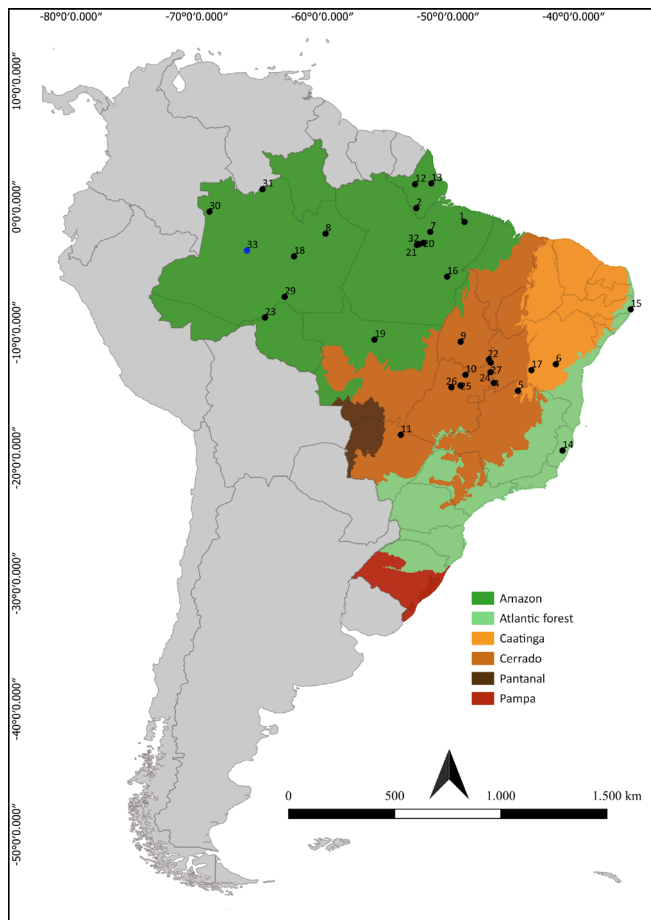


Fig. 4 - Geographic distribution of *Lionycteris spurrelli* in Brazil. Numbers indicate localities are listed in Table 2 in Supplementary Material. The black circles represent known records, and the blue circle represents the new record.

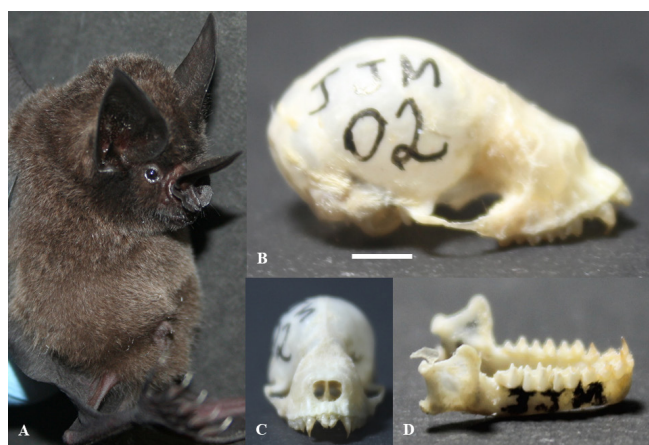


Fig. 5 - Specimen of *Macrophyllum macrophyllum* collected in Juami-Japurá Ecological Station (A), state of Amazonas (Brazil), lateral view of skull (B), protruding upper incisors (C), lateral view of the mandible(D). Scale bar: 20 mm.

Dasypterus ega

We found 151 records of this species in Brazil (Table 4 in Supplementary Mateiral; Fig. 8) and 14 occurrences for the Amazon. The most recent publication on the records of this species in Brazil reported 96 records for Brazil, with only three records for the Amazon (Leal & Gomes-Silva 2015).

One adult female of *D. ega* (IDSM1217) (Fig. 7) was captured in the Amanã Sustainable Development Reserve, in the Bom Jesus do Baré community (-2.481508S; -64.711647W), next to a terra firme secondary forest on 17 November 2014. Although there are records by identifying echolocation calls in the same location (Marques et al. 2016), we consider this record important because this species has few vouchers in the Amazon.

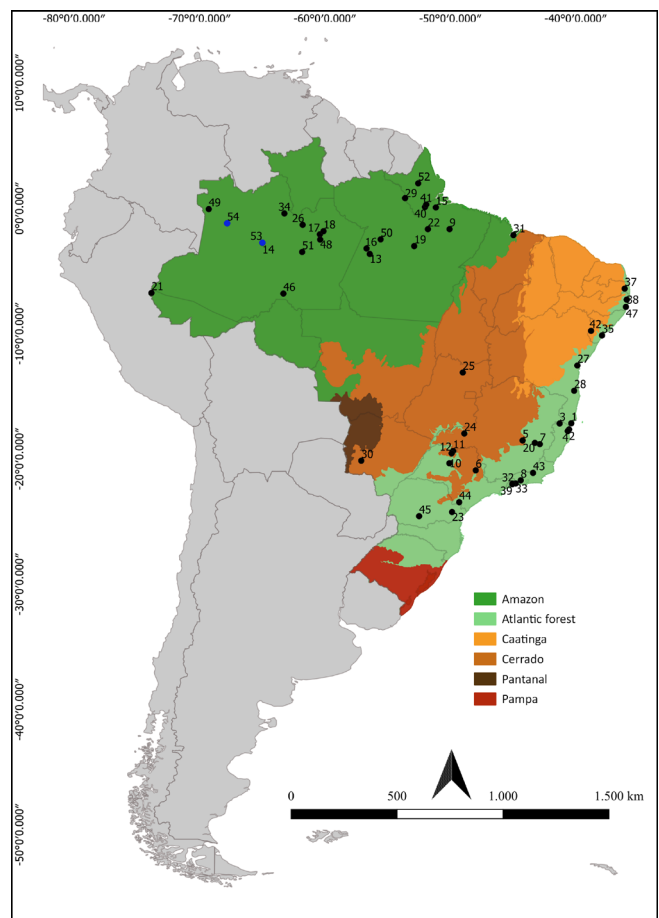


Fig. 6 - Geographic distribution of *Macrophyllum macrophyllum* in Brazil. Numbers indicate localities listed in Table 3 in Supplementary Material. The black circles represent known records, the blue circles represent new records.



Fig. 7 - *Dasypterus ega* specimen captured at Amanã Sustainable Development Reserve: (A) lateral view of skull, (B) protruding upper incisors (C) dorsal view of the mandible. Scale bar: 10 mm.

DISCUSSION

Here we show that four poorly sampled bat species have more records in Brazil than previously suggested and also that recent publications focused on those species missed several available records. Our records and literature search for *L. spurrelli*, *T. nicefori*, *M. macrophyllum*, and *D. ega* allowed us to update the occurrence map of these species, as well to fill the distribution gaps of bats in Amazonia.

Despite having many records in the Amazon, *T. nicefori* presents gaps throughout its distribution, probably associated with its low capture rate in bat inventories (e.g., Rocha et al. 2017). In fact, in Tefé, where we conduct annual surveys, we have captured only one individual so far. Our search in databases added twice as many records of this species than what has been compiled in recent publications for its distribution (Rocha et al. 2013, Lima et al. 2018, Silva et al. 2020a). In addition, several publications on this species have been ignored in the most recent compilations. For example, Silva et al. (2020a) placed only 27 records for Brazil. These authors ignored an extensive list of studies for the Amazon (e.g., Handley 1967, Mok et al. 1982, Robinson 1998, Nunes et al. 2007, Pereira et al. 2009, Tavares et al. 2012, 2017, Costa et al. 2013, 2016, Bobrowiec et al. 2014, Castro & Michalski 2015, Silva & Bobrowiec 2015, Silva et al. 2015, Martins et al. 2017, Rocha et al. 2017, Capaverde et al. 2018, Lima et al. 2018, Carvalho et al. 2019). This was also found for the other species.

For *L. spurrelli*, ours is the sixth record for the state of Amazonas. Even in places intensively sampled in the state, such as the Biological Dynamics of Forest Fragments Project, where bat surveys have been taking place since 1996, the species has been recorded only twice (Sampaio et al. 2003, Rocha et al. 2017, Farneda et al. 2018, Silva et al. 2020b). The new record provided here extends its occurrence to more than 400 km west from the nearest record in the state, which is Uauaçú Lake (Bobrowiec et al. 2014).

For *M. macrophyllum*, a species that hunts over water, our active search under a floating house, as well as the fact that we placed mist nets above water, turned to be crucial to detect it. This indicates that the use of multiple sampling methods increases the efficiency of bat inventories (Flaquer et al. 2007).

Our compilations for *M. macrophyllum* and *D. ega* indicated significant increases in records for both species, filling large gaps in the distribution of these species in the Amazon. Although widely distributed, records for those species are scattered, probably due to the low capture rates in studies based on the sole use of ground-level mist nets (Kalko et al. 1996, Voss & Emmons 1996).

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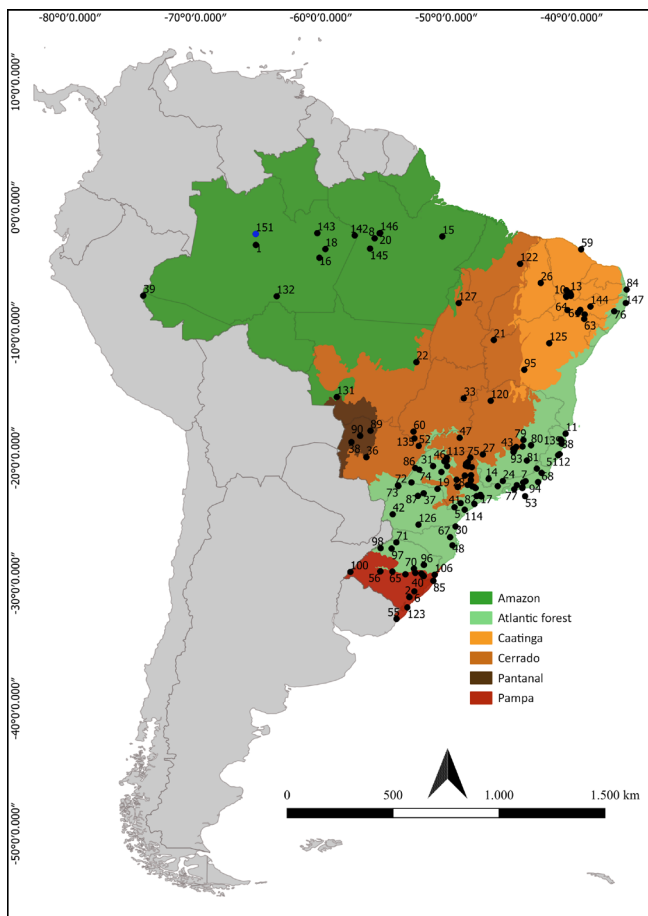


Fig. 8 - Geographic distribution of *Dasypterus ega* in Brazil. Numbers indicate localities listed in Table 4 in Supplementary Material. The black circles represent known records, and the blue circle represents the new record.

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