



## Potential seed dispersers: a new facet of the ecological role of *Boa constrictor constrictor* Linnaeus 1758

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**Abstract:** The boa (*Boa constrictor*) is considered a top predator and its diet includes a wide variety of birds, mammals, and other reptiles, all related directly to their availability in the environment inhabited by the snake. Seven boas were found roadkilled on highways adjacent to conservation units in the semi-arid region of Rio Grande do Norte state, in northeastern Brazil. Their digestive tract was analyzed to identify food items and classify them according to their orientation in the tract. Among the food items found, the white-eared opossum (*Didelphis albiventris*) and the black-and-white tegu (*Salvator merianae*) were ingested head-first, while teeth of a punaré (*Thrichomys laurentius*) and a Spix's yellow-toothed cavy (*Galea spixii*) and hairs of an unidentified rodent were found in the intestinal tract. In addition, two novel items were identified: the plain-breasted ground-dove (*Columbina minuta*), which were ingested tail-first, and carnauba palm seeds (*Copernicia prunifera*). The orientation of the prey (head-first or tail-first) followed what was expected for each type of prey. In addition, the presence of carnauba palm seeds indicates that, while being a top predator, the boa may also be a potential disperser of seeds, which would constitute a previously unrecorded ecological role for this species.

**Keywords:** Caatinga, Feeding behavior, Snake, Seed dispersal, Zoochory.

## Potenciais dispersores de sementes: uma nova faceta do papel ecológico de *Boa constrictor constrictor* Linnaeus 1758

**Resumo:** A jiboia (*Boa constrictor*) é considerada um predador de topo e a sua dieta inclui uma grande variedade de aves, mamíferos e outros répteis, todos relacionados diretamente com a sua disponibilidade no ambiente por ela habitado. Sete jiboias foram encontradas atropeladas em estradas no entorno de unidades de conservação da região semiárida do estado do Rio Grande do Norte, no nordeste brasileiro. O seu trato digestivo foi analisado de modo a identificar itens alimentares e a classificá-los de acordo com a sua orientação no trato. De entre os itens alimentares encontrados, o gambá-de-orelha-branca (*Didelphis albiventris*) e o teju (*Salvator merianae*) foram ingeridos no sentido ântero-posterior, enquanto que dentes de punaré (*Thrichomys laurentius*) e de preá (*Galea spixii*) e pelos de um roedor não identificado foram encontrados do trato intestinal. Dois itens novos foram identificados: a rolinha-de-asa-de-canela (*Columbina minuta*), que foi ingerida no sentido pósterio-anterior, e sementes de carnaúba (*Copernicia prunifera*). A orientação dos itens (ântero-posterior ou pósterio-anterior) seguiu o que era esperado para cada tipo de presa. A presença de sementes de carnaúba indica que, além de ser um predador de topo, a jiboia também pode ser um potencial dispersor de sementes, o que constitui um papel ecológico previamente não descrito para esta espécie.

**Palavras-chave:** Caatinga, Comportamento alimentar, Serpente, Dispersão de sementes, Zoocoria.

## Introduction

The boa constrictor (*Boa constrictor* Linnaeus, 1758) is a large, robust snake of the Boidae family Gray, 1825a (Pizzatto et al. 2009, Mesquita et al. 2013) and is widely distributed in the Neotropical region, including South and Central America (Hynková et al. 2009, Card et al. 2016). In Brazil, this species can be commonly found in several biomes such as the Amazon and Atlantic forests (Pizzatto et al. 2009) the Cerrado (Pizzatto et al. 2009) and the Caatinga (Loebmann & Haddad 2010, Marques et al. 2017). Boas are primarily nocturnal, although daylight activity has also been reported, and present terrestrial or semi-arboreal behavior (Strüssmann & Sazima 1993, Martins & Oliveira 1999, Freitas 2003, Pizzatto et al. 2009, Bernarde 2012, Mesquita et al. 2013, Guedes et al. 2014). They are non-venomous snakes with aglyphous dentition and are considered mostly ambush predators, although they can also actively forage (Montgomery & Rand 1978, Greene 1997, Martins & Oliveira 1999, Freitas 2003). Similarly to other members of the Boidae family, boas detect their prey using infrared radiation, visual cues and chemoreception (Buning 1983, Gracheva et al. 2010). Once the prey is captured, it is subdued by constriction (Vanzolini et al. 1980, Scartozzoni & Molina 2004, Bernarde 2012) which causes circulatory arrest followed by death (Boback et al. 2015).

*Boa constrictor* is considered a dietary generalist, typically consuming preys available in its local environment (Pizzatto et al. 2009) such as birds, mammals, and reptiles (Vitt & Vangilder 1983, Martins & Oliveira 1999, Freitas 2003, Quick et al. 2005, Mesquita et al. 2013, Guedes et al. 2014, Marques et al. 2017). Hence, boas usually reside in higher trophic levels (Campbell & Campbell 2001) and they are considered top-predators, since they are generally free of predation, particularly in the adult stage (Sergio et al. 2014). Because of their ecological role as top-predators, boas can exert strong top-down pressure on the food webs associated with trophic cascades (Schmitz et al. 2000), affecting both species abundance and composition. This effect on community structure has also been observed in areas where boas were introduced (Martínez-Morales & Cuarón 1999, Snow et al. 2007).

The present study describes prey items found in the digestive tracts of boas found roadkilled on highways in the vicinity of two federal conservation units in the state of Rio Grande do Norte, northeastern Brazil. These records provide a preliminary analysis of the diet composition of *Boa constrictor*, for this region of the Brazilian Caatinga biome. In addition, a review on the feeding habits of this species for its Brazilian distribution was conducted to provide a general overview on the ecological role of this species, also considering the new data presented.

## Materials and Methods

The *B. c. constrictor* specimens were obtained from highways adjacent to two federal conservation units in the Brazilian state of Rio Grande do Norte: (i) the Açu National Forest (5°03'15.53"S, 37°30'39.85"W, altitude: 123 m) in the municipality of Açu, located in the Vale do Açu microregion (IBGE 1992), and (ii) the Seridó Ecological Station (6°35'15.43"S, 37°15'19.63"W, altitude: 214 m) in the municipality of Serra Negra do Norte, located in the West Seridó microregion (IBGE 1992). Highways were surveyed by motor vehicle, traveling at a speed of 40–60 km/hour. Collected specimens

were preserved in ice-filled coolers, and then taken to the Laboratory of Wildlife Ecology and Conservation at the Federal Rural University of the Semi-Arid in Mossoró, Rio Grande do Norte, Brazil.

In the laboratory, snout-vent length (SVL) and tail length (TL) were measured using a ruler (precision 1 mm) and body mass was determined using a 5000 g Pesola Macro-Line Spring Scale (precision 50 g). Sex of specimens was determined through the examination of the gonads. Food items were weighed using a Shimadzu AUW220 digital bench-top balance (precision 0.0001 g) and then identified to the lowest taxonomic level possible. Those items still largely intact were measured with a caliper (precision 0.05 mm) and their direction of ingestion classified as head-first or tail-first (Sazima 1989, Ruffato et al. 2003), according to the orientation of the prey item in relation to the body of the snake. The intestinal content in an advanced digestive stage was placed in a plastic sieve (1 mm mesh) and washed under running water. Its remaining content was then examined using a stereomicroscope (PHYSIS) with a WF10X wide-angle lens for the identification of hairs, feathers, bones, and teeth to the lowest possible taxonomic level. All material collected was fixed in 70% alcohol and stored in glass containers in the laboratory.

## Results

Of the seven boas obtained from highways adjacent to the two federal conservation units considered, two of them were found in the Açu region and five in the Seridó region (Table 1). Two of the boas (B3 and B5) presented empty stomachs and digestive tracts, while the remaining five presented food items at different states of digestion (Table 2). Specimens B1 and B6 contained food items, in both the stomach and the digestive tract, that could be classified quantitatively and qualitatively. In specimen B1, both the white-eared opossum (*Didelphis albiventris* Lund, 1840) and the black-and-white tegu (*Salvator merianae* Duméril & Bibron, 1839) were ingested head-first. Additionally, its digestive tract also contained two seeds of the carnauba palm tree (*Copernicia prunifera* (Miller) H.E. Moore, 1963; Arecales: Arecaceae). The digestive tract of specimen B6 contained two plain-breasted ground-doves (*Columbina minuta* Linnaeus, 1766), which were both ingested tail-first. Although specimens B2 and B4 had empty stomachs, their digestive tracts did contain hairs and teeth of rodents (order Rodentia). In specimen B2, the material could only be identified to order (Greene 1959), whereas in specimen B4, the teeth could be identified as belonging to a punaré (*Thrichomys laurentius* Thomas, 1904) and a Spix's yellow-toothed cavy (*Galea spixii* Wagler, 1831), based on available identification keys (Neves & Pessôa 2011, D'Elia & Myers 2014, Ubilla & Rinderknecht 2014).

## Discussion

Both the reviewed (Table 2) and the new data here presented confirm that *Boa constrictor constrictor* acts as a top predator in the Caatinga by preying upon higher vertebrate taxa, such as mammals, birds and other reptiles. Some food items identified in this study had already been described for the diet of boas in the Caatinga, including *Galea spixii* (Vitt & Vangilder 1983) and rodents (Mesquita et al. 2013). Moreover, other food items identified were already recorded as part of the diet of Boas in other biomes (Table 1): rodents of the family Echimyidae



**Table 1.** Collection sites, sex, morphometric measurements and diet of the seven *Boa constrictor* specimens retrieved from highways of the semi-arid zone of the Brazilian Northeast. Measurements are given in millimeters (mm) and body mass in grams (g). SVL = Snout-Vent Length; TL = Tail Length; Missing values of mass, SVL and TL indicate measurements that could not be determined due to the advanced stage of decomposition of either the boa (predator found roadkilled) or its food items (digestive decomposition).

Ind.	location	Lat/long.	sex	Mass (g)	SVL (mm)	TL (mm)	Food items		
							identification	TL (mm)	Mass (g)
B1	BR 304, Açu	5°37'06.95" S, 36°53'13.77" W	F	4250	1770	185	<i>Didelphis albiventris</i>	339	123.22
							<i>Salvator merianae</i>	–	–
							<i>Copernicia prunifera</i> (seed)	20.9	1.997
							<i>Copernicia prunifera</i> (seed)	17.5	1.456
B2	RN 118, Açu	5°29'33.01" S, 36°51'16.15" W	F	–	–	91.7	Rodentia: teeth and hairs	–	–
B3	RN 288, Seridó	6°22'59.63" S, 37°19'41.16" W	M	–	572	70	–	–	–
B4	RN 288, Seridó	6°22'03.20" S, 37°24'36.90" W	M	2500	1435	205	<i>Thrichomys laurentius</i>	–	–
							<i>Galea spixii</i>	–	–
B5	RN 288, Seridó	6°23'57.72" S, 37°16'20.42" W	F	1200	1129	189	–	–	–
B6	BR 427, Seridó	6°26'24.55" S, 37°10'58.71" W	F	3000	1510	161	<i>Columbina minuta</i>	137	39.24
							<i>Columbina minuta</i>	–	–
							<i>Galea spixii</i>	–	–
							<i>Thrichomys laurentius</i>	–	–
B7	RN 118, Seridó	6°37'01.37" S, 37° 08'57.80" W	F	318	842	81	Mammal: hairs	–	–

**Table 2.** Known diet of *Boa constrictor*, *B. constrictor amarali* and *B. constrictor constrictor* for their Brazilian distribution, including biome, prey category (Aves, Mammalia and Reptilia/Squamata) and food items.

Species/subspecies	Biome	Locality, State	Prey category	Food items	Reference
<i>B. constrictor</i>	Amazon Forest	Ilha de Germoplasma, PA	Mammalia	<i>Chiropotes satanas utahicki</i> Herskovitz, 1985	Ferrari et al. (2004)
<i>B. constrictor</i>	Amazon Forest	Oriximiná, PA	Reptilia/Squamata	<i>Iguana iguana</i> Linnaeus, 1758	Oliveira et al. (2015)
<i>B. constrictor</i>	Amazon Forest	Espigão do Oeste, RO	Aves	<i>Volatinia jacarina</i> Linnaeus, 1766	Bernarde and Abe (2010)
			Aves	unspecified	
			Mammalia	Rodentia Bowdich, 1821 (undet.)	
			Reptilia/Squamata	<i>Ameiva ameiva</i> Linnaeus, 1758	
<i>B. constrictor</i>	Amazon Forest	Cacaulândia, RO	Aves	<i>Ara severus</i> Linnaeus, 1758	Begotti and Marcos Filho (2012)
<i>B. constrictor</i>	Amazon Forest	Rolim de Moura, RO	Mammalia	<i>Alouatta puruensis</i> Lönnberg, 1941	Quintino and Biccamarques (2013)
<i>B. constrictor</i>	Amazon Forest	Manaus, AM	Aves	unspecified	Martins and Oliveira (1999)
			Mammalia	unspecified	
			Reptilia/Squamata	unspecified	
<i>B. constrictor</i>	Caatinga	Fortaleza, CE	Aves	<i>Troglodytes musculus</i> Naumann, 1823	Gondim et al. (2012)
<i>B. constrictor</i>	Caatinga	Pentecoste, CE	Aves	<i>Passer domesticus</i> Linnaeus, 1758	Mesquita et al. (2013)
			Mammalia	Rodentia (undet.)	
			Reptilia/Squamata	<i>Cnemidophorus ocellifer</i> Spix, 1825	
<i>B. constrictor</i>	Caatinga	Exu, PE	Aves	<i>Tinamus</i> sp. Hermann, 1783	Vitt and Vangilder (1983)
			Mammalia	<i>Galea spixii</i> Wagler, 1831	

Continuation Table 2.

Species/subspecies	Biome	Locality, State	Prey category	Food items	Reference
<i>B. constrictor</i>	Cerrado	Distrito Federal, DF	Mammalia Reptilia/Squamata	Muridae Illiger, 1811 (undet.) <i>Ameiva ameiva</i>	França et al. (2008)
<i>B. constrictor</i>	Cerrado	Campos Belos, GO	Mammalia	<i>Callithrix penicillata</i> É. Geoffroy, 1812	Teixeira et al. (2016)
<i>B. constrictor</i>	Cerrado	Campo Grande, MS	Aves Aves	<i>Turdus rufiventris</i> Vieillot, 1818 <i>Pitangus sulphuratus</i> Linnaeus, 1766	Rocha-Santos et al. (2014)
<i>B. constrictor</i>	Cerrado	Itirapina e Brotas, SP	Aves Mammalia	unspecified <i>Didelphis albiventris</i> Lund, 1840	Sawaya et al. (2008)
<i>B. constrictor</i>	Atlantic Forest	Cachoeiro de Itapemirim, ES	Aves	<i>Furnarius</i> sp. Vieillot, 1816	Giori et al. (2016)
<i>B. constrictor</i>	Atlantic Forest	São Paulo, SP	Aves	<i>Diopsittaca nobilis</i> Linnaeus, 1758	Travaglia-Cardoso et al. (2016)
<i>B. constrictor</i>	Pantanal	Cuiabá, MT	Mammalia	<i>Noctilio albiventris</i> Desmarest, 1818	Esbérard and Vrcibradic (2007)
<i>B. constrictor amarali</i>	Atlantic Forest	Botucatu, SP	Mammalia	Erethizontidae Bonaparte, 1845 (undet.)	Cherubini et al. (2003)
<i>B. constrictor amarali</i>	—	Several unspecified locations	Aves	<i>Gallus gallus</i> Linnaeus, 1758	Pizzatto et al. (2009)
			Aves	<i>Zonotrichia capensis</i> Cabanis & Heine, 1850	
			Aves	Passeriformes (undet.)	
			Aves	unspecified	
			Mammalia	<i>Akodon cursor</i> Winge, 1887	
			Mammalia	<i>Didelphis albiventris</i> Lund, 1840	
			Mammalia	Cricetidae G. Fischer, 1817 (undet.)	
			Mammalia	Echimyidae Gray, 1825b (undet.)	
<i>B. constrictor constrictor</i>	Amazon Forest	Alta Floresta, MT	Mammalia	Rodentia (undet.)	Rocha and Bernarde (2012)
			Reptilia/Squamata	<i>Ameiva ameiva</i>	
			Reptilia/Squamata	<i>Tupinambis teguixin</i> Linnaeus, 1758	
			Aves	unspecified	
			Mammalia	Echimyidae (undet)	
			Mammalia	<i>Didelphis marsupialis</i> Linnaeus, 1758	
			Mammalia	Rodentia (undet.)	
			Mammalia	unspecified	
<i>B. constrictor constrictor</i>	—	Several unspecified locations	Reptilia/Squamata	<i>Ameiva ameiva</i>	Pizzatto et al. (2009)
			Reptilia/Squamata	<i>Tropidurus</i> sp Wied-Neuwied, 1825	
			Reptilia/Squamata	unspecified	
			Reptilia/Squamata	unspecified	

(Pizzatto et al. 2009), represented by *Thrichomys laurentius*; marsupials of the family Didelphidae Gray, 1821 (Sawaya et al. 2008, Pizzatto et al. 2009), represented by *D. albiventris*; and lizards of the family Teiidae (Pizzatto et al. 2009, Bernarde & Abe 2010, Rocha & Bernade 2012, França & Braz 2013, Mesquita et al. 2013), represented by *Salvator merianae*. However, while boas are known to prey on birds, *Columbina minuta* doves (family Columbidae Leach, 1820) had not been previously recorded in the diet of *B. constrictor*.

Food items such as *S. merianae* (this study) and *Iguana iguana* Linnaeus, 1758 (Oliveira et al. 2015) emphasizes the capacity of *Boa constrictor* for the ingestion of relatively large prey, which is a

characteristic of the boids (Sazima & Martins 1990), and reinforces their ecological role of top predators. Furthermore, snakes may ingest their prey head-first, tail-first or sideways, depending on the type of animal being preyed upon (Greene 1976, Rodríguez-Robles & Leal 1993, Rodríguez-Robles et al. 1999). Boas tend to ingest more reactive animals (which are able to respond to attacks with defensive behaviors, such as biting) head-first, as observed by Scartozzoni & Molina (2004). This would be consistent with the head-first orientation of the *D. albiventris* and *S. merianae* prey items recorded in the present study. By contrast, columbiform birds, such as *C. minuta*, are more passive, and much less likely to wound a predator such as the boa. Hence, the tail-first ingestion

position of *C. minuta* observed in this study is consistent with previous observations regarding preys with low relative mass ratios and small diameters when compared to snake head size, albeit for other serpent species (Greene 1976, Rodríguez-Robles & Leal 1993, Rodríguez-Robles et al. 1999).

The two seeds of the carnauba palm tree (*C. prunifera*) found in the digestive tract of a boa (B1, Table 2) raise the question of whether its ingestion was voluntary or accidental. The carnauba palm fruit presents a dark colored epicarp (when it is mature), a fleshy mesocarp (rich in nutrients) and a hard endocarp that protects the seed (Braga 2001). Since dark-colored items heat more and faster than lighter ones, it is possible that carnauba palm fruits retain more heat due to their dark coloration. Even though boas detect their prey's heat using infrared radiation (Buning 1983, Gracheva et al. 2010), they also use visual and Jacobson's organs (chemoreception), which makes the scenario of a boa mistaking these fruits for a potential prey and purposefully ingesting them highly unlikely to occur.

A more likely scenario is that the ingestion of the carnauba palm seeds/fruits by the boa was accidental, occurring during the maneuvering and swallowing of a prey (direct ingestion) or they were already within the stomach and digestive tract of the prey (indirect ingestion). Boas are known to prey on relatively large animals, such as the marsupial *D. albiventris*, which are capable of ingesting and dispersing *C. prunifera* seeds. Cantor et al. (2010) and Cáceres & Lessa (2012) found seeds of a size similar to those of *C. prunifera* in the digestive tracts of *D. albiventris*. In a camera-trap study of two areas of Caatinga (Furna Feia National Park and the TRIPOL trail on the Rafael Fernandes Experimental Farm in the municipality of Mossoró), Torquato (Torquato 2015) recorded the ingestion of *C. prunifera* seeds by: two birds, the white-naped jay (*Cyanocorax cyanopogon* Linnaeus, 1766) and rufous-bellied thrush (*Turdus rufiventris* Vieillot, 1818); a mammal, the crab-eating fox (*Cerdocyon thous* Linnaeus, 1766); and a lizard, the black-and-white tegu (*S. merianae*). While it is unlikely that *C. thous* can constitute part of the boa's diet, *S. merianae* certainly is (Table 2) and the bird species most likely are. Indirect ingestion of seeds/fruits and other plant structures can be relatively common, as the two plain-breasted ground-doves found in the digestive tract of another boa (B6, Table) also contained unknown seeds.

Prior to the present study, there are no published records that describe the ingestion of fruits or seeds by boas, in its Brazilian distribution (Table 1). This lack of records could be the result of no seeds being found on the stomach and digestive tract of boas or simply because they were not considered food items, and thus unreported. However, regardless of how these fruits/seeds are ingested, the boa could have benefited from its nutritional content, in particular if the fruits possess a fleshy mesocarp. Furthermore, seeds may survive the digestive process of these snakes, in particular those which present a fruit with a hard endocarp that protects the seed, such as the carnauba palm (Braga 2001). Even though boas can carry seeds, the effectiveness of dispersal must still be confirmed, including by assessing whether these seeds can survive and become adult plants (Schupp 1993, Schupp et al. 2010). If the effectiveness of seed dispersal is confirmed, these snakes can become seed rescuers and secondary dispersers of different plants, as seen in other species (Reiserer et al. 2018). Hence, the findings of this study not only expand the list of prey known to be exploited by *B. constrictor*, reinforcing its

capacity to adapt to different environments and the availability of prey (Martins & Oliveira 1999, Pizzatto et al. 2009), but also indicate that, in addition to being a top predator that feeds on terrestrial and arboreal vertebrates, the boa may also be a potential disperser of carnauba palm seeds, a previously unrecorded ecological role.

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## Author Contributions

Sofia de O. Cabral: Bibliographical survey; Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation; Contribution to manuscript preparation.

Itainara da S. Freitas: Contribution to data collection.

Viviane Morlanes: Contribution to data collection; Bibliographical survey.

Marco Katzenberger: Contribution to data analysis and interpretation; Contribution to critical revision, adding intellectual content.

Cecilia Calabuig: Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

## Conflicts of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

## Ethics

All studied specimens of *Boa constrictor constrictor* were found roadkilled (dead) and were collected under the license 40620, attributed by Instituto Chico Mendes de Conservação da Biodiversidade – ICMBio. Further ethic requirements do not apply to this study.

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## Patterns of avian malaria in tropical and temperate environments: testing the “The enemy release hypothesis”

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**Abstract:** According to the enemy release hypothesis (ERH) the spread of invasive species will be facilitated by release from their enemies as they occupy new areas. However, the ERH has rarely been tested by comparing populations of native (non-invasive, long established) species with expanding or shifting ranges, to the same species as invasive in another area. We tested the ERH with respect to blood parasite levels (prevalence and intensity of *Plasmodium* spp. and *Haemoproteus* spp.) of (a) two closely related, widely distributed species of thrush (*Turdus leucomelas* and *T. merula*), and (b) an invasive sparrow (*Passer domesticus*) whose range has expanded from the Old World to the New World since the 18<sup>th</sup> century. A total of 158 birds were sampled in Portugal and 99 in Brazil. All bird species were parasitized, and 55% of the individuals collected were parasitized, and the mean intensity of infection was of 28 parasites per 10,000 erythrocytes. We assessed whether differences in levels of infection (prevalence and intensity) were due to site (tropical/New World and temperate/Old World) or host species. The ERH was supported: *Passer domesticus* and *Turdus merula* had higher levels of parasitism in the Old World than in the New World. Thus, *P. domesticus* seems to be benefitting from its “recent” range expansion, compared to *T. leucomelas*, through ecological release from its native parasites and because the parasites of the recently invaded area seem to be infesting native species instead.

**Keywords:** Portugal, Brazil, *Turdus*, House sparrow, invasive bird, range expansion.

## Padrões de malária aviária em região tropical e temperada: testando a “hipótese da liberação do inimigo”

**Resumo:** De acordo com a hipótese da liberação do inimigo (HLI), a disseminação de espécies invasoras será facilitada pela liberação de seus inimigos ao ocuparem novas áreas. No entanto, a HLI raramente é testada comparando-se as populações de espécies nativas (não invasivas, estabelecidas há muito tempo) que apresentam expansão ou alteração de habitats, com populações das mesmas espécies em habitats que foram invadidos. Testamos a HLI com relação aos níveis de parasitas no sangue (prevalência e intensidade de *Plasmodium* spp. e *Haemoproteus* spp.). De (a) duas espécies estreitamente relacionadas e amplamente distribuídas de *Turdus* (*Turdus leucomelas* e *T. merula*), e (b) um pardal invasor (*Passer domesticus*) cujo alcance se expandiu do Velho Mundo para o Novo Mundo desde o século 18. Um total de 158 aves foram amostradas em Portugal e 99 no Brasil. Todas as espécies foram parasitadas e 55% dos indivíduos foram parasitados, sendo que a intensidade média da infecção foi de 28 parasitas por 10.000 eritrócitos. Avaliamos se as diferenças nos níveis de infecção (prevalência e intensidade) foram devidas ao local (tropical/Novo Mundo e temperado/Velho Mundo) ou espécies hospedeiras. A HLI foi corroborada: *Passer domesticus* e *Turdus merula* apresentaram valores mais elevados de parasitismo no Velho Mundo do que no Novo Mundo. Assim, *P. domesticus* parece estar se beneficiando de sua expansão “recente” em comparação com *T. leucomelas*, através da liberação ecológica de seus parasitas nativos porque os parasitas da área recentemente invadida parecem infestar espécies nativas.

**Palavras-chave:** Portugal, Brasil, *Turdus*, pardal, ave invasora, expansão de habitat.

## Introduction

Avian Haemosporidian infections is caused by blood parasites of the phylum Apicomplexa, which includes the genera *Plasmodium*, *Haemoproteus*, *Leucocytozoon* and *Fallisia* (Valkiūnas 2005). Parasites of the genus *Plasmodium* causes avian malaria and are transmitted by mosquitoes of the family Culicidae, while parasites of the genus *Haemoproteus* are transmitted by flies of the families Hippoboscidae and Ceratopogonidae (Santiago-Alarcon et al. 2012). *Plasmodium* and *Haemoproteus* infections have negative consequences to host fitness by reducing clutch sizes and nest defense behavior, increasing probability of clutch desertion, reducing hatching and fledging success and weaning nestlings with poorer body condition (Korpimäki et al. 1993, Dulfa 1996, Hakkarainen et al. 1998, Sanz et al. 2004, Andrezej 2005, Marzal et al. 2005). They also pose a physiological cost to the host birds (Norte et al. 2009, Dallas et al. 2016), and may ultimately lead to death (Warner 1968, Cannell et al. 2013, Vanstreels et al. 2014).

Due to climatic stability, it has been predicted that parasite pressure should be greater in tropical than temperate species since parasites and their vectors can persist throughout the year (Janzen 1970, Connell 1971). However, most avian studies have found the opposite pattern (Ricklefs 1992, Martin et al. 2007), with greater Haemosporidian prevalence in birds from temperate areas than those from tropical areas. Therefore, the Enemy Release Hypothesis (ERH) could help explain the success of temperate avian species introduced into tropical regions in relation to their blood hemoparasites (Lima et al. 2010, Marzal et al. 2011). The ERH states that the abundance and distribution of an invasive species will be facilitated by the absence of natural enemies in the new habitats (Keane & Crawley 2002). Thus, we expected that colonizers or exotic populations will have competitive advantages due to lower parasite loads and consequently better health conditions than endemic populations. According to Marzal et al. (2011), range shifts of host or parasite species may result in species interactions not previously experienced in the wild. Consequently, introduced species might encounter novel parasites whose virulence will partly determine whether they become invasive.

Only a few studies have tested the role of Haemosporidian parasites in invasive birds by comparing their prevalence in invasive birds living in temperate and tropical environments (Marzal et al. 2011), and they have generally produced inconclusive results. For example, the prevalence of Haemosporidian parasites did not differ significantly between populations of the common myna, *Acridotheres tristis*, in India (Ishtiaq et al. 2006) compared to an invasive population in Australia (Clark et al. 2015), whereas parasite prevalence in house sparrows in their native range was higher when compared to those introduced to central Brazil (Lima et al. 2010, Marzal et al. 2011). Small sample sizes were recognized as the main reason for these inconsistent conclusions because other variables, such as age, sex and body condition, can also influence parasite prevalence.

We used microscopy and PCR to examine the prevalence of avian Haemosporidian parasites in populations of two thrush species (*Turdus*) and the house sparrow (*Passer domesticus*) that inhabit tropical and temperate environments. We focused on addressing three questions: (1) Do Haemosporidian prevalence and density differ between host species? (2) Do Haemosporidian prevalence and density differ between populations of the same host species/genus, sampled in temperate (Western Europe, Portugal) and tropical (South America, Brazil)

environments? (3) Does Haemosporidian infection cause negative consequences to host health in both temperate and tropical areas?

In testing the potential contributions of the Enemy Release Hypothesis to explain the prevalence and intensity of infection, we hypothesize that *Turdus* spp. and *P. domesticus* will have greater Haemosporidian infection and parasitemia in birds sampled from their historical range (Europe) compared to birds sampled in their recent range (Brazil). Furthermore, we hypothesize that infected “invasive” birds will have a better health condition than those infected in their native area.

## Material and Methods

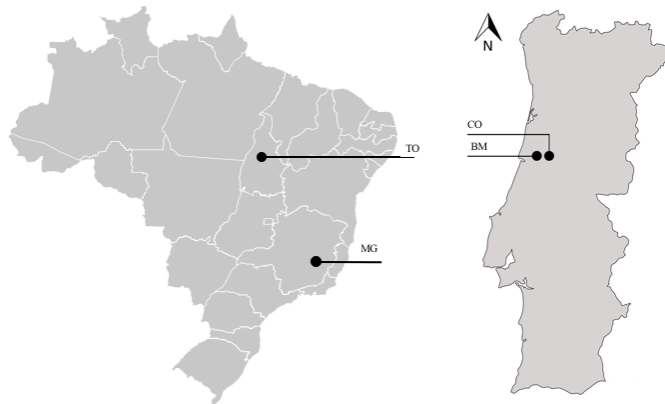
### 1. Bird species

The study was conducted with three bird species: the house sparrow *Passer domesticus*, the Pale-breasted thrush *Turdus leucomelas* and the Eurasian blackbird *T. merula*. Over the last two centuries, and with human assistance, the house sparrow has spread to all continents (except Antarctica) and many oceanic islands (Summers-Smith 1988). It expanded its range into South America during the 18th century, reaching Brazil in the same century, according to Efe et al. (2001). The avian genus *Turdus* is one of the largest and most widely distributed passerine genera, with 65 recognized extant species occurring throughout South America, Central and North America, Africa, and Eurasia; one species (*Turdus merula*) has been introduced to Australia. This is one of the few genera of passerines that has radiated extensively both in Africa, South America, and the Palearctic and Oriental regions (Clement & Hathway 2000, Collar 2005). The Eurasian blackbird is a species of true thrush that breeds in Europe, Asia, and North Africa, and was introduced to the Americas in the beginning of 20<sup>th</sup> century. The pale-breasted thrush is also a species of true thrush, and is found in a wide range of wooded habitats in eastern and northern South America, with localized populations in the West (Nylander et al. 2008). According to Nylander et al. (2008) *Turdus merula* and *T. leucomelas* shares a common ancestor.

### 2. Study area and sampling

The study was carried out during both the breeding and non-breeding seasons between May 2008 and February 2010 in Brazil (House sparrows in state of Minas Gerais and Pale-breasted thrush in Tocantins) and March and November 2010 in Portugal (Coimbra and Baixo Mondego, Eurasian blackbird and House sparrow) (Figure 1).

Birds were captured using mist-nets and banded with a metallic ring provided by Centro de Pesquisa para a Conservação de Aves Silvestres and Instituto Chico Mendes de Conservação da Biodiversidade (CEMAVE/ICMBio) in Brazil and Centro de Estudos de Migrações e Proteção de Aves and Instituto da Conservação da Natureza e das Florestas (CEMPA/ICNF) in Portugal. Approximately 50 µL of blood was collected from each individual through puncture of the brachial vein using a disposable hypodermic needle and microcapillary tubes. Measured morphometric characters included body weight, which was measured with spring scales (Pesola AG from Switzerland) with a precision of 0.1 g, and tarsus length, measured with a digital calliper to the nearest 0.01 mm. To evaluate host health, body condition was calculated using the residuals from the regression of body weight on tarsus length (Brown 1996). All collected samples were tested by microscopic and PCR-based methods in parallel.



**Figure 1.** Study areas in Brazil: TO-Tocantins, MG-Minas Gerais and Portugal CO-Coimbra, BM-Baixo Mondego Region.

### 3. Microscopic analyses

Three blood smears were prepared per bird for identification of the morphological stages of Haemosporidian parasites. Blood smears were air-dried, fixed in absolute methanol and stained with 10% Giemsa. A minimum of 100 fields was examined at high magnification (1000x) and the parasite intensity (i.e. parasitemia or density) was calculated by counting the number of infected erythrocytes per 10,000 erythrocytes (Godfrey et al. 1987, Valkiūnas 2005). Parasites (*Plasmodium* and *Haemoproteus*) were identified microscopically according to the morphological characteristics of the different blood stages (Valkiūnas 2005).

### 4. DNA extraction and screening

The remaining blood, after microscopic analyses, was immediately transferred into new sterile microtubes, with and without heparin, containing 300µL of cell lysis solution (Promega, MA, EUA) for DNA extraction.

Genomic DNA was extracted from bird blood samples using Wizard Genomic DNA Purification Kit (PROMEGA®, Madison, Wisconsin, USA) according to the manufacturer's protocol. In order to determine Haemosporidian infection, 50 to 100 ng of the extracted DNA was used for a screening PCR that amplifies a 154 nucleotide segment of ribosomal RNA coding sequence within the mitochondrial DNA of *Plasmodium* and/or *Haemoproteus* in a single reaction. We used the primers 343F (5' GCTCACGCATCGCTTCT 3') and 496R (5' GACCGGTCATTTTCTTTG 3') according to the protocol described by Fallon et al. (2003). The positive control consisted of genomic DNA from *Plasmodium gallinaceum* (obtained from experimentally infected chickens) and negative controls were DNA samples obtained from non-infected chickens maintained at the Veterinary School (UFMG). The overall prevalence of Haemosporidian infection was considered as the proportion of individuals infected, in each population of each species/region, based on the combination of the results of microscopic examination of blood smears and PCR analysis.

### 5. Statistical analyses

To investigate if there were any differences in the prevalence and infection intensity of avian malaria and body condition between house sparrows and thrushes from Brazil and Portugal, generalized linear mixed models (GLMM) were constructed considering a binomial

error distribution and logit link function for infection prevalence and quasi-poisson error distribution for infection intensity. In order to account for non-independence due to capture, random effects included individuals and location, and fixed effects included site (Portugal and Brazil), host species (*Passer domesticus* – BR and PT, *Turdus merula* and *T. leucomelas*), sex (Male and Female) age (Juvenile and Adult). To investigate if there were differences in the Haemosporidian infection on body condition, were constructed a model using parasitized and non-parasitized birds species from Brazil and Portugal as explanatory variables and body condition as response variable.

GLMM was used because it allowed us to pool all bird species by using individual infection status (infected or not) as the dependent variable, while controlling for sample size difference. Thus, information is not lost due to sample size restrictions, since more weight will be given to data with larger sample sizes (Paterson & Lello 2003, Jovani & Tella 2006). In addition, GLMM is a powerful method for analyzing parasitological data because it allows for the analyses of binomial data while controlling for correlations between measures that occur as a result of grouped observations (Paterson & Lello 2003). We used software R for all analyses (R Development Core Team 2016).

## Results

A total of 257 adult birds were captured: 40 house sparrows and 59 pale-breasted thrushes in the tropical region of Brazil and 66 House sparrows and 92 Eurasian blackbirds in the temperate region of Portugal. Taking into account all bird species in both regions, the total parasite prevalence was 55% and the mean parasite intensity was 27.96 parasites/10,000 erythrocytes. *Plasmodium* spp. and *Haemoproteus* spp. were detected. *Haemoproteus* spp. were found both in *Turdus merula* and *P. domesticus* in temperate region and only in *Turdus leucomelas* in Tropical region. *Plasmodium* and *Haemoproteus* prevalence in the temperate region (Portugal = 49% and 12% respectively) were higher than in the tropical region (Brazil = 37% and 10%) (Table 1).

Even when we analyzed bird species separately, this regional pattern remained the same. Trophozoites were the predominate form and were observed in all positive blood smears. Since these forms (Trophozoites) are not present in *Haemoproteus* parasites in blood smears, *Plasmodium* parasites were found in all the samples.

Other forms, such as meronts and gametocytes, were occasionally observed only in slides prepared from birds inhabiting temperate areas.

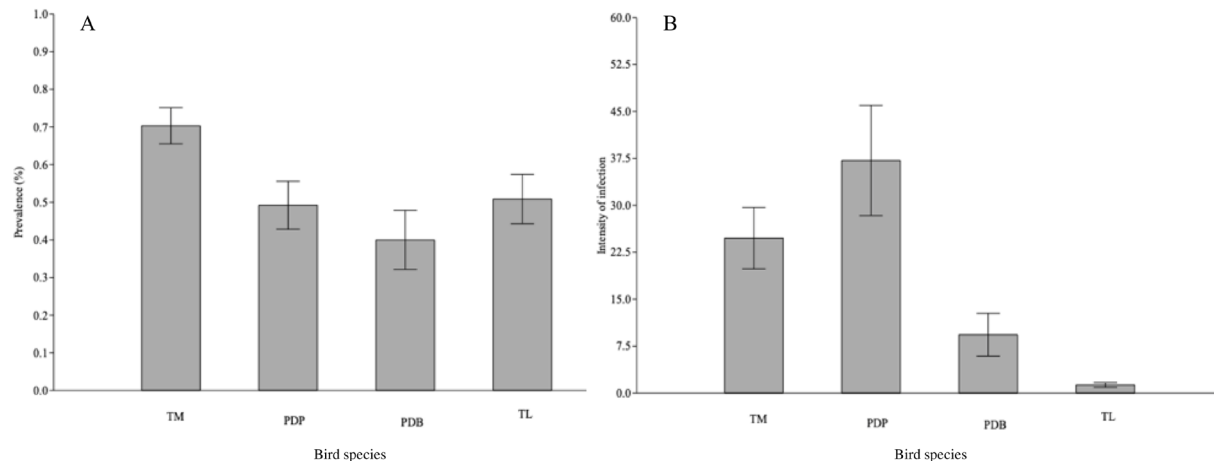
Prevalence and intensity were significantly higher in both House sparrows and thrushes from the temperate region when compared to birds from the tropical region ( $F_{2,257}=4.44$ ,  $P=0.036$  and  $F_{2,257}=5.34$ ,  $P=0.002$ ; Figure 2 A and B). Thrushes were significantly more infected ( $F_{2,257}=5.54$ ,  $P=0.032$ ) and had a higher mean parasite intensity than house sparrows ( $F_{1,257}=3.15$ ,  $P=0.004$ ) in both regions (Figure 2 A and B). Sex and age were not good predictors for Haemosporidian infection because we found no significant differences in the prevalence and intensity of infection between birds of different sex and among birds of different ages.

Haemosporidian infection had a significant influence on the body condition only for birds from the tropical region ( $F_{7,257}=79.203$ ,  $P<0.001$ ; Figure 3) when comparing positive and negative groups from both regions.

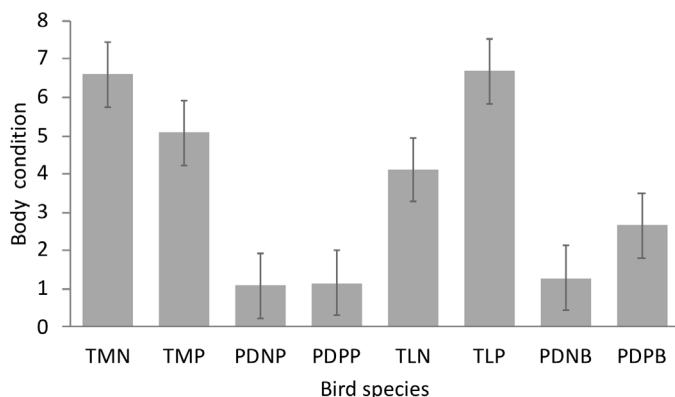
**Table 1.** Prevalence and parasite intensity of *Plasmodium* spp., using combined PCR and microscopy, in bird species from Temperate (Portugal) and Tropical (Brazil) regions. – TM = *Turdus merula*; TL = *Turdus leucomelas*; PD = *Passer domesticus*; H = *Haemoproreus*; P = *Plasmodium*.

Bird species	Country	Individuals (n)	Prevalence (%)		Parasite intensity (%) (Mean ± SE) <sup>a</sup>
			H	P	
TM	Portugal	92	16.0	54.0	24.76 ± 46.75
PD	Portugal	66	5.0	42.0	1.43 ± 2.14
TL	Brazil	59	10.0	40.0	1.30 ± 2.94
PD	Brazil	40	0.0	40.0	0.48 ± 0.64
Total		257	9.0	91.0	27.97 ± 30.32

<sup>a</sup> Prevalency was calculated by counting the number of infected erythrocytes per 10,000 erythrocytes.



**Figure 2.** Prevalence (A) and Intensity of infection (B) of Haemosporidian on bird species from Temperate (Portugal) and Tropical region (Brazil) after the combination of PCR and microscopic analysis. TM = *Turdus merula*, TL = *T. leucomelas*, PDP: *Passer domesticus* Portugal, PDB: *P. domesticus* Brasil. Error bars are mean ± sd.



**Figure 3.** Body condition of positive and negative birds species from Temperate (Portugal) and Tropical region (Brazil). TMN = *Turdus merula* negative, TMP = *Turdus merula* positive, TLP = *T. leucomelas* positive, TLN = *T. leucomelas* negative, PDPP: *Passer domesticus* positive Portugal, PDNP: *Passer domesticus* negative Portugal, PDPB: *P. domesticus* positive Brazil, PDNB: *P. domesticus* negative Brazil. Error bars are mean ± sd.

## Discussion

The Enemy Release Hypothesis predicts that a species whose range is expanding should have lower levels of parasitism in ‘new’ sites versus ‘old’ sites. Corroborating the ERH, we found that both house sparrows and thrushes in their native range (temperate zone) exhibited a higher prevalence of Haemosporidian infection than in their non-native or range expanding (tropical zone). Similar results

were obtained by Marzal et al. (2011), who pointed out that the most common lineage of Haemosporidian parasite, *Haemoproreus passeris*, was not observed in house sparrows in introduced areas. In addition, we found that *T. leucomelas* had lower Haemosporidian parasite infections than *T. merula* (the species considered as expanding range). Furthermore, our results support another prediction of the ERH, which is that non-indigenous species in their introduced range should exhibit lower parasite prevalence than in their native range. Indeed, we found that the prevalence and intensity of infection in the House sparrow were lower than those found in the thrush species in both sampled areas (Lafferty & Holt 2003, Torchin et al. 2003, Lafferty et al. 2005). Although native to Mediterranean areas, the range of the House sparrow has changed considerably. The success of the House sparrow in colonizing novel environments is unquestionably linked to the widespread occurrence of favorable agricultural and urban habitats resulting from human activities. It is important to notice that the main difference on the intensity of infection among the House sparrow from temperate versus tropical region, could be more important, since the difference on prevalence was small, but significant.

This same pattern of parasite infection was found by Ricklefs & Sheldon (2007) studying *Turdus* spp. from temperate and tropical zones. Although not significant, they found a Haemosporidian prevalence of 57% in *Turdus migratorius* from Missouri and a prevalence of 63% in this host species from Michigan, whereas the prevalence in *Turdus gray* from Panama was only 41%. Moreover, they found a significantly higher infection intensity in thrushes from temperate than from tropical



zones (Ricklefs & Sheldon 2007). Our results are also in accordance with those of Lima et al. (2010), who found, despite their low sample size for native urban birds, a low prevalence of Haemosporidian in the house sparrow compared to native bird species in Brazil.

Considering the three-bird species together, we found the prevalence of Haemosporidian infection to be significantly higher for the tropical regions compared to temperate (70% and 61% respectively). Discussion if the prevalence of Haemosporidian are lower in tropical regions, compared to temperate regions are frequent in the literature. Several studies have suggested that Haemosporidian infections are generally less prevalent in the tropics than in temperate regions (Ricklefs 1992, Valkiūnas et al. 2006). A lower prevalence was found in other temperate areas, such as Southeast Mediterranean (42%, Marzal et al. 2011) and France (41%, Loiseau et al. 2011), but higher values for prevalence was also found in Southeast and Southwest Europe, with 61.6% and 52.9%, respectively (Marzal et al. 2011). The total prevalence for the tropical areas of our study was also higher than that found by Lima et al (2010) (20%), but was similar to that reported by Belo et al. (2011) (55%) and Lobato et al. (2011) (58%).

Haemosporidian prevalence for the Eurasian blackbird (70%) was higher than previously described for this host species, with approximately 31% in Germany (Geue & Parteche 2008) and 53% on the island of São Miguel, Azores (Hellgren et al. 2011). For the Pale breasted thrush, Haemosporidian prevalence (50%) was also higher than previously described in Brazil (20% Lima et al. 2010; 23% (Sebaio et al. 2012), however, it was lower than that described in another study in Brazil (58% Lobato et al. 2011). For the House sparrow, we also found higher prevalence among Old World birds, while the prevalence in Brazil was higher than that previously described by Lima et al. (2010) and by Marzal et al. (2011). Clearly, parasite prevalence differed drastically between native bird species and populations of the non-native House sparrow on different continents and from different study areas, possibly resulting from different trade-offs between immune function against parasites and other energetically costly activities such as predator defense and competition, among others (Navarro et al. 2004). This finding also supports the idea that prevalence varies geographically and from population to population (Merilä et al. 1995), and could be a result of variation in exposure among localities, which should be related to vector presence, abundance and competence (Bennett et al. 1998, Norte et al. 2009).

In addition to host richness, climatic conditions and lack of suitable vectors could explain the low parasitemia among the House sparrows in Brazil. One hypothesis is that the lower prevalence of blood parasites is due to the lack of suitable vectors for the completion of the parasite's life cycle. However, since native urban bird species were infected with Haemosporidian, it appears that suitable vectors are present in the urban localities where the birds were sampled (Valkiūnas et al. 2006).

Haemosporidian vectors require specific conditions of temperature and humidity to complete their life cycle (Hopp & Foley 2001). Although Portugal has lower mean temperatures and pluviosity than Brazil, temperate environments have higher thermal variation throughout the year, with distinct seasons and high temperatures only during summer, resulting in an increase in vectors during this period. However, tropical environments, where the temperature and rainfall are distributed throughout the year, offer better conditions for maintaining

active vector populations that could infect hosts throughout the year. This could explain why we observed trophozoites in most of the samples, even though they are difficult to detect.

Body condition was not a good predictor of Haemosporidian infection since it did not differ between infected and non-infected hosts from the temperate region, and although the differences were greater between infected and non-infected birds from the tropical areas, they were not significant. Other studies also failed to find a relationship between parasitemia and body condition, such as in *Ploceus capensis* from South Africa (Schultz et al. 2010). Norte et al. (2009), however, found that body condition was related to haemoparasite infection in birds from Portugal. In their study, the body condition index decreased with infection by *Leucocytozoon* and *Plasmodium*, but was dependent upon the year and season. The low body condition of the birds in our study could be explained by the large energy expenditure by breeding individuals, followed by the dry season when weather conditions are more adverse. Negative effects of haematozoan infection on body mass are difficult to detect in wild birds that co-evolve with blood parasites (Bennett et al. 1988), and may also be related to food availability throughout the year and/or reproductive period (Schrader et al. 2003).

The pathogenicity of blood parasites in wild birds has been difficult to demonstrate because of the capacity of the hosts to maintain infections below a threshold at which effects become apparent (Marzal et al. 2005, Norte et al. 2009, Lobato et al. 2011). We show that the house sparrow in non-native regions has a lower prevalence and intensity of haemosporidia infections than in their native range, which is consistent with the ERH. As the ERH is still controversial, more detailed comparative studies of house sparrows, as well as other bird species, and their malaria parasites and co-existing host species in native and non-native ranges are necessary to shed light on these important questions and to better understand the impact of invasive species on the biodiversity of the invaded communities.

"Parasite prevalence" refers to the proportion of a host population that is infected, which is traditionally measured by visual inspection of blood smears (Godfrey et al. 1987, Valkiūnas 2005). However, prevalence data have not been compared broadly with adequate control for host taxonomy. Moreover, prevalence might be confounded by immune response to the extent that it reflects the ability of host individuals to control chronic infections below the level of detection by visual examination of blood smears. Thus, the present study has contributed to a better understanding of variation in haematozoa prevalence and ecology in host bird species of two genus, in two geographic regions.

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## Author Contributions

Yasmine Antonini: Contributed to data analysis and interpretation, manuscript preparation and critical revision; Contributed to data collection.

Debora Nogueira Campos Lobato: Contributed to data analysis and interpretation, manuscript preparation and critical revision; Contributed to data collection.

Ana Cláudia Norte: Contributed to data analysis and interpretation, manuscript preparation and critical revision; Contributed to data collection.

Jaime A. Ramos: Contributed to data analysis and interpretation, manuscript preparation and critical revision.

Patrícia de Abreu Moreira: Contributed to data analysis and interpretation, manuscript preparation and critical revision.

Erika Martins Braga: Contributed to data analysis and interpretation, manuscript preparation and critical revision.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## A preliminary overview of the Brazilian Apioninae (Coleoptera: Brentidae) with an illustrated key for genera, and a checklist with distribution information

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**Abstract:** Here we examine the taxonomic diversity of the Brazilian Apioninae (Coleoptera: Brentidae) and provide an update based on the literature and through examination of material in primary Brazilian collections. Ten genera and 89 species are valid and we include 30 (25 genera, 5 species) new distribution records. *Chrysapion* Kissinger, 1968 is first reported in Brazil, restricted to the northeast. Ranges now include the Amazon (three genera and one subgenus) and Atlantic (four genera and one subgenus) forests, the Pantanal (five genera) and Cerrado (three genera). The ranges of several genera now include more than one new biome. An identification key (including images of adults and illustrations of male genitalia) also includes the 10 Brazilian genera of Apioninae for the first time.

**Keywords:** Curculionoidea, Neotropical Region, taxonomy, systematic.

## Uma visão geral preliminar dos Apioninae (Coleoptera: Brentidae) brasileiros: chave ilustrada para identificação de gêneros, lista de espécies e registros de distribuição

**Resumo:** O conhecimento sobre a diversidade taxonômica da fauna de Apioninae (Coleoptera: Brentidae) do Brasil é atualizado com base na literatura disponível e exame de material depositado nas principais coleções públicas do país. Dez gêneros e de 89 espécies válidas foram incluídas em 30 novos registros de distribuição (25 gêneros e cinco espécies). *Chrysapion* Kissinger, 1968 é registrado pela primeira para o Brasil, restrito aos biomas do Nordeste do Brasil. A distribuição geográfica inclui Amazônia (três gêneros e um subgênero) e Mata Atlântica (quatro gêneros e um subgênero), o Pantanal (cinco gêneros) e Cerrado (três gêneros). A distribuição de vários gêneros é atualizada para mais de um bioma. Uma chave de identificação (com imagens dos adultos e desenhos da genitália masculina) é apresentada pela primeira vez para os 10 gêneros de Apioninae que ocorrem no Brasil.

**Palavras-chave:** Curculionoidea, Região Neotropical, sistemática, taxonomia.

## Introduction

The subfamily Apioninae (Brentidae *sensu lato*) comprises a monophyletic group (Alonso-Zarazaga & Wanat 2014; Marvaldi et al. 2002; Kuschel 1995; Wanat 2001) of very small beetles (0.75-13.00 mm in length, >95% < 3 mm). This subfamily differs from the other Curculionoidea by the following: a markedly convex to globose, pear-shaped (*Apion* is Greek for 'small pear') body (except Antliarhinitae); straight antennae with a compact club that appear geniculate in some Rhadinocybini; short, two-segmented, palps; nine-striate elytra (Apionini) with some genera having remainders of a tenth; brentid-type abdomen; two large, usually convex, first ventrites, the second almost vertical at apex; the last three ventrites small, forming a valve-like structure that opens to extrude genitalia; usually long, cylindrical trochanters but which may be short in several basal lineages; tegmen with fully developed and diverse parameral plate (Alonso-Zarazaga & Wanat 2014; Anderson & Kissinger 2002; Jones et al. 2012; Kissinger 2005a; Ptaszynska et al. 2012).

Adults feed on living plant tissues, including all green parts as well as fruit. Larvae are usually endophagous and also feed on different plant parts, including stems, roots, inflorescences, fruit, seeds and tissues forming gall-like structures. In contrast to Arctic fauna of both the Old and New Worlds, the fauna of the Neotropical Region is very poorly known (Alonso-Zarazaga & Wanat 2014). While some few basal lineages of the Apioninae are associated with gymnosperms (Kissinger 1968; Alonso-Zarazaga and Wanat 2014), most basal groups are associated with plants in the clade Magnoliid as hosts (Alonso-Zarazaga & Wanat 2014). More apical apionines (Apionitae: Apionini) have at least 23 families in 15 orders of dicot angiosperms (Adoxaceae, Apiaceae, Asteraceae, Polygonaceae, Caprifoliaceae, Ebenaceae, Euphorbiaceae, Fabaceae, Lamiaceae, Malvaceae, Melastomataceae, Proteaceae, Rhamnaceae, Rutaceae, Salicaceae and Verbenaceae; Alonso-Zarazaga & Wanat 2014; Anderson & Kissinger 2002; Badenes-Perez & Jhonson 2007; Lima et al. 2008; Maia 2012). However, host associations of the Apioninae in South America are largely unknown (Alonso-Zarazaga & Wanat 2014).

More than 2,200 species of Apioninae have been described (Alonso-Zarazaga & Wanat, 2014), where they are found from high altitudes to sea level throughout the region (Alonso-Zarazaga 2004; Anderson & Kissinger 2002; Oberprieler et al. 2007; Alonso-Zarazaga & Wanat 2014) and where the species comprise about 205 genera and subgenera (Alonso-Zarazaga & Wanat 2014), seven supertribes and nine tribes (Bouchard et al. 2011; Alonso-Zarazaga 1990).

Kissinger (1968) pioneered the study of the genera and subgenera of *Apion* while focusing on the fauna of North and Central America, where he described an extraordinary diversity of forms and demonstrated that the tegmen of male genitalia provides important characters to determine their taxonomy. In South America, the Apioninae comprise 190 species (Wibmer & O'Brien 1986) in 19 genera, 10 of which were thought to be endemic (Kissinger 1968, 2002, 2003, 2005a, b). Among other South American countries, Brazil also has no recent studies of the Apioninae and the little taxonomic information available can be found in 17 published studies that include 86 species (summarized and listed under the genus *Apion* Herbst, 1797 in the catalog of Wibmer and O'Brien 1986).

Some subgenera of *Apion* were elevated to genera after Kissinger (1968) and Wibmer and O'Brien (1986) laid the groundwork (Alonso-Zarazaga 1990; Kissinger 1992). Subsequently, new genera

and subgenera were proposed (Alonso-Zarazaga 1990; Kissinger 1990, 1991, 1998, 1999b, 2002, 2005a). In the catalog of the families and genera of Curculionoidea (Alonso-Zarazaga & Lyal 1999), only five genera in the Apioninae were reported in Brazil, each of which only included the type species: *Neapion* Alonso-Zarazaga, 1990 (Ixapiini), *Apionion* Kissinger, 1998, *Bothryoapteron* Wagner, 1912a, *Coelopterapion* Wagner 1912a and *Stenapion* Wagner 1912a.

Currently, the only classification for suprageneric and generic Apioninae taxa is based on Palearctic fauna (Alonso-Zarazaga 1990, Ptaszynska et al. 2012, Winter et al. 2017). The Alonso-Zarazaga classification scheme does not include many North and Central American and most South American taxa, nor do the molecular studies of Ptaszynska et al. (2012) and Winter et al. (2017) include North, Central and South American taxa. Consequently, the basis for identification of the Apioninae of South America continues to be Kissinger (1968), even though obsolete. The genus *Apion* is now considered to be a monophyletic genus of 16 species that is restricted to the Palearctic (Alonso-Zarazaga 1990) and so many species listed by Kissinger (1968) remain in incorrect genera.

Today, hundreds of specimens have accumulated in collections in South American institutions under the genus *Apion*. This material is very important for taxonomic and systematic understanding of the Apioninae. Thus, we present a first step in understanding the Apioninae of Brazil by providing a checklist of genera and species, with their geographic distributions, host plants, citations of the relevant literature, and a key for the genera including photographs and illustrations.

## Material and Methods

Specimens of the Apioninae were examined between August 2016 and March 2017 at the following institutions: Coleção Entomológica Pe. J. S. Moure, Departamento de Zoologia, Universidade Federal do Paraná, Curitiba (DZUP); Coleção Entomológica do Instituto de Biociências, Universidade Federal de Mato Grosso, Cuiabá (UFMT); Instituto Nacional de Pesquisas da Amazônia, Manaus (INPA); Laboratório de Ecologia e Taxonomia de Artrópodes Terrestres, Universidade Federal de Mato Grosso, Cuiabá (LETA); Museu Nacional, Universidade Federal do Rio de Janeiro, Rio de Janeiro (MNRJ); Museu de Zoologia, Universidade de São Paulo, São Paulo (MZSP).

Taxa identification, development of the dichotomous key and the species checklist of Brazilian Apioninae were based on Wagner (1910, 1912a, b, c, 1914, 1915), Kissinger (1968, 1998, 2002, 2003, 2005a, b), O'Brien & Wibmer (1982), Wibmer & O'Brien (1986) and Alonso-Zarazaga & Lyal (1999). We adapted and modified the genera identification key from Alonso-Zarazaga (2004). We based the identification key on male characteristics because they are relevant for specific, generic and suprageneric classifications and diagnoses (Anderson & Kissinger 2002; Wanat et al. 2001). To better examine the sexes and genitalia morphology, dried specimens were softened in lukewarm water with detergent for 24 hours. Genitalia extraction methods follow Alonso-Zarazaga (1989) and terminology for external morphology follows Alonso-Zarazaga & Wanat (2014).

We examined the type material of the following 13 species: *Apion cydoniae* Bondar, 1950 and *A. zikani* Heller, 1922 (Museu Nacional, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil - MNRJ); *Apion argentinum* Béguin-Billecocq 1909, *A. contrarium*



Béguin-Billecocq, 1909, *A. desbordesii* Béguin-Billecocq, 1911, *A. ingratum* Béguin-Billecocq, 1909, *A. lativentre* Béguin-Billecocq, 1909, *A. salpingoides* Béguin-Billecocq, 1909, *A. simplex* Béguin-Billecocq, 1909, *A. rufonigrum* Béguin-Billecocq, 1911 and *A. tucumanense* Béguin-Billecocq, 1909 (Museu Argentino de Ciencias Naturales “Bernardino Rivadavia”, Buenos Aires, Argentina - MACN); *Neapion (Neotropion) diringsi* De Sousa & Ribeiro-Costa, 2018 (MZSP) and *Neapion (Neotropion) marquesae* De Sousa & Ribeiro-Costa, 2018 (DZUP).

We examined photographic images of the type material of the following species: *Apion areolatum* Kirsch, 1874, *A. luteipes* Kirsch, 1874 and *A. opacum* Kirsch, 1874 deposited in the Staatliches Museum für Tierkunde, Dresden (SMTD); *A. novaeteutoniae* Voss, 1959b, *A. phaseoli* Voss, 1958 and *A. sesbaniae* Voss, 1958 (Zoological Museum, Hamburg); *Apion clarki* Kissinger, 1990 (Smithsonian National Museum of Natural History, Washington, USA - USMN); *Apion hibisci* Fall, 1918, *A. neolentum* Kissinger, 1968, *A. umboniferum* Fall, 1898, *A. xanthoxyli* Fall, 1898 (Database of Zoological Collections - CVZBASE of the Museum of Comparative Zoology - Harvard University, <http://mczbase.mcz.harvard.edu/name/Apion>). We also examined photographic images and redescrptions in Kissinger (1968) of the following species: *Apion americanum* Wagner, 1908, *Apion aurichalceum* Wagner, 1912b, *Apion bryanti* Wagner, 1914, *Apion constricticollis* Sharp, 1890, *Apion chiriquense* Sharp, 1890, *Apion crassum* Fall, 1898, *Apion grallarium* Sharp, 1891a, *A. peculiare* Wagner, 1909, *A. spinitarse* Wagner, 1912a and *A. spretissimum* Sharp, 1890. We examined photographic images of material named *Chrysapion auctum* (Sharp, 1890) and *Chrysapion tantillum* (Sharp, 1890) from Arizona State University Hasbrouck Insect Collection (<http://symbiota4.acis.ufl.edu/scan/portal/>). Some species are very conspicuous, well-illustrated or labeled in such a way as to make examination of the type material unnecessary (e.g. *Apion binodosum* Wagner, 1912a, *A. cionoides* Wagner, 1912a, *A. testaceum* Wagner, 1912a, *A. luteirostre* Gerstaecker, 1854, *A. brevicorne* Gerstaecker, 1854, *A. ensirostre* Gerstaecker, 1854, *A. periscelis* Gyllenhal, 1839, *A. pseudampullula* Voss, 1940, *A. undulipenne* Wagner, 1912a).

We indicate new records in the geographic distribution of the checklist by placing them in parentheses. Geographical records from the literature were complemented by the examination of the labels of the specimens deposited in the Brazilian public institution collections.

Classification for the suprageneric taxa is in alphabetical order in the checklist, following Bouchard *et al.* (2011). Genera and species determined to be *Incertae sedis* are placed in a special section at the end of the checklist.

Suprageneric group names are typed in bold capital letters, while genera, subgenera and species group names are both in italic and bold capitals, followed by author, date and page of publication preceded by a colon (:). All genera and subgenera include: type-species, synonyms, distribution in bold type face, country in parentheses, host plants, references and page of publication preceded by an colon (:), remarks explaining how the taxa was recognized. Genera are left-justified while subgenera are indented and in parentheses. The most current species name is followed by author, date and page of publication in parentheses. We include synonyms in chronological order, followed by comments (if needed, in parentheses) and references between square brackets; distribution in bold type faces and states between in parentheses, references and page of publication, remarks about the species and material examined when studied.

Photographic images were taken using a Leica MZ16 stereomicroscope, Leica DFC 500 camera, Leica 3D viewer module and version montage 4.7 (software). Drawings and images were edited in Photoshop CS6. Schematics of the male genitalia were drawn using a stereomicroscope or a microscope with a light camera coupled system or image capture system.

## Results

From ca 230 specimens in collections in Brazil, we identified 10 genera and two subgenera of Apioninae in a total of 43 species. We identify 25 Brazilian species, 15 of which are placed in “*Apion*” in the broadest sense. Thirty new records are included, one for Brazil: *Chrysapion* Kissinger, 1968. Another 24 records are new for Brazil: the geographic distribution of *Neapion* Alonso-Zarazaga, 1990, subgenus *Neotropion* Alonso-Zarazaga, 1990 is extended to include Amazon (Acre and Pará) and Atlantic (São Paulo and Paraná) forests; *Coelocephalapion* Wagner, 1914 is extended to the Pantanal (Mato Grosso and Mato Grosso do Sul); *Apionion* Kissinger, 1998 now includes the Pantanal (Mato Grosso) and Atlantic Forest (Rio de Janeiro); *Bothryopteron* Wagner, 1912 is extended to the Amazon (Maranhão), Pantanal (Mato Grosso), Cerrado (Tocantins and Goiás) and Atlantic Forest (Santa Catarina); *Coelopterapion* Wagner, 1912 to the Pantanal (Mato Grosso), Cerrado and Atlantic Forest (Minas Gerais, São Paulo and Paraná); *Mythapion* Kissinger, 2005 now includes the Amazon (Amazonas); *Stenapion* Wagner, 1912 is also extended to include Amazon Forest (Pará), the Pantanal (Mato Grosso and Mato Grosso do Sul), Cerrado and Atlantic Forest (Minas Gerais and Santa Catarina). *Apionion samson* is reported for the first time from Brazil (Minas Gerais), two species of *Coelopterapion*, *C. cionoides* (Wagner 1912a) and *C. testaceum* (Wagner 1912a) are first records from the state of São Paulo; *Trichapion lativentre* (Béguin-Billecocq, 1909) is a first record for Paraná; one species of *Apion* in the broadest sense, *A. ensirostre* Gerstaecker, 1854, is a first record for the state of Mato Grosso do Sul.

### Key to the genera of Brazilian Apioninae (males)

1. Middle coxae contiguous, not separated by junction of meso- and metaventral processes (Fig.1) ..... *Chrysapion* Kissinger, 1968
  - Middle coxae separated by junction of meso- and metaventral processes (Fig. 2) ..... 2
2. Elytra expanded and nodose apically; apical prominences in the intervals 2 + 3 and 8 interrupted by large deep pit (Fig. 6); if elytra not nodose apically, the apex of intervals 2 + 3 and 8 interrupted by punctures; pronotum conical to subconical and without basal flange when in dorsal view ..... *Bothryopteron* Wagner, 1912a
  - Elytra not expanded and nodose apically; apex at intervals 2 + 3, and 8 not interrupted by large deep pit or punctures; pronotum cylindrical, conical or campaniform to subturbinate or tripartite, with or without basal flange in dorsal view ..... 3
3. Rostrum straight in side view (Fig. 8), stout and cylindrical pronotum subcylindrical (Fig. 7) to tripartite (Fig. 7) ..... *Stenapion* Wagner, 1912a



- Rostrum more or less distinctly curved or tapering towards apex in side view; pronotum conical or campaniform to subtrubinate ..... 4
- 4. Elytra with fasciate pattern formed by light and/or dark scales (Fig.9); first protarsomere at most isodiametric, second clearly transverse; mesocoxae widely separated (0.25-0.30 times middle coxae diameter) *Neapion* Alonso-Zarazaga, 1990 ..... 5
- Elytra without fasciate pattern; first protarsomere distinctly longer than wide; distance between mesocoxae usually not separated by more than 0.25 mesocoxal diameter (Fig. 2) ..... 6
- 5. Pronotum subconical, sides in basal two-thirds straight (Fig. 9); body robust; elytra convex to strongly convex, with or without raised areas in intervals (Fig. 10). On Rutaceae ..... Subgenus *Neotropion* Alonso-Zarazaga, 1999
- Pronotum subcylindrical, sides rounded; elongated body; elytra never convex to strongly convex, without raised areas in intervals. On Caprifoliaceae ..... Subgenus *Neapion* Alonso-Zarazaga, 1990
- 6. Body stout, pyriform and sub-sphaerical elytra in dorsal view; pronotum conical (Fig. 11); elytra strongly convex in lateral view (Fig. 12) ..... *Coelopterapion* Wagner, 1912a
- Body more elongate; pronotum conical (Fig. 17) to subconical (Fig. 20) or campaniform to subtrubinate (Figs. 22, 27); elytra less convex in lateral view (Figs. 17, 18, 21, 23, 28) ..... 7
- 7. Ventral region of the rostrum with a high median keel, sulci between the median and lateral keel punctate and with semierect pilosity; pronotum campaniform with vestiture perpendicular to the apical margin; elytra with base of the intervals 3 with a dense pubescent spots. On Malvaceae ..... *Alocentron* Schilsky 1901 (subgenus *Nearctalox* Alonso-Zarazaga, 1990)
- With another character combination ..... 8
- 8. Tegminal plate fused with free ring (Fig. 13-14) ..... *Coelocephalapion* Wagner, 1914
- Tegminal plate articulated with free ring (Fig. 15-16) ..... 9
- 9. All tibia not mucronate ..... *Apionion* Kissinger, 1998 (in part)
- At least median tibia mucronate ..... 10
- 10. Pronotum conical to subconical, without basal flange (Fig. 20); fore tibia not mucronate ..... *Apionion* Kissinger, 1998 (in part)
- Pronotum otherwise; fore tibia mucronate ..... 11
- 11. Pronotum with basal flange (Fig. 22) and/or prescutellar fovea; if fore tibia not mucronate, the pronotum is always campaniform (Fig. 22) to subtrubinate; first tarsomere of one of the legs modified in the form of plantar spine (Fig. 26) *Trichapion* Wagner, 1912b
- Pronotum without basal flange and prescutellar fovea (Fig. 27); fore tibiae mucronate; tarsomeres not modified in the form of plantar spine ..... *Mythapion* Kissinger, 2005a

#### Checklist of the Apioninae recorded in Brazil

Subfamily Apioninae Schönherr, 1823:1136

Supertribe Apionitae Schönherr, 1823

Tribe Apionini Schönherr, 1823

Subtribe Aspidapiina Alonso-Zarazaga, 1990:34

Genus *Alocentron* Schilsky, 1901:G (in key)

Type-species: *Apion curvirostre* Gyllenhal, 1833.

#### Subgenus *Nearctalox* Alonso-Zarazaga, 1990:54

Type-species: *Apion hibisci* Fall, 1918:219.

Synonyms: *Apion* subgenus *Alocentron* sensu Kissinger 1968:234, 258 [Alonso-Zarazaga 1990: 54].

Distribution: **North, Central and South America (Brazil).**

Host plants: Malvaceae.

References: Kissinger 1968:234, 1988:304, 1992:65; Alonso-Zarazaga 1990:54, 2004:696.

Remarks: examined images of the type species available in The Database of the Zoological Collections (CVZBASE) of the Museum of Comparative Zoology - Harvard was compared with the studied material.

#### *Alocentron chiriquense* (Sharp, 1890)

Synonyms: *Apion chiriquense* Sharp, 1890:70; *Apion chiriquense* sbsp. *curvinasus* Wagner, 1912b: 100; *Apion chiriquense* var. *curvinasus* Wagner, 1912b:118.

Distribution: **Mexico, Guatemala, Nicaragua, Panama, Brazil (Rio de Janeiro).**

References: Blackwelder 1947:828; Kissinger 1959a:28, 1968:235, 236; O'Brien and Wibmer 1982:21; Wibmer and O'Brien 1986:40.

#### Subtribe Ixapiina Alonso-Zarazaga, 1990:71

##### Genus *Neapion* Alonso-Zarazaga, 1990:75

Type-species: *Apion umboniferum* Fall, 1898:174.

Synonyms: *Apion* subgenus *Ixias* sensu Kissinger 1968:41, 257, not Sainte-Claire Deville, 1924:122 (= *Ixapion* Roudier et Tempère, 1973:80) [Alonso-Zarazaga 1990: 54]; *Xixias* Kissinger, 1991:35.

Remarks: examined images of the type species available in The Database of the Zoological Collections (CVZBASE) of the Museum of Comparative Zoology - Harvard University was compared with the studied material.

#### Subgenus *Neotropion* Alonso-Zarazaga, 1990:75

Type-species: *Apion xanthoxyli* Fall, 1898:176.

Distribution: **North, Central and South America (Colombia, Venezuela, Trinidad, Brazil and Paraguay), Antilles.**

Host plants: Rutaceae.

References: Kissinger 1959a:24, 31, 1968:42 (as *peculiare* species group), 1990:37, 1992:65; Alonso-Zarazaga 1990:75, 2004:696; Alonso-Zarazaga and Lyal 1999:58.

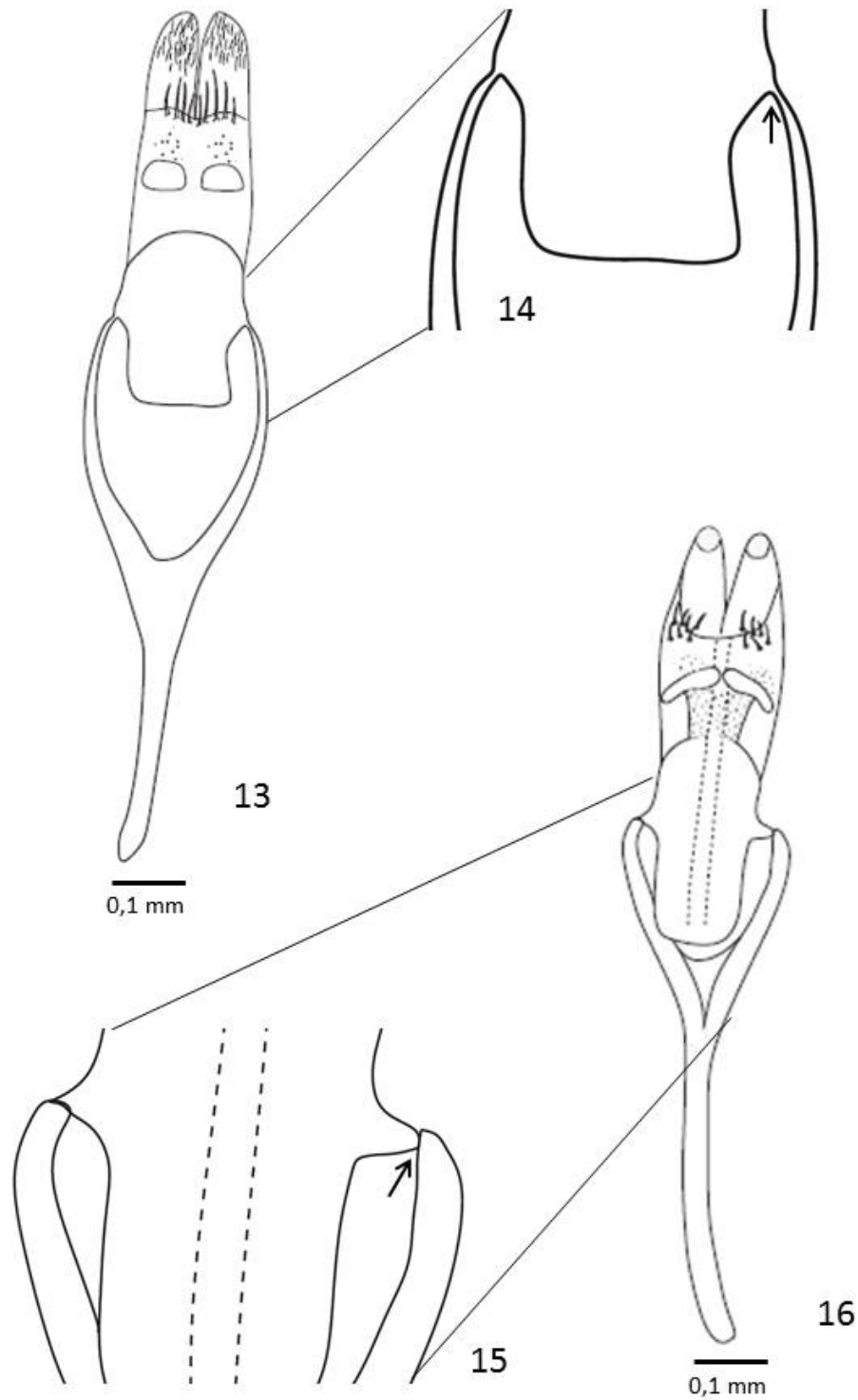
Remarks: examined images of the type species available in The Database of the Zoological Collections (CVZBASE) of the Museum of Comparative Zoology - Harvard University and of the *Apion clarki* Kissinger, 1990 deposited in the Smithsonian National Museum of Natural History, Washington, EUA (USMN) was compared with the studied material.

Material examined: *Neapion* (*Neotropion*) undetermined species.

**Brazil: Amazonas:** Rio Solimões, Lago do José: 1 ind., IIIV-09-1979, J. Adis, Fumigação (fogging), W. O. Sousa det. (INPA); **Acre (New record):** Rio Branco: 1 ind., IV-1954, 1 ind., X-1954, M. Alvarenga col., W. O. Sousa det. (MZSP); **Pará (New record):** Tapará: 1 ind., IX-1922, H. L. Boy, W. O. Sousa det. (MNRJ);

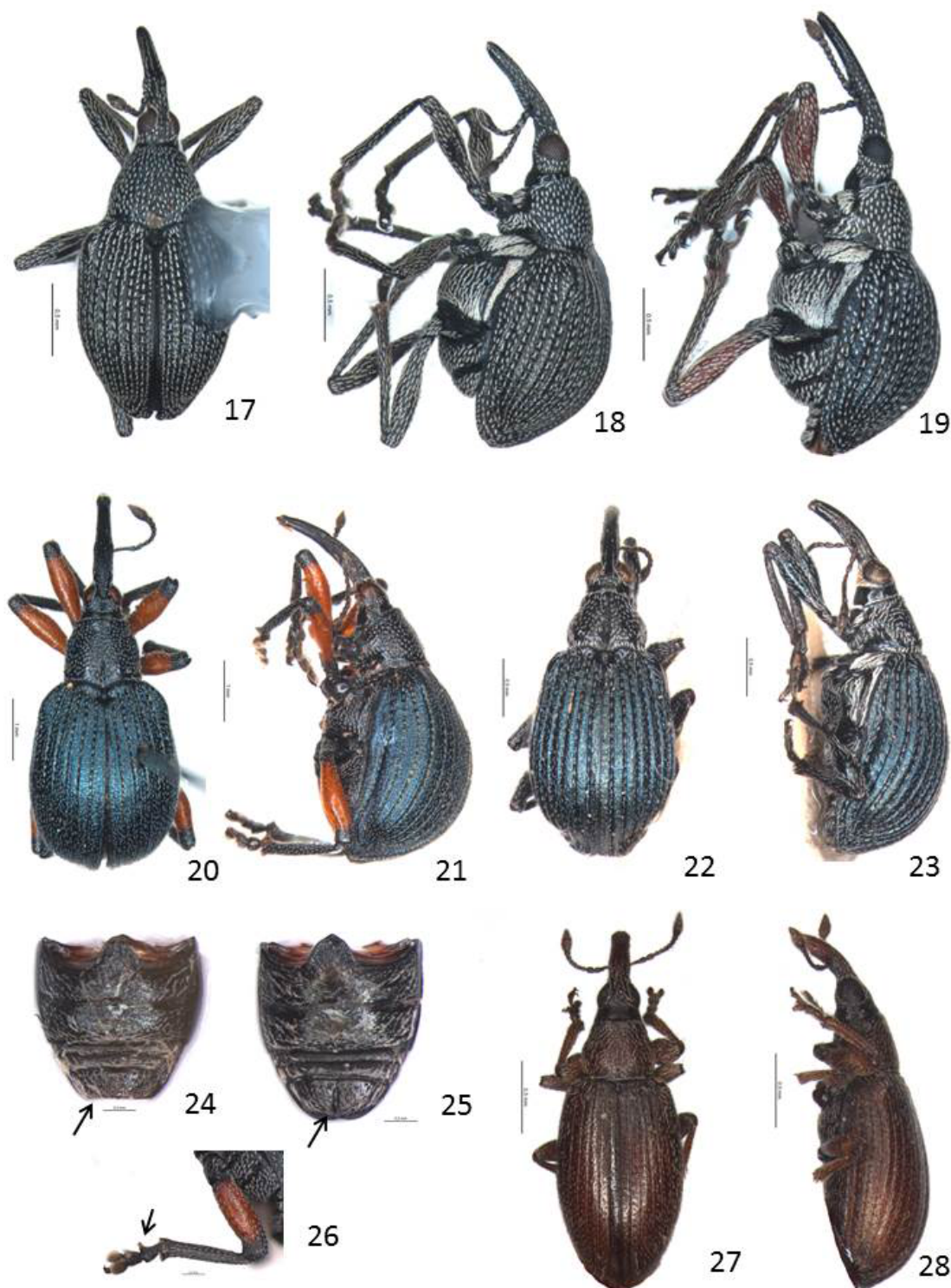


**Figures 1-12.** *Chrysapion* sp.: 1, prothorax, meso- and metaventrite ventral view; 3, dorsal view; 4, lateral view. *Trichapion* sp.: 2, prothorax, meso- and metaventrite ventral view. *Bothryopteron binodosum* (Wagner): 5, dorsal view; 6, lateral view. *Stenapion* sp.: 7, dorsal view; 8, lateral view. *Neapion* (*Neotropion*) *marquesae* De Sousa & Ribeiro-Costa: 9, dorsal view; 10, lateral view. *Coeloapterapion testaceum* (Wagner): 11, dorsal view; 12, lateral view.



**Figures 13-16.** *Neapion (Neotropion) marquesae* De Sousa & Ribeiro-Costa: **13**, Male genitalia: tegmen, dorsal view; **14**, prostegium fused with free ring. *Trichapion* sp. (Béguin-Billecocq): **15**, Male genitalia: tegmen, dorsal view; **16**, prostegium articulated with free ring.





**Figures 17-28.** *Coelocephalapion* sp.: 17, male, dorsal view; 18, male, lateral view; 19, female, lateral view. *Apionion samson* (Sharp): 20, dorsal view; 21, lateral view. *Trichapion* sp. (Béguin-Billecocq): 22, dorsal view; 23, lateral view; 24, male, abdomen with apical margin of 5<sup>th</sup> sternite truncate; 25, female, abdomen with rounded margin and median area of 5<sup>th</sup> sternite marked by a median sulcus. *Trichapion* sp.: 26, trochanter, femur, tibia, tarsus 3 and tarsomere 1 modified in the form of plantar spine of male. *Mythapion simplex* (Béguin-Billecocq): 27, dorsal view; 28, lateral view.

**Mato-Grosso:** Barra do Tapirapés: 1 ind., I-30-31-1964, B. Malkin col. (MZSP); **São Paulo (New record):** Itú: Fazenda Pau D'Alho: 1 ind., I-59, col. U. Martins, W. O. Sousa det. (DZUP); **Paraná (New record):** Curitiba: 4 ind., XII-1935, 103 (MZSP). **Paraguay: Misiones:** Villa Florida: 1 male (dissected), IV-17-1988, Colr. C. Aguilar, W. O. Sousa det. (DZUP); **San Ignacio,** Tahyity: 1 ind., II-20-22-2005, B. Garcete col., W. O. Sousa det. (DZUP).

***Neapion (Neotropion) americanum* (Wagner, 1908)**

Synonyms: *Apion americanum* Wagner, 1908:5; *Apion conicicollae* Sharp, 1890:65 (not Gerstaecker 1854:270) [Wagner, 1908:5].

Distribution: **Mexico, Belize, Guatemala, Panama, Costa Rica, Venezuela, Brazil (Goiás: Jataí).**

References: Wagner 1910:11, 1912c:117; Blackwelder 1947:828; Kissinger 1963a:156, 1968:41, 42, 1990:37; O'Brien and Wibmer 1982:20; Wibmer and O'Brien 1986:40.

***Neapion (Neotropion) diringsi* De Sousa & Ribeiro-Costa, 2018**

Distribution: **Brazil (Amazônias: Itacoatiara).**

Type material: Brazil, Itacoatiara, Amazonas, Dirings, Januare 1965 (Holotype) (MZUSP). Paratype: 4 females, 1 with genitalia in a separate microvial (MZUSP), glued on paper triangle, same holotype data, 1 female (MZUSP), glued on paper triangle, same holotype data except III/1964.

Material examined: Brazil, Itacoatiara, Amazonas, Dirings, Januare 1965, 1 male without head, glued on paper triangle, with genitalia in a separate microvial (MZUSP).

***Neapion (Neotropion) marquesae* De Sousa & Ribeiro-Costa, 2018**

Distribution: **Brazil (Mato Grosso: Poconé).**

Type material: Brasil, Mato Grosso: Poconé, (Canopy), 25/10/2012, Bonatti, J., seca, Cord. 04, Funil 50, Quadrante F2 (Holotype) (DZUP). Paratype: 1 female dissected (DZUP), same holotype information except Cord. 3, Funil 44, 1 female (LETA), same holotype information except 05/09/2013, Funil 36.

***Neapion (Neotropion) peculiare* (Wagner, 1909)**

Synonyms: *Apion peculiare* Wagner, 1909:766; *Apion gibbosum* Sharp, 1891a:82 (not Herbst 1797:103, not Faust 1887) [Wagner, 1909:766].

Distribution: **Costa Rica, Panama, Brazil (Goiás: Jataí).**

References: Wagner 1912c:117; Blackwelder 1947:830; Kissinger 1963a:158, 1968:41, 44, 1990:38; O'Brien and Wibmer 1982:26; Wibmer and O'Brien 1986:42.

**Subtribe Piezotrachelina Voss, 1959a:51**

**Genus *Chrysapion* Kissinger, 1968:23**

Type-species: *Apion auctum* Sharp, 1890:78.

Distribution: **North, Central, and South America (Colombia, Venezuela and Brazil (New record)), Antilles.**

Host plants: Fabaceae.

References: Kissinger 1968:23, 1988:302; O'Brien and Wibmer 1982:19; Wibmer and O'Brien 1986:38; Alonso-Zarazaga 1990:79, 88, 2004:696; Alonso-Zarazaga and Lyal 1998:59.

Remarks: images examined of the material determined by D. G. Kissinger of the species *Chrysapion auctum* (Sharp, 1890) and *Chrysapion tantillum* (Sharp, 1890:55) of the Arizona State University Hasbrouck Insect Collection and (<http://symbiota4.acis.ufl.edu/scan/portal/>).

Material examined: *Chrysapion* undetermined species. **Brazil (New record): Ceará:** Barbalha: 10 ind., V-1969, M. Alvarenga, Coleção M. Alvarenga, W. O. Sousa det. (DZUP), Crato, S. Araripe: 17 ind. V-1969, M. Alvarenga, Coleção M. Alvarenga, W. O. Sousa det. (DZUP); **Rio Grande do Norte:** Natal: 7 ind., X-1951, M. Alvarenga col., Coleção M. Alvarenga, W. O. Sousa det. (MZSP); **Pernambuco:** Petrolina: 3 ind., V-1969, M. Alvarenga, Coleção M. Alvarenga, W. O. Sousa det. (DZUP), Caruaru: 2 ind., IV-1972, M. Alvarenga, Coleção M. Alvarenga, W. O. Sousa det. (DZUP).

**Subtribe Oxystomatina Alonso-Zarazaga, 1990:110**

**Genus *Coelocephalopion* Wagner, 1914:145**

Type-species: *Apion bryanti* Wagner, 1914:147, by subsequent designation of Kissinger 1968:29.

Distribution: **North, Central and South America (Colombia, Venezuela, Trinidad, Brazil, Bolivia, Chile and Argentina), Antilles. Introduced in Thailand and Australia.**

Host plants: Asteraceae, Euphorbiaceae, Fabaceae.

References: Blackwelder 1947-828, Kissinger 1968:29, 151, 257, 258, 1988:303, 1992:66; O'Brien and Wibmer 1982:20; Wibmer and O'Brien 1986:39; Alonso-Zarazaga 1990:88, 2004:696; Forno *et al.* 1994:147; Alonso-Zarazaga and Lyal 1999:61; Vergara-Pineda *et al.* 2014.

Material examined: *Coelocephalopion* undetermined species.

**Brazil: Pernambuco:** Caruaru: 3 ind., IV-1972, M. Alvarenga, Coleção M. Alvarenga, W. O. Sousa det. (DZUP); **Mato Grosso (New record):** Claudia: 1 male, 3 female, IV-2015, Mod. I, Gap. 35, 11°35'45.0" S, 55°16'59.0" W, 1 male, Gap. 21, 11°36'07.5" S, 55°15'59.0" W, 1 female, Mod. II, Gap. 12, 11°25'43.3" S, 55°18'52.0" W, Puça, Vicente R. E., W. O. Sousa det. (UFMT); **Mato Grosso do Sul (New record):** Corumbá, Porto Esperança: 1 ind., XII-7-1960, K. Lenko col., *Apion nodicorne* grp., det Kissinger (MZSP); **Rio de Janeiro:** Tijuca, Esc. Nac. Agr.: 2 ind. (1 damaged), IV-24- 1935, Charles Hathamay, 11.305, *Apion* sp. in *nodicorne* grp. (MZSP), Represa Rio Grande, Guanabara: 1 ind., VII-1972, 3 male, VII-1972, F. M. Oliveira, Coleção M. Alvarenga, W. O. Sousa det. (DZUP); **Paraná:** Guarapuava, Est. Águas Claras: 1 ind., I-02-1987, Lev. Ent. PROFAUPAR LAMPADA, Ponta Grossa: 1 ind. VII-20-87, Prof. Mal., W. O. Sousa det. (DZUP).

***Coelocephalopion atirostre* (Fabricius, 1802)**

Synonyms: *Atelabus atirostre* Fabricius, 1802:424; *Apion luteirostre* Gerstaecker, 1854:254 [Alonso-Zarazaga, 2014:75]; *Apion acarinum* Sharp 1890:64 [Wagner 1912a:30]; *Apion argentinum* Béguin-Billecocq 1909:464 [Wagner 1912a:30]; *Apion cydoniae* Kissinger 1957:40, error [not Bondar, 1950:454].

Distribution: **Mexico, Guatemala, Panama, Colombia, Venezuela, Brazil (Rio de Janeiro, Santa Catarina), Surinam, Bolivia, Argentina.**



References: Wagner 1910:8; Blackwelder 1947:829; Kissinger 1959a:30, 1968::196, 210; O'Brien and Wibmer 1982:24; Wibmer and O'Brien 1986:42.

Type material: 1 ind., *Apion argentinum* L. B. B. Rep. Argentina, Prov. Buenos Aires, VII-09-1905, C Bruch (Holotype) (MACN).

Material examined: **Brazil: Rio de Janeiro:** Represa Rio Grande, Guanabara: 1 ind., VII-1962, F. M. Oliveira, Coleção M. Alvarenga, W. O. Sousa det. (DZUP).

***Coelocephalapion brachyrrhinum* (Wagner, 1914)**

Synonyms: *Apion brachyrrhinum* Wagner, 1914:150.

Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:828; Kissinger 1968:168; Wibmer and O'Brien 1986:40.

Remarks: lost type material (Kissinger 1968:168).

***Coelocephalapion coelocephalum* (Wagner, 1914)**

Synonyms: *Apion coelocephalum* Wagner, 1914:146.

Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:828; Kissinger 1968:168; Wibmer and O'Brien 1986:40.

Remarks: lost type material (Kissinger 1968:168).

***Coelocephalapion dissimilipes* (Sharp, 1890:49)**

Synonyms: *Apion dissimilipes* Sharp, 1890:49.

Distribution: **Mexico, Guatemala, Panama, Brazil (Pernambuco e Bahia).**

References: Wagner 1912a:32; Blackwelder 1947:829; Kissinger 1963b:122, 125, 1968:171, 172; O'Brien and Wibmer 1982:22; Wibmer and O'Brien 1986:41.

***Coelocephalapion longipenne* (Wagner, 1912a:13)**

Synonyms: *Apion longipenne* Wagner, 1912a:13; *Apion cydoniae* Bondar, 1950:454 [Kissinger 1968:195]; *Apion luteirostre* Kissinger 1957:40 [not Gerstaecker, 1854:254].

Distribution: **Mexico, Brazil (Pernambuco e Bahia).**

Host plants: *Croton* sp. (Euphorbiaceae).

References: Blackwelder 1947:829; Kissinger 1959a:30, 1968:195, 210; D'Araujo e Silva 1968:461; O'Brien and Wibmer 1982:24; Vaurie 1953:14; Wibmer and O'Brien 1986:42.

Type material: 6 ind., *Apion cydoniae* Bond., Estado da Bahia, Brazil, G. Bondar Leg. (Cotype) (MNRJ).

***Coelocephalapion rugifrons* (Boheman, 1839)**

Synonyms: *Apion rugifrons* Boheman, 1839:373.

Distribution: **Brazil.**

References: Wagner 1910:55; Blackwelder 1947:830; Kissinger 1968:168, Wibmer and O'Brien 1986:43.

***Coelocephalapion spretissimum* (Sharp, 1890)**

Synonyms: *Apion spretissimum* Sharp, 1890:48.

Distribution: **Mexico, Belize, Costa Rica, El Salvador, Guatemala, Nicaragua, Panama, Brazil (Rio de Janeiro).**

References: Blackwelder 1947:830; Kissinger 1963b:122, 126, 1968:174, 209; O'Brien and Wibmer 1982:27; Wibmer and O'Brien 1986:43.

**Subtribe Trichapiina Alonso-Zarazaga, 1990:116**

**Genus *Apionion* Kissinger, 1998:93**

Type-species: *Apion crassum* Fall, 1898:165.

Distribution: **North, Central and South America (Colombia, Trinidad, Brazil, Peru and Bolivia).**

Host plants: Asteraceae, Euphorbiaceae, Fabaceae.

References: Kissinger 1968:152 (as *annulatum* species group), 1998:93; Alonso-Zarazaga and Lyal 1999:60; Whitehead 1977:165 (as *Coelocephalapion annulatum* species group).

Remarks: examined images of the type of the *Apion neolentum* Kissinger, 1968 available in The Database of the Zoological Collections (CVZBASE) of the Museum of Comparative Zoology - Harvard University was compared with the studied material.

Material examined: *Apionion* undetermined species. **Brazil: Manaus (New record):** Reserva Duke, 26 Km NE Manaus: 1 male, Plot C Malaise 3, IV-95, Barbosa, M. G. V., W. O. Sousa det. (INPA), Rio Solimões, Lago do José, Prox. Manaus: 2 male, VIII-09-1979, fumigação (fogging), J. Adis e outros, W. O. Sousa det. (INPA); **Mato Grosso (New record):** Nossa Senhora do Livramento, Baía dos Coqueiros: 2 female, IX-25-1988, Marinez I. Marques, W. O. Sousa det. (UFMT); **Rio de Janeiro (New record):** Rio de Janeiro, (Corcovado) GB: 2 male, IX-1958, 1 male, X-1958, 1 male, 1 female, XI-1958, 1 male., X-1959, 4 male (1 dissected), 2 female, VIII-1960, Corcovado, D. Federal: 1 male., 1 female, VIII-1958, M. Alvarenga, Ex-coleção M. Alvarenga, W. O. Sousa det. (MZSP).

***Apionion annulatum* (Gerstaecker, 1854)**

Synonyms: *Apion annulatum* Gerstaecker, 1854:256.

Distribution: **Brazil.**

References: Wagner 1910:11, Blackwelder 1947:828, Kissinger 1968:152, 1998:93; Wibmer and O'Brien 1986:40.

***Apionion faldermanni* (Gyllenhal, 1839)**

Synonyms: *Apion faldermanni* Gyllenhal, 1839:370.

Distribution: **Brazil.**

References: Wagner 1910:27; Blackwelder 1947:829; Kissinger 1968:152; Wibmer and O'Brien 1986:41.

***Apionion samson* (Sharp, 1891)**

Synonyms: *Apion samson* Sharp, 1891:84.

Distribution: **Belize, Panamá, Bolivia, Brazil.**

References: Kissinger 1968:49; O'Brien and Wibmer 1982:21; Wibmer and O'Brien 1986:43.

Material examined: **Brazil (New record): Minas Gerais:** Rio Verde: 440m, 1 male, Cu, 447, 11.60, W. O. Sousa det. (DZUP).

**Genus *Trichapion* Wagner, 1912b:116**

Type-species: *Apion aurichalceum* Wagner, 1912b:103, by subsequent designation Kissinger 1959b:248.

Distribution: **North, Central and South America (Colombia, Venezuela, Brazil, Peru, Bolivia, Chile, Paraguay, Argentina and Uruguay). Introduced in South Africa.**

Host plants: Asteraceae, Fabaceae.

References: Blackwelder 1947:828, Kissinger 1959a:24, 32, 1959b:247, 1968:49; 1989a:271; O'Brien and Wibmer 1982:20; Wibmer and O'Brien 1986:39; Alonso-Zarazaga 1990:116,

2004:696; Alonso-Zarazaga and Lyal 1999:59; Alonso-Zarazaga and Wanat 2014:399, 405.

Material examined: *Trichapion* undetermined species. **Brazil: Amazonas:** Parque Nacional do Jaú, Rio Carabinani, mg. Dir 1°59'S/61°32'W: 1 male, IV-06-07-1994, Motta, C, *et al.* Col., Luz mista mercúrio Luz negra BL e BLB Lençol, W. O. Sousa det. (INPA), Manaus, Reserva Duke, 26 Km NE Manaus: 1 male, Plot C Malaise 4, 1 female, Plot C Malaise 1, 1 female, Plot C Malaise 5, 1 female, 372, 0031505, Plot C Malaise 4, VI-95, 1 female, 372, 0031506, 1 female, 372, 0031507, Plot C Malaise 4, V-95, Barbosa, M. G. V., W. O. Sousa det. (INPA).

#### *Trichapion acanthopus* (Wagner, 1912)

Synonyms: *Apion acanthopus* Wagner, 1912c:110.

Distribution: **Brazil (Amazonas), Bolivia.**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:39.

#### *Trichapion fusconitidum* (Wagner, 1912)

Synonyms: *Apion fusconitidum* Wagner, 1912a:23.

Distribution: **Brazil (Rio de Janeiro: Petrópolis).**

References: Blackwelder 1947:829; Kissinger 1959b:250, 332, 338; Wibmer and O'Brien 1986:41.

#### *Trichapion lativentre* (Béguin-Billecocq, 1909)

Synonyms: *Apion lativentre* Béguin-Billecocq, 1909:451.

Distribution: **Brazil (Rio de Janeiro), Paraguay, Argentina, Uruguay. Introduced in South Africa.**

References: Wagner 1912a:32; Blackwelder 1947:829; Kissinger 1959b:250, 369, 374; Wibmer and O'Brien 1986:41; Alonso-Zarazaga 1990:148; Bachmann and Lanteri 2013:210; Alonso-Zarazaga and Wanat 2014:339, 405.

Type material: 1 ind., *Apion lativentre* tipo L. B. B, Rep. Argentina, Prov. Buenos Aires, III-1897, C Bruch (Syntype) (MACN).

Material examined: **Brazil: Paraná (New record):** Curitiba: 6 male, IV-12-1980, 13 male (1 dissected), 19 female (1 dissected), XII-5-1980, Rosado Neto col., W. O. Sousa det. (DZUP).

#### *Trichapion longicorne* (Wagner, 1912)

Synonyms: *Apion longicorne* Wagner, 1912c:107.

Distribution: **Brazil (Amazonas), Bolivia.**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

#### *Trichapion spinitarse* (Wagner, 1912)

Synonyms: *Apion spinitarse* Wagner, 1912a:18.

Distribution: **Nicaragua, Brazil (Rio de Janeiro).**

References: Blackwelder 1947:830; Kissinger 1959b:250, 254, 321, 1968:68, 70, 74; Wibmer and O'Brien 1986:43.

#### *Trichapion suffusum* (Wagner, 1912)

Synonyms: *Apion suffusum* Wagner, 1912b:113.

Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

### **Incertae sedis genera**

#### **Genus *Bothryopteron* Wagner, 1912a:8**

Type-species: *Apion grallarium* Sharp, 1891a:83, by subsequent designation Kissinger 1968:29 (examined images of the material available in Kissinger (1968)).

Distribution: **North, Central and South America (Brazil and Bolivia).**

Host plants: unknown.

References: References: Kissinger 1959a:26, 28, 1968:32, 243; O'Brien and Wibmer 1982:20; Wibmer and O'Brien 1986:39; Alonso-Zarazaga 2004:696; Alonso-Zarazaga and Lyal 1998:61.

Material examined: *Bothryopteron* undetermined species.

**Brazil: Maranhão (New record):** Igarape Gurupi - Uma aldeia

Araçú: 1 ind., V-1968, B. Malkin col., W. O. Sousa det. (MZSP);

**Mato-Grosso (New record):** Utariti (325 m), Rio Papagaio:

1 ind., CII-VIII-961, K. Lenko col., W. O. Sousa det. (MZSP),

Chapada dos Guimarães, Chapada Aventura: 1 ind., VIII-23-

2016, UmbrellaEnt, A. O. Lira leg., W. O. Sousa det. (LETA), Rio

Caraguata: 3 ind., III-1953, F. Plaumann, Coleção Campos Seabra,

W. O. Sousa det. (MNRJ); **Tocantins (New record):** Dianópolis:

1 ind., I-16-22-1962, J. Bechyné col., W. O. Sousa det. (MZSP);

**Goiás (New record):** Pirineus: 1 ind., II-2-1962, J. Bechyné col.,

W. O. Sousa det. (MZSP); **Rio de Janeiro:** Rio de Janeiro, Recreio

dos Bandeirantes, Baía da Guanabara: 2 ind., dz 48/60, V-8-60, J.

C. M. Carvalho, W. O. Sousa det. (MNRJ); **Santa Catarina (New**

**record):** Nova Teutônia: 1 ind., 27° 11' B. 52° 23' L, XI-1953, 1 ind.

I-1954, Fritz Plaumann col., W. O. Sousa det., (MZSP), 1 ind., XII-

1976, 300-500 m, Fritz Plaumann col., W. O. Sousa det. (DZUP).

#### *Bothryopteron balzani* (Wagner, 1912)

Synonyms: *Apion balzani* Wagner, 1912a:9.

Distribution: **Brazil (Rio de Janeiro), Bolivia.**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

#### *Bothryopteron binodosum* (Wagner, 1912)

Synonyms: *Apion binodosum* Wagner, 1912a:10.

Distribution: **Brazil (Rio de Janeiro, Bahia).**

References: Wagner 1912c:117; Wibmer and O'Brien 1986:40.

Material examined: **Brazil: Rio de Janeiro:** Represa Rio Grande,

Guanabara: 1 ind., X-1960, Coleção Campos Seabra, F. M. Oliveira,

W. O. Sousa det. (MNRJ), 1 ind., IX-1961, Coleção M. Alvarenga,

F. M. Oliveira, W. O. Sousa det. (DZUP).

#### *Bothryopteron praestabile* (Wagner, 1914)

Synonyms: *Apion praestabile* Wagner, 1914:159.

Distribution: **Brazil.**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

#### *Bothryopteron steinbachi* (Wagner, 1912)

Synonyms: *Apion steinbachi* Wagner, 1912a:11.

Distribution: **Brazil (Amazonas), Bolivia.**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

**Genus *Coelopterapion* Wagner, 1912a**

Type-species: *Apion testaceum* Wagner, 1912a:3, by subsequent designation Kissinger 1968:29.

Distribution: **South America (Brazil and Paraguay).**

Host plants: Euphorbiaceae.

References: References: Lucas 1920:196; Kissinger 1968:33; O'Brien and Wibmer 1982:20; Wibmer and O'Brien 1986:39; Alonso-Zarazaga 2004:696; Alonso-Zarazaga and Lyal 1998:61.

Material examined: *Coelopterapion* undetermined species. **Brazil: Mato Grosso (New record):** Rio Caraguata: 1 ind., VI-1953, 218° 48'B, 52° 27'W, 400 m, Fritz Plaumann, W. O. Sousa det. (MZSP); **Minas Gerais (New record):** Pouso Alegre: 3 ind., XII-1965, F. S. Pereira col. Sousa, W. O., det. (MZSP); **Espirito Santo:** Sta. Tereza: 1 ind., XII-7-64, C. Elias leg. W. O. Sousa det. (DZUP); **São Paulo (New record):** Botucatu: 3 ind. III-1967, Dirings, W. O. Sousa det. (MZSP), Barueri: 1 ind., X-15-1954, 1 ind., I-22-1961, 1 ind., IV-9-1964, K. Lenko col., W. O. Sousa det. (MZSP), São Paulo, Cantareira: 1 ind. II-17-1960, J. Halik, 3626, W. O. Sousa det. (MZSP), Ferraz de Vasconcelos: 1 ind., XI-19-1971, J. J. Ferracioli, 3117, Coleção Ferracioli, W. O. Sousa det. (MZSP), Salesópolis, E. B. da Boracéia: 1 ind., X-2007, Col. Uehara M., W. O. Sousa det. (UFMT), Ilha dos Búzios: 1 ind., X-16-XI-04-963, Exp. Dep. Zool., W. O. Sousa det. (MZSP); **Rio de Janeiro:** Itatiaia, Est. Biológica: 1 ind., XI-29-32, 700m, W. Zikán, W. O. Sousa det. (MNRJ); Rio de Janeiro: Realengo, Guanabara: 1 ind., X-9-1949, M. Alvarenga leg., Ex-coleção M. Alvarenga, W. O. Sousa det. (MZSP), Corcovado, Rio, Guanabara: 1 male (dissected), IX-1958, Alvarenga e Seabra, Ex-coleção M. Alvarenga, W. O. Sousa det. (MZSP), (Corcovado) GB: 1 ind., X-1958, M. Alvarenga, Ex-coleção M. Alvarenga, W. O. Sousa det. (MZSP), Inst. Biolog. Entomologia Agrícola: 1 ind., III-1930, coll Dario Mendes, W. O. Sousa det. (MNRJ); **Paraná (New record):** Londrina: 1 ind., II-14-75, Pe. Moure, Rosado leg. 1 ind., XII-6-8-75, Pe. Moure leg., W. O. Sousa det. (DZUP), Jundiá do Sul, Fazenda Monte Verde: 1 ind., X-5-1986, Lev. Ent. PROFAUPAR, Lampada, W. O. Sousa det. (DZUP).

***Coelopterapion cionoides* (Wagner, 1912)**

Synonyms: *Apion cionoides* Wagner, 1912a:5.

Distribution: **Brazil (Rio de Janeiro, Pernambuco).**

Host plants: *Croton floribundus* (Euphorbiaceae).

References: Blackwelder 1947:828; D'Araujo e Silva 1968:461; Kissinger 1968:33; Wibmer and O'Brien 1986:40.

Material examined: **Brazil: São Paulo (New record):** Barueri: 1 ind., X-7-1954, 97, leg K. Lenko, det. DKiss 1957 (MZSP).

***Coelopterapion fuscipenne* (Wagner, 1912)**

Synonyms: *Apion fuscipenne* Wagner, 1912a:7.

Distribution: **Brazil (Bahia).**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Coelopterapion globulum* (Wagner, 1912)**

Synonyms: *Apion globulum* Wagner, 1912a:6.

Distribution: **Brazil (Rio de Janeiro, Bahia).**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Coelopterapion subulirostre* (Gyllenhal, 1833)**

Synonyms: *Apion subulirostre* Gyllenhal, 1833:266; *Apion daimio* Sharp, 1891b:296 [Kissinger 1989b:24]; *Apion fryi* Wagner, 1912a:4 [Kissinger 1989b:24].

Distribution: **Brazil (Pernambuco, Rio de Janeiro).**

References: Wagner 1910-61; Blackwelder 1947-829, 830; Wibmer and O'Brien 1986-43.

***Coelopterapion testaceum* (Wagner, 1912)**

Synonyms: *Apion testaceum* Wagner, 1912a:3.

Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:830; Kissinger 1968:29, 33; Wibmer and O'Brien 1986:43.

Material examined: **Brazil: São Paulo (New record):** Barueri: 1 ind., X-14-1955, K. Lenko col., det Kissinger (MZSP).

**Genus *Mythapion* Kissinger, 2005a:85**

Type-species: *Mythapion trifolium* Kissinger, 2005a:86.

Distribution: **South America (Brazil, Chile, Argentina and Uruguay).**

Host plants: unknown.

References: Kissinger 2005a:85.

Material examined: *Mythapion* undetermined species. **Brazil: Amazonas (New record):** Manaus: ZF-2. Km-19: 1 male, VIII-18-1979, J. Adis e outros, fumigação (fogging), W. O. Sousa det. (INPA).

***Mythapion simplex* (Béguin-Billecocq, 1909)**

Synonyms: *Apion simplex* Béguin-Billecocq, 1909:459.

Distribution: **Brazil (Bahia, Rio de Janeiro), Argentina, Uruguay.**

References: Wagner 1912a:31; Blackwelder 1947:830; Kissinger 2005a:71, 86, 88; Wibmer and O'Brien 1986:43; Bachmann and Lanteri 2013:210.

Material examined: **Argentina:** Buenos Aires: 1 ind., I, 2-y2, 373, Coleção F. Justus Jor. (DZUP), 2 ind., II-1942, Dirings (MZSP).

**Genus *Stenapion* Wagner, 1912a:20**

Type-species: *Apion constricticollis* Sharp, 1890:59, by subsequent designation Kissinger 1968:29 (examined images of the material available in Kissinger (1968)).

Distribution: **North, Central and South America (Trinidad, Brazil, Peru, Bolivia, Paraguay and Argentina).**

Associated plants: Polygonaceae.

References: Wagner 1915:35; Kissinger 1959a:23, 32, 1968:29, 39, 127; O'Brien and Wibmer 1982:20; Wibmer and O'Brien 1986:39; Alonso-Zarazaga 2004-696; Alonso-Zarazaga and Lyal 1999:61.

Type material: 1 ind., *Apion salpingoides* = *mediocre* L. B. B, Rep. Argentina, Prov. Buenos Aires, II-2-1905, C Bruch (Syntype), 1 ind., *Apion contrarium* L. B. B, Rep. Argentina, Prov. Buenos Aires, XI-27-1904, C Bruch (Syntype) (MACN).

Material examined: *Stenapion* undetermined species. **Brazil: Pará (New record):** Camargo-Itaituba, Rio Tapajós: 1 ind., XI-1963, Dirings, W. O. Sousa det. (MZSP); **Mato-Grosso (New record):** Poconé: Pirizal: 4 ind., Armadilha luminosa, Cambará-Borda,

I-15-2000, W. O. Sousa col., W. O. Sousa det. (LETA), Baía do Coqueiro: 1 ind. X-2000 P.4, Gaiola de ADIS, W. O. Sousa col., W. O. Sousa det. (LETA), Barão de Melgaço: 1 ind., Baía/P03 Litorânea, IX-02-09, Brizzola and Sousa col., W. O. Sousa det. (LETA); **Mato Grosso do Sul (New record)**: Costa Rica: 1 ind., XII-18-87, F. Sta Cruz. Exp. IQ. MZ DZIUUSP, W. O. Sousa det. (MZSP); **Minas Gerais (New record)**: Santa Barbara, Serra da Caraça: 1 ind. XI-23-25-960, Araujo e Martins (MZSP); **Rio de Janeiro**: Represa Rio Grande, Guanabara: 2 ind., VII-1972, F. M. Oliveira, W. O. Sousa det. (DZUP); **Santa Catarina (New record)**: Nova Teutônia: 2 ind., 27° 11'B. 52° 23'L, XI-1953, Fritz Plaumann col., W. O. Sousa, det. (MZSP), 1 ind., XII-1976, 300-500 m, Fritz Plaumann col., W. O. Sousa, det. (DZUP). **Paraguay: Puerto La Esperanza**: La Victoria, Alto-Paraguay: 1 ind. XI-30-XII-3-2006, B. Garcete col., 79 msnm 22° 01'30,7"S - 50° 01'34,9"W, Malaise trap 1 in dense *Schinopsis-Ruprechtia* forest, W. O. Sousa det. (DZUP); **Asuncion**: 2 ind., I-30-22-1985, col. R. T. Bonace, Bahia de Asuncion vegetation flotante, W. O. Sousa det. (DZUP).

#### ***Stenapion brevinasus* (Wagner, 1912)**

Synonyms: *Apion brevinasus* Wagner, 1912a:20.

Distribution: **Brazil (Pará: Monte Alegre, S. Antonio-Brasília), Paraguay, Argentina, Uruguay.**

References: Wagner 1915:41, 52; Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

#### ***Stenapion carinifrons* (Wagner, 1914)**

Synonyms: *Apion carinifrons* Wagner, 1914:152.

Distribution: **Brazil.**

References: Wagner 1915:39, 59; Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

#### ***Stenapion divergens* (Wagner, 1912)**

Synonyms: *Apion divergens* Wagner, 1912a:21.

Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:829; Wagner 1915:41, 46; Wibmer and O'Brien 1986:41.

#### ***Stenapion heydeni* (Wencker 1863)**

Synonyms: *Apion heydeni* Wencker 1863:181.

Distribution: **Brazil (Monte Alegre), Bolivia, Paraguay, Argentina.**

References: Wagner 1910:33, 1915:40, 52; Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

#### ***Stenapion yatahyanum* (Wagner, 1912)**

Synonyms: *Apion yatahyanum* Wagner, 1912c:116.

Distribution: **Brazil (Goiás: Jataí).**

References: Wagner 1915:38, 59; Blackwelder 1947:829; Wibmer and O'Brien 1986:44.

#### **Incertae sedis species ("Apion" in the broadest sense)**

##### ***Apion alocorhinum* Wagner, 1914:141**

Distribution: **Brazil (Goiás: Jataí).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion ampullula* Gerstaecker 1854:255**

Synonyms: *Apion macromerum* Kirsch, 1874:419 [Wagner 1912a:31].

Distribution: **Peru, Brazil.**

References: Wagner 1910:11; Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion angulatum* Gerstaecker 1854:245**

Distribution: **Brazil (Goiás: Jataí).**

References: Wagner 1910:11; Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion angustifrons* Wagner, 1914:162**

Distribution: **Brazil (Rio de Janeiro: Teresópolis).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion areolatum* Kirsch, 1874:423**

Distribution: **Peru, Brazil (Amazonas, Rio de Janeiro).**

References: Wagner 1912a:32, Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

Remarks: examined images of the type of the *A. areolatum* deposited in the Staatliches Museum für Tierkunde, Dresden.

##### ***Apion atronitidum* Wenker, 1863:178**

Distribution: **Brazil (São Paulo).**

References: Wagner 1910:14; Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion bicoloratum* Wagner, 1914:138**

Distribution: **Brazil (Pernambuco).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion brachypterum* Wagner, 1912b:130**

Distribution: **Brazil (Amazonas, Rio de Janeiro).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion brasilianum* Wagner, 1912a:14**

Distribution: **Brazil (Pernambuco, Rio de Janeiro).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion brevicorne* Gerstaecker 1854:249**

Distribution: **Brazil.**

References: Wagner 1910:15; Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

Material examined: **Brazil: Rio de Janeiro**, Tijuca, Esc. Nac. Agr., Brasil Central: 1 ind., IX-1945, C. de Araujo, 11.308, det. Kissinger (MZSP).

##### ***Apion brevipes* Wagner, 1912a:25**

Distribution: **Brazil (Pernambuco).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.

##### ***Apion coeruleosquamosum* Wagner, 1912b:131**

Distribution: **Peru, Bolivia, Brazil (Amazonas).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:40.



***Apion convergens* Wagner, 1912a:28**Distribution: **Brazil (Pernambuco, Rio de Janeiro).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:41.

***Apion convexirostre* Wagner, 1912b:135**Distribution: **Brazil (Amazonas).**

References: Blackwelder 1947:828; Wibmer and O'Brien 1986:41.

***Apion dentinasus* Wagner, 1912b:133**Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion desbordesii* Béguin-Billecocq, 1911:131**Distribution: **Brazil (Ceará), Argentina.**

References: Wagner 1912a:31; Blackwelder 1947:829; Wibmer and O'Brien 1986:41; Bachmann and Lanteri 2013:209.

Material examined: Syntype: 1 female, *Apion desbordesii* L. B. B, Rep. Argentina, Prov. Cordoba, 190, C Bruch (MACN).***Apion ensirostre* Gerstaecker, 1854:251**Synonyms: *Apion columbianum* Faust, 1894:319 [Wagner 1912a:31].Distribution: **Colombia, Bolivia, Brazil.**

References: Wagner 1910:19, 1912c:118; Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

Material examined: **Brazil: Mato Grosso do Sul (New record):** Corumbá, Porto Esperança: 1 ind., XII-7-1960, K. Lenkko col., 129, det. Kissinger (MZSP).***Apion exophthalmum* Wencker, 1863:179**Distribution: **Brazil.**

References: Wagner 1910:26; Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion fulvicorne* Wagner, 1912a:29**Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion gilvirostre* Wagner, 1912a:26**Distribution: **Brazil (Amazonas).**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion inaequale* Gyllenhal, 1833:256**Distribution: **Brazil.**

References: Wagner 1910:34; Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion informe* Béguin-Billecocq, 1911:131**Distribution: **Bolivia, Brazil (Rio de Janeiro).**

References: Wagner 1912a:32, 1912c:117; Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion ingratum* Béguin-Billecocq, 1909:461**Distribution: **Brazil (Amazonas), Argentina.**

References: Wagner 1912a:32; Blackwelder 1947:829; Wibmer and O'Brien 1986:41; Bachmann and Lanteri 2013:210.

Type material: 1 ind., *Apion ingratum* L. B. B, Rep. Argentina, Prov. Buenos Aires, 190, C. Bruch (Syntype) (MACN).***Apion latifrons* Gerstaecker, 1854:246**Distribution: **Brazil.**

References: Wagner 1910:37; Blackwelder 1947:829; Wibmer and O'Brien 1986:41.

***Apion lividipes* Wencker, 1863:180**Distribution: **Brazil.**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

***Apion longitarse* Wagner, 1914:141**Distribution: **Brazil.**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

***Apion longulum* Gerstaecker, 1854:239**Distribution: **Brazil.**

References: Wagner 1910:39; Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

***Apion luridipes* Wagner, 1912a:24**Distribution: **Brazil.**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

***Apion luteipes* Kirsch, 1874:419**Distribution: **Peru, Brazil.**

References: Wagner 1910:39; Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

Remarks: examined images of the type of the *A. luteipes* deposited in the Staatliches Museum für Tierkunde, Dresden.***Apion macrostylum* Wagner, 1914:144**Distribution: **Brazil (Rio de Janeiro: Rio de Janeiro; Minas Gerais: Matusinho, Caraça).**

References: Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

***Apion nigroaeneum* Gerstaecker, 1854:255**Distribution: **Brazil.**

References: Wagner 1910:43; Blackwelder 1947:829; Wibmer and O'Brien 1986:42.

***Apion nigrosuturatum* Béguin-Billecocq, 1909:459**Distribution: **Brazil (São Paulo), Argentina.**

References: Wagner 1912:32; Blackwelder 1947:829; Wibmer and O'Brien 1986:42.



***Apion novaeteutoniae* Voss, 1959b:315 [in footnote]**Distribution: **Brazil (Nova Teotonia).**

References: Weidner 1976:147; Wibmer and O'Brien 1986:42.

Remarks: examined images of the holotype, 1 female of the *A. novaeteutoniae* deposited in the Zoological Museum, Hamburg.***Apion opacum* Kirsch, 1874:420**Distribution: **Peru, Brazil.**

References: Wagner 1910:45; Blackwelder 1947:830; Wibmer and O'Brien 1986:42.

Remarks: examined images of the type of the *A. opacum* deposited in the Staatliches Museum für Tierkunde, Dresden.***Apion parviceps* Wagner 1914:160**Distribution: **Brazil.**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:42.

***Apion parvulum* Gerstaecker, 1854:248**Synonyms: *Apion motabile* Faust, 1894:317 [Wagner, 1910:47].Distribution: **West Indies, Colombia, Venezuela, Brazil.**

References: Wagner 1910:47, 1912a:31; Blackwelder 1947:830; Wibmer and O'Brien 1986:42.

***Apion periscelis* Gyllenhal, 1839:391**Synonyms: *Apion strangulatum* Kirsch, 1874:413 [Wagner, 1910:47].Distribution: **Peru, Bolivia, Brazil.**

References: Wagner 1910:47, 1912a:31; Blackwelder 1947:830; Wibmer and O'Brien 1986:42.

Material examined: **Brazil: Minas Gerais:** Pouso Alegre: 1 ind., IX-962, F. S. Pereira col., 139, det. Kissinger (MZSP).***Apion phaseoli* Voss, 1958:921**Distribution: **Brazil (Bahia).**

References: Weidner 1976:147; Wibmer and O'Brien 1986:43.

Remarks: examined images of the paratype of the *A. phaseoli* deposited in the Zoological Museum, Hamburg.***Apion pseudampullula* Voss, 1940:12**Synonyms: *Apion pseudampulla* [Bondar 1950:452, error].Distribution: **Brazil.**

Host plants: Leguminosae.

References: D'Araujo e Silva 1968:461; Wibmer and O'Brien 1986:43.

Material examined: 2093: 2 ind., Voss (MNRJ).

***Apion pulverulentum* Wencker, 1863:177**Distribution: **Brazil.**

References: Wagner 1910:51; Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

***Apion pygmaeum* Wagner, 1912a:29**Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

***Apion retusipenne* Wagner, 1912a:28**Distribution: **Brazil.**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

***Apion rubrirostre* Wagner, 1912a:13**Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

***Apion rubronigrum* Wagner, 1912b:132**Distribution: **Brazil (Bahia, Rio de Janeiro).**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

***Apion sesbaniae* Voss, 1958:919**Distribution: **Brazil (Rio Grande do Sul: Porto Alegre).**

References: Weidner 1976:147; Wibmer and O'Brien 1986:43.

Remarks. Examined images of the paratype, 1 female of the *A. sesbaniae* deposited in the Zoological Museum – Hamburg.***Apion subnudum* Wagner, 1912a:21**Distribution: **Brazil (Goiás: Jataí), Bolivia.**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:43.

***Apion tropidorrhynchum* Wagner, 1914:143**Distribution: **Brazil.**

References: Blackwelder 1947:830; Wibmer and O'Brien 1986:44.

***Apion tucumanense* Béguin-Billecocq, 1909:458**Distribution: **Brazil (Rio de Janeiro), Bolivia, Argentina.**

References: Wagner 1912c:118; Blackwelder 1947:830; Wibmer and O'Brien 1986:44; Bachmann and Lanteri 2013:211.

Type material: 1 female, *Apion tucumanense* L. B. B. Rep. Argentina, Prov. Tucuman, I-20-1904, C. Bruch (Syntype) (MACN).***Apion undulipenne* Wagner, 1912a:11**Distribution: **Brazil (Bahia).**Host plants: *Cassia apoucuita* (Fabaceae).

References: Voss 1940:12; Blackwelder 1947:830; D'Araujo e Silva 1968:461; Wibmer and O'Brien 1986:44.

Material examined: **Brazil: Bahia:** 5 ind., 1977, G. Bondar leg., Voss deter., 5 ind., 1977 (MNRJ).***Apion wingelmulleri* Wagner, 1912a:14**Distribution: **Brazil (Rio de Janeiro).**

References: Blackwelder 1947:831; Wibmer and O'Brien 1986:44.

***Apion xanthopus* Gyllenhal, 1839:375**Synonyms: *Apion sulcipenne* Gyllenhal, 1839:401 [Wagner, 1912a:31].Distribution: **Brazil.**

References: Wagner 1910:61,67, 1912a:31; Blackwelder 1947:831; Wibmer and O'Brien 1986:44.

***Apion xanthorhynchum* Wagner, 1912a:27**Distribution: **Brazil.**

References: Blackwelder 1947:831; Wibmer and O'Brien 1986:44.

***Apion zikani* Heller, 1922:52**

Distribution: **Brazil (Minas Gerais: Passa Quatro).**

Host plants: *Dalbergia glaucescens* (Mart. ex. Benth), *D. foliolosa* (Benth) (Fabaceae).

References: Blackwelder 1947:831; Costa-Lima 1956:126; D'Araujo e Silva 1968:461; Wibmer and O'Brien 1986:44.

Type material: 2 ind., *Apion zikani* Heller, Para, S<sup>oc</sup> Dario Mendes, Passa Quatro Minas Zikan (Cotype) (MNRJ).

**Discussion**

We recognize 10 genera of Brazilian Apioninae, one of which is a first record for Brazil (*Chrysapion*) and which appears to be restricted to northeastern Brazil. Three genera are widely distributed in the New World: *Alocentron*, *Coelocephalapion* and *Trichapion*. The latter two genera are found throughout Brazil. Five genera (*Apionion*, *Bothropterion*, *Coelopterapion*, *Neapion* and *Stenapion*) have more restricted distributions in South America (Alonso-Zarazaga and Lyal 1999) but also are found throughout Brazil. *Coelopterapion* is the only genus with most species subendemic to Brazil (Kissinger 1968) and whose distributions are restricted to the Cerrado and Atlantic Forest. The recently described *Mythapion* is monospecific and found in Argentina, Brazil and Uruguay (Wibmer and O'Brien 1986; Kissinger 2005a), from the Amazon to the Pampas of South America.

Only 86 species of Apioninae from Brazil were described from 1833 to 1959, mostly by Hans Wagner, with the recent addition of two new species (De Sousa and Ribeiro-Costa 2018). Yet, Alonso-Zarazaga (pers. comm. 2017) reported that Don Whitehead estimated that the number of Brazilian Apioninae species is closer to 1,500, most of which are endemic. Alonso-Zarazaga and Wanat (2014) suggested 800 species for the Neotropical region, which is likely to be a large underestimation. More than 50 years have passed since the last Brazilian species were described. Thus, our descriptions and keys provide a new landmark for the taxonomy of the Apioninae in South America. Brazil is among the most biodiverse countries with many hotspots (Aleixo *et al.* 2010; Marques and Lamas 2006; Myers *et al.* 2000) and so the Apioninae is a very promising group and much more promising comparing parasitoid hymenopterans wasp or dipterans in South America.

We identified ca 230 especimens to genus or species. Taxonomic impediments, such as brief, incomplete, original descriptions, often without illustrations that would facilitate species recognition, prevent identification at the species level for most. Additionally, many small specimens apparently comprise a large number of morphotypes, but each morphotype included only a few specimens without marked sexual dimorphism. Thus, many specimens remain to be identified.

Upon examination of the Apioninae in Brazilian collections we found a large quantity of unidentified material, many specimens of which are only labelled *Apion* sp., while others were mixed Apioninae. We found no specimen that was identified with any generic name other than *Apion*, and there were no subgenera. Hence the challenge of this study, in which we provide a list for the Brazilian taxa and place them in genera and subgenera. All identification was through comparison with relevant diagnostic characters of the original descriptions, with the addition of currently available types or images of type material or with material previously identified by specialists.

External morphology and genitalia of males, both of which are fundamental for identification of taxa in the Apioninae, are described in Alonso-Zarazaga (1989, 1990, 2004) and Alonso-Zarazaga & Wanat (2014). Also, efficient use of the key requires the separation of morphotypes and identification of the sex of each morphotype. Species identification based only on females is essentially impossible and so males are required. For that end, an adequate collection with many specimens that can be dissected to identify sex is required (Alonso-Zarazaga 2004). If dissection is not possible, secondary sexual dimorphism must be identified and which can be observed in the rostrum (shorter in males, Fig. 18, wider and generally more coarsely sculptured and pubescent than in females, Fig. 19; Anderson & Kissinger 2002). Some males have mucro in at least one pair of tibiae, or the first tarsomere of one of the legs is modified as plantar spines (Fig. 26). The apical margin of ventrite 5 is truncated with a part of the pygidium being visible externally, or the pygidium is formed by the 8th tergite (7th in females, Fig. 24). In females, the legs have no special characters and ventrite 5 (Fig. 25) has a rounded margin (Alonso-Zarazaga 1990). Rarely, ventrite 5 of females (in some species) may have a median glabrous area or a median sulcus (Alonso-Zarazaga and Wanat 2014), such as in *Trichapion* sp. (Fig. 25).

Hundreds of specimens in South American collections are still waiting to be studied, many of them likely to be new taxa. Thus, these specimens may be among the 10 genera comprising the key, or they may be unknown genera or even be among the non-monophyletic hyperdiverse genera (*Trichapion* and *Coelocephalapion*). Thus, they demonstrate the potential of the group for future systematic research at all taxonomic levels. Additionally, studies must be carried out to recover monophyly of the genera and tribes and to clarify relationships of *incertae sedis*, all based on morphology, molecular, biogeography and host plant associations.

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**Author Contributions**

Wesley Oliveira de Sousa: substantial contribution in the concept and design of the study; data collection; analysis and interpretation; manuscript preparation and critical revision, adding intellectual content.

Cibele Stramare Ribeiro-Costa: contribution in the concept and design of the study; manuscript preparation and critical revision, adding intellectual content.

Germano Henrique Rosado-Neto: contribution in the concept and design of the study and critical revision, adding intellectual content.

## Conflicts of interest

The author(s) declare(s) that they have no conflict of interest related to the publication of this manuscript.

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## Odonata of the state of Maranhão, Brazil: Wallacean shortfall and priority areas for faunistic inventories

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**Abstract:** Environmental changes are worrying in a scenario with large knowledge gaps on species diversity and distribution. Many species may become extinct before they are known to science. Considering this scenario, the present study aims to evaluate the known distribution of the species recorded for Maranhão state in Brazilian northeast region and discuss knowledge gaps about Odonata indicating the priority areas for faunistic inventories. Using primary and secondary data together, we present convex minimum polygons of the distribution of all the species registered for the state. In addition, we created maps with the richness of species and number of records of Odonata in the Maranhão state. In primary data sample 269 specimens, represented by 17 genera and 30 species were collected. Of the 30 species collected, 17 are new records for the state of Maranhão; of these, 35.29% are geographically widespread species, occurring in practically all regions of Brazil. Considering the records in the literature, there was a 68% increase in the number of Odonata species known for Maranhão. The most unexplored region is the Cerrado of the state of Maranhão. Furthermore, the transition regions between Cerrado and Amazônia and between Cerrado and Caatinga are also unknown. All these areas are a priority for faunistic inventories.

**Keywords:** Anisoptera, checklist, patterns of diversity, Zygoptera.

## Odonata do estado do Maranhão, Brasil: Déficit wallaceano e áreas prioritárias para inventários faunísticos

**Resumo:** As mudanças ambientais são preocupantes em um cenário com muitas lacunas de conhecimento sobre a distribuição das espécies. Várias espécies podem ser extintas antes mesmo de serem conhecidas pela ciência. Considerando este cenário, o presente estudo tem como objetivo avaliar a distribuição conhecida das espécies de Odonata registradas para o Maranhão, na região nordeste do Brasil e discutir sobre as lacunas de conhecimento sobre Odonata, indicando áreas prioritárias para inventários faunísticos. Usando os dados primários e secundários juntos, nós apresentamos polígonos mínimos convexos da distribuição de todas as espécies registradas para o estado neste estudo. Além disso, criamos mapas com a riqueza de espécies e número de registros de Odonata no Maranhão. Para os dados primários foram coletados 269 indivíduos, representando 17 gêneros e 30 espécies. Das 30 espécies amostradas, 17 são novos registros para o Maranhão; dessas, 35,29% são espécies com ampla distribuição geográfica, ocorrendo em praticamente todas as regiões do Brasil. Considerando os registros na literatura, houve um aumento de 68% no número de espécies conhecidas para o Maranhão. A região mais desconhecida é o Cerrado do Maranhão. Outrossim, a região de transição Cerrado e Amazônia, e a transição entre Cerrado e Caatinga são também desconhecidas. Todas essas áreas são prioritárias para inventários faunísticos.

**Palavras-chave:** Anisoptera, checklist, padrões de diversidade, riachos urbanos, Zygoptera.

## Introduction

Historically, environmental changes have become increasingly frequent and intense, posing a serious problem for the maintenance of biodiversity (Bush et al. 2008). The consequences are more worrying in places where the knowledge about the geographic distribution of the species is restricted or nonexistent (De Marco & Vianna 2005). This gap is called Wallacean shortfall (Hortal et al. 2015). As a result, many species may be extinct before they are known to science (Pimm et al. 2014). These uncertainties make it difficult to define priority areas for conservation due to the lack of knowledge of the true biodiversity.

In Brazil, the geographical distribution of Odonata is poorly known. Despite of increase of collection effort in many regions the geographical distribution still remains poorly known. In details, some species have already been collected or studied in only approximately 29% of the country's total area (Miguel et al. 2017). The Northeast region of the country, which contains parts of the Cerrado, Caatinga and Amazon biomes, is one of its most environmentally diverse regions (Leal et al. 2005; MMA 2009). However, the biodiversity of most organisms is not well known within this region, such as aquatic insects of the orders Odonata (De Marco 2008), Ephemeroptera (Shimano et al. 2013) and Heteroptera (Dias-Silva et al. 2013). In this context, Maranhão state stands out negatively among the states of the northeast region as there are few published works on Odonata records (De Marco 2008), which may be a reflection of the shortage of professionals specialized in aquatic entomology in this region of the country (Shimano et al. 2013; Nessimian et al. 2014), as well as the lack of financial resources for the public promotion of science in Brazil (Gibney 2015).

It is known that Odonates are sensitive to environmental alterations in all of their life stages (Mendes et al. 2017). Due to the fact that they are thermoregulators, characteristics related to their ecophysiology, light, which influences the environmental temperature (De Marco et al. 2015) and environmental integrity (Oliveira-Junior et al. 2017) are very important variables for the distribution of species in tropical streams (Monteiro-Júnior et al. 2014; Brasil et al. 2017). Therefore, total or partial removal of riparian vegetation alters light/temperature levels and the environmental integrity of the streams, and consequently destabilizes the communities of Odonata (Carvalho et al. 2013; Oliveira-Junior et al. 2015, 2017). Furthermore, changes in Odonata communities may adversely affect other communities. The absence of these predators should change the trophic organization of riparian zones and this causes loss of ecological integrity (Samways 1993).

Considering that little knowledge exists about Odonata in the Maranhão state (De Marco 2008; Miguel et al. 2017); which is part of the last Brazilian agricultural frontier called MATOPIBA (an acronym of the names of the states of Maranhão, Tocantins, Piauí and Bahia) where there is intense agricultural pressure on natural landscapes for the expansion of soybean cultivation (Spera et al. 2016), studies of this order within this region are extremely relevant. Considering this scenario, the present study's objective evaluates the known distribution of the species recorded for Maranhão state in Brazilian northeast region and discuss knowledge gaps about Odonata indicating the priority areas for faunistic inventories as a conservation allowance.

## Material and Methods

### 1. Study area

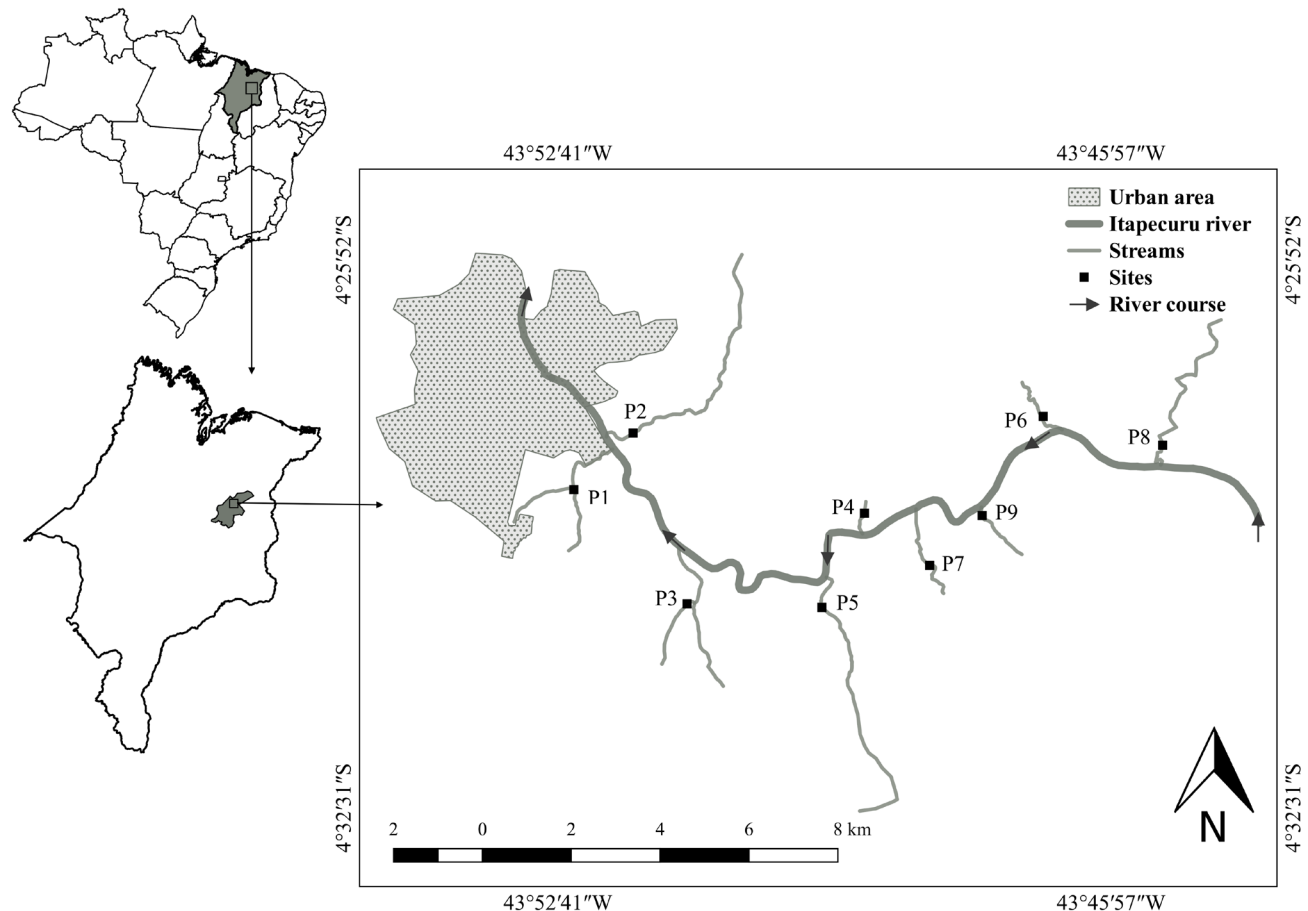
The state of Maranhão has an area of 331.937 km<sup>2</sup>, and includes the phytogeographical domains of Amazonian Forest, Cerrado and Caatinga (Correia-Filho et al. 2011; MMA 2009), according to the Köppen classification system is *Aw* climate type (tropical savanna). This type of climate is primarily characterized by a dry winter (from December to May) and rainy summer (from July to November). In contrast, the state's remaining area, which is predominantly Amazon rainforest, is classified as climate type *Am* (tropical rainy), characterized by a long rainy season throughout the year (Peel et al. 2007; Huete et al. 2006).

The study was carried out in tributaries of the Itapecuru River, in the municipality of Codó, located in the eastern meso-region of Maranhão state, Brazil. This municipality is in a Cerrado-Caatinga transition zone, and has an area of 4,698 km<sup>2</sup> (Feitosa & Almeida 2002). The predominant phytogeographical domain is the Cerrado, presenting Cerrado *stricto sensu* phytophysionomies, composed mainly of shrub vegetation and medium-sized trees such as Cajueiro (*Anacardium occidentale* L.); Cerradão, which presents denser vegetation with larger trees such as Bacuri (*Platonia insignis* Mart.); and riparian forest on the banks of rivers and streams, generally composed of medium and large trees, pioneers, non-pioneers and some shrubs. It also presents the Mata de Cocal formation, where Babaçu (*Orbignya phalerata* Mart.) (Sousa et al. 2016) and Carnaúba [*Copernicia prunifera* (Mill.) H.E. Moore] palm trees predominate. The Mata dos Cocais, which extends from the Amazon in central-west Maranhão to the Caatinga in western Piauí, is a type of transitional vegetation cover between tropical, equatorial and semi-arid climates (Nunes et al. 2012). The region's topography varies between flat and gently undulating with modest altitudes relative to the Brazilian relief, with an average of 47 m (Vasconcelos Gama et al. 2007).

The municipality is drained by the Itapecuru river basin and micro basins of the Codozinho and Saco rivers (Feitosa & Almeida 2002; Sousa et al. 2016). Riparian vegetation of the Itapecuru River tributaries within the study area is mostly characterized by the Riparian Forest physiognomy. However, streams present considerable levels of alteration resulting from the intensive use of soil for crops and pasture, and the use of water resources for food and leisure (Feitosa & Almeida 2002). In general, streams' riparian vegetation is narrow or absent and many small streams are ephemeral, containing running water only during periods of high rainfall (such as from January to April) due to the prolonged dry season, typical of northeast Brazil.

### 2. Primary data collection

Odonata adults were collected from nine tributaries belonging to the Itapecuru river basin in the northeast region of Brazil (Fig. 1). The collects were realized twice in each stream, both being in the 2016 year. The streams are perennial, however, eventually becoming temporary in extreme dry events, common in northeastern Brazil. The first collect was realized in May, and the second in June. Specimens were collected along the margins of each stream using an entomological net, during a one hour collection period along a 100 m section. Sampling at each



**Figure 1.** Location of Odonata collection points in tributaries of the Itapecuru River in Codó, Maranhão, Brazil.

point was always performed on sunny days between 11:00 am and 2:00 pm (Monteiro-Júnior et al. 2015). The collected specimens were conditioned and preserved in accordance with the protocol of Lencioni (2006). For the taxonomic identification of the collected specimens, specific keys were used (Heckman 2006; Lencioni 2005, 2006, 2013; Garrison et al. 2006; Belle 1988, 1996; Pessacq 2014; Garrison & Ellenrieder 2015); when necessary, comparisons were made with specimens already identified and deposited in the collection of the Ecology and Conservation Laboratory, Universidade Federal do Pará; and, expert confirmation.

### 3. Secondary data collection

In order to create species distribution polygons, we used georeferenced distribution data for collected species already used in other studies (Brasil et al. 2018; Calvão et al. 2014), in addition to data that has been compiled from articles, books and museums by De Marco & Vianna (2005) and Juen & De Marco (2012) and recently updated by Martins (2016).

### 4. Data analysis

The occurrence data for species collected during this sampling event was tabulated along with the other records for the Brazil. We then selected only the species that occurred in this study and, subsequently,

the occurrences plotted and geoprocessed in QGIS 2.14.0-Essen (QGIS 2011) software. The occurrence points of each species were used to create the Minimum Convex Polygon (Nilsen et al. 2008). This type of polygon is limited by the outermost points of the occurrence set, graphically demonstrating the probable areas of occurrence of these species.

To define the priority areas for faunal inventories, we used all Odonata records for the state of Maranhão (primary and secondary data) for the preparation of two maps, one with species richness values and another with the number of records. We followed the same method used by De Marco & Vianna (2005), the maps were divided into a grid with 1-degree cells. The maps of the species richness and records were made in the R program using the `lets.presab.points` functions of the `letsR` package (R Core Team 2014; Vilela & Villalobos 2015).

## Results

### 1. Description of communities

A total of 269 specimens represented by 17 genera and 30 species were collected. The suborder Anisoptera presented the highest species richness, with 17 species, representing 56.6% of the total sampled. The suborder Zygoptera presented only 13 species, with 43.4% of the total collected. However, considering the abundance of individuals,

Zygoptera were more representative, with 170 specimens, or 63% of the total collected; while Anisoptera presented only 99 individuals, representing 37% of the total.

The most frequently occurring taxa were *Acanthagrion aepiolum* Tennessen, 2004 found in seven streams (77% of streams), followed by *Acanthagrion kennedii* Williamson, 1916 and *Erythrodiplax basalis* Kirby, 1889 in five streams (56% of the total), *Argia reclusa* Selys, 1865 in four streams (or 44%), and *Perithemis* sp.1 in three streams (or 33%). The remaining taxa represents less than 25% of the total species, whose distribution was restricted to a maximum of two streams (Table 1).

## 2. Spatial distribution of species

Of the 30 Odonata species reported in this study, 17 are new records for the state of Maranhão. Of these new records, 35.29% are geographically widespread species, occurring in practically all regions of Brazil, from the southernmost Cerrado to the northern Amazon. The remaining 64.71% are species with a more restricted geographic distribution, occurring in a limited fashion or in a specific region of the country (Table 1; Fig. 2), however the occurrence of *Micrathyria paruensis* Geijskes, 1963 is the first record of this specie to Brazil.

**Table 1.** List of Odonata species sampled in the study with points of occurrence and known distribution for the Brazilian states, and species already been recorded in other works for the state of Maranhão. Acronym of the Brazilian states: AC = Acre. AL = Alagoas. AM = Amazonas. AP = Amapá. BA = Bahia. ES = Espírito Santos. GO = Goiás. MA = Maranhão. MG = Minas Gerais. MS = Mato Grosso do Sul. MT = Mato Grosso. PA = Pará. PB = Paraíba. PE = Pernambuco. PR = Paraná. RJ = Rio de Janeiro. RO = Rondônia. RR = Roraima. RS = Rio Grande do Sul. SC = Santa Catarina. SP = São Paulo. TO = Tocantins.

Species sampled in the study with known distribution for the Brazilian states and other records for the state of Maranhão.				
Suborder/Species	Points of occurrence	Distribution in Brazil	Other records for Maranhão	Reference
<b>Anisoptera</b>				
<i>Diastatops obscura</i> Fabricius, 1775	P1	AC. AM. AP. BA. ES. GO. MA. MG. MS. MT. PA. PB. PE. PR. RJ. RO. RR. SP and TO	<i>Brachymesia herbida</i> Gundlach, 1889	De Marco (2008)
<i>Erythemis haematogastra</i> Burmeister, 1839	P1	AC. AM. AP. BA. ES. GO. MA. MG. MS. MT. PA. PB. PE and SP	<i>Erythemis plebeja</i> Burmeister, 1839	De Marco (2008)
<i>Erythrodiplax basalis</i>	P1. P3. P4. P6 and P7	AC. AM. GO. MA. MS. MT. PA. PE. PR. RJ. RO. RR. RS and SP.	<i>Erythemis carmelita</i> Williamson, 1923	De Marco (2008)
<i>Erythrodiplax fusca</i>	P4 and P7	AC. AM. BA. ES. PA. PE. RJ. SP. GO. MA. MG. MS. MT. PR. RO. RR. RS and SC	<i>Erythemis peruviana</i> Rambur, 1842	De Marco (2008)
* <i>Erythrodiplax latimaculata</i> Ris, 1911	P7	AM. GO. MG. MS. MT. PE. RJ. RS and SP	<i>Erythemis vesiculosa</i> Fabricius, 1775	De Marco (2008)
<i>Erythrodiplax</i> sp.1	P3	-	<i>Erythrodiplax media</i> Borrer, 1942	De Marco (2008)
* <i>Erythrodiplax umbrata</i>	P2 and P9	AC. AM. AP. BA. ES. GO. PA. PE. PR. MG. MS. MT. RJ. RO. RR. RS and SP	<i>Erythrodiplax paraguayensis</i> Förster, 1905	De Marco (2008)
** <i>Micrathyria artemis</i> Ris, 1911	P7	AM. AP. BA. ES. GO. MG. MS. MT. PA. RJ. RO and SP	<i>Micrathyria marcella</i> Selys, 1857	De Marco (2008)
** <i>Micrathyria divergens</i> Westfall, 1992	P8	MG	<i>Micrathyria ocellata</i> Martin, 1897	De Marco (2008)
** <i>Micrathyria paruensis</i>	P4	-	<i>Nephepeltia phryne</i> Perty, 1834	De Marco (2008)
<i>Micrathyria pseudeximia</i> Westfall, 1992	P1	AM. GO. ES. MA. PA. PR. MG. MS. MT. RJ and RO	<i>Orthemis discolor</i> Burmeister, 1839	De Marco (2008)
* <i>Nephepeltia flavifrons</i> Karsch, 1889	P7	AC. MG. MS. MT. PA. PE. RJ. RR. and SP	<i>Perithemis laís</i> Perty, 1834	De Marco (2008)
<i>Orthemis ambinigr</i> Calvert, 1909	P6	BA. ES. MA. PE. RJ. RS and SC	<i>Zenithoptera anceps</i> Pujol-luz, 1993	De Marco (2008)
<i>Perithemis</i> sp.1	P1. P5 and P7	-	<i>Uracis fastigiata</i> Burmeister, 1839	Costa (1997)
<i>Perithemis</i> sp.2	P7	-	<i>Uracis imbuta</i> Burmeister, 1839	Costa (1997)
** <i>Perithemis thais</i> Kirby, 1889	P1	AM. AP. ES. PA. MS. MT. RJ. RO and SP	<i>Micrathyria mengeri</i> Ris, 1919	Costa (2002)

Continuation Table 1.

Species sampled in the study with known distribution for the Brazilian states and other records for the state of Maranhão.				
Suborder/Species	Points of occurrence	Distribution in Brazil	Other records for Maranhão	Reference
<i>Zenithoptera lanei</i> Santos, 1941	P3 and P4	AC. AM. BA. ES. GO. MA. MS. MT. PA. PE. RJ. RO. SC. SP and TO	<i>Uraxis siemensi</i> Kirby, 1897	Pujol-luz and Fonseca (1997)
<b>Zygoptera</b>				
** <i>Acanthagrion aepiolum</i>	P1. P2. P4. P5. P6. P7 and P8	MS. PA. PR and SP	<i>Ischnura fluviatilis</i> Selys, 1876	De Marco (2008)
** <i>Acanthagrion kennedii</i>	P1. P2. P3. P4 and P7	PA		
** <i>Acanthagrion truncatum</i> Selys, 1876	P8	BA. GO. MG. MS. MT. SP and TO		
* <i>Argia reclusa</i>	P1. P5. P6 and P8	AL. GO. MG. MS. MT. RS and SP		
<i>Argia</i> sp.1	P7	-		
<i>Epipleoneura metallica</i> Rácenis, 1955	P3 and P5	AM. BA. GO. MA. MG. MT. PA and TO		
** <i>Epipleoneura westfalli</i> Machado, 2001	P5	MT. PA and RO		
** <i>Hetaerina curvicauda</i> Garrison, 1990	P5	GO. ES. MS. MT and RO		
<i>Hetaerina sanguinea</i> Selys, 1853	P6 and P8	AM. MA. PA. RO and RR		
* <i>Ischnura capreolus</i>	P8	AC. AM. BA. ES. GO. MG. MS. MT. PA. PE. RJ. RR. RS and SP		
** <i>Neoneura fulvicollis</i> Selys, 1886	P7	GO. PA and SP		
** <i>Perilestes solutus</i> Williamson & Williamson, 1924	P6 and P9	GO. MT. PA. RO and RR		
* <i>Telebasis coccinea</i> Selys, 1876	P3	BA. GO. MS. MT. SP and TO		

\* First record for Maranhão state. \*\* First record for Maranhão state and Northern region.

When we analyze each suborder separately, were found to have similar spatial distributions. The two suborders have records of common species with wide spatial distribution. For instance, *Erythrodiplax umbrata* Linnaeus, 1758 occurs from the southernmost Cerrado to the northern Brazilian Amazon. *Ischnura capreolus*, 1861 Hagen occurs the southernmost Cerrado to the northern Brazilian Amazon.

Analysing the Odonata records to Maranhão state we can see that every state practically has priority for faunal inventories. Furthermore, the knowledge gaps are even greater when we consider the distribution of records by Biomes. This happens because although there are few records it focuses on the Amazon portion. The portion of Cerrado, mainly the east of Maranhão is a region where knowledge about the Odonata fauna is almost non-existent (Figure 3).

## Discussion

### 1. Community description

The species with the highest frequency of occurrence and abundance in this study are known as common organisms in altered environments. For example, *A. aepiolum* has a strong relationship with the loss of native vegetation (Rodrigues et al. 2016). Similarly, *E. basalis* has been shown to be a common species in lentic environments (Calvão et al. 2013), being an excellent indicator of impacted areas (Monteiro-Júnior et al. 2013; Monteiro-Júnior et al. 2015; Oliveira-Junior et al. 2015). *A. reclusa* was also related to altered environments in previous studies (Dutra & De Marco 2015). However, *A. reclusa* was related to environments preserved in the studies of Carvalho et al. (2013) and



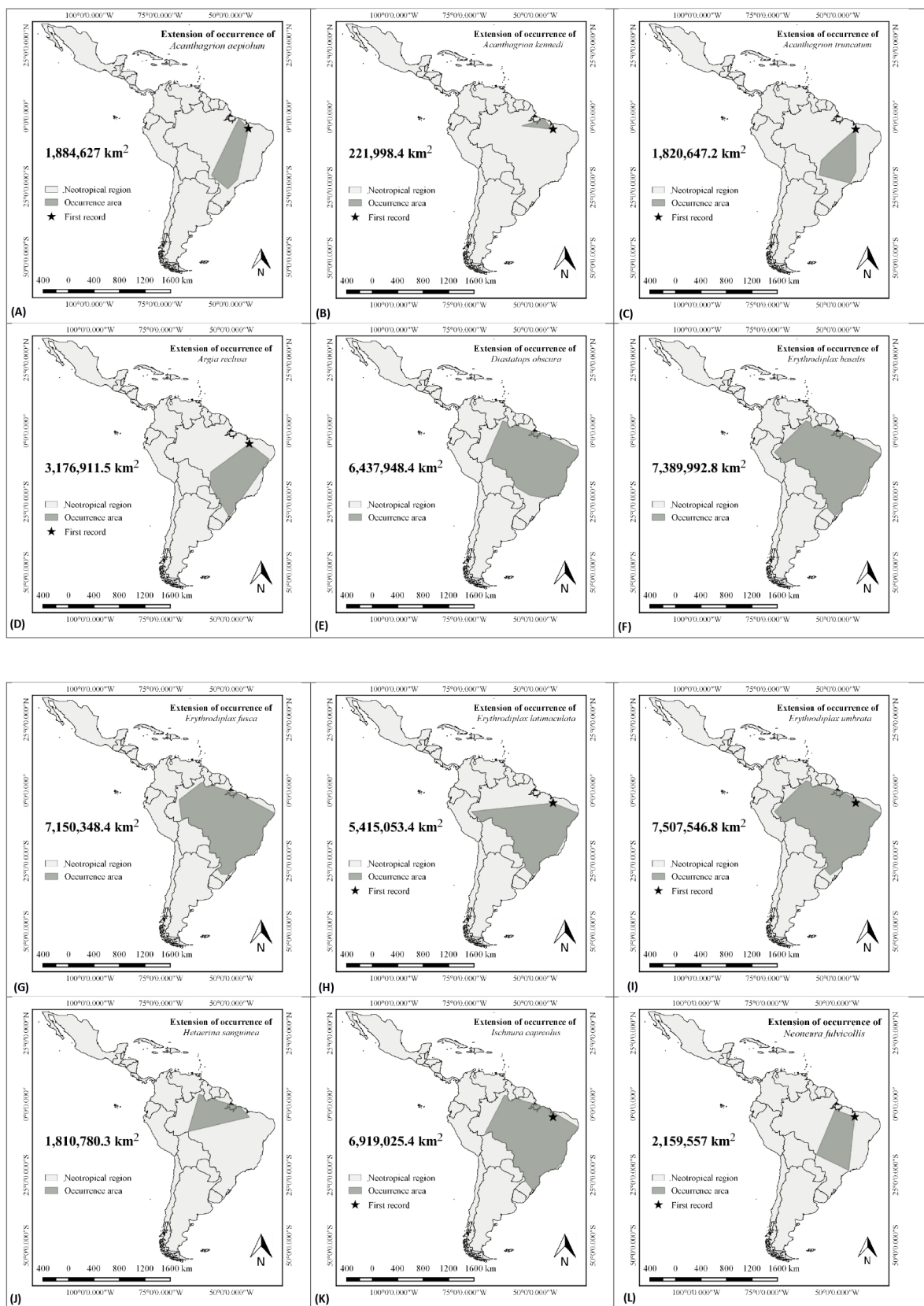


Figure 2. Spatial distribution and new records of Odonata species from Maranhão state, Brazil.

## Odonata of the Maranhão: Areas for inventories

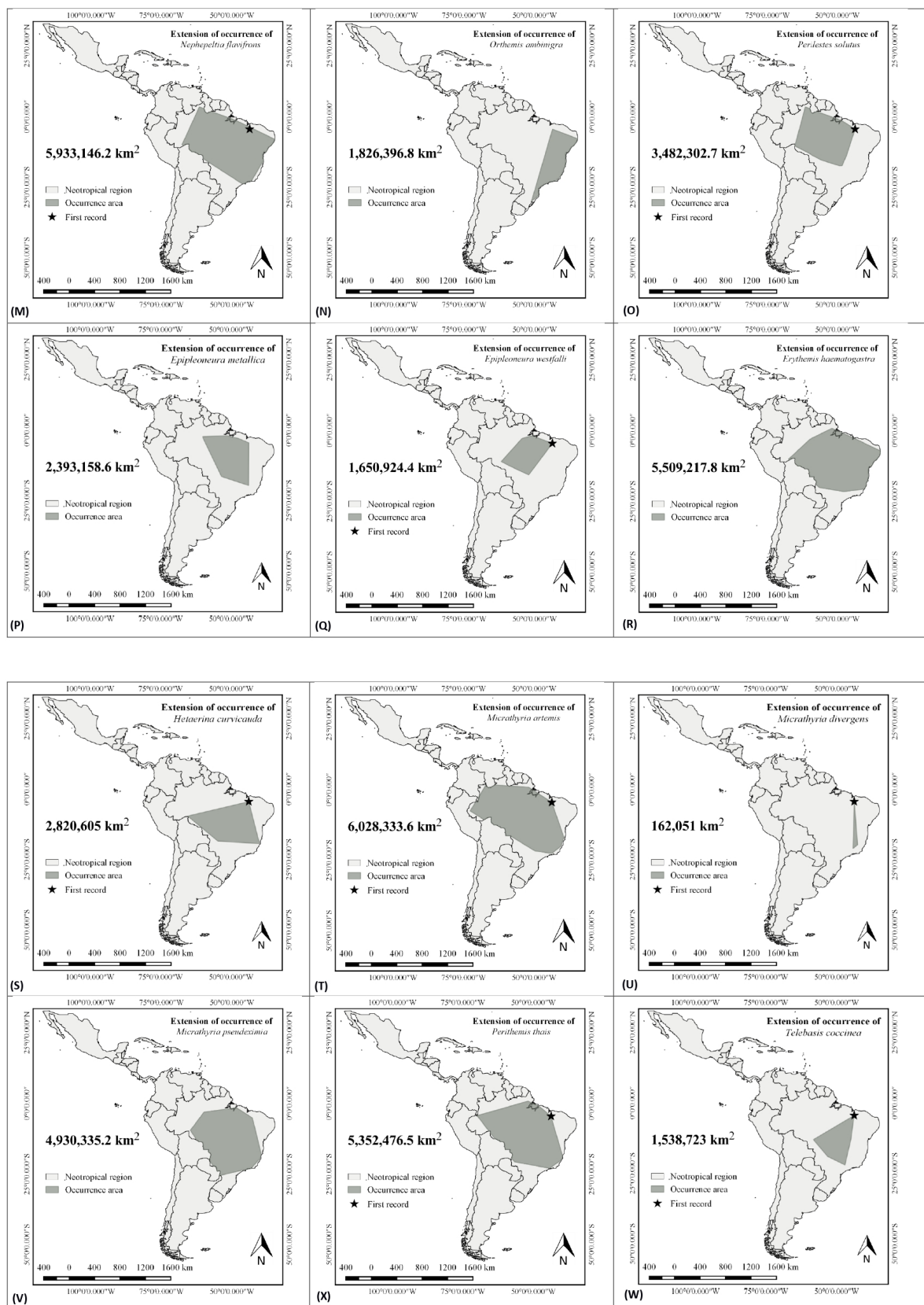
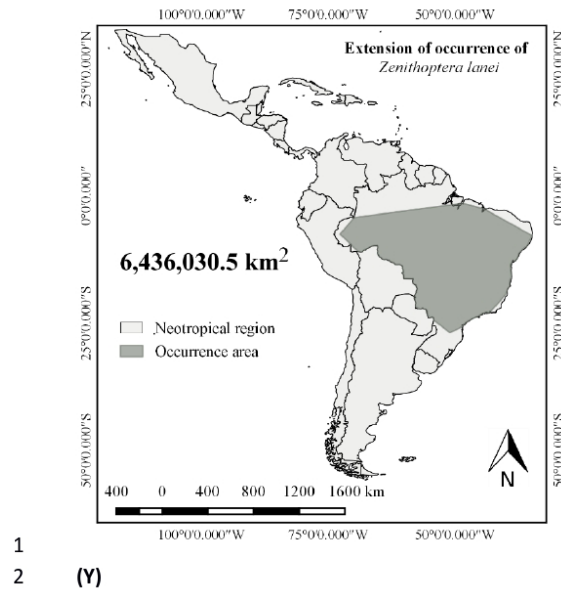
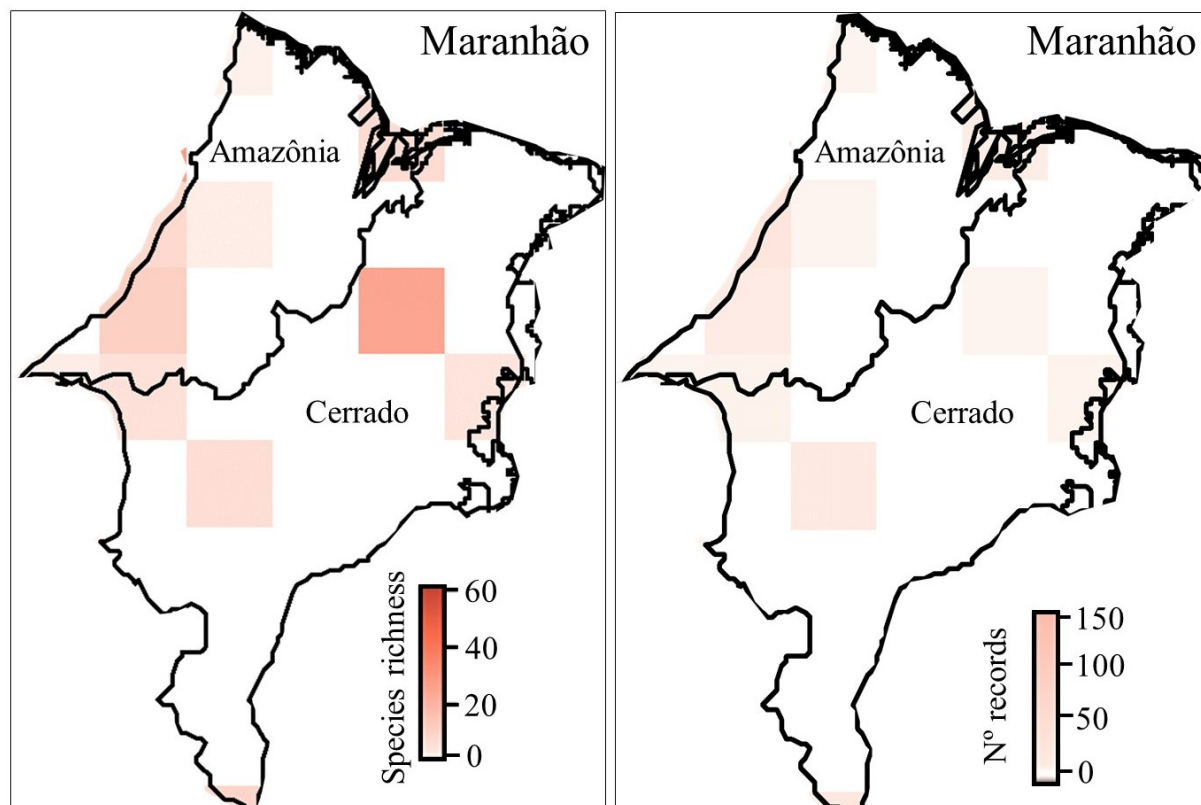


Figure 2. Spatial distribution and new records of Odonata species from Maranhão state, Brazil.



**Figure 2.** Spatial distribution and new records of Odonata species from Maranhão state, Brazil.



**Figure 3.** Records and species richness of Odonata in Maranhão state.

Juen et al. (2014) conducted in the Cerrado and Cerrado–Amazon Forest transition areas. This suggests that other factors, such as resource availability, climate and/or temperature, or even biogeographic patterns (Brasil et al. 2018) may affect the territorial behavior of this specie or the way they perceive environmental variation. It is possible that *A. reclusa* can occur in altered areas, but that have small strip of riparian forest for maintaining a spot of propitious habitat.

It is also worth mentioning that many owners in the region build private pools for family leisure, causing water damming in the channels. In some cases, the stream banks are widened and lined with bags of sand and/or concrete. The water flow in these places is controlled according to the owner's interest. This activity may be favoring the occurrence of generalist organisms, which have a greater tolerance for open areas, such as some species of the genera *Erythrodiplax* and *Micrathyria* (De Marco 2008). The change in stream flow may over time lead to a change in species composition, locally wiping out those that are typical of more pristine lotic environments and favoring the entry of more generalist species from more lentic or semi-lentic environments (Juen et al. 2014).

## 2. Spatial distribution of species

Most of the species registered in Maranhão state have a distribution in Cerrado areas. The species *M. pseudeximia* Westfall, *E. latimaculata* Ris and *E. basalis* have occurrence records in practically all the domains, however, they occur in greater concentration in Cerrado. In contrast to these results, *M. divergens* Westfall, which is reported in this study, is recorded only in regions of the Atlantic Forest. Another case similar to this involves the species *Orthemis ambinigra* Calvert previously reported for Maranhão in the study of De Marco (2008), which also concentrates most of its records in areas of the Atlantic Forest. This results shows the collection gaps in this region (Almeida et al. 2010).

The majority of records for *H. curvicauda* Garrison, *T. coccinea* Selys, and *Epipleoneura westfalli* Machado were concentrated in the midwest region of the country, whereas *M. divergens* and *A. kennedii* were recorded only in the Southeast and Northern regions of the country, respectively. *A. aepiolum*, which had a large occurrence in the Southern Neotropical region (Lozano 2013), was recently reported for the state of Pará in the study by Calvão et al. (2016), Rondônia state in Brazil and Peru and Bolivia (Lencioni 2017) state and now for Maranhão in this study.

Only 29% of the Brazil's total area has geographic distribution data for Odonata and, considering the production of scientific articles, 45%, 20.6% and 16% were published in the Southeast, North and Central West regions respectively (Miguel et al. 2017). For this reason, it is possible that the highest number of records in these regions (Southeast, North and Central West), when compared to the Northeast, is a bias related to the greater collection effort in these regions. Therefore, it is important to have a considerable increase in Odonata collections in the Northeast of Brazil so that it is possible to reduce the Wallacean shortfall. This would also allow for a more robust biogeographic analysis of the order (De Marco 2008).

In the present study, 42.31% of the species are reported for the first time in the Northeast region of Brazil. In addition, the occurrence of *M. paruensis* was recorded, which until then had no certainty in its occurrence for Brazil and was only previously suggested by Heckman (2006). These results suggest that in addition to the Odonata

biogeographic factors, a lack of research investments are generating species distribution data that is strongly skewed by the spatial distribution of universities, research centers and specialists (De Marco & Vianna 2005; Nóbrega & De Marco 2011).

Greater knowledge of species distribution and environments can be used as a tool for selecting new conservation units (Nóbrega & De Marco 2011), and also contributes to robust analyses of the socioeconomic and environmental situation on a regional scale. This work contributes to the knowledge of the fauna of Maranhão state, where high species diversity is expected due to its biogeographic and environmental characteristics, however, in contrast it is one of the states where there are fewer studies on Odonata fauna in the country (De Marco & Vianna 2005). In addition, it enhances the importance of the Northeast region of Brazil for biodiversity conservation, considering that the region is home to several common Odonata species from the Cerrado, Amazon and even the Atlantic Forest. Therefore, despite being an initial effort, with only nine points sampled, this study contributes to minimizing the Wallacean shortfall of the Northeast region and indicates the priority areas. The most unknown region is the Cerrado of the state of Maranhão. Furthermore, the transition regions between Cerrado and Amazônia and between Cerrado and Caatinga are also unknown. All these areas are a priority for faunistic inventories.

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## Author Contributions

Rafael Costa Bastos: Contribution to data collection; Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Leandro Schlemmer Brasil: Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Fernando Geraldo De Carvalho: Substantial contribution in the concept and design of the study; Contribution to critical revision, adding intellectual content.

Lenize Batista Calvão: Substantial contribution in the concept and design of the study; Contribution to critical revision, adding intellectual content.

José Orlando De Almeida Silva: Substantial contribution in the concept and design of the study; Contribution to critical revision, adding intellectual content.

Leandro Juen: Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.



## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Morphological abnormality in a Longnose Stingray *Hypanus guttatus* (Bloch & Schneider, 1801) (Myliobatiformes: Dasyatidae)

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**Abstract:** A Longnose stingray *Hypanus guttatus* (Bloch & Schneider, 1801) embryo with a major asymmetrical morphological abnormality to its pectoral fin was obtained from commercial shrimp fisher's bycatch, off the coast of the Amazon River Mouth in northern Brazil. The specimen and the deformity, which would presumably have impeded its long-term survival, are described and documented in detail. We herein provide the first report of an abnormal individual of this species for the Brazilian coast.

**Keywords:** Fish, Elasmobranchs, Atlantic, Teratology, Deformity.

## Anormalidade morfológica em uma Arraia-bicuda *Hypanus guttatus* (Bloch & Schneider, 1801) (Myliobatiformes: Dasyatidae)

**Resumo:** Um embrião de Arraia-bicuda *Hypanus guttatus* (Bloch & Schneider, 1801) com uma anomalia morfológica assimétrica grave na nadadeira peitoral foi obtido como captura-acidental de um barco de pesca de camarão da costa da foz do rio Amazonas, no norte do Brasil. O indivíduo e sua deformação, que provavelmente teria impedido sua sobrevivência ao longo prazo, são descritos e documentados em detalhe. Apresentamos com esse estudo o primeiro registro de um indivíduo anormal dessa espécie para a costa do Brasil.

**Palavras-chave:** Peixes, Elasmobrânquios, Atlântico, Teratologia, Deformação.

## Introduction

The longnose stingray *Hypanus guttatus* (Bloch & Schneider, 1801), Dasyatidae, is widespread throughout the tropical waters of the western Atlantic Ocean from the southern Gulf of Mexico to the southern coast of São Paulo in Brazil. This aplacental viviparous stingray inhabits shallow water areas to at least 36 m in depth, has been found primarily on sandy bottoms and feeds predominantly on mobile invertebrates (Anderson et al. 2015, Froese & Pauly 2018). *Hypanus guttatus* is a commercially important species in certain regions, such as the Caribbean coast of Colombia, Venezuela and Northeastern Brazil, where it is primarily targeted by artisanal fisheries (Cervigón & Alcalá 1999, Ramírez-Hernandez et al. 2011).

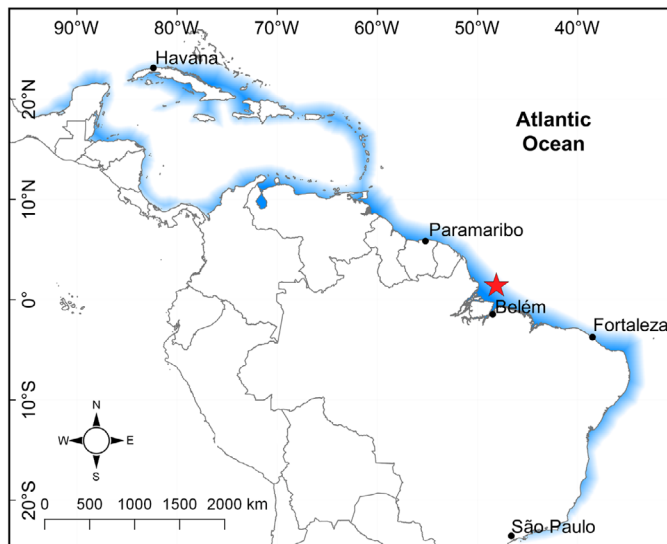
There have been several reports of morphological abnormality in specimens of the genera *Dasyatis* and *Hypanus*, from both the Atlantic and Pacific Oceans. The vast majority of the deformities have been found in the head, rostrum and pectoral fin (Gomes et al. 1991, Lamilla et al. 1995, Escobar-Sánchez et al. 2009, Blanco-Parra & Niño-Torres 2011), while there have been fewer reports of abnormality in other parts, such as gonads, internal organs, tail and teeth (Ribeiro-Prado

et al. 2008, Ramírez-Hernandez et al. 2011). Here we describe in detail the morphological abnormalities of a *H. guttatus* embryo, captured off the Brazilian Amazon Coast, as the first record for this taxon from this region.

## Material and Methods

The *H. guttatus* embryo with its yolk sac still present was collected approximately 200-km off the coast of the Amazon River mouth (1°23'54.4" N 48°07'37.2" W) in February 2017, as bycatch during a bottom trawl for the southern brown shrimp *Farfantepenaeus subtilis* (Pérez-Farfante, 1967), in approximately 43 m depth (Figure 1). The embryo was identified based on Carpenter and Niem (1999), measured, weighed, fixed in 10% formalin, and deposited in the fish collection of the Grupo de Ecologia Aquática (GEA 3677). No other individuals (e.g. more embryos or a pregnant female) of *H. guttatus* were captured during this trawl. A morphological description of the embryo and measurements were conducted based on Gomes et al. (2000) (Table 1). Additionally, measurements were taken for: (1) preorbital length (snout tip along sagittal axis of body to the eye level), (2) snout to aperture

end of the pectoral fin, a linear measurement from the snout tip through the sagittal axis to the aperture end of the pectoral fin level, and (3) anterior margin of cranium to aperture end of the pectoral fin, measured as in the previous item (Figure 2). The internal anatomy was visualized by radiography performed on a Phillips Aquilla Plus 300 device at 45 kVolts and 50 mA for 2.5mAs at the Universidade Federal Rural da Amazônia (UFRA).



**Figure 1.** Distribution of *Hypanus guttatus* (Bloch & Schneider, 1801) (blue shading) according to Rosa and Furtado (2004), and the collection site (red star) at ~200 km offshore Amazon River mouth, Brazil.

## Results

The *H. guttatus* embryo had a morphological abnormality that formed an aperture from the anterior margin of the snout around the left margin of the disc to the fourth gill slit (Figure 2, Table 1). In addition to the clearly separated pectoral fin, the left branchial arches (on the deformed side) appeared to be more widely interspaced, albeit more regularly formed than on the right side (Figure 3). The pectoral fins presented convex anterior margins and slightly straightened posterior margins. The embryo's disc length was slightly shorter than its width and its eye diameter was smaller than the spiracle length. Although the x-ray imaging (Figure 3) does not provide full evidence, the cranium and jaw musculature do not seem to present an evident abnormality. Moreover, other morphological features of the specimen are consistent with other accounts of the species, such as that by Bigelow & Schroeder (1953) and Last et al. (2016).

## Discussion

The cause of the pectoral fin separation found in the *H. guttatus* embryo remains unknown. Pectoral fin separation from the head has been described as a common feature in the early stages of batoid embryonic development, with possible causes including parasitic infection, natural developmental mutation as well as injury- and environmental stressor-induced (e.g. long-term exposure to chemical pollution) teratogenic responses (Mancini et al. 2006, Ramírez-Hernandez et al. 2011).

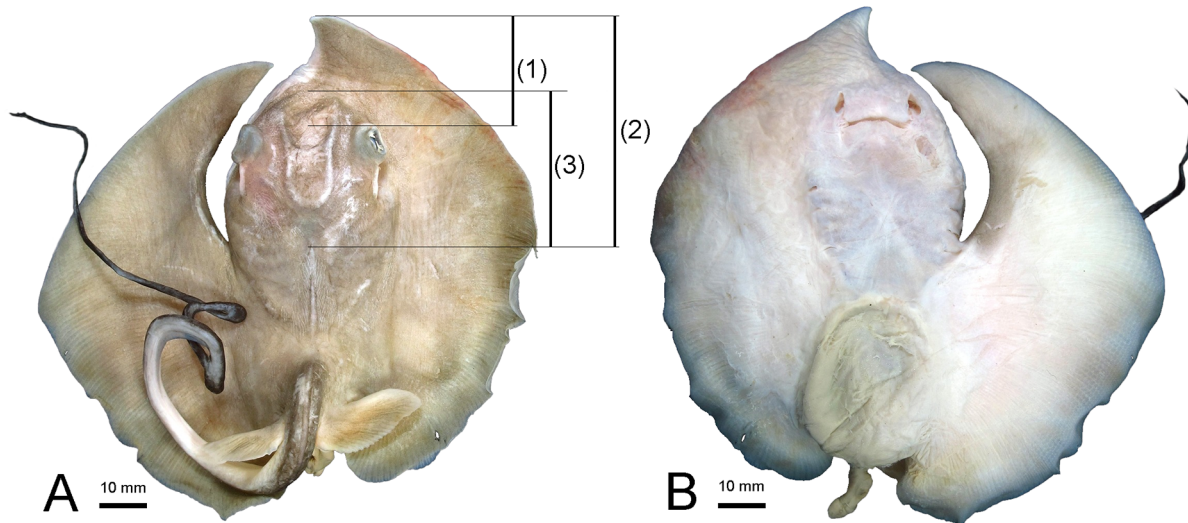
**Table 1.** Measurements of the *Hypanus guttatus* (Bloch & Schneider, 1801) specimen (GEA 3677).

Measurement	(mm)
Disc width	95.9
Disc length	96.6
Total length	271.6
Disc length to axil of pectoral fin	44.4
Preoral length	27.2
Preorbital length	27.6
Snout to cloaca	89.9
Snout to aperture end	50.4
Cloaca to sting base	39.8
Tail length	199.2
Tail height	4.9
Height of dorsal finfold	0.5
Eye diameter	7.2
Spiracle length	8.8
Cranial interorbital	15.6
Cranium anterior margin to aperture end	30.9
Interspiracular	24.7
Internarial	6.4
Mouth width	5.4
Distance between first pair of gill slits	12.3
Distance between first and fifth gill slits	9.0
Pelvic fin anterior margin	4.1
Pelvic fin posterior margin	10.2

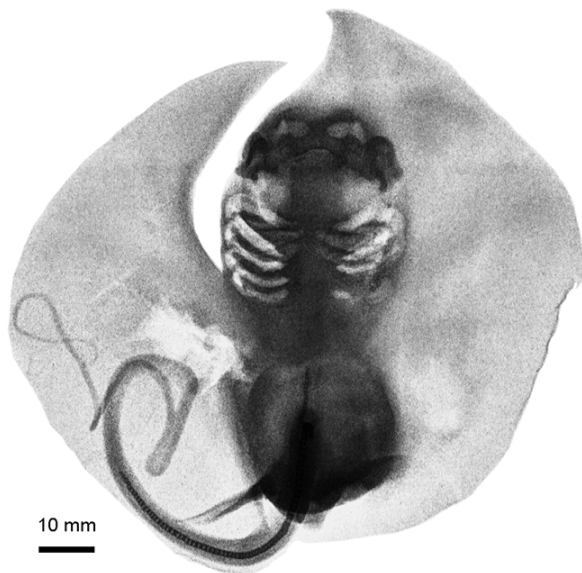
Morphological abnormalities of the fins could directly impact a fish's mobility, affecting both its ability to forage and/or avoid predators, thereby reducing its likelihood of survival (Mancini et al. 2006). Although the morphological abnormality found in the embryo from this study could have presumably impeded its long-term survival, a similar malformation was recorded in a mature male *H. guttatus* captured in the Colombian Caribbean (Ramírez-Hernandez et al. 2011). This mature male had a disc width of 520 mm; however, the abnormality was symmetrical and divided the snout equally, which presumably had a reduced impact on the swimming kinematics of the individual, thus allowing it to reach adult size. The abnormality of the *H. guttatus* embryo was asymmetrical and would presumably have had a negative impact on the swimming capacity of the individual, post-embryonically. That being said, Ribeiro-Prado et al. (2008) reported a female *Pteroplatytrygon violacea* (Bonaparte, 1832) (Dasyatidae) specimen which reached maturity, carrying an embryo, regardless of exhibiting very similar asymmetrical anomaly—a one-sided deeply unfused pectoral fin.

Scientific records of abnormalities for elasmobranchs are generally much rarer than for teleost fishes. The present report is only the second for an abnormal *H. guttatus* specimen, and the first from the Brazilian coast and for an embryo of this species thus providing an important biogeographic insight on abnormalities for this genus and species, given that existing and future reports contribute to the detection of





**Figure 2.** Embryo *Hypanus guttatus* (Bloch & Schneider, 1801), collected off the Brazilian Amazon Coast (GEA 3677), disc width = 95.9 mm, (a) dorsal view, (b) ventral view. New measurements depicted by (1) preorbital length, (2) snout to aperture end of the pectoral fin, and (3) anterior margin of cranium to aperture end of the pectoral fin; see material and methods for more details.



**Figure 3.** Radiograph of the embryo *Hypanus guttatus* (Bloch & Schneider, 1801) (GEA 3677), disc width = 95.9 mm.

potential spatial and/or species-related patterns and, eventually, may help uncovering possible causes.

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### Author Contributions

Kurt Schmid is the main (first) author of the manuscript and made substantial contribution in the concept and design of the study.

Marcelo Andrade made contribution to data collection, data analysis and interpretation and manuscript preparation.

Fabiola Machado made contribution to data collection, data analysis and interpretation.

Juliana Araujo made contribution to data collection, data analysis and interpretation.

Eglê Corrêa made contribution to data collection.

Tommaso Giarrizzo made substantial contribution in the concept and design of the study, to critical revision, adding intellectual content.

### Conflicts of interest

The author(s) declare(s) that they have no conflict of interest related to the publication of this manuscript.

### Data availability

All data used and analyzed in this study are included in the main text of the manuscript.

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## Ichthyofauna from floodplain lakes of Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), lower rio Purus

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**Abstract:** The ichthyofauna of the rio Purus has been little investigated, especially in its lower portion, characterized by diverse aquatic environments, especially in flooded areas. We inventoried the ichthyofauna of floodplain lakes with different management efforts in Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), which represent important resource for commercial fishing in North region, in the first illustrated inventory of the fish fauna from lower rio Purus. We surveyed 20 lakes, classified as open access (eight lakes) and protected (12 lakes) during 2009 low water levels. A total of 2,299 individuals were collected, represented by seven orders, 25 families and 74 species. Characiformes was the most representative order in number of species and families, followed by Siluriformes, Cichliformes, Gymnotiformes, Clupeiformes, and Osteoglossiformes. Most abundant and frequent species were *Pygocentrus nattereri*, *Triportheus angulatus*, *Serrasalmus* sp. “2n=58”, *Pterygoplichthys gibbiceps*, and *Osteoglossum bicirrhosum*. Protected lakes presented higher species richness compared to open access lakes. Similarly, protected lakes possessed 26 species occurring exclusively, with emphasis on *Colossoma macropomum*, an important species for fisheries due to its commercial importance. We added 44 new records of fish species for the lower rio Purus. Our results indicate the potential efficiency of zoning systems of open access and protected lakes established by local population and ruled by RDS-PP for fisheries management. Therefore, we strongly suggest its maintenance for conservation of ichthyofauna of floodplain lakes of lower rio Purus.

**Keywords:** Amazonian fish fauna, Conservation, Inventory, Neotropical.

## Ictiofauna de lagos de várzea da Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), baixo rio Purus

**Resumo:** A ictiofauna do rio Purus tem sido pouco investigada, especialmente em seu curso inferior, caracterizada pela diversidade de ambientes aquáticos, especialmente em áreas de inundação. Inventariamos a ictiofauna dos lagos de várzea com diferentes tipos de manejo na Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), que representa importante recurso para pesca comercial na região Norte, no primeiro inventário ilustrado da fauna de peixes do baixo rio Purus. Amostramos 20 lagos classificados como de uso (oito lagos) e proteção (12 lagos), durante o período de águas baixas de 2009. Foi coletado um total de 2.299 indivíduos representados por sete ordens, 25 famílias e 74 espécies. Characiformes foi a ordem mais representativa em número de espécies e famílias, seguida por Siluriformes, Cichliformes, Gymnotiformes, Clupeiformes e Osteoglossiformes. As espécies mais abundantes e frequentes foram *Pygocentrus nattereri*, *Triportheus angulatus*, *Serrasalmus* sp. “2n=58”,

*Pterygoplichthys gibbiceps* e *Osteoglossum bicirrhosum*. Lagos de proteção apresentaram maior riqueza de espécies comparados aos lagos de uso. Similarmente, lagos de proteção possuíam 26 espécies ocorrendo exclusivamente nesta categoria, com ênfase no *Colossoma macropomum*, uma espécie importante para pesca devido ao seu valor comercial. Adicionamos 44 novos registros de espécies de peixes para o baixo rio Purus. Nossos resultados indicam a potencial eficácia do sistema de zoneamento de lagos de uso e proteção para o manejo da pesca estabelecido pela população local e regulamentada pela RDS-PP. Portanto, sugerimos fortemente sua manutenção para conservação da ictiofauna dos lagos de várzea do baixo rio Purus.

**Palavras-chave:** Fauna de peixes amazônicos, Conservação, Inventário, Neotropical.

## Introduction

The Amazon basin is the largest and most diverse hydrographic system on Earth (Goulding *et al.* 2003), covering an area of over 8,000,000 km<sup>2</sup> (Sioli 1984), comprising almost 20% of the total river water in the world (Reis *et al.* 2016). Diversity of ichthyofauna inhabiting the Amazon basin comprises 60 families, 529 genera, and almost 2,720 described species, from which more than 60% are endemic to the basin (Dagosta & de Pinna 2019). However, this number might be underestimated, because new species descriptions are increasing fish species richness abruptly (Hoorn & Wesselingh 2010, Dagosta & de Pinna 2017).

The Western Amazon (or Lowland Amazon) is drained by white- or muddy-water rivers with headwaters in the Andes (*i.e.*, Solimões, Madeira and Japurá) and by tributaries draining the western lowlands as Purus and Juruá (Lima & Ribeiro 2011), composing the floodplains popularly known as *várzeas* (Sioli 1984). This region is characterized by elevated load of sediments and dissolved inorganic solids (Sioli 1984, Goulding *et al.* 2003). The fish species richness in Lowland Amazonia is directly related to the exceptional diversity of the floodplains (Crampton 2011) and might also be influenced by the tributary effect that enriches the species diversity of Amazon mainstream, especially the white water tributaries from the Western region (Fernandes *et al.* 2004).

The rio Purus is one of the largest tributaries of the rio Solimões, with an extension of 2,765.65 km (Melo 2012) from its sources in Peru until its confluence on rio Solimões, at Amazonas State. It represents the largest fishing productivity of Amazon basin compared to other major white-water tributaries (Petrere 1978, Batista 1998, 2003), however, the efforts are concentrated in a few species, from which only 12 represent 80% of the fish landed in the main ports of Amazonas State (Barthem & Fabre 2003). Despite this potential, few studies were conducted in order to investigate the ichthyofauna of this basin. Most focused on the upper portion of the river, with 48 species recorded in the rio Macauhan (tributary of rio Yaco) (La Monte 1935), 86 species in Caeté and Macapá rivers (Anjos *et al.* 2008), and 94 species in rio Acre (Claro-García *et al.* 2013). The investigations in lower rio Purus registered higher number of species: 180 by Rapp Py-Daniel & de Deus (2003) in flooded areas, and approximately 110 species in two sandy beaches of Reserva Biológica de Abufari and Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP) (Duarte *et al.* 2010).

The RDS-PP was created in 2003 by the Amazonas State Government, to conciliate usage of natural resources with socio-environmental sustainability. The region was deeply exploited for terrestrial and aquatic resources during the first half of twentieth century (Vieira *et al.* 2015). Activities as fishing, hunting, and practice of small-scale agriculture and extractivism are allowed to approximately

60 communities with 4,000 residents (de Deus *et al.* 2003, IPI 2010). Fishing rules agreement determining management practices and fisheries areas were established in 2008 by local population and signed in agreements. Floodplain lakes were divided in a zoning model into two categories: open access and protected. In open access lakes subsistence and commercial fishing are allowed, while in protected lakes commercial fishing is prohibited (IPI 2010). The zoning model adopted in RDS-PP is similar to the model pioneered in RDS-Mamirauá (Queiroz & Crampton 1999).

The reserve is comprised of both upland *terra firme* landscapes and flooded forests, the latter representing 45% of its area, more concentrated on its North region, on the interfluvium of Solimões and Purus rivers (de Deus *et al.* 2003; IPI 2010). The confluence of Purus and Solimões rivers generates a large floodplain area in the lower portion of the basin, with many environments still unexplored, which require huge sampling efforts to improve the fish fauna knowledge and fisheries consequences in natural sources. In this study, we sampled, for the first time, floodplain lakes in RDS-PP, with different management efforts, that represent important resources for commercial fishing in North region. Therefore, the main goals of this study were: (i) register the composition of fish species that occur in the RDS-PP floodplain lakes of different zoning model categories: open access and protected; and (ii) provide the first illustrated inventory of the ichthyofauna from floodplain lakes of lower rio Purus basin.

## Material and Methods

### 1. Study area

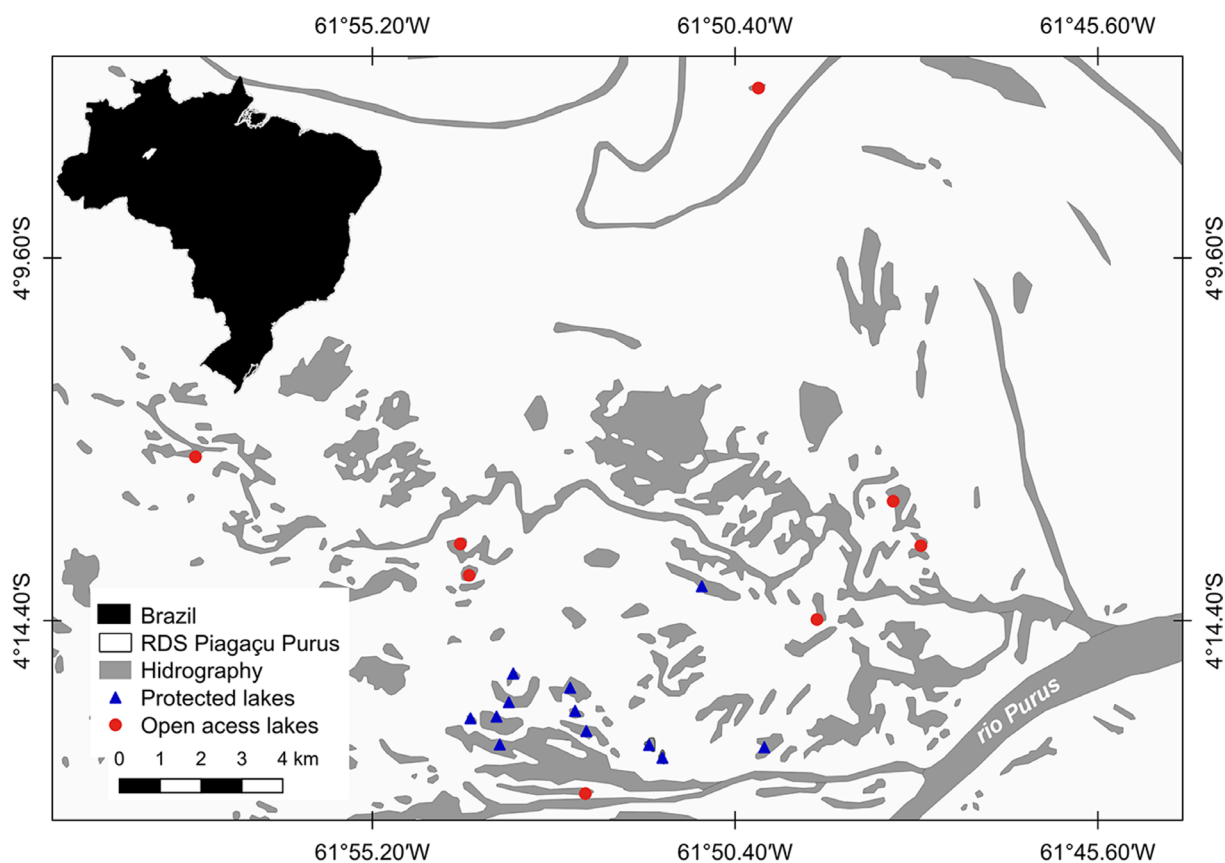
The Reserva de Desenvolvimento Sustentável Piagaçu-Purus (4°03'–5°25'S and 61°40'–63°30'W) comprises 834,245 ha situated along the lower rio Purus basin (Fig. 1), 223 km upstream from Manaus, capital of Amazonas State (Vieira *et al.* 2015). Surveys were carried in 20 floodplain lakes of the RDS-PP between October and November 2009, during low water levels on lower rio Purus (Tab. 1). Lakes were chosen according to access availability, and were previously classified by the zoning model as eight open access and 12 protected (Fig. 1).

### 2. Data Collection

Sampling efforts were standardized for all lakes. Fishing gear consisted in a set of stationary gillnets (10 m length) with mesh sizes 40, 60, 80, 100 and 120 mm between opposite knots, exposed for 24 hours starting at 6:00 AM, with harvests at four hours. The exposure time was defined in order to minimize the damage caused by piscivorous fish, dolphins and alligators to gillnet and fish caught.



## Ichthyofauna from floodplain lakes of rio Purus



**Figure 1.** Map of the study area showing the location of the Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP) in Amazonas State, and sampled floodplain lakes from rio Purus.

**Table 1.** Sampled floodplain lakes from rio Purus at Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP) in Amazonas State.

Site	Name of the lake	Category	Coordinates
1	Cobra	Open access	4°13'24.5"S/61°47'56.6"W
2	Pepu Grande	Open access	4°12'49.1"S/61°48'18.6"W
3	Adão	Open access	4°14'23.1"S/61°49'19.1"W
4	Panela	Open access	4°12'11.5"S/61°57'30.0"W
5	Macacão	Open access	4°13'20.2"S/61°53'53.3"W
6	Macaco	Open access	4°13'48.0"S/61°53'55.2"W
7	Igarapé Açu	Open access	4°16'40.7"S/61°52'20.7"W
8	Fortuna	Open access	4°07'22.5"S/61°50'00.8"W
9	Marajá	Protected	4°13'52.8"S/61°50'48.0"W
10	Cedro	Protected	4°15'45.9"S/61°53'56.9"W
11	Caverna	Protected	4°15'25.0"S/61°53'33.7"W
12	Caverninha	Protected	4°15'08.8"S/61°53'18.2"W
13	Tucuxi	Protected	4°16'14.8"S/61°50'03.9"W
14	Redondo 1	Protected	4°15'50.5"S/61°52'30.7"W
15	Redondo 2	Protected	4°15'35.6"S/61°52'31.1"W
16	Redondo 3	Protected	4°15'28.6"S/61°52'28.5"W
17	Redondo do Caverna 1	Protected	4°15'48.2" S/61°53'17"W
18	Redondo do Carvena 2	Protected	4°15'50.3"S/61°53'25.4"W
19	Henrique	Protected	4°16'05.4"S/61°51'29.3"W
20	Mureruzinho	Protected	4°16'12.0"S/61°51'23.5"W

In field, fish specimens were anesthetized in benzocaine hydrochloride, and fixed in 10% formalin and weighted to define the individual biomass. Total biomass of each species was represented by the sum of the weight of all specimens collected. Posteriorly, specimens were transferred to 70% ethanol in the laboratory and deposited in the Coleção de Peixes do Instituto Nacional de Pesquisas da Amazônia (INPA, Manaus, Amazonas State).

### 3. Fishfauna

The classification of families and subfamilies were presented in alphabetical order due to the many changes in the current classifications, and the lack of congruence or resolution regarding the relationships between them. The nomenclatural arrangement and classification of groupings in levels higher than family and family names followed Betancur-R. et al. (2017). Sub-families of Characidae followed Mirande (2018), and of Loricariidae followed Armbruster (2004), with the exception of Rhineleptinae, which is according to Lujan et al. (2015). Identification follows Menezes & Géry (1983), Kullander & Ferreira (2006), Santos *et al.* (2006), Bristki & Birindelli (2008), Zarske & Géry (2008), Queiroz et al. (2013), Littmann et al. (2015), Andrade et al. (2016), Ferraris et al. (2017), Ramirez et al. (2017), Ribeiro et al. (2017), and Mateussi et al. (2018).

## Results

A total of 2,299 individuals were collected, distributed in seven orders, 25 families and 74 species (Tab. 2, Figs. 2-4). Characiformes was the most representative order in number of species and families (44 and 14, respectively), followed by Siluriformes with 20 species and six families (Fig. 5), Cichliformes, with only one family but five species, and Gymnotiformes (two species in two families). Clupeiformes, Osteoglossiformes and Perciformes were less representative, with only one species each.

Characiformes and Siluriformes presented major number of species. The families with higher species richness were Serrasalminae (12 species), Curimatidae and Pimelodidae (7 species), and Loricariidae (5 species) (Fig. 6). Highest abundances were also observed for Characiformes and Siluriformes with 1,888 and 279 individuals respectively.

Most abundant and frequent species were *Pygocentrus nattereri* (piranha caju) with 880 individuals collected in 90% of sampled lakes, followed by *Triportheus angulatus* (sardinha-papuda) (276 individuals and 80% of lakes), *Serrasalmus* sp. 2n=58 (piranha preta) (157 individuals in 80% of lakes), *Pterygoplichthys gibbiceps* (bodó) (100 individuals in 65% of lakes), and *Osteoglossum bicirrhosum* (aruanã) with 76 individuals captured in 65% of lakes. Species represented by only one individual represented 28.5% of the total catch. With exception of *Serrasalmus maculatus*, with scarce number of individuals (59) but collected in 65% of lakes, the other species were found in less than 45% of the lakes.

Four species presented high biomass levels, representing 72.7% of total 359.5 kg collected, *P. nattereri* (32.4%), *O. bicirrhosum* (20.8%), *Colossoma macropomum* (tambaqui) (11.8%), and *P. gr. gibbiceps* (7.7%). Despite the great contribution of *C. macropomum* biomass it occurred in only 20% of the sampled sites (all protected lakes), differing from the others that occurred in at least 60% of the lakes.

Protected lakes presented higher species richness (62 species) compared to open access lakes with 48 species collected (Table 3). Similarly, protected lakes possessed 26 species occurring exclusively in this category (e.g., *Brycon* spp., *Colossoma macropomum*, *Curimata inornata*, *Hypophthalmus* spp., and *Pseudoplatystoma* spp.), while in open access lakes this number was reduced to 12 species (e.g. *Loricariichthys acutus*, *Mylossoma aureum*, and *Semaprochilodus taeniurus*). However, beyond the dominance of Characiformes and Siluriformes in both type of lakes, Cichliformes did not occur in protected lakes, while in open access lakes five species were collected, including commercially important as (*Astronotus ocellatus* and *Cichla monoculus*). Open access lakes presented higher abundance of Characiformes than protected lakes, mainly due to the participation of *P. nattereri* (Tab. 3).

Species accumulation curve is concerned with accumulation rates of new species over the sampled area and depends on species identity. Considering all lakes sampled, the species accumulation curve did not completely reach the asymptote (Fig. 7). Species richness expectation obtained by nonparametric estimators indicated that was collected 72% of the species expected to lakes by Jackknife estimate 1 (Jack1 = 107.4), 60% by Jackknife estimate 2 (Jack2 = 12, 5), 57.5% by Chao estimate (Chao = 133.89) and 85.8% according to the Bootstrap estimator.

## Discussion

The ichthyofauna of floodplain lakes of RDS Piagaçu-Purus on lower rio Purus, as remaining freshwater regions of the world, is composed mostly by Ostariophysi, representing 88.2% of total richness. Characiformes presented higher families representativeness from which Serrasalminae and Curimatidae composed 25% of all diversity surveyed by us. The predominance of these two families is different from previous results registered for lower rio Purus. From the 90 species inventoried using gillnets by Rapp Py-Daniel & de Deus (2003), mostly belong to Anostomidae (10), Serrasalminae (8) and Hemiodontidae (6), whereas only *Hemiodus* sp. “rabo de fogo” and five species of anostomids were captured in the present study. The former study was developed during high water levels (June) and fish were sampled in different types of environments (lakes, paranás, igarapés and river channel), while the present investigation was performed in low water levels (October and November) strictly in lakes.

The lower rio Purus, next to its confluence with rio Solimões generates a large floodplain. Flooded areas are known by suffer seasonally dramatic changes in landscapes, modeled by the flood pulse (Junk et al. 1989). During wet period flooded forest consists an important feeding source of fruits and seeds, providing a wider diversity of habitat options, decreasing the density of possible predators (Winemiller & Jepsen 1998, Lowe-McConnell 1999). In low water ichthyofauna migrates to available habitats, including main river channel and permanent lakes, increasing predators occurrence in consequence of the reduction of suitable occupancy areas (Goulding 1980). These environmental differences are essential to the maintenance of life cycle of many fish species and biological interactions (Duarte et al. 2010). Despite the efficiency and selectivity of the use of a unique sampling effort (gillnet) we reached a representative number of species (74) and complemented available information of ichthyofauna from lower rio Purus, surveying for

**Table 2.** List of fish species captured in floodplain lakes of Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), lower rio Purus, in 2009 drought. The references for the classification used are listed in the Material and Methods section. (Locality: 1. Cobra; 2. Pepu grande; 3. Adão; 4. Panela; 5. Macacão; 6. Macaco; 7. Igarapé Açu; 8. Fortuna; 9. Marajá; 10. Cedro; 11. Caverna; 12. Caverninha; 13. Tucuxi; 14. Redondo 1; 15. Redondo 2; 16. Redondo 3; 17. Redondo do caverna 1; 18. Redondo do caverna 2; 19. Henrique; 20. Mureruzinho. Bold numbers are for Protected lakes).

Classification	Locality	Voucher INPA
<b>TELEOSTEI</b>		
<b>OSTEOGLOSSOCEPHALAI</b>		
<b>OSTEOGLOSSIFORMES</b>		
<b>Osteoglossidae</b>		
<i>Osteoglossum bicirrhosum</i> (Cuvier, 1829)	3,4,5,6,7,9,10,11,12,14,15,19,20	17228, 39950
<b>CLUPEIFORMES</b>		
<b>Pristigasteridae</b>		
<i>Pellona flavipinnis</i> (Valenciennes, 1837)	1,7,17,18	34030, 36729
<b>CHARACIFORMES</b>		
<b>Acestrorhynchidae</b>		
<i>Acestrorhynchus microlepis</i> (Jardine, 1841)	1	17152
<i>Acestrorhynchus cf. pantaneiro</i> Menezes, 1992	1,2,4,6,7,9,17,18,20	23749, 36738
<b>Anostomidae</b>		
<i>Leporinus friderici</i> (Bloch, 1794)	1,4,5,8,12,18,19	36714, 48861
<i>Leporinus parae</i> Eigenmann, 1907	6,10	36734, 36737
<i>Megaleporinus trifasciatus</i> (Steindachner, 1876)	2,3,12	36724
<i>Rhytiodus microlepis</i> Kner, 1858	3,4,5,6,7,9,10,11,12,14,15,19,20	41882
<i>Schizodon fasciatus</i> Spix & Agassiz, 1829	1,4,5,9,13,14,15,16,19	36599, 36720
<b>Bryconidae</b>		
<i>Brycon amazonicus</i> (Agassiz, 1829)	13	17122, 23628
<i>Brycon melanopterus</i> (Cope, 1872)	13	36665
<b>Chalceidae</b>		
<i>Chalceus erythrurus</i> (Cope, 1870)	1,2,4,5,8,9,13,19,20	17257, 17235
<b>Characidae</b>		
<b>Characinae</b>		
<i>Roeboides myersii</i> Gill, 1870	1,3,4,13,14,17	36573
<b>Tetragonopterinae</b>		
<i>Tetragonopterus argenteus</i> Cuvier, 1816	13	36718
<i>Tetragonopterus chalceus</i> Spix & Agassiz, 1829	17	42420
<b>Ctenoluciidae</b>		
<i>Boulengerella maculata</i> (Valenciennes, 1850)	13	42394
<b>Curimatidae</b>		
<i>Curimata inornata</i> Vari, 1989	19	23638, 36707
<i>Curimatella alburna</i> (Müller & Troschel, 1844)	1,7,17,18,19	36708, 36715
<i>Potamorhina altamazonica</i> (Cope, 1878)	1,2,7,9,16,17,18,19,20	36746
<i>Potamorhina pristigaster</i> (Steindachner, 1876)	4	23617, 36730
<i>Psectrogaster amazonica</i> Eigenmann & Eigenmann, 1889	1,2,7,17,18,19	36594, 36736
<i>Psectrogaster rutiloides</i> (Kner, 1858)	1,2,16	36558, 36740
<i>Steindachnerina bimaculata</i> (Steindachner, 1876)	1,2,4,8,10,16,17,19	36600, 36626
<b>Cynodontidae</b>		
<i>Cynodon gibbus</i> (Spix & Agassiz, 1829)	1,2,16	23746
<i>Rhaphiodon vulpinus</i> Spix & Agassiz, 1829	1,3,4,7,11,12,15,17,18	18551, 32043

Continuation Table 2.

Classification	Locality	Voucher INPA
<b>Erythrinidae</b>		
<i>Hoplias malabaricus</i> (Bloch, 1794)	1,2,3,6,10,12,15,16	33740, 48374
<b>Hemiodontidae</b>		
<i>Hemiodus</i> sp. "rabo de fogo"	5,13	36706
<b>Iguanodectidae</b>		
<i>Bryconops melanurus</i> (Bloch, 1794)	19	36709
<b>Prochilodontidae</b>		
<i>Prochilodus nigricans</i> Spix & Agassiz, 1829	1,3,6,11	41661
<i>Semaprochilodus insignis</i> (Jardine, 1841)	1,4,5,8,10,13,17,20	41691
<i>Semaprochilodus taeniurus</i> (Valenciennes, 1821)	18	42456
<b>Serrasalminae</b>		
<i>Colossoma macropomum</i> (Cuvier, 1816)	12,15,19,20	58771
<i>Metynnis longipinnis</i> Zarske & Géry, 2008	13	39502
<i>Myloplus aff. asterias</i> (Müller & Troschel, 1844)	13	58772
<i>Myloplus torquatus</i> (Kner, 1858)	9	36702
<i>Mylossoma albiscopum</i> (Cope, 1872)	1,2,3,5,6,11,13	36683, 36686
<i>Mylossoma aureum</i> (Spix & Agassiz, 1829)	4	16776, 41655
<i>Pristobrycon striolatus</i> (Steindachner, 1908)	13	36703, 41917
<i>Pygocentrus nattereri</i> Kner, 1858	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18	41663, 41689
<i>Serrasalmus elongatus</i> Kner, 1858	1,2,4,5,6,7,8,9,10,13,17,18	36672, 36705
<i>Serrasalmus maculatus</i> Kner, 1858	1,2,3,4,5,7,8,9,11,12,15,18,19	36698, 36699
<i>Serrasalmus rhombeus</i> (Linnaeus, 1766)	13	36704, 36693
<i>Serrasalmus</i> sp. "2n=58"	1,2,3,4,5,7,8,9,10,11,13,15,16,17,18,19	36670, 36673
<b>Triporthidae</b>		
<i>Triporthus angulatus</i> (Spix & Agassiz, 1829)	1,2,3,4,5,6,7,9,10,11,12,13,15,17,18,19	36713, 36716
<i>Triporthus auritus</i> (Valenciennes, 1850)	1,2,5,7,13,17,18,20	36666, 48380
<i>Triporthus rotundatus</i> (Jardine, 1841)	13	36744
<b>GYMNOTIFORMES</b>		
<b>Rhamphichthyidae</b>		
<i>Rhamphichthys pantherinus</i> Castelnau, 1855	3,17	27614, 42430
<b>Sternopygidae</b>		
<i>Eigenmannia macrops</i> (Boulenger, 1897)	14	36717
<b>SILURIFORMES</b>		
<b>Auchenipteridae</b>		
<i>Ageneiosus inermis</i> (Linnaeus, 1766)	11,15,16	36728, 41700
<i>Trachelyopterus galeatus</i> (Linnaeus, 1766)	12,14,15	36661, 41790
<i>Trachychorystis porosus</i> Eigenmann & Eigenmann, 1888	2,12	36732, 36735
<b>Doradidae</b>		
<i>Anadoras grypus</i> (Cope, 1872)	10,12,14,15	36733
<i>Oxydoras niger</i> (Valenciennes, 1821)	2,3,5,11	36731
<b>Callichthyidae</b>		
<i>Dianema urostriatum</i> (Miranda Ribeiro, 1912)	20	36722
<i>Hoplosternum littorale</i> (Hancock, 1828)	1,3,4,5,6,10,12	36710



Continuation Table 2.

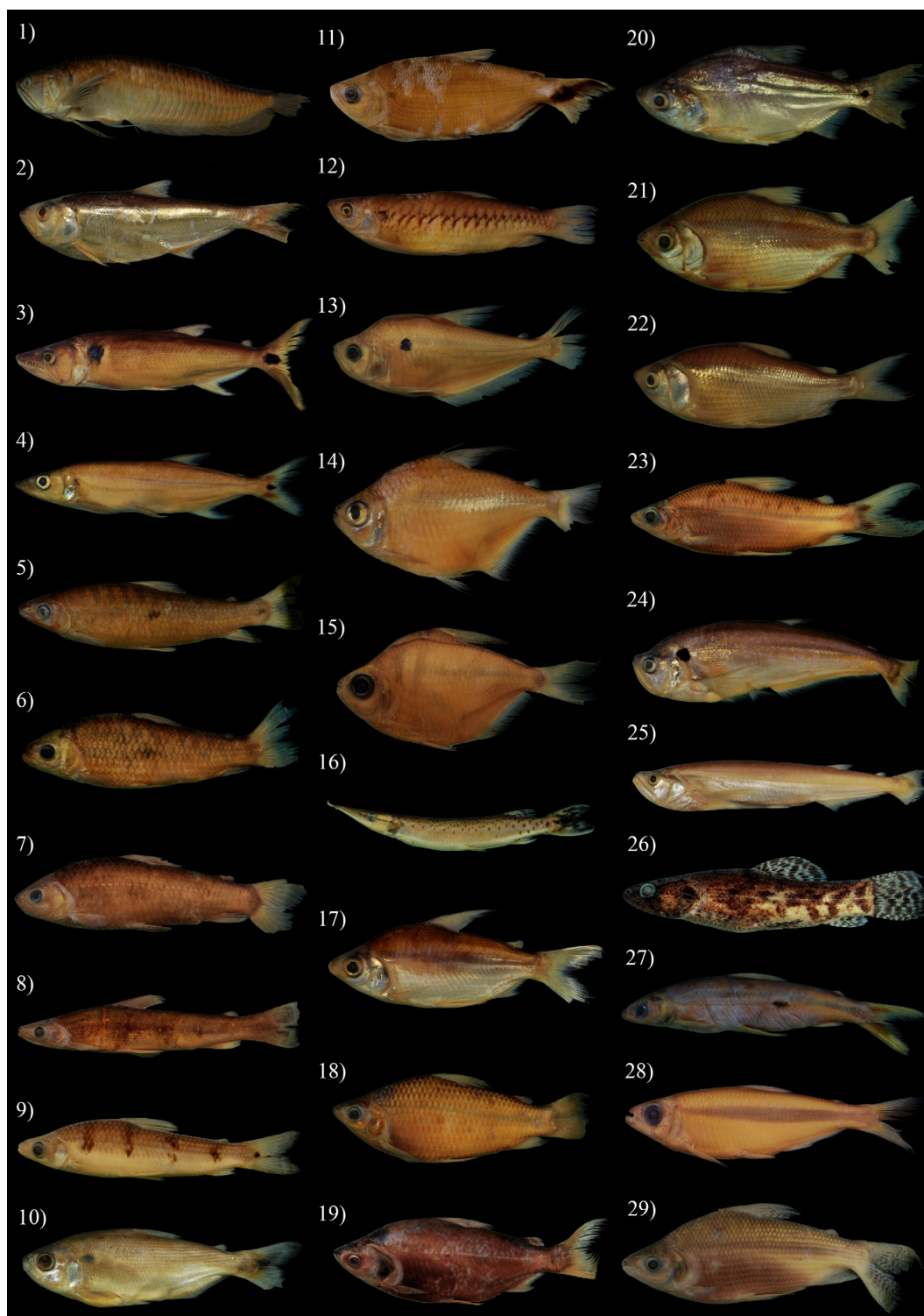
Classification	Locality	Voucher INPA
<b>Loricariidae</b>		
<b>Hypostominae</b>		
<i>Dekeyseria amazonica</i> Rapp Py-Daniel, 1985	13	17328, 29206
<i>Pterygoplichthys gibbiceps</i> (Kner, 1854)	2,3,4,6,8,10,11,12,14,15,16,19,20	36721, 36664
<b>Loricariinae</b>		
<i>Loricariichthys acutus</i> (Valenciennes, 1840)	1,2,3	36712
<i>Loricariichthys nudirostris</i> (Kner, 1853)	4	34633
<b>Rhinelepinae</b>		
<i>Pseudorinelepis genibarbis</i> (Valenciennes, 1840)	4,8,10,12,20	36745
<b>Heptapteridae</b>		
<i>Pimelodella</i> gr. <i>cristata</i> (Müller & Troschel, 1849)	7,17	36741, 36748
<b>Pimelodidae</b>		
<i>Calophysus macropterus</i> (Lichtenstein, 1819)	10,11,15,18	36743
<i>Hypophthalmus edentatus</i> Spix & Agassiz, 1829	12,17	41720
<i>Hypophthalmus oremaculatus</i> Nani & Fuster, 1947	16,17	36747
<i>Pimelodus blochii</i> Valenciennes, 1840	1,2,3,4,7,15,18	36711, 36723
<i>Pseudoplatystoma punctifer</i> (Castelnau, 1855)	16	36663
<i>Pseudoplatystoma tigrinum</i> (Valenciennes, 1840)	16	36662
<i>Sorubim lima</i> (Bloch & Schneider, 1801)	1,4,7,18	28028
<b>CICHLIFORMES</b>		
<b>Cichlidae</b>		
<i>Acarichthys heckelii</i> (Müller & Troschel, 1849)	2	28716, 36719
<i>Astronotus ocellatus</i> (Agassiz, 1831)	5	17092, 17387
<i>Cichla monoculus</i> Spix & Agassiz, 1831	2,4,6	17363, 49977
<i>Heros spurium</i> Heckel, 1840	1,2	36742, 41831
<i>Pterophyllum scalare</i> (Schultze, 1823)	2	23711
<b>PERCIFORMES</b>		
<b>Sciaenidae</b>		
<i>Plagioscion squamosissimus</i> (Castelnau, 1855)	1,2,7,9,12,15,17,18	32078, 48866

the first time flooded lakes from RDS-PP during the dry season. As expected, composition of ichthyofauna changed significantly, with 44 new species recorded by us for this region.

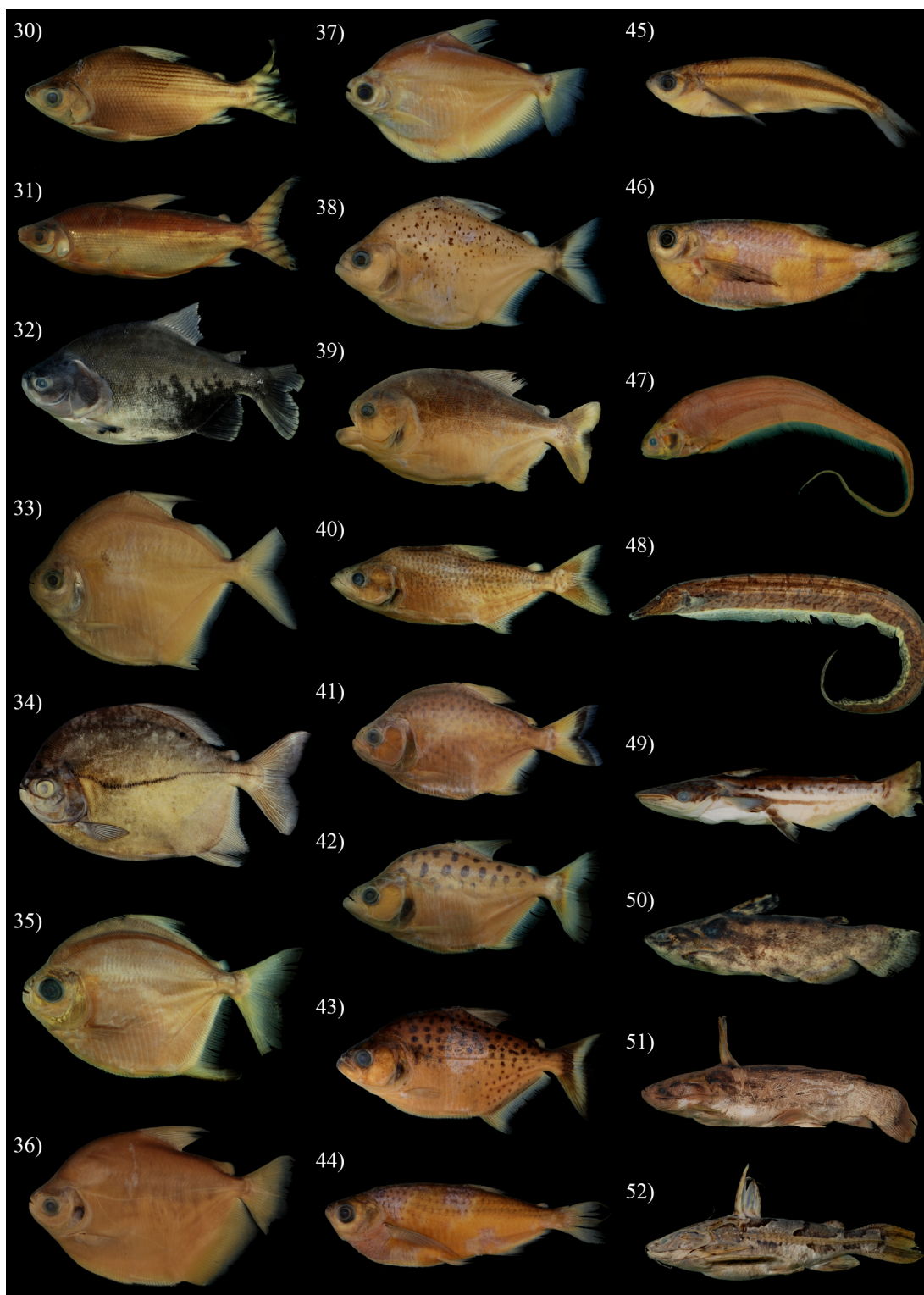
The most abundant and frequent species, occurring in more than 60% of sampled lakes, including open access and protected, were *Pygocentrus nattereri*, *Triportheus angulatus*, *Serrasalmus* sp. 2n=58, *Pterygoplichthys gibbiceps*, and *Osteoglossum bicirrhosum*, all consumed by local population of RDS-PP. Similarly, in floodplain lakes of another reserve (RDS Mamirauá) in Solimões river, piranhas (*Serrasalmus* spp. and *P. nattereri*) and aruanã (*Osteoglossum bicirrhosum*) were the main collected species (Silvano et al. 2009). *Osteoglossum bicirrhosum* is a relevant artisanal fisheries resource in the entire Amazon (Lowry et al. 2005), and is also used as ornamental in aquarium trades (Santos et al. 2006). Apart from its intense exploitation, the aruanã was the third most abundant with the second highest biomass registered by us. This success can be attributed to the generalist feeding behavior, hunting along lake borders and flooded forests, allied to

capacity of exploiting allochthonous food items during the high-water season (Saint-Paul et al. 2000, Lowry et al. 2005, Castillo et al. 2012), by leaping from the water to seek allochthonous prey, including terrestrial invertebrates (Aragão 1984, Goulding 1980) and small vertebrates (Verba et al. 2018).

In the comparison between the two lakes categories, open access presented less number of species (48 vs. 62 in protected). Nevertheless, from the 12 exclusively captured in these lakes, several possess elevated importance for artisanal and commercial fisheries, as *Pellona flavipinnis*, *Semaprochilodus taeniurus*, *Mylossoma aureum*, and the cichlids *Astronotus ocellatus* and *Cichla monoculus*, indicating the efficacy of choice of these lakes for fish exploration. On the other hand, the exclusive capture of *Colossoma macropomum* in protected lakes in RDS Piagaçu-Purus, indicates the efficiency of fishing management model adopted, that had already registered a major probability of capture and higher abundance of this species in protected lakes of RDS Mamirauá (Silvano et al. 2009).

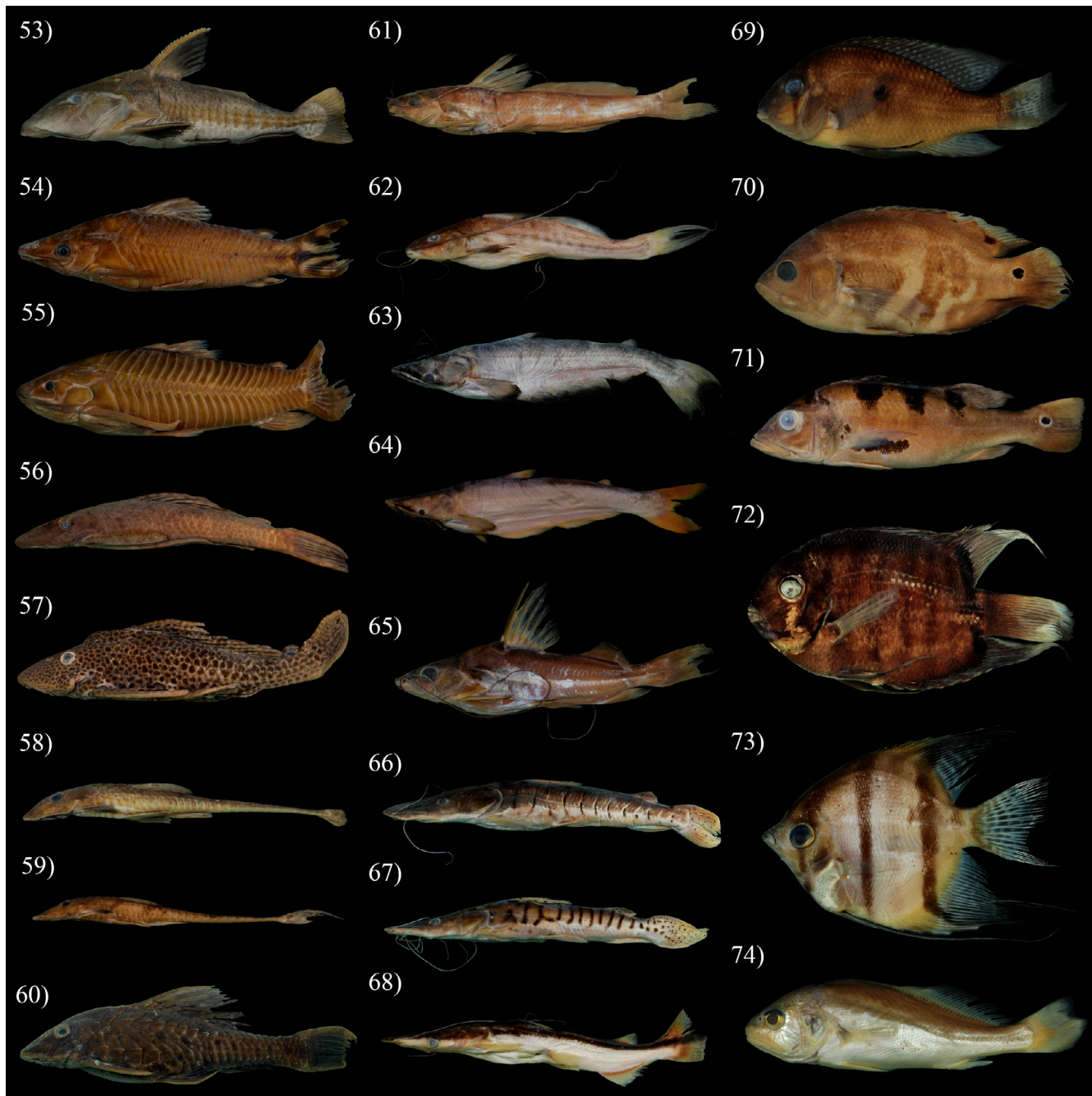


**Figure 2.** Fish species from floodplain lakes of Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), lower rio Purus, Amazonas State, Brazil. 1) *Osteoglossum bicirrhosum* INPA 17228, 278.8 mm SL; 2) *Pellona flavipinnis* INPA 34030, 231.0 mm SL; 3) *Acestrorhynchus* cf. *pantaneiro* INPA 23749, 212.8 mm SL; 4) *Acestrorhynchus microlepis* INPA 17152, 133.0 mm SL; 5) *Leporinus friderici* INPA 48861, 186.7 mm SL; 6) *Leporinus parae* INPA 36734, 132.5 mm SL; 7) *Megaleporinus trifasciatus* INPA 36724, 178.5 mm SL; 8) *Rhytiodus microlepis* INPA 41882, 166.8 mm SL; 9) *Schizodon fasciatus* INPA 36599, 131.4 mm SL; 10) *Brycon amazonicus* INPA 17122, 135.8 mm SL; 11) *Brycon melanopterus* INPA 36665, 163.7 mm SL; 12) *Chalceus erythrurus* INPA 17235, 174.2 mm SL; 13) *Roeboides myersii* INPA 36573, 108.1 mm SL; 14) *Tetragonopterus argenteus* INPA 36718, 64.2 mm SL; 15) *Tetragonopterus chalceus* INPA 42420, 91.1 mm SL; 16) *Boulengerella maculata* INPA 42394, 192.9 mm SL; 17) *Curimata inornata* INPA 23638, 130.5 mm SL; 18) *Curimatella alburna* INPA 36708, 148.8 mm SL; 19) *Potamorhina altamazonica* INPA 36746, 172.3 mm SL; 20) *Potamorhina pristigaster* INPA 36730, 172.2 mm SL; 21) *Psectrogaster amazonica* INPA 36594, 114.2 mm SL; 22) *Psectrogaster rutiloides* INPA 36558, 126.3 mm SL; 23) *Steindachnerina bimaculata* INPA 36626, 93.0 mm SL; 24) *Cynodon gibbus* INPA 23746, 207.7 mm SL; 25) *Rhaphiodon vulpinus* INPA 32043, 220.0 mm SL; 26) *Hoplias malabaricus* INPA 48374, 100.8 mm SL; 27) *Hemiodus* sp. "rabo de fogo" INPA 36706, 151.0 mm SL; 28) *Bryconops melanurus* INPA 36709, 94.4 mm SL; 29) *Prochilodus nigricans* INPA 41661, 155.6 mm SL.



**Figure 3.** Fish species from floodplain lakes of Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), lower rio Purus, Amazonas State, Brazil. 30) *Semaprochilodus insignis* INPA 41691, 128.4 mm SL; 31) *Semaprochilodus taeniurus* INPA 42456, 222.5 mm SL; 32) *Colossoma macropomum* INPA 58771, 357.6 mm SL; 33) *Metynnis longipinnis* INPA 39502, 98.9 mm SL; 34) *Myloplus* aff. *asterias* INPA 58772, 189.3 mm SL; 35) *Myloplus torquatus* INPA 36702, 78.8 mm SL; 36) *Mylossoma aureum* INPA 41655, 129.6 mm SL; 37) *Mylossoma albiscopum* INPA 36686, 124.8 mm SL; 38) *Pristobrycon striolatus* INPA 41917, 102.5 mm SL; 39) *Pygocentrus nattereri* INPA 41689, 168.9 mm SL; 40) *Serrasalmus elongatus* INPA 36705, 134.4 mm SL; 41) *Serrasalmus maculatus* INPA 36699, 116.8 mm SL; 42) *Serrasalmus rhombeus* INPA 36693, 109.0 mm SL; 43) *Serrasalmus* sp. "2n=58" INPA 36670, 104.0 mm SL; 44) *Triportheus angulatus* INPA 36713, 126.9 mm SL; 45) *Triportheus auritus* INPA 36666, 94.4 mm SL; 46) *Triportheus rotundatus* INPA 36744, 94.8 mm SL; 47) *Eigenmannia macrops* INPA 36717, 183.1 mm SL; 48) *Rhamphichthys pantherinus* INPA 27614, 573.9 mm SL; 49) *Ageneiosus inermis* INPA 41700, 126.9 mm SL; 50) *Trachelyopterus galeatus* INPA 41790, 80.4 mm SL; 51) *Trachychorystis porosus* INPA 36732, 102.0 mm SL; 52) *Anadoras grypus* INPA 36733, 136.1 mm SL.

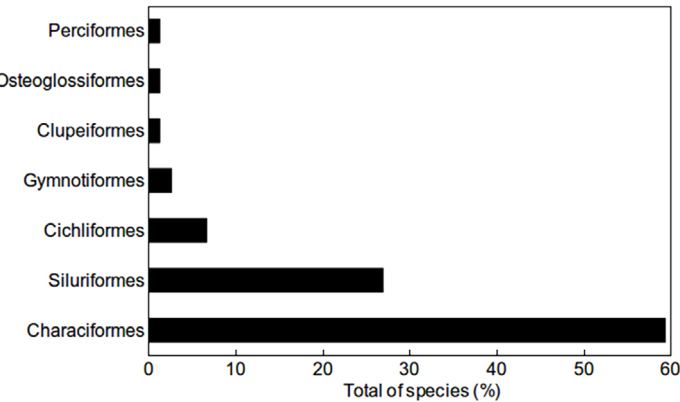




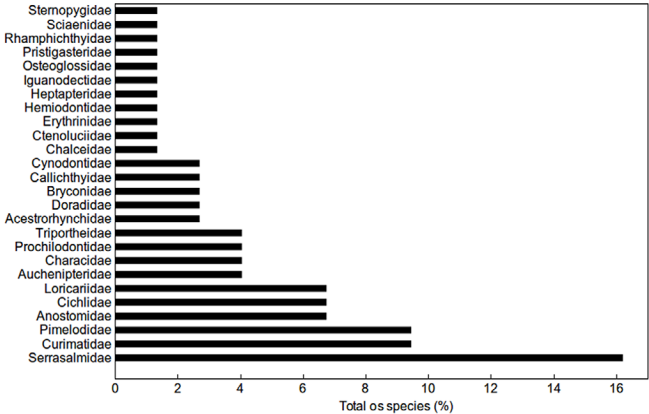
**Figure 4.** Fish species from floodplain lakes of Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), lower rio Purus, Amazonas State, Brazil. 53) *Oxydoras niger* INPA 36731, 163.0 mm SL; 54) *Dianema urostriatum* INPA 36722, 99.6 mm SL; 55) *Hoplosternum littorale* INPA 36710, 140.9 mm SL; 56) *Dekeyseria amazonica* INPA 29206, 173.9 mm SL; 57) *Pterygoplichthys gibbiceps* INPA 36664, 251.2 mm SL; 58) *Loricariichthys acutus* INPA 36721, 208.1 mm SL; 59) *Loricariichthys nudirostris* INPA 34633, 95.9 mm SL; 60) *Pseudorinelepis genibarbis* INPA 36745, 140.3 mm SL; 61) *Pimelodella gr. cristata* INPA 36741, 120.1 mm SL; 62) *Calophysus macropterus* INPA 36743, 113.6 mm SL; 63) *Hypophthalmus oremaculatus* INPA 36747, 185.2 mm SL; 64) *Hypophthalmus edentatus* INPA 41720, 198.7 mm SL; 65) *Pimelodus blochii* INPA 36711, 120.6 mm SL; 66) *Pseudoplatystoma punctifer* INPA 36662, 208.2 mm SL; 67) *Pseudoplatystoma tigrinum* INPA 36663, 275.5 mm SL; 68) *Sorubim lima* INPA 28208, 215.7 mm SL; 69) *Acarichthys heckelii* INPA 28716, 101.4 mm SL; 70) *Astronotus ocellatus* INPA 17387, 170.4 mm SL; 71) *Cichla monoculus* INPA 49977, 240.2 mm SL; 72) *Heros spurius* INPA 41831, 123.4 mm SL; 73) *Pterophyllum scalare* INPA 23711, 80.3 mm SL; 74) *Plagioscion squamosissimus* INPA 32078, 149.6 mm SL.



Ichthyofauna from floodplain lakes of rio Purus



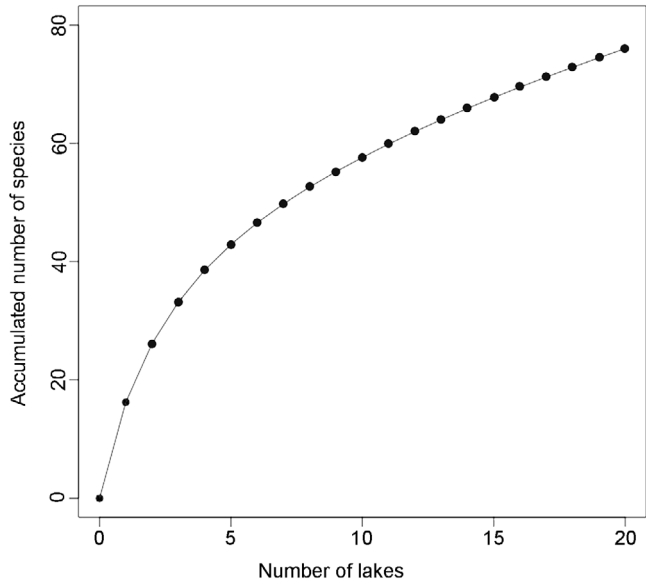
**Figure 5.** Percentages of the total number of collected species of each order in floodplain lakes from rio Purus, Amazonas State, Brazil.



**Figure 6.** Percentages of the total number of collected species of each family in floodplain lakes from rio Purus, Amazonas State, Brazil.

**Table 3.** Participation of Teleostei orders in total abundance and species richness observed in protected, open access lakes and shared by protected and open access lakes (overlap) of the lower rio Purus.

Orders	Protected		Open access		Protected + Open access	
	Species Richness	Abundance	Species Richness	Abundance	Species Richness	Abundance
Characiformes	15	69	5	7	24	1816
Siluriformes	10	79	2	6	8	194
Cichliformes	0	0	5	9	0	0
Perciformes	0	0	0	0	1	12
Clupeiformes	0	0	0	0	1	27
Osteoglossiformes	0	0	0	0	1	76
Gymnotiformes	1	1	0	0	1	3
Total	26	149	12	22	36	2128



**Figure 7.** Species accumulation curve of ichthyofauna from floodplain lakes of Reserva de Desenvolvimento Sustentável Piagaçu-Purus (RDS-PP), considering the total number of sample units.

However, this can be a consequence of the overexploitation of *C. macropomum* in open access lakes due to its relevance as feed source of protein for entire Amazon population, but not necessarily mean that such management actions have been ineffective. In marine systems, for example, the main goal of major management is to improve fisheries, allowing some fish landings in managed areas (common-based management), without affecting fish species diversity and ecological integrity (Mcclanahan et al. 1997). Though, the model of integral protection of the lakes adopted in RDS-PP focuses in whole habitat protection, preserving ecological patterns and complexity, and seems to be the factor supporting the ecological stability, beyond providing the local population the most appreciated species for feed consumption.

RDS Piagaçu-Purus provides legal context for monitoring wildlife along with the local population, gathering data and developing strategies for sustainable use. Additionally, it forms a mosaic of protected areas ruled by several different regimes, with two indigenous reserves (Terra Idígena Lago Ayapua and Itixi Mitari), Reserva Biológica Abufari (REBIO) and Parque Nacional Nascentes do Lago Jari (PARNA), consisting an important region for conservation of ichthyofauna, especially considering fishing productivity of rio Purus. However, recent models of future deforestation in Amazonia predict an increase in the

loss of vegetation cover for the RDS-PP, related to the reactivation of the road BR-319 (Manaus - Porto Velho), which percuss the interfluvium between Madeira and Purus rivers (Soares-Filho et al. 2006, Fearnside et al. 2009, Vieira et al. 2015).

Considering all registers published for the rio Purus basin (*i.e.* La Monte 1935, Rapp Py-Daniel & de Deus 2003, Fernandes et al. 2004, Anjos et al. 2008, Duarte et al. 2010; Claro-García et al. 2013, and the present) 360 currently valid species compose the ichthyofauna of this river basin. Still, we strongly believe that this number is underestimated, with much of the basin still unexplored or only sparsely sampled. Our surveys beyond assessing and illustrating for the first time the ichthyofauna composition of floodplain lakes of RDS Piagaçu-Purus, revealed the occurrence of at least three new species for science in rio Purus, *Hemiodus* sp. “rabo de fogo”, *Myloplus* aff. *asterias* (under description by RPO et al.), and *Serrasalmus* sp. 2n=58. Principally, our results indicate the potential efficiency of zoning systems of open access and protected lakes established by local population and ruled by RDS-PP for fisheries management. Therefore, we strongly suggest its maintenance for conservation of ichthyofauna of floodplain lakes of lower rio Purus.

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## Author Contributions

Bruno Ferezim Morales: Substantial contribution in the concept and design of the study, contribution to data collection, analysis and interpretation, contribution to manuscript preparation, contribution to critical revision, adding intellectual content.

Rafaela Priscila Ota: Substantial contribution in the concept and design of the study, contribution to data collection, analysis and interpretation, contribution to manuscript preparation, contribution to critical revision, adding intellectual content.

Victória Dandara Pereira e Silva: Contribution for data collection, analysis and interpretation, contribution to critical revision, adding intellectual content.

Cláudia Pereira de Deus: Substantial contribution in the concept and design of the study, contribution for data collection, contribution to critical revision, adding intellectual content.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Survey of fish species from the Lower Roosevelt River, Southwestern Amazon basin

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**Abstract:** This study presents an inventory of the ichthyofauna of the lower Roosevelt River sub-basin and its associated tributaries. Fish sampling with fishing nets and measurements of environmental parameters of water occurred in November/2012 (rising water), February/2013 (flooding), May/2013 (falling water) and August/2013 (drought). Depth mean was 8.86 m, water transparency was 0.6 m, conductivity was 22.7  $\mu\text{S}\cdot\text{cm}^{-1}$ , pH was 6.59, dissolved oxygen was 7.63  $\text{mg}\cdot\text{l}^{-1}$  and temperature was 28°C. The total estimated capture area was 68,829.6  $\text{m}^2$  during 2,880 hours. The catch per unit Effort (CPUE) was 0.37 individuals  $\text{m}^{-2}\cdot\text{day}^{-1}$ . Species were spatially aggregated in all sampling points and river water levels. A total of 5,183 individuals distributed in 7 orders, 29 families, 104 genders and 188 species were sampled in this survey. The diversity index was 4.121 and equitability index was 0.789. The Characiforms order was the most abundant with 106 species, followed by Siluriforms with 63 species and Cichliforms with 23 species. The most abundant species was *Serrasalmus rhombeus* (Linnaeus, 1766) with 327 individuals (5.9%), followed by *Chalceus epakros* (Cope, 1870) with 309 individuals (5.6%) and *Acestrorhynchus microlepis* (Schomburgk, 1841) with 250 individuals (4.5%). Trophicity was characterized by omnivorous (28.6%), piscivorous (14.3%), carnivorous (13.8%) and detritivorous (12.8%). According to IBAMA's regulation, 29.25% of captured species presents ornamental potential.

**Keywords:** Ichthyofauna; Inventory; Biodiversity; Madeira River Basin.

## Levantamento de espécies de peixes do Baixo Rio Roosevelt, Sudoeste da Bacia Amazônica

**Resumo:** Este estudo apresenta um inventário da ictiofauna da sub-bacia do baixo Rio Roosevelt e seus tributários associados. As coletas de peixes com malhadeiras e as medições de parâmetros ambientais da água ocorreram em Novembro/2012 (enchente), Fevereiro/2013 (cheia), Maio/2013 (vazante) e Agosto/2013 (seca). A média da profundidade foi 8,86 m, da transparência da água foi 0,6 m, da condutividade foi 22,7  $\mu\text{S}\cdot\text{cm}^{-1}$ , do pH foi 6,59, do oxigênio dissolvido foi 7,63  $\text{mg}\cdot\text{l}^{-1}$  e da temperatura da água foi 28°C. A área total de captura estimada foi 68.829,6  $\text{m}^2$  durante 2880 horas. A captura por unidade de esforço (CPUE) foi 0,37 indivíduos  $\text{m}^{-2}\cdot\text{dia}^{-1}$ . As espécies foram espacialmente agregadas em todos os pontos de coleta e períodos de coleta. Um total de 5183 peixes em 7 ordens, 29 famílias, 104 gêneros e 188 espécies foram coletados. O índice de diversidade foi 4,121 e o índice de equidade foi 0,789. As ordens Characiforme, Siluriforme e Cichliforme foram as mais abundantes. As espécies *Serrasalmus rhombeus* Linnaeus 1766 com 327 indivíduos (5,9%), *Chalceus epakros* (Cope 1870) com 309 indivíduos (5,6%) e *Acestrorhynchus microlepis* Schomburgk 1841 com 250 indivíduos (4,5%) foram as mais abundantes. Os onívoros (28,6%), piscívoros (14,3%), carnívoros (13,8%) e detritívoros (12,8%) foram os indivíduos mais abundantes. De acordo com o IBAMA, 29,25% das espécies capturadas tem potencial ornamental.

**Palavras-chave:** Ictiofauna; Inventário; Biodiversidade; Bacia do Rio Madeira.



## Introduction

Roosevelt River is a clear water tributary of the right-bank of the Aripuanã River which is an important tributary of the east side of the Madeira River basin (Pedroza et al., 2012). Nine different protected areas in the Southeast of Amazonas state comprises the Mosaic of Apuí with approximately 2.5 million hectares (Ribeiro et al., 2011). This mosaic has an important role to contain the spread of the arc of deforestation, minimizing the loss of biodiversity. Unsustainable human practices such as hydropower expansion (Lees et al., 2016), deforestation (Soares-Filho et al., 2014) and mining (Meira et al., 2016) are imperiling the remarkable biodiversity of the Amazon River Basin.

Neotropical freshwater fishes are the most diverse on the planet with more than 4,000 species described (Toussaint et al., 2016), representing about one-third of all freshwater fishes worldwide (Reis et al., 2016). National policies in most countries in the Latin America historically encouraged unsustainable practices over the preservation of fish biodiversity (Pelicice et al., 2017). In this case, Neotropical region can be considered a hotspot for fish conservation. However, 28% of the known fauna was described in just the past 11 years and most reasonable estimates for the actual total number of freshwater fishes in the Neotropical region exceed 8000 species (Reis et al., 2016).

Nearly half of the Neotropical fish species are known to occur in Brazil, with at least 2,587 species (Buckup et al. 2007), but probably more than 1,000 fish species were not yet described (Junk et al., 2007). On the other hand, São Francisco River Basin has 200 fish species (Alves & Pompeu, 2001) and Paraguay River Basin has about 330 estimated species (Reis et al., 2003) which is a reasonable well-studied Brazilian basin. Studies over the Brazilian ichthyofauna are still recent (Camargo & Giarizzo, 2007; Perin et al., 2007; Rapp Py-Daniel et al., 2007; Araújo et al., 2009; Pedroza et al., 2012; Queiroz et al., 2013) when compared to another Amazonian region (Lauzanne & Loubens, 1985; Lauzanne et al., 1991; Chernoff et al., 2000). Recent studies indicate high species richness in Madeira River tributaries (Rapp Py-Daniel et al., 2007; Torrente-Vilara et al., 2011; Pedroza et al., 2012), numbering over 900 species (Queiroz et al., 2013).

Most studies of the Amazonian ichthyofaunal diversity have concentrated in the floodplains adjacent to large rivers and next to urban areas, but there are few reports in areas of high conservation value (Costa et al., 2017). Ichthyological surveys assess the biodiversity of water bodies (Silveira et al., 2010), resulting in new discoveries of undescribed species (Frota et al., 2016) and basis for conservation actions (Ferreira et al., 2017). Improving scientific information from conservation sites is crucial for guiding policy and management decisions (Willink et al., 2013), such as for fishery management (Agostinho et al., 2016). In order to know the ichthyofauna from part of Southwestern Amazon basin, this study provides a survey of fish in the Lower Roosevelt River and some of its tributaries.

## Material and Methods

### 1. Study area

The study area is located at the lower Roosevelt River sub-basin and its small associated tributaries (Figure 1). The Roosevelt River is a clear water tributary on the right bank of the Aripuanã River, one of

the most important tributaries on the east side of the Madeira River Basin (Anjos et al., 2016). The 30 sampling points were distributed over 168 km between parallels 7° and 8° S, and meridians 60° and 61° (Table 1). Riparian forest established along its shores and Open Ombrophilous forest over the sub-basin were well preserved. According to Köppen classification, the regional climate is Am which represents a tropical monsoon climate with annual rainfall around 2,800 mm per year (Alvares et al., 2014). The wet season is from October to March and the dry season from June to August (Vidotto et al., 2007).

### 2. Stream and fish sampling

A graduated ruler and Secchi disk were used to measure water depth and transparency, respectively. A portable multiparameter probe (YSI 6600, YSI Environmental Company, Bahrain) was used in each point to measure conductivity, pH, dissolved oxygen and temperature. Fish sampling lasted 20 days and occurred following river water levels: rising water (November/2012), flooding (February/2013), falling water (May/2013) and drought (August/2013). Chico Mendes Institute for Biodiversity Conservation granted a fishing license (35382-1) for fish collection and transportation.

Fish were sampled using fishing nets of mesh sizes of 20, 30, 40, 50, 60, 70, 80, 90, 100, 120, 140, 160, 180 and 200 mm with 10 m long and height varying between 1.5 to 4.0 m. Total capture area was 573.58 m<sup>2</sup>.day<sup>-1</sup> per sampling point. Nets were visited every six hours. Sampled fish were anesthetized with Eugenol solution and subsequently fixed by immersion in 4% formaldehyde solution for at least 48 hours. Specimens were then washed and transferred to 70% ethanol.

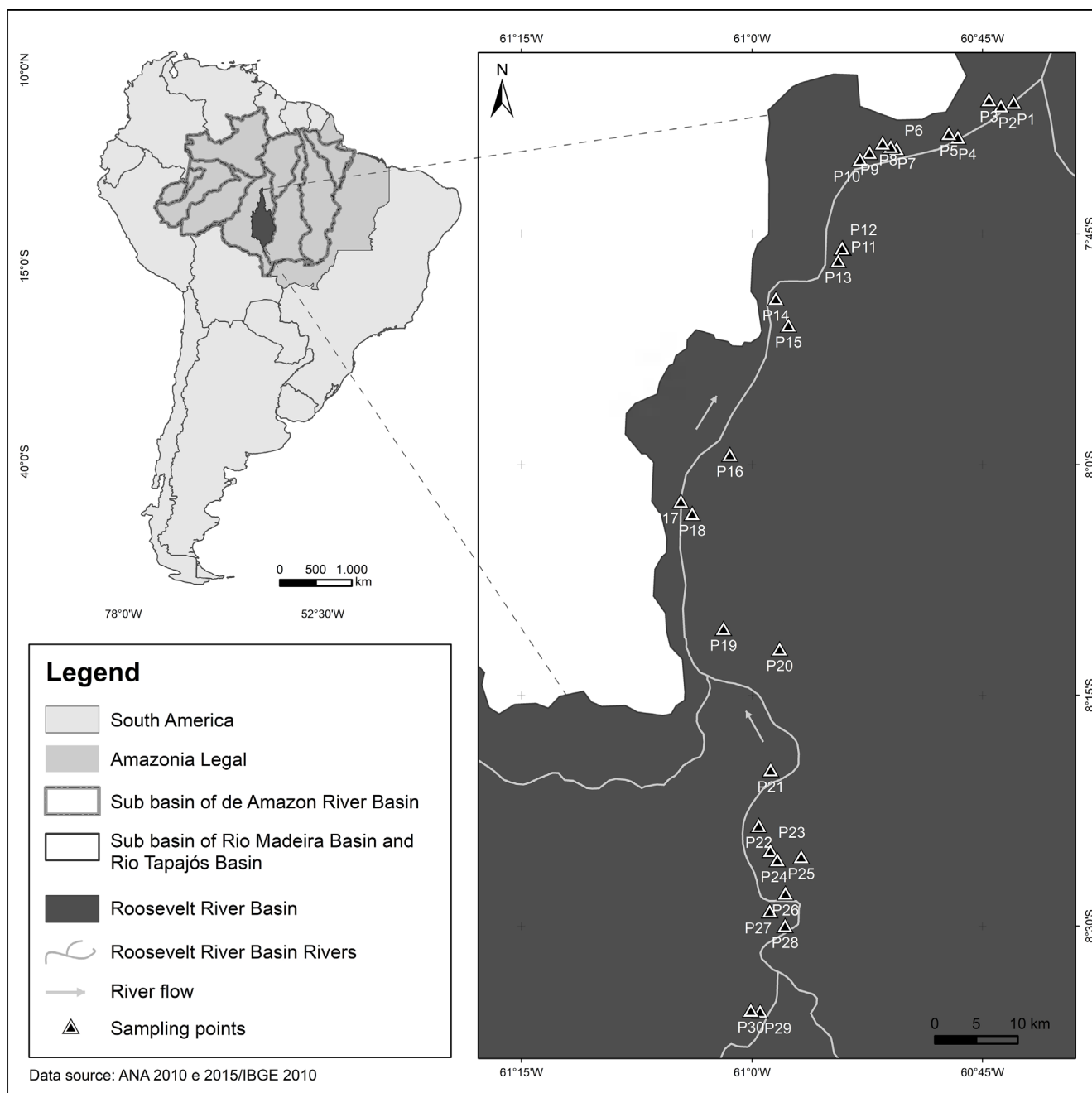
Fish identification was performed mainly using Lauzanne & Loubens (1985), Ferreira et al. (1998), Silvano (2001), Reis et al. (2003), Menezes et al. (2003), Buckup et al. (2007), Fricke & Eschmeyer (2019), Queiroz et al. (2013), and Van Der Laan, Fricke & Eschmeyer (2019). Voucher specimens were cataloged with labels which contained information on location, geographic coordinates, date and time of capture, type of environment, and fishing equipment used. They were deposited in the fish collection at Laboratório de Ictiologia e Ordenamento Pesqueiro do Vale do Rio Madeira (LIOP) in the Federal University of Amazonas (UFAM).

### 3. Data analysis

Fish with ornamental potential were defined according to IBAMA Normative Instruction number 001, from January 3, 2012. Fish species was checked on the Brazilian Red List established by Ordinances number 444/14 and 445/14 of the Ministry of the Environment.

## Results

Depth mean was 8.86 m, water transparency was 0.6 m, conductivity was 22.7 µS.cm<sup>-1</sup>, pH was 6.59, dissolved oxygen was 7.63 mg.l<sup>-1</sup> and temperature was 28°C (Table 2). The total estimated capture area was 68,829.6 m<sup>2</sup> during 2,880 hours. The catch per unit effort (CPUE) was 0.37 individuals m<sup>-2</sup>.day<sup>-1</sup>. Species were spatially aggregated in all sampling points and river water levels. A total of 5,183 individuals distributed in 7 orders, 29 families, 104 genders and 188 species were sampled in this survey (Table 3). The diversity index was 4.121 and equitability index was 0.789. The Characiforms order was the



**Figure 1.** Lower Roosevelt River sub-basin sampling points location, Brazilian Southwest Amazon.

most abundant with 106 species and 4,246 individuals, followed by Siluriforms with 63 species and 863 individuals and Cichliforms with 23 species and 276 individuals (Figure 2 and Table 3). The most abundant species was *Serrasalmus rhombeus* (Linnaeus, 1766) with 327 individuals (5.9%), followed by *Chalceus epakros* (Cope, 1870) with 309 individuals (5.6%) and *Acestrorhynchus microlepis* (Schomburgk, 1841) with 250 individuals (4.5%). Trophicity (Figure 3) was characterized by omnivorous (28.6%), piscivorous (14.3%), carnivorous (13.8%) and detritivorous (12.8%). According to IBAMA's regulation, 29.25% of captured species presents ornamental potential (Table 3).

## Discussion

Few species (about 2.13%) found in this survey were recorded by Pedroza et al. (2012) in the Roosevelt River, indicating a total of 209 species for this study area. The total number of species found in the present study is in accordance with other studies in the Amazon River Basin. Some studies recorded 67 fish species in the Tapajós River (Keppeler et al., 2016), 86 species in the Purus River (Anjos et al., 2008), 90 species in the Juruá River (Silvano et al., 2000), 90 species in the Teles Pires River (Dary et al., 2017), 133 species in the Madeira

**Table 1.** Description of sampling points and its location at Lower Roosevelt River, Southwestern Amazon basin.

Point	Locality	Latitude	Longitude	Environment
P1	Sereia Stream	S 07°36'26.3"	W 60°42'57.5"	Lotic
P2	Macimiano Lake	S 07°36'40.2"	W 60°43'47.2"	Lentic
P3	Piquiá Backwater	S 07°36'16.1"	W 60°44'34.1"	Lentic
P4	Pium Stream	S 07°38' 42.3"	W 60°46'35.8"	Lotic
P5	Ariranha Stream	S 07°38'28.5"	W 60°47'10.9"	Lotic
P6	Pium Backwater	S 07°39'27.0"	W 60°50'35.0"	Lentic
P7	Cutia Stream	S 07°39'18.4"	W 60°50'58.2"	Lotic
P8	Tracajá Backwater	S 07°39'05.4"	W 60°51'29.9"	Lentic
P9	Goiaba Brava Stream	S 07°39'41.9"	W 60°52'20.6"	Lotic
P10	Pedral Stream	S 07°40'08.9"	W 60°52'57.6"	Lotic
P11	Apuí Grande Stream	S 07°45'55.6"	W 60°54'04.0"	Lotic
P12	Apuizinho Stream	S 07°45'54.1"	W 60°54'07.7"	Lotic
P13	Sombra Backwater	S 07°46'43.8"	W 60°54'23.3"	Lentic
P14	Torre da Lua Stream	S 07°49'11.4"	W 60°58'26.3"	Lotic
P15	Piranha Stream	S 07°50'55.1"	W 60°57'36.9"	Lotic
P16	Gavião Stream	S 07°59'20.2"	W 61°01'25.7"	Lotic
P17	Praia Stream	S 08°02'22.1"	W 61°04'36.8"	Lotic
P18	Camponesa Stream	S 08°03'09.6"	W 61°03'52.8"	Lotic
P19	Machadinho Stream	S 08°10'38.0"	W 61°01'50.9"	Lotic
P20	Cujubim Stream	S 08°11'57.3"	W 60°58'11.6"	Lotic
P21	Morcega Stream	S 08°19'49.0"	W 60°58'46.5"	Lentic
P22	Zé Comprido Pit	S 08°23'26.0"	W 60°59'33.2"	Lentic
P23	Inferninho Pit	S 08°25'04.2"	W 60°58'47.6"	Lentic
P24	Diogo Pit	S 08°25'40.2"	W 60°58'20.4"	Lentic
P25	Perneta Pit	S 08°25'28.0"	W 60°56'47.3"	Lentic
P26	Glória Pit	S 08°27'49.9"	W 60°57'48.9"	Lentic
P27	Esperança Pit	S 08°28'59.7"	W 60°58'50.4"	Lentic
P28	Santa Rita Pit	S 08°29'56.6"	W 60°57'50.3"	Lentic
P29	Pirarara Pit	S 08°35'30.4"	W 60°59'27.7"	Lentic
P30	Tucunaré Lake	S 08°35'25.8"	W 61°00'04.1"	Lentic

River Basin (Camargo & Giarrizzo, 2007), 148 species in the Xingu River (Fitzgerald et al., 2017) and 160 species in the Guariba River (Pedroza et al., 2012).

The Amazon River Basin contains the highest fish species diversity of any region on earth (Reis et al., 2003). The biodiversity results from processes operating at multiple spatial and temporal scales (Peláez & Pavanelli, 2018). Heterogenous environments can contribute to maintain biodiversity (Peláez et al., 2017). A strong environmental control on species composition is expected at intermediate spatial scales, where dispersal is neither too high to mask the effects of environmental variables (Heino et al., 2015) nor too low for the differences in species composition to be related to historical processes (Villéger et al., 2013). A major environmental factor on the Amazon Basin system is the water seasonal variation that constitutes an annual hydrological cycle, with changes in water level that can exceed 15 m between high and low water periods that can strongly affect fish assemblages (Scarabotti et al., 2011). Changes in environmental variables over the hydrologic seasons of the

year are likely to change the relative importance of biotic interactions such as predation and competition, which may increase when low water crowds populations, creating non-random assortments of fish species (Fernandes et al., 2009). Abiotic influences such as temperature, oxygen concentration, and transparency also change over the hydrologic cycle and differ among water bodies, which can be the basis of habitat selection among fish (Freitas et al., 2010; Miyazono et al., 2010; Van der Wolfshaar et al., 2011).

Characiformes and Siluriformes were the predominant orders, following the Neotropical pattern for freshwater fish diversity (Lowell-McConnell, 1999). We emphasize that none of the sampled species are on the Brazilian Red List. The higher number of species registered in this study is probably due to the environmental heterogeneity (Teresa et al., 2010). However, the diversity may have been underestimated. Several sampled species were discriminated with the use of "cf", indicating that the number of new species may be higher. Ten taxa were provisionally identified, due to their uncertain taxonomic status. They may be records

**Table 2.** Environmental variables and Morisita index (If) to the survey of fish species from the Lower Roosevelt River, Southwestern Amazon basin.

Points	Depth (m)	Transparency (cm)	Conductivity ( $\mu\text{S}\cdot\text{cm}^{-1}$ )	pH	Dissolved O2 ( $\text{mg}\cdot\text{L}^{-1}$ )	Temperature (°C)	If
Min-max(mean)							
P1	2.1-8.2 (4.9)	35.0-80.0 (58.8)	9.3-26.8 (17.0)	6.3-7.4 (6.9)	8.5-9.8 (9.0)	25.6-31.1 (28.2)	10.53
P2	3.8-8.6 (5.6)	30.0-75.0 (53.0)	8.0-32.5 (17.9)	6.4-7.5 (6.7)	8.4-9.8 (8.9)	25.6-31.4 (28.5)	7.68
P3	3.1-13.5 (7.9)	33.0-80.0 (56.0)	8.8-28.0 (17.1)	6.6-7.3 (6.9)	9.2-9.8 (9.4)	26.4-31.2 (28.7)	7.39
P4	2.7-14.3 (7.8)	56.0-80.0 (69.0)	9.3-34.0 (26.6)	6.6-7.3 (6.9)	7.3-9.7 (8.2)	25.5-31.1 (28.3)	9.62
P5	3.8-15.7 (8.9)	38.0-85.0 (65.8)	9.3-35.5 (27.3)	6.2-7.3 (6.9)	7.4-9.9 (8.2)	24.7-31.2 (27.9)	12.26
P6	4.2-5.4 (4.6)	38.0-86.0 (64.8)	11.3-36.3 (27.5)	6.0-7.1 (6.5)	9.5-10.0 (9.7)	27.2-30.3 (28.5)	8.04
P7	3.3-12.3 (7.0)	45.0-85.0 (65.0)	10.5-36.0 (29.6)	6.4-7.1 (6.8)	7.1-9.9 (8.1)	26.2-30.4 (28.4)	7.27
P8	5.2-13.8 (8.1)	53.0-85.0 (68.3)	9.8-31.0 (22.3)	6.6-7.2 (7.0)	9.1-9.7 (9.3)	25.4-30.6 (27.8)	10.92
P9	4.1-12.9 (7.5)	55.0-100.0 (76.3)	9.8-25.5 (16.9)	6.6-7.3 (7.0)	8.6-9.9 (9.1)	25.7-30.8 (28.0)	22.83
P10	2.3-9.1 (5.1)	60.0-100.0 (77.5)	9.8-26.0 (16.8)	6.5-7.3 (6.9)	7.3-9.8 (8.2)	25.9-30.8 (28.3)	6.90
P11	3.2-8.5 (5.1)	50.0-85.0 (67.5)	7.3-25.5 (16.2)	5.9-7.5 (6.8)	8.8-10.0 (9.4)	25.5-30.5 (26.8)	7.61
P12	1.9-12.6 (6.5)	36.0-70.0 (56.5)	8.8-28.0 (16.7)	6.2-7.7 (6.6)	8.5-9.9 (9.4)	26.8-31.7 (28.8)	9.12
P13	2.4-11.3 (5.9)	38.0-80.0 (60.8)	10.0-27.3 (17.1)	6.3-8.1 (7.2)	7.4-10.4 (8.5)	24.8-31.5 (27.9)	7.86
P14	4.4-11.9 (7.9)	40.0-80.0 (65.0)	12.0-27.0 (17.6)	6.4-7.0 (6.7)	6.3-7.0 (6.6)	26.5-30.7 (27.9)	15.73
P15	4.1-7.3 (5.7)	40.0-80.0 (62.5)	13.5-25.3 (17.6)	6.6-6.9 (6.7)	6.7-7.7 (7.1)	26.0-29.1 (27.7)	7.20
P16	3.4-10.1 (6.0)	38.0-70.0 (53.3)	10.0-31.3 (25.9)	6.4-7.2 (6.8)	6.6-9.6 (7.4)	25.5-30.2 (27.3)	11.33
P17	4.1-9.1 (5.8)	40.0-80.0 (61.3)	15.3-35.8 (30.6)	6.5-6.7 (6.7)	5.2-6.8 (5.8)	25.4-30.2 (27.6)	10.68
P18	2.8-13.7 (7.1)	37.0-70.0 (56.8)	15.5-35.8 (30.7)	6.2-6.6 (6.4)	5.7-9.2 (7.0)	25.3-29.4 (27.9)	8.94
P19	2.2-9.3 (5.5)	50.0-150.0 (95.0)	16.0-32.0 (28.0)	5.5-6.1 (5.8)	5.7-8.3 (6.7)	26.4-29.1 (27.5)	7.89
P20	1.7-12.2 (6.3)	20.0-30.0 (27.5)	15.3-29.3 (24.5)	5.5-6.7 (6.1)	6.1-9.4 (7.3)	24.7-28.9 (26.9)	21.24
P21	3.7-12.6 (6.9)	35.0-80.0 (62.5)	15.0-35.8 (30.6)	6.5-7.5 (6.8)	6.3-8.9 (7.3)	24.7-30.6 (27.5)	8.56
P22	6.8-16.9 (10.3)	35.0-75.0 (61.3)	13.8-32.3 (27.6)	5.7-6.6 (6.3)	6.6-8.4 (7.4)	25.6-29.2 (26.8)	12.39
P23	4.1-7.3 (6.0)	37.0-85.0 (65.5)	14.0-32.5 (24.9)	5.4-6.7 (6.3)	4.6-8.4 (5.9)	24.7-29.9 (28.0)	15.33
P24	5.1-16.6 (9.7)	36.0-80.0 (62.8)	13.5-32.5 (23.6)	5.7-6.6 (6.4)	6.6-9.3 (7.7)	25.7-29.2 (27.9)	6.02
P25	8.3-21.5 (12.8)	34.0-75.0 (58.5)	11.3-30.5 (18.7)	5.3-6.7 (6.2)	5.6-8.0 (6.5)	24.7-30.0 (27.7)	5.46
P26	9.4-27.4 (15.8)	38.0-85.0 (63.3)	16.5-27.3 (20.1)	5.3-6.7 (6.2)	5.5-8.2 (6.5)	25.6-29.0 (27.4)	6.38
P27	7.5-25.6 (14.1)	39.0-90.0 (64.8)	11.3-26.8 (17.0)	5.5-6.8 (6.2)	6.1-8.4 (6.9)	26.3-29.9 (28.1)	8.43
P28	3.3-11.3 (6.5)	55.0-75.0 (65.0)	10.3-27.3 (17.3)	6.0-6.7 (6.3)	6.0-8.5 (6.9)	25.7-29.7 (27.8)	6.09
P29	4.3-14.9 (8.5)	15.0-30.0 (23.8)	12.0-27.5 (18.5)	5.7-6.7 (6.2)	6.5-8.6 (7.2)	26.6-30.0 (28.0)	6.99
P30	5.5-18.1 (10.3)	15.0-30.0 (23.8)	11.5-24.5 (16.9)	5.5-6.6 (6.1)	6.5-8.5 (7.2)	25.5-30.2 (27.8)	12.56

**Table 3.** Survey of fish species from the Lower Roosevelt River, Southwestern Amazon basin, indicating number of captured individuals (N), ornamental potential (OP) and trophicity.

Order/Family/Specie	N	OP	Trophicity
<b>BELONIFORMS: Belonidae</b>			
<i>Potamorhaphis</i> sp.	19	yes	unknown
<i>Pseudotyloturus microps</i> (Günther, 1866)	04		unknown
<b>CHARACIFORMS: Acestrorhynchidae</b>			
<i>Acestrorhynchus falcirostris</i> (Cuvier, 1819)	9	yes	piscivorous
<i>Acestrorhynchus heterolepis</i> (Cope, 1878)	1		piscivorous
<i>Acestrorhynchus microlepis</i> (Schomburgk, 1841)	250	yes	piscivorous
<b>CHARACIFORMS: Anostomidae</b>			
<i>Anostomoides laticeps</i> (Eigenmann, 1912)	10		omnivorous
<i>Hypomasticus pachycheilus</i> (Britski, 1976)	3		unknown



Continuation Table 3.

Order/Family/Specie	N	OP	Trophicity
<i>Laemolyta proxima</i> (Garman, 1890)	9		omnivorous
<i>Laemolyta taeniata</i> (Kner, 1859)	7	yes	omnivorous
<i>Leporellus vittatus</i> (Valenciennes, 1850)	1	yes	omnivorous
<i>Leporinus aripuanensis</i> (Garavello & dos Santos, 1981)	2		omnivorous
<i>Leporinus brunneus</i> (Myers, 1950)	63		omnivorous
<i>Leporinus cylindriiformes</i> (Borodin, 1929)	14		omnivorous
<i>Leporinus desmotes</i> (Fowler, 1914)	70		omnivorous
<i>Leporinus fasciatus</i> (Bloch, 1794)	83	yes	omnivorous
<i>Leporinus friderici</i> (Bloch, 1794)	82		omnivorous
<i>Leporinus jamesi</i> (Garman, 1929)	4		omnivorous
<i>Leporinus polymaculatus</i> (Géry, 1977)	1		omnivorous
<i>Pseudanos gracilis</i> (Kner, 1859)	1	yes	omnivorous
<i>Schizodon fasciatus</i> (Spix & Agassiz, 1829)	1		herbivorous
<b>CHARACIFORMS: Bryconidae</b>			
<i>Brycon amazonicus</i> (Spix & Agassiz, 1829)	24		omnivorous
<i>Brycon</i> cf. <i>pesu</i> (Müller & Troschel, 1845)	14		omnivorous
<i>Brycon falcatus</i> (Müller & Troschel, 1844)	54		omnivorous
<i>Brycon melanopterus</i> (Cope, 1872)	3		omnivorous
<i>Brycon pesu</i> (Müller & Troschel, 1845)	55		omnivorous
<i>Brycon</i> sp.	12		omnivorous
<b>CHARACIFORMS: Characidae</b>			
<i>Acestrocephalus pallidus</i> (Menezes, 2006)	5		carnivorous
<i>Astyanax</i> cf. <i>anterior</i> (Eigenmann, 1908)	12		omnivorous
<i>Astyanax</i> cf. <i>maximus</i> (Steindachner, 1876)	5		omnivorous
<i>Astyanax maximus</i> (Steindachner, 1876)	4		omnivorous
<i>Astyanax</i> sp.	3		omnivorous
<i>Charax</i> sp. "cuniã" (Peixes R. Madeira, 2013)	4		carnivorous
<i>Ctenobrycon spilurus</i> (Valenciennes, 1850)	1		omnivorous
<i>Jupiaba citrina</i> (Zanata & Ohara, 2009)	3		omnivorous
<i>Moenkhausia grandisquamis</i> (Müller & Troschel, 1845)	14		invertivorous
<i>Moenkhausia lata</i> (Eigenmann, 1908)	1	yes	unknown
<i>Moenkhausia</i> sp. "lepidura longa" (Peixes R. Madeira, 2013)	5	yes	omnivorous
<i>Tetragonopterus chalceus</i> (Spix & Agassiz, 1829)	21	yes	omnivorous
<b>CHARACIFORMS: Chalceidae</b>			
<i>Chalceus epakros</i> (Cope, 1870)	308		omnivorous
<b>CHARACIFORMS: Chilodontidae</b>			
<i>Caenotropus</i> cf. <i>schizodon</i> (Scharcansky & Lucena, 2007)	16		omnivorous
<i>Caenotropus labyrinthicus</i> (Kner, 1858)	5		iliophagus
<b>CHARACIFORMS: Ctenoluciidae</b>			
<i>Boulengerella cuvieri</i> (Agassiz, 1829)	217		piscivorous
<i>Boulengerella amaculata</i> (Valenciennes, 1850)	50	yes	piscivorous
<b>CHARACIFORMS: Curimatidae</b>			
<i>Curimata inornate</i> (Vari, 1989)	29		detritivorous
<i>Curimata ocellata</i> (Eigenmann & Eigenmann, 1889)	1		

Continuation Table 3.

Order/Family/Specie	N	OP	Trophicity
<i>Curimata roseni</i> (Vari, 1989)	19		detritivorous
<i>Curimatella alburna</i> (Müller & Troschel, 1844)	29	yes	detritivorous
<i>Potamorhina latior</i> (Spix & Agassiz, 1829)	12		detritivorous
<b>CHARACIFORMS: Cynodontidae</b>			
<i>Cynodon gibbus</i> (Agassiz, in Spix & Agassiz, 1829)	3		piscivorous
<i>Hydrolycus scomberoides</i> (Cuvier, 1816)	137		piscivorous
<i>Hydrolycus tatauaia</i> (Toledo-Piza, Menezes & Santos, 1999)	102	yes	piscivorous
<i>Rhaphiodon vulpinus</i> (Agassiz, in Spix & Agassiz, 1829)	18		piscivorous
<b>CHARACIFORMS: Erythrinidae</b>			
<i>Hoplerethrinus unitaeniatus</i> (Agassiz, in Spix & Agassiz, 1829)	4		carnivorous
<i>Hoplia saimara</i> (Valenciennes, 1847)	1		piscivorous
<i>Hoplias malabaricus</i> (Bloch, 1794)	8	yes	piscivorous
<b>CHARACIFORMS: Hemiodontidae</b>			
<i>Argonectes longiceps</i> (Kner, 1858)	167		omnivorous
<i>Bivibranchia fowleri</i> (Steindachner, 1908)	11		invertivorous
<i>Hemiodus atranalis</i> (Fowler, 1940)	32		herbivorous
<i>Hemiodus gracilis</i> (Günther, 1864)	1	yes	herbivorous
<i>Hemiodus semitaeniatus</i> (Kner, 1858)	18		herbivorous
<i>Hemiodus unimaculatus</i> (Bloch, 1794)	132		herbivorous
<b>CHARACIFORMS: Iguanodectidae</b>			
<i>Bryconops alburnoides</i> (Kner, 1858)	40		omnivorous
<i>Bryconops cf. caudomaculatus</i> (Günther, 1864)	4	yes	omnivorous
<i>Bryconops giacopinii</i> (Fernández-Yépez, 1950)	1		omnivorous
<i>Iguanodectes geisleri</i> (Géry, 1970)	10	yes	insectivorous
<i>Iguanodectes spilurus</i> (Günther, 1864)	30		unknown
<b>CHARACIFORMS: Prochilodontidae</b>			
<i>Prochilodus nigricans</i> (Agassiz, 1829)	218		detritivorous
<i>Semaprochilodus brama</i> (Valenciennes, 1850)	6		detritivorous
<i>Semaprochilodus insignis</i> (Jardine, 1841)	4		detritivorous
<b>CHARACIFORMS: Serrasalminidae</b>			
<i>Catoprion mento</i> (Cuvier, 1819)	3	yes	lepidophagus
<i>Colossoma macropomum</i> (Cuvier, 1818)	4		omnivorous
<i>Myleus micans</i> (Müller & Troschel, 1844)	2		frugivorous
<i>Myleus schomburgkii</i> (Jardine, 1841)	38	yes	frugivorous
<i>Myleus setiger</i> (Müller & Troschel, 1844)	3		frugivorous
<i>Myleus</i> sp.	10		unknown
<i>Myleus torquatus</i> (Müller & Troschel, 1845)	38		frugivorous
<i>Myloplus asterias</i> (Müller & Troschel, 1844)	227	yes	frugivorous
<i>Myloplus cf. rubripinnis</i> (Müller & Troschel, 1844)	121		frugivorous
<i>Myloplus lobatus</i> (Valenciennes, 1850)	9		frugivorous
<i>Myloplus rubripinnis</i> (Müller & Troschel, 1844)	147	yes	frugivorous
<i>Mylossoma duriventre</i> (Cuvier, 1818)	5		omnivorous
<i>Piaractus brachypomus</i> (Cuvier, 1818)	1		frugivorous
<i>Pristobrycon striolatus</i> (Steindachner, 1908)	12	yes	carnivorous

Continuation Table 3.

Order/Family/Specie	N	OP	Trophicity
<i>Pygocentrus nattereri</i> (Kner, 1858)	30	yes	omnivorous
<i>Serrasalmus</i> cf. <i>maculatus</i> (Kner, 1858)	1		carnivorous
<i>Serrasalmus compressus</i> (Jégu, Leão & Santos, 1991)	1		piscivorous
<i>Serrasalmus eigenmanni</i> (Norman, 1929)	8	yes	piscivorous
<i>Serrasalmus elongatus</i> (Kner, 1858)	14	yes	piscivorous
<i>Serrasalmus</i> gr. <i>humeralis</i> (Valenciennes, 1850)	20		piscivorous
<i>Serrasalmus</i> gr. <i>rhombus</i> (Linnaeus, 1766)	14		carnivorous
<i>Serrasalmus humeralis</i> (Valenciennes, 1850)	14	yes	piscivorous
<i>Serrasalmus maculatus</i> (Kner, 1858)	1		carnivorous
<i>Serrasalmus manueli</i> (Fernández-Yépez & Ramírez, 1967)	160		piscivorous
<i>Serrasalmus rhombus</i> (Linnaeus, 1766)	323	yes	carnivorous
<i>Serrasalmus spilopleura</i> (Kner, 1858)	76	yes	piscivorous
<i>Tometes</i> sp.	9		unknown
<i>Utiaritchthys longidorsalis</i> (Tito de Moraes & Santos, 1992)	4		unknown
<i>Utiaritchthys sennaebragai</i> (Miranda Ribeiro, 1937)	3		herbivorous
<b>CHARACIFORMS: Triportheidae</b>			
<i>Agonia teshalecinus</i> (Müller & Troschel, 1845)	143		carnivorous
<i>Triportheus angulatus</i> (Spix & Agassiz, 1829)	4	yes	omnivorous
<i>Triportheus auritus</i> (Valenciennes, in Cuvier & Valenciennes, 1850)	138		omnivorous
<i>Triportheus</i> cf. <i>auritus</i> (Valenciennes, in Cuvier & Valenciennes, 1850)	80		omnivorous
<b>CICHLIFORMS: Cichlidae</b>			
<i>Acarichthys heckelii</i> (Müller & Troschel, 1849)	1	yes	herbivorous
<i>Acaronia nassa</i> (Heckel, 1840)	1	yes	herbivorous
<i>Biotodoma cupido</i> (Heckel, 1840)	3	yes	omnivorous
<i>Caquetaia spectabilis</i> (Steindachner, 1875)	6	yes	unknown
<i>Cichla</i> cf. <i>pinima</i> (Kullander & Ferreira, 2006)	32		piscivorous
<i>Cichla monoculus</i> (Agassiz, 1831)	33		piscivorous
<i>Cichla ocellaris</i> (Bloch & Schneider, 1801)	18		piscivorous
<i>Cichla pinima</i> (Kullander & Ferreira, 2006)	4		piscivorous
<i>Cichla</i> sp.	1		unknown
<i>Crenicichla</i> cf. <i>marmorata</i> (Pellegrin, 1904)	4		carnivorous
<i>Crenicichla johanna</i> (Heckel, 1840)	12	yes	carnivorous
<i>Crenicichla marmorata</i> (Pellegrin, 1904)	5	yes	carnivorous
<i>Crenicichla strigata</i> (Günther, 1862)	1	yes	carnivorous
<i>Geophagus mirabilis</i> (Deprá et al., 2014)	1	yes	omnivorous
<i>Geophagus megasema</i> (Heckel, 1840)	22	yes	omnivorous
<i>Geophagus surinamensis</i> (Bloch, 1791)	4		omnivorous
<i>Mesonauta festivus</i> (Heckel, 1840)	9	yes	omnivorous
<i>Retroculus lapidifer</i> (Castelnau, 1855)	1	yes	insectivorous
<i>Satanoperca jurupari</i> (Heckel, 1840)	14	yes	detritivorous
<i>Satanoperca lilith</i> (Kullander & Ferreira, 1988)	2	yes	detritivorous
<b>CICHLIFORMS: Sciaenidae</b>			
<i>Pachyurus schomburgkii</i> (Günther, 1860)	1		invertivorous
<i>Petilipinnis grunniens</i> (Jardine in Schomburgk, 1843)	9		piscivorous

Continuation Table 3.

Order/Family/Specie	N	OP	Trophicity
<i>Plagioscion squamosissimus</i> (Heckel, 1840)	59		carnivorous
<b>CLUPEIFORMS: Engraulidae</b>			
<i>Lycengraulis batesii</i> (Gunther, 1868)	47		omnivorous
<b>CLUPEIFORMS: Pristigasteridae</b>			
<i>Pellona castelnaeana</i> (Valenciennes, 1847)	32		piscivorous
<i>Pellona flavipinnis</i> (Valenciennes, 1836)	3		piscivorous
<i>Pristigaster cayana</i> (Cuvier, 1829)	6		invertivorous
<b>GYMNOTIFORMS: Gymnotidae</b>			
<i>Electrophorus electricus</i> (Linnaeus, 1766)	1		piscivorous
<b>MYLIOBATIFORMS: Potamotrygonidae</b>			
<i>Potamotrygon motoro</i> (Müller & Henle, 1841)	1		carnivorous
<b>SILURIFORMS: Auchenipteridae</b>			
<i>Ageneiosus inermis</i> (Linnaeus, 1766)	22		carnivorous
<i>Ageneiosus</i> sp.	1		carnivorous
<i>Ageneiosus ucayalensis</i> (Castelnau, 1855)	20		carnivorous
<i>Auchenipterichthys longimanus</i> (Günther, 1864)	128		omnivorous
<i>Auchenipterichthys thoracatus</i> (Kner, 1858)	6		omnivorous
<i>Auchenipterus ambyiacus</i> (Fowler, 1915)	17		insectivorous
<i>Auchenipterus brachyurus</i> (Cope, 1878)	4		carnivorous
<i>Centromochlus heckelii</i> (De Filippi, 1853)	10		carnivorous
<i>Centromochlus schultzi</i> (Rössel, 1962)	2		unknown
<i>Tatia aulopygia</i> (Kner, 1857)	1		unknown
<i>Trachelyopterichthys taeniatus</i> (Kner, 1858)	1	yes	carnivorous
<i>Trachelyopterus galeatus</i> (Linnaeus, 1766)	13	yes	carnivorous
<b>SILURIFORMS: Cetopsidae</b>			
<i>Cetopsis coecutiens</i> (Lichtenstein, 1819)	2	yes	necrophagous
<b>SILURIFORMS: Doradidae</b>			
<i>Leptodoras linnelli</i> (Eigenmann, 1912)	3	yes	invertivorous
<i>Lithodoras dorsalis</i> (Valenciennes, 1840)	9		herbivorous
<i>Nemadora strimaculatus</i> (Boulenger, 1858)	2	yes	insectivorous
<i>Oxydoras niger</i> (Valenciennes, 1821)	1		omnivorous
<i>Platyodoras costatus</i> (Linnaeus, 1758)	2		unknown
<i>Pterodoras granulosus</i> (Valenciennes, 1821)	4		omnivorous
<b>SILURIFORMS: Heptapteridae</b>			
<i>Pimelodella steindachneri</i> (Eigenmann, 1917)	1		unknown
<b>SILURIFORMS: Loricariidae</b>			
<i>Ancistrus</i> sp.	1	yes	unknown
<i>Aphanotrutulus rubrocauda</i> (Oliveira, Py-Daniel & Zawadski, 2017)	15		
<i>Hypoptopoma gulare</i> (Cope, 1878)	3		detritivorous
<i>Hypoptopoma incognitum</i> (Aquino & Schaefer, 2010)	9		detritivorous
<i>Hypostomus</i> cf. <i>plecostomus</i> (Linnaeus, 1758)	7		detritivorous
<i>Hypostomus</i> cf. <i>pyrineusi</i> (Miranda Ribeiro, 1920)	13		detritivorous
<i>Hypostomus emarginatus</i> (Valenciennes, 1840)	2		detritivorous
<i>Hypostomus</i> gr. <i>cochliodon</i> (Kner, 1854)	12		detritivorous

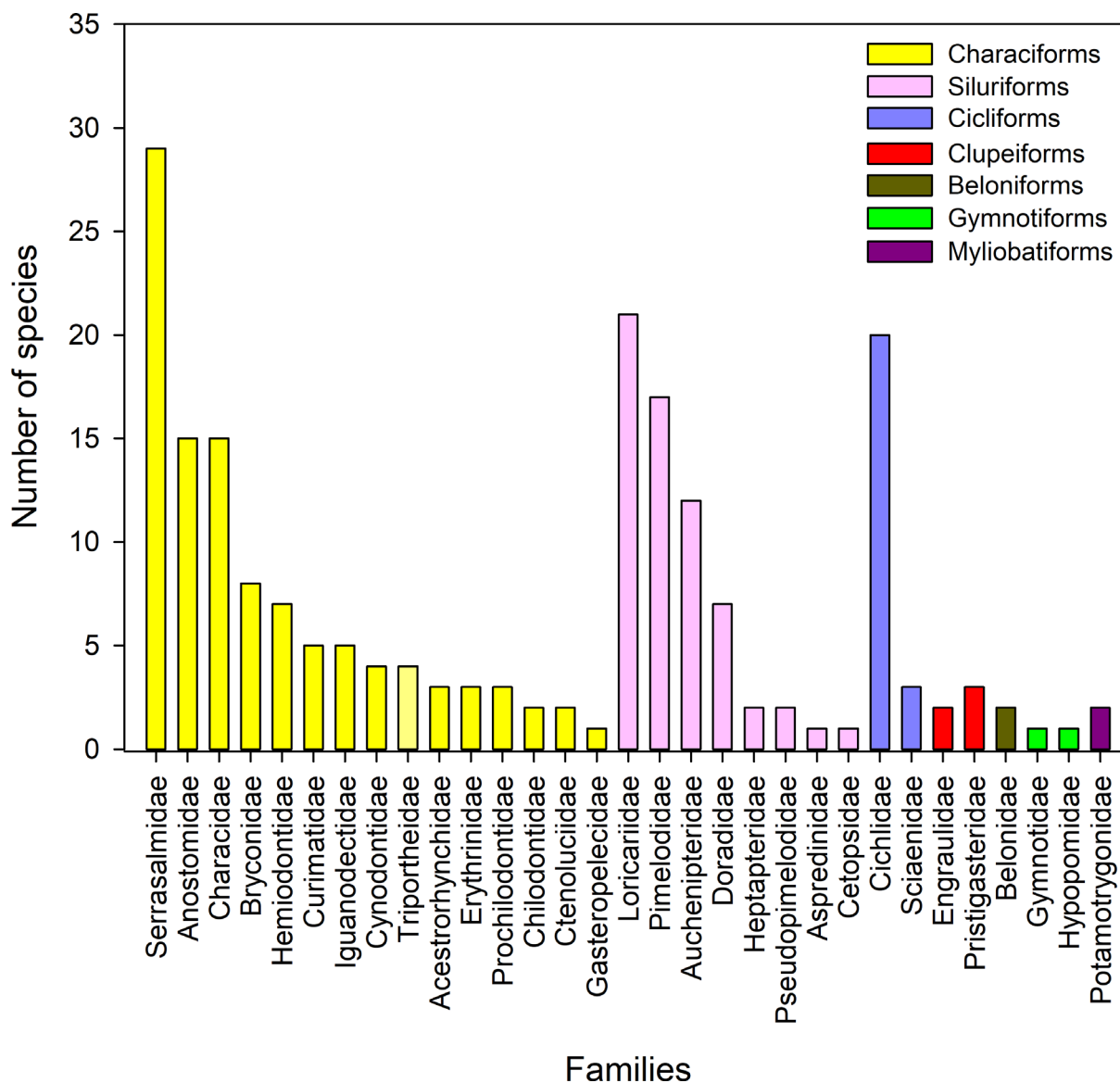
Continuation Table 3.

Order/Family/Specie	N	OP	Trophicity
<i>Hypostomus Plecostomus</i> (Linnaeus, 1758)	1	yes	detritivorous
<i>Hypostomus pyrineusi</i> (Miranda Ribeiro, 1920)	55		detritivorous
<i>Hypostomus</i> sp.	9		detritivorous
<i>Lasiancistrus schomburgkii</i> (Günther, 1864)	6		detritivorous
<i>Lasiancistrus scolymus</i> (Gunther, 1864)	3	yes	detritivorous
<i>Limatulichthys griseus</i> (Eigenmann, 1909)	4		detritivorous
<i>Loricaria cataphracta</i> (Linnaeus, 1758)	1		detritivorous
<i>Loricariichthys nudirostris</i> (Kner, 1853)	2		detritivorous
<i>Panaque armbrusteri</i> (Lujan, Hidalgo & Stewart, 2010)	3		perifitivorous
<i>Pterygoplichthys pardalis</i> (Castelnau, 1855)	1	yes	unknown
<i>Squaliformae marginata</i> (Valenciennes, 1840)	8	yes	unknown
<b>SILURIFORMS: Pimelodidae</b>			
<i>Aguarunichthys torosus</i> (Stewart, 1986)	1		unknown
<i>Brachyplatystoma filamentosum</i> (Lichtenstein, 1819)	1		carnivorous
<i>Calophysus macropterus</i> (Lichtenstein, 1819)	2		carnivorous
<i>Hemisorubim platyrhynchos</i> (Valenciennes, 1840)	7		carnivorous
<i>Hypophthalmus marginatus</i> (Valenciennes, 1840)	5		planctophagus
<i>Hypophthalmus</i> sp.	1		planctophagus
<i>Leiarius marmoratus</i> (Gill, 1870)	1		carnivorous
<i>Phractocephalus hemiliopterus</i> (Bloch & Schneider, 1801)	8		omnivorous
<i>Pimelodus blochii</i> (Valenciennes, 1840)	48	yes	omnivorous
<i>Pimelodus</i> cf. <i>blochii</i> (Valenciennes, 1840)	1		omnivorous
<i>Pimelodus</i> cf. <i>maculatus</i> (Lacepède, 1803)	2		omnivorous
<i>Pimelodus ornatus</i> (Kner, 1857)	10	yes	omnivorous
<i>Pinirampus pirinampu</i> (Spix & Agassiz, 1829)	17		carnivorous
<i>Pseudoplatystoma punctifer</i> (Castelnau, 1855)	4		piscivorous
<i>Pseudoplatystoma tigrinum</i> (Valenciennes, 1840)	3		piscivorous
<i>Sorubim elongatus</i> (Littmann, Burr, Schmidt & Isern, 2001)	12		carnivorous
<i>Sorubim lima</i> (Bloch & Schneider, 1801)	152		carnivorous
<b>SILURIFORMS: Pseudopimelodidae</b>			
<i>Batrochoglanis villosus</i> (Eigenmann, 1912)	1	yes	unknown

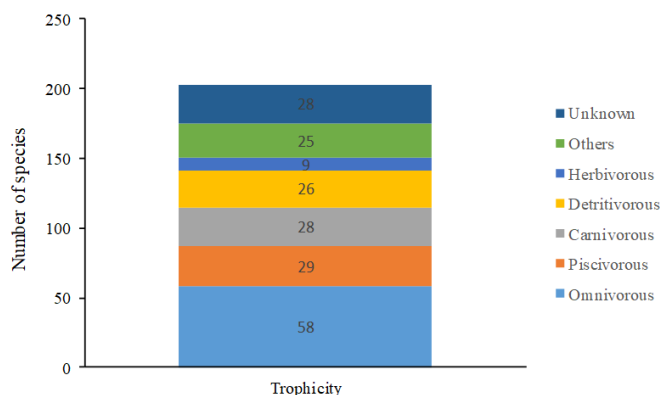
of new species, such as *Hypophthalmus* sp., *Cichla* sp. and *Astyanax* sp. Among the sampled species in Lower Roosevelt River sub-basin are included in the ornamental fish list of IBAMA such as *Acestrorhynchus microlepis* (Schomburgk, 1841), *Leporinus fasciatus* (Bloch, 1794), *Boulengerella maculata* (Valenciennes, 1850), *Hydroly custatauaia* (Toledo-Piza, Menezes & Santos, 1999); *Mylo plusasterias* (Müller & Troschel, 1844), *Mylo plusrubripinnis* (Müller & Troschel, 1844), *Serrasalmus rhombeus* (Linnaeus, 1766); *Serrasalmus spilopleura* (Kner, 1858) e *Pimelodus blochii* (Valenciennes, 1840). Some species considered rare due to their shortage in ichthyological collections were sampled in this study, including the Characiform species such as *Acestrorhynchus heterolepis* (Cope, 1878) and *Acestrocephalus pallidus* (Menezes, 2003), and the Siluriform species such as *Pimelodella steindachneri* (Eigenmann, 1917) and *Panaque armbrusteri* (Lujan, Hidalgo & Stewart, 2010).

The most abundant species were *Serrasalmus rhombeus* (Linnaeus, 1766) (Serrasalmidae) and *Chalceus epakros* (Zanata & Toledo-Piza, 2004) (Characidae). *S. rhombeus* is the largest piranha species, with adults reaching 50 cm in length, and is considered to be one of the most successful fish species in Amazonian reservoirs (Santo & Santos, 2005). It has non-migratory habit, is predominantly carnivorous, and is considered a top-chain species (Goulding, 1988; Lowell-McConnell, 1987); therefore, it reflects the environmental quality of the aquatic ecosystem (Borges et al., 2018). This piranha species is a Neotropical predator that occur in many environments of the Amazon Basin (Sá-Oliveira et al., 2017). On the other hand, *C. epakros* has a much wider distribution throughout the central and lower portions of the Amazon Basin (including the lower course of the Madeira River), middle and upper Orinoco River Basin, the Essequibo River in Guyana and the Nanay River in Peru (Zanata & Toledo-Piza, 2004).





**Figure 2.** Number of species per families per order from the survey of fish species from the Lower Roosevelt River, Southwestern Amazon basin.



**Figure 3.** Trophicity from the survey of fish species from the Lower Roosevelt River, Southwestern Amazon basin.

Our work highlights the importance of conducting fish survey within Roosevelt River Basin. Fish have an important socio-economic role for human communities living along tropical rivers and are a major protein source for these people (Fabr  & Alonso, 1998; Cerdeira et al., 2008; Santos & Santos, 2005; Santos et al., 2014). It is important to monitor native fish diversity in this region, both to preserve biodiversity and to ensure sustainable levels of fish stocks for harvesting.

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## Authors' Contributions

**Marcelo Rodrigues dos Anjos:** Contributions to: elaboration of study's concept and design; data collection, analysis, and interpretation; and preparation of manuscript.

**Nadja Gomes Machado:** Substantial contribution to: study's concept and design; data interpretation; manuscript preparation; and critical revision.

**Mizael Andrade Pedersoli:** Contributions to: data collection, analysis, and interpretation.

**Nátia Regina Braga Pedersoli:** Contributions to: data collection, analysis, and interpretation.

**Bruno Stefany Barros:** Contributions to: data analysis, and interpretation; manuscript preparation; and critical revision.

**Igor Hister Lourenço:** Contributions to: data collection; and manuscript preparation.

**João Pedro Barreiros:** Contributions to: critical revision and addition of intellectual content.

## Conflicts of Interest

The authors declares that there are no conflict of interests related to the publication of this manuscript.

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## Diversity of *Biomphalaria* spp. freshwater snails and associated mollusks in areas with schistosomiasis risk, using molecular and spatial analysis tools

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**Abstract:** The Middle Paranapanema River region of São Paulo, Brazil is home to significant diversity of *Biomphalaria* species and is very vulnerable to health and environmental impacts such as schistosomiasis. This study updates freshwater malacological surveys for ecosystems in one portion of the Middle Paranapanema River Basin, with emphasis on the genus *Biomphalaria*. Snails were collected from 114 distinct bodies of water between 2015 and 2018. *Biomphalaria* specimens were identified according to morphological and molecular characteristics, while animals in other genera (*Drepanotrema*, *Lymnaea*, *Melanoides*, *Physa* and *Pomacea*) were identified solely according to shell characteristics. A geographic information system was used to update intermediate host colonization sites and consequently assist in identifying probable hotspots for intermediate hosts of schistosomiasis. The sequences of the COI gene relating to the DNA barcode stretch were tested for similarity against sequences found in GenBank, for monophyly through Maximum Likelihood phylogenetic inference, and analyzed in ABDG, bPTP and GMYC for the delimitation of putative species. Of the 10,722 snails collected, 86.7% were in the Planorbidae family (75.5% *Biomphalaria* and 11.2% *Drepanotrema*) and 13.3% were other non-Planorbidae species (*Lymnaea*, *Melanoides*, *Physa* and *Pomacea*). The taxonomic COI reference sequences in the NCBI nucleotide database used for DNA sequence comparison, and phylogenetic analysis used to test the monophyly of the groups, resulted in more reliable taxonomic units than delimitation of the COI sequences in MOTUs using statistical taxonomic models. Analysis of the species distribution shows that *B. glabrata* and *B. tenagophila* are heterogeneously distributed in the study area. *B. glabrata* colonizes only five water bodies, in the study area, most of them in Ourinhos, while *B. tenagophila* predominates in water bodies in Ipaussu. Contrasting with this, *B. straminea*, *B. occidentalis* and *B. peregrina* are evenly distributed throughout the study area.

**Keywords:** *Biomphalaria*, freshwater snails, biodiversity, DNA barcode, distribution.

## Diversidade de caramujos de água doce *Biomphalaria* spp. e outros moluscos associados, em áreas de risco da esquistossomose, empregando ferramentas moleculares e espaciais

**Resumo:** A região do Médio Rio Paranapanema, em São Paulo, Brasil abriga uma diversidade significativa das espécies de *Biomphalaria*. É também uma região vulnerável a impactos ambientais e de saúde, como a esquistossomose. Este estudo atualiza dados sobre a distribuição de caramujos de água doce em ecossistemas de uma porção da Bacia do Médio Rio Paranapanema, com ênfase no gênero *Biomphalaria*. Os caramujos foram coletados de 114 corpos distintos de água doce, entre 2015 e 2018. Exemplares pertencentes ao gênero *Biomphalaria* foram identificados de acordo com características morfológicas e moleculares, enquanto animais de outros gêneros (*Drepanotrema*, *Lymnaea*, *Melanoides*, *Physa* e *Pomacea*) foram identificados somente de acordo com características da concha. Ferramentas de análise geoespaciais foram utilizadas para atualizar os sítios de colonização dos caramujos e, consequentemente, auxiliar na identificação de possíveis pontos críticos para



hospedeiros intermediários da esquistossomose. As sequências do gene COI relacionadas ao DNA barcode foram testadas quanto à similaridade com sequências encontradas no GenBank, por análise filogenética sob máxima verossimilhança, e analisadas em ABDG, bPTP e GMYC para a delimitação de espécies putativas. Dos 10.722 moluscos coletados, 86,7% pertenciam a família Planorbidae (75,5% *Biomphalaria* e 11,2% *Drepanotrema*) e 13,3% a *Lymnaea* spp., *Melanoides* spp., *Physa* spp. e *Pomacea* spp. A comparação das sequências taxonômicas de COI com o banco de dados de nucleotídeos do NCBI, e a análise filogenética usada para testar a monofilia dos grupos, resultaram em delimitações taxonômicas comparáveis à delimitação morfológica. As espécies *B. glabrata* e *B. tenagophila* estão heterogeneamente distribuídas ao longo da área de estudo. *B. glabrata* foi identificada em apenas cinco coleções de água doce, quatro delas em Ourinhos, enquanto *B. tenagophila* predominou em Ipaussu. Por outro lado, *B. straminea*, *B. occidentalis* e *B. peregrina* estão distribuídas uniformemente na área de estudo.

**Palavras-chave:** *Biomphalaria*, caramujos de água doce, biodiversidade, código de barras DNA, distribuição.

## Introduction

The midsection of the Paranapanema River in the state of São Paulo, Brazil is located in areas with mostly flat topography and features shallow lentic ecosystems such as streams, marshes, and wetlands colonized by macrophytes. These ecosystems are home to diverse freshwater mollusk fauna including the genus *Biomphalaria*; species described in this region are *Biomphalaria glabrata* (Say 1818), *B. tenagophila* (d'Orbigny 1835), *B. straminea* (Dunker 1848), *B. occidentalis* (Paraense 1981), and *B. peregrina* (d'Orbigny 1835), as well as *Pomacea* (Perry 1810), *Drepanotrema* (Fischer & Crosse 1880), *Lymnaea* (Lamarck 1799), *Melanoides* (Olivier 1804), and *Physa* (Draparnaud 1801). (Piza et al. 1972, Vaz et al. 1989, Luz et al. 1998, Teles 1989, Fernandez et al. 2003, Carvalho et al. 2008, Tuan 2009, Maltchik et al. 2010, Scholte et al. 2012, Medeiros et al. 2014, Pereira et al. 2018).

This species richness combined with the volume and extension of water in the Paranapanema River Basin presents a significant opportunity to explore snail diversity and their relationship with potential transmission of waterborne diseases. Specifically, with regard to the genus *Biomphalaria*, the Middle Paranapanema has been the source of human cases of schistosomiasis; *B. glabrata*, *B. tenagophila*, and *B. straminea* are found in bodies of fresh water in this region, and larvae of the trematode *Schistosoma mansoni* Sambon, 1907 develop in these three species causing schistosomiasis in humans.

Taxonomic identification of species in the genus *Biomphalaria* uses binary anatomical characters with a high coefficient for retention and consistency within a single species, which are associated with the characters that exhibit major variations in number and shape (Paraense 1961, 1975, 1981). Traditional taxonomy requires meticulous observation of snails from various bodies of fresh water and an extensive geographical area.

Intraspecific variations in morphological characters that are used to identify *Biomphalaria* species are usually minimized, and consequently identification is exclusively based on the snails' anatomy, although this method cannot decisively address the natural diversity of the species in question.

Results obtained for species of freshwater and land mollusks which are medically relevant in the neotropics show the importance of combining traditional methods of morphological analysis with molecular methods to identify known species as well as potential cryptic

species (Langand et al. 1999, Vidigal et al. 2000, 2013, Campbell et al. 2000, Dejong et al. 2003, Wethington et al. 2007, Teodoro et al. 2010, Tuan et al. 2012, Palasio et al. 2017, Bezerra et al. 2018).

Molecular analysis shows significant interspecific genetic differentiation in some species of *Biomphalaria* such as *B. tenagophila*, *B. glabrata*, *B. peregrina*, and *B. straminea*, suggesting the presence of differentiated lineages in contiguous geographic areas (Tuan & Santos 2007, Palasio et al. 2018). These results contrast with the phenotypic uniformity found in morphological assessment, and demonstrate that the molecular characters used to differentiate *Biomphalaria* species could broaden knowledge on the diversity of freshwater snails.

The close relationship between snails of the genus *Biomphalaria* and human health makes diversity studies of this group essential, since taxonomy and even the mapping of aquatic ecosystems are necessary to monitor and control schistosomiasis. Within the context of public health, the inclusion of new approaches can affect the adoption of new strategies in the future (Salked et al. 2015).

Spatial analysis techniques are being used systematically to analyze distribution patterns of snail species in the genera *Bulinus* and *Biomphalaria* which colonize specific regions of Africa (Simoonga et al. 2009; Moser et al., 2014). In Brazil, geographic information systems (GIS) have been utilized in studies correlating the distribution patterns of snails which are intermediate hosts of *S. mansoni* in areas where schistosomiasis is highly endemic, including Bahia (Bavia et al. 1999, Cardim et al. 2011), Pernambuco (Almeida et al. 2003; Gomes et al. 2012), Minas Gerais (Guimarães et al. 2009, 2010, 2012, Fonseca et al. 2014), and Sergipe (Barboza et al. 2012, Santos et al. 2016).

The distribution of *Biomphalaria* species in the state of São Paulo was mapped using traditional methods which utilize mollusk checklists to record species distribution in macro-geographic administrative divisions (Piza et al. 1972, Vaz et al. 1983, 1986, 1987, 1989, 1992). There is no record of a mollusk checklist for the middle region of the Paranapanema River. It is important to update snail distribution of this region and gather this data into a mollusk checklist based on geospatial analysis methodology.

With this in mind, our analysis was developed to investigate the diversity of mollusk fauna, particularly the distribution patterns of *Biomphalaria* species, using geospatial analysis and taxonomy tools that combine morphological and molecular approaches.



## Material and Methods

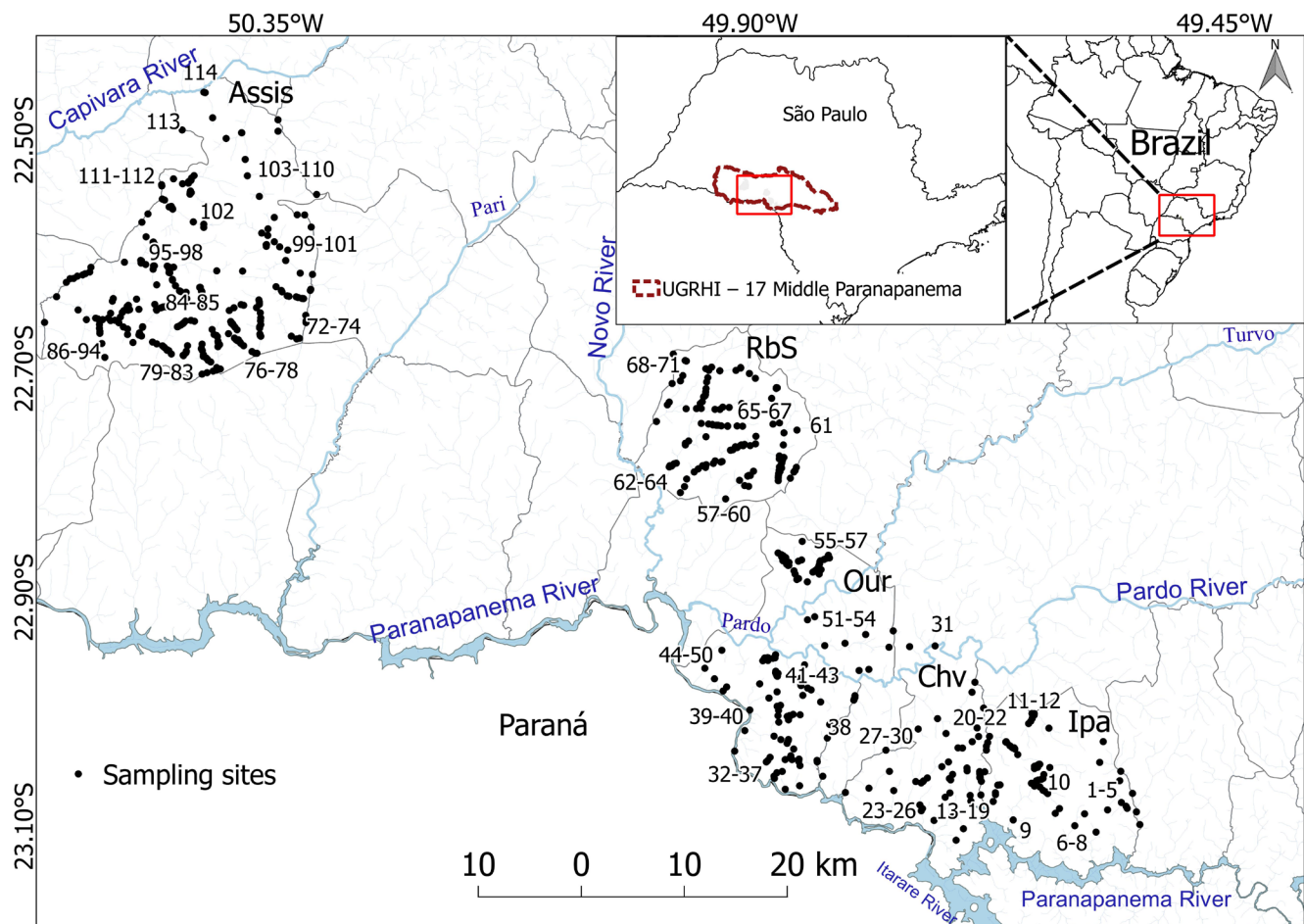
### 1. Study area and snail collection

The study area is located in the Middle Paranapanema River Basin, which is located in the west/southwest region of the state of São Paulo, on the border with Paraná, Brazil (Figure 1). This region has anthropogenic landscapes located between large expanses of sugarcane, soy, and coffee plantations as well as pasture areas (Peron & Piroli 2011, CBH-MP, 2018).

The Paranapanema, Pardo, Turvo, Novo, Capivara, and Pari Rivers traverse the region and comprise the Middle Paranapanema River Water Resources Management Unit (UGRHI-17) (Safre & Manzione 2015, CBH-MP, 2018).

Mollusks were collected from 114 freshwater ecosystems located in rural and urban areas across the municipalities of Ipaussu, Chavantes, Ourinhos, Ribeirão do Sul, and Assis, from ecosystems where medically relevant snails had previously been reported (Figure 1). The location and boundaries of the larger and smaller bodies of water were identified utilizing physical maps obtained from each of the five municipalities.

The snails were collected from a minimum of 1 and a maximum of 44 collection points spaced from 250 m to 1000 m in each of the 114 bodies of fresh water (additional information file), totaling 654 sampling points. Variations in the collection points along the same body of fresh water resulted from variations in total length (Km) in the bodies of water which were the object of study. The results ranged from 1 to 159 snails per sample, depending on the productivity of the breeding site and the extension of the collection points. The snails were collected using a standardized capture scoop made of metal mesh (BRASIL 2008) during March, June, September, and December of 2015–2018. The samples were placed into containers with water from their own ecosystems to keep the specimens alive. Once they arrived in the laboratory in Ourinhos, the snail specimens obtained from the collection points were grouped and packed in dry gauze and sent to the Biochemistry and Molecular Biology Laboratory at the São Paulo State Center for Endemic Diseases Control (SUCEN). All the collection points were georeferenced using a Garmin eTrex GPS device (Garmin, Olathe, USA), with an exact margin of error of  $\pm 3.6$  meters (12 feet).



**Figure 1.** Points where snails and associated mollusk fauna were collected from 114 bodies of water between 2015 and 2018 in the municipalities of Ourinhos, Ipaussu, Chavantes, Ribeirão do Sul, and Assis in the Middle Paranapanema region of São Paulo, Brazil. \* numbers correspond to the bodies of water (see additional information file).

The diversity of species and genus of snails sampled across areas of each municipality was expressed as the frequency of each species or genus (*i*) in relation to the number of water bodies inhabited by species or genus sampled.

## 2. Morphological and molecular identification of the snails

The characteristics of the shells of the snails collected were used for identification to the genus level. All snails in the genus *Biomphalaria* were exposed to artificial light for 4 hours to stimulate the release of *S. mansoni* cercariae (BRASIL 2008).

From each of the samples containing *Biomphalaria* snails, which are distinguishable by examining their shells, a subsample of up to five adult and/or juvenile specimens were separated for morphological and molecular identification.

The process of morphological identification in the adult snails began with separating the soft parts from the shells after the columellar muscle of the animal was relaxed in water at 70 °C. Next, a 1-mm or approximately 10-mm section of the foot was excised from the cephalopodal region in the juvenile and adult specimens, respectively, for subsequent molecular analysis. The other soft parts from the adult specimens were maintained in Raillet-Henry solution for at least three days and then dissected and identified according to the morphological characteristics of their reproductive systems, as described in Deslandes (1959) and Paraense (1975, 1981). The parts used for morphological identification were kept as “vouchers” in tubes with Raillet-Henry fixation at SUCEN’s Molecular Biology Laboratory (LBMSU) (LBMSU-670-674, 675-722, 727-829, 864-899, 935-957, 973-977, 1007-1026, 1042-1063, 1094-1099 and 1118-1252). Non-*Biomphalaria* vouchers were filed with the Brazilian National Reference Laboratory for Schistosomiasis [LRNEM] at the Oswaldo Cruz Institute under number LRNEM (NA77-87/17, NA 89-108/17, NA 193/17, NA 199-201/17, NA194-195/18, NA196/19, NA198/17 and NA197/20).

The cephalopodal section taken from the adults and juveniles then underwent procedures to extract and purify the genomic DNA using a Qiagen Blood and Tissue Kit (Qiagen, Hilden, Germany). After extraction and purification, the genetic material was frozen at a temperature of -18°C at the SUCEN laboratory.

The purified genomic DNA then underwent amplification of the mitochondrial cytochrome oxidase I gene using LCO-1490 and HCO-2198 primers (Folmer 1994), according to DNA Barcode protocol (Hebert et al. 2003) and PCR conditions described by Tuan et al. 2012.

The amplified products were then sequenced in an ABI3100 sequencer (Applied Biosystems, Foster City, CA, USA) at the Molecular Biology Laboratory of the Butantan Institute.

The electrochromatograms were visualized and analyzed in Chromas software (Technelysium Pty Ltd., South Brisbane, Australia) and aligned in MAFFT version 7 software (Kato et al. 2017, <<https://mafft.cbrc.jp/alignment/server/>>), using the highly rigorous Q-INS-I parameter. The sequences were visually corrected using the BioEdit 7.2.5 tool (Hall 1999) and individually analyzed in the NCBI GenBank database (<<https://www.ncbi.nlm.nih.gov/genbank/>>) to obtain the similarity values between the target sequence and sequences which had been previously filed in GenBank.

COI sequences which had previously been identified according to morphology, were used to classify the species into taxonomic units, along with sequences from immature specimens and those with ambiguous

morphology. This process utilized the following statistical tests: (i) ABGD (Puillandre et al. 2012), which uses distance as a parameter to infer the barcoding gap; ii) bPTP (Zhang et al. 2013), which calculates the highest probability of branching through Bayesian analysis and (iii) GMYC (Fujisawa & Barraclough 2013), with an algorithm using models of speciation and coalescence to infer putative species.

The results based on genetic distance between the sequences were obtained using the online version of ABGD software (<<http://www.wabi.snv.jussieu.fr/public/abgd/>>). The results for bPTP were obtained using the online version of the software (<<https://species.h-its.org/>>), with a tree containing sequences grouped by the NJ method as the input file, and the GTR+G model generated in MEGA X. The models were selected using the Akaike information criterion (AIC) via Modelgenerator software (Keane et al. 2006).

The results for the GMYC model were obtained from the online version (<<https://species.h-its.org/gmyc/>>) using an ultrametric input tree calculated from the grouping of the COI sequences according to the Neighbor-Joining method (NJ), with the GTR+G model in MEGA X (Kumar et al. 2018). To test the hypothesis of monophyly, the COI-sequences were tested under the Maximum-Likelihood (ML) principle in PHYML 3.2 software (Guindon & Gascuel 2003), which uses the NNI heuristic method to obtain the initial tree and exhaustive search (SSR) for obtaining the tree with the highest likelihood value. The values of support for the branches were obtained by parametric method approximate Likelihood Ratio Test (aLRT) associated to the non-parametric method Shimodaira-Hasegawa (SH) (Shimodaira & Hasegawa 1999, Anisimoiva et al. 2011). The analyzes were done under General Time Reversible (GTR) model. The trees were visualized in Treeview (Page 2001).

Interspecific and intraspecific genetic divergence values were calculated using MEGA X (Kumar et al. 2018).

## 3. GIS Mapping

Together with the coordinates from the collection sites, the data on identification of the snails at the species and genus levels were used to create thematic maps showing the distribution of *Biomphalaria* species and the other mollusk groups. This was done in QGIS software version 2.18.22 (QGIS Development Team, 2018). The cartographic materials showing river layers and census tracts were obtained from the Brazilian Institute of Geography and Statistics (IBGE 2010a, 2010b, 2010c) and the São Paulo State Secretary of the Environment (SMA 2013).

## Results

### 1. Taxonomic composition

Between 2015 and 2018, 10,722 snails were collected with conchological characteristics of animals in the genera *Biomphalaria*, *Drepanotrema*, *Physa* spp., *Melanoides* spp., *Lymnaea* spp., and *Pomacea* spp. These snails were found at 258 collection points and totaled 473 positive samples. During this same period, negative results were obtained at 396 collection points, totaling 421 samples.

Of the 473 positive samples, in 298 only snails in the genus *Biomphalaria* were collected (N=7092). In 108 samples (N=1960) snails from the following genera were collected: *Drepanotrema* spp., *Physa* spp., *Melanoides* spp., *Lymnaea* spp., and *Pomacea* spp.

In the remainder of the 67 positive samples, 1006 snails in the genus *Biomphalaria* were collected; these were randomly associated with the snails belonging to other genera (N=664) described above. Snails in the genus *Lymnaea* were found at only 13 collection points; 3 of these specimens were *Lymnaea columella* (Say, 1817) and trematodes were not present. No *Biomphalaria* snails shed *S. mansoni* cercarian flukes.

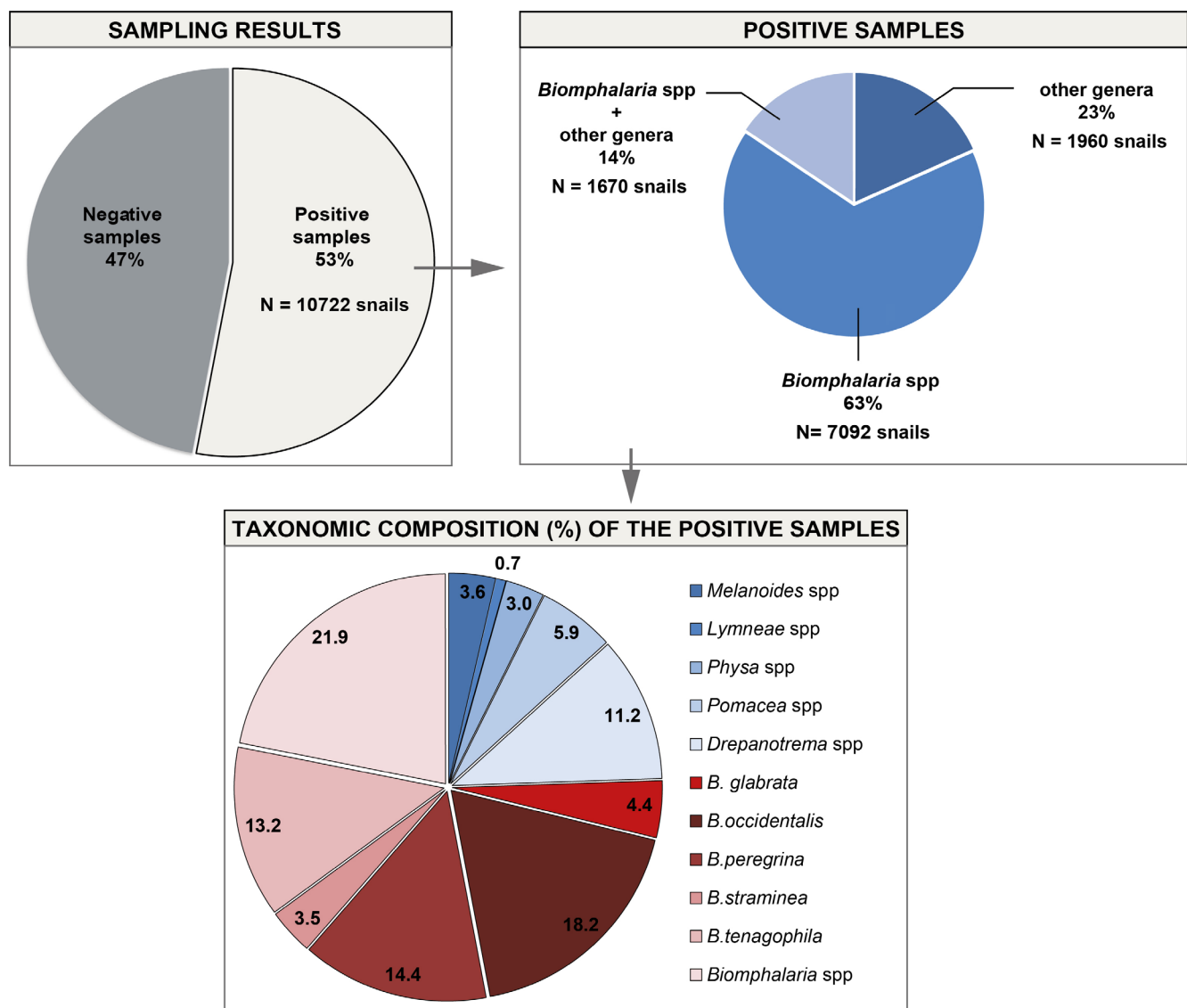
The complete taxonomic composition according to conchological analysis is depicted in Figure 2.

Of the total of 365 positive samples containing individuals of *Biomphalaria*, specific identification methods could be applied to individuals from 276 samples. In the 89 remaining samples, specific identification was not viable because the specimens had died during transport from the field to the laboratory.

Of the total of 276 samples of snails in the genus *Biomphalaria*, taxonomic identification by morphological and molecular characters was applied to subsamples of 144 snails. Because of technical problems

extracting and purifying the genomic DNA, subsamples of 63 snails were identified by morphological characters alone. Immature snails (N=4 samples) with ambiguous morphology and wrinkling in the reproductive system due to problems obtaining the soft tissue (N=65 samples) offered a further opportunity to apply the DNA barcode, because it was impossible to dissect or morphologically identify these specimens.

The samples which were identified using molecular biology or molecular as well as morphological analysis (4+65+144=213 samples) produced 275 COI gene sequences which were compared with the sequences deposited in GenBank for similarity (Table 1). All the COI sequences obtained in the present study corresponded to the nominal species present in GenBank filed under access numbers KX354433-KX354444 and MK395801-MK396064. Data related to the geographic coordinates, reference numbers for the COI sequences in GenBank and each voucher specimen are presented in the additional information file.



**Figure 2.** Sampling results and observed frequency of species of *Biomphalaria*, and snails in the genera *Drepanotrema*, *Physa*, *Pomacea*, *Melanoides*, and *Lymnaea*, observed in the period 2015-2018 in the area under study.

**Table 1.** Similarity indexes between the 275 COI gene sequences for *Biomphalaria*, the sequences in GenBank, and the number of molecular taxonomic units obtained using different statistical methods (ABGD, bPTP, GMYC).

	Morphological species	Number of seqs	Bodies of water	Genbank			Number of MOTUs		
				Number seqs	E Best fit	Best fit MOTU	ABGD	bPTP*	GYMC**
Adults	<i>B. glabrata</i>	17	4	29	100	<i>B. glabrata</i>	1	1	1
	<i>B. occidentalis</i>	58	23	21	100	<i>B. occidentalis</i>	1	1	1
	<i>B. peregrina</i>	45	18	14	95-99	<i>B. peregrina</i>	2	3	5
	<i>B. straminea</i>	11	7	96	99	<i>B. straminea</i>	1	2	2
	<i>B. tenagophila</i>	46	9	107	100	<i>B. tenagophila</i>	1	1	1
	Total	177	45				6#	8	10
Ambiguous morphology (Ma)		8	4	29	100	<i>B. glabrata</i>	1	1	1
		52	19	21	100	<i>B. occidentalis</i>	1	1	1
	<i>Biomphalaria</i> spp.	13	10	15	95-100	<i>B. peregrina</i>	2	3	4
		7	6	96	99	<i>B. straminea</i>	1	2	2
		11	8	107	100	<i>B. tenagophila</i>	1	1	1
Immature (Im)		2	1	29	100	<i>B. glabrata</i>	1	1	1
	<i>Biomphalaria</i> spp.	4	2	21	100	<i>B. occidentalis</i>	1	1	1
		1	1	107	100	<i>B. tenagophila</i>	1	1	1
	Total	98	32				6#	8	9
Adults + Im+ Ma	<i>B. glabrata</i> +Ma + IM	27	5	29	100	<i>B. glabrata</i>	1	1	1
	<i>B.occidentalis</i> +Ma+IM	114	28	21	100	<i>B. occidentalis</i>	1	1	1
	<i>B. peregrina</i> +Ma	58	26	14	95-99	<i>B. peregrina</i>	2	3	6
	<i>B. straminea</i> +Ma	18	9	96	99	<i>B. straminea</i>	1	2	2
	<i>B.tenagophila</i> +Ma+IM	58	12	107	100	<i>B. tenagophila</i>	1	1	1
	Total	275	65				6#	8	11

## 2. Integrated morphological and molecular taxonomy

In order to separate the species into taxonomic units (DNA barcode), 275 sequences were evaluated; 177 of those sequences were obtained from the COI gene in adult individuals which had previously been morphologically identified, while the remaining 98 came from immature specimens or those with ambiguous morphology.

The similarity between the COI gene sequences and the sequences filed in GenBank was 99–100% for *B. tenagophila*, *B. glabrata*, *B. occidentalis*, and *B. straminea*. Minimum similarity values of 95% were obtained for comparison of some sequences from *B. peregrina*. This similarity value was also obtained for sequences derived from immature specimens, adults or those with ambiguous morphology (Table 1).

When analyzed in ABGD, bPTP, and GMYC, the COI sequences from *B. glabrata*, *B. tenagophila*, and *B. occidentalis* with the lowest values for intra-species genetic diversity produced three different groups of molecular operational taxonomic units (MOTUs). Nevertheless, the three statistical methods differ with regard to the number of MOTUs in species with higher rates of intraspecific genetic diversity. In ABGD, *B. peregrina* was identified in two groups of putative species. In bPTP, *B. peregrina* and *B. straminea* were identified in three and two groups, respectively, while in GMYC there was statistical support for the identification of *B. peregrina* in 5 putative species. (Table 1).

The lowest values (5%) for interspecific genetic divergence were observed when comparing sequences from *B. tenagophila* vs. *B. occidentalis*, and the highest values (12%) were seen in comparisons of sequences from *B. peregrina* vs. *B. glabrata* and *B. peregrina* vs. *B. tenagophila* (Table 2).

Analysis of the distribution of K2P distance values calculated for the pairs of COI sequences displayed in Figure 3 shows an area of intersection between the intra- and interspecies values. This intersection zone is comprised of 10% of the sequences from the *B. peregrina* taxon.

Phylogenetic analysis shows the results of grouping the sequences into five monophyletic branches with high statistical support (Figure 4), and was the method with closest approximation with the taxonomic results obtained from analysis of the morphological variables.

## 3. Mapping and distribution of the taxonomic groups identified

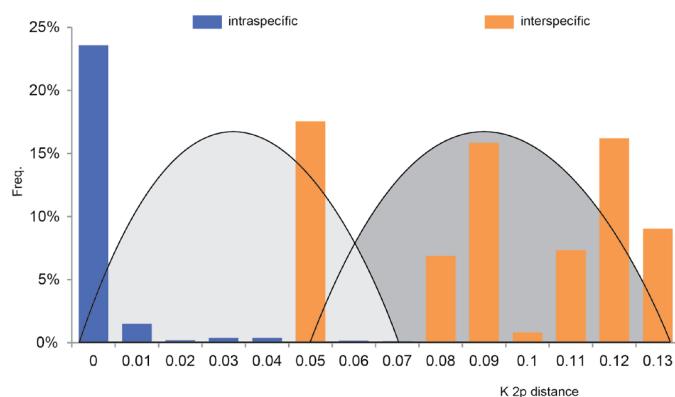
Snails in *Biomphalaria* species colonized 76 of the 114 bodies of fresh water sampled, with *B. glabrata* and *B. tenagophila* being most frequent in the region between the Pardo and Paranapanema Rivers.

*B. glabrata* was concentrated in five of the 114 bodies of water, while *B. straminea*, *B. occidentalis*, and *B. peregrina* were evenly distributed across the study area (Figure 5 and Table 3).



**Table 2.** Inter (light grey) and intraspecies genetic distances (dark gray) for 275 COI gene sequences from *Biomphalaria* species collected in the Middle Paranapanema River Basin (São Paulo, Brazil), using the Kimura 2-parameter model (K2P, Kimura, 1980). The values were obtained in MEGA X.

Species	<i>B. occidentalis</i>	<i>B. tenagophila</i>	<i>B. glabrata</i>	<i>B. straminea</i>	<i>B. peregrina</i>
<i>B. occidentalis</i>	0.00-0.00				
<i>B. tenagophila</i>	0.05	0.00-0.00			
<i>B. glabrata</i>	0.08	0.08	0.00-0.01		
<i>B. straminea</i>	0.08	0.08	0.08	0.00-0.01	
<i>B. peregrina</i>	0.11	0.12	0.12	0.10	0.00-0.08

**Figure 3.** Distribution of 275 sequences of the COI gene from *Biomphalaria* specimens collected from the Middle Paranapanema River (São Paulo, Brazil) from 2015 to 2018 in pair comparisons according to the K2P distance calculated in MEGA X.

*Drepanotrema* spp., *Melanoides* spp., *Lymnaea* spp., *Physa* spp., and *Pomacea* spp. were found in 68 of the 114 bodies of water sampled (Figure 6, Table 3). In 39 of these bodies of water, species in these genera were found in sympatry with *Biomphalaria* spp. Nevertheless, only ten of these 39 bodies of water contained *Biomphalaria* species which can act as intermediate hosts for *Schistosoma mansoni* (Table 3).

Considering the results of identification by morphology, morphology+DNA barcode, and DNA barcode, in addition to the results obtained by shell analysis of the other taxa found in the study region, we can see from our results (Table 3) that freshwater bodies in Ourinhos contain 80% of all *B. glabrata* snails and 40% of *B. occidentalis* sampled in the whole study area. *B. tenagophila* snails mostly inhabits freshwater bodies at Ipaussu (39%), while *B. straminea* and *B. peregrina* are spread over the study area. It is worth to note the high frequency of *Lymnaea* spp. in freshwater bodies at Assis.

## Discussion

The taxonomic diversity of species in the genus *Biomphalaria*, as assessed using morphological methods and DNA barcoding, is consistent with the malacological survey conducted in freshwater bodies of Ourinhos, Ipaussu (Tuan 2009) and Assis (Piza et al. 1972, Teles & Vaz 1987), along the Middle Paranapanema River Basin.

In general, all the COI sequences obtained are highly similar to the nominal species sequences in GenBank. The lowest similarity value with the GenBank sequences was observed in *B. peregrina*; this species included a group of 9 sequences with high genetic distance, which as of this writing is the greatest intraspecies distance obtained for

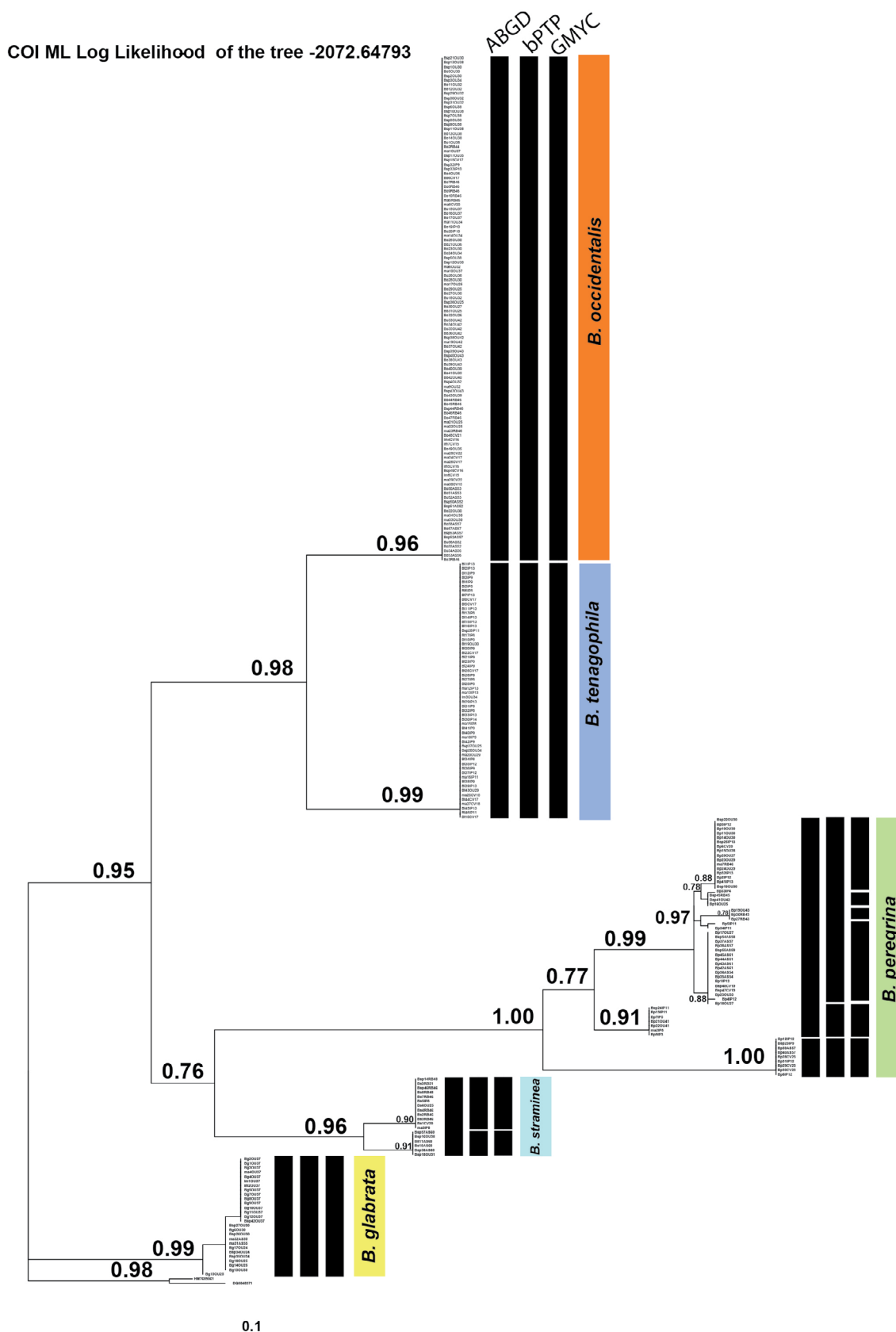
*Biomphalaria* species (Spatz et al. 2000, Vidigal et al. 2000, Collado et al. 2011, Collado & Mendez 2012, Tuan & Santos 2007, Standley et al. 2011, Rumi et al. 2017). Although the genetic variation observed within morphologically recognized as *B. peregrina* specimens, all the sequences comprised a highly supported monophyletic group, suggesting that this taxon probably contains cryptic lineages. Detailed morphological analysis of specimens from a wide geographic area involving nuclear molecular markers is necessary to test this hypothesis.

As we found in this study, a lack of genetic variation in sequence groups in *B. occidentalis* is commonly diagnosed in this species (Tuan et al. 2012). What was uncommon was the lack of genetic diversity in *B. tenagophila*, since previous studies have shown that high levels of genetic diversity of this species were found in subpopulations within other regions (Palasio et al. 2018). The genetic homogeneity observed in the study region in *B. tenagophila* may be a result of incomplete sampling of all of the genetic variants of this species. Furthermore, *Biomphalaria* species are subject to cycles of local extinction followed by recolonization, experiencing dramatic declines in genetic variability (Buckling et al. 2000) with only a fraction of total species variability remaining at the end of several cycles. This type of narrowing event may have extinguished a significant portion of total genetic diversity for this species, which could explain underestimation of *B. tenagophila* diversity in this region.

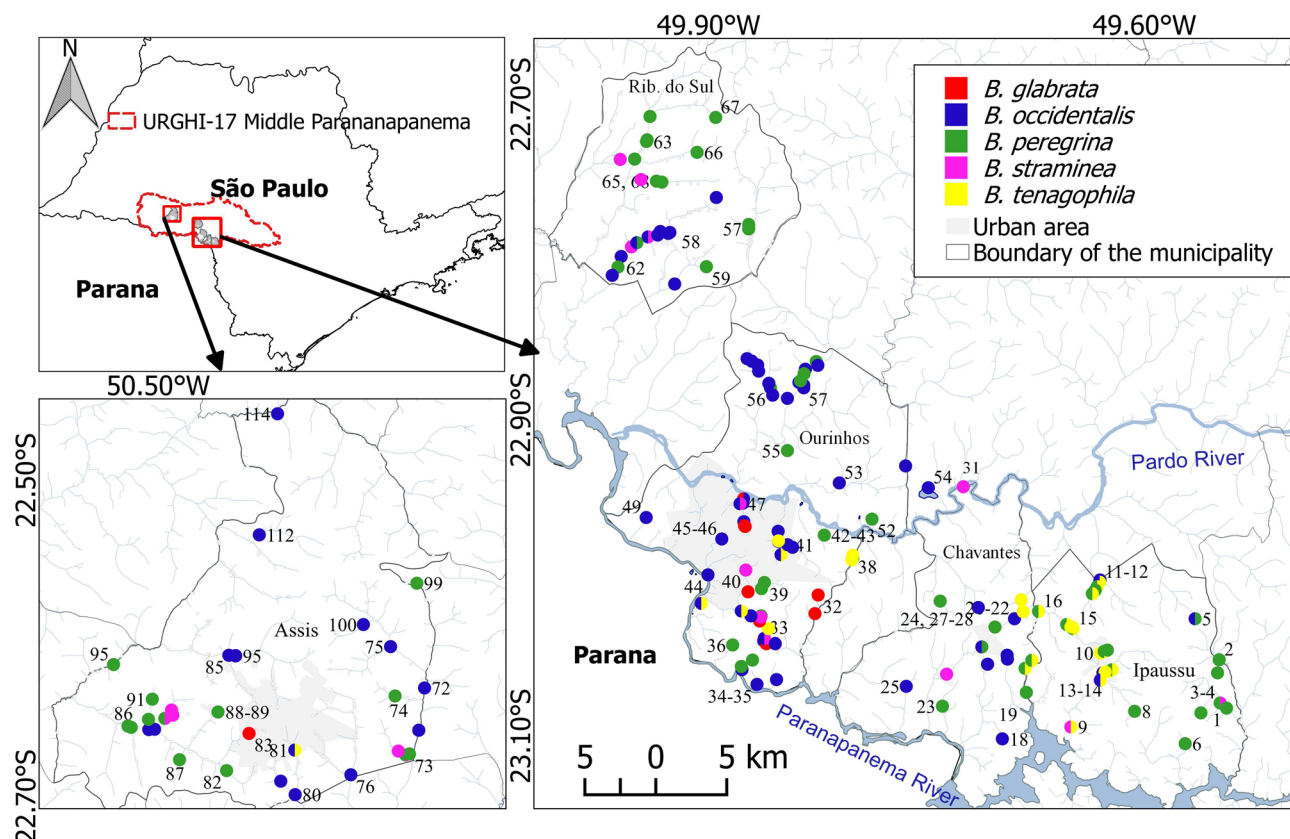
The methods based on distance (ABGD) and trees (bPTP and GMYC), ABGD approached demarcation according to morphology-based taxonomy which discriminate *B. glabrata*, *B. tenagophila* and *B. occidentalis*. The lack of a distinct barcode gap in our dataset, as showed in the frequency histogram for intra and interspecific distributions, suggests, at first, that distance-based methods are unsuitable for *Biomphalaria* species delimitation. However, intra and interspecific values overlapped only for a set of sequences of *B. peregrina* with the higher values for intraspecific genetic diversity. This result might also explain the subdivision of this species into three molecular taxonomic units in bPTP and five in GMYC. Overall, the three statistical methodologies applied (ABGD, bPTP, and GMYC) for delimitation of the 177 COI sequences, produced numbers of molecular taxonomic units that most probably reflected variations in intraspecies genetic diversity. In fact, the differences seen in the number of MOTUs between the bPTP and GMYC methods are commonly observed; the most logical explanation is that these methods reflect differences in each species with regard to effective population size and the rates of mutation and speciation processes (Dellicour & Flot 2018).

*B. tenagophila* colonized 14 of the 92 bodies of water positive for *Biomphalaria*, seven of which are located in Ipaussu. This information contrasts sharply with previous studies (Vaz 1989, Teles 1989, SUCEN





**Figure 4.** Phylogenetic tree with 275 COI sequences, analyzed by maximum likelihood in PhyML using the GTR model. Two sequences were used as an outgroup. The branch support values were calculated using Shimodaira-Hasegawa [SH]-aLRT. The black bars indicate the species boundaries based on ABGD, bPTP, and GMYC, respectively. The colored bars correspond to the boundaries based on morphology.



**Figure 5.** Distribution of *Biomphalaria* species identified by collection point during the period 2015–2018 in Ourinhos, Ribeirão do Sul, Ipaussu, Chavantes, and Assis. The numbers correspond to the bodies of water where the snails were collected.

**Table 3.** The species and genera distribution according to their frequency (%) in each area compared to all positive water bodies sampled (N=92) along five areas between 2015–2018, in the Middle Paranapanema, São Paulo, Brazil. Frequency (%) = (number of water bodies with taxon *i* sampled in each area/total number of water bodies in the five areas sampled) × 100. \*Source IBGE.

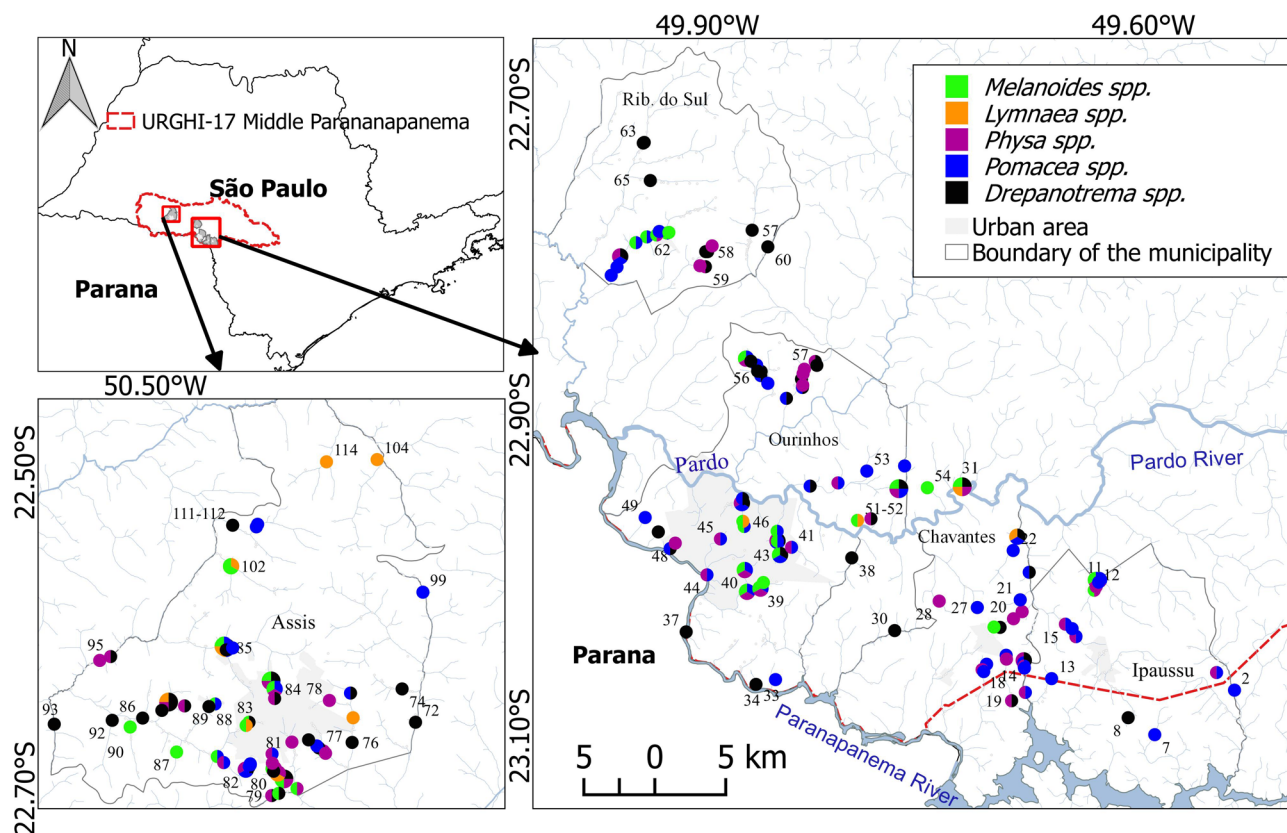
Area	Extension (km <sup>2</sup> )*	N water bodies with taxon <i>i</i>	% of water bodies in each area with taxon <i>i</i>									
			<i>B. glabrata</i>	<i>B. tenagophila</i>	<i>B. straminea</i>	<i>B. occidentalis</i>	<i>B. peregrina</i>	<i>Drepanotrema spp.</i>	<i>Lymnaea spp.</i>	<i>Physa spp.</i>	<i>Pomacea spp.</i>	<i>Melanoideis spp.</i>
Ipaussu	209,6	16	0	39	17	8	27	5	0	11	14	8
Chavantes	188,7	12	0	25	17	16	13	14	17	17	17	8
Ourinhos	296,8	25	80	29	25	41	22	25	17	33	36	36
Ribeirão do Sul	203,2	9	0	0	25	7	15	15	0	10	4	4
Assis	460,6	30	20	7	17	29	23	40	67	29	29	44

2012) indicating that this species was present in bodies of water in Chavantes and Ourinhos. Today, the bodies of water in Ourinhos (more specifically the Christoni, Fundo, and Água da Veada streams) are not colonized by *B. tenagophila*, which may indicate a change in the distribution area of this species in the Middle Paranapanema River Basin region.

A significant change was seen in the distribution of *B. glabrata* compared with studies from other periods and at specific points in the Middle Paranapanema River region. Although *B. glabrata* was described in previous research involving bodies of water in Água de Jacu in Assis and Furninhas in Ourinhos (SUCEN 1980, Tuan 2009) in this current study we found that this species had been replaced by *B. tenagophila*.

Contrasting with previous studies (Piza & Ramos 1960, Teles & Vaz 1987), in bodies of water in the municipalities of Ipaussu, Chavantes, and Ribeirão do Sul no specimens of *B. glabrata* were identified. However, it is difficult to compare the current fauna and species identified in previous studies because of the lack of precise information about the bodies of water and locations where the animals were collected.

In this study we confirmed the existence of five *B. glabrata* hotspots using georeferencing techniques, one in Assis (Fortuninha) and four in Ourinhos (Sobra, Lageadinho, Jacu, and Christoni). This finding allows for the adoption of prophylaxis activities in the affected bodies of water, and also encourages educational efforts to restrict access to these sites. It will also allow comparisons of mollusk species in future studies.



**Figure 6.** Distribution of non-*Biomphalaria* snails collected by collection point during the period 2015–2018, in Ourinhos, Ribeirão do Sul, Ipaussu, Chavantes, and Assis, in the state of São Paulo, Brazil. The numbers correspond to the bodies of water where the snails were collected.

Previous reports show colonization of *B. straminea* in few bodies of water, restricted to the municipalities of Ourinhos (Teles 2005). The current study shows that the geographical area occupied by *B. straminea* has expanded to bodies of water in Assis, Chavantes, and Ipaussu, which is new information. *B. straminea* is a species with a high potential for colonization of new environments far from the neotropical region, as demonstrated by data on colonization of this species in Asia (Woodruff et al. 1985). The current expansion of *B. straminea* throughout the bodies of water in the Middle Paranapanema River region is consequently not surprising.

As for the Planorbidae species, *Drepanotrema* spp. was most commonly found, followed by *Physa* spp., *Pomacea* spp., and *Melanoides* spp. These snails are common benthic mollusk fauna in the continental waters of southeastern Brazil (Paraense 1975, Thiengo et al. 2006, Ohlweiller et al. 2010, Medeiros et al. 2014, Fernandez et al. 2018). They are often associated with polluted environments, due to their high degree of resilience (Moreno & Callisto 2006); their presence in the Middle Paranapanema River near human-altered areas is reasonable. Representatives of these Planorbidae species were easily found in sympatry with *Biomphalaria*. For this reason, *Pomacea* spp. and *Melanoides* spp. have been used in schistosomiasis prevention efforts involving biological control of snails in the genus *Biomphalaria* (Guimarães et al. 2001, Fernandez et al. 2001, Thiengo et al. 2005, BRASIL 2008). In the present study we observed that these vector and non-vector species inhabit the same bodies of fresh water, without any evidence that any one species predominates.

A notable finding was the presence of *Lymnaea columella*, one of three species in the genus *Lymnaea* in which *Fasciola hepatica* (the common liver fluke) develops; this parasite causes fasciolosis in Brazil (Barbosa 1995, Maure et al. 1998, BRASIL 2008).

Our collection method, which subdivided the entire total watershed comprising 114 bodies of water into various sampling points, provided a comprehensive qualitative analysis of freshwater snail species in the Middle Paranapanema River Basin. Additionally, the use of geospatial tools permitted unique and highly effective mapping of the bodies of water.

The region features the intermediate hosts *B. glabrata*, *B. tenagophila*, and *B. straminea* (CVE 2011ab), but *B. occidentalis* and *B. peregrina* predominate in this area and are refractory to *S. mansoni* under natural conditions. These two species were identified in 62 bodies of water, without previous mention in the literature.

Can DNA barcoding replace traditional identification methods? Validation of the DNA barcode as a routine mollusk identification technique requires comparisons of the results obtained using the same technique and methodology by independent laboratories. The results from sampling the Middle Paranapanema River indicate that the base data of COI-Folmer sequences for *Biomphalaria* in GenBank must be broadened in order to represent specimens collected in different geographical areas and cover the entire distribution of each species. There are just 14 COI-Folmer sequences for *B. peregrina*, with the majority of specimens collected from the state of São Paulo. Sequences from species such as *B. orbignyi*, *B. kuhniana*, *B. oligoza*, and *B. schrammi* are absent from GenBank.

Molecular methodology should be used in mollusk analysis alongside traditional methods involving morphological identification of adult snails in order to provide a more comprehensive understanding of each species. The application of DNA barcoding is promising for juvenile and adult specimens in which morphological identification is not precise, and may consequently help improve mollusk research.

In conclusion, the identification of *Biomphalaria* snails to the species level through DNA Barcode complements and enhances the traditional morphological taxonomy. The COI sequences dataset already available in GenBank is valuable to identify immature snails at species level. The COI sequences were grouped into five monophyletic groups, all of them convergent with morphological identification. *B. occidentalis* and *B. peregrina* predominate along the freshwater environments along the portion of the middle Paranapanema river assessed in this study. We identified hotspots for *B. glabrata* and *B. tenagophila* with highest probability for reproducing these two key species for schistosomiasis infection, which can be used in the planning of the disease control.

## Supplementary material

The following online material is available for this article:  
Additional information

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## Author Contributions

Raquel Gardini Sanches Palasio: The manuscript was part of the Doctoral thesis of RGS Palasio. The author contributed with field and molecular data collection, analysis and interpretation of molecular data, geospatial mapping and manuscript preparation adding intellectual content.

Iara Giordano Xavier: Contribution to Data Collection in the field and processing of geographical data.

Francisco Chiaravallotti-Neto: Contribution to Geospatial Data Analysis and interpretation, manuscript preparation, adding intellectual content.

Roseli Tuan: Concept, design and coordination of the research project funded by SUCEN. Molecular and morphological data analysis and interpretation. Manuscript preparation. Critical revision, adding intellectual content.

## Conflicts of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Streblid bat flies (Diptera) and other ectoparasites on bats (Mammalia: Chiroptera) from French Guiana

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GRACIOLLI, G., GUERRERO, R., CATZEFLIS, F. **Streblid bat flies (Diptera) and other ectoparasites on bats (Mammalia: Chiroptera) from French Guiana**. *Biota Neotropica*. 19(4): e20180724. <http://dx.doi.org/10.1590/1676-0611-BN-2018-0724>

**Abstract:** Recent field surveys of bats (Chiroptera) in various localities of French Guiana have been accompanied by the collection and preservation of ectoparasites, mainly bat flies (Diptera: Streblidae and Nycteribiidae). Most specimens of ectoparasites was collected haphazardly during the course of bats inventories, but systematic surveys on the whole chiropteran community were realized at five opportunities. Concerning Streblidae, 813 individuals have been examined, which represent 46 species and/or subspecies belonging to 15 genera and 6 taxa for confirmation and/or future description. For Nycteribiidae, 44 individuals of 3 identified species and 2 for confirmation and/or future description. Other ectoparasites have been found (Hemiptera and acarids), which are also listed with details on their bat-host, place and date of collect. For six species of bats in which at least 10 animals were carrying ectoparasites, a brief description of the frequencies of their ectoparasites provides some preliminary characteristics of their infracommunities.

**Keywords:** *Infracommunity, host-parasite relationship, Guianan bioregion.*

## Moscas estréblidas (Diptera) e outros ectoparasitos de morcegos (Mammalia: Chiroptera) na Guiana Francesa

**Resumo:** Pesquisas de campo recentes de morcegos (Chiroptera) em várias localidades da Guiana Francesa foram acompanhadas pela coleta e preservação de ectoparasitas, principalmente moscas ectoparasitas (Diptera: Streblidae e Nycteribiidae). A maioria dos espécimes de ectoparasitos foi coletada aleatoriamente durante o curso dos inventários de morcegos, mas pesquisas sistemáticas em toda a comunidade de quirópteros foram realizadas em cinco oportunidades. Sobre Streblidae, 813 indivíduos foram examinados, dos quais representam 46 espécies e/ou subespécies pertencentes a 15 gêneros e 6 táxons para posterior confirmação específica e/ou descrição. Para Nycteribiidae, 44 indivíduos de 3 espécies e 2 para posterior confirmação específica e/ou descrição. Outros ectoparasitos foram coletados (hemípteros polictenídeos e ácaros), os quais também foram listados com detalhes sobre seus hospedeiros, localidade e data de coleta. Para seis espécies de morcegos com mais de 10 morcegos infestados a descrição da composição de cada infracomunidade encontrada e sua frequência são apresentados.

**Palavras-chave:** *Infracomunidade, relação hospedeiro-parasito, biorregião da Guiana.*

## Introduction

Bats (Mammalia: Chiroptera) harbour a varied group of ectoparasitic arthropods, most of them occur exclusively on these mammalian hosts. Bat flies (Streblidae and Nycteribiidae), bat fleas (Ischnopsyllidae and one species of *Hectopsylla* [Tungidae]), bat bugs (Cimicidae and Polyctenidae), mites (Cheyletidae, Chirorhynchobiidae, Macronyssidae, Myobiidae, Spelaorhynchidae, and Spinturnicidae), chiggers (Trombiculidae and Leeuwenhoekiidae) and tick (Argasidae and Ixodidae) are found parasiting bats in the Neotropical region (Whitaker et al. 2009). Important taxonomic surveys of ectoparasitic arthropods were carried out in Panama (Wenzel et al. 1966), Venezuela (Guimarães 1972, Jones et al. 1972, Ueshima 1972, Herrin & Tipton 1975, Wenzel 1976, for example), Paraguay (Dick & Gettinger 2005), Peru (Guerrero 1996-a, b), Argentina (see Autino et al. 2009, for example) and Brazil (see Graciolli et al. 2008). But in several areas of South America only occasional collections were made, resulting in a poor knowledge of the biodiversity of these ectoparasites and their host relationship. In French Guiana 12 species of streblid bat flies from six genera have been so far registered (Guerrero 1997).

On the hosts side, bats are rather common in all ecosystems of French Guiana, and several studies have described some aspects of their communities (Simmons & Voss 1998, Charles-Dominique et al. 2001, Catzefflis et al. 2013).

Here we provide new data on streblid bat flies and their hosts and infracomunities, as per a combination of anecdotic and systematic collections carried out in French Guiana. We also provide data on nycteribiid bat flies, bat bugs (Hemiptera: Polyctenidae) and ticks and mites (Acari: Argasidae, Ixodidae, and Spinturnicidae) found on French Guianan bats.

## Material and Methods

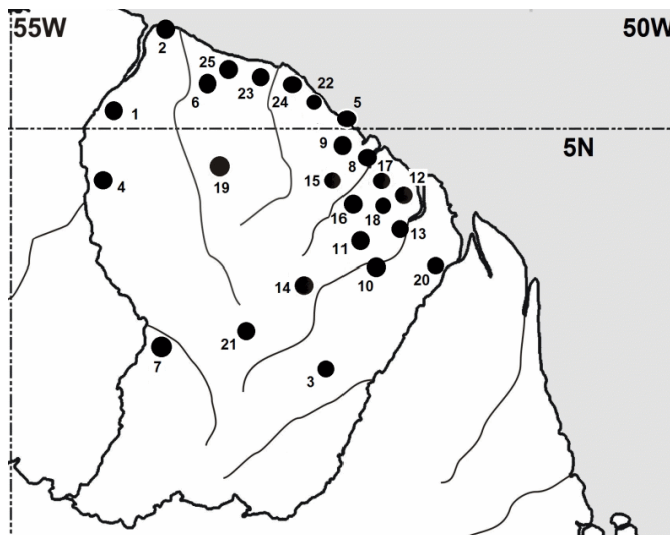
Ectoparasites were collected either haphazardly through the course of various bats inventories by experienced field naturalists or systematically during five surveys aimed at collecting Streblidae.

Altogether, the preserved ectoparasites come from 25 geographic localities spanning most of French Guiana (Figure 1).

Capture methods of Chiroptera included essentially mist nets set across nearby corridors such as trails in the forest. Mist nets of 2.6 × 6 m and 2.6 × 10 m (mesh size = 16 mm) were employed at ground level. In addition, we used at Cacao (locality 15 in Fig. 1) a three-frame harp trap (AUSTBAT Research Equipment, Victoria, Australia), with a catching surface of 1.0 m<sup>2</sup> erected across a trail acting as a corridor for bats flying out of a nearby cave system.

Bats were handled individually in cloth bags, and first examined for the presence of ectoparasites, which were gently collected with the aid of tweezers and brushes through, and preserved in 70% ethanol.

Collected Diptera were taken to the laboratory and identified down to the specific level by Gustavo Graciolli and Ricardo Guerrero, with the aid of the following literature: Guimarães & D'Andretta (1956), Guimarães (1966, 1972), Guerrero (1993, 1994, 1995a-b, 1996a). For Polyctenidae, Ixodidae and Spinturnicidae were consulted Ueshima (1972), Guerrero (1996c), Herrin & Tipton (1975), respectively. For Acarids were mounted in Hoyer medium, and other taxa, when necessary, were examined in Lactophenol. The nomenclature of Streblidae and



**Figure 1.** Map of French Guiana with the localities from where bats were caught with preserved ectoparasites. Localities are numbered 1 to 25 and correspond to: 1 = Apatou; 2 = Awala-Yalimapo; 3 = Camopi; 5 = Kourou; 6 = Mana: Crique Gargoulette along National Road; 7 = Maripasoula: Atachi Bakka; 8 = Matoury: Lagune Concorde; 9 = Montsinery: Crique Cascade; 10 = Regina: Approuague: Saut Athanase; 11 = Regina: Grande Montagne Tortue, piste de Belizon, pK-13; 12 = Regina: Grotte Mathilde; 13 = Regina: Montagne des Gouffres; 14 = Regina: Nouragues; 15 = Roura: Cacao; 16 = Roura: Crique Boulanger; 17 = Roura: grotte Fourgassie; 18 = Roura: Trésor Nature Preservation; 19 = Saint-Elie: Trinité Nature Preservation; 20 = Saint-Georges-Oyapock: Trois-Palétuviers; 21 = Saül: near the village; 22 = Sinnamary: Paracou at CIRAD; 23 = Sinnamary: Piste de Saint-Elie, pK-15; 24 = Sinnamary: National Road, pK-106; 25 = Sinnamary: Yiyi.

Nycteribiidae follows Guerrero (1997) and Graciolli et al. (2007), respectively. For Polyctenidae, Ixodidae and Spinturnicidae, we follow the proposal taxonomic showed in Ueshima (1972), Guerrero (1996c) and Herrin & Tipton (1975).

Bat flies are deposited mainly at Coleção Zoológica de Referência da Universidade Federal de Mato Grosso do Sul, Campo Grande (Brazil), Colección de Parasitología del Museo de Ciencias de la Universidad Central de Venezuela (MBUCV) and also at Institut des Sciences de l'Evolution, Université de Montpellier (France). The other ectoparasites are deposited at MBUCV.

Identification of bats was done primarily based on Simmons & Voss (1998) and on Charles-Dominique et al. (2001), but complementary literature and specimens for comparison were used as well. The nomenclature of bats follows Simmons (2005), with slight changes due to more recent taxonomical studies. Specifically, according to de Thoisy et al. (2014) and to Pavan et al. (2018), all Common Mustached Bats are identified either as *Pteronotus rubiginosus* (Wagner, 1843) - emitting at 53 kHz - or as *Pteronotus alitonus* Pavan, Bobrowiec & Percequillo, 2018 - emitting at 59 kHz -. When no voucher has been preserved, and/or no molecular or bioacoustic character is available for a proper taxon identification, we name the Common Mustached Bats *Pteronotus* group-*parnellii*, as they were known in French Guiana before the study of de Thoisy et al. (2014). Following Velazco & Patterson (2013; 2019), we name *Sturnira giannae* Velazco & Patterson, 2019 all Little Yellow-shouldered Bats who were previously known as *Sturnira lilium* throughout the Guianan countries.

External measurements (forearm, weight) were recorded for all captured bats after which they were released in the same spot. A small biopsy punch (Worthington & Barratt 1996) used for DNA analyses was also taken from some individuals prior to release, and a small selection of specimens were preserved as scientific vouchers for further cranial measurements. These vouchered specimens were euthanized, fixed in 10% buffered formalin and finally stored in 70% ethanol. Appendix 1 lists the origin and institutions housing these vouchers. Bats were caught, handled, kept and euthanized following the guidelines of the American Society of Mammalogists for the use of wild mammals in research (Sikes et al. 2011). As no specific decree concerning Chiroptera outside protected areas exist in French Guiana, no specific legal authorization was required for captures and handling of bats. Vouchered bats were deposited in the collection of Museum d'Histoire Naturelle de Genève (MHNG).

The material examined is sorted by alphabetical order of species within subfamilies and families of ectoparasites. For each ectoparasite taxon, a few relevant comments are presented below, including at least the host species, locality and date of collect. Three species in the genus *Trichobius* (*longipes* group of taxa) appear new for science, and will be described in a separate paper.

For species of hosts with more than 10 infected individuals, we show the frequency of infracommunities found.

## Results

During the five systematic surveys, 660 bats (of 56 species) were caught and examined for ectoparasites: 129 individuals (or 20%) were found carrying bat flies. When considering only the eight species of bats caught each by at least 20 individuals, prevalence of bats harbouring ectoparasites ranges from zero (on two molossid species: *Molossus coibensis* and *M. molossus*) to 37% (17 out of 46 *Pteronotus alitonus*), with an average of 13.5% (62 infested hosts out of 458 captured bats).

The ectoparasites listed here derive from 204 infested bats belonging to 37 species. Regarding hosts, we found ectoparasites on 75 (opportunistic) and 129 (systematic) individuals, which were from 24 (opportunistic) and 31 (systematic) species of bats.

## Insecta

### DIPTERA

#### Streblidae Kolenati, 1863

We examined 813 streblid bat flies of for a total of 46 species/subspecies and 15 genera and 6 taxa for confirmation and/or future description, of which 35 species/subspecies and 8 genera are registered for the first time in French Guiana.

#### Nycterophiliinae Wenzel, 1966

##### *Nycterophilia coxata* Ferris, 1916

Examined material: French Guiana: Regina: Nouragues, 1 male, ex *Pteronotus* group-*parnellii*, Maxime Cobigo leg., 6/III/2013.

Remarks: New geographic record for French Guiana.

##### *Nycterophilia parnellii* Wenzel, 1966

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Pteronotus* group-*parnellii*, F. Catzefflis and M. Ruedi legs., 13/VII/2012.

Remarks: New geographic record for French Guiana.

#### Streblinae Speiser, 1900

##### *Anastrebla caudiferae* Wenzel, 1976

Examined material: French Guiana: Roura: Cacao, 1 male and 1 female, ex *Anoura caudifer*, Francois Catzefflis leg., 9/VII/2012.

Remarks: New geographic record for French Guiana.

##### *Anastrebla modestini* Wenzel, 1966.

Examined material: French Guiana: Cacao, Va-Joua, 1 male, ex *Anoura geoffroyi*, Benoit de Thoisy leg., 11/VI/2011.

Remarks: New geographic record for French Guiana.

##### *Anastrebla spurrelli* Wenzel, 1976

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 female, ex *Lionycteris spurrelli*, M. Delaval and V. Ruffray leg., 10/IX/2011; same locality, 1 male, same host and collectors but 12/IX/2011.

Remarks: New geographic record for French Guiana.

##### *Metelasmus pseudopterus* Coquillett, 1907

Examined material: French Guiana: Regina: Nouragues, 1 female, ex *Artibeus planirostris*, M. Cobigo leg., 14/VIII/2013. Roura: Cacao, 1 male, same host, Francois Catzefflis leg., 10/VII/2012.

Remarks: New geographic record for French Guiana.

##### *Strebla alvarezii* Wenzel, 1966

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Micronycteris megalotis*, Francois Catzefflis leg., 11/VII/2012. Saint-Elie: Trinité Nature Preservation, 1 female, same host, J. F. Cosson leg., 12/IX/1991.

Remarks: Previously quoted by Guerrero (1997).

##### *Strebla christinae* Wenzel, 1966

Examined material: French Guiana: Regina: Grande Montagne Tortue, piste de Belizon, pK-13, 2 males, ex *Phyllostoma stenops*, Margot Delaval leg., 13/X/2010. Maripasoula, Atachi Bakka, 13 males and 7 females, ex 2 *P. stenops*, Sylvain Uriot leg., IX/2011.

Remarks: New geographic record for French Guiana.

##### *Strebla consocia* Wenzel, 1966

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 female, ex *Phyllostoma elongatus*, M. Delaval and V. Ruffray leg., 6/IX/2011; same locality, 1 male, same host and collectors but 12/IX/2011; same locality, 1 male and 1 female, same host and collectors but 14/IX/2011; same locality, 1 male, ex *Phyllostoma hastatus*, M. Delaval and V. Ruffray leg., 10/IX/2011; same locality, 1 female, same host and collectors but 11/IX/2011; same locality, 5 males and 5 females, ex 3 *P. hastatus*, same collectors but 15/IX/2011. Maripasoula: Atachi Bakka, 3 males and 4 females ex 2 *P. hastatus*, Sylvain Uriot leg., IX/2011. Sinnamary: Paracou at CIRAD-camp, 1 male, ex *Artibeus obscurus*, Francois Catzefflis leg., 10/X/2013. Sinnamary: Piste de Saint-Elie, pK-15, 1 female, ex *P. hastatus*, J.-F. Cosson leg., 23/I/1990; same locality, 1 male and 1 female, ex *Pteronotus rubiginosus*, J.-F. Cosson leg., 8/XI/1991. Awala-Yalimapo, 1 male, ex *P. hastatus*, M. Delaval leg., 29/X/2011. Roura: Cacao, 1 male and 2 females, same host, 14/VII/2012.

Remarks: Previously quoted by Guerrero (1997).



*Strebla diaemi* Wenzel, 1966

Examined material: French Guiana: Regina: Approuague, Saut Athanase, 2 males and 3 females, ex 2 *Diaemus youngi*, Benoit de Thoisy leg., 18/VIII/2012.

Remarks: New geographic record for French Guiana.

*Strebla guajiro* (García & Casal, 1965)

Examined material: French Guiana: Roura: Cacao, 3 females, ex 2 *Carollia perspicillata*, Francois Catzefflis leg., 19/VI/2011; same locality, 1 female, same host and collector but 21/VI/2011; same locality, 4 females, ex 2 *C. perspicillata*, same collector, 7/VII/2012; same locality, 5 males and 2 females, same host and collector but 9/VII/2012; same locality, 5 males and 2 females, same hosts and collector but 9/VII/2012; same locality, 1 male and 1 female, same host and collector but 12/VII/2012; same locality, 1 female, same host, Benoit de Thoisy leg., 11/XI/2011. Sinnamary: Piste de Saint-Elie, pK-15, 1 male, same host, J.-F. Cosson leg., 1990.

Remarks: Previously quoted by Guerrero (1997).

*Strebla harderi* Wenzel, 1976

Examined material: French Guiana: Roura: Cacao, 2 males, ex *Lonchophylla thomasi*, Francois Catzefflis leg., 9/VII/2012.

Remarks: New geographic record for French Guiana.

*Strebla hertigi* Wenzel, 1966

Examined material: French Guiana: Sinnamary: Piste de Saint-Elie, pK-15, 1 male, ex *Phyllostomus elongatus*, J.-F. Cosson leg., 1/VI/1990. Roura: Cacao, 2 males, ex *Phyllostomus discolor*, Francois Catzefflis leg., 14/VII/2012.

Remarks: Previously quoted by Guerrero (1997).

*Strebla mirabilis* (Waterhouse, 1879)

Examined material: French Guiana: Roura: grotte de Fourgassier, Route de Kaw, 1 male and 1 female, ex *Trachops cirrhosus*, Benoit de Thoisy leg., 12/VI/2011. Saint-Elie: Trinité Nature Preservation, 1 male, same host, M. Delaval and V. Ruffray legs., 10/IX/2011.

Remarks: Previously quoted by Guerrero (1997).

*Strebla obtusa* Wenzel, 1976

Examined material: French Guiana: Apatou: 1 male and 1 female, ex *Trinycteris nicefori*, Margot Delaval leg., 04/09/2010. Roura, Cacao: 3 males and 1 female, ex *T. nicefori*, Francois Catzefflis leg., 9/VII/2012; same locality, 7 males and 1 female, same host and collector but 21/VII/2012.

Remarks: New geographic record for French Guiana.

*Strebla wiedemanni* Kolenati, 1856

Examined material: French Guiana: Regina: Nouragues, Arataye, 2 males, ex *Desmodus rotundus*, Margot Delaval leg., 30/X/2010. Roura: grotte de Fourgassier (Route de Kaw), 6 males and 3 females, ex 3 *D. rotundus*, Benoit de Thoisy leg., 12/VI/2011. Saut-Athanase, Fleuve Approuague, 2 males, ex *D. rotundus*, Benoit de Thoisy leg., 26/VI/2011; same locality, 2 males and 1 male, same host, M. Delaval leg., 20/XI/2011. Saint-Elie, Trinité Nature Preservation, 2 males and 2 females, same host, M. Delaval and V. Ruffray legs., 14/IX/2011. Regina, grotte Mathilde, 4 males and 4 females, same host, M. Delaval

leg., 27/I/2013. Regina, Approuague, Saut Athanase, 2 males, same host, Benoit de Thoisy leg., 26/VI/2011

Remarks: New geographic record for French Guiana.

*Strebla* cf. *paramirabilis*

Examined material: French Guiana: Roura: Cacao, 1 female, ex *Mimon bennetti*, Francois Catzefflis leg., 12/VII/2012.

*Strebla* sp.

Examined material: French Guiana: Roura: Cacao, 1 female, ex *Lonchophylla thomasi*, Francois Catzefflis, 13/VII/2012.

## Trichobiinae Jobling, 1936

*Aspidoptera falcata* Wenzel, 1976

Examined material: French Guiana: Roura: Cacao, 2 males and 2 females, ex *Sturnira giannae*, Francois Catzefflis leg., 19/VI/2011, same locality, 9 males and 8 females, same host and same collector, 21/VI/2011; same locality, 1 male and 1 female, same host, F. Catzefflis and M. Ruedi legs., 6/VII/2012; same locality, 3 females, same hosts and collectors but 9/VII/2012.

Remarks: New geographic record for French Guiana. This represents a new host record. *Aspidoptera falcata* is often found on bats of the genus *Sturnira* (Guerrero 1995b).

*Aspidoptera phyllostomatis* (Perty, 1833)

Examined material: French Guiana: Roura: Cacao, 1 male and 2 females, ex 3 *Artibeus planirostris*, Francois Catzefflis leg., 10/VII/2012; same locality, 1 male, same host and collector but 21/VI/2011. Regina, Nouragues, 1 male and 4 females, same host, M. Cobigo leg., 14/VIII/2013.

Remarks: New geographic record for French Guiana.

*Mastoptera minuta* (Costa Lima, 1921)

Examined material: French Guiana: Saint-Elie, Trinité Nature Preservation, 1 male, ex *Lophostoma sylvaticum*, M. Delaval and V. Ruffray legs., 14/IX/2011; same locality, 4 males and 7 females, ex *Phyllostomus hastatus*, same collectors, 10/IX/2011; same locality, 1 male and 2 females, same host, same collectors but 15/IX/2011. Roura: Cacao, 3 males and 3 females, same host, F. Catzefflis and M. Ruedi legs., 10/VII/2012.

Remarks: New geographic record for French Guiana.

*Megistopoda aranea* (Coquillett, 1899)

Examined material: French Guiana: Roura: Cacao, 1 male and 1 female, ex 2 *Artibeus planirostris*, F. Catzefflis leg., 10/VII/2012.

Remarks: New geographic record for French Guiana.

*Megistopoda proxima* (Séguy, 1926)

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Sturnira giannae*, F. Catzefflis leg., 19/VI/2011; same locality, 4 males ex 3 *Sturnira giannae*, same collector but 21/VI/2011; same locality, 1 male and 1 female, same host, Catzefflis and M. Ruedi legs., 9/VII/2012; same locality, 1 male, ex *Sturnira tildae*, same collectors, 12/VII/2012; same locality, 1 male, *Sturnira giannae*, Francois Catzefflis leg., 23/VII/2012. Kourou, 1 female, ex *Sturnira* sp., J.-F. Cosson leg., no data.

Remarks: Previously quoted by Guerrero (1997). This represents a new host record. *Megistopoda proxima* is often found on bats of the genus *Sturnira* (Guerrero 1995b).

*Neotrichobius bisetosus* Wenzel, 1976

Examined material: French Guiana: Saint-Elie, Trinité Nature Preservation, 1 male, ex *Artibeus planirostris*, M. Delaval and V. Ruffray legs., 14/IX/2011. Regina, Nouragues, 2 males and 1 female, same host, M. Cobigo leg., 14/VIII/2013.

Remarks: New geographic record for French Guiana.

*Neotrichobius delicatus* (Machado-Allison, 1966)

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Rhinophylla pumilio*, F. Catzefflis leg., 11/VI/2011; 1 male and 2 females, ex 2 *R. pumilio*, F. Catzefflis and M. Ruedi legs., 7/VII/2012. Saint-Elie: Trinité Nature Preservation, 1 female, same host, M. Delaval and V. Ruffray legs., 12/IX/2011.

Remarks: New geographic record for French Guiana.

*Neotrichobius delicatus* complex

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Dermanura gnoma*, F. Catzefflis leg., 26/VI/2011.

*Neotrichobius stenopterus* Wenzel & Aitken, 1966

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Pteronotus group-parnelli*, F. Catzefflis and M. Ruedi legs., 13/VII/2012; same locality, 1 female, ex *Dermanura gnoma*, Francois Catzefflis leg., 18/VII/2012.

Remarks: New geographic record for French Guiana.

*Noctiliostrebla aitkeni* Wenzel, 1966

Examined material: French Guiana: Awala-Yalimapo, 4 males and 6 females, ex 3 *Noctilio leporinus*, M. Delaval leg., 30/X/2011.

Remarks: New geographic record for French Guiana. Recently Alcantara et al. (2019) published a taxonomic revision of *Noctiliostrebla*, restricting the records of *N. aitkeni* for Brazil, Ecuador, Trinidad and Tobago and Venezuela.

*Noctiliostrebla maai* Wenzel, 1966

Examined material: French Guiana: Awala-Yalimapo, 4 males and 5 females, ex 4 *Noctilio albiventris*, M. Delaval leg., 30/X/2011.

Remarks: New geographic record for French Guiana. Previously this species was recorded in Colombia, Panama and Venezuela (Alcantara et al. 2019).

*Paradyschiria lineata* Kessel, 1925

Examined material: French Guiana: Awala-Yalimapo, 7 males and 6 females, ex 3 *Noctilio leporinus*, M. Delaval leg., 30/X/2011. Matoury: Lagune Concorde, 2 males and 1 female, same host and collector, 4/XI/2011. Sinnamary: Yiyi, 2 females, same host, J.-F. Cosson leg., 8/XI/1991.

Remarks: New geographic record for French Guiana.

*Paradyschiria parvula* Falcoz, 1931

Examined material: French Guiana: Awala-Yalimapo, 18 males and 16 females, ex 7 *Noctilio albiventris*, M. Delaval leg., 31/X/2011.

Remarks: New geographic record for French Guiana.

*Parastrebla handleyi* Wenzel, 1966

Examined material: French Guiana: Apatou, *Trinycteris nicefori*, 3 males, Margot Delaval leg., 04/09/2010. Roura: Cacao, 1 female, same host, Francois Catzefflis leg., 21/VII/2012.

Remarks: New geographic record for French Guiana.

*Pseudostrebla riberoi* Costa Lima, 1921

Examined material: French Guiana: Sinnamary: Piste de Saint-Elie, pK-15, 1 female, ex *Micronycteris megalotis*, J.-F. Cosson leg., 13/I/1990.

Remarks: Previously quoted by Guerrero (1997) as *Pseudostrebla microtis* litt. error, a *nomem nudum*.

*Speiseria ambigua* Kessel, 1925

Examined material: French Guiana: Sinnamary: Piste de Saint-Elie pK-15, 1 female, ex *Carollia perspicillata*, 1990. Roura: Cacao: Va-Joua, ex *C. perspicillata*, 3 males and 3 females, ex 4 *C. perspicillata*, Benoit de Thoisy leg., 11/VI/2011; same locality, 1 male, same host, F. Catzefflis leg., 19/VI/2011; same locality, 1 male and 2 females, same host and collector but 21/VI/2011. Maripasoula: Atachi Bakka, 1 female, ex *Trachops cirrhosus*, Sylvain Uriot leg., 1/IX/2011. Saint-Elie: Trinité Nature Preservation, 1 male, same host, M. Delaval and V. Ruffray legs., 10/IX/2011. Roura: Cacao, 1 male, ex *Trinycteris nicefori*, F. Catzefflis leg., 9/VII/2012; same locality, 1 male and 1 female, ex *C. perspicillata*, same collector and date; same locality, 1 female, same host and collector but 7/VII/2012. Saint Georges-Oyapock: Trois Paletuviers, 2 females, same host and collector, 5/VII/2013.

Remarks: Previously quoted by Guerrero (1997).

*Speiseria magniocularis* Wenzel, 1976

Examined material: French Guiana: Maripasoula: Atachi Bakka, 1 female, ex *Trachops cirrhosus*, Sylvain Uriot leg., 1/IX/2011.

Remarks: New geographic record for French Guiana.

*Trichobioides perspicillatus* (Pessôa and Galvão, 1937)

Examined material: French Guiana: Roura: Cacao, 1 male and 1 female, ex *Phyllostomus discolor*, Francois Catzefflis leg., 14/VII/2012.

Remarks: New geographic record for French Guiana.

*Trichobius anducei* Guerrero, 1998

Examined material: French Guiana: Roura: Cacao: Va-Joua, 8 males and 4 females, ex 4 *Carollia perspicillata*, Benoit de Thoisy leg., 11/VI/2011; same locality, 4 males and 6 females, ex 2 *C. perspicillata*, Francois Catzefflis leg., 19/VI/2011; same locality, 3 males and 1 female, same host and collector, 21/VI/2011; same locality, 4 females, ex 2 *C. perspicillata*, same collector, 7/VII/2012; same locality, 1 male, same host and collector but 9/VII/2012.

Remarks: New geographic record for French Guiana.

*Trichobius caecus* Edwards, 1918

Examined material: French Guiana: Sinnamary: Piste de Saint-Elie, pK-15, 4 males, 1 female, ex *Pteronotus rubiginosus*, J.-F. Cosson leg., 8/XI/1991. Grand-Santi: Gaa Kaba (Montagne Francaise), 1 male and 3 females, same host, M. Delaval leg., 22/IX/2010. Regina: Grande Montagne Tortue, piste de Belizon, pK13, 3 males, same host, M. Delaval leg., 15/X/2010. Regina: Grotte Mathildae, 1 male, 1 female, ex *P. rubiginosus*, K. Pineau and M. Delaval legs., 2011; same locality,

2 males and 2 females, ex 2 *Pteronotus* group-*parnelli*, same collectors and date. Roura: Trésor Nature Preservation, 3 males and 3 females ex 3 *Pteronotus* group-*parnelli*, K. Pineau and M. Delaval legs., 2011; same locality, 1 female, ex *P. rubiginosus*, same collectors and date. Saint-Elie: Trinité Nature Preservation, 6 males and 10 females, ex 2 *Pteronotus alitonus*, M. Delaval and V. Ruffray legs., 9/IX/2011; same locality, 25 males and 13 females, ex 4 *Pteronotus alitonus*, same collectors, 10/IX/2011; same locality, 7 males and 3 females, ex *Natalus tumidirostris*, same collectors, 12/IX/2011; same locality, 2 males and 2 females, ex *Pteronotus alitonus*, same collectors, 14/IX/2011; same locality, 8 males and 3 females, same host and collectors, 15/IX/2011. Awala-Yalimapo, 1 male, ex *Noctilio albiventris*, M. Delaval leg., 31/X/2011. Regina: Montagne des Gouffres, 2 males and 2 females, M. Delaval leg., 3 males and 1 female, ex *Pteronotus alitonus*, same collector, 22/XI/2011; same locality, 1 male and 1 female, ex *P. rubiginosus*, same collector and date. Roura: Cacao, 2 males and 2 females, ex *Pteronotus alitonus*, F. Catzefflis and M. Ruedi legs., 9/VII/2012; same locality, 3 males, ex 2 *Pteronotus alitonus*, same collectors, 11/VII/2012; same locality, 1 female, same host and collector, 12/VII/2012; same locality, 8 males and 13 females, ex 4 *P. rubiginosus*, same collectors, 13/VII/2012; same locality, 9 males and 15 females, 4 ex *Pteronotus alitonus*, same collectors and date; same locality, 6 males and 4 females, same host and collectors, 14/VII/2012; same locality, 1 male and 1 female, ex *P. rubiginosus*, same collectors and date; same locality, 17 males and 18 females, same host and collectors, 19/VII/2012. Regina: Nouragues, 6 males and 3 females ex *P. rubiginosus*, Maxime Cobigo leg., 12/X/2012; same locality, 4 males and 3 females, same host and collector, 6/III/2013; same locality, 11 males and 5 females, same host and collector, 12/VIII/2013.

Remarks: In French Guiana, Guerrero (1997) had already recorded this species on *Pteronotus* group-*parnelli* without information about locality. The occurrence of *T. caecus* on *P. alitonus* represents a new host record. Other species of *Pteronotus* parasitized by *T. caecus* are *P. davyi* and *P. parnellii* (Guerrero 1994).

#### *Trichobius diaemi* Wenzel, 1976

Examined material: French Guiana: Regina. Approuague: Saut Athanase. 10 males and 6 females, ex 2 *Diaemus youngi*, Benoit de Thoisy, 18/VIII/2012.

Remarks: New geographic record for French Guiana.

#### *Trichobius dugesioides dugesioides* Wenzel, 1966

Examined material: French Guiana: Saul: near the village, 1 female, ex *Carollia perspicillata*, J.-F. Cosson, leg., 18/VII/1990. Regina: Grande Montagne Tortue, piste de Belizon, pK-13, 2 females, *Trachops cirrhosus*, Margot Delaval leg., 16/X/2010. Roura: grotte de Fourgassier (Route de Kaw), 2 females, same host, Benoit de Thoisy leg., 12/VI/2011. Maripasoula: Atachi Bakka, 1 male and 5 females, ex 3 *T. cirrhosus*, Sylvain Uriot leg., IX/2011. Saint-Elie: Trinité Nature Preservation, 1 female, same host, M. Delaval and V. Ruffray legs., 10/IX/2011. Roura: Cacao, 1 male and 2 females, ex *Mimon bennettii*, Francois Catzefflis leg., 14/VII/2012.

Remarks: In French Guiana, Guerrero (1997) had already recorded this species on *Carollia perspicillata* from Saül.

#### *Trichobius dugesioides phyllostomus* Guerrero, 1998

Examined material: French Guiana: Regina: Grande Montagne Tortue, piste de Belizon, pK-13, 1 male, ex *Phyllostomus elongatus*, Margot Delaval leg., 13/X/2010. Maripasoula: Atachi Bakka, 7 males and 3 females, same host, Sylvain Uriot leg., IX/2011. Saint-Elie: Trinité Nature Preservation, 1 male and 1 female, same host, M. Delaval and V. Ruffray leg. 6/IX/2011; Saint-Elie: Trinité Nature Preservation, 1 male and 1 female, same host, M. Delaval and V. Ruffray leg. 12/IX/2011; Saint-Elie: Trinité Nature Preservation, 1 male and 3 females, same host, M. Delaval and V. Ruffray leg. 14/IX/2011.

Remarks: New geographic record for French Guiana.

#### *Trichobius joblingi* Wenzel, 1966

Examined material: French Guiana: Sinnamary: Piste de Saint-Elie, pK-15, 2 males and 3 females, ex *Carollia perspicillata*, J.-F. Cosson leg., 1990. Maripasoula: Atachi Bakka, 1 male and 1 female, ex *Trachops cirrhosus*, Sylvain Uriot leg., IX/2011. Roura: Cacao: Va-Joua, *C. perspicillata*, 1 female, Francois Catzefflis leg., 19/VI/2011; same locality, 1 female, same collector, 7/VII/2012; same locality, 2 males and 4 females, ex 2 *C. perspicillata*, Francois Catzefflis leg., 9/VII/2012; same locality, 3 males and 3 females, same host and collector but 12/VII/012; same locality, 2 males, ex *Micronycteris megalotis*, Francois Catzefflis leg., 11/VII/2012. Saint Georges-Oyapock: Trois-Paletuviers, 3 females, ex *Carollia perspicillata*, Francois Catzefflis leg., 5/VII/2013.

Remarks: In French Guiana, Guerrero (1997) had already recorded this species on *Carollia perspicillata* without information about locality.

#### *Trichobius jonhsonae* Wenzel, 1966

Examined material: French Guiana: Roura: Cacao: Va-Joua, 1 male and 1 female, *Pteronotus rubiginosus*, Francois Catzefflis leg., 11/VI/2011; same locality, 11 males and 24 females, *Pteronotus personatus*, F. Catzefflis & M. Ruedi legs., 23/VII/2012; same locality 2 males and 1 female, same host and collectors, 25/VII/2012; same locality 9 males and 3 females, same host and collectors, 27/VII/2012.

Remarks: New geographic record for French Guiana.

#### *Trichobius keenani* Wenzel, 1966

Examined material: French Guiana: Roura: Cacao, 1 male, ex *Micronycteris megalotis*, Francois Catzefflis leg., 11/VII/2012.

Remarks: New geographic record for French Guiana.

#### *Trichobius lionycteridis* Wenzel, 1966

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 male and 1 female, ex 2 *Lionycteris spurrelli*, M. Delaval and V. Ruffray legs., 10/IX/2011; same locality, 2 males and 1 female, ex 2 *L. spurrelli*, same collectors, 12/IX/2011; same locality, 6 males and 3 females, ex 3 *L. spurrelli*, same collectors, 15/IX/2011. Roura: Cacao, 1 male and 1 female, same host, F. Catzefflis leg., 10/VII/2012.

Remarks: New geographic record for French Guiana.

#### *Trichobius longipes* (Rudow, 1871)

Examined material: French Guiana: Roura: Cacao, 4 males, ex *Phyllostomus hastatus*, F. Catzefflis and M. Ruedi legs., 14/VII/2012. Maripasoula: Atachi Bakka, 1 male, ex *Phyllostomus elongatus*, Sylvain

Uriot leg., IX/2011. Saint-Elie: Trinité Nature Preservation, 2 males and 2 females, ex *P. hastatus*, M. Delaval and V. Ruffray legs., 9/IX/2011; same locality, 1 male and 2 females, same host and collectors, 10/IX/2011; same locality, 2 males and 5 females, ex 3 *P. hastatus*, same collectors, 15/IX/2011: Awala-Yalimapo, 1 female, same host, M. Delaval leg., 28/X/2011.

Remarks: New geographic record for French Guiana.

*Trichobius parasiticus* Gervais, 1844

Examined material: French Guiana: Regina: Grande Montagne Tortue, piste de Belizon, pK-13, 1 male, ex *Desmodus rotundus*, Margot Delaval leg., 14/X/2010. Roura: grotte de Fourgassier (Route de Kaw), 9 males and 5 females, ex 4 *D. rotundus*, Benoit de Thoisy leg., 12/VI/2012; same locality, 3 males and 4 females, same host, M. Delaval leg., 20/XI/2011. Régina: Saut-Athanase, Fleuve Approuague, *D. rotundus*, 1 male, Benoit de Thoisy leg., 26/VI/2011. Regina: Approuague: Saut Athanase, 1 male, same host and collector but 26/VI/2011. Saint-Elie: Trinité Nature Preservation, 1 female, same host, M. Delaval and V. Ruffray legs., 14/IX/2011.

Remarks: New geographic record for French Guiana.

*Trichobius sparsus* Kessel, 1925

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 2 males, ex *Pteronotus alitonus*, M. Delaval and V. Ruffray legs., 10/IX/2011; same locality, 1 female, same host and collectors but 14/IX/2011; 1 female, same host and collectors but 15/IX/2011.

Remarks: New geographic record for French Guiana.

*Trichobius tiptoni* Wenzel, 1976

Examined material: French Guiana: Sinnamary Piste de Saint-Elie pK-15, 2 males, ex *Anoura caudifer*, J.-F. Cosson, 1990.

Remarks: Previously quoted by Guerrero (1997).

*Trichobius* sp. (*parasiticus* complex)

Examined material: French Guiana: Cacao: Va-Joua, ex *Micronycteris* sp., 1 male and 2 females, Benoit de Thoisy leg., 11/VI/2011.

*Trichobius* sp. n. 1 (*longipes* group)

Examined material: French Guiana: Mana: Crique Gargoulette along National Road, 2 males and 1 female, ex *Molossus rufus*, M. Delaval leg., 2/XI/2011.

*Trichobius* sp. n. 2 (*longipes* group)

Examined material: French Guiana: Roura: Cacao, 3 males and 1 female, ex *Lonchophylla thomasi*, F. Catzefflis leg., 9/VII/2012.

*Trichobius* sp. n. 3 (*longipes* group)

Examined material: French Guiana: Sinnamary: Paracou at CIRAD camp, 3 males, 1 female, ex *Artibeus obscurus*, F. Catzefflis leg., 10/X/2013.

Nycteribiidae Samouelle, 1819

*Basilia carteri* Scott, 1936

Examined material: French Guiana: Montsinery: Crique Cascade, ex *Myotis albescens*, 2 males and 6 females, M. Dewynter leg., 31/VII/2011. Regina: Grotte Mathilde, 1 female, ex *Myotis cf riparius*, M. Delaval leg., 2/VII/2011; same locality, 1 male and 1 female, ex *Myotis nigricans*, Francois Catzefflis leg., 25/VII/2012; same locality, 1 male and 1 female, ex *Myotis* sp., same collector and date. Awala-Yalimapo, 1 male, ex *Myotis* sp., same collector, 29/X/2011. Roura: Crique Boulanger, 1 female, same host and collector, 7/XI/2011. Sinnamary: National Road, pK-106, 3 males and 4 females, ex 3 *Myotis* sp., F. Catzefflis leg., 1/X/2013; same locality, 1 female, same host and collector but 7/X/2013.

Remarks: New geographic record for French Guiana.

*Basilia ortizi* Machado-Allison, 1963

Examined material: French Guiana: Awala-Yalimapo, 6 males and 2 females, ex *Eptesicus furinalis*, M. Delaval leg., 31/X/2011.

Remarks: New geographic record for French Guiana.

*Basilia* sp.

Examined material: French Guiana: Sinnamary: Yiyi, 2 females, ex *Eptesicus furinalis*, M. Delaval leg., 3/XI/2011.

Remarks: This specimen could be an undescribed species belongs to *ferruginea* group.

*Basilia travassosi* Guimarães, 1938

Examined material: French Guiana: Awala-Yalimapo, 2 males and 4 females, ex 3 *Myotis* sp., M. Delaval leg., 29/X/2011.

Remarks: New geographic record for French Guiana. Previously, this species was restricted Northeastern of Brazil (Ceará, Maranhão, Paraíba, and Pernambuco states) on species of *Myotis* (Gracioli et al., 2007; Santos et al., 2013; Barbier et al. 2016; Bezerra & Bocchiglieri, 2018; Barbier et al., 2019). This extending the distribution of *B. travassosi* more than 1,500 km (distance between Awala-Yalimapo and Bairrerinhas, Maranhão).

*Basila* cf. *mimoni* Theodor & Peterson, 1964

Examined material: French Guiana: Camopi: Pic Coudreau du Sud, 1 male and 4 females, ex *Mimon crenulatum*, M. Delaval leg., 4/II/2013.

HEMIPTERA

Polycetenidae Westwood, 1874

*Hesperoctenes cartus* Jordan, 1922

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 male and 1 female, ex *Cynomops paranus*, M. Delaval and V. Ruffray legs., 10/IX/2011. Mana: Crique Gargoulette along National Road, 1 female, ex *Cynomops abrasus*, M. Delaval leg., 2/XI/2011. Sinnamary: Yiyi, 1 female, same host and collector, 3/XI/2011; same locality, 2 males and 6 females, ex 3 *Cynomops planirostris*, same collector and date.

Remarks: New geographic record for French Guiana.



*Hesperoctenes fumarius* (Westwood, 1874)

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 female, ex *Molossus rufus*, M. Delaval and V. Ruffray legs., 14/IX/2011. Mana: Crique Gargoulette along National Road, 1 male, ex *Cynomops abrasus*, M. Delaval leg., 2/XI/2011; 1 male, 1 female and 2 nymphs, ex 2 *M. rufus*, same collector but 1/XI/2011.

Remarks: New geographic record for French Guiana.

**Acarina**

## IXODOIDEA

## Argasidae Canestrini 1890

*Ornithodoros hasei* (Schulze, 1935)

Examined material: French Guiana: Awala-Yalimapo, 3 larvae, ex 2 *Noctilio leporinus*, M. Delaval leg., 30/X/2011; same locality, 6 larvae, ex *Noctilio albiventris*, same collector, 31//2011.

Remarks: New geographic record for French Guiana.

*Ornithodoros marinkellei* Kohls, Clifford & Jones, 1969

Examined material: French Guiana: Roura: Cacao, 1 larva, ex *Ptenotus rubiginosus*, F. Catzeffis and M. Ruedi legs., 13/VII/2012.

Remarks: New geographic record for French Guiana.

*Ornithodoros* sp.

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 larva, ex *Micronycteris megalotis*, J.-F. Cosson leg., 12/IX/1991. Same locality, 2 larvae, ex 2 *Pteronotus alitonus*, M. Delaval and V. Ruffray legs., 9/IX/2011. Roura: Trésor Nature Preservation, 1 larva, ex *Pteronotus rubiginosus*, K. Pineau and M. Delaval legs., 2011.

## DERMANYSSOIDEA

## Spinturnicidae Oudemans, 1901

*Spinturnix americanus* Banks, 1902

Examined material: French Guiana: Montsinery: Crique Cascade, 1 female, ex *Myotis albescens*, M. Dewynter leg., 31/VII/2011. Roura: Cacao, 1 female, ex *Myotis nigricans*, F. Catzeffis leg., 25/VII/2012. Sinnamary: National Road, pK106, 1 male and 1 protonymph, ex *Myotis* sp., same collector, 1/X/2013.

Remarks: New geographic record for French Guiana.

*Spinturnix surinamensis* Dusbabek and Lukoschus, 1971

Examined material: French Guiana: Sinnamary: Yiyi, 1 female, ex *Eptesicus furinalis*, M. Delaval leg., 3/XI/2011.

Remarks: New geographic record for French Guiana.

*Periglischrus micronycteridis* Furman, 1966

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 1 female, ex *Micronycteris megalotis*, J.-F. Cosson leg., 12/IX/1991. Roura: Cacao, 1 male, ex *Trinycteris nicefori*, F. Catzeffis leg., 21/VII/2012.

Remarks: New geographic record for French Guiana.

*Periglischrus paravargasi* Herrin and Tipton, 1975

Examined material: French Guiana: Saint-Elie: Trinité Nature Preservation, 6 females, ex *Anoura caudifer*, J.-F. Cosson leg., 20/V/1992.

Remarks: New geographic record for French Guiana.

## Spelaeorhynchidae Oudmans, 1902

*Spelaeorhynchus* sp.

Examined material: French Guiana: Regina: Grotte Mathilde, 16 females, ex 4 *Pteronotus rubiginosus*, K. Pineau and M. Delaval legs., 2011.

**Discussion**

Previously Guerrero (1997) cited and registered 12 species of 6 genera from French Guiana, most of which originating from Piste de Saint-Elie (locality number 23 on Figure 1). Adding our results, the number of species of streblid bat flies increases to 58 (Table 1). But the number of streblid bat flies should be much higher, if we consider the 102 species of bat recorded in French Guiana (Lim & Tavares 2012). In neighboring countries that harbour a high richness of bats there has also been recorded higher richness of streblid bat flies. For example in Brazil there are 178 species of bats (Nogueira et al. 2014) and 83 species of streblid bat flies (Gracioli 2018), in Venezuela 166 bats (Gardner 2007) and 121 bat flies (Guerrero 1997) and in Colombia 180 species of bats (Gardner 2007) and 54 species of Streblidae (Marinkelle & Grose 1981; Dick et al. 2016), respectively.

We show the infracommunities found on six species of host with a sample size of at least 10 infested individuals. For *Carollia perspicillata*, we found nine infracommunities with one to three species. The most frequent infracommunities have two species. Few information is available for infracommunities composition and frequency on bats' ectoparasites. Santos et al. (2013), considering only infracommunities with two or more species, found three infracommunities with two and three species on *C. perspicillata* in Maranhão state, Brazil. There *T. joblingi* was the species that occurred in all infracommunities, whereas in French Guiana, *T. joblingi* and *T. anducei* were found in four out of nine infracommunities. These two *Trichobius* species were observed together in only one infracommunity.

Regarding the species of the mormoopid genus *Pteronotus*, we note that *P. rubiginosus* showed the most infracommunities with one species (16 out of 17 infracommunities); to the contrary, on the species *P. alitonus*, the majority of the infracommunities was composed of two species. On both species of mustached bats, the most frequent infracommunity was composed only by *Trichobius caecus*.

On *Desmodus rotundus* and *Sturnira giannae* were found the same number of infracommunities with same numbers of species. But on *D. rotundus*, the most frequent infracommunity had two species, whereas on *S. giannae*, infracommunities were composed by only one species (*Aspidoptera falcata*). Komeno & Linhares (1999) showed a negative correlation between *A. falcata* and *M. proxima* on *Sturnira lilium* and this could be an evidence of competition between these species. The

**Table 1.** Species of bat host with more than 10 parasitized specimens (number of specimens between brackets) and infracommunities of ectoparasitic arthropods frequencies and relative frequencies.

Host	Infracommunity	Frequency	Relative Frequency (%)
<i>Carollia perspicillata</i> (14)	<i>Speiseria ambigua</i>	1	7.1
	<i>Trichobius anducei</i>	2	14.3
	<i>S. ambigua</i> + <i>T. anducei</i>	2	14.3
	<i>S. ambigua</i> + <i>T. joblingi</i>	1	7.1
	<i>Strebla guajiro</i> + <i>T. anducei</i>	1	7.1
	<i>Strebla guajiro</i> + <i>T. joblingi</i>	2	14.3
	<i>S. ambigua</i> + <i>S. guajiro</i> + <i>T. anducei</i>	3	21.4
	<i>S. ambigua</i> + <i>S. guajiro</i> + <i>T. joblingi</i>	1	7.1
	<i>S. guajiro</i> + <i>T. anducei</i> + <i>T. joblingi</i>	1	7.1
<i>Desmodus roduntus</i> (11)	<i>Strebla wiedemanni</i>	3	27.3
	<i>Trichobius parasiticus</i>	2	18.2
	<i>S. wiedemanni</i> + <i>T. parasiticus</i>	6	54.5
<i>Phyllostomus hastatus</i> (13)	<i>Mastoptera minuta</i>	2	15.4
	<i>Strebla consocia</i>	4	30.8
	<i>Trichobius longipes</i>	1	7.7
	<i>S. consocia</i> + <i>T. longipes</i>	3	23.1
	<i>S. consocia</i> + <i>Trichobius</i> sp.	1	7.7
	<i>M. minuta</i> + <i>S. consocia</i> + <i>T. longipes</i>	2	15.4
<i>Pteronotus rubiginosus</i> (17)	<i>Ornithodoros</i> sp.	1	5.9
	<i>Spelaeorhynchus</i> sp.	4	23.5
	<i>Trichobius caecus</i>	10	58.8
	<i>Trichobius johnsonae</i>	1	5.9
	<i>Ornithodoros marinkellei</i> + <i>T. caecus</i>	1	5.9
<i>Pteronotus alitonus</i> (22)	<i>Trichobius caecus</i>	17	77.3
	<i>Antricola</i> sp. + <i>T. caecus</i>	1	4.5
	<i>Orbithodoros</i> sp. + <i>T. caecus</i>	1	4.5
	<i>T. caecus</i> + <i>Neotrichobius stenopterus</i>	1	4.5
	<i>T. caecus</i> + <i>Nycterophilina parnellii</i>	1	4.5
	<i>T. caecus</i> + <i>Trichobius sparsus</i>	1	4.5
<i>Sturnira giannae</i> (15)	<i>Aspidoptera falcata</i>	8	53.3
	<i>Megistopoda proxima</i>	5	33.3
	<i>A. falcata</i> + <i>M. proxima</i>	2	13.3

infracommunities characteristics (richness and composition) could be influenced by several factors such as host availability, competition including host available, geographic area, bat flies species composition, competition with other ectoparasites, or else roost quality (Wenzel et al. 1966). Furthermore, along the geographic range of a bat species, the various populations can harbor different infracommunities.

## Conclusion

Despite the new field surveys bringing 41 additional species to the Streblidae fauna of French Guiana, it appears that the number of streblid bat flies and other species of bat 's ectoparasites remains less than expected based onto comparisons with neighboring countries.

Clearly, new field work in various environments, savannahs, marshes, terra firme forests, mountains, of the whole of French Guiana are needed for a better understanding of the biodiversity, the distribution and the host relationship of arthropods ectoparasites of bats in this part of the Guianan Shield.

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## Author contributions

Gustavo Graciolli: Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Ricardo Guerrero: Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Francois Catzefflis: Substantial contribution in the concept and design of the study; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Biodiversity of aquatic environments in a peri-urban Atlantic Forest protected remnant: a checklist

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**Abstract:** We present a checklist for the aquatic biodiversity from two reservoirs within a PPBio (Biodiversity Research Program) site in a peri-urban forest fragment, the Dois Irmãos State Park (PEDI), in Pernambuco, Brazil. We obtained the data via extensive field collection and information from a specialized literature survey. We recorded 397 species in 156 families; the animal was the most abundant group (140 species) followed by fungi taxa (103), periphyton (69), aquatic macrophytes (44), and terrestrial plants in flooded areas (41). This review reflects different sample efforts toward selected groups and allows the definition of a long-term protocol for guiding new research based on the identified knowledge gaps revealed. Future ecological research should address the influence of the trophic state of the reservoirs, as well as the effects of competitive exclusion and predation on the long-term viability of the local diversity.

**Keywords:** PPBio, PEDI site, Aquatic macrophytes, Fungi, Aquatic Fauna.

## Biodiversidade de ambientes aquáticos em remanescente protegido da Mata Atlântica periurbana: um checklist

**Resumo:** Apresentamos uma lista da biodiversidade aquática de dois reservatórios em um sítio do PPBio (Programa de Pesquisa em Biodiversidade) em um fragmento de floresta peri-urbana, o Parque Estadual Dois Irmãos (PEDI), em Pernambuco, Brasil. Obtivemos os dados através de extensa coleta de campo e informações de pesquisa bibliográfica especializada. Registramos 397 espécies em 156 famílias; os animais foram o grupo mais abundante (140 espécies), seguido pelos fungos (103), perifíton (69), macrófitas aquáticas (44) e plantas terrestres em áreas alagadas (41). Esta revisão reflete diferentes esforços de amostra para grupos selecionados e permite a definição de um protocolo de longo prazo para orientar novas pesquisas com base nas lacunas de conhecimento identificadas. Pesquisas ecológicas futuras devem abordar a influência do estado trófico dos reservatórios, bem como os efeitos da exclusão competitiva e predação na viabilidade a longo prazo da diversidade local.

**Palavras-chave:** PPBio, site PEDI, macrófitas aquáticas, fungos, fauna aquática.

## Introduction

Continental aquatic ecosystems have been experiencing unprecedented disturbances in recent decades (Saunders et al. 2002) and are among the most threatened ecosystems in the world (Dudgeon et al. 2006) with 65% of aquatic habitats threatened (Vörösmarty et al. 2010). Changes in the flow regime of water bodies negatively impact the primary structure of river systems and wetlands (Bunn & Arthington 2002). Excessive withdrawal of water for agricultural, industrial (Szollosi-Nagy et al. 1998) and urban uses (Urban et al. 2006) may lead to water shortages in coming years (Szollosi-Nagy et al. 1998).

As a result, aquatic biodiversity is under pressure (Januchowski-Hartley et al. 2016), leading to both population decline and range

reduction of freshwater species (Dudgeon et al. 2006). Between 1990 and 2010, 123 freshwater animal species were considered to be extinct in North America alone (Ricciardi & Rasmussen 1999) while 10,000 to 20,000 freshwater species are estimated to be under risk of extinction or already extinct worldwide (Vörösmarty et al. 2010). Knowledge of biodiversity is insufficient for tropical regions where a high number of species are distributed, but species loss rates have still not been accurately calculated (Dudgeon et al. 2006).

In addition to local threats to biodiversity, there are also global impacts such as nitrogen deposition, changes in rainfall patterns, and global warming (Dudgeon et al. 2006). Long-term monitoring programs are essential for evaluating possible changes in the composition and abundance of biodiversity and in the main abiotic factors that affect

the dynamics of populations and habitats (Pezzini et al. 2012). With this goal in mind, the Biodiversity Research Program (PPBio) was created in 2004, aiming at intensifying studies on biodiversity in Brazil and integrating research findings and actions for various targets, including environmental management. A PPBio site was established in Pernambuco in 2013 at the Dois Irmãos State Park (PEDI) where there are two artificial water bodies, Prata and Dois Irmãos reservoirs.

An initial diagnosis of the existing biodiversity is required to subsidize and initiate long-term studies and monitoring, identify knowledge gaps, and guide management actions. Thus, the objective of this work is to present a checklist of the up-to-date aquatic biodiversity of the reservoirs in order to support conservation actions and a subsequent long-term research planning for this area.

## Material and Methods

### 1. Study site

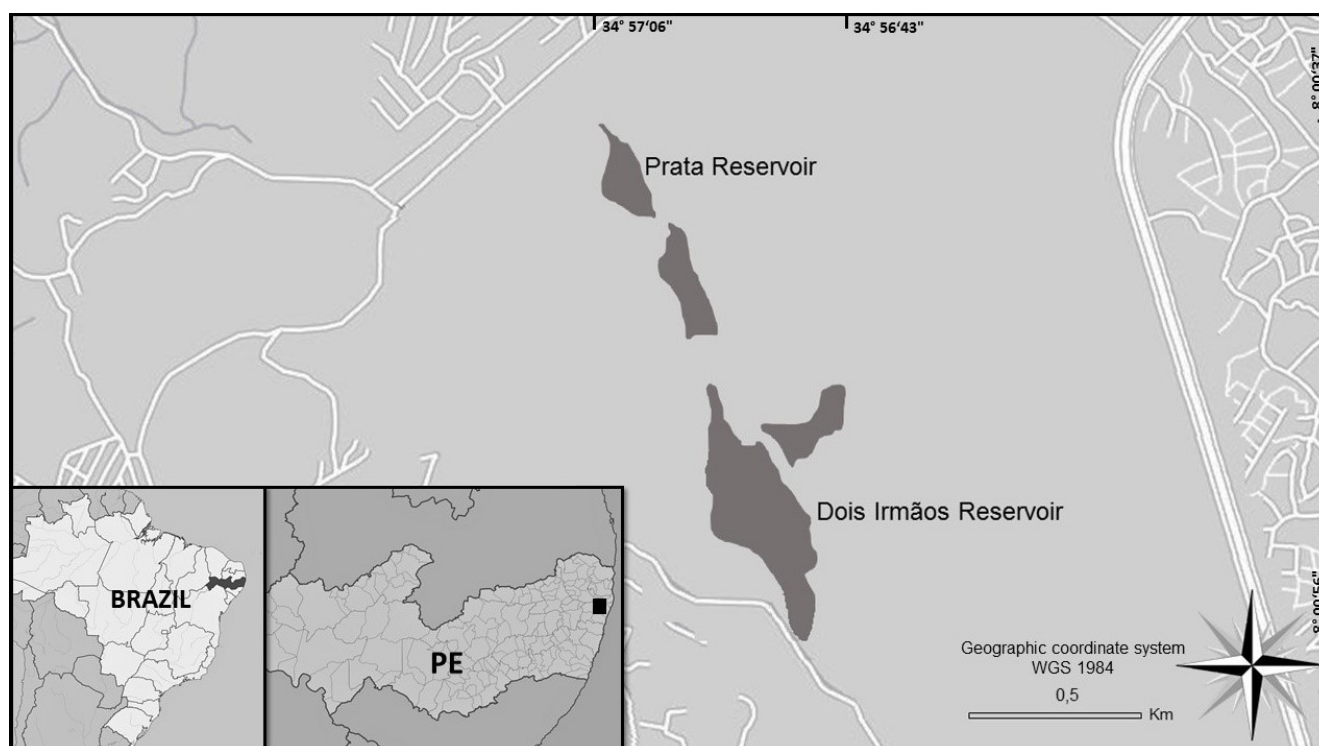
The PEDI comprises 1157.44 hectares of forest which 384.42 hectares are mature. It is located in the metropolitan region of Recife (8° 04' 03" S/34° 55' 00" W; Figure 1), Pernambuco, Northeast Brazil. The climate is tropical As' (Alvares et al. 2013) with a mean annual precipitation of 2460 mm and an average monthly temperature of 23°C (Silvestre & Carvalho 1998). The dry period occurs between September and February, and the rainy period is from March to August (Coutinho et al. 1998).

There are four reservoirs (Dentro, Dois Irmãos, Meio, and Prata) in the mature forest portion of the PEDI—all built in the first half of the nineteenth century to supply water to the city of Recife (Silvestre & Carvalho, 1998). Of these four water bodies, two occur within the

grid of the PPBio-PEDI monitoring site. The Dois Irmãos reservoir (8°00'42.6" S/34°56'48.2" W) is a eutrophic reservoir with 12 hectares and covered with floating macrophytes. It is situated within an intensely managed area of the Park, where there is a zoo, and is surrounded by roads, services, and building facilities for visitors and recreational uses (Silvestre & Carvalho 1998, Thomas & Middleton 2003). The Prata reservoir (8°00'16.2" S/34°57'00.8" W) covers 1.7 hectares and is oligotrophic. It is surrounded by the primitive or core zone of the park, where natural processes dominate, and the best forest values are preserved. Despite this, the reservoir is used for public water supply in the metropolitan region of Recife (Silvestre & Carvalho 1998).

### 2. Data collection

We obtained data through field collection and literature survey, which initially included scientific papers published in journals with an editorial board. As a starting point for the survey, we searched through the "Portal de Periódicos Capes/MEC", a free access Brazilian search tool for indexed journals and scientific literature provided by the Ministry of Education (MEC), containing more the 45,000 available journals, 130 reference bases, books and encyclopedias. We applied the following search terms in advanced search options, in combinations of taxonomic group (i) AND the target location (ii), in English and Portuguese: (i) biodiversity (fungi; aquatic macrophyte; ichthyofauna, fish; microalgae, periphyton, phytoplankton; herpetofauna; mastofauna; birds; zooplankton, Cladocera, Copepoda, Rotifera; fauna; flora); and (ii) Dois Irmãos State Park (PEDI), Prata Reservoir and Dois Irmãos Reservoir. Since this initial search returned only seven records, we expanded the search to encompass grey literature, including book chapters, simple summaries, expanded abstracts published in meetings and congresses, monographs, theses, and dissertations in addition to



**Figure 1.** Location of the studied areas in the Dois Irmãos State Park (PEDI), Recife, Pernambuco, Brazil.

technical reports available online. This search returned a total of 22 records from 1993 to 2019. The database WikiAves (2008) was also searched for bird occurrences, and personal observations from the group Birdwatchers of Pernambuco (OAP) were included.

We compiled the lists of species per taxonomic groups and habits: plants were categorized as macrophytes, terrestrial herbs, shrubs or periphyton. The identities were checked with specialized literature for each group. For plant species, we used the Flora do Brasil (2018); for birds, Piacentini et al. (2015); reptiles and amphibians, Pereira et al. (2013), Pereira et al. (2016) and Santos et al. (2017); for fish species, Fish Base (Froese & Pauly 2018); and for mammal species, Paglia et al. (2012).

Whenever possible, species occurrences were assigned to the Prata or Dois Irmãos reservoir. There were cases, however, when surveys were carried out in two, three, or all water bodies in the forest with no distinct sample points. In these cases, species lists were included if the Prata and Dois Irmãos reservoirs were cited as the study area. We also incorporated information on species sampled by the PPBio team through fieldwork or querying local informants.

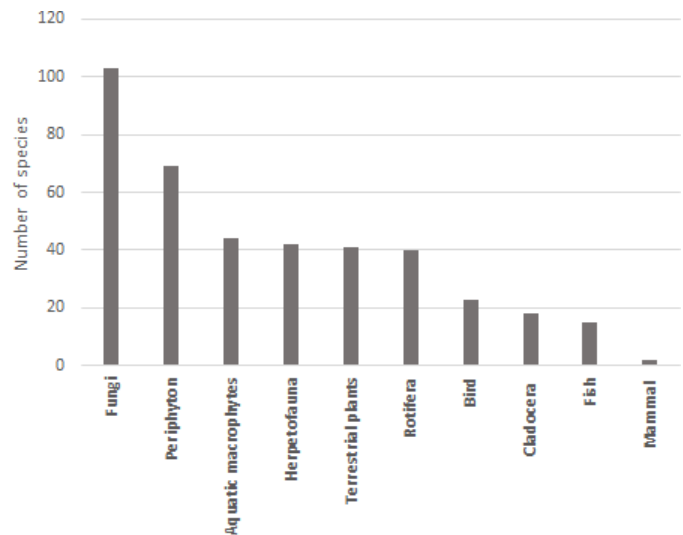
We assessed the conservation status of each species on the Red List of the International Union for Conservation of Nature (IUCN 2018) and on the Brazilian List of Threatened Species for Animals (MINISTÉRIO 2014) and plants (MARTINELLI 2013). The biogeographic origin of each species (native or non-native) was examined. Non-native species were also searched if the species had been classified as an invasive species according to the invasive non-native species database I3N Brasil (Instituto Hórus de Desenvolvimento e Conservação Ambiental 2018) as well as the Global Invasive Species Database (GISD) managed by the Invasive Species Specialist Group (ISSG) of the International Union for Conservation of Nature (IUCN 2018).

## Results

We registered 397 species distributed in 156 families and 82 orders for the aquatic biodiversity of the Dois Irmãos and Prata reservoirs (Figure 2). The animal was the most abundant group with 140 species divided into herpetofauna (42 species, 12 families, Table S1), Rotifera (40 species, 12 families, Table S1), birds (23 species, 09 families, Table S1), Cladocera (18 species, 04 families, Table S1), fish (15 species, 10 families, Table S1), and mammals (02 species, 02 families, Table S1). Fungi represented 28.93% of the local richness (103 species in 46 families; Table S2) followed by periphyton (19.38%, 69 species in 23 families; Table S3), aquatic macrophytes (44 species in 17 families, Table S4), terrestrial plants (41 species, 23 families, Table S4).

Although much smaller in size, the Prata Reservoir held a higher number of exclusive occurrences: 204 species against 94 in the Dois Irmãos; however, this reflects a bias towards the sampling of the less impacted and more conserved Prata reservoir. Research about Fungi and periphyton, for instance, are exclusive to the Prata water body although most of the studies in the PEDI were performed in both reservoirs (36.36%) or on the Dois Irmãos reservoir (36.36%). Fauna groups were sampled in both reservoirs and were less selective.

The two reservoirs have no plant (terrestrial, macrophytes, periphyton) or fungi species under risk of extinction according to the IUCN Red List. Of the total plants accessed, most are not evaluated, and 17 are categorized as of Least Concern (LC). On the Brazilian



**Figure 2.** The number of species in different taxonomic groups recorded for the aquatic biodiversity of Dois Irmãos and Prata reservoirs, Pernambuco, Brazil.

Red List for plant species, only six are included, but five are listed as LC and one as Data Deficient (DD). Among animal taxa, there are 62 species evaluated as LC on the IUCN Red List—mostly Anuran or Bird species. The neotropical river otter (*Lontra longicaudis*, Olfers, 1818) is near threatened (NT) on the IUCN list and Vulnerable in the Brazilian Atlantic Forest and the yellow-spotted river turtle (*Podocnemis unifilis*, Troschel, 1948) is considered vulnerable and near threatened (NT) for the same lists, respectively. In addition, three species were found to be endemic, although not restricted to the region or the biome: two plants (*Rolandra fruticosa* – endemic to North and Northeast Brazil – and *Homolepis aturensis* – endemic to North, Northeast and Southeast Brazil), and a mammal (*Hydrochoerus hydrochaeris* – endemic to South America east of the Andes).

Regarding the origin, there were six fungi, 13 plant taxa, and 15 animal species listed as non-native in the country or in the region. These include six potential invaders: the two aquatic herbs *Vallisneria americana* (Hydrocharitaceae) and *Nymphoides indica* (Menyanthaceae), and the terrestrial plants *Lantana camara* (Verbenaceae), *Urochloa* sp. (Poaceae), *Nephrolepis cordifolia* (Davalliaceae), and *Pteris vittata* (Pteridaceae). Eight species are native to Brazil, but not native to the studied region. These include the aquatic *Eichhornia crassipes* (Pontederiaceae), an Amazonian taxon, noted as one of the worst aquatic weeds in the world. Of the 21 non-native plants, 16 are recorded only in the Dois Irmãos Reservoir while five occur only in the Prata, including two potential invaders.

Six species of fungi (*Penicillium dipodomyis*, *Pseudopithomyces sacchari*, *Purpureocillium lilacinum*, *Talaromyces funiculosus*, *T. verruculosus* and *Trichoderma aureoviride*) and ten species of fish are non-native. Among fish species, nine come from other water basins in Brazil (*Astronotus ocellatus*, *Astyanax bimaculatus*, *Cichla ocellaris*, *Electrophorus electricus*, *Gymnotus omarorum*, *Hoplias malabaricus*, *Hypostomus affinis*, *Pterophyllum scalare*, *Serrasalmus rhombeus*) and one from Africa and Israel (*Oreochromis niloticus*). Three of them are potentially invasive species (*A. ocellatus*, *C. ocellaris* and *O. niloticus*) with predatory behaviour. Four non-native species of reptiles (*Trachemys dorbigni*, *T. scripta*, *Podocnemis expansa* and

*P. unifilis*) are recorded. The two *Trachemys* species, only found in the Dois Irmãos reservoir, are considered potential invaders with known ecological impacts in other areas due to higher competitive abilities and predator status.

## Discussion

The water bodies had a reduced group diversity of fauna and flora, probably due to the minor amount of scientific work performed and published in this area. These data reinforce the importance of programs such as the PPBio that focuses on the formation of a reliable database, grants for studies on the biological community structure, and biodiversity knowledge (Pezzini et al. 2012). Although not much is known about the continental aquatic biota in general, this survey revealed that the list of aquatic fungi in PEDI comprises more than 27% of the total number of known species in Brazil (Rocha 2003).

The aquatic flora recorded for PEDI represents approximately 22% of the aquatic biodiversity registered for the Northeastern Brazilian area (Moura-Junior et al. 2013). Aquatic flora has been a topic of recent studies due to its important link with water quality. The presence of the two invasive species and eutrophication indicators *Eichhornia crassipes* (Holm.) Solms and *Salvinia auriculata* Aubl. (Barreto et al. 2000) is a matter of serious concern as these species potentially have adverse effects not only on the aquatic biodiversity functioning, but can also prevent the use of the reservoir for leisure activities and offer favourable environments for disease vectors (Pedralli 2003).

The neotropical river otter is listed as “near threatened” but has a wide distribution. It can be found in rivers and streams (Kasper et al. 2004) and is a bioindicator for these environments because of its sensitivity to pollution. It is considered “vulnerable” in the Atlantic Forest biome due to extreme habitat degradation and susceptible to regional extinction within the next 50 years (Rodrigues et al. 2013). These animals are frequently hunted due to commercial interest on their skin or by fishers and fish farmers due to predation in fish farming tanks (Quadros 2009). Other activities can contribute to the decline of this species, such as fish introduction, deforestation, dredging, metals, and contamination by pesticides (Quadros 2009). Some of these impacts have already been observed in PEDI, especially the introduction of non-native fishes.

The introduction of non-native species to natural ecosystems represents one of the greatest threats to biological diversity on the planet (Simberloff 2003). In PEDI, we have recorded introduced species—mostly fish species, with particular attention to *Cichla ocellaris* (Bloch & Schneider, 1801) and *Oreochromis niloticus* (Linnaeus, 1758). The former species is popularly known as tucunaré, originated from the Amazon basin. This species has an aggressive behaviour and preys on other fish species (Pelicice & Agostinho 2009). The latter species, also known as *tilápia*, is omnivorous, with high reproductive efficiency, and is adaptable to environmental variability. It can increase water turbidity and lead to a considerable increase in phytoplankton biomass and abundance of cyanobacteria leading to adverse effects on water quality (Starling et al. 2002). In other reservoirs in Pernambuco (Lazzaro et al. 2003), there are already economic losses recorded due to increased costs for water treatment intended for public supply.

In general, most studies indicate that for conservation policy, including protection, restoration and management, all sizes of water bodies are important, as a set of small size ponds may contain higher conservation value than large ponds of the same size (Oertli et al. 2002, Scheffer et al. 2006), thus the diversity of aquatic habitats at PEDI is essential to biodiversity maintenance. Our findings suggest that studies on different biodiversity groups are still needed—particularly for wild fauna. Ecological questions on these studies should address the influence of the trophic state of the reservoirs in determining the composition of biodiversity. These efforts should also consider the effects of non-native species on the long-term viability of local diversity, particularly concerning competitive exclusion and predation.

## Supplementary material

The following online material is available for this article:

**Table S1** - List of species of fauna identified at Prata and Dois Irmãos reservoirs, Pernambuco, Brazil.

**Table S2** - List of species of fungi identified at Prata reservoir, Pernambuco, Brazil.

**Table S3** - List of species of periphyton identified at Prata and Dois Irmãos reservoirs, Pernambuco, Brazil.

**Table S4** - List of species of plants identified at Prata and Dois Irmãos reservoirs, Pernambuco, Brazil.

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## Authors' contributions

Karine Matos Magalhães: Formulated the idea. Correia performed the field work and identification. All authors performed data analysis. Contributed to critical revision adding intellectual content and writing of the manuscript.

Maria Cecília Santana de Lima: Contributed to critical revision adding intellectual content and writing of the manuscript.

Ednilza Maranhão Santos: Performed the field work and identification. All authors performed data analysis.

Jozélia Maria de Sousa Correia: Performed the field work and identification. All authors performed data analysis.

Ana Carolina Borges Lins e Silva: Contributed to critical revision adding intellectual content and writing of the manuscript.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

## Data availability

The data of this paper will follow the PPBio metadata politics.



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## Wild pig (*Sus scrofa* L.) occupancy patterns in the Brazilian Atlantic forest

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**Abstract:** Despite the great impacts of invasive wild pig (*Sus scrofa*) to natural ecosystems, habitat use by this species in the neotropics remains poorly studied. Here, we investigated the effects of local habitat and landscape covariates (vegetation types, running watercourses and roads) on occupancy patterns of wild pig in the Atlantic Forest of southern Brazil. We used single season occupancy modeling to estimate detection ( $p$ ) and occupancy ( $\psi$ ) probabilities, using 8-day camera-trap monitoring of 100 sampled sites. The cameras detected wild pig in 64 sites (naïve occupancy = 64 %). The four best models explained 72.7 % of the occupancy patterns, and the top model (with “water” variable) had a weight of 28.5 %. Even though none of the tested variables had high explanatory power of wild pig occupancy, the water variable had a negative effect trend ( $\beta = -1.124$ ; SE = 0.734), with 59 % of occupancy when water was present and 82 % when it was absent around the sampling sites. Vestiges of the presence of wild pig in different vegetation types revealed that they used plantations of *Pinus* sp., native forests, and corn and oat crops. The occupation pattern shows that wild pig are generalist at our study site at the Atlantic Forest being found everywhere, raising ecological and economic concerns about the high potential negative effects of its invasion.

**Keywords:** Occupancy modeling, feral pig, wild boar, landscape, Neotropics.

## Padrões de ocupação do javali (*Sus scrofa* L.) na Mata Atlântica brasileira

**Resumo:** Apesar dos grandes impactos da invasão do javali (*Sus scrofa*) nos ecossistemas naturais, o uso de habitats por esta espécie nos neotrópicos ainda permanece pouco estudado. Aqui, nós investigamos os efeitos do habitat local e de covariáveis da paisagem (tipos de vegetação, cursos d’água e estradas) sobre os padrões de ocupação do javali na Mata Atlântica do sul do Brasil. Utilizamos a modelagem de ocupação de estação única para estimar as probabilidades de detecção ( $p$ ) e de ocupação ( $\psi$ ) dos javalis, usando monitoramento de armadilha fotográfica por 8 dias em 100 locais. As câmeras detectaram javalis em 64 locais (ocupação ingênua = 64%). Os quatro melhores modelos explicaram 72,7% dos padrões de ocupação, e o melhor modelo (com variável “água”) teve um peso de 28,5%. Embora nenhuma das variáveis testadas apresentaram alto poder explicativo na ocupação do javali, a variável água foi a que contribuiu com uma tendência de efeito negativo ( $\beta = -1,124$ ; SE = 0,734), com 59% de ocupação quando a água estava presente e 82% quando estava ausente nos pontos de amostragem. Vestígios da presença de javali em diferentes tipos de vegetação revelaram que eles utilizaram plantações de *Pinus* sp., florestas nativas e culturas de milho e aveia. O padrão de ocupação mostra que o javali é extremamente generalista em nosso local de estudo na Mata Atlântica, sendo encontrado em todos os lugares, o que levanta preocupações ecológicas e econômicas sobre os potenciais efeitos negativos de sua invasão.

**Palavras-chave:** Modelagem de ocupação, porcos asselvajados, javalis, paisagem, Neotrópicos.

## Introduction

Non-native and invasive species are found in almost all ecosystems worldwide, a number that has increased markedly in the last two decades (Blackburn et al. 2011). These accidentally or intentionally introduced species are causing several damages to native species (Vitule et al. 2012), changes in community structure and in dynamics of natural ecosystems (Martin et al. 2009) and even reduction in biological diversity (Chapin III et al. 2000). Furthermore, anthropogenic disturbances of natural environments favor the success of invasive species (Gurevitch & Padilla 2004), and many changes promoted by invasive species are gradual and unnoticed (Simberloff et al. 2013).

Wild pig (*Sus scrofa* L.) are those pig invasive/non-native/introduced (Keiter et al. 2016, Melletti & Meijaard 2017), currently considered one of the 100 most invasive species of the world (Lowe et al. 2000, IUCN 2019). After humans, wild pig comprises the large-bodied size mammal with the broadest distribution in the world (Massei & Genov 2004; Barrios-García & Ballari 2012). Since the wild pig had a wide native geographical distribution, it can be considered pre-adapted to a large array of environmental conditions (Baskin & Danell 2003). Wild pig can move long distances in one single day in search for food (Leaper et al. 1999) and are able to cause major impacts on native plants and animals, on crop plantations and domestic animals, and on ecological processes (Oliver & Brisbin 1993; Hadjisterkotis 2004; Massei & Genov 2004; Barrios-García & Ballari 2012; Myrphy et al. 2014). However, resource abundance and distribution can have strong impacts on population dynamics and survival rates of wild pig (Ostfeld & Keesing 2000). Wild pig population growth and abundance can be determined by presence or absence of food resources (Jedrzejewska et al. 1997; Honda 2009), landscape structure (Acevedo et al. 2006) and climatic factors (Honda 2009).

Models have been used recently to predict the distribution of wild pig and to understand their occupancy patterns in native and non-native areas (Bosch et al. 2012, Bosch et al. 2014, Acevedo et al. 2014, Gantchoff & Belant 2015, McClure et al. 2015, Forsyth et al. 2016, Sales et al. 2017, Pittiglio et al. 2018). Such studies were based on models using camera traps and indirectly on signs and presence/absence data of wild pig associated with environmental (vegetation type and topography) and climatic variables, besides anthropogenic effects. These analyses revealed that areas occupied or with a potential to be occupied by wild pig are those where food and shelter are most abundant. Also, niche shifts in non-native areas might be explained mainly by the existence of unoccupied areas where the climate is similar to its native areas (Sales et al. 2017).

Native to Eurasia and north of Africa, the wild pig were introduced in South America at the beginning of the 20th century, invading Brazil by late 1980s from Uruguay into the southern part of the State of Rio Grande do Sul (Deberdt & Scherer 2007). In the wild, it interbred with the domestic pig (*Sus scrofa domesticus* Erxleben) resulting in fertile hybrids (Grossi et al. 2006), called “wild pig” (Keiter et al. 2016, Melletti & Meijaard 2017). Wild pig are one of the several invasive species present in the Brazilian Atlantic Forest (Deberdt & Scherer 2007, Hegel & Marini 2013, Pedrosa et al. 2015). These native forests offer resources such as water, food, and humid areas, and have nowadays low density of large predators, like jaguars or pumas (Machado et al. 2008), which have the potential to prey upon large ungulates (Hegel & Marini 2018). Wild pig impact on the Atlantic Forest is poorly known, but at

a forest fragment in south Brazil, the impact on the native vegetation inside a reserve was evaluated and showed intense herbivory, rooting and soil overturning (Hegel & Marini 2013). Also worryingly, wild pig consumes and destroy the seeds and cones (Deberdt & Scherer 2007, Hegel & Marini 2013) of the critically endangered Parana Pine (*Araucaria angustifolia*) (Thomas 2013), which has already lost 97% of its geographical distribution (Gantzel 1982, Guerra et al. 2002). The increasing abundance and economic damage, such as partial loss of crop plantations, caused by wild pig in Brazil have promoted a series of laws by Brazilian governmental agencies allowing wild pig hunting (see IBAMA nº 03 of 31 January 2013, reissued in IBAMA nº 12 of 25 March 2019). However, there are no current estimates of wild pig densities and expansion rates at the Atlantic Forest. Finally, it is worthy to mention that the Atlantic Forest has lost approximately 90% of its original distribution (Ribeiro et al. 2009), is a world hotspot (Myers et al. 2010), and is still being deforested in the last decades (SOS Mata Atlântica 2014).

Thus, considering the potential threat of wild pig to native species and habitats, the high endangerment of the Atlantic Forest, and the scarcity of studies about the recent invasion of wild pig in the region, we tested the hypothesis that wild pig occupation patterns in the Atlantic Forest are related to vegetation types and landscape variables, resembling the patterns found in other native and non-native areas. To accomplish that, we estimated wild pig detection ( $p$ ) and occupancy ( $\psi$ ) probabilities in the Atlantic Forest. The findings provide unique information about how wild pig occupy an altered Atlantic Forest landscape, with potential applications to conservation and management plans.

## Material and Methods

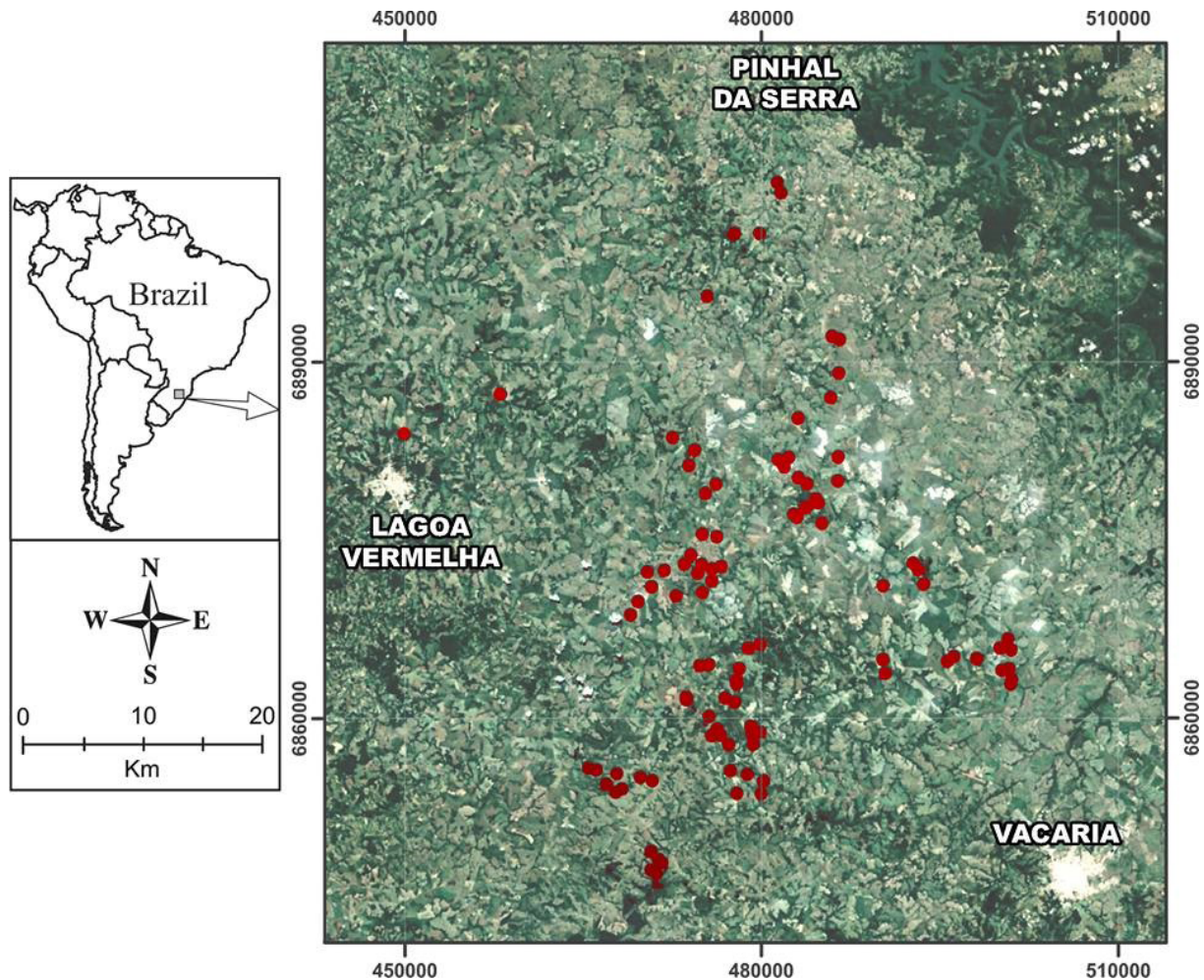
### 1. Study site

We conducted this study at the ‘Campos de Cima da Serra’ region, southern Brazil (28°13’54.2” S and 51°10’14.9” W), at the southern part of the Atlantic Forest. We studied a region up to 50 km centered at the reserve ‘Estação Ecológica de Aracuri-Esmeralda’ (EEAE, with 275 ha), municipality of Muitos Capões, Rio Grande do Sul (Figure 1, Supplementary Table S1). Today, the vegetation of the region is composed of patches of disturbed Mixed Ombrophilous Forest, a type of Atlantic forest of southern Brazil with *Araucaria angustifolia* as the most emblematic tree, in a matrix of native grasslands, wetlands, secondary vegetation, ‘vassorais’ (*Baccharis* dominated vegetation) and crop plantations (Brasil 2008). The study region is located around 700-950 m elevation with mean annual rainfall ranging from 1,700 to 2,200 mm well distributed along the year and mean annual temperature ranging from 14° and 16°C (Brasil 2008), with four well defined seasons.

### 2. Sampling sites and variables

We conducted a 10-day sampling design preliminary study at the end of July 2015, using one camera-trap in each of 16 sites. Then, we used this result to simulate in program MARK (White & Burnham 1999) the number of days and sampling units necessary to estimate our parameters of interest (i.e., occupancy ( $\psi$ ) and detection ( $p$ ) probabilities) during one single season. Based on the results of this simulation we designed our study to register wild pig with camera traps during 8 days at 100





**Figure 1.** Wild pig (*Sus scrofa*) occupancy study region showing the sampling sites (red dots) in the Atlantic Forest, state of Rio Grande do Sul, southern Brazil.

sampling sites distributed up to 50 km around EEAE, at least 1 km apart from each other (a distance also used by Gantchoff & Belant 2015 and Forsyth et al. 2016) (Figure 1). To improve detectability, all 100 sampling sites were selected based on previous vestiges of the presence of wild pig at each site, such as hair, feces, footprints, overturned soil, trunk damage, and sightings. We set up 16 cameras during six consecutive 8-day periods, and four cameras during the 8-day period from September 8 to November 3, 2015 (adding to 800 camera/days). This period of sampling was used for logistical reasons spanning the end of winter and the beginning of spring.

At each site, the percentage of four vegetation types (native forests, native grasslands, wetlands, and crop plantations) was estimated around 500 m from the point that each camera was mounted using Google Earth images treated with ArcGIS (Esri 2011). Overall, the native forest was the most common vegetation type (56.5% of the area), followed by crop plantations (25.7%), native grasslands (9.9%), and wetlands (7.8%). To run the single-season occupancy modeling analysis, we used six variables: two landscape variables (distance from roads - categorical, up to 50 m and further than 50 m, and distance from running watercourses - categorical, up to 30 m and further than 30 m) and the four vegetation types, each one as a variable (native forests, native grasslands, wetlands, and crop plantations (encompassing oat,

corn, soybean, wheat, apple and grape orchards). We excluded *Pinus* sp. plantations from the analyses since they represented only a very small portion (0.12%) of total vegetation.

### 3. Statistical analyses

We used a single-season occupancy modeling approach to estimate occupancy ( $\psi$ ) and detection ( $p$ ) probabilities of wild pig (Mackenzie et al. 2002). The assumptions of the method are that (1) within the sampling period the occupancy status of the species was closed (no colonization or extinction occur during the sampling) (Mackenzie et al. 2006); (2) the probability of detecting the species was independent among sampled sites; and (3) the species was not falsely detected. We considered occupancy as a measure of habitat use, because home ranges of wild pig may exceed the size of our sampling unit (0.7 – 6 km<sup>2</sup>) (Baber & Coblenz 1986, Ilse & Hellgren 1995, Gabor et al. 1999). The assumption that sites are close to changes in occupancy during the sampling occasions may be relaxed if changes in the occupancy status of sites are random. In this case, occupancy should be interpreted as 'use' and movement throughout the sampled sites (Mackenzie et al. 2004; Mackenzie & Royle 2005). The detection probability incorporated to the models accounts for imperfect detection, reducing bias in parameters estimation (Mackenzie et al. 2006).

Our modeling process followed three steps in program MARK (White & Burnham 1999). First, we built a global additive model with occupancy varying by the presence of running watercourses (water), roads (road), amount of forest (forest), grassland (grass), wetlands (wetlands), and crop plantations (crops). In this global model, we maintained detection constant because the temporal version did not estimate all parameters, and because we did not have specific hypotheses on detection variation. Next, we built a set of 64 models with all possible combinations. This resulted in a balanced model set to estimate the importance (cumulative weights, hereafter  $w_i^+$ ) of each landscape variable, following the recommendation of Burnham & Anderson (2002). Finally, to have a reliable set of candidate models, we excluded from the analysis models with non-informative parameters, following Arnold (2010). Although vegetation variables appeared as non-informative parameters, we decided to maintain models with these variables based on our field observations and the importance of vegetation to the presence and distribution of wild pig. We conducted goodness-of-fit analysis with program PRESENCE (Hines 2006) to evaluate the global model fit and to estimate the variance inflation factor (c-hat), which we used to adjust the Akaike's Information Criterion for small sample size (QAIC<sub>c</sub>, see Mackenzie & Bailey 2004). We used QAIC<sub>c</sub> to rank competing models and we considered models with  $\Delta\text{QAIC}_c$  values  $< 2$  equally supported and used them to make inferences (Burnham & Anderson 2002). We considered the QAIC<sub>c</sub>  $w_i$  (hereafter  $w_i$ ) as the relative weight of support of each model and we model-averaged occupancy ( $\psi_i$ ) across the final set of models (Burnham & Anderson 2002, Doherty et al. 2012). Finally, we did not use null  $p$ -values to clarify uncertainties in the modeling to avoid mixing the paradigms "hierarchical model selection" and "null hypothesis testing", following Wasserstein et al. (2019).

## Results

We recorded wild pig in 64 of the 100 sampled sites in the Atlantic Forest. We built all possible combinations of additive models, resulting in a set of 64 models (Supplementary Table S2). In these models, the variable "water" had the highest cumulative weight ( $w_i^+ = 0.60$ ), followed by "crops" ( $w_i^+ = 0.34$ ), "grass" ( $w_i^+ = 0.31$ ), "forest" ( $w_i^+ = 0.29$ ), "wetlands" ( $w_i^+ = 0.27$ ), and "roads" ( $w_i^+ = 0.25$ ). After excluding from the analysis models with non-informative parameters, only 11 models remained with the most important variables affecting detection and occupancy probabilities of wild pig. From these, the four top-ranked models accounted for 72.7 % of the total model weight and  $\Delta\text{QAIC}_c < 2$  (Table 1). We considered them to explain variation in occupancy probability of wild pig in the Atlantic Forest. The top model ( $w_i = 0.285$ ) had "water" as a covariate on occupancy, in the second model the occupancy was "constant" ( $w_i = 0.208$ ), and in the third and fourth models, the occupancy was explained by "crops" and "grass" ( $w_i$  of 0.123 and 0.111), respectively (Table 1).

No single variable highly explained wild pig occupancy in the Atlantic Forest (Tables 1 and 2). The two best models answered for 49.3% of the weight of all models ("water" – 28.5% and "constant" – 20.8%). However, the top model showed a slight tendency towards a negative effect of "water" (running watercourses), with a decrease of wild pig occupation at sampling sites close to running watercourses ( $\beta = -1.12$ ; SE = 0.58). When the "water" variable was analyzed alone,

the results indicated that in the presence of running watercourses the percentage of occupation by wild pig was lower (59%) than in the absence of running watercourses (82%). The second model, "constant", reinforces the generalist habit of wild pig indicating a random pattern of occupation of the landscape. The next models with the variables "crops", "grass", "forest", and "wetlands" had inconclusive tendencies with weak explanatory weights (between 7.2 and 12.3%) (Table 1) and confidence intervals of the  $\beta$  parameters overlapping zero (Table 2). Furthermore, "roads" did not contribute to explaining the occupation of wild pig ( $\beta < 0.001$ ; SE = 0.51). Wild pig were detected equally in areas with (N = 32) and without (N = 32) roads near the sampling sites with cameras.

## Discussion

Our results showed uncertainties about the influencing variables of wild pig occupation on South Atlantic Forest, indicating a random pattern of occupation of the landscape that reinforces the generalist habit of the species (Mayer & Brisbin 2009; West et al. 2009). Nevertheless, we detected a slight tendency for a negative effect of running watercourses and wetlands on wild pig occupation. This is opposite to expected since McClure et al. (2015), in a macro-spatial study in the USA, found that both distance to water and landscape heterogeneity were important in their models, with localities far away from the water having lower occurrence of wild pig. This lower occupancy near running watercourses could represent a threat to young pig because of a higher danger of drowning when crossing deeper watercourses, because of hypothermia due to the low-fat content in the first months of live (Rosell et al. 2001). Also, wild pig tends to avoid areas near watercourses probably because of lower protection from predators (Kurz & Marchinton 1972; Massei et al. 1997), which can prey upon juveniles and piglets (Hegel & Marini 2018). However, wetlands are known to be used as a shelter, for breeding, feeding and mainly regulation of body temperature by mud baths (Mendina Filho et al. 2015), which can also help clean out ectoparasites (West et al. 2009). Other studies have shown that the only environmental condition that can effectively avoid the presence of wild pig in an area is the lack of superficial water (Mayer & Brisbin 2009; Beasley et al. 2014). Also, wild pig prefer to construct nests in areas with dense cover and water nearby (Fernández-Llario 2004). One explanation for this contradiction between our results and previous studies is probably related to the 45% above average rainfall at our study site in 2015 (INPE 2016), related to an "El Niño" effect in the southern Neotropical region. Thus, the excess of rain, and of humid areas, might have changed landscape use by wild pig during our sampling, allowing them to occur in areas independently of local water availability.

The single-season occupancy modeling analysis indicated that there is no specific preference for any vegetation type by wild pig. Thus, the occupation patterns of wild pig showed that it is a habitat-generalist at the Atlantic Forest, similar to other native and introduced regions of the world, being found at several vegetation types, such as native and planted forests, grasslands, humid areas, and plantations (Spitz 1986 apud Oliver & Leus 2008; Mayer et al. 2000; Wilson 2004). Accordingly, we also observed vestiges of wild pig in different vegetation types such as *Pinus* sp. plantations, and corn and oat crops. Similarly, in the USA, wild pig preferred *Pinus* sp. trees (Graves 1984), indicating that this species might be beneficial for wild pig at introduced localities. Wild

**Table 1.** Single-season occupancy modeling: 11 models generated to explain detection and occupancy of wild pig in the south Atlantic Forest, where  $p$  = detection probability, and  $\psi$  = occupancy probability. The table presents the values of QAICc,  $\Delta$ QAICc, AICc weights ( $w_i$ ) of each model and the number of parameters (K).

Model	QAICc	$\Delta$ QAICc	Weights ( $w_i$ )	K
$\Psi(\text{water}) p(.)$	470.802	0	0.285	3
$\Psi(.) p(.)$	471.431	0.629	0.208	2
$\Psi(\text{crops}) p(.)$	472.486	1.684	0.123	3
$\Psi(\text{grass}) p(.)$	472.683	1.880	0.111	3
$\Psi(\text{forest}) p(.)$	473.055	2.252	0.092	3
$\Psi(\text{wetlands}) p(.)$	473.205	2.403	0.085	3
$\Psi(\text{road}) p(.)$	473.558	2.755	0.072	3
$\Psi(\text{water} + \text{wetlands} + \text{grass} + \text{forest} + \text{crops}) p(.)$	477.495	6.692	0.010	7
$\Psi(\text{wetlands} + \text{grass} + \text{forest} + \text{crops}) p(.)$	478.567	7.765	0.005	6
Global $\Psi(\text{road} + \text{water} + \text{wetlands} + \text{grass} + \text{forest} + \text{crops}) p(.)$	479.844	9.041	0.003	8
$\Psi(\text{road} + \text{wetlands} + \text{grass} + \text{forest} + \text{crops}) p(.)$	480.881	10.078	0.001	7

**Table 2.** Single-season occupancy modeling: seven first models with one variable to explain beta values each parameter, where  $\beta$  = value of the variable beta parameter, SE = standard error, CI = lower and upper limits of 95% Confidence Interval.

Model	$\beta$	SE	CI
$\Psi(\text{water}) p(.)$	-1.1219	0.5895	-2.2773 0.0335
$\Psi(\text{crops}) p(.)$	-2.6349	2.1169	-6.7840 1.5142
$\Psi(\text{grass}) p(.)$	2.6147	2.3915	-2.0727 7.3021
$\Psi(\text{forest}) p(.)$	3.8165	4.8289	-5.6482 13.2812
$\Psi(\text{wetlands}) p(.)$	-3.5177	4.6996	-12.7290 5.6935
$\Psi(\text{roads}) p(.)$	0.26 E-5	0.5166	-1.0125 1.0125

pig are attracted to areas with these trees, at least in part because of their behavior of rubbing their bodies against trees to remove parasites after mud baths (Campbell & Long 2009). The preference of wild pig for conifers might also be related to antimicrobial activity against bacteria and fungi, properties of the resin that helps heal wounds (Sipponen et al. 2012). A native conifer, the Parana pine *Araucaria angustifolia*, which occurs at southern Atlantic forests, is also used by wild pig for rubbing (C. Hegel, pers. obs.), and as a food source (Deberdt & Scherer 2007, Hegel & Marini 2013). Wild pig presented a seasonal variation in occupation in coniferous forests of New Zealand, being present in more places in the summer than in the winter (Forsyth et al. 2016).

In turn, because of wild pig broad diet, food available in the forests is not expected to be a limiting factor (Ballari & Barrios-García 2014). We observed vestiges of the presence of the wild pig especially in corn and oat plantations, but not in other cultures and plantations (soybean, wheat, and apple and grape orchards), though they were poorly sampled. Thus, proper year-round use of habitats is necessary to evaluate seasonal and spatial use of the landscape. Wild pig are known to consume large amounts of several crops (oat, corn, sugar cane, wheat, sorghum, barley, and oilseeds) as well as tree saplings in the USA (Mayer et al. 2000). In Spain, wild pig often occurred in large forest fragments surrounded by crops, and adjacent to other large forests close to mountains or riparian forests (Virgós 2002). Also, Caley (1993) found that wild pig consumes not only standing crops but also rooting crop residues after harvest, evidencing its food flexibility. Since wild pig have a generalist diet, the potential impact on specific crops should be evaluated throughout the year and at different stages of each crop.

Areas of grasslands also had no effect on the detection or occupancy of wild pig at out study site. Native grasslands at our study site might have been used, like roads, only for movement among adjacent vegetation types. However, wild pig caused vegetation disturbance in pasturelands and plantations adjacent to forests in southern England (Wilson 2004). Roads were used by wild pig to move among preferred habitats, such as humid areas and crops (Mayer & Brisbin 2009; Beasley et al. 2014). In Argentinian Patagonia, wild pig were present only in humid lands, and occupancy was lower closer to settlements but higher closer to roads (Gantchoff & Belant 2015).

Although our study has seasonal limitations of sampling and the possible influence of a climatic phenomenon that increases the precipitation in the south of Brazil, our results are similar to other regions either where wild pig are native or introduced. Here, wild pig showed an overall broad and unselective use of the landscape occupying most vegetation types, with a poor relationship with running waterbodies and roads. The tendency of higher occupation of some vegetation types demonstrates only weak preferences, such as for forests, and some crops, such as corn and oat. This broad occupation pattern stresses the major potential of invasion of wild pig even at subtropical regions like the southern Atlantic Forest. The rich and highly fragmented and altered Atlantic Forest, a world hotspot, seems prone to be invaded by wild pig since wild pig can benefit from and occupy the current mosaic of vegetation types in the region. The fact that wild pig can cause economic and environmental impacts is worrisome, requiring urgent attention by governmental authorities to manage and control wild pig, especially in pine forests and other forest types in the Atlantic Forest domain,



before their populations increase even further. We also recommend the financial incentive to year-round studies of wild pig occupation patterns not only in the Atlantic Forest but also in other Brazilian environments and their transition areas.

## Supplementary material

The following online material is available for this article:

Table S1 - Geographical coordinates and name of the localities of the 100 sampling sites.

Table S2 - Single-season occupancy modeling: 64 models generated to explain detection and occupancy of wild pig in the south Atlantic Forest.

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## Author Contributions

Carla Grasielle Zanin Hegel - Substantial contribution in the concept and design of the study; Contribution to data collection; Contribution to data analysis and interpretation; Contribution to manuscript preparation.

Luane Reis dos Santos - Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation; Contribution to manuscript preparation.

Mauro Pichorim - Contribution to data analysis and interpretation; Contribution to manuscript preparation, Contribution to critical revision, adding intellectual content.

Miguel Ângelo Marini - Substantial contribution in the concept and design of the study; Contribution to data analysis and interpretation, Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Flora of Pernambuco, Brazil: Tabebuia alliance and tribe Jacarandae (Bignoniaceae)

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**Abstract:** Bignoniaceae is a Pantropical plant family that includes 82 genera and 830 species of trees, lianas, and shrubs. The Tabebuia alliance (14 genera and 147 species) and tribe Jacarandae (2 genera and 50 species) are both Neotropical and represent the largest clades of trees and shrubs in the family. Here, we present a taxonomic treatment for these two clades for the state of Pernambuco, Brazil. Overall, we documented 23 species distributed in seven genera, i.e., *Cybistax* Mart. ex Meisn., *Godmania* Hemsl., *Jacaranda* Juss., *Handroanthus* Mattos, *Sparattosperma* Mart. ex Meisner, *Tabebuia* Gomes ex DC., and *Zeyheria* Mart. Six taxa are new records for the state, i.e., *Cybistax antisiphilitica* (Mart.) Mart., *Handroanthus capitatus* (Bureau & K. Schum) Mattos, *Handroanthus umbellatus* (Sond.) Mattos, *Jacaranda cuspidifolia* Mart., *Sparattosperma catingae* A.H. Gentry, and *Tabebuia stenocalyx* Sprague & Stapf. Furthermore, *S. catingae*, previously thought to be endemic to the Caatinga of Bahia, was documented for the first time in the Atlantic Forest domain. We present identification keys and taxonomic descriptions for all genera and species, as well as provide illustrations and information on the geographic distribution, habitat, and phenology for all species.

**Keywords:** Botanical inventories; Brazilian flora; Caatinga; Atlantic Forest.

## Flora de Pernambuco, Brasil: Aliança Tabebuia e tribo Jacarandae (Bignoniaceae)

**Resumo:** Bignoniaceae é uma família de plantas pantropicais que inclui 82 gêneros e 830 espécies de árvores, lianas e arbustos. A aliança Tabebuia (14 gêneros e 147 espécies) e tribo Jacarandae (2 gêneros e 50 espécies) são ambas neotropicais e representam os maiores clados de árvores e arbustos na família. Aqui, apresentamos um tratamento taxonômico para estes dois clados para o estado de Pernambuco, Brasil. No geral, nós documentamos 23 espécies distribuídas em 7 gêneros, i.e., *Cybistax* Mart. ex Meisn., *Godmania* Hemsl., *Jacaranda* Juss., *Handroanthus* Mattos, *Sparattosperma* Mart. ex Meisner, *Tabebuia* Gomes ex DC., e *Zeyheria* Mart. Seis táxons são novos registros para o estado, i.e., *Cybistax antisiphilitica* (Mart.) Mart., *Handroanthus capitatus* (Bureau & K. Schum) Mattos, *Handroanthus umbellatus* (Sond.) Mattos, *Jacaranda cuspidifolia* Mart., *Sparattosperma catingae* A.H. Gentry, e *Tabebuia stenocalyx* Sprague & Stapf. Além disso, *S. catingae*, que anteriormente se pensava ser endêmica da Caatinga da Bahia, foi documentada pela primeira vez no domínio da Mata Atlântica. Nós apresentamos chaves de identificação e descrições taxonômicas para todos os gêneros e espécies, assim como fornecemos ilustrações e informações sobre a distribuição geográfica, habitat e fenologia para todas as espécies.

**Palavras-chave:** Inventários botânicos; Flora brasileira; Caatinga; Mata Atlântica.

## Introduction

The Bignoniaceae comprises about 830 species distributed in 82 genera (Lohmann & Ulloa Ulloa 2006 continuously updated). This plant family is Pantropical, but predominantly Neotropical, where about 80% of the species occur, with only a few species occurring in the Temperate zones (Lohmann 2004). The Bignoniaceae is centered in Brazil, where 33 genera and 413 species are found, of which two genera (*Neojobertia* Baill. and *Paratecoma* Kuhl.) and 199 species are endemic (Lohmann 2010). The family includes lianas, trees, and shrubs that generally occur in humid or dry forests, sometimes in open dry areas, or rocky outcrops (Lohmann 2004).

Members of the family are characterized by the following synapomorphies: a lack of endosperm in the mature seeds, and by two placental ridges, each bearing one to several rows of ovules (Spangler & Olmstead 1999). Species of the Bignoniaceae are also recognized by the woody habitat, opposite and compound leaves, showy hermaphrodite and gamopetalous flowers, with four didynamous stamens and one staminode (reduced or elongated), and dehiscent fruits with winged seeds (Lohmann 2004).

Gentry (1980) divided the family into eight tribes based on habit, distribution and fruit dehiscence: Bignonieae, Coleae, Crescentieae, Eccremocarpeae, Oroxyleae, Schlegelieae, Tecomeae, and Tourrettieae.

Subsequent phylogenetic studies (Spangler & Olmstead 1999, Olmstead et al. 2009) indicated that Tecmeae is not monophyletic and that the supra-generic classification needed to be reformulated. Six monophyletic tribes (i.e., Bignoniaceae, Catalpeae, Jacarandeae, Oroxyleae, Tecmeae, and Tourrettiae) and two informally named clades (i.e., Tabebuia alliance and Paleotropical clade) are currently recognized (Olmstead et al. 2009).

The Tabebuia alliance is restricted to the Neotropics and includes 14 genera and 147 species, representing the second largest clade of the family (Olmstead et al. 2009). The clade includes genera composed predominantly of large trees such as *Handroanthus* Mattos and *Tabebuia* Gomes ex DC., as well as treelets and shrubs (Olmstead et al. 2009). Taxa included in this clade are characterized by the palmate-compound leaves (synapomorphy of the clade), flowers with spathaceous to cupular calyces, and infundibuliform corollas with varied colors, and linear to ovate ovaries (Grose & Olmstead 2007, Olmstead et al. 2009). In Brazil, this group is represented by seven genera and 44 species (*Handroanthus*, *Tabebuia*, *Zeyheria* Mart., *Godmania* Hemsl., *Paratecoma* Kuhlman, *Cybistax* Mart ex Meisn. and *Sparattosperma* Mart. ex Meisner) most of which are broadly distributed throughout the national territory, except from *Paratecoma* and *Godmania* that have more restricted distributions (Lohmann 2010).

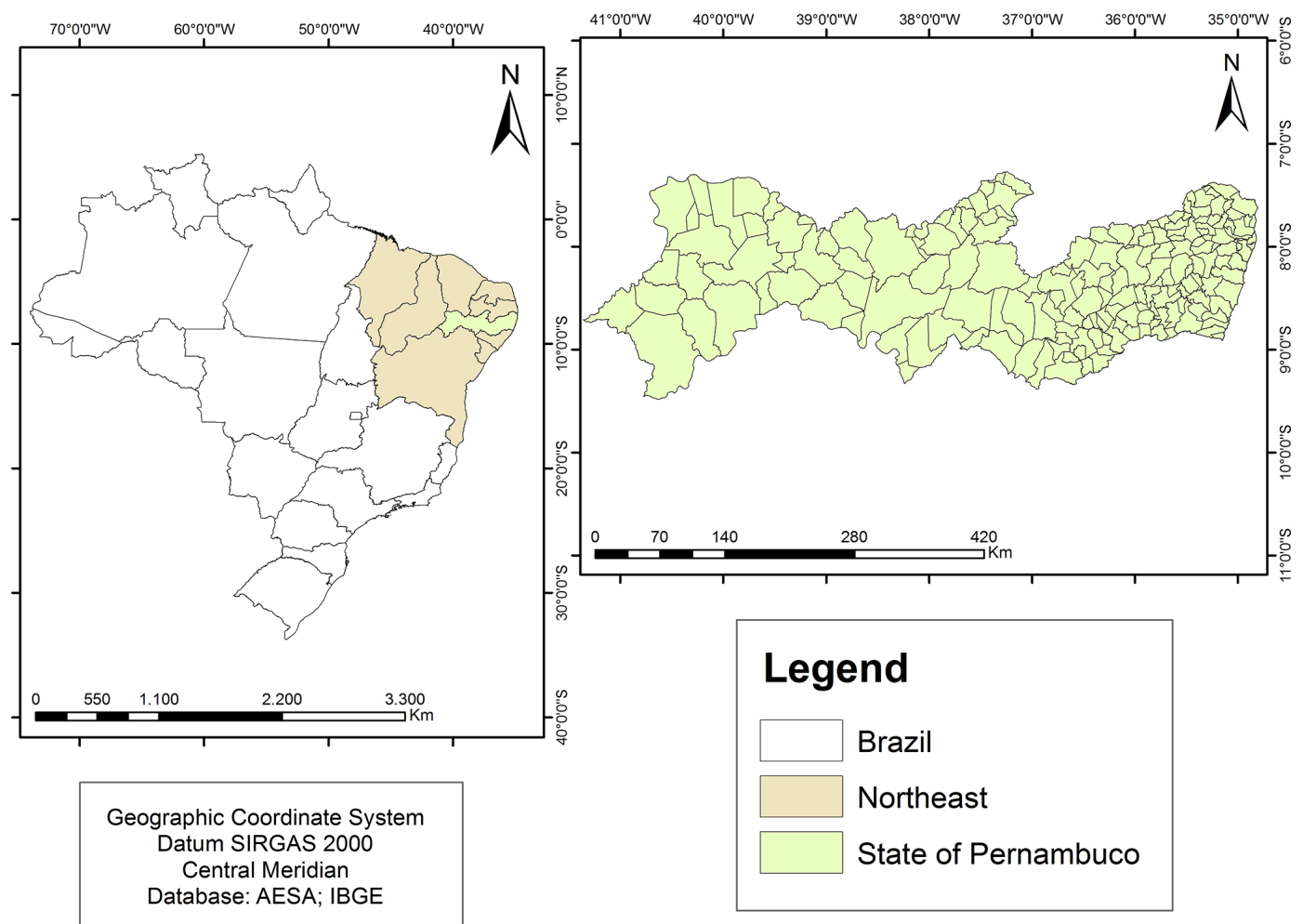
Tribe Jacarandeae includes two genera (*Jacaranda* Juss. and *Digomphia* Benth.), and around 55 species distributed from Guatemala to Argentina (Olmstead et al. 2009). Both genera and 38 species are

found in Brazil (Lohmann 2010). This clade is sister to the rest of the family and easily distinguished by the elongated and glandular staminodes, pinnately or bipinnately compound leaves, calyx with deeply divided lobes, and oblong to elliptic flattened woody capsules that dehisce perpendicularly to the septum (Olmstead et al. 2009).

The Northeast of Brazil concentrates around 215 species, representing one of the regions with the greatest diversity of Bignoniaceae within the country (Lohmann 2010). The state of Pernambuco comprises 60 of the 215 species found in the region (Lohmann 2010). However, there is still little information about the family in Northeastern Brazil, especially in the state of Pernambuco. The objective of this work is to inventory all members of the Tabebuia alliance and tribe Jacarandeae in the state of Pernambuco (Northeastern Brazil), and present a taxonomic treatment for all taxa. Our findings will help reduce the taxonomic impediment and contribute knowledge that is relevant for the conservation of the Brazilian flora. This study provides useful information for future studies on the ecology, evolution, and biogeography of this iconic group of Brazilian plants.

## Material and Methods

Pernambuco is located in Northeastern Brazil (Figure 1), being limited in the south by the states of Alagoas and Bahia, in the north by the states of Ceará and Paraíba, and in the west by the state of Piauí.



**Figure 1.** Map indicating the location of the state of Pernambuco (in light green), within the Northeastern region of Brazil (in orange). Prepared by Thais Mara Souza.



Pernambuco is situated between 7°15'45" and 9°28'18"S, and between 34°48'35" and 41°19'54"W, including 98.311 km<sup>2</sup> of surface area (Andrade-Lima 1960). The state includes heterogeneous climate, soil, and phytophysionomies, although the regions with semi-arid climate, covered with Caatinga vegetation occupy around 80% of the state. Fragments of ombrophilous and seasonal forests, forested highlands, restingas, mangroves, dunes and associated ecosystems are available in the coastal areas and "Zona da Mata" (Andrade-Lima 1960).

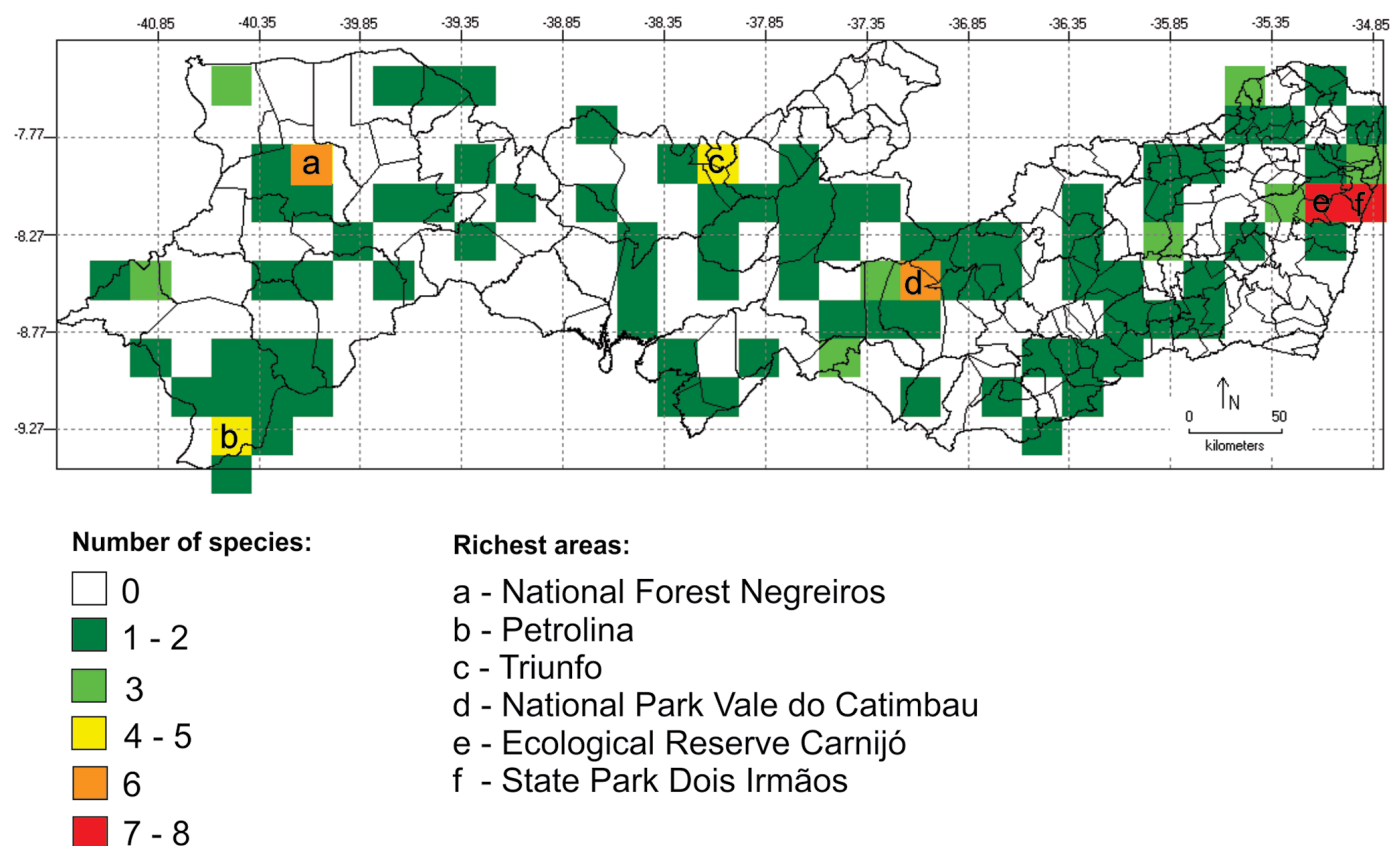
Field trips were carried out between 2017 and 2018 to collect and observe living material, as well as to obtain data on the ecological characteristics of the individual species. The timing of field trips was defined according to the species phenological data obtained from herbarium collections and literature. In the field, collection and herborization procedures followed Bridson & Forman (1998). Specimens were deposited in the PEUFR herbarium. In addition to the materials collected in the field, materials deposited at thirteen herbaria located in the state of Pernambuco were analyzed, i.e., ACAM, EAC, HESBRA, HST, HUEFS, HUFRN, HVASF, IPA, JPB, MOSS, PEUFR, RB and UFP (herbarium acronyms follow Thiers, continuously updated).

Specimen identification was based on comparisons with specimens previously identified by specialists, images of Type materials, species protologues, and published taxonomic treatments and floristic inventories (e.g., Bureau & Schumann 1896, Espirito-Santo et al. 2014, Gentry 1992,

2009, Lohmann 2004, 2010, Lohmann & Pirani 1996a, 1996b, 2003, Pereira & Mansano 2008, Scudeller 2004, Silva-Castro et al. 2007). We also consulted online collections of the Virtual Herbarium REFLORA (Brazil), Missouri Botanical Garden (MOBOT), and The New York Botanical Garden (NY) to support identifications and complement information on the species distribution and morphology.

Morphological descriptions were based on materials collected in the field and those deposited in herbaria. Descriptions follow the terminology of Harris & Harris (2000) and Gonçalves & Lorenzi (2007) and focus on diagnostic reproductive and vegetative traits. Due to the large amount of examined materials, we selected the best quality fertile specimens from each phytogeographic domain (Caatinga and/or Atlantic Forest) and list those as "selected material." The remaining specimens analyzed are included in an overall list of examined specimens (Appendix 1). Information on the geographic distribution, flowering and fruiting periods, ecological preferences, altitude, and common names were obtained from specimen labels. Ornamental species are not included.

The map of the region (Figure 1) and the species richness maps (Figure 2) were both prepared using DIVA-GIS, while figure captions were prepared in Corel Draw X7. The coordinates were obtained through field collections and original coordinates indicated in specimen labels or using *speciesLink*.



**Figure 2.** Map showing the species richness of members of the Tabebuia alliance and tribe Jacarandae in the state of Pernambuco. Prepared by Silmara Nepomuceno.

## Taxonomic treatment

### Identification key for the *Tabebuia* alliance and *Jacarandae* genera occurring in the state of Pernambuco

1. Leaves pinnate or bipinnate; staminode longer than fertile stamens, covered by glandular trichomes ..... *Jacaranda*
- 1'. Leaves simple or palmate; staminode shorter than fertile stamens, glabrous
2. Calyx tubular, densely lepidote to glabrous
3. Leaflets discolorous; inflorescences in thyrses; corolla white to light pink ..... *Sparattosperma*
- 3'. Leaflets concolorous; inflorescences in panicle; corolla yellow or white ..... *Tabebuia*
- 2'. Calyx cupular, pubescent or tomentose
4. Corolla infundibuliform
5. Calyx membranaceous; corolla green; capsules elliptic to oblong, ribbed ..... *Cybastax*
- 5'. Calyx coriaceous; corolla yellow, pink, lilac or purple; capsules linear-cylindrical, not-ribbed ..... *Handroanthus*
- 4'. Corolla campanulate or urceolate
6. Calyx green, with simple trichomes; corolla cream externally, with purple spots internally; capsules linear-twisted, smooth ..
- 6'. Calyx dark brown, with stellate trichomes; corolla brown externally, orange internally; capsules wide-elliptic, muricate ..... *Zeyheria*

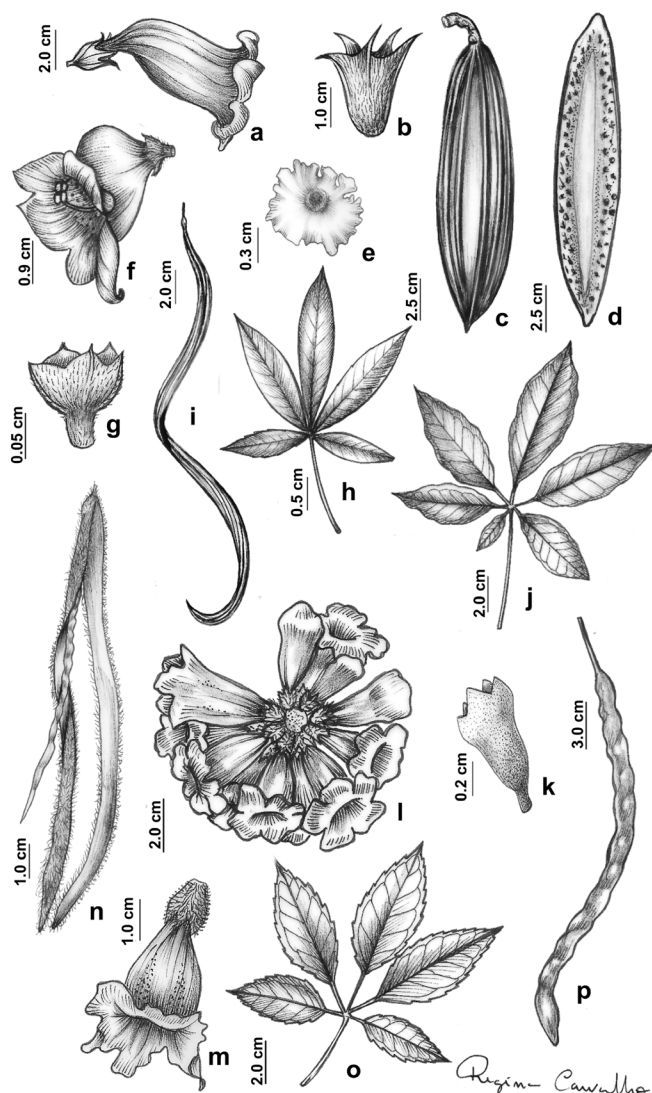
***Cybastax*** Martius ex Meisner, Pl. vasc. gen. 2: 208, 1840.

*Cybastax* is a monospecific genus, represented in the state of Pernambuco by *Cybastax antisiphilitica*.

1. *Cybastax antisiphilitica* (Mart.) Mart., Syst. Mat. Med. Bras., 66, 1843. Figure 3 a–e.

**Tree:** branchlets cylindrical, striated, without lenticels, glabrous.

**Leaves** palmate, 5–6 foliolate; petiole 6.1–16 cm long, glabrous; petiolule 0.4–1.6 cm long, glabrous; leaflets chartaceous, 11–13.4 × 4.7–5 cm, elliptic to obovate, base cuneate, apex acuminate, margin entire, slightly revolute, concolorous, adaxial surface glabrous, abaxial surface lepidote, with sparse simple trichomes; venation camptodromous. **Inflorescence** in thyrses, terminal; bracts 0.5–1.5 cm long, oblanceolate; bracteoles 0.5–0.8 cm long, oblanceolate to narrow-elliptic. **Calyx** cupular, membranaceous, 1.1–2.8 × 0.5–1.0 cm, 5-dentate, prolonged acuminate teeth, yellow to green, sparsely pubescent in length and velutinous at apex, with simple trichomes, internally velutinous, with simple trichomes, caducous. **Corolla** infundibuliform, 2.2–6.8 × 0.9–1.3 cm, both sides green, externally pubescent, with simple trichomes; stamens included, anthers ca. 0.3 cm long, glabrous, dorsal filaments 2.2–2.3 cm long, ventral filaments 1.7–1.9 cm long, staminode shorter than fertile stamens, ca. 0.2 cm long; ovary sessile, ovate-oblong, 0.3 × 0.1 cm, lepidote, style ca. 2.7 cm long, stigma ca. 0.2 cm long, lanceolate. **Capsule** 15.3 × 4.2 cm, elliptic to oblong, longitudinally 12-ribbed, prominent ribs, base and apex acuminate, glabrous surface, woody, slightly inflated. **Seeds** winged, 0.6–1.5 × 0.7–1.8 cm, wide-elliptic, wings hyaline, membranaceous.



**Figure 3.** *Cybastax antisiphilitica* (Mart.) Mart.: a. Flower. b. Calyx. c. Fruit. d. Fruit replum. e. Seed. *Godmania dardanoi* (J.C. Gomes) A.H. Gentry: f. Flower. g. Calyx. h. Leaf. i. Fruit. *Handroanthus capitatus* (Bureau & K. Schum) Mattos: j. Leaf. k. Calyx. *Handroanthus chrysotrichus* (Mart. ex DC.) Mattos: l. Inflorescence. m. Flower. n. Fruit. *Handroanthus heptaphyllus* (Vell.) Mattos: o. Leaf. p. Fruit.

**Habitat and Distribution:** *Cybastax antisiphilitica* occurs in Peru, Bolivia, Paraguay, Argentina and, disjointly, in Suriname (Gentry 1992). In Brazil it is distributed in all phytogeographical domains, occurring from Pará to Santa Catarina (Lohmann 2010). This species is a new record for the state of Pernambuco. It was found in a Caatinga area, at high altitudes (ca. 875 m).

**Phenology:** Collected with flowers and fruits in December.

**Taxonomic Notes:** *Cybastax antisiphilitica* can be recognized by the calyx 5-dentate, with teeth long acuminate (Figure 3a-b), and oblong fruit, longitudinally ribbed (Figure 3c-d). This is the only species in the Bignoniaceae with light green corollas, which led to the popular name “ipê verde.”

**Examined Material:** BRAZIL. PERNAMBUCO: Serrita, Serra de Brejinho, 14.12.2012, fl. e fr., R.A. Silva 2493 (HVASF 18878).

**Additional Examined Material:** BRAZIL. CEARÁ: Santana do Cariri, 04.12.1974, fl., Academia Brasileira de Ciências 1154 (IPA 21491).

*Godmania* Hemsley, Diagn. pl. nov. mexic. 35, 1879.

The genus includes two species, *G. aesculifolia* and *G. dardanoi*, distributed from Mexico to Brazil and Bolivia. In Pernambuco the genus is represented by *Godmania dardanoi*.

2. *Godmania dardanoi* (J.C. Gomes) A.H. Gentry, Ann. Missouri Bot. Gard., 63: 74, 1936. Figure 3 f–i.

**Tree**, 3–10 m alt.; branchlets cylindrical, striated, with lenticels, pubescent, with simple trichomes. **Leaves** palmate, 4–6 foliolate; petiole 1.5–6.7 cm long, pubescent, with simple trichomes; petiolule absent; leaflets chartaceous, 1.7–4.9 × 0.7–1.5 cm, elliptic, base cuneate, apex acute, margin entire to irregularly serrate, discolorous, pubescent, with simple trichomes in both sides, abaxial surface grayish; venation camptodromous. **Inflorescence** in thyrses, terminal; bracts 0.1–0.3 cm long, lanceolate; bracteoles 0.1–0.2 cm long, lanceolate. **Calyx** cupular, 0.1–0.3 × 0.1–0.2 cm, 5–dentate, teeth attenuated, green, pubescent, with simple trichomes, caducous. **Corolla** campanulate to urceolate, 2.0–3.9 × 0.9–2.1 cm, externally cream and internally with purple spots, lobes cuneate and facing out, pubescent externally, with simple and glandular trichomes; stamens included, anthers ca. 0.2 cm long, pubescent, dorsal filaments 1.7–1.8 cm long, ventral filaments 1.4–1.5 cm long, staminode shorter than fertile stamens, ca. 0.2 cm long; ovary sessile, linear-oblong, 0.3 × 0.1 cm, lepidote, style ca. 1.7 cm long, stigma ca. 0.2 cm long, lanceolate. **Capsule** 22.3–33.4 × 0.4–0.6 cm, linear-twisted, flattened, smooth, with longitudinal ridges along the entire length, pubescent surface, with simple trichomes, margin entire. **Seeds** winged, 4.5 × 0.5 cm, elliptic, wings hyaline, lengthy, membranaceous.

**Habitat and Distribution:** *Godmania dardanoi* is endemic to the Brazilian Northeast, occurring in the states of Bahia, Ceará, Pernambuco, Piauí, and Paraíba, in Caatinga and Cerrado environments (Lohmann 2010; Brito et al. 2018). In Pernambuco this species was found in areas of Caatinga, near rivers and roadside.

**Phenology:** Collected with flowers from June to January and with fruits in February.

**Taxonomic Notes:** *Godmania dardanoi* is recognized by the sessile leaflets, trait that differentiates this species from the other species in the genus (Figure 3h), corolla campanulate to urceolate (Figure 3f), internally with purple spots and fruit twisted and longitudinally ridged (Figure 3i). This species is popularly known in the study area as “chifre-de-carneiro” or “tapioca.”

**Selected Material:** BRAZIL. PERNAMBUCO: Araripina, 19.11.1992, fl., A.M. Miranda et al. 671 (HUFNR 2157); Ipubi, 08.02.1983, fl. and fr. G. Fotius 3345 (HUEFS 173093).

*Handroanthus* Mattos, Loefgrenia, 50: 2, 1970.

Small and large trees. Leaves palmate, 3–6 foliolate. Inflorescence terminal, a panicle or cyme. Calyx cupular, coriaceous, pubescent, tomentose, lepidote or villose. Corolla infundibuliform, yellow, pink, lilac or purple; stamens included, anthers glabrous, staminode shorter than fertile stamens. Capsule linear-cylindrical, flattened. Seeds winged, wings hyaline, membranaceous.

*Handroanthus* is represented in the state of Pernambuco by 30 species distributed from Central and South America and Antilles (Gentry 1992). In Brazil 27 species (15 endemic) are found (Lohmann 2010).

In the study area eight species were found, *H. capitatus*, *H. chrysotrichus*, *H. heptaphyllus*, *H. impetiginosus*, *H. ochraceus*, *H. serratifolius*, *H. spongiosus*, and *H. umbellatus*.

#### Identification key for the *Handroanthus* species occurring in the state of Pernambuco

1. Corolla pink or lilac
  2. Leaflets with margins entire, sometimes irregularly serrate, pubescent in adaxial surface and glabrescent to tomentose in abaxial surface, with simple trichomes; calyx pubescent with stellate and simple trichomes; capsule without irregular constrictions ..... 6. *H. impetiginosus*
  - 2'. Leaflets with margins serrate, glabrous; calyx sparsely lepidote; capsule with irregular constrictions ..... 5. *H. heptaphyllus*
- 1'. Corolla yellow
  3. Inflorescence in cyme or fascicle
    4. Leaves 3-foliate, strongly discolorous; calyx deeply 5-dentate with a thin membrane between teeth ..... 9. *H. spongiosus*
    - 4'. Leaves 5-foliate, concolorous; calyx irregularly 3–5 dentate with longitudinal ribs along this ..... 10. *H. umbellatus*
  - 3'. Inflorescence in panicle
    5. Leaflets glabrescent or lepidote
      6. Branchlets tetragonal, with stellate trichomes, without lenticels; ovary linear-oblong, densely lepidote ..... 3. *H. capitatus*
      - 6'. Branchlets cylindrical, with simple trichomes, with lenticels; ovary ovate, pubescent with glandular trichomes ..... 8. *H. serratifolius*
    - 5'. Leaflets pubescent or tomentose
      7. Leaflets with abaxial surface rusty; sessile flowers; capsule with lanuginous indument ..... 4. *H. chrysotrichus*
      - 7'. Leaflets with abaxial surface whitish; pedicellate flowers; capsule with floccose indument ..... 7. *H. ochraceus*

3. *Handroanthus capitatus* (Bureau & K. Schum) Mattos, Loefgrenia, 50: 4, 1970. Figure 3 j–k.

**Tree**; branchlets tetragonal, striated, without lenticels, pubescent, with stellate trichomes. **Leaves** 5–6 foliolate; petiole 5.5–6.2 cm long, pubescent, with stellate trichomes; petiolule 2.0–2.6 cm long, pubescent, with stellate trichomes; leaflets chartaceous, 7.2–14.1 × 4.2–5.8 cm, elliptic, base rounded, apex acuminate, margin entire, concolorous, adaxial surface glabrescent, abaxial surface pubescent only along the main vein and in the axils of the secondary veins, with stellate trichomes; venation brochidodromous. **Inflorescence** in panicle. **Calyx** 0.7–1.2 × 0.5–0.8 cm, irregularly dentate, pubescent, with stellate trichomes. **Corolla** 4.9–7.2 × 0.9–1.5 cm, yellow, glabrous externally; anthers ca. 0.2 cm long, dorsal filaments ca. 1.7 cm long, ventral filaments ca. 1.4 cm long, staminode ca. 0.2 cm long; ovary linear-oblong, 0.2 × 0.1 cm, densely lepidote, style ca. 2.0 cm long, stigma ca. 0.2 cm long. **Fruits and seeds** not seen.

**Habitat and Distribution:** *Handroanthus capitatus* is distributed through Guyana, Suriname, Peru, Venezuela, and Brazil. This species is restricted to the Amazonian domain, where it occurs in north and northeastern Brazil (Maranhão only) (Gentry 1992, Lohmann 2010).



Despite that, a specimen from the Atlantic Forest of Brazil (D. Andrade-Lima 52-1000) was identified by Gentry (1979) as *H. capitatus* due to the densely lepidote ovary. Even though we were able to verify the identity of this specimen, it is possible that this specimen may have been cultivated in this region.

**Phenology:** Collected with flowers in March.

**Taxonomic Notes:** *Handroanthus capitatus* can be recognized by the branchlets and calyx with stellate trichomes (Figure 3k), by the leaflets entire, pubescent along the main vein and axils of the secondary veins (abaxial surface), and by the densely lepidote ovary.

**Examined Material:** BRAZIL. PERNAMBUCO: São Lourenço da Mata, Usina Tiuma, 13.03.1952, fl., D. Andrade-Lima 52-1000 (IPA 2482).

4. *Handroanthus chrysotrichus* (Mart. ex DC.) Mattos, Loefgrenia, 50: 4, 1970. Figure 3 l–n.

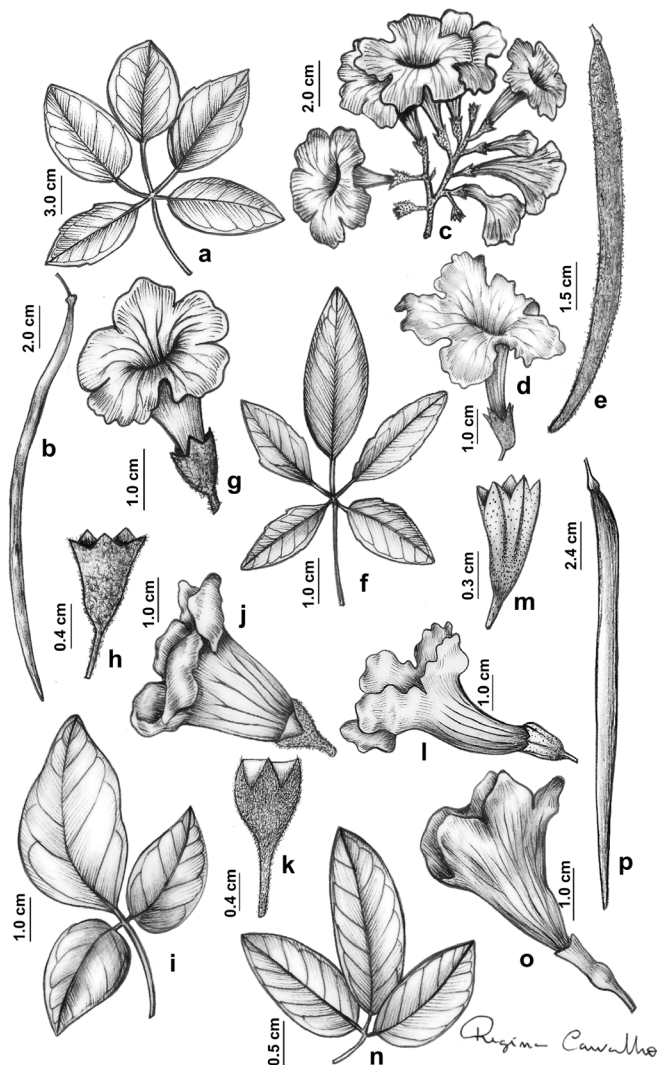
**Tree**, 8–10 m; branchlets cylindrical, striated, with lenticels, pubescent, with stellate trichomes. **Leaves** 3–5 foliolate; petiole 1.3–2.9 cm long, tomentose, with stellate trichomes; petiolules 0.5–1.8 cm long, tomentose, with stellate trichomes; leaflets chartaceous, 3.0–7.2 × 1.7–3.3 cm, obovate to elliptic, base cuneate, apex cuneate to cuspidate, margin entire, discolorous, adaxial surface tomentose, with stellate trichomes, abaxial surface rusty, tomentose, with simple and stellate trichomes; venation brochidodromous. **Inflorescence** in panicle, congest, sessile flowers. **Calyx** 0.5–1.9 × 0.3–0.8 cm, 5-dentate, teeth acute, rust, tomentose, with rust stellate and dendritic trichomes, persistent. **Corolla** 2.8–6.5 × 0.5–2.1 cm, yellow, externally pubescent, with simple trichomes; anthers ca. 0.3 cm long, dorsal filaments 2.1–2.3 cm long, ventral filaments 1.8–1.9 cm long staminode ca. 0.2 cm long; ovary sessile, linear-oblong, 0.3 × 0.2 cm, pubescent, with glandular trichomes, style ca. 2.0 cm long, stigma ca. 0.2 cm long, lanceolate. **Capsule** 10.2–15.2 × 0.9–1.7 cm, rusty, lanuginous surface, with simple and stellate trichomes, margin entire. Seeds not seen.

**Habitat and Distribution:** *Handroanthus chrysotrichus* is found in Argentina and in Cerrado and Atlantic Forest environments along the Brazilian coast (Lohmann 2010). In Pernambuco it occurs in areas of Caatinga and Atlantic Forest, in rocky outcrops and “brejos de altitude,” with altitudes varying from 400 to 1080 m.

**Phenology:** Collected with flowers in September, October and January and with fruits in January.

**Taxonomic Notes:** *Handroanthus chrysotrichus* and *H. ochraceus* are morphologically similar due to the shared leaflet shape (obovate to elliptic), texture (chartaceous), trichomes type (stellate), inflorescence type (a congest panicle; Figure 3l and 4c) and shape (linear-cylindrical), and capsule color (rusty) (Figure 3n and 4e). These species can be differentiated by the leaflet with rusty abaxial surface (vs. whitish abaxial surface in *H. ochraceus*), sessile flowers (Figure 3l) (vs. pedicellate flowers; Figure 4c), and capsule with indument lanuginous (vs. with floccose indument).

**Selected Material:** BRAZIL. PERNAMBUCO: Maraial, Engenho Curtume, 25.11.2007, fr., M. Sobral-Leite 561 (UFP 50292); Caruaru, Brejo dos Cavalos, 09.01.1999, fl., E. Locatelli & P. Medeiros s.n. (UFP 39390)



**Figure 4.** *Handroanthus impetiginosus* (Mart. ex DC.) Mattos: a. Leaf. b. Fruit. *Handroanthus ochraceus* (Cham.) Mattos: c. Inflorescence. d. Flower. e. Fruit. *Handroanthus serratifolius* (Vahl.) S. Grose: f. Leaf. g. Flower. h. Calyx. *Handroanthus spongiosus* (Rizzini) S. Grose: i. Leaf. j. Flower. k. Calyx. *Handroanthus umbellatus* (Sond.) Mattos: l. Flower. m. Calyx. *Sparattosperma catingae* A.H. Gentry: n. Leaf. o. Flower. p. Fruit.

5. *Handroanthus heptaphyllus* (Vell.) Mattos, Loefgrenia, 50: 2, 1970. Figure 3 o–p.

**Tree;** branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** 5–6 foliolate; petiole 2.8–9.8 cm long, glabrous; petiolule 0.8–2.8 cm long, glabrous; leaflets chartaceous, 3.0–10.9 × 1.3–5.2 cm, obovate to elliptic, base obtuse, apex acute to attenuate, margin serrate, concolorous, glabrous in both sides; venation brochidodromous. **Inflorescence** in panicle, pedicellate flowers. **Calyx** 0.3–0.9 × 0.2–0.5 cm, 5-dentate, teeth obtuse, lilac, sparsely lepidote, caducous. **Corolla** 2.2–5.8 × 1.2–2.7 cm, lilac with yellow nectar guides, externally pubescent, with simple trichomes; anthers ca. 0.3 cm long, dorsal filaments 2.2–2.3 cm long, ventral filaments 1.5–1.6 cm



long, staminode ca. 0.3 cm long; ovary sessile, oblong,  $0.3 \times 0.1$  cm, pubescent, with glandular trichomes, style ca. 2.7 cm long, stigma ca. 0.3 cm long, lanceolate. **Capsule**  $24.5 \times 1.3$  cm, glabrous surface, margin with irregular constrictions, with lenticels. **Seeds**  $0.5\text{--}1.9 \times 0.2\text{--}0.3$  cm, narrow-elliptic.

**Habitat and Distribution:** *Handroanthus heptaphyllus* occurs in wet forests of Paraguay, western Bolivia and northeastern Argentina (Gentry 1992). In Brazil it is found in Cerrado and Atlantic Forest environments and is distributed in all states of the South, Southeastern and Midwestern regions, as well as in the Northeastern states of Bahia, Pernambuco and Ceará (Lohmann 2010). In the study area this species was found in areas of Caatinga with altitude around 400 m.

**Phenology:** Collected with flowers in September and October and with fruits in January.

**Taxonomic Notes:** Most specimens of *H. heptaphyllus* deposited in herbaria are identified as *H. impetiginosus*. Both species have glabrous branchlets, terminal panicle inflorescence, cupular calyx, 5-dentate and pink to lilac flowers with yellow to orange nectar guides. However, these species can be differentiated by leaflets glabrous (vs. pubescent with simple trichomes in *H. impetiginosus*), with serrate margin (Figure 3o) (vs. entire margins; Figure 4a), calyx sparsely lepidote (vs. pubescent with stellate trichomes), and fruit with irregular constrictions (Figure 3p) (vs. fruit with linear margin; Figure 4b).

**Selected Material:** BRAZIL. PERNAMBUCO: Tacaratu, 13.09.1990, fl., R. Pereira & A. Bocage 101 (IPA 53701); Tacaratu, 15.01.2009, fr., J.G. Carvalho-Sobrinho et al. 1802 (HVASF 2845).

6. *Handroanthus impetiginosus* (Mart. ex DC.) Mattos, *Loefgrenia*, 50: 2, 1970. Figure 4 a–b.

**Tree:** branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** 3–5 foliolate; petiole 2.2–9.2 cm long, glabrous; petiolule 0.8–3.9 cm long, glabrous; leaflets chartaceous,  $1.2\text{--}15.7 \times 1.3\text{--}6.9$  cm, elliptic, narrow-elliptic or obovate, base rounded, apex acute to acuminate, margin entire, sometimes irregularly serrate, concolorous, rarely discolorous, adaxial surface pubescent, with simple trichomes, abaxial surface glabrescent to tomentose, with simple trichomes; venation brochidodromous. **Inflorescence** in panicle, congest, glabrous to densely tomentose; bracts and bracteoles 0.1–0.2 cm long, narrow-elliptic. **Calyx**  $0.2\text{--}1.0 \times 0.2\text{--}0.7$  cm, 5-dentate to truncate, teeth cuneate, lilac, densely pubescent, glandular, with stellate and simple trichomes, persistent. **Corolla**  $2.2\text{--}6.7 \times 0.9\text{--}3.3$  cm, pink to lilac with yellow, pink or orange nectar guides, externally pubescent, with simple trichomes; anthers ca. 0.2–0.3 cm long, dorsal filaments 1.8–2.0 cm long, ventral filaments 1.5–1.6 cm long, staminode ca. 0.2 cm long; ovary sessile, oblong,  $0.3 \times 0.2$  cm, lepidote, style ca. 2.5 cm long, stigma ca. 0.2 cm long, lanceolate. **Capsule**  $10.9\text{--}32.3 \times 1.0\text{--}1.3$  cm, glabrous surface, green to dark brown, margin without irregular constrictions. **Seeds**  $0.5\text{--}1.7 \times 0.2\text{--}0.4$  cm, narrow-elliptic.

**Habitat and Distribution:** *Handroanthus impetiginosus* is distributed from Northeastern Mexico to Argentina (Gentry 1992), and is widely distributed throughout the Brazilian territory (except for the southern region), where it occurs in Amazonia, Caatinga, Cerrado, Atlantic Forest, and Pantanal (Lohmann 2010). In Pernambuco it was found in areas of Caatinga, Atlantic Forest, “brejos de altitude,” anthropic areas, and rocky outcrops, between 300 to 1200 m alt.

**Phenology:** Collected with flowers from April to December and fruits from July to March.

**Taxonomic Notes:** *Handroanthus impetiginosus* can be recognized by leaflets with entire margins (Figure 4a), pubescent with simple trichomes, pink to lilac flowers with yellow to orange nectar guides, by the calyx pubescent with stellate trichomes, and by the fruit with linear margins (Figure 4b). The similarity with *H. heptaphyllus* is discussed under that species comments.

**Selected Material:** BRAZIL. PERNAMBUCO: São Joaquim do Monte,  $8^{\circ}23'43''\text{S } 35^{\circ}51'02''\text{W}$ , 27.I.2014, fr., M. Oliveira 5736 (IPA); Buíque, Parque Nacional do Catimbau, 14.IX. 2011, fl., A.C.G. Costa et al. 45 (IPA).

7. *Handroanthus ochraceus* (Cham.) Mattos, *Loefgrenia*, 50: 2, 1970. Figure 4 c–e.

**Tree:** branchlets cylindrical, striated, with lenticels, pubescent, with stellate trichomes. **Leaves** 5-foliate; petiole 3.1–5.6 cm long, tomentose, stellate trichomes; petiolules 1.2–3.3 cm long, stellate trichomes; leaflets chartaceous,  $4.2\text{--}10.1 \times 3.1\text{--}7.3$  cm, obovate to elliptic, base cordate, apex retuse to rounded, margin entire, rarely serrated, sometimes sinuate, discolorous, adaxial surface pubescent only along of main veins and in axils of secondary veins, with stellate trichomes, abaxial surface pubescent, with whitish stellate trichomes; venation brochidodromous. **Inflorescence** in panicle, congest, pedicellate flowers. **Calyx**  $0.5\text{--}1.5 \times 0.5\text{--}1.0$  cm, 5-dentate, teeth acute, rust, villose, with simple and stellate trichomes, persistent. **Corolla**  $3.0\text{--}5.9 \times 0.9\text{--}2.8$  cm, yellow with wine ribs, externally glabrous and internally pubescent, with simple and stellate trichomes; anthers 0.2–0.3 cm, dorsal filaments ca. 2.1 cm long, ventral filaments ca. 1.8 cm long, staminode ca. 0.2 cm long; ovary sessile, oblong,  $0.2 \times 0.1$  cm, pubescent, style ca. 2.0 cm long, stigma ca. 0.2 cm long, lanceolate. **Capsule**  $9.2\text{--}13.2 \times 0.8\text{--}1.5$  cm, floccose surface, with stellate trichomes, rust, margin entire, without lenticels. **Seeds**  $1.6 \times 0.6$  cm, elliptic.

**Habitat and Distribution:** *Handroanthus ochraceus* is found in dry forests from Guatemala to Argentina, from sea level up to 1600 m (Gentry 1992). It is widely distributed throughout Brazil, where it occurs in Amazonia, Caatinga, Cerrado, and Atlantic Forest domains (Lohmann 2010). In the study area it was found in Atlantic Forest and Caatinga, “brejos de altitude,” and rocky outcrops.

**Phenology:** Collected with flowers in September to February, and fruits from September to November.

**Taxonomic Notes:** *Handroanthus ochraceus* can be recognized by obovate to elliptic leaflets with a whitish abaxial surface, by the pedicellate flowers (Figure 4c), and the capsule with floccose indumentum (Figure 4e). The similarity and differences with *H. chrysotrichus* are discussed under that species comments.

**Selected Material:** BRAZIL. PERNAMBUCO: Brejo dos cavalos, 01.III.1996, fr., D.S. Pimentel 55 (PEUFR 25148); Pesqueira, 28.IX.1995, fl., M. Correia 371 (IPA 58254)

8. *Handroanthus serratifolius* (Vahl.) S. Grose, *Syst. Bot.*, 32: 666, 2007. Figure 4 f–h.

**Tree:** branchlets cylindrical, striated, with lenticels, pubescent, with simple trichomes. **Leaves** 3–5 foliolate; petiole 1.5–3.9 cm long, pubescent, with simple trichomes; petiolule 0.9–1.3 cm long, pubescent,

with simple trichomes; leaflets chartaceous,  $3.1\text{--}7.3 \times 1.9\text{--}4.1$  cm, elliptic to obovate-elliptic, base cuneate to rounded, apex acuminate, margin entire, serrated or irregularly serrated, concolorous, lepidote on both sides; venation brochidodromous. **Inflorescence** in panicle; bracts and bracteoles absent. **Calyx**  $0.7\text{--}1.4 \times 0.5\text{--}0.9$  cm, 3–5 dentate to irregularly dentate, teeth cuneate, green, pubescent, with simple and stellate trichomes, caducous. **Corolla**  $4.8\text{--}7.9 \times 0.8\text{--}2.2$  cm, yellow, externally glabrous; anthers ca. 0.3 cm long, dorsal filaments  $2.2\text{--}2.4$  cm long, ventral filaments  $1.7\text{--}1.8$  cm long, staminode ca. 0.3 cm long; ovary sessile, ovate,  $0.3 \times 0.2$  cm long, pubescent, glandular trichomes, style ca. 3.1 cm long, stigma ca. 0.3 cm long. **Capsule**  $18.3\text{--}39.8 \times 0.7\text{--}1.5$  cm, glabrous surface, margin entire, without lenticels. **Seeds** not seen.

**Habitat and Distribution:** *Handroanthus serratifolius* occurs from Colombia to Bolivia in wet and dry forests (Gentry 1992). In Brazil this species is widely distributed throughout most of the territory (except the states of Rio Grande do Sul and Santa Catarina in the southern region), where it occurs in areas of Caatinga, Cerrado, Atlantic Forest, Pantanal, and Amazon forests (Lohmann 2010). In Pernambuco it occurs in the Atlantic Forest and Caatinga.

**Phenology:** Collected with flowers from November to January and with fruits in August.

**Taxonomic Notes:** *Handroanthus serratifolius* can be identified by the leaflets with entire, serrated or irregularly serrate margins (Figure 4f), and the calyx with cuneate teeth (Figure 4h), with simple and stellate trichomes.

**Selected Material:** BRAZIL. PERNAMBUCO: Exu, Serra da Gameleira, 06.VIII.1986, fr., V.C. Lima 337 (IPA 49180); Bonito, 29.I.1970, fl., Andrade-Lima 70–5683 (IPA 20913).

9. *Handroanthus spongiosus* (Rizzini) S. Grose, *Syst. Bot.*, 32: 666, 2007. Figure 4 i–k.

**Tree:** branchlets cylindrical, striated, without lenticels, pubescent, with stellate trichomes. **Leaves** 3-foliolate; petiole  $1.1\text{--}4.2$  cm long, tomentose, with stellate trichomes; petiolules  $0.5\text{--}1.0$  cm long, tomentose, with stellate trichomes; leaflets membranaceous,  $4.5\text{--}8.7 \times 1.9\text{--}3.3$  cm, elliptic to obovate-elliptic, base obtuse to rounded, apex attenuate, margin entire, strongly discoloured, adaxial surface tomentose, with stellate trichomes, abaxial surface densely tomentose, with whitish stellate trichomes, sometimes rusty; venation brochidodromous. **Inflorescence** a dicaxial cyme, densely tomentose, with stellate rusty trichomes; bracts and bracteoles absent. **Calyx**  $0.3\text{--}1.7 \times 0.3\text{--}0.8$  cm, deeply 5-dentate with a thin membrane between the teeth, rusty, densely tomentose, with rusty stellate trichomes, persistent. **Corolla**  $1.8\text{--}4.9 \times 1.2\text{--}1.5$  cm, yellow with red nectar guides, externally glabrous; anthers ca. 0.2 cm long, dorsal filaments  $1.9\text{--}2.1$  cm long, ventral filaments  $1.7\text{--}1.8$  cm long, staminode ca. 0.2 cm long; ovary sessile, linear, ca. 0.2 cm long, lepidote, style ca. 3.5 cm long, stigma ca. 0.2 cm long. **Capsule**  $13.1\text{--}22.5 \times 0.6\text{--}0.8$  cm, base cuneate, apex attenuate, glabrous surface, margin with irregular constrictions, without lenticels. **Seeds**  $2.1 \times 0.5$  cm, oblong-elliptic.

**Habitat and Distribution:** This species is endemic to Northeastern Brazil, where it occurs in the states of Alagoas, Bahia, Paraíba, Pernambuco, Piauí and Sergipe (Lohmann 2010). In Pernambuco it occurs in Caatinga, between 300 to 400 m of altitude.

**Phenology:** Collected with flowers in May, August and October to January, and with fruits from November to February.

**Taxonomic Notes:** *Handroanthus spongiosus* is characterized by 3-foliolate leaves (Figure 4i), densely tomentose leaflets, inflorescence and calyx, with stellate rusty trichomes, and calyx deeply 5-dentate with a thin membrane between the teeth (Figure 4j–k). In the field, it can be identified by the yellow flowers with red nectar guides, and by the flaky bark. This species is known in the region by the common names “sete cascas” and/or “cascudo.”

**Selected Material:** BRAZIL. PERNAMBUCO: Lagoa Grande, Fazenda Cabana,  $08^{\circ}30'25.84''\text{S } 40^{\circ}15'33.64''\text{W}$ , 10.XII.2012, fr., A.C.P. Oliveira et al. 2044 (HVASF 18918); Petrolina,  $08^{\circ}48'07''\text{S } 40^{\circ}48'04''\text{W}$ , 534 m, 21.XI.2011, fl., T.S. Oliveira et al. 73 (HVASF 13431).

10. *Handroanthus umbellatus* (Sond.) Mattos, *Loefgrenia*, 50: 2, 1970. Figure 4 l–m.

**Tree:** branchlets cylindrical, striated, with lenticels, sparsely pubescent, with stellate trichomes. **Leaves** 5-foliolate; petiole  $3.0\text{--}4.2$  cm long, densely pubescent, with stellate trichomes; petiolules  $1.5\text{--}2.3$  cm long, densely pubescent, with simple trichomes; leaflets membranaceous,  $5.5\text{--}8.9 \times 1.7\text{--}4.6$  cm, elliptic, base obtuse, apex acuminate, margin entire, concolorous, pubescent on both sides, with stellate trichomes; venation brochidodromous. **Inflorescence** a fascicle; bracts and bracteoles absent. **Calyx**  $0.6\text{--}2.0 \times 0.4\text{--}0.9$  cm, irregularly 3–5 dentate, teeth cuneate, with longitudinal ribs along to calyx, green to yellow, densely tomentose on base and on ribs, sparsely pubescent on apex, with stellate trichomes. **Corolla**  $4.2\text{--}7.7 \times 1.5\text{--}2.1$  cm, yellow, glabrous externally; anthers ca. 0.3 cm long, dorsal filaments  $2.0\text{--}2.1$  cm long, ventral filaments  $1.4\text{--}1.5$  cm long, staminode ca. 0.3 cm long; ovary sessile linear-cylindrical,  $0.3 \times 0.1$  cm, lepidote, style ca. 2.2 cm long, stigma ca. 0.3 cm long, lanceolate. **Fruit and seeds** not seen.

**Habitat and Distribution:** *Handroanthus umbellatus* is endemic to Brazil, where it occurs in Atlantic Forest vegetation along the Brazilian coast, from Rio Grande do Sul to Bahia (Lohmann 2010). It represents a new record for the state of Pernambuco, where it occurs in the Atlantic Forest, at forest edges, with altitudes ranging between 400–600 m.

**Phenology:** Collected with flowers in January and February.

**Taxonomic Notes:** *Handroanthus umbellatus* is recognized by the calyx densely tomentose at the base and sparsely pubescent at apex, longitudinally ribbed (Figure 4l–m).

**Examined Material:** BRAZIL. PERNAMBUCO: Bezerros, Parque Ecológico Serra Negra, 08.02.1996, fl., E. Inácio et al. 139 (MO); São Vicente Férrer, Mata do Estado, 29.01.1999, fl., E.M.N. Ferraz et al. 583 (MO).

*Jacaranda* Jussieu, Gen. Pl. 138, 1789.

Trees or shrubs. Leaves bipinnate or pinnate. Inflorescence a panicle, terminal or axillar. Calyx campanulate, pubescent, glabrescent, villous or lepidote; corolla infundibuliform with the narrow base, purple, blue, wine or lilac; stamens included, anthers mono-thecae or di-thecae, staminode larger than fertile stamens, pubescent from half to apex, glandular trichomes. Capsule wide-elliptic, elliptic or ovate, flattened or inflated. Seeds winged, wings hyaline, membranaceous.

*Jacaranda* includes 49 species distributed from Guatemala to Argentina (Gentry 1992). In Brazil, 36 species (32 endemic) are found (Lohmann 2010). Six species are found in the study area (i.e., *J. brasiliensis*, *J. cuspidifolia*, *J. jasminoides*, *J. microcalyx*, *J. puberula*, and *J. rugosa*).

### Identification key for *Jacaranda* species occurring in the state of Pernambuco

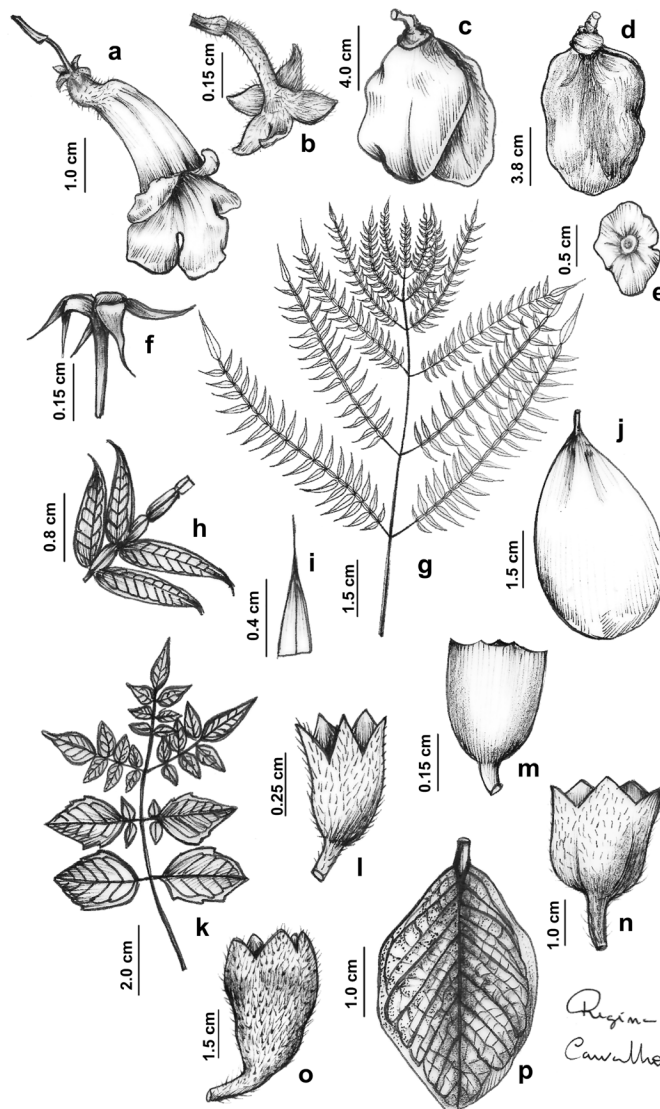
1. Anthers mono-thecae
  2. Pinnules concolorous; calyx lobes ovate with acute apex, not reflexed, divided almost to the base; capsule inflated and undulate at dehiscence ..... 11. *J. brasiliiana*
  - 2'. Pinnules discolorous; calyx lobes lanceolate with acuminate apex, reflexed, divided to the base; capsule flattened and smooth at dehiscence ..... 12. *J. cuspidifolia*
- 1'. Anthers bi-thecae
  3. Leaves pinnate completely or pinnate on base and bipinnate at apex; leaflets coriaceous; inflorescence axillary
    4. Leaflets with smooth surface; bracts and bracteoles narrow-elliptic; calyx sparsely pubescent ..... 13. *J. jasminoides*
    - 4'. Leaflets with rugose surface; bracts and bracteoles absent; calyx villose ..... 16. *J. rugosa*
  - 3'. Leaves bipinnate; leaflets chartaceous; inflorescence terminal
    5. Branchlets tetragonal; leaflets concolorous and lepidote on both sides; bracts and bracteoles absent ..... 14. *J. microcalyx*
    - 5'. Branchlets cylindrical; leaflets discolorous and glabrous on adaxial surface and densely pubescent on abaxial surface; bracts and bracteoles present ..... 15. *J. puberula*

#### 11. *Jacaranda brasiliiana* (Lam.) Pers., Syn. Pl. 2: 174, 1807. Figure 5 a–e.

**Tree**, 10 m; branchlets cylindrical, winged, striated, with lenticels, sparsely pubescent, with simple trichomes. **Leaves** bipinnate; petiole 4.1–4.8 cm long, pubescent, with simple trichomes; pinnules sessile, chartaceous, 0.7–1.2 × 0.3–0.6 cm, elliptic to oblong-elliptic, base cuneate, apex obtuse to acute, margin entire, concolorous, adaxial surface glabrous, abaxial surface pubescent only in main veins, with simple trichomes; venation brochidodromous. **Inflorescence** terminal; bracts and bracteoles absent. **Calyx** 0.3–0.5 × 0.2–0.3 cm, 5-lobed, lobes divided almost to the base, lobes ovate with acute apex, not reflexed, purple, pubescent, with glandular trichomes, caducous. **Corolla** 3.9–5.7 × 1.5–1.7 cm, purple to blue, externally densely pubescent on base and sparsely pubescent on tube and lobes, with glandular trichomes; anthers ca. 0.2 cm long, mono-thecae, dorsal filaments 2.2–2.5 cm long, ventral filaments 1.7–1.8 cm long, staminode 3.0–3.3 cm long; disc annular, 0.2 × 0.2 cm; ovary ovate, 0.2 × 0.1 cm, glabrous, style ca. 2.4 cm long, stigma ca. 0.3 cm long, elliptic. **Capsule** 8.6–19.2 × 7.7–16.8 cm, wide-elliptic, inflated, woody, undulate at dehiscence, surface glabrous, margin entire, without lenticels. **Seeds** 2.1 × 1.8 cm, wide-elliptic.

**Habitat and Distribution:** *Jacaranda brasiliiana* is endemic to Brazil, where it occurs in the Amazon, Cerrado, and Caatinga domains, from Mato Grosso to Minas Gerais and from Pará to Bahia (Lohmann 2010). In Pernambuco it occurs in environments of Caatinga with altitudes ranging from 500–890 m.

**Phenology:** Collected with flowers from November to February, and with fruits in February.



**Figure 5.** *Jacaranda brasiliiana* (Lam.) Pers.: a. Flower. b. Calyx. c–d. Fruit. e. Seed. *Jacaranda cuspidifolia* Mart.: f. Calyx. g. Leaf. h. Detail of the leaf rachis and leaflets. i. Detail of leaflet apex. j. Fruit. *Jacaranda jasminoides* (Thunb.) Sandwith: k. Leaf. l. Calyx. *Jacaranda microcalyx* A.H. Gentry: m. Calyx. *Jacaranda puberula* Cham.: n. Calyx. *Jacaranda rugosa* A.H. Gentry: o. Calyx. p. Leaf.

**Taxonomic Notes:** *Jacaranda brasiliiana* is morphologically very close to *J. cuspidifolia* with which it shares the cylindrical branchlets, sessile pinnules, entire pinnules margin, terminal inflorescence, and purple calyx. Despite that, *J. brasiliiana* can be differentiated by the concolorous pinnules (vs. discolorous pinnules in *J. cuspidifolia*), pinnae not winged (vs. pinnae slightly winged; Figure 5h), calyx lobes ovate with acute apex, divided almost to the base (Figure 5b) (vs. lobes lanceolate with acuminate apex, divided to the base; Figure 5f), and the inflated, undulate wide-elliptic fruit (Figure 5c–d) (vs. ovate, flattened wide-elliptic fruit; Figure 5j).



**Selected Material:** BRAZIL. PERNAMBUCO: Arcoverde, Serra das Varas, 890 m, 22.II.2006, fl. and fr., R. Pereira et al. 2707 (IPA 73344); São José do Belmonte, 07°44'20,63"S 38°41'32,69"W, 570 m, 30.VII.2013, fl., A.C.P. Oliveira et al. 2945 (HVASF 21170).

12. *Jacaranda cuspidifolia* Mart., *Prodr.*, 9: 228, 1845. Figure 5 f–j.

**Shrubs**, 8–10 m; branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** bipinnate; petiole 2.0–4.4 cm long, glabrous; pinna slightly winged; pinnules sessile, membranaceous, 1.5–2.5 × 0.3–0.6 cm, lanceolate to oblong, base obtuse to cuneate, apex long acuminate, margin entire, discolorous, adaxial surface glabrous, abaxial surface pubescent, with simple trichomes; venation brochidodromous. **Inflorescence** terminal; bracts and bracteoles absent. **Calyx** 5-lobed, lobes divided to the base, lobes lanceolate with apex acuminate, reflexed, 0.2–0.5 × 0.1–0.3 cm, purple, glabrescent, with simple trichomes on lobes margin, caducous. **Corolla** 2.4–5.4 × 0.6–1.5 cm, lilac, externally pubescent, with glandular trichomes; anthers 0.2–0.3 cm long, mono-thecae, dorsal filaments 2.5–2.6 cm long, ventral filaments 2.0–2.1 cm long, staminode ca. 2.8 cm long; disc annular, 0.2 × 0.2 cm; ovary ovate, 0.2 × 0.1 cm, glabrous, style ca. 2.6 cm long, stigma ca. 0.2 cm long, elliptic. **Capsule** 4.9–6.8 × 3.3–5.7 cm, ovate to wide-elliptic, flattened, smooth at dehiscence, base rounded, apex acute, surface glabrous, margin entire, with lenticels. **Seeds** 1.2 × 1.0 cm, wide-elliptics.

**Habitat and Distribution:** *Jacaranda cuspidifolia* occurs from Argentina to Bolivia (Gentry 1992). In Brazil it is distributed through Cerrado, Atlantic Forest, and Pantanal vegetation of the Midwestern and Southeastern states (Lohmann 2010). This species is a new record for the state of Pernambuco and consequently for Northeastern Brazil. It is found in high altitude forest (above 1000 m).

**Phenology:** Collected with flowers and with fruits in November.

**Taxonomic Notes:** *Jacaranda cuspidifolia* is recognized by the pinna slightly winged (Figure 5h), the sessile leaflets (Figure 5g) that are lanceolate to oblong, with long and acuminate apices (Figure 5i), and by the calyx 5-lobed, with lanceolate and acuminate lobes divided all the way until the base and reflexed (Figure 5f). The similarity and differences with *J. brasiliensis* are discussed under that species comments.

**Selected Material:** BRAZIL. PERNAMBUCO: Recife, Santuário dos Três Reinos, 07°57'41,4"S 34°56'20,9"W, 99 m, 24.X.2013, fl., M. Sobral-Leite et al. 1336 (UFP 79079); Triunfo, Sítio Jardim, 17.XI.1998, fl. and fr., A.M. Miranda 3080 (HUEFS 185187).

13. *Jacaranda jasminoides* (Thunb.) Sandwith, Figure 5 k–l.

**Tree or shrubs**, 3–8 m; branchlets cylindrical, striated, with lenticels, pubescent, with simple and glandular trichomes. **Leaves** pinnate on base and bipinnate on apex, with terminal leaflet bigger; petiole 3.8–5.0 cm long, pubescent, with simple trichomes; leaflets and pinnules sessile, coriaceous, 0.9–5.2 × 0.5–2.1 cm, ovate-elliptic to ovate, base cuneate to rounded, apex acute to obtuse, margin entire, discolorous, adaxial surface sparsely pubescent, with simple trichomes, abaxial surface densely pubescent, simple trichomes, smooth surface; venation brochidodromous. **Inflorescence** axillar; bracts and bracteoles 0.2–0.3 cm long, narrow-elliptics. **Calyx** 0.4–0.9 × 0.3–0.5 cm, 5-dentate, teeth attenuate, purple to vinaceous, sparsely pubescent, with glandular trichomes, persistent. **Corolla** 3.1–4.2 × 1.2–1.9 cm, wine, externally sparsely pubescent, with glandular trichomes; anthers ca.

0.2 cm long, di-thecae, dorsal filament 2.6–2.8 cm long, ventral filament 1.9–2.1 cm long, staminode ca. 4.0 cm long; ovary ovate, 0.2 × 0.2 cm, glabrous, style ca. 3.5 cm long, stigma ca. 0.3 cm long, elliptic. **Capsule** 4.4 × 2.5 cm, elliptic to wide-elliptic, woody, base and apex rounded, surface glabrous, margin entire, with lenticels. **Seeds** 1.2 × 1.5 cm, wide-elliptic.

**Habitat and Distribution:** *Jacaranda jasminoides* is endemic to Brazil, where it occurs along the Brazilian coast, from Ceará to Rio de Janeiro, in Caatinga, Cerrado, and Atlantic Forest vegetation (Lohmann 2010). In Pernambuco it was found in areas of Caatinga, in mountains up to 1000 m altitude, and rocky outcrops up to 600 m.

**Phenology:** Collected with flowers from November to February, and fruits from May to December.

**Taxonomic Notes:** Morphologically close to *J. rugosa* with which it shares cylindrical branchlets, discolorous, and coriaceous leaflets, axillar inflorescences, elliptic to wide-elliptic capsules, and persistent calyx in the fruit (unusual feature in the genus). These species can be differentiated by the pinnate leaves at the base and bipinnate at the apex (Figure 5k) (vs. bipinnate leaves in throughout the leaf extension in *J. rugosa*), leaflets with smooth surface (Figure 5k) (vs. surface densely rugose; Figure 5p), and calyx 5-dentate with attenuate teeth (Figure 5l) (vs. teeth cuneate to rounded; Figure 5o).

**Selected Material:** BRAZIL. PERNAMBUCO: Araripina, 08.VIII.1986, fr., V.C. Lima 383 (IPA 49191); Tacaratu, Serra Grande, 09°04'20,70"S 38°07'34,20"W, 802 m, 23.XI.2009, fl. and fr. A.P. Fontana & G. Rodrigues 6247 (HVASF 6460).

14. *Jacaranda microcalyx* A.H. Gentry, *Fl. Neotrop. Monogr.*, 25(2): 87, 1992. Figure 5 m.

**Tree**, 8–10 m; branchlets tetragonal, striated, with lenticels, glabrous. **Leaves** bipinnate; petiole 3.5–7.0 cm long, canaliculate, lepidote; petiolule 1.2–1.5 cm long, canaliculate, lepidote; pinnule subsessile, decreasing as it reaches the base; chartaceous, 3.3–9.5 × 1.6–4.7 cm, elliptic to obovate-elliptic, base obtuse to cuneate, apex attenuate to cuneate, margin entire, concolorous, adaxial surface lepidote, abaxial surface densely lepidote; venation brochidodromous. **Inflorescence** terminal; bract and bracteoles absent. **Calyx** 0.3–0.5 × 0.3–0.4 cm, truncate to minutely 5-dentate, purple, sparsely lepidote. **Corolla** ca. 2.5–4.7 × 0.9–1.2 cm, lilac to wine, externally densely pubescent, with glandular and simple trichomes; anthers ca. 0.2 cm long, di-thecae, dorsal filaments 2.1–2.3 cm long, ventral filaments 1.7–1.8 cm long, staminode ca. 3.0 cm long; ovary oval, 0.2 × 0.1 cm, glabrous, style ca. 2.5 cm long, stigma ca. 0.2 cm long. **Capsule** 6.1–7.3 × 4.7–5.5 cm, elliptic to oblong-elliptic, flattened, woody, base attenuate, apex rounded, surface lepidote, margin slightly undulate, without lenticels. **Seeds** not seen.

**Habitat and Distribution:** Endemic to the Brazilian Northeast, where it is known from two populations growing within Atlantic Forest vegetation in Bahia and Pernambuco, respectively (Gentry 1992, Lohmann 2010).

**Phenology:** Collected with flowers from May to June, and fruits in February.

**Taxonomic Notes:** *Jacaranda microcalyx* is morphologically close to *J. puberula*, with which it shares chartaceous, elliptic to obovate leaflets, terminal inflorescences, and purple calyces.



These species can be differentiated by the tetragonal branchlets (vs. cylindrical in *J. puberula*), leaflets subsessile and lepidote on both sides (vs. sessile and glabrous on the adaxial surface and densely pubescent on the abaxial surface), calyx truncate to minutely 5-dentate (Figure 5m) (vs. 5-dentate with cuneate teeth; Figure 5n), and bracts and bracteoles absent (vs. bracts and bracteoles lanceolate).

**Examined Material:** BRAZIL. PERNAMBUCO: Ipojuca, Engenheiro Maranhão, 10.VI.1967, fl., Andrade-Lima 67-5032 (IPA 16223).

**Additional Examined Material:** BRAZIL. BAHIA: (Type Specimen) Ubaitaba-Itacaré, 14°20'S 39°20'W, 09.II.1985, fl. And fr., A.L. Gentry & E. Zardini 49962 (MO).

15. *Jacaranda puberula* Cham. *Linnaea*, 7: 550, 1832. Figure 5 n.

**Tree**, 8 m; branchlets cylindrical, striated, with lenticels, pubescent, with simple trichomes. **Leaves** bipinnate; petiole 4.6–5.8 cm long, pubescent, with simple trichomes; pinnules sessile, chartaceous, 1.8–5.5 × 1.0–2.7 cm, obovate to elliptic, base cuneate, apex rounded to attenuate, margin entire to irregularly serrate, slightly discolored, adaxial surface glabrous, abaxial surface densely pubescent, with simple trichomes; venation brochidodromous. **Inflorescence** terminal; bracts and bracteoles ca. 0.2 cm long, lanceolate. **Calyx** 0.6–1.8 × 0.3–0.7 cm, 5-dentate, teeth cuneate, purple, sparsely pubescent, with simple trichomes. **Corolla** 4.4–6.3 × 0.8–1.1 cm, lilac, externally pubescent, with glandular and simple trichomes; anthers ca. 0.2 cm long, di-thecae, dorsal filaments 2.2–2.3 cm long, ventral filament ca. 1.9 cm long, staminode ca. 3.8 cm long; disc annular, 0.1 × 0.2 cm; ovary ovate, 0.2 × 0.1 cm, glabrous, style ca. 2.5 cm long, stigma ca. 0.2 cm long, elliptic. **Fruits and seeds** not seen.

**Habitat and Distribution:** *Jacaranda puberula* is widely distributed in Atlantic Forest environments, where it occurs from Argentina and Rio Grande do Sul in Brazil, to the states of Bahia, Pernambuco, and Ceará (Gentry 1992). In the study area, it was found in Atlantic Forest vegetation.

**Phenology:** Collected with flowers from November to January.

**Taxonomic Notes:** *Jacaranda puberula* is recognized by the sessile pinnules, with abaxial surface densely pubescent and margin entire to irregularly serrate, as well as by the calyx 5-dentate with cuneate teeth (Figure 5n). The similarity and differences with *J. microcalyx* are discussed within the comments of that species.

**Selected Material:** BRAZIL. PERNAMBUCO: Bom Conselho, Fazenda Arabari, 01.XI.1966, fl., E. Tenório 66-206 (IPA 14658); Recife, Mata de Dois Irmãos, 06.I.1953, fl., Ducke & Andrade-Lima 149 (IPA 5524).

16. *Jacaranda rugosa* A.H. Gentry, *Fl. Neotrop. Monogr.*, 25(2): 102, 1992. Figure 5 o–p.

**Tree**; branchlets cylindrical, striated, without lenticels, densely pubescent, with simple trichomes. **Leaves** pinnate; petiole 2.1–2.6 cm long, pubescent, with simple trichomes; leaflets sessile, coriaceous, 1.9–4.0 × 1.7–2.6 cm, elliptic, base cuneate to obtuse, apex cuneate to rounded, margin entire and revolute, surface densely rugose, discolored, adaxial surface pubescent, with simple and short trichomes, abaxial surface densely tomentose, with simple and long trichomes; venation brochidodromous. **Inflorescence** axillar; bracts and bracteoles absent. **Calyx** 0.5–0.7 × 0.4–0.5 cm, 5-dentate, teeth cuneate to rounded, lilac, villose, with glandular and

simple trichomes, persistent. **Corolla** 3.1–5.2 × 1.2–1.7 cm, lilac with tube white, externally pubescent, with glandular trichomes; anthers ca. 0.3 cm long, di-thecae, dorsal filaments ca. 1.8 cm long, ventral filaments ca. 1.5 cm long staminode ca. 3.7 cm long; disc annular, 0.2 × 0.2 cm; ovary ovate, 0.2 × 1.1 cm, glabrous, style ca. 2.5 cm long, stigma ca. 0.2 cm long, elliptic. **Capsule** 3.5–5.3 × 2.2–4.2 cm, elliptic to wide-elliptic, flattened, base attenuate, apex rounded, surface pubescent, with simple trichomes, margin entire, without lenticels. **Seeds** 1.7 × 0.9 cm, elliptic.

**Habitat and Distribution:** *Jacaranda rugosa* was first described by Gentry (1992) based on a material from the National Park Vale do Catimbau in Buíque (Pernambuco). Even though materials from other Brazilian states have been identified as *J. rugosa*, these identifications were later shown to be erroneous. As such, *J. rugosa* is presumed endemic from the Caatinga of Pernambuco, where it is known from the National Park Vale do Catimbau exclusively.

**Phenology:** Collected with flowers from September to May, and fruits from October to January and May to June.

**Taxonomic Notes:** *Jacaranda rugosa* is recognized by the densely rugose leaflets (Figure 5p), a trait exclusive to this species in the state of Pernambuco. This species is also characterized by the calyx 5-dentate with cuneate to rounded teeth (figure 5o). The similarity and differences with *J. jasminoides* are discussed within that species comments.

**Selected Material:** BRAZIL. PERNAMBUCO: Buíque, Vale do Catimbau, 06.XI.2002, fr. and fr., A. Lopes & M.J. Santos 27 (UFP 45437).

*Sparattosperma* Martius ex Meisner, Pl. vasc. Gen. 2: 208, 1840.

Trees. Leaves palmate, 3–5 foliolate. Inflorescence terminal, a thyrs. Calyx tubular, caducous, lepidote or glabrous; corolla campanulate to infundibuliform, white to light pink; stamens included, anthers glabrous, staminode shorter than fertile stamens. Capsule linear, margin entire, without lenticels.

*Sparattosperma* is represented by two species (*S. catingae* and *S. leucanthum*), distributed from South America, both of which occur in Pernambuco (Gentry 1992).

**Identification key for *Sparattosperma* species occurring in Pernambuco state**

1. Leaves 3-foliate; calyx bilabiate with lobes cuspidate and indument lepidote; capsule with surface lepidote, without longitudinal ribs ..... *S. catingae*
- 1'. Leaves 5-foliate; calyx 2-dentate with teeth acuminate to apiculate and glabrous; capsule with surface glabrous, with longitudinal ribs ..... *S. leucanthum*

17. *Sparattosperma catingae* A.H. Gentry, *Fl. Neotrop. Monogr.*, 25(2): 115, 1992. Figure 4 n–p.

**Tree**; branchlets cylindrical, striated, with lenticels, lepidote. **Leaves** 3-foliate; petiole 1.2–3.8 cm long, sparsely pubescent, with simple trichomes; petiolule 0.5–1.8 cm long, sparsely pubescent, with simple trichomes; leaflets chartaceous, 2.3–6.8 × 2.0–3.2 cm, elliptic, base rounded, apex attenuate to obtuse, margin entire, discolored, densely lepidote on both sides; venation brochidodromous. **Inflorescence** terminal; bracts and bracteoles 0.3–1.5 cm long, foliaceous, lanceolate,

lepidote and pubescent, with simple trichomes. **Calyx** 0.8–1.5 × 0.3–0.4 cm, bilabiate, lobes cuspidate, densely lepidote. **Corolla** campanulate, 2.3–3.8 × 0.8–1.3 cm, with to light pink ribbed wine, externally glabrous; anthers ca. 0.2 cm long, dorsal filaments 1.7–1.9 cm long, ventral filaments 1.4–1.5 cm long, staminode ca. 0.2 cm long; disc annular; ovary sessile, oblong, 0.3 × 0.1 cm, with longitudinal ribs, pubescent, glandular trichomes, style 1.8–2.3 cm long, stigma ca. 0.2 cm long. **Capsule** 23.3 × 0.8 cm, slightly inflated, base obtuse, apex attenuate, surface lepidote, without longitudinal ribs. **Seeds** not seen.

**Habitat and Distribution:** *Sparattosperma catingae* has always been thought to be an endemic species of the Caatinga vegetation from the state of Bahia (Gentry 1992, Lohmann 2010). However, this study documented the occurrence of this species in the state of Pernambuco and in the Atlantic Forest domain, where this species was found in “Mata das Negras” within the municipality of Glória de Goitá.

**Phenology:** Collected with flowers in January.

**Taxonomic Notes:** *Sparattosperma catingae* differs from *S. leucanthum* by the 3-foliolate leaves (Figure 4n) (vs. 5-foliolate in *S. leucanthum*; Figure 4b), bilabiate calyx with cuspidate lobes (Figure 4o) and densely lepidote (vs. 2-dentate calyx with acuminate to apiculate teeth and glabrous; Figure 6a), and fruit with lepidote surface, without longitudinal ribs (Figure 4p) (vs. capsule with glabrous surface and with longitudinal ribs; Figure 6c).

**Examined Material:** BRAZIL. PERNAMBUCO: Glória de Goitá, Mata das Negras, 27.I.2012, fl., L.L.S. Melo & V.F. Silva 10 (HUEFS 191800).

**Additional Examined Material:** BRAZIL. BAHIA: Jequié, Fazenda Brejo Novo, 13°56'41"S 40°06'33,W, 750 m, 30.IV.2004, fr., G.E.L. Macedo 853 (PEUFR 48828); Boa Nova, 23.III.2013, fl. and fr., A.F.P. Machado et al. 1218 (HUEFS 193968).

18. *Sparattosperma leucanthum* (Vell.) K. Schum, Nat. Pflanzenfam. [Engler & Prantl], 4(3b): 235, 1894. Figure 6 a–c.

**Tree;** branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** 5-foliolate; petiole 3.2–9.3 cm long, sparsely pubescent, with simple trichomes; petiolule 0.9–3.3 cm long, sparsely pubescent, with simple trichomes; leaflets chartaceous, 5.5–8.9 × 1.5–4.8 cm, elliptic, base obtuse, apex attenuate to acuminate, margin entire, discolorous, glabrous on both sides, only with simple trichomes on axils of second veins; venation brochidodromous. **Inflorescence** terminal; bracts and bracteoles 0.2–2.1 cm long, foliaceous, lanceolate, pubescent, with glandular and simple trichomes. **Calyx** 1.1–3.0 × 0.3–0.7 cm, 2-dentate, teeth acuminate to apiculate, glabrous. **Corolla** infundibuliform, 2.2–5.0 × 0.5–0.9 cm, with to light pink ribbed wine, externally glabrous; anthers ca. 0.2 cm long, dorsal filaments 1.7–1.8 cm long, ventral filaments 1.3–1.4 cm long, staminode ca. 0.2 cm long; disc annular; ovary sessile, oblong to ovate, 0.3 × 0.1 cm, with longitudinal ribs, pubescent, glandular trichomes, style 2.0–2.7 cm long, stigma ca. 0.4 cm long. **Capsule** 23.4 × 1.1 cm, flattened, base obtuse, apex attenuate, surface glabrous, with longitudinal ribs. **Seeds** not seen.

**Habitat and Distribution:** *Sparattosperma leucanthum* is widely distributed from Venezuela and Peru to Southeastern Brazil, where it occurs in the Amazon, Caatinga, Cerrado, Atlantic Forest, and Pantanal (Gentry 1992, Lohmann 2010). In the state of Pernambuco it was found growing on roadside, in Caatinga.

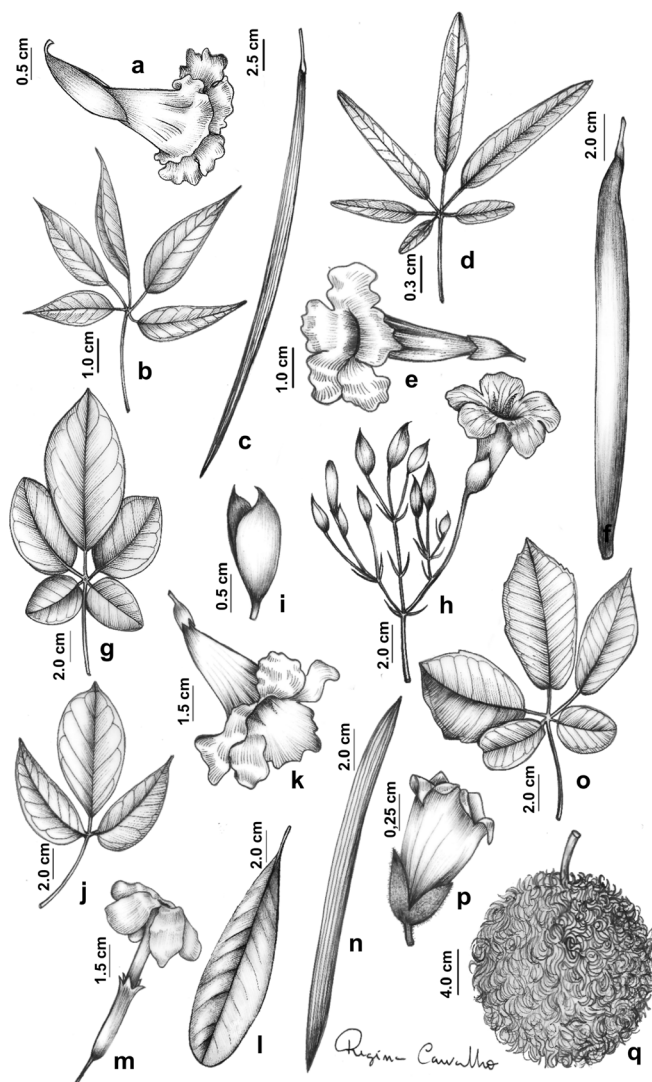
**Phenology:** Collected with flowers in October.

**Taxonomic Notes:** *Sparattosperma leucanthum* is morphologically close to the other species of the genus, being differentiated mainly by the number of leaflets (Figure 6b), shape, calyx indument, and fruit surface (Figure 6c) (see *S. catingae*).

**Examined Material:** BRAZIL. PERNAMBUCO: Canhotinho, 07.X.1956, fl., Andrade–Lima 56–2600 (IPA 11021).

**Additional Examined Material:** BRAZIL. BAHIA: Cravolândia, 30.V.1994, F. França et al. 1053 (HUEFS 16945); Nova Viçosa, Argolo, 20.VII.1988, fr., G. Hatschbach & M. Hatschbach 52260 (MO).

*Tabebuia* Gomes ex A. P. de Candolle, Biblioth. Universelle Genève, ser. 2, 17: 139, 1838.



**Figure 6.** *Sparattosperma leucanthum* (Vell.) K. Schum: a. Flower. b. Leaf. c. Fruit. *Tabebuia aurea* (Silva Manso) Benth. & Hook f. ex S. Moore: d. Leaf. e. Flower. f. Fruit. *Tabebuia elliptica* (DC.) Sandwith: g. Leaf. h. Inflorescence. i. Calyx. *Tabebuia roseoalba* (Ridl.) Sandwith: j. Leaf. k. Flower. *Tabebuia stenocalyx* Sprague & Stapf: l. Leaf. m. Flower. n. Fruit. *Zeyheria tuberculosa* (Vell.) Bureau ex Verl.: o. Leaf. p. Flower. q. Fruit.

Trees. Leaves simple or palmate, 3–6 foliolate. Inflorescence terminal, a panicle. Calyx tubular, coriaceous, lepidote; corolla infundibuliform or hipocrateriform, yellow or white; stamens included, anthers glabrous, staminode shorter than fertile stamens, ovary linear. Capsule linear, flattened or inflated. Seeds winged, wings hyaline, membranaceous.

*Tabebuia* includes 67 species that are widely distributed from Central and South America and Antilles (Gentry 1992). In Brazil, 15 species (5 endemic) are found (Lohmann 2010). Four species were documented in the state of Pernambuco: *T. aurea*, *T. elliptica*, *T. roseoalba* and *T. stenocalyx*.

#### Identification key for *Tabebuia* species occurring in the state of Pernambuco

1. Simple leaves; corolla hipocrateriform ..... *T. stenocalyx*
- 1'. Palmate leaves, 3–6 foliolate; corolla infundibuliform ..... 2
2. Corolla yellow; bracts and bracteoles absent; capsules wide with calyx caducous ..... *T. aurea*
- 2'. Corolla white; bracts and bracteoles present; capsule narrow with calyx persistent
3. Leaves 5-foliate; bracts and bracteoles filiform ..... *T. elliptica*
- 3'. Leaves 3-foliate; bracts and bracteoles triangular ..... *T. roseoalba*

19. *Tabebuia aurea* (Silva Manso) Benth. & Hook f. ex S. Moore, *Trans. Linn. Soc. London, Bot.*, 4: 423, 1895. Figure 6 d–f.

**Tree**, 3–10 m; branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** palmate, 5–6 foliolate; petiole 1.4–8.7 cm long, glabrous; petiolule 1.0–4.4 cm long, glabrous; leaflets coriaceous, 5.3–22.3 × 0.5–9.1 cm, elliptic, oblong-elliptic or lanceolate, base rounded to obtuse, apex rounded to cuneate, margin entire, concolorous, glabrescent to lepidote on both sides; venation brochidodromous. **Inflorescence** a panicle; bracts and bracteoles absent. **Calyx** 0.8–1.7 × 0.5–0.7 cm, irregularly dentate, teeth cuneate, light brown to yellow, densely lepidote, with sparsely distributed glands, caducous. **Corolla** infundibuliform, 4.3–8.1 × 1.1–1.7 cm, yellow, externally glabrous; anthers ca. 0.3 cm long, dorsal filaments 1.9–2.1 cm long, ventral filaments 1.6–1.7 cm long, staminode ca. 0.3 cm long; ovary 0.3 × 0.1 cm, densely lepidote, style 2.8–3.0 cm long, stigma ca. 0.2 cm long. **Capsule** 10.3–20.3 × 1.7–3.7 cm, flattened, base attenuate, apex acute, surface densely lepidote, margin entire, without lenticels. **Seeds** 1.7–1.9 × 3.5–4.1 cm, oblong-elliptic.

**Habitat and Distribution:** *Tabebuia aurea* is widely distributed in South America, occurring in dry forests and savannas of Argentina, western Bolivia, and occurring disjunctly in southern Suriname (Gentry 1992). In Brazil it is found in all phytogeographical domains extending from the Amazon to Paraná (Lohmann 2010). Due to its exuberant yellow tree-top while flowering, it is widely used in public ornamentation and landscaping. In the state of Pernambuco it is found in Caatinga environments, with altitudes varying from 300–500 m.

**Phenology:** Collected with flowers from September to March, and fruits from October to December.

**Taxonomic Notes:** *Tabebuia aurea* is easily recognized by the yellow corollas (the only species of *Tabebuia* in Pernambuco with this character), calyx densely lepidote, with sparse glands, and wide variation in leaf shape (Figure 6e).

**Selected Material:** BRAZIL. PERNAMBUCO: Águas Belas, Território Indígena Fulni-ô, 01.X.2015, fl., W. Torres et al 213 (IPA 91002); Floresta, Riacho Caraibas, 08°43'09"S 38°29'39,39"W, 354 m, 11.X.2012, fl. and fr. A.C.P. Oliveira & N.M. Almeida 1979 (HVASF 18529).

20. *Tabebuia elliptica* (DC.) Sandwith, *Candollea*, 7: 253, 1937. Figure 6 g–i.

**Tree**, 4–5 m; branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** palmate, 5-foliate; petiole 1.5–10.2 cm long, sparsely pubescent, with simple trichomes; petiolule 0.6–2.5 cm long, sparsely pubescent, with simple trichomes; leaflets coriaceous, 3.3–11.2 × 2.8–7.1 cm, elliptic to oblong-elliptic, base rounded to truncate, apex acuminate to cuspidate, margin entire, concolorous, lepidote on both sides; venation brochidodromous. **Inflorescence** a panicle; bracts and bracteoles 0.5–1.9 cm long, filiform. **Calyx** 1.1–2.3 × 0.4–1.0 cm, 2–3 lobed, lobes acuminate, green, densely lepidote, persistent. **Corolla** infundibuliform, 4.3–7.2 × 1.2–1.7 cm, white, internally yellow and white tube, externally glabrous; anthers ca. 0.3 cm long, dorsal filaments 1.6–1.7 cm long, ventral filaments 1.4–1.5 cm long, staminode ca. 0.3 cm long; disc annular; ovary 0.4 × 0.1 cm, lepidote, style 2.1–3.2 cm long, stigma ca. 0.2 cm long. **Capsule** 8.7–20.9 × 0.3–0.7 cm, flattened, base and apex acute, glabrous, with longitudinal ribs, margin entire, without lenticels. **Seeds** 0.5–1.4 × 0.4–0.5 cm, oblong-elliptic.

**Habitat and Distribution:** *Tabebuia elliptica* is endemic to Brazil, where it occurs in “restingas” and Atlantic Forest vegetation, from Paraíba to Espírito Santo (Lohmann 2010). In Pernambuco it was found in the Atlantic Forest.

**Phenology:** Collected with flowers from December to March, and fruits in December.

**Taxonomic Notes:** *Tabebuia elliptica* is morphologically similar to *T. roseoalba*, with which it shares the cylindrical branchlets, leaflets that are concolorous, elliptic to oblong-elliptic, with entire margins (Figure 6g and 6j), infundibuliform and glabrous corollas (Figure 6h and 6k). These species are distinguished by the leaves 5-foliate (Figure 6g) (vs. 3-foliate in *T. roseoalba* (Figure 6j), bracts and bracteoles filiform (Figure 6h) (vs. triangular and numerous bracts and bracteoles), calyx 2–3 lobed acuminate (Figure 6i) (vs. irregularly lobed with cuneate lobes (Figure 6k), and corolla white externally, with yellow internally and white tube (vs. white externally, yellow internally and pink tube).

**Selected Material:** BRAZIL. PERNAMBUCO: Aldeia, 17.III.1952, fl., D. Andrade-Lima 52–1006 (IPA 2488); Goiana, 28.XII.1965, fl. and fr., D. Andrade-Lima 65–4344 (IPA 13709).

21. *Tabebuia roseoalba* (Ridl.) Sandwith, *Kew Bull.*, 9: 597, 1955. Figure 6 j–k.

**Tree**, 8–10 m; branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** palmate, 3-foliate; petiole 1.3–10.3 cm long, glabrous; petiolule 0.3–2.9 cm long, glabrous; leaflets chartaceous, 3.4–11.2 × 1.5–9.1 cm, elliptic to oblong-elliptic, base obtuse to cuneate, apex acuminate, margin entire, concolorous, glabrous on both sides; venation brochidodromous. **Inflorescence** a panicle; bracts and bracteoles 0.3–0.7 cm long, triangular, numerous.



**Calyx** 0.8–2.3 × 0.3–0.7 cm, irregularly lobed, lobes cuneate, brown, densely lepidote, persistent. **Corolla** infundibuliform, 2.9–6.1 × 0.5–1.3 cm, white, internally yellow and pink tube, externally glabrous; anthers ca. 0.3 cm long, dorsal filaments 1.5–1.6 cm long, ventral filaments 1.3–1.4 cm long, staminode ca. 0.2 cm long; disc annular; ovary 0.4 × 0.1 cm, densely lepidote, style 2.4–2.7 cm long, stigma ca. 0.2 cm long. **Capsule** 13.2–28.8 × 0.3–0.7 cm, flattened, base obtuse, apex acute, lepidote, margin entire, without lenticels. **Seeds** 0.4–0.5 × 0.2–0.3 cm, oblong–elliptic.

**Habitat and Distribution:** *Tabebuia roseoalba* occurs in dry forests of Paraguay, Bolivia, and disjunctly in Peru (Gentry 1992). In Brazil, it is distributed through most of the national territory (except the southern region), in the areas of Caatinga, Cerrado, and Atlantic Forest vegetation (Lohmann 2010). In the state of Pernambuco it occurs in the Atlantic Forest domain.

**Phenology:** Collected with flowers from November to June, and with fruits in January and February.

**Taxonomic Notes:** *Tabebuia roseoalba* can be recognized by the leaves 3-foliolate (Figure 6j), triangular and numerous bracts and bracteoles, and calyx irregularly lobed with cuneate lobes (Figure 6k). This species is morphologically similar to *T. elliptica*. The similarities and differences among those species are discussed under that species comments.

**Selected Material:** BRAZIL. PERNAMBUCO: Ipojuca, Sítio Oiteiro, 26.V.2012, fl., M.C. Moraes s.n. (IPA 88620); Olinda, São Bento, 01.I.1915, fl. and fr., B. Pickel 434 (IPA 4928).

22. *Tabebuia stenocalyx* Sprague & Stapf, *Bull. Misc. Inform. Kew*, 1910: 196, 1910. Figure 6 l–n.

**Tree;** branchlets cylindrical, striated, with lenticels, glabrous. **Leaves** simple; petiole 0.3–3.2 cm long, glabrous; leaves coriaceous, 12.1–23.3 × 2.9–9.1 cm, obovate to oblong–obovate, base attenuate, apex cuneate to attenuate, margin entire, concolorous, lepidote on both sides; venation brochidodromous. **Inflorescence** a panicle; bracts and bracteoles 0.5–0.9 cm long, filiform. **Calyx** 1.5–4.7 × 0.3–0.6 cm, 5-dentate, teeth acute to cuneate, green, densely lepidote with sparse glands, persistent. **Corolla** hipocrateriform, 7.2–8.5 × 0.3–0.7 cm, white, externally glabrous; anthers ca. 0.3 cm long, dorsal filaments ca. 1.1 cm long, ventral filaments ca. 0.8 cm long, staminode ca. 0.3 cm long; disc cupular; ovary 0.5 × 0.1 cm, densely lepidote, style ca. 1.2 cm long, stigma ca. 0.2 cm long. **Capsule** 10.8–15.3 × 0.7–1.0 cm, inflated, base and apex cuneate, glabrous, with longitudinal ribs, margin entire. **Seeds** not seen.

**Habitat and Distribution:** *Tabebuia stenocalyx* is found in wet areas of the Guyana, Venezuela, and Brazil, where it occurs in the Atlantic Forest of Bahia, Espírito Santo, and Rio de Janeiro (Gentry 1992, Lohmann 2010). This species represents a new record for Pernambuco, where it is found in Atlantic Forest vegetation.

**Phenology:** Collected with flowers in September and December, and fruits in February.

**Taxonomic Notes:** *Tabebuia stenocalyx* is often confused with *T. obtusifolia*, with which it shares simple obovate to oblong–obovate leaves (Figure 6l). However, these species can be separated by the long (> 7.2), 5-dentate tubular calyx (Figure 6m) (vs. campanulate and bilabiate calyx in *T. obtusifolia*), and corolla hipocrateriform (Figure 7m) (vs. corolla infundibuliform).

**Selected Material:** BRAZIL. PERNAMBUCO: Recife, Beberibe, IX.1937, fl., Vasconcelos–Sobrinho s.n. (IPA 542); Recife, Dois Irmãos, 14.II.1952, fr., D. Andrade–Lima 52–979 (IPA 2450).

*Zeyheria* Martius, Nov. gen. Spec. pl.

The genus includes two species (*Z. montana* and *Z. tuberculosa*) distributed in Bolivia and Brazil (Gentry 1992). In Pernambuco it is represented by *Zeyheria tuberculosa*.

23. *Zeyheria tuberculosa* (Vell.) Bureau ex Verl., *Vidensk. Meddel. Dansk Natuhist. Foren. Kjøbenhavn*, 1863: 115, 1863. Figure 6 o–q.

**Tree or shrubs;** branchlets subtetragonal, striated, without lenticels, densely tomentose, with stellate trichomes. **Leaves** palmate 5-foliolate; petiole 5.6–18.2 cm long, densely tomentose, with stellate trichomes; petiolule 0.2–0.6 cm long, densely tomentose, with stellate trichomes; leaflets chartaceous, 3.4–16.7 × 2.1–6.3 cm, elliptic to obovate, base truncate or cordate, apex acuminate, margin entire, rarely irregularly serrated, strongly discolorous, densely tomentose on both sides, with stellate and dendritic trichomes, adaxial surface dark green, rugose, abaxial surface whitish; venation brochidodromous. **Inflorescence** a panicle, terminal; bracts and bracteoles 1.1–2.7 cm long, narrow–elliptics. **Calyx** cupular, 0.3–0.5 × 0.3–0.4 cm, bilabiate to truncate, lobes rounded, dark brown, densely tomentose, with stellate trichomes, caducous. **Corolla** wide–campanulate, 1.1–1.6 × 0.7–1.0 cm, externally brown, internally orange, externally densely tomentose, with stellate trichomes; stamens exserted, anthers ca. 0.1 cm long, glabrous, dorsal filaments 0.9–1.0 cm long, ventral filaments 0.8–0.9 cm long, staminode shorter than fertile stamens, ca. 0.2 cm long; ovary stipitate, obovate, 0.2 × 0.2 cm, pubescent, stellate trichomes, style 0.9–1.0 cm long, stigma ca. 0.2 cm long. **Capsule** 8.3–12.5 × 8.1–12.3 cm, wide–elliptic, inflated, base and apex rounded, muricate, without lenticels. **Seeds** not seen.

**Habitat and Distribution:** *Zeyheria tuberculosa* occurs in all states of Northeastern and Southeastern Brazil, growing disjunctly in Bolivia in areas of Caatinga, Cerrado, and Atlantic Forest (Gentry 1992). In Pernambuco it was found growing in Atlantic Forest vegetation, growing up to 1100 m.

**Phenology:** Collected with flowers from April to July, and fruits in February.

**Taxonomic Notes:** *Zeyheria tuberculosa* can be easily recognized by the strongly discolorous 5-foliolate leaves (Figure 6o), corolla wide–campanulate (Figure 6p), brown externally and orange internally, and the wide–elliptic fruit with muricate surface (typical of the genus; Figure 6q). This species is commonly known as “ipê felpudo” or “bolsa de pastor.”

**Selected Material:** BRAZIL. PERNAMBUCO: Lagoa dos Gatos, Serra do Urubu, 22.II.2011, fr., F. Gadelha 10 (HUFN 15835); Arcoverde, Serra do Mimoso, 20.IV.1996, fl., A.M. Miranda et al. 2349 (HUEFS 97160).

## Discussion

In this study we documented 23 species of the *Tabebuia* alliance and tribe Jacarandae in Pernambuco, distributed among seven genera, i.e., *Cybistax*, *Godmania*, *Jacaranda*, *Handroanthus*, *Sparattosperma*, *Tabebuia*, and *Zeyheria*. Of these, *Handroanthus* (8 spp.), *Jacaranda* (6 spp.), *Tabebuia* (4 spp.), and *Sparattosperma* (2 spp.) are the most diverse.



The genera *Cybistax*, *Godmania*, and *Zeyheria* are represented by a single species each (Table 1). The most frequent species in the area are: *Handroanthus impetiginosus* (Mart. ex DC.) Mattos and *Tabebuia aurea* (Silva Manso) Benth. & Hook f. ex S. Moore.

In the checklist of the Bignoniaceae from Brazil, Lohmann (2010) documented 16 species of the Tabebuia alliance and tribe Jacarandae for the state of Pernambuco. The occurrence of all of these taxa in Pernambuco was confirmed during this study. Besides those, *Cybistax antisiphilitica* (Mart.) Mart., *Handroanthus capitatus* (Bureau & K. Schum) Mattos, *Handroanthus umbellatus* (Sond.) Mattos, *Jacaranda cuspidifolia* Mart., *Sparattosperma catingae* A.H. Gentry, and *Tabebuia stenocalyx* Sprague & Stapf were documented for the first time in the state of Pernambuco, thus constituting new records.

Furthermore, *S. catingae*, previously thought to be endemic to the Caatinga of Bahia, was documented for the first time in the Atlantic Forest domain (Table 1). *Jacaranda cuspidifolia* was also documented for the first time in Northeastern Brazil. In addition, *Jacaranda rugosa* was confirmed as an endemic species of Pernambuco caatinga, being found only in the National Park Vale do Catimbau (see the comments of this species).

The diversity map (Figure 2) indicated that the State Park Dois Irmãos (Figure 2f), the National Park Vale do Catimbau (Figure 2d), Ecological Reserve Carnijó (Figure 2e), and the National Forest Negreiros (Figure 2a) represent the three most diverse areas in the state of Pernambuco. These areas are located within conservation units, highlighting the importance of these units for species preservation. The diversity map also highlights other priority areas for conservation within the state, all of which include high diversity but are outside of conservation units, such as Triunfo (Figure 2c) and Petrolina (Figure 2b).

## Supplementary material

The following online material is available for this article:

Appendix 1 - List of examined materials

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## Author Contributions

Swami Costa: Data collection; species identification; data analysis and interpretation; manuscript preparation; and, study design.

Lúcia G. Lohmann: Data interpretation; species identification; manuscript preparation and critical revision, adding intellectual content.

Maria Teresa Buril: Data collection; manuscript preparation and critical revision, adding intellectual content; and study design.

## Conflict of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Unfolding additional massive cutback effects of the Native Vegetation Protection Law on Legal Reserves, Brazil

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**Abstract:** The Native Vegetation Protection Law – 2012 - (NVPL) is the main Brazilian regulation for protecting native vegetation (NV) on private land. The NVPL, currently in the implementation phase, reduced Legal Reserves (LR) requirements compared to its previous version, the 1965's Forest Act (FA), through several legal mechanisms. Among them, Article 68 (Art.68) exempts landholders from LR obligations if NV was converted without offending the legislation in place at the time of the conversion. The technical implementation of Art. 68 is controversial and its effects are still unknown. We developed a model to estimate the effects of Art.68 on LR using São Paulo State (Brazil) as case study. We analyzed former environmental laws to identify key periods in which NV preservation requirements had changed. After, we searched for past spatial data on NV cover with sufficient accuracy for each legal benchmark. Combining legal benchmarks with spatial data, we created two scenarios for Art.68 effects, plus a baseline scenario. The first scenario considered a single legal benchmark, the 1965's FA (scenario "1965"), while the other included the 1989 Cerrado's protection Federal Law as a second benchmark (scenario "1965/89"). The baseline scenario did not include Art.68 effects. Scenario "1965" reduced LR deficits in 49% compared to the baseline scenario, waiving landholders from restoration or offsetting needs in 423 thousand hectares (kha) of NV. Scenario "1965/89" waved 507 kha of NV from restoration needs and represented a 59% reduction in LR deficit compared to the baseline scenario. The LR reduction by scenario "1965/89" assumed particular importance considering that the additional cutback was concentrated on Cerrado, an already very fragmented and impacted region. Together with reductions from other NVPL rules, the additional effects of Art. 68 unfolded great concerns about the role of LR as a tool for NV preservation on private land, threatening governmental restoration commitments, and pointing that conservation command and control approaches should be complemented with incentive policies to achieve the desired and committed standards.

**Keywords:** Native Vegetation, Biodiversity Conservation, New Forest Act, São Paulo State, Environmental Regularization Program.

## Revelando reduções adicionais de Reserva Legal da Lei de Proteção da Vegetação Nativa, Brasil

**Resumo:** A Lei de Proteção da Vegetação Nativa – 2012 - (LPVN) é a principal lei brasileira para proteção da vegetação nativa (VN) em terras privadas. A LPVN, atualmente em fase de implementação, reduziu os requerimentos de Reserva Legal (RL) presentes no Código Florestal (CF) de 1965 através de uma série de mecanismos legais. Entre eles, o Artigo 68 (Art.68) elimina a obrigação de recomposição ou restauração da VN convertida sem violação

da lei vigente à época da conversão. O Art.68 é um dos mais controversos mecanismos da LPVN e cujos efeitos ainda não são conhecidos. Nós desenvolvemos um modelo para estimar os efeitos do Art.68 utilizando o estado de São Paulo, Brasil, como estudo de caso. Para isso, levantamos marcos legais nos quais os requerimentos mínimos de preservação da VN foram alterados. Em seguida, levantamos a existência de dados espaciais da cobertura de VN com a precisão necessária para cada marco legal. Combinando os marcos legais com os dados espaciais encontrados, criamos dois cenários incluindo os efeitos do Art.68 e um cenário linha de base para controlar tais efeitos. O primeiro cenário considerou apenas um marco legal, o CF de 1965 (cenário “1965”), enquanto o segundo incluiu a Lei Federal de proteção ao Cerrado de 1989 (cenário “1965/89”). O cenário “1965” reduz os déficits de RL em 49% quando comparado ao cenário de base, dispensando os proprietários de terra da obrigação de restaurar ou recompor 423 mil hectares (kha) de VN. O cenário “1989/65” dispensa da obrigação de restauração ou recomposição 507 kha de VN, representando uma redução de 59% do déficit de RL em comparação ao cenário base. A redução apresentada pelo cenário “1965/89” assume grande importância uma vez que se concentra em áreas de Cerrado, bioma já extremamente fragmentado e impactado. Em conjunto com as reduções promovidas por outros Artigos da LPVN, estes efeitos revelam grande preocupação sobre o papel das RL como uma ferramenta para a conservação de VN em terras privadas, ameaçando compromissos governamentais de restauração e indicando que estratégias de comando e controle deverão ser complementadas por políticas de incentivo para atingir os objetivos de conservação desejados.

**Palavras-chave:** *Vegetação Nativa, Conservação da Biodiversidade, Novo Código Florestal, Estado de São Paulo, Programa de Regularização Ambiental.*

## Introduction

Native vegetation (NV) on private land is worldwide recognized as essential for biodiversity conservation, climate regulation and maintenance of ecosystem services (Norton 2001, Doremus 2003, Tikka & Kauppi 2003, Nunes et al. 2016). Strategies that balance NV conservation and economic activities, such as agriculture and animal production, are essential to involve landholders in conservation efforts (Harvey et al. 2008, Blom et al. 2010, Pacheco et al. 2017).

In Brazil, 54% of the remaining NV occurs in private lands (Sparovek et al. 2015). The “Native Vegetation Protection Law” (NVPL) (Brasil 2012) is the main national regulation for protecting NV on private land (Soares-Filho et al. 2014, Brancalion et al. 2016, Garcia et al. 2016). The NVPL replaced the previous Brazilian Forest Act (FA) (Brasil 1965) through a long process of disputes among multiple stakeholders until its approval by the National Congress in 2012 (Metzger et al. 2010, Nazareno et al. 2012, Sparovek et al. 2016). The NVPL’s implementation is behind schedule after several delays of initially foreseen deadlines. For instance, the entering-step of the NVPL, the “Rural Environmental Registry” (Portuguese acronym: CAR), was postponed three times and, currently, landholders have until December 2019 to register to CAR and, this way, benefit from the “Environmental Regularization Program” (Portuguese acronym: PRA). PRA defines several rules that reduce protection of NV to promote easier compliance comparing the 2012 NVPL with the previous FA (1965). Some States have not defined the PRA regulations so far (SFB 2018).

The NVPL kept the Legal Reserve (LR) from the 1965’s FA (Brasil 1965), one of the main mechanisms to foster conservation on private lands. LR corresponds to a land fraction of the farm for NV maintenance but allow NV sustainable management. Its size depends on the biome and the vegetation type, varying from 20 to 80% of the farm (Brasil 2012). LR areas have a crucial role in biodiversity conservation (Beca et al. 2017, Farah et al. 2017) and on the provision of environmental services, including water and soil protection, carbon storage, pollination, and agricultural pest control (Brancalion et al. 2016, Garcia et al. 2016,

Saturni et al. 2016, Librán-Embid et al. 2017, Oakleaf et al. 2017). Further, LR vegetation patches act as stepping-stones between public Protected Areas (Conservation Units). Since Brazilian Conservation Units are usually far from each other, the LR network is essential to functionally connect landscapes (Metzger 2001, Ribeiro et al. 2009, Tambosi et al. 2013).

However, the NVPL reduced substantially the total amount of protected NV in Brazil by granting partial amnesty for illegal deforestation prior to 2008 and allowing the reduction of the required LR in several situations (Brancalion et al. 2016). Previous studies suggested that NVPL reduced 37 Mha of LR total area (Sparovek et al. 2012, Freitas et al. 2017, Guidotti et al. 2017). Those estimations did not consider the controversial Article 68 (Art.68), which promotes additional LR reduction. Art.68 specifies that if the NV was converted without offending the legislation effective at the time of the conversion, the landholder should be waived from LR obligation. The effects of Art.68 are still unknown and may represent a huge cutback in NV protection, mainly in areas of long-established agricultural production, therefore also more degraded. Art.68 effects, consequently, may represent a major threat to the maintenance of ecosystem services and biodiversity protection. Further, knowing the effects of Art.68 over LR deficits may guide States strategies for the “Program for Recovery of Degraded Areas” (Portuguese acronym: PRADA), another requirement from the NVPL. It can drive, for example, policies to promote LR compensation enabling a market for the trade of NV surpluses that also result in additional environmental protection (May et al. 2015).

We developed a model to estimate the potential effects of Art.68 on LR using São Paulo State, Brazil, as a case study. São Paulo represents an extreme situation of a State with a long history of consolidated agriculture and early deforestation, what turns it into a valuable proxy of Art.68 maximum effects. Similar conditions would apply to other long-time consolidated agriculture areas in South, Southeast, and part of Central West Brazilian regions (Barretto et al. 2013). We considered two scenarios of law interpretation and application. For this, we analyzed the



historical development of the Brazilian environmental legislation and identified periods in which accurate spatial data of NV were available.

## Material and Methods

The effects of Art.68 were determined in two steps. First, the percentage of NV in 2008 was determined for each farm and checked against the LR requirement of the NVPL in São Paulo State (i.e., 20%). The 2008 benchmark refers to the date set by the NVPL to exempt of restoration requirements or offsetting deforestation in disagreement with the 1965 FA. If this percentage was not reached, the farm was considered as non-compliant and potentially eligible to access Art.68 benefits. In a second step, the model verified if the percentage of past NV decreased between the chosen legal benchmark and 2008. If a reduction was observed, the farm loses the Art.68 benefit and the LR deficit was considered to be the same computed for 2008 (described in the first step). If no reduction was observed, the farm was considered eligible to access Art.68 benefit and LR deficit was computed as the area of 1965 NV subtracted by the area of 2008 NV.

The model takes into account that “Areas of Permanent Protection” (APP); i.e. areas that protect fragile environments such as hill tops, steep slopes and riparian forests; can be computed into the LR percentage as established by the NVPL. A detailed description of the modeling procedure and the combination with previous NVPL models is described in Supporting Information (Tables S1, S2, S3). It is also important to note that the model does not include farms smaller than four Fiscal Modules (FM) since NVPL discharge them from LR restoration. In São Paulo State, each FM varies from 5 to 40 ha (INCRA, 2013).

To model Art.68 we first analyzed previous environmental laws to identify key periods in which the minimal requirements for NV preservation in private lands had changed. This step was also important to determine the spatial scope of NV protection of past legislation. A second step was to search for past spatial data on NV cover with sufficient accuracy for each legal benchmark. Finally, we matched historical NV preservation requirements with historical spatial data on NV cover, creating two scenarios for Art.68 application and one baseline scenario.

### 1. Native vegetation spatial data availability and previous preservation requirements

Brazilian legal requirements for NV preservation in private lands changed over time, determining different levels of protection through LR (Table 1). The processes of checking the spatial scope of such legislation was ambiguous by the various terminologies and definitions used to describe NV over time (e.g. “matta”, “arvoredo”). These differences lead to several possible legal interpretations about the past legislation amplitude. For instance, it is possible to interpret that all NV types, or that only the forested ones, were protected in the legislations previous to 1989.

Our search for past NV cover databases showed that the first spatial data with the necessary accuracy and precision for the NVPL modeling was generated in the 1960s. This database consists of maps made by the “Brazilian Institute of Geography and Statistics” (IBGE) at the scale 1:50,000 (IBGE 1965). Thus, we do not have a spatial solution for NVPL spatial explicit modeling of Art.68 before the 1965’s FA. To estimate Art.68 effects for previous laws, it would be necessary to rely

on spatial equation models (Dias et al. 2016) and, by these, move from a spatial explicit solution to a still comprehensive, but probabilistic approach. Another option would be to keep the spatial explicit approach but narrow it to spatial data documents that are not comprehensively available, such as old farms’ sketches, in some cases only available in old registry office books. In the first case, we would not reach the accuracy to access precise data for each farm, limiting this approach for implementation purpose by the responsible authority. Such a reference may be useful for regional planning or assessment, but improper for farm-level decisions. For the second, we would depend on one-by-one document analysis, where available, that would result in a partial and very time-consuming approach impossible to be applied for the whole State in a modelling research project.

Therefore, we adopted the 1965’s Brazilian FA as the initial legal benchmark, disregarding all previous laws, and addressing the study with a spatial explicit large-scale model solution. The FA from 1965 (Federal Law 4.771/1965) introduces the term “Legal Reserve” for the first time and changes the percentage of protected NV to 20%. Again, the interpretation about the comprehensiveness of NV protection given by the law is subjective, being uncertain if the protection applies to all physiognomies of NV or only to the forest types.

This outcome has a critical effect over São Paulo State “Environmental Regularization Program” (PRA) (São Paulo 2015). The State legislation indicates that in 1934, farms should keep as LR at least 25% of the existing forests. However, there is no precise spatial information on land cover available for 1934. The manual analysis based on information supplied by landholders may delay even more the implementation of the NVPL in the State, foster juridical queries and legal contests. Further, it could open an over-the-counter one by one negotiation opportunity that favors interpretation errors, administrative misconduct, and corruption. Other Brazilian States that are still deciding on how to define Art.68 interpretation rules for their PRAs should take into account the availability of accurate spatial data on the past NV cover in order design rules that allows a precise and systematic solution for the Art. 68 application.

In one of the scenarios we included the Federal Law from 1989 (Federal Law 7.803/1989) that complements the 1965 FA and reassures the protection of a 20% LR area for farms located in Cerrado regions.

### 2. Scenarios for Article 68 application

We considered two scenarios to access the effects of Art.68 over LR deficit in SP: scenario “1965” and scenario “1965/89”, plus a baseline scenario to control for such effects (Table 2). The baseline scenario includes LR reduction mechanisms that were modeled by previous studies and based in other articles of the NVPL (i.e. Art. 13, 15 and 67) but do not account for the effects of Art.68 (Sparovek et al. 2012, 2015, Freitas et al. 2017, 2016). This scenario represents a control to isolate the effects of these two possible Art.68’s interpretation rules.

For the scenario “1965”, we considered a single legal benchmark: the 1965 Brazilian FA (Table 1), acknowledging that LR requirements were applied to all types of NV and, consequently, since 1965, NV outside APPs should represent at least 20% of the farm (Table 2).

For the scenario named “1965/89”, we used two legal benchmarks: the Brazilian FA from 1965, and the 1989 Cerrado’s protection Federal law (Law n° 7.803/1989) (Table 1). In this scenario, farms with forested types of NV should comply with a 20% LR since 1965 and, for other

**Table 1.** Brazilian main native vegetation protection mechanisms related to Article's 68 modeling and availability of accurate spatial data.

Regulation	Year	Description	Data
São Paulo State Law 2.223	1927	Sets the São Paulo State Forest Service and establishes that properties with vegetation and larger than 100 ha must keep a forest cover over 10% of its area, except when it is homogeneous vegetation ("mattas" in the Portuguese original) or vegetation with spontaneous regrowth (Art. 5).	no
Federal Decree 23.793	1934	First Brazilian Forest Act. Compels landholders to protect 25% of the vegetation present in the property ("mattas" in the Portuguese original) (Art. 23).	no
Federal Law 4.771	1965	Second Brazilian Forest Act. Establishes different percentages of native vegetation protection accordingly to the region where the property is located. Landholders from the southeast, south and central-west Brazilian regions of the country must protect 20% of forests and other forms of native vegetation in their lands (Art. 16).	yes
São Paulo State Decree 49.141	1967	Sets the protection of 20% for a specific Cerrado vegetation type ("Cerradão", in the Portuguese original) (Art. 7).	yes
Federal Law 7.803	1989	Changes the Law 4.771/1965 and reassures the protection of a 20% Legal Reserve for Cerrado areas (Art. 16).	yes
Provisional Act 2.166.67	2001	Sets new limits for Legal Reserves: 80% for forests and 35% for "Cerrado" inside the Legal Amazon boundaries and 20% for forests, "Campos Gerais" and other types of native vegetation outside the Legal Amazon (Art. 16).	yes
Federal Law 11.428	2006	Atlantic Forest Protection Law. Sets special protection mechanisms for the Atlantic Forest biome.	yes
Federal Decree 6.514	2008	Environmental infractions decree. Benchmark used by the Law 12.651/2012 as a cutting line to establish differences in conservation and restoration requirements for Areas of Permanent Preservation and Legal Reserves.	yes
Federal Law 12.651	2012	Third Brazilian Forest Act named "Native Vegetation Protection Law". Sets the Legal Reserve limits of 80% for forests, 35% for "Cerrado" and 20% for "Campos Gerais" inside the Legal Amazon Boundaries and 20% for other biomes (Art. 12). Establishes several Legal Reserve reduction mechanisms (Art. 12, 13, 15, 67 and 68).	yes

**Table 2.** Description of the main requirements and data used to perform the three considered scenarios of Article 68 implementation.

Scenario	Past Native Vegetation database	Native Vegetation classification	Legal Reserve requirements
Baseline	n.a. <sup>1</sup>	n.a. <sup>1</sup>	Compliance in 2008 with the NVPL
1965	1:50000 IBGE maps <sup>2</sup>	n.a. <sup>1</sup>	NV outside APP $\geq$ 20% since 1965 Compliance in 2008 with the NVPL
1965/89	1:50000 IBGE maps <sup>2</sup>	RADAM Project maps <sup>3</sup>	NV outside APP $\geq$ 20% since 1965 for forested NV NV outside APP $\geq$ 20% since 1989 for other types of NV Compliance in 2008 with the NVPL

Notes: <sup>1</sup> n.a. = does not apply; <sup>2</sup> IBGE, 1965; <sup>3</sup> IBGE, 2015.

types of NV, with more open canopies such as savannahs (all Cerrado vegetation types, excluding the "Cerradão", which was considered as a forested vegetation) or grasslands (Campo) should comply with a 20% LR only after 1989. To identify the type of NV in each farm we used maps from the RADAM Brasil project (IBGE 2015) and classified it as "forested NV" or "other types of NV".

For scenarios "1965" and "1965/89", we used the legal interpretation which states that landholders who, at some time, have not complied with the law in force, lose the benefits from Art.68, demanding 20% of LR at current time.

These two scenarios represent the two most common interpretations of the 1965 and 1989 legal references, being the "1965/89" scenario in line with the State PRA (São Paulo 2015) and the "1965" scenario, that is more protective, in line with the interpretation of the environmental Civil Society and Public Attorney agencies (Loubet 2014, Chiavari & Lopes 2016).

## Results and Discussion

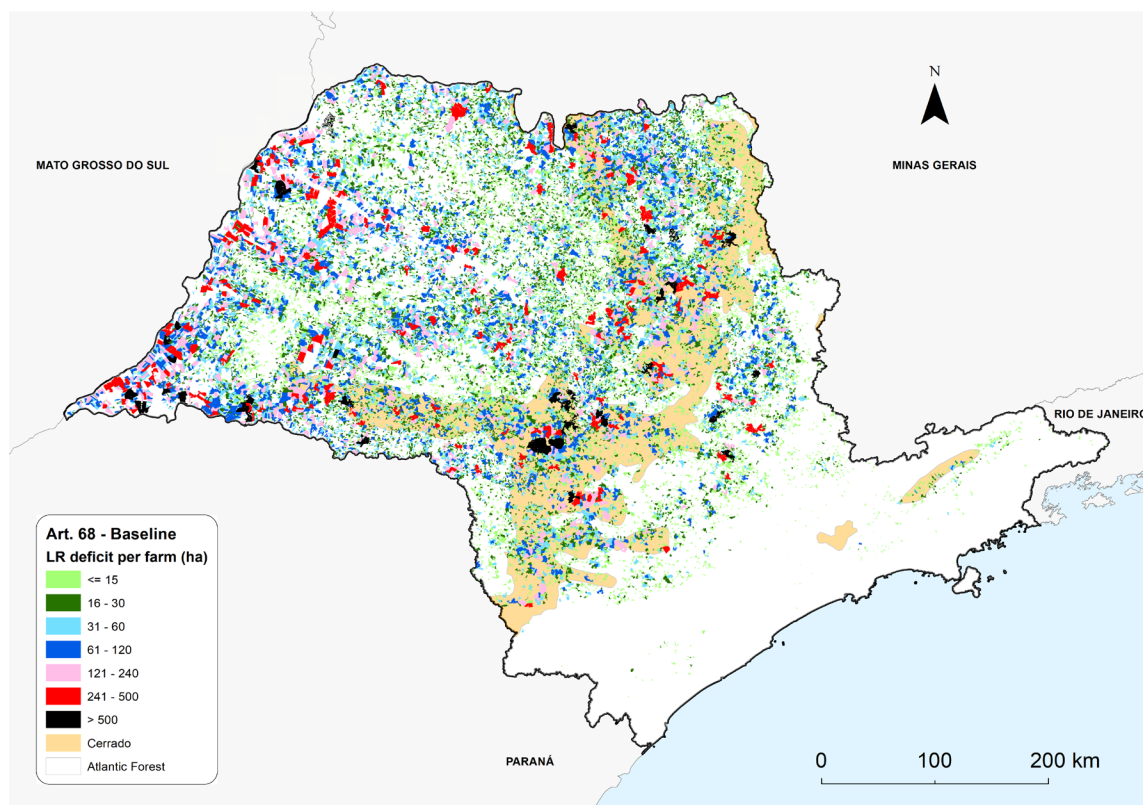
### 1. São Paulo State LR deficits

For all three scenarios, LR deficits were unevenly distributed among São Paulo State, with a higher concentration at West, Northwest and Mid-West regions (Figures 1, 2 and 3). The "baseline" scenario created a total LR deficit of 865 thousand hectares (kha) (Figure 1), of which 635 kha were located at Atlantic Forest and 230 kha in the Cerrado biome (Table 3).

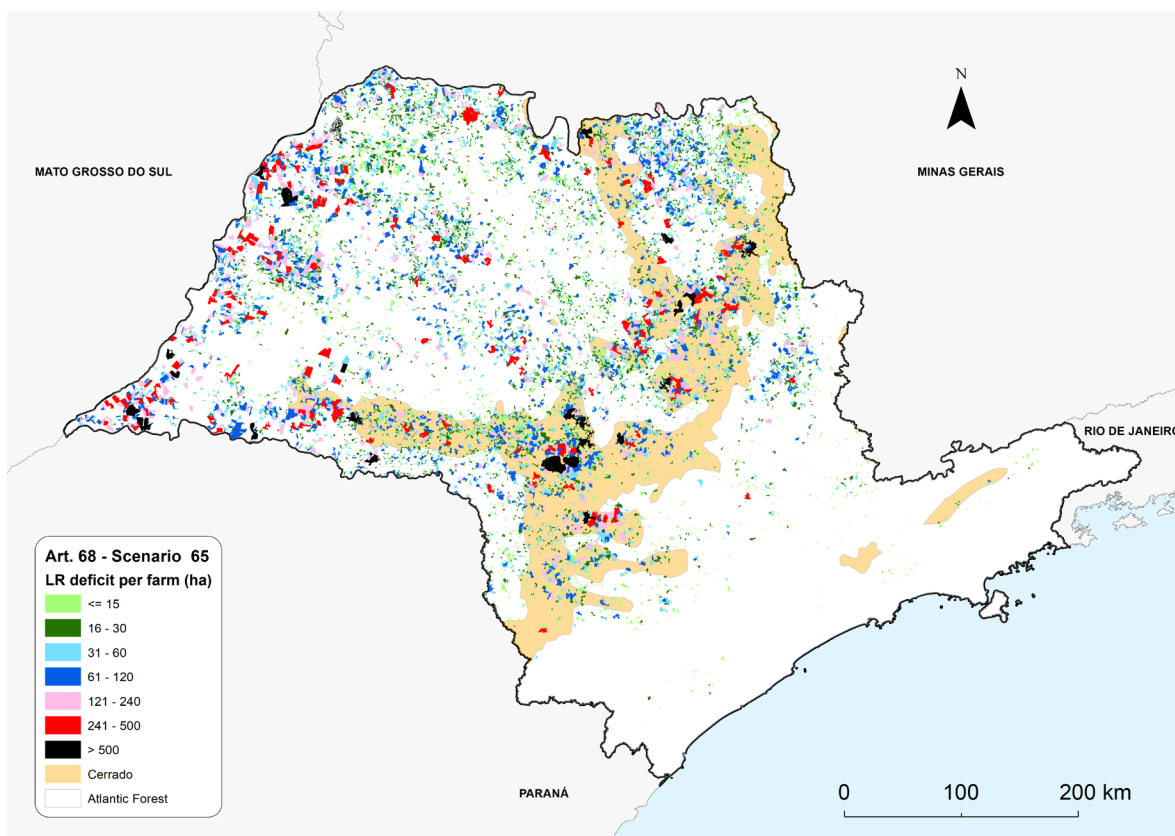
Scenario "1965" reduced LR deficits in almost 50% in relation to the baseline scenario, waiving landholders from restoration needs in 423 kha (Figure 2). Scenario "1965/89" reduced additional 84 kha from restoration needs when compared to scenario "1965", a 59% reduction in the LR deficit in relation to the baseline scenario (Figure 3).

The reduction in LR brought about by scenario "1965/89" was particularly important because the geography of the additional reduction

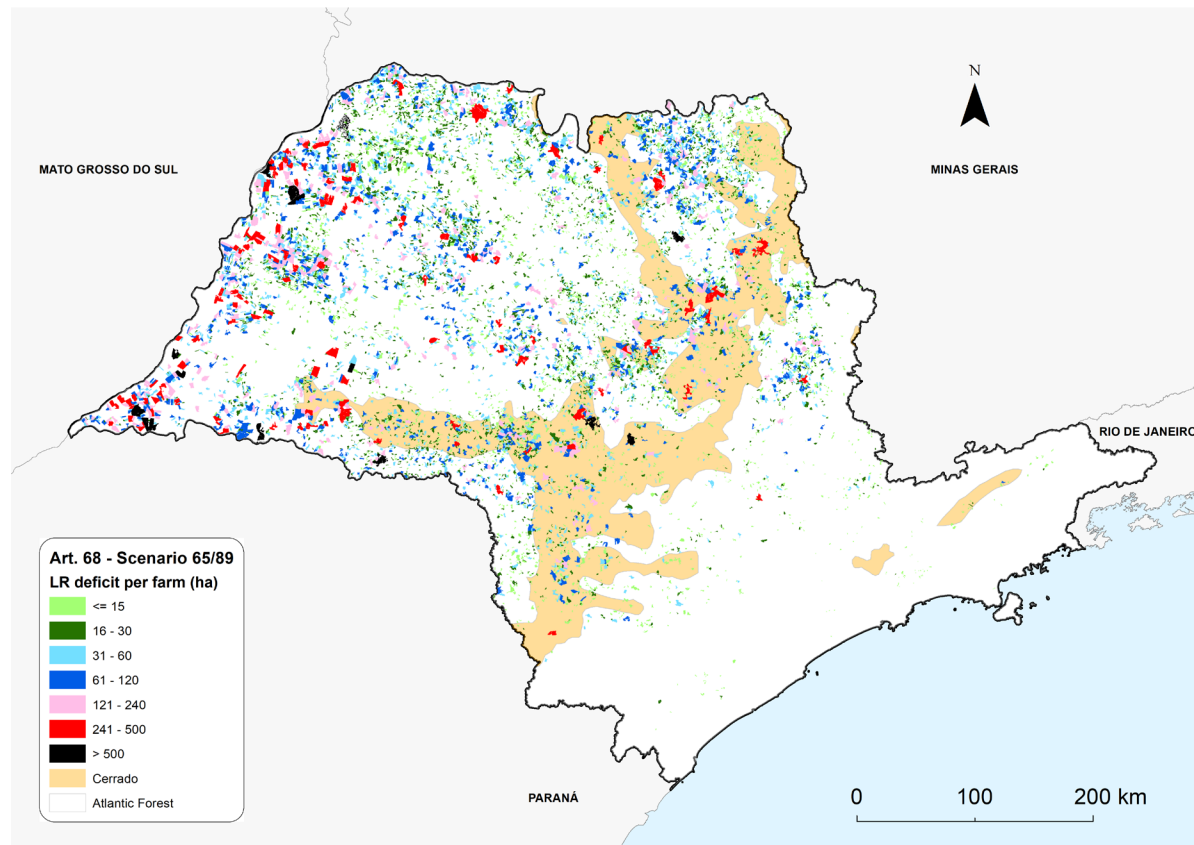
## Legal Reserves cutbacks



**Figure 1.** São Paulo State Legal Reserve deficits per farm, in hectares, for the baseline scenario (i.e. without art. 68 inclusion), total deficit of 865 thousand hectares from a total of 30,417 farms with deficit. Each polygon represents one farm.



**Figure 2.** São Paulo State Legal Reserve deficits per farm, in hectares, for the scenario 1965, total deficit of 443 thousand hectares from a total of 12,324 farms with deficit. Each polygon represents one farm.



**Figure 3.** São Paulo State Legal Reserve deficits, hectare, for the “1965/89” scenario, total deficit of 358 thousand hectares from a total of 10,477 farms with deficit. Each polygon represents one farm.

**Table 3.** Legal Reserve deficit and native vegetation surplus in thousand hectares (kha), for the three considered scenarios.

Native Vegetation Surplus <sup>1</sup> (kha)	Legal Reserve Deficit (kha) Scenario	
	Baseline	1965
776	635	294
119	230	149
895	865	443

Note: <sup>1</sup> Native Vegetation (NV) surplus accounts for the total amount of NV available for LR offsetting, including large farmlands (more than 4 Fiscal Modules) with more than 20% of NV and any existing NV fragments in small properties (less than 4 Fiscal Modules).

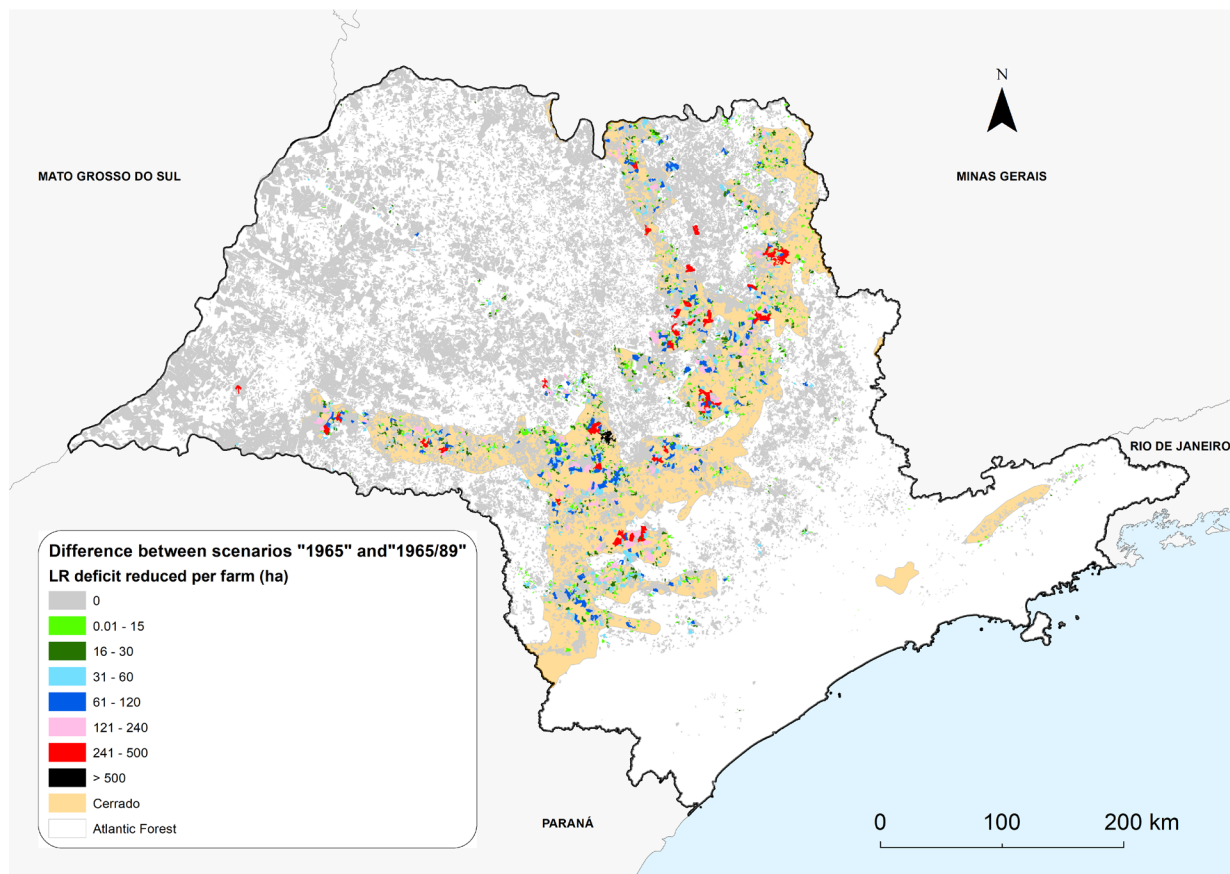
occurs on the Cerrado biome (Figure 4). While the implementation of Art.68, considering scenario “1965/89”, reduced the LR deficit in only 4% for Atlantic Forest, it reduced by 50% LR deficit in the Cerrado biome in SP (Table 3). The difference between both scenarios (“1965” and “1965/89”) can be explained by the fact that scenario “1965/89” adds a second legal benchmark to the model. In this scenario, for forested NV types, the model follows the 1965’s FA (Brasil 1965), for other NV types, it follows the Cerrado’s protection law (Brasil 1989). Thus, suppression of non-forested NV between 1965 and 1989, more common in the Cerrado biome, was exempted from LR restoration or offsetting to the percentage required by the NVPL in scenario “1965/89” (Brasil 2012).

In this period NV was converted mainly for sugarcane plantations, which was expanding quickly in the region, boosted by incentives given through the “National Alcohol Program” (Pró-Alcool) (Bastos 2007, Natale Netto 2007, Camara & Caldarelli 2016). By that time, driven by favorable soil and climate conditions, sugarcane expanded over a large extent of Cerrado NV, increasing deforestation rates at this biome (Durigan et al. 2004, Kronka et al. 2005, Takaaki et al. 2015). Considering that Cerrado has a remarkable low occurrence of protection through Conservation Units – 24 Sustainable Use Conservation Units, representing a total area of 433.674 ha; while Atlantic Forest has 142, representing a total area of 3.412.517 ha (DATAGEO 2018) - the exemption of restoration or offsetting of non-forested NV can have a negative impact over the conservation of this already very fragmented biome (Durigan et al. 2007, Strassburg et al. 2017). Presumably, this effect will also occur in other Brazilian States that had a similar history of agriculture development (e.g. Paraná, South of Minas Gerais, South of Mato Grosso do Sul).

## 2. São Paulo State native vegetation surplus

According to the NVPL, LR deficits may be restored within non-compliant farms or offset in another farm with NV surplus in the same biome. Presumably, most farmers will opt for offsetting to avoid the conversion of productive farmland to nature protection or the costs with a restoration plan (Bernasconi et al. 2016, Freitas et al. 2017).





**Figure 4.** São Paulo State Legal Reserve deficits, difference between scenarios “1965” and “1965/89”. Each polygon represents one farm.

In the Atlantic Forest, for scenarios “1965” and “1965/89”, the available NV for offsetting—NV surplus—was substantially higher than the LR total deficits (Table 3). This means that the entire LR deficit in the Atlantic Forest could be offset within the State without the need for NV restoration or conversion of productive lands. On the other hand, for Cerrado, only for scenario “1965/89” it would be possible to overcome the need for NV restoration or conversion of productive land.

However, since both biomes are already protected by NV conservation laws (São Paulo 1967, Brasil 2006), LR offsetting would not lead to additionality in nature protection. Thus, in order to increase NV conservation and the supply of ecosystem services, the only scenario that promoted these by legal enforcement was scenario “1965” and restricted to the Cerrado biome. For scenario “1965/89” it is essential to promote incentives for NV restoration, such as payments for environmental services (PES) and other policies to foster restoration of NV on private land or the creation of public owned Conservation Units.

## Conclusions

We could not identify an accurate model solution for Art.68 before 1965. The suggested solutions allow systematic analysis of Art. 68 reducing legal contests and the negative effects of subjectivity in the one-by-one analysis by governmental officials. Since the spatial data we used for 1965 NV cover in São Paulo State was based on the first nationwide aero photographs, we believe this is also the earliest date

for other Brazilian States. Thus, any attempt to apply Art.68 before the 1965’s Brazilian FA, e.g. the 1934 FA (Brasil 1934) would have to rely on probabilistic NV maps or one-by-one manual analysis of data provided by the landholders. These options would challenge the development of a large-scale and accurate tool for decision making to be used by governmental agencies during the implementation phase of the NVPL. This fact should be considered for Art.68 definition in the States PRAs, to avoid the undesired effects of unrealistic legal rules that undermine the applicability of the NVPL.

Regarding the effects of Art.68 it decreased the LR deficits between 49% and 59%. This effect adds substantially to the already important reductions caused other rules from the NVPL (Freitas et al. 2016), unfolding great concerns on the role of LR as a conservation aid for NV preservation on private land. Furthermore, we observed a higher LR deficit reduction in areas of Cerrado biome changing from scenario “1965” to “1965/89”. This biome had high deforestation rates in the past and has only a small amount of land protected by public Conservation Units.

It is very likely that the trends observed in São Paulo also apply to other States with a long history of agricultural occupation (e.g. South Region, Minas Gerais, South of Mato Grosso do Sul). In such conditions, the enforcement capacity of command and control mechanisms to promote NV preservation on private land outside APPs is currently largely overestimated by modeling due to the absence of Art. 68 effects.

In order to keep the benefits of NV restoration, the command and control approach should be complemented by incentive policies. The assessment of NVPL effects on NV conservation and the planning of NVPL implementation could substantially benefit from a national wide modelling of the effects of Art.68.

## Supplementary material

The following online material is available for this manuscript:

Table S1 - Datasets used to generate São Paulo State land tenure and boundaries map.

Table S2 - Datasets used to spatialize and classify past NV cover in the state of São Paulo.

Table S3 - Possibilities of native NV cover at the propriety (in %) along legal benchmarks and applicability of Article 68 (Federal Law n° 12.651/2012) benefits.

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## Author Contributions

Paulo André Tavares: substantial contribution in the concept and design of the study; contribution to data collection; contribution to data analysis and interpretation and contribution to manuscript preparation.

Alice Dantas Brites: substantial contribution in the concept and design of the study; contribution to manuscript preparation; contribution to data analysis and contribution to critical revision, adding intellectual content.

Gerd Sparovek: substantial contribution in the concept and design of the study; contribution to data collection; contribution to data analysis and interpretation and contribution to manuscript preparation.

Vinicius Guidotti: contribution to data analysis and interpretation and contribution to critical revision, adding intellectual content.

Felipe Cerignoni: contribution to data analysis.

Daniel Aguiar: contribution to data collection and contribution to data analysis and interpretation.

Jean Paul Metzger: substantial contribution in the concept and design of the study and contribution to critical revision, adding intellectual content.

Ricardo Ribeiro Rodrigues: substantial contribution in the concept and design of the study and contribution to critical revision, adding intellectual content.

Luís Fernando Guedes Pinto: contribution to critical revision, adding intellectual content.

Kaline de Mello: substantial contribution in the concept and design of the study and contribution to critical revision, adding intellectual content.

Paulo Guilherme Molin: contribution to data analysis and interpretation and contribution to critical revision, adding intellectual content.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Interactions between cetaceans and artisanal fishermen from Ilhéus, Bahia - Brazil

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**Abstract:** Ethnobiological studies are necessary to understand the relationships between fishers and cetaceans. The aim of this study was to describe the interactions between cetaceans and artisanal fishers and the possible conflicts that can arise as a result. Semi-structured interviews were conducted from February to September 2017 with 35 fishermen from the municipality of Ilhéus, Brazil. All fishermen reported both positive and negative interactions with *Megaptera novaeangliae*, *Tursiops truncatus* and *Sotalia guianensis* and only negative interactions with *Balaenoptera acutorostrata* (n=14) and *Stenella frontalis* (n=4). The positive interactions consisted of playful relationships and collaborative fishing. Negative interactions (bycatch, entanglement, collisions) affect both the fishermen and cetaceans. *S. guianensis* is the species most affected by bycatch in fishing nets and *M. novaeangliae* by entanglement and collision with vessels. The results of this study highlight the value of bringing together scientific and fishing communities to understand conflicts by monitoring interactions in order to assess the impact of fishing activity on cetacean populations. Fishers have a positive perception of cetaceans, know the areas where accidents occur and would like to avoid them. They can contribute to research on these animals and to the development of management plans appropriate to the local reality. This points out the need to integrate scientific and local knowledge for the conservation of cetaceans and for the sustainability of fishing practices.

**Keywords:** ethnobiology marine mammals, bycatch, entanglement, collision.

## Interações entre cetáceos e pescadores artesanais de Ilhéus, Bahia - Brasil

**Resumo:** Estudos etnobiológicos são necessários para conhecer as relações entre pescadores e cetáceos. O objetivo deste trabalho foi descrever as interações entre cetáceos e pescadores artesanais e possíveis conflitos decorrentes destas. Entrevistas semiestruturadas foram aplicadas de fevereiro a setembro de 2017 a 35 pescadores do município de Ilhéus, Brasil. Todos os pescadores relataram interações positivas e negativas com *Megaptera novaeangliae*, *Tursiops truncatus* e *Sotalia guianensis* e interações somente negativas com *Balaenoptera acutorostrata* (n=14) e *Stenella frontalis* (n=4). As interações positivas se referem a relações lúdicas e pesca colaborativa. Interações negativas (emalhe, emaranhamento, colisão) afetam os pescadores e os cetáceos. *S. guianensis* é a mais afetada por emalhes em redes de pesca e *M. novaeangliae* por emaranhamento e colisão com embarcações. Os resultados deste estudo destacam o valor de reunir comunidades científicas e pesqueiras para compreender os conflitos através da monitorização das interações, a fim de avaliar o impacto das atividades de pesca nas populações de cetáceos. Os pescadores possuem uma percepção positiva sobre os cetáceos, conhecem as áreas onde ocorrem os acidentes e gostariam de evita-los. Eles podem contribuir para as pesquisas sobre estes animais e a elaboração de planos de manejo adequados a realidade local. Isto aponta a necessidade da integração de conhecimentos científicos e locais para a conservação dos cetáceos e para a sustentabilidade das práticas da pesca.

**Palavras-chave:** etnobiologia, mamíferos marinhos, emalhe, emaranhamento, colisão.



## Introduction

Interactions occur between fishers and cetaceans as a result of fishing grounds overlapping the areas frequented by the animals. The intensity and frequency of these interactions may be increasing as a result of an increasing fishing effort and the growth of cetacean populations (Northridge 1984, De Master et al. 2001, Read et al. 2006, Silva et al. 2014). In Brazil, the banning of commercial whaling in 1987 (Law 7643), along with other conservation measures, has resulted in the growth of humpback whale (*Megaptera novaeangliae*, Borowski, 1781) (Andriolo et al. 2010, Ward et al. 2011) and southern right whale (*Eubalaena australis*, Desmoulins, 1822) (Groch 2005) populations. This population growth, in turn, is a potential source of conflict.

Ethnobiological studies emphasize the importance of deepening the analysis of interactions between fishers and cetaceans in order to estimate the extent of these interactions and provide new information about these animals (e.g. Alarcon et al. 2009, Zappes et al. 2013a, Lenney et al. 2015). These interactions may lead to insecurity and economic losses for fishers as a result of reduced catches and damage to fishing gear (Zambonim et al. 2009, Zappes et al. 2011b); injuries or death of cetaceans from collisions with fishing boats (Marcondes & Engel 2009, Zappes et al. 2013b); intentional (Barbosa-Filho et al. 2016) or accidental capture (bycatch) (Freitas-Netto & Di Benedetto 2008, Silva et al. 2014, Manzan & Lopes 2015, Revuelta et al. 2018). Damage to fishing gear usually occurs when cetaceans are caught accidentally (Freitas-Netto & Di Benedetto 2008, Zappes et al. 2011b) or when travelling migratory species drag and destroy fishing equipment (Pinheiro & Cremer 2003, Zambonim et al. 2009, Zappes et al. 2013a). In most cases, the carcasses of cetaceans that are accidentally killed are discarded at sea, but they may be used for human consumption or for use as bait (Zappes et al. 2009, Souza 2011, Lenney et al. 2015, Barbosa-Filho et al. 2018).

There are also benefits for fishers arising from their interaction with cetaceans. For example, it has been reported that bottlenose dolphins (*Tursiops truncatus*) can indicate the location of fish shoals to fishers (Peterson et al. 2008, Zappes et al. 2011a). On the other hand, the interaction can be positive for the cetaceans, which can catch fish directly from the fishing gear and thus save energy that would be needed for fishing (e.g. Orca, *Orcinus orca*, Dalla-Rosa & Secchi 2007).

Integrating fishers's knowledge into scientific research may be the only way to ensure the quality and accessibility of the data that only they can provide. A contextualized analysis that is connected to local realities allows better understanding of the natural systems and people's relations with them (Diegues 2000). This understanding can support the planning of more appropriate management strategies for local natural resources (Carlsson & Berkes 2005). Scientific knowledge of the majority of cetacean species occurring in Bahia is still scarce: 14 species are classified as "Insufficient Data" with regards to their conservation status (Cassano et al. 2017). The increase in humpback whale sightings reported by fishers in the region (Souza 2015) suggests that encounters with the species have increased. Fishing is small-scale and artisanal in the Ilhéus region: the fishing fleet is made up of *saveiro* boats varying from 6-9 m in length for line fishing and from 10-15 m in length for trawling (Barbosa-Filho & Cetra 2007). In light of the above, this study aims to assess whether there is an overlap between

the fishing grounds and the areas visited by cetaceans, to describe and analyze interactions and to identify possible conflicts between cetaceans and the artisanal fishers of Ilhéus.

## Material and Methods

### 1. Study area

The municipality of Ilhéus (14° 48' S; 39° 01' W) is located on the southern coast of the state of Bahia, in the northeast of Brazil. In this municipality there are two Fishers's Colonies (Z-19 and Z-34), as well as the Fishers and Shellfish Association of São Miguel (A-87) (Figure 1). The municipality of Ilhéus was chosen for this study because fishing is an important economic activity (Queiroz 2012) and because of the occurrence of a wide variety of cetacean species in the region (Cassano et al. 2017, Batista et al. 2012).

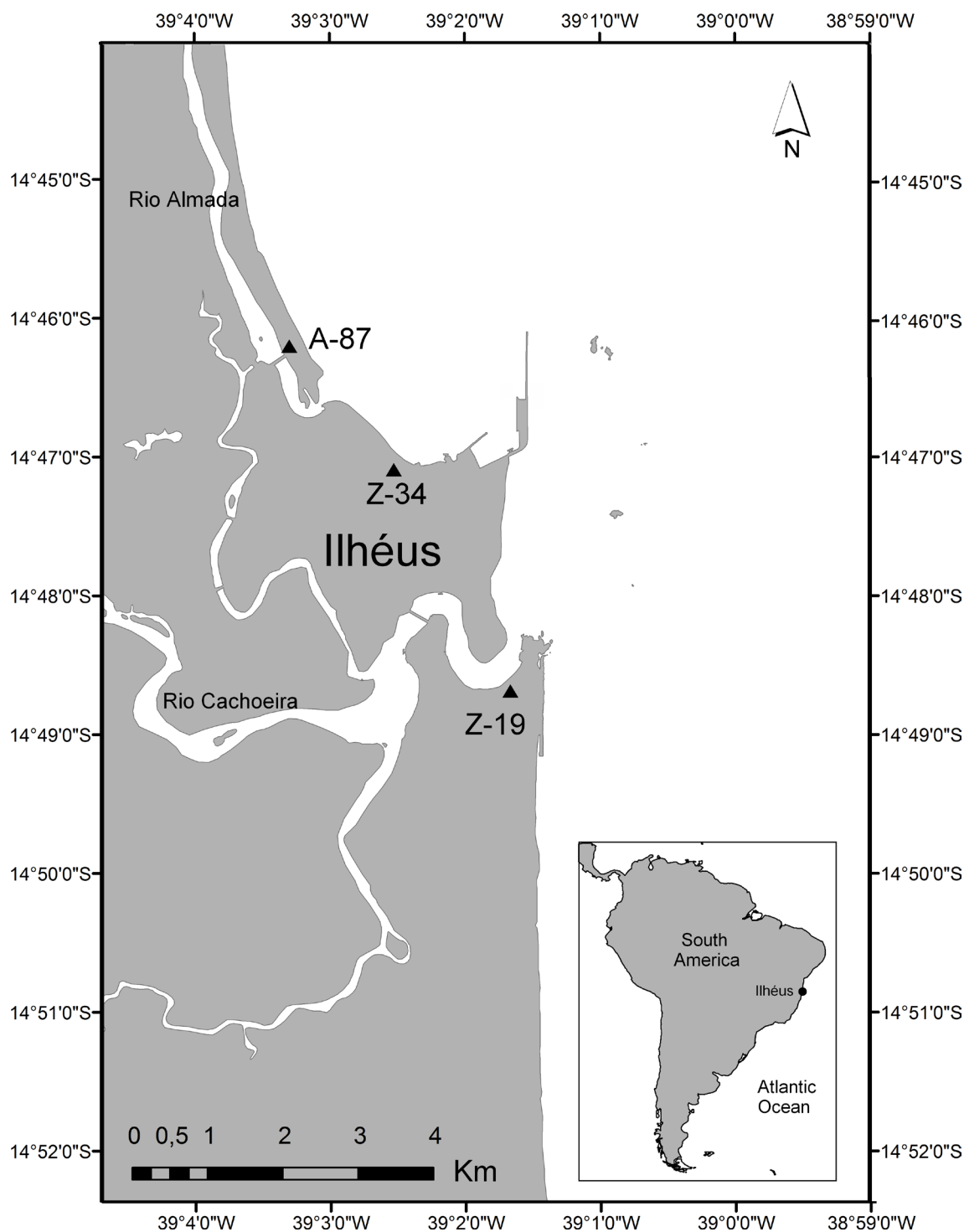
The width of the continental shelf ranges between 6 km in the municipality of Itacaré, located 50 km to the north of Ilhéus and 17.5 km on the Royal Charlotte Bank, situated 130 km south of Ilhéus. The shelf consists of a sand and sandy mud seabed in the shallower areas (up to 10 m depth), followed by mixed muddy sediments (up to 20 m depth). At the edge of the continental shelf break (50 to 70 m depth) muddy sediments and biotrititic sands predominate (Bittencourt et al. 2000).

### 2. Data collection

A qualitative approach is appropriate to conduct studies on cultural perception of members of local community, because the method allow the researchers to approximate the subjects of study and better understand the social, historical and cultural context of the community (Zappes et al. 2013a). According to Mason (2010) a qualitative research is concerned with meaning. Thus, frequencies are rarely important, the occurrence of a data is as useful as many in the understanding of the subjective process of a community.

Sample size in qualitative research designs is highly variable (Marshall et al. 2013). In this study, the researcher (first author) conducted oral interviews with 35 fishermen. This number of interviewees is comparable to other ethnobiological studies on perceptions of cetaceans by communities in Brazil, as: Souza & Begossi (2007) with an average of five respondents in each community in São Sebastião, state of São Paulo; Zappes et al. (2009) with 20 interviews in each area studied in Bahia, Espírito Santo, Rio de Janeiro and São Paulo; Zappes et al. (2011a) with 22 artisanal fishermen in Barra de Imbé/Tramandaí, Southern Brazil; Zappes et al. (2011b) with 22 respondents in Rio de Janeiro; Amorim et al. (2012) with 20 interviews in Rio de Janeiro, Costa et al. (2012) with 22 in Canavieiras, Bahia; Zappes et al. (2013a) with 33 ethnographic interviews in Garopaba, Santa Catarina; Manzan & Lopes (2015) with an average of 38 in each community of Rio Grande do Norte.

Data collection began after approval was given by the Ethics Committee (CEP-CONEP nº 68007516.9.0000.5526). The nature and purpose of the research was explained to each fisher and they were each provided with a Free and Informed Consent Form, in order to establish whether they would like to contribute to the research, with permission requested for anonymous recording of information.



**Figure 1.** Geographical location of Ilhéus and the Z-19 Fishing Colony in Pontal, Fishing Colony Z-34 in Malhado, and the Fishers and Shellfish Association of São Miguel (A-87) in Ilhéus, Bahia, Brazil.

We conducted a pilot study in February 2017 to test and calibrate the questionnaire. The questions were adjusted to improve the understanding, cultural acceptance and the usual vocabulary used by the fishers in the study area and to improve the interpretation of the data (Fontanella et al. 2011). The interviews considered for analyses were conducted between March and September 2017 and took place on streets, beaches, squares, and fishers' association headquarters. Each interview lasted for about an

hour. The interruption of the interviews occurred when the researcher verified the scarcity of new types of statements, based on the empirical data already collected and on her analytical and interpretative attributes. The collection of new data by additional interviews would not necessarily add new information for discussion in relation to the theoretical density already obtained for the research objectives (Mason 2010; Fontanella et al. 2011; Marshall et al. 2013).

The first interviewees were the fishers considered by the presidents of the colonies as having the greatest and most detailed knowledge of the regional fishing context. The non-probability “snowball” sampling method (Biernacki & Waldorf 1981) was then applied, which involved each fisher indicating another with recognized skill as a fisher. Only artisanal fishers with at least 15 years of fishing experience and who have resided in the municipality for more than 10 years were interviewed. These criteria were considered reasonable for acquiring accurate knowledge of the dynamics and components of the marine ecosystem (Souza & Begossi 2007).

Semi-structured interviews were conducted (Albuquerque et al. 2014), the questionnaire consisted of open (25) and closed (20) questions, addressing socioeconomic aspects of local fishing (age, time as a fisher, time of residence in the study area, schooling level, characteristics of the vessels) and the interactions between fisheries and cetaceans: local name, positive and negative interactions, local of interactions, behavior, accidents related to vessels and animals (bycatch, intentional use of cetaceans by fishers for baiting and other uses).

Fishermen were visually stimulated (Medeiros et al. 2014) to investigate their ability to distinguish the cetaceans that occur in their fishing territory and to indicate interactions with fishing activity). We used 19 boards each containing four different photographs of 19 cetacean species previously recorded in the study area (Rocha-Campos et al. 2011, Monteiro-Filho et al. 2013, Cassano et al. 2017): Suborder Mysticeti: *Eubalaena australis*, *Megaptera novaeangliae*, *Balaenoptera acutorostrata* (Lacépède, 1804), *B. edeni* (Anderson, 1879) and *B. physalus* (Linnaeus, 1758); Suborder Odontoceti: *Physeter macrocephalus* (Linnaeus, 1758), *Orcinus orca* (Linnaeus, 1758), *Globicephala macrorhynchus* (Gray, 1846), *Feresa attenuata* (Gray, 1870), *Pseudorca crassidens* (Owen, 1846), *Peponocephala electra* (Gray, 1846), *Tursiops truncatus* (Montagu, 1821), *Sotalia guianensis* (Van Beneden, 1864), *Steno bredanensis* (Gray, 1846), *Stenella attenuata* (Gray, 1846), *S. frontalis* (Cuvier, 1829), *S. clymene* (Gray, 1846), and *S. longirostris* (Gray, 1828). The franciscana dolphin *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844), which has not been recorded in Bahia, was included as a control species in order to verify whether fishers would recognize this fact. Each board was shown to the fisherman. When he affirmed to recognize the depicted animal, he was questioned about the common name and the interactions with that animal.

Nine of the 35 interviewees were invited to elaborate a participatory mapping (da Silva & Verbicaro 2016, Gerhardinger et al. 2010). The fishermen drew on the nautical chart 1200 (Port of Ilhéus to Ponta Cumuruxatiba) scale 1:305090 (BRASIL 1979), provided by themselves, the location of the fishing areas previously cited during the interviews and navigation routes. They were also encouraged to mark the places where they usually observe humpback whales and where accidents with this species have already occurred. The participatory mapping of marine territory and accident areas has been chosen as a way to contribute to the conservation goals and actions of the IWC (International Whaling Commission) and the National Plan of Action for the Conservation of Marine Mammals, among them, to evaluate the impacts of the fishing activity on cetaceans and to identify the main areas of overlap (Rocha-Campos et al. 2011, IWC 2016). The local knowledge of fishermen is an efficient way to quickly fill this gap.

### 3. Data analysis

The data obtained from the interviews was analyzed using the model of identification of covert categories, in which all information pertinent to the research is considered (Hays 1976). The fishers could choose not to respond or to provide more than one answer to each question. Descriptive statistics (means and frequencies) were calculated using the number of responses as the criterion (Silvano & Begossi 2002).

The nautical chart drawn by the nine fishermen was digitalized and georeferenced using ArcGis 10.3, generating a map of the marine territory used by the community for fishing and the main areas of occurrence of accidents with humpback whales.

A Kernel density map was generated with the interpolation tool of Geostatistical Analyst Tools, an extension of ArcGis 10.3, using the fishing ground where the 35 fishermen sighted cetaceans. These coordinates were georeferenced on a map of the cartographic base in a SIG environment; then the Kernel density estimator was applied, which generated a density surface of the visual identification of areas of sightings, based on the number of citations for each fishing ground. The GPS points of the humpback whale accident sites were provided by the fishermen's colony and plotted on the map.

## Results

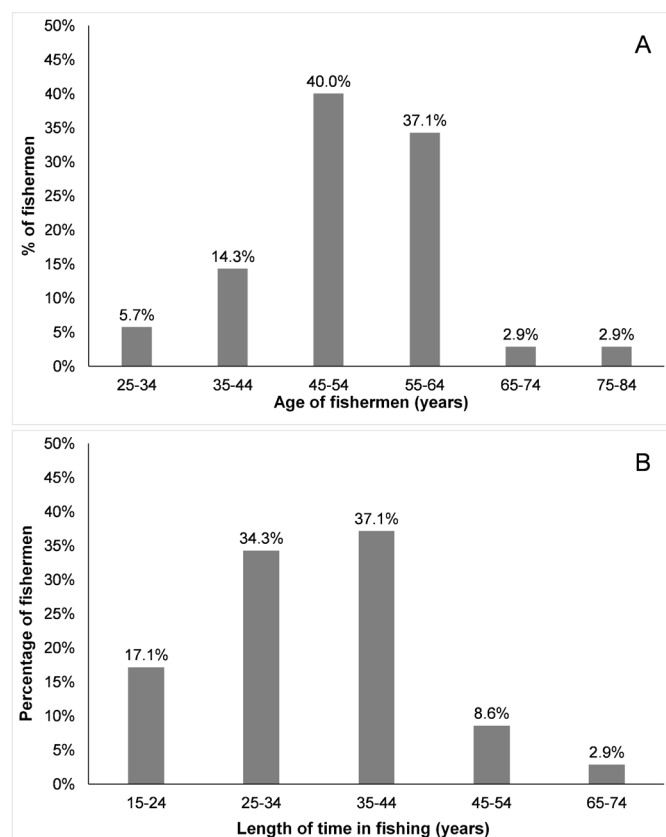
Thirty-five male fishers aged 25-82 years (mean = 51, standard deviation = 10) were interviewed, predominantly in the 45-54 years age group (Figure 2a). Time spent fishing ranged from 16 to 74 years (mean = 33, standard deviation = 11), with fishermen with 35 to 44 years of experience being the most frequent (Figure 2b). The schooling level of the interviewees is considered low, with 75% (n = 26) currently illiterate, having never been to school or not having completed elementary school. The majority of the fishermen (n = 21; 60%) were born in Ilhéus, while the others are from neighboring municipalities in the interior of Bahia (n = 8) and other states (n = 6).

The fishing grounds used by fishermen from Ilhéus cover 190 km of coastline from the municipality of Itacaré (14° 16' 36" S; 38° 59' 56" W) in the north to Belmonte (15° 51' 47" S; 38° 52' 58" W) in the south. The time spent at sea by the boats per trip varies from 7 to 9 days. The fishermen also carry out one-day trips, called “bate e volta” (“there and back”), to check the nets. The crew usually consists of four fishermen. Most vessels are equipped with VHF radio for communication and with GPS, compass and fishfinder for the location of the fishing grounds, located mainly on the continental shelf and continental slope.

The fishermen interviewed reported fishing-related interactions with five of the 19 species of cetaceans presented: *M. novaeangliae* (humpback whale), *T. truncatus* (common bottlenose dolphin), *S. guianensis* (Guiana dolphin), *B. acutorostrata* (dwarf minke whale) and *S. frontalis* (Atlantic spotted dolphin), known locally as: *Baleia*, *Golfinho*, *Boto*, *Tauaçu* and *Pinta-preta*, respectively. None of the participants reported having seen the franciscana dolphin *P. blainvillei* (control species) in the region.

### 1. Positive interactions

All of the respondents believe that the existence of cetaceans is important for conservation of the marine environment and most of



**Figure 2.** Age (A) and length of time in fishing (B) of the 35 artisanal fishermen interviewed in Ilhéus.

them ( $n = 32$ ; 91.43%) stated that they enjoy watching these animals and their behavioral displays. Positive interactions were reported with bottlenose dolphins, Guiana dolphins and humpback whales.

All of the fishermen cited at least one type of behavior for humpback whales and bottlenose dolphins, and 40% of them mentioned some kind of display for Guiana dolphins. The behaviors observed most often among Guiana dolphins were: “*jump, roll, play, pass by swimming, show the upper flipper and dive*” ( $n = 14$ ) (Table 1). They described both diurnal and nocturnal behavior for bottlenose dolphins (Table 2) and for humpback whales (Table 3). The most frequent behavior for bottlenose dolphins during day and night was “*swimming alongside the boat, at about 6 knots, then swimming near the prow of the boat and jumping clear of the water before going back to swimming alongside the boat again, it's funny, they are our friends, they distract us*” (1. 50 years). Two of the behaviors reported refer to the use of parts of the boat: “*When the boat is anchored, they wrap themselves around the anchor line to scratch themselves, they circle around it, and then go down*” and “*they wrap themselves around the anchor line and pull the boat along, they are strong, they pull it about 3 m, and then let go, and then do it again*”.

Breaching was the most common behavior for humpback whales during the day, while “*at night they are quieter*” (Table 2).

Fishermen fishing on the riverbanks or in the river reported that Guiana dolphins show them where the fish are and drive them towards the bank ( $n = 3$ ), they describe how “*the boto swims above the fish and*

**Table 1.** Number of mentions ( $n = 14$ ) for behaviors displayed by Guiana dolphins and reported by fishermen from Ilhéus-BA.

Observed behaviors	Number of citations
Jump	10
Play	6
Roll	2
Pass by swimming	2
Show the upper flipper	2
Dive	1

**Table 2.** Number of mentions of diurnal ( $n = 35$ ) and nocturnal ( $n = 25$ ) behaviors displayed by bottlenose dolphins and observed by fishermen from Ilhéus, Bahia.

Observed behaviors	Number of citations	
	Day	Night
Bow-riding	31	9
Breaching	26	1
Playing with the anchor line	11	5
Play	11	-
Whistling, making "piiii" sound	10	-
Twisting, pulling the boat	3	2
Tail-slapping	2	2
Lifting head in and out of the water	1	-
Spyhopping	1	-
Spinning in the air	1	-

**Table 3.** Number of mentions of diurnal ( $n = 35$ ) and nocturnal ( $n = 25$ ) behaviors displayed by the humpback whale and observed by fishermen from Ilhéus, Bahia.

Observed behaviors	Number of citations	
	Day	Night
Breach, spinner-breaching	35	9
Tail-slapping	14	-
Tail up, head down	15	1
Flipper-slapping	7	1
Play with others	4	-
Stopped and looking at us	5	-
Floating on one side with one flipper out	4	-
Swimming/passing by	4	7
Lifting head and slapping	3	-
Lifting both flippers out of the water, open	3	-
Floating	2	-
Spraying	2	8
Becoming quieter, sleeping on the surface	-	6
Bellowing	13	6

*starts to jump, when this happens, we know that there are fish*” (M. 25 years). They also reported that bottlenose dolphins round up schools of fish ( $n = 4$ ), “*they swim around the boat, they round up the school of small fish that we use for bait, then we catch them*” (N. 57 years).



Guiana and bottlenose dolphins were mentioned as being protectors of fishermen at sea: “If you are drowning the boto pushes you to land” (n = 2). “The golfinho saves us, he does not let sharks attack you, he is the bravest animal in the sea” (n = 11). According to the respondents (n = 6), Guiana dolphins, bottlenose dolphins and humpback whales also warn of a change in the weather:

When a thunderstorm is coming, the baleia breaches, and swims around in circles. The golfinho is the same, it slaps the water with its tail, after this a cold front will arrive within two days. When it is windy they like to play, when the weather is getting worse they are happy. The boto is the same, they beat their tails at the Pedra da Concha (A. 63 years).

## 2. Negative interactions

Most of the respondents (n = 33; 94.29%) know of the existence of the law protecting these animals from capture and harassment (Law 7643/1987) and say that they respect it. However, negative interactions were reported.

Four fishermen reported the intentional harpooning of bottlenose and Atlantic spotted dolphins and explained that this still happens today:

The pinta-preta is often harpooned for use as bait for bottom longlining, it is good for snapper, grouper, shark, swordfish (A. 46 years).

They catch them for shark bait, the smell attracts sharks. The golfinho come very close to the boat, it's the same with the people here from Ilhéus and from elsewhere too, Valença, Alcobaça and Vitoria. It happens all the time, even today (P. 48 years).

The animals most cited for disrupting fishing were: the humpback whale for getting caught on the line or rope and the minke whale for coming close to the boat and because there is a belief that they attack fishermen (Figure 3). The minke whale was described as “the most

distressing, a very angry creature”, considered “very dangerous”, it scares all of the respondents who recognized the animal (n = 14; 100%),

It is treacherous, if someone is sitting on the edge, it comes from behind, it wants to devour the person. It is the most dangerous animal in the sea. You have to move to another spot, it does not flee when it hits the boat, it pulls the anchor and dives, in 35 m deep water, then it leaves. Every fisherman is afraid of this “fish”. It scares the fish away and the people too (N. 47 years).

Most of the respondents (n = 33; 94.3%) have developed strategies to prevent cetaceans from disrupting fishing, to try to avoid accidents and, in the case of humpbacks and minke whales, also as a result of the fear they instill. The most frequently adopted behavior is to move to another fishing site or remove the line from the water and wait for the animals to move away from the proximity of the boat. They also hit the sides of the boat or start the engine, and at night, “we turn off the light of the boat, so that they move away, the light attracts them”. Sometimes they are able to get rid of the animal without hurting it, “when the golfinho gets to be too much I throw flour; they don't like it and go away, I get rid of them without hurting them” (M. 82 years). However, sometimes the animals are injured, as quoted for the minke whale, “I threw a lead weight at its back to get it away, its nose was already wanting to pass the boat” (J. 55 years).

Despite the fishermen's strategies to ward off the animals, accidents do happen and are the result of bycatch, entanglement or collision. Bycatch in nets was mentioned for the bottlenose dolphin, humpback whale, Atlantic spotted dolphin and Guiana dolphin, with the latter being the most frequent victim (Figure 4). One fisherman (C. 55 years) explained that, “it doesn't see the net at night and its tail and fins become entangled; it cannot swim, and it dies, plenty die from the net or a hook line in the tail, the net causes a lot of harm”. Respondents (n = 24) reported that accidental capture (bycatch) often happens and

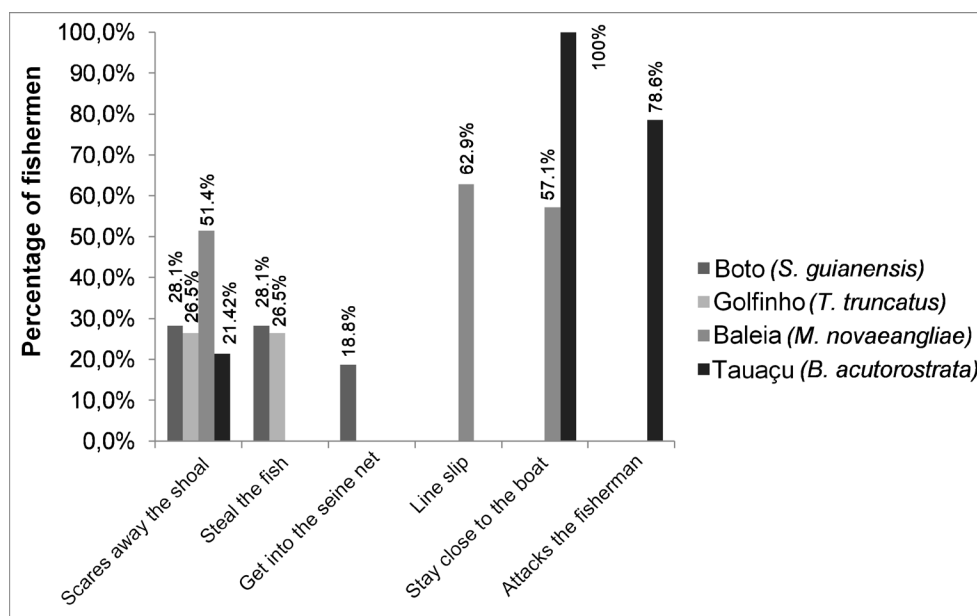
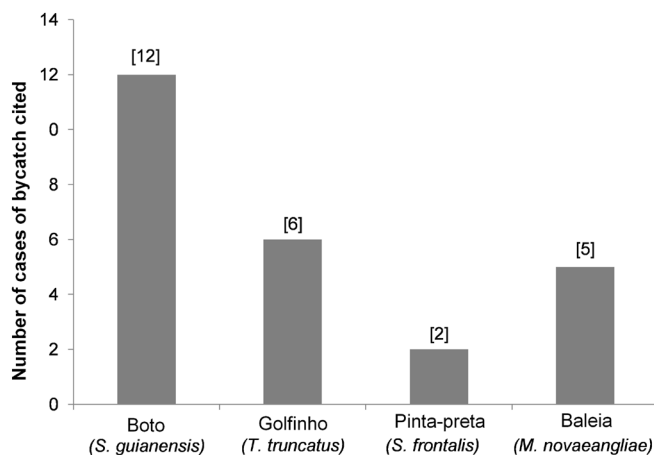


Figure 3. Ways in which the animals disrupt fishing, according to fishermen from Ilhéus, BA.

that sometimes the animals can be released while still alive. They can also be used as bait for shark fishing ( $n = 12$ ) or the meat is cooked and eaten ( $n = 5$ ) (Figure 5). They also discuss bycatch in ghost nets (nets lost in the sea), where they found: “turtles, golfinho, infant baleia and decomposing fish, the fish caught is wasted” (J. 45 years).

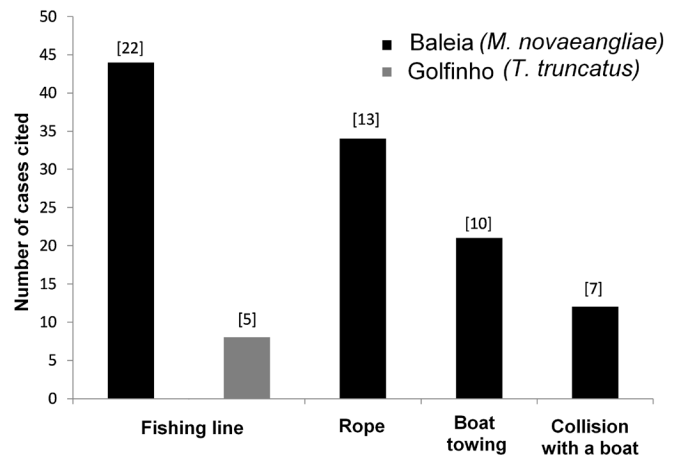


**Figure 4.** Number of cases of cetacean bycatch in fishing nets cited by fishermen from Ilhéus. The number of fishermen who cited the cases is enclosed in square brackets.



**Figure 5.** An adult Guiana dolphin carcass with transversal cuts in the anal region in the estuary of the Rio Cachoeira in Ilhéus (Cecília Inês Seminara, 2013, December 26).

Entanglement was mainly cited for the humpback whale. Everyone explained that as it swims by it gets caught and drags the line until it breaks. However, the rope does not break “when it gets caught in the anchor line it gets scared and dives, pulling the boat. The fisherman who fishes on the bow of the boat has to act quickly in order to cut the anchor line and we lose the line and the iron (anchor), otherwise it can sink the vessel” (M. 48 years). This interaction may cause injury to the animal too, as one reported “the anchor cut a hole in her face” (Figure 6).



**Figure 6.** Number of entanglements of dolphins and whales, cited by fishermen from Ilhéus, Bahia. The number of accidents is based on the memories of the fishermen.

I have been dragged four times. It got caught on the rope, it was cool, but scary, it got tired and we cut the rope. It was a race, we passed in front of the other ships. It lasted for more than an hour, it goes by and once or twice per trip breaks our line, we lose everything (A. 63 years).

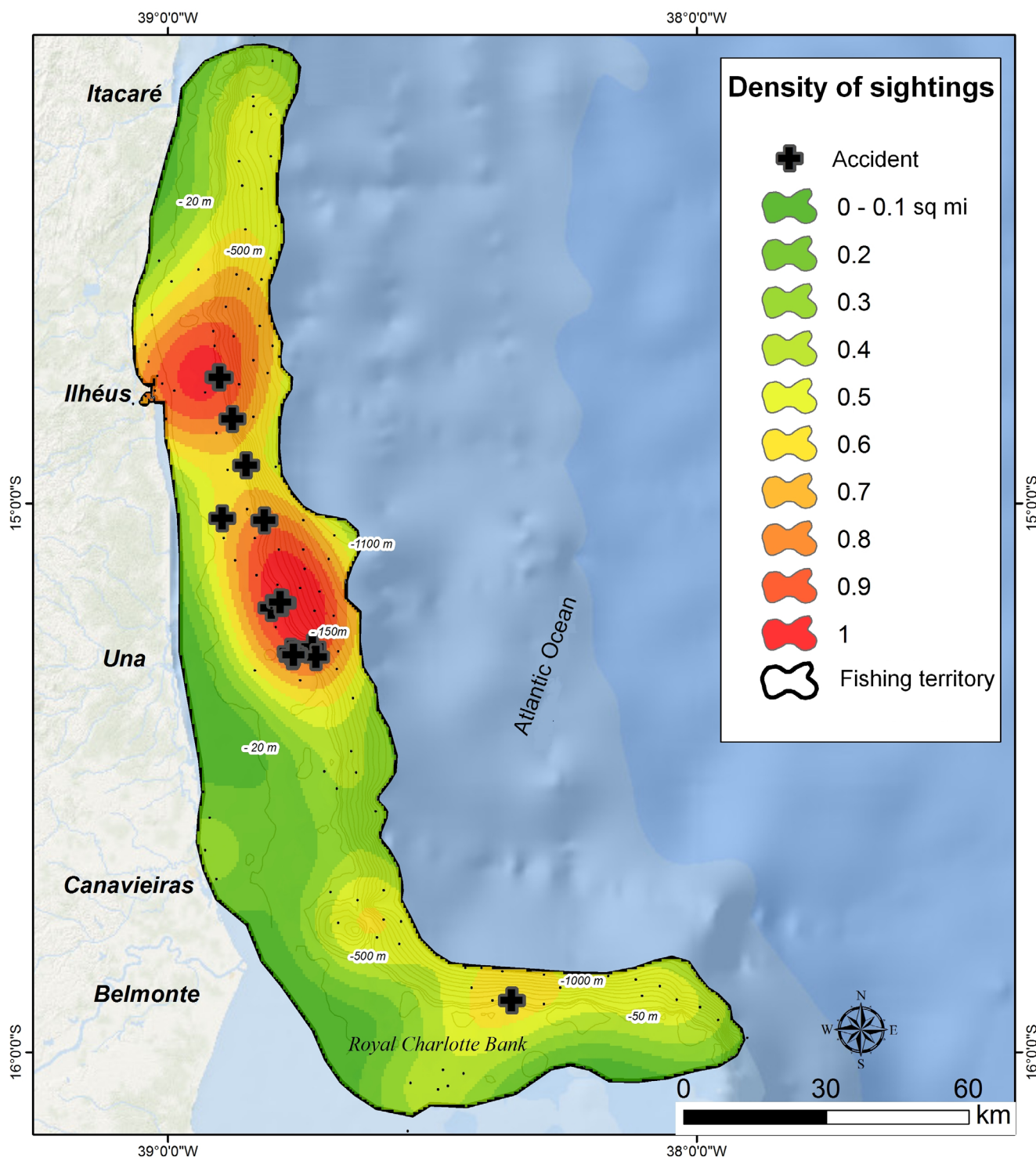
Some of the respondents ( $n = 25$ ) stated that the quantity of humpback whales has increased in the last 10 years, mostly because “they have stopped hunting them”, and, if the population continues to increase, they won’t be able to work anymore because “it is harming our fishing, there has to be some control”. Most fishermen ( $n = 24$ ; 68.57%) believe that the risk of accidents is high, especially at night, and have felt threatened by humpback whales before. The main reason is a fear of the humpbacks breaking or sinking the vessel, “It is very frightening, they are so big, it is dangerous to travel at night, it smacks its tail, it scrapes against the boat, it sinks the boat, if the young one comes alongside the boat the mother throws herself onto it” (P. 52 years). The areas where there were the most reported accidents with whales are between the coast of Ilhéus and Una (Figure 7).

The majority of respondents ( $n = 33$ ; 94.3%) would like to avoid accidents with cetaceans “to avoid losing equipment and to protect the animals”, but explain that “it happens, it cannot be avoided”. With regards to nets, one of them said that “whoever uses nets knows that at some point a dolphin will get stuck there. Nets should be banned” (N. 47 years).

Of the fishermen interviewed, 83% ( $n = 24$ ) would work with whale-watching, as they already have the example of whale watching in Abrolhos, “they earn well in Caravelas” (C. 55 years), but also stated that their vessels could not carry tourists and that “my life is fishing”.

## Discussion

The comparison between the time spent as a fisherman and the age of the interviewees indicates that they started fishing as a child (minimum 8 years old) or adolescent. This fact evidences the difficulty of articulating



**Figure 7.** Map of the density of cetacean sightings by square nautical mile (SNM) and the location of occurrences of accidents with whales reported by fishermen ( $n = 35$ ) from Ilhéus, Bahia.

formal education with the socio-cultural activity, demonstrated by the low level of education presented in the study. The low level of education of this social segment is found in many of the surveys conducted on the Brazilian coast, where most fishermen have incomplete primary education (Souza 2011, Ramires et al. 2012, Silva et al. 2014). The need to dedicate oneself to fishing to obtain income and the difficulty

of schools to follow a fishing calendar causes the early abandonment of formal education in the school.

The vessels used in Ilhéus are small (gross tonnage of 20 or less, BRASIL 2009), with reduced fishing technology, limited fishing autonomy and are practiced by professional fishermen. Most of the interviewees are native of Ilhéus and have all practice fishing for more



than 15 years, which is their main source of economic income. Since they spend more than half the month at sea, they acquire detailed knowledge of the marine environment, including non-target species (Mourão et al. 2006, Alarcon et al. 2009, Zappes et al. 2010, Silva et al. 2014).

### 1. Positive interactions

The fact that the fishers enjoy watching cetacean behavior indicates that they have a positive view of them. In fact, the beauty and social behavior of cetaceans can attract people's interest and generate friendly feelings among diverse fishing communities (Zappes et al. 2010, Rodrigues & Silva 2012).

The respondents cited a range of behaviors observed with cetaceans. The displays mentioned for *S. guianensis* correspond to the activities of socialization described by Izidoro & Le Pendu (2012) near the Port of Ilhéus. The Guiana dolphin is mentioned proportionally less because most of the fishers operate on the continental shelf and slope while the Guiana dolphins stay closer to the coast (Borobia et al. 1991).

The behaviors cited for bottlenose dolphins are described as socialization (Simões-Lopes 1998) and bow-riding (Janik 2015). Bow-riding was reported as occurring during both daytime and nighttime by fishers from the Cagaras Archipelago (Rio de Janeiro) (Zappes et al. 2010) and without specifying the time of day by fishers in Rio Grande do Norte, São Paulo and Santa Catarina (Souza 2011). According to Janik (2015) bottlenose dolphins frequently travel at the bow of boats, as either a playful behavior or a sexual display. According to the same author, the behaviors of "rolling themselves up in the anchor line, circling and diving" and "pulling the boat" can be play too.

Despite humpback whales are only being present for five months of the year in the region, fishermen mentioned most of the behavioral displays that can be found in the literature (see Engel et al. 2016), indicating considerable knowledge of the behavior of this cetacean. Humpback whales are mentioned more often due to their body size, which attracts attention, and due to more frequent interaction with this animal, which permits greater recognition (Mourão et al. 2006).

Collaborative fishing between fishers and dolphins has been reported in different parts of the world, including Brazil, for Guiana dolphins (Souza 2011, Costa et al. 2012, Brito 2012, Manzan & Lopes 2015) and bottlenose dolphins (Simões-Lopes et al. 1991, Peterson et al. 2008, Zappes et al. 2011a, Souza 2011). This type of behavior increases the fishers's contact with the animal, allowing them to build up empirical knowledge of the animals with which they coexist (Mourão et al. 2006).

The fishermen from Ilhéus believe that Guiana dolphins and bottlenose dolphins save people, including defending them from other potentially dangerous animals, such as sharks. These beliefs seem to generate positive attitudes among fishers towards these dolphins. Similar beliefs were recorded in Bahia (Zappes 2007), Pará (Bruto 2012) and São Paulo (Souza 2011) for the Guiana dolphin and in Pará and São Paulo for the bottlenose dolphin (Souza 2011). The reports related to Guiana dolphins, bottlenose dolphins and humpback whales reveal changes over time, demonstrating that fishermen recognize the existence of a behavioral pattern associated with certain environmental conditions. For example, humpback whales breach and perform fin and tail slapping with greater frequency when the wind speed increases, permitting communications between and within groups when the

noise from wind and waves interferes with acoustic communication (Kavanagh et al. 2017).

### 2. Negative interactions

Fishermen reported the intentional capture of small cetaceans by harpooning to serve as bait for shark fishing. In several regions of the world harpooning affects a number of cetacean species (Weir & Pierce 2012, Quintana-Rizzo 2011). In Brazil, harpooning has been previously recorded, mainly in Pará (Siciliano 1994, 2008, Alves & Rosa 2008, Brito et al. 2012). In Bahia, evidence of harpooning has been found in Itacaré (Alarcon et al. 2009), and in Canavieiras fishers explained that, in the past, cetaceans were often harpooned when following boats (Barbosa-Filho et al. 2016). Barbosa-Filho et al. (2018) recorded the use of dolphin fat for shark fishing by fishers in the south of Bahia.

According to the responses, there is a belief that the minke whale attacks people, mainly because of its behavior of approaching vessels. This has generated a negative attitude that is reflected in behaviors that can harm the animal. Confirming this behavior, researchers have characterized the minke whale (*Balaenoptera acutorostrata*) to be "friendly" due to its custom of approaching boats and swimmers on the Great Barrier Reef in Australia and interacting for hours (Arnold & Birtles 1999). It is likely that the respondents' lack of knowledge about the animal causes fear. The exchanges of knowledge between researchers and fishers can contribute to changing beliefs and improving the coexistence of fishers with minke whales and other cetaceans. This example illustrates the importance of understanding the community's perception of local fauna and the conflicts involved in order to efficiently contribute to educational programs (Silvano & Begossi 2012).

When animals interfere with fishing, fishermen use a range of methods to drive them away or move to another fishing site, which requires more fuel and travel time. Fishers from a number of different communities in Brazil have reported driving away cetaceans to avoid accidents (e.g. Alarcon et al. 2009, Zappes et al. 2011b, 2013a, Silva et al. 2014).

Bycatch was mostly mentioned in relation to *S. guianensis*, which is considered to be the most vulnerable species to fishing nets along the Brazilian coast for being a coastal species (Borobia et al. 1991): there are records of bycatch of Guiana dolphins in the southeastern, northeastern and northern regions of Brazil (Siciliano 1994, Freitas-Netto & Di Benedetto 2008, Alarcon et al. 2009, Zappes et al. 2009, Manzan & Lopes 2015). Accidental capture (bycatch) of *T. truncatus* was reported by few fishermen; however, reports and memories of accidents suggest that bycatch does occur. Bycatch of *T. truncatus* in fishing nets has been reported by fishers in Brazil (Simões-Lopes 1998, Zappes et al. 2011b) and in other countries around the world (Weir & Pierce 2012, Leeney et al. 2015, Revuelta et al. 2018). Fishers also denounced the bycatch of animals in "ghost nets". These pose a serious threat to marine animals as they continue to capture a diverse range of organisms in the coastal areas of the United States (Arthur et al. 2014) and Guatemala (Quintana-Rizzo 2011), amongst others. The most frequently mentioned accidents involved the entanglement of humpback whales in the line or rope. The fact that humpbacks get caught up in the line and usually tow the vessel causes insecurity and fear among the fishermen. Entanglement in the line has been described by fishers in the south of Bahia, Espírito Santo and Rio de Janeiro (Alarcon et al. 2009, Zappes et al. 2013b, Silva



et al. 2014), who report that the presence of humpback whales hinders the use of this equipment. The loss or damage of fishing equipment causes financial losses for the fisherman, in addition to the loss of the fish catch, time and fuel. Humpback whales are found in the region in winter, which is not a good fishing season. The loss of fishing gear during a period of low profits may be especially difficult to handle for artisanal fishers.

For whales, the very thin nylon fishing line may not represent as serious a threat as the anchor rope or nets, which may hinder their swimming and feeding capacity (Johnson et al. 2005, Cassof et al. 2011). The friction of the nylon line dragging in the water may result in deep wounds that can penetrate through several layers of tissues, eventually reaching the bones (Moore et al. 2006). According to these authors, the wounds remain open, facilitating infection by pathogens. Pinheiro et al. (2015) reported a case of severe mutilation of the peduncle of a small whale entangled in a longline fishing gear off the southeastern coast of Brazil. In Bahia, humpback whales with nets or ropes attached to their bodies have been observed several times (Rocha-Campos et al. 2011).

The fishermen reported that collisions are more common at night. According to Zappes et al. (2013b), the whale-spotting capability of boat operators is practically zero during nighttime navigation. Although the vessels are small and easily maneuverable, accidents with boats have previously been recorded in Bahia, Espírito Santo and Rio de Janeiro (Marcondes & Engel 2009, Zappes et al. 2013b, Silva et al. 2014). Collisions are reported in various different regions of the world, such as Africa (Weir & Pierce 2012); Central America (Guzman et al. 2012) and the North Atlantic (Van Der Hoop et al. 2012). Accidents can cause internal or external injuries and even the death of the animals (Lagueux et al. 2011, Zappes et al. 2013b), as well as damaging the hull of the vessels. The damage caused to the vessel can generate a major economic loss for artisanal fishers, who are low-income workers (Queiroz 2012).

The participatory mapping aggregated relevant information represented graphically, an approach that facilitates management discussions among stakeholders (da Silva & Verbicaro 2016). The map allowed to identify the areas where humpback whales are most frequently seen, where accidents occur, and the areas of overlap in the use of marine space by fishermen and whales. Opening the dialogue and allowing the exchange of information with local fishers may result in their greater commitment and help in the implementation of conservation objectives. A spatial understanding of accident sites and the types of accidents that occur provides a very rapid method of visualizing accident sites and identifying areas of potential conflict between fisheries and cetaceans. Furthermore, it can contribute to a more refined dialogue between institutions and fishing communities (Gerhardinger et al. 2010).

The impression of the fishermen is that the number of humpbacks has increased in the last decade. According to Souza (2015), fishers from Ilhéus have noticed the population growth of humpbacks in the region. According to Andriolo et al. (2010), the increasing population of humpback whales result in the occupation of new areas. As a consequence, the frequency of negative interactions is likely to increase and the consequences for both fishers and humpbacks needs to be considered. Studies carried out with fishing communities in Rio

Grande do Sul and southern Bahia indicate that conflicts between fishers and humpbacks are increasing (Zambonim et al. 2009, Zappes et al. 2013a, b).

The fishermen responded that they do not know how to avoid accidents. According to Zappes et al. (2013a), to avoid conflicts with *E. australis*, fishers in Santa Catarina proposed the implementation of a closed season during the months that the species is present, alongside the development of whale watching activities. This last suggestion was also cited by Alarcon et al. (2009) as a way to reduce accidents with humpbacks in Itacaré. Nonetheless, the development perspectives of whale watching in Ilhéus are currently limited due to the low number of tourists during the austral winter and the low economic income of the local population. However, the development of whale watching with tourists can even replace fishing, an old activity, and result in new socioeconomic and cultural problems for the community. The implementation of a closed season would bring changes to the fishing industry. Fish is an important part of the culinary tradition of Bahia. If fishing is reduced or stopped during winter, then other sources of the product will have to be found to supply the municipality of Ilhéus. This would directly affect the local economy, which is partly based on the productive cycle of the local and regional fishing industry, involving economic dynamics that generate work and income for a large number of families in the municipality (Queiroz 2012). Alternative economic activities that may favor the valuation and conservation of cetaceans in the region are desirable, but according to Alves & Rosa (2008), understanding the socioeconomic aspects involved is important for the development of any successful management plan.

Other possible solutions to avoid accidents should be analyzed and discussed among the local fishing community, researchers and environmental agencies, such as IBAMA (Brazilian Institute of the Environment and Renewable Natural Resources), directing efforts to improve the commitment of stakeholders to successfully implement co-management strategies. According to Souza & Begossi (2007), socio-environmental conflicts can only be resolved with the contribution of local perception, which is a product of the beliefs and practices in the territory in order to value and strengthen the culture of the local community (Gerhardinger et al. 2010).

Many fishermen interviewed in Ilhéus have a positive perception about cetaceans, know the areas where the accidents occur and want to avoid them. This study shows that fishers can contribute substantially to the development of research on cetaceans and that they are essential in the development of management plans suited to the local reality. To minimize negative interactions between fishermen and cetaceans in Ilhéus, it is necessary to understand the socio-environmental relations involved in the interactions, in order to support the elaboration of measures that contribute to the conservation of cetaceans and the sustainability of fishing (Silvano & Begossi 2012). We recommend the development of a participatory environmental education program (e.g. Zappes et al. 2016) to increase understanding of the importance of cetaceans in the ecosystem, as well as to value local knowledge and traditions related to marine mammals. Systematic monitoring programs should be conducted with fishers on negative interactions in order to identify the species and number of animals involved in accidents.

## Conclusion

The fishermen from Ilhéus described both positive and negative interactions with cetaceans in their fishing grounds. The negative interactions affect both the practice of fishing in the region, through damage to fishing equipment, and the cetaceans, causing injury or death. The animals most cited as sources of negative interactions were: the Guiana dolphin as bycatch and the humpback whale for entanglement, dragging of boats and collision.

Estimates of accidents based on the fisher's memories provide a sense of the scale of negative interactions. Considering the increase in the humpback whale population and the "vulnerable" conservation status of the Guiana dolphin, it is necessary to strengthen relations between researchers and the fishing community in Ilhéus to develop studies focusing on these conflicts. As a result, interactions could be monitored to assess the impact on fishing activity and cetacean populations. With the exception of the minke whale, the fishermen have a relatively positive view of cetaceans and would like to avoid accidents. They have shown themselves to be participative and open with the research. This indicates that they are willing to contribute to research with these animals.

We also suggest strengthening knowledge exchange between researchers and fishers, since academic knowledge about the behavior of some species, such as the minke whale, could facilitate the coexistence of fishers with these animals. Information provided by fishers and reported in this paper could contribute to the definition of participatory conservation strategies to reduce accidents between artisanal fishers and cetaceans.

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## Author Contributions

Cecilia Inés Seminara data collection; Substantial contribution to all steps of the research from concept and design of the study to manuscript preparation.

Márcio Luiz Vargas Barbosa-Filho: Substantial contribution to all steps of the research from concept and design of the study to manuscript preparation.

Yvonnick Le Pendu: Substantial contribution to all steps of the research from concept and design of the study to manuscript preparation.

## Conflict of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

## Ethics

As informed in the manuscript Material and Methods section of the manuscript, data collection began after approval was given by the Ethics Committee (CEP-CONEP nº 68007516.9.0000.5526). The nature and purpose of the research was explained to each fisher and they were each provided with a Free and Informed Consent Form, in order to establish whether they would like to contribute to the research, with permission requested for anonymous recording of information.

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## Brazilian assessment on biodiversity and ecosystem services: summary for policy makers

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**Abstract:** Biodiversity and ecosystems are important elements for addressing national and global socioeconomic and environmental crises, since they provide new development opportunities, for example, as source of job and income creation, and reduction in poverty and socioeconomic inequity. Brazilian biological diversity is also expressed in its immense cultural diversity, with a great variety of knowledge holders. These peoples possess vast knowledge on agrobiodiversity, fishing, fire management, natural medicine, among others of commercial, cultural and spiritual value. The main conclusions of this Summary for Policy Makers is that land use changes and climate changes have been - and will continue to be throughout this century - the main drivers that result in the loss of biodiversity and ecosystem services in the country. Political interventions at different levels (from local to national, from public to private) and the enforcement of existing laws (regulatory mechanisms and incentives) are required to cope with the mitigation of the negative impacts of biodiversity and ecosystem services loss. Brazil has already a wide variety of policy instruments and socioenvironmental governance options, as well as global commitments (ODS, Aichi Targets, Paris Agreement) related to the objective of a sustainable future. However, inefficient management control or lack of incentive to comply with the rules pose risks to consolidating the path to this future. The country has strong and capable institutions, but infrastructural problems, slow processes, inefficient measurements and judicial, social and ecological conflicts obstruct a proficient performance. There is a lack of communication between science and society which needs to be improved by establishing an effective flow that makes communication inclusive and representative, reaching public and private decision makers. Permanent efforts to integrate Science and policy knowledges are desirable to build confidence between policy makers and researchers.

**Keywords:** Sustainable use, direct and indirect drivers; land use; climate change; policy instrument; global commitments.

## Diagnóstico brasileiro sobre biodiversidade e serviços ecossistêmicos: sumário para tomadores de decisão

**Resumo:** A biodiversidade e os ecossistemas são elementos importantes para enfrentar as crises socioeconômicas e ambientais nacionais e globais, uma vez que proporcionam novas oportunidades de desenvolvimento. Incorporar a biodiversidade na vida cotidiana é uma oportunidade de ouro para promover o uso sustentável da biodiversidade e dos serviços ecossistêmicos. A conservação e a restauração da biodiversidade, dos ecossistemas e de seus serviços associados mostram potencial para um novo desenvolvimento social e econômico, como fonte de geração de emprego e renda, redução da pobreza e da desigualdade socioeconômica. A diversidade biológica brasileira também se expressa em sua imensa diversidade cultural, com uma grande variedade de detentores de conhecimento indígenas e tradicionais. Esses povos possuem vasto conhecimento sobre agrobiodiversidade, pesca, manejo do fogo, medicina natural, entre outros de valor comercial, cultural e espiritual. As principais conclusões deste Sumário para Tomadores de Decisão é que as mudanças no uso da terra e as mudanças climáticas tenham sido – e continuarão sendo ao longo deste século – os principais vetores da perda de biodiversidade e serviços ecossistêmicos no país. Intervenções políticas em diferentes níveis (do local ao nacional, do público ao privado) e a aplicação das leis existentes (mecanismos regulatórios e incentivos) são necessárias para promover a mitigação dos impactos negativos sobre a biodiversidade e a perda de serviços ecossistêmicos. O Brasil já possui uma ampla variedade de instrumentos de política e opções de governança socioambiental, bem como compromissos globais (ODS, Metas de Aichi, Acordo de Paris) relacionados à possibilidade de um futuro sustentável. Entretanto, o controle ineficiente da gestão ou a falta de incentivo para cumprir as regras traz riscos para a consolidação do caminho para esse futuro. O país tem instituições fortes e capazes, mas problemas de infraestrutura, processos lentos, medidas ineficazes e conflitos judiciais, sociais e ecológicos impedem a realização de um desempenho eficiente. Há uma falta de comunicação entre a ciência e a sociedade que precisa ser melhorada por meio do estabelecimento de um fluxo efetivo que torne a comunicação inclusiva e representativa, alcançando os tomadores de decisão públicos e privados. Esforços permanentes para integrar essas duas esferas de conhecimento na sociedade são desejáveis para criar confiança entre os formuladores de políticas e os pesquisadores.

**Palavras-chave:** *Uso sustentável, vetores diretos e indiretos; uso da terra; mudanças climáticas; instrumentos de política; compromissos globais.*

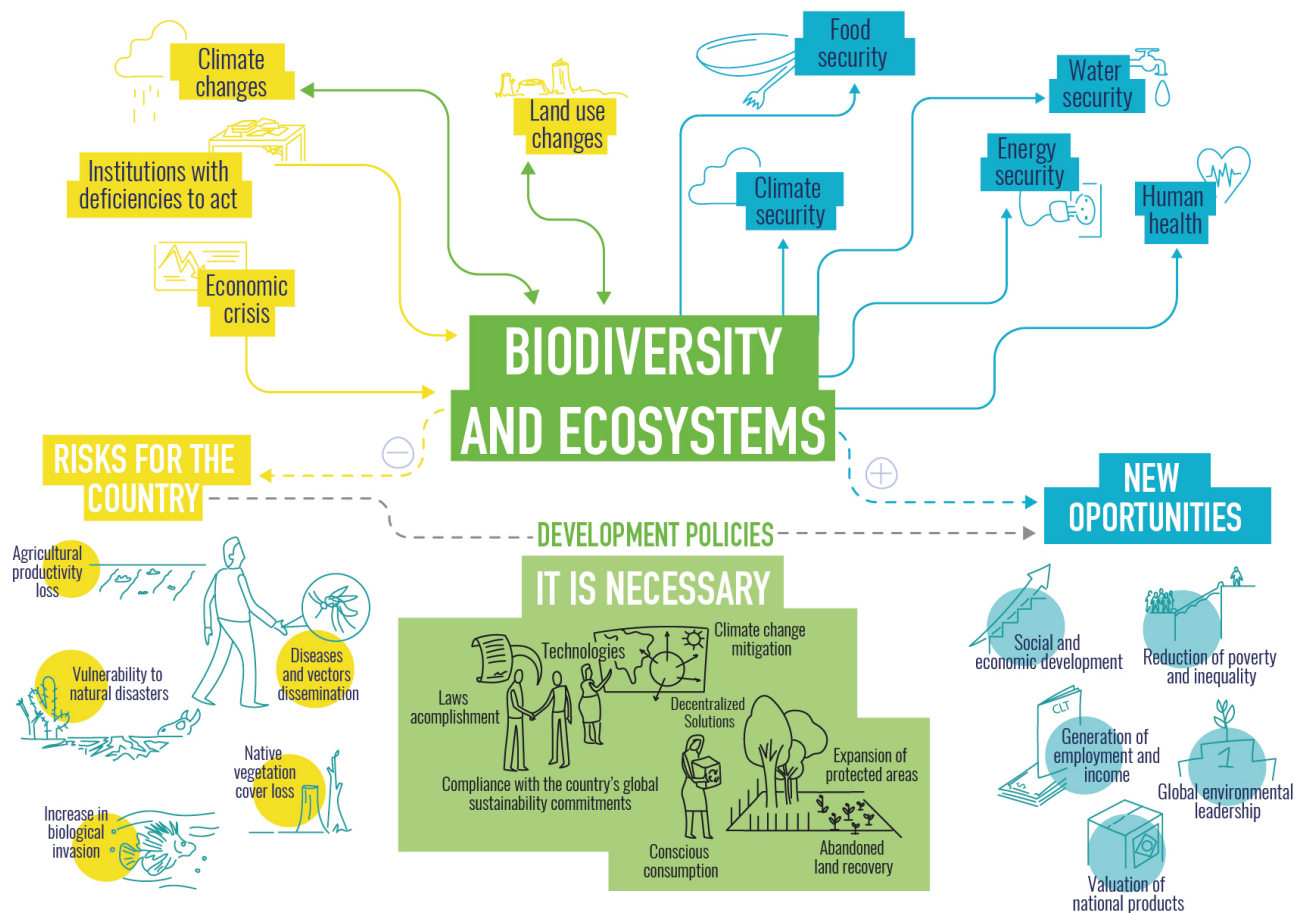
### About the Summary for Policy Makers

This summary for policy makers (SPM) objectively summarizes the 1<sup>st</sup> Assessment on Biodiversity and Ecosystem Services (Joly et al. 2019) written by the Brazilian Platform of Biodiversity and Ecosystems (BPBES). The BPBES is an independent group formed by around 100 authors, including professors, researchers, environmental managers and/or decision makers, who have met regularly since November 2015. During the assessment preparation process, sectoral working meetings were held with groups of interest (federal government, non-governmental organizations, companies, indigenous representatives and journalists) to share the main results, as well as hear, discuss and assimilate other points of view and suggestions. The platform receives funding from the CNPq (National Council for Scientific and Technological Development) and the Biota Program/Fapesp, and institutional support from the SBPC (Brazilian Society for Progress in Science), ABC (Brazilian Academy of Sciences) and FBDS (Brazilian Foundation for Sustainable Development).

The report in which this SPM was drawn (Joly et al. 2019) consists of 5 chapters - 1) The context of the Brazilian Assessment on Biodiversity and Ecosystem Services; 2) Nature's contributions to the quality of life; 3) Trends and impacts of degradation and recovery of biodiversity and ecosystem services drivers; 4) Interactions between nature and society: trajectories from the present to the future; and 5) Options for governance and decision-making across scales and sectors – and is available for download at the platform webpage [www.bpb.es.net.br/en/](http://www.bpb.es.net.br/en/).

### 1. Contextualization

- **The loss of biodiversity caused by the conversion of native ecosystems and the unsustainable use of natural resources threatens essential processes for human well-being. The growing systemic crisis (economic, climatic, hydrological, food and migratory) in Brazil is contrasted by the country's vast range of natural resources and opportunities to address these demands.** Its enormous natural capital provides the conditions needed to transform the conservation and sustainable use of environmental assets into development opportunities capable of dealing with future climate changes, while promoting socioeconomic prosperity. This unusual combination results from the fact that the high potential for economic production (present and future) depends on maintaining biodiversity resources and associated ecosystem services (Figure 1).
- **A prosperous future for the Brazilian population will depend on the choices and measures taken now, in terms of the country's sustainable development.** To construct a sustainable future, it is essential to understand the intrinsic and monetary value of biodiversity and ecosystem services for creating jobs and income, as well as reducing social and economic inequities. This scenario will only be possible, however, if the contribution of biodiversity in achieving social and economic development is recognized and encouraged. Sustainable Brazilian products could



**Figure 1.** Brazilian Platform on Biodiversity and ecosystem Services framework.

be viewed as unique by using a label such as *made in Brazil*, *environmentally friendly*, for example. It is urgent to halt the current unsustainable use of natural resources, given the various signs of environmental collapse.

- **Land use and climate change are the main pressures that result in loss of biodiversity and ecosystem services.** Due to its nature and the history of agriculture to date, it has been the primary agent of change in land use and the impact on biodiversity and ecosystem services. Given the current global agreements and market demands, the challenge and opportunity is to bring the agricultural sector closer to the biodiversity and ecosystem services agenda, thereby making them major allies.
- **Although Brazil is rich in fresh water, harbouring 12% of the world's reserves, water availability varies significantly between the subregions. There has been a per capita decline and generalized unsustainable use of surface and subterranean waters in many parts of the country** (see [https://www.bpb.es.net.br/wp-content/uploads/2018/11/BPBES\\_SPM\\_Agua.pdf](https://www.bpb.es.net.br/wp-content/uploads/2018/11/BPBES_SPM_Agua.pdf)). Water quality is declining in most watershed and coastal areas, and dependence on infrastructure for water supply is increasing. In urban areas, especially large centers, around 39% of the places monitored by the National Water Agency exhibited fair, poor or very poor quality. Moreover, despite its overall abundance, freshwater supply

can be scarce locally. This unequal availability, combined with an inadequate distribution infrastructure and inefficient wastewater treatment plants, make water security a problem for a large portion of the Brazilian population, reducing reliable access to sufficient amounts of potable water, with impacts on human health.

- **Throughout this century, the intensification of climate change will increase the loss of biodiversity and ecosystem services.** Combined with the decline in natural vegetation cover, alterations in temperature and rainfall patterns are forecast in the different regions of the country due to global climate change (see [https://www.bpb.es.net.br/wp-content/uploads/2019/01/Relat%C3%B3rioTem%C3%A1tico\\_ClimaCompleto.pdf](https://www.bpb.es.net.br/wp-content/uploads/2019/01/Relat%C3%B3rioTem%C3%A1tico_ClimaCompleto.pdf)). In the case of a “business as usual” scenario, the result will be loss of agricultural production, change in species habitats and distribution - affecting the dissemination of diseases and their vectors - and increasing the aggressiveness of alien species invasions.
- **The official governance of biodiversity and ecosystem services in Brazil is bipolar: on one hand, there are strong and capable institutions; on the other, infrastructural problems, slow processes, inefficient measures and judicial, social and ecological conflicts.** Ability and efficiency also vary, tending to decline from federal to state to municipal levels. The economic crisis forecasts a reduction in funding,

resulting in the loss of organizations and significant layoffs of technical personnel, thereby aggravating the situation.

## 2. Sustainability: a mandatory and promising path

- **The window of time and opportunity to consolidate a sustainable future is rapidly closing. We must at least ensure enforcement of existing laws through regulatory mechanisms and incentives, in line with the global sustainability commitments made by other countries.** Choices for this sustainable future must be made as soon as possible, and scientific knowledge, always in partnership with other forms of knowledge, will be essential for good decision making. Current global and national pressures in the social, economic and environmental fields are numerous and growing, and the current development model is becoming obsolete. A new model is needed to incorporate the challenges of a planet in rapid socioeconomic and climatic transformation.
- **Biodiversity and ecosystems are important elements for addressing national and global socioeconomic and environmental crises, since they provide new development opportunities. As such, they should be incorporated into the country's development policies.** Natural resources are currently being controlled by sectoral policies. Biodiversity and ecosystem services are considered an obstacle or even a barrier to the development process when, in fact, they are the basis for improved global competitiveness. The productive sectors will increasingly depend on inclusive and sustainable practices, circumscribing the challenge of creating a new agenda for the conservation and sustainable use of natural resources.
- **Political interventions at different levels (from local to national) may lead to successful mitigation of the negative impacts on biodiversity.** Given the complexity of issues related to biodiversity and ecosystem services and the numerous possible political interventions, there are a number of different options to follow. For example, the Globio modeling platform to support policies, uses three paths: global technology (large-scale technologically optimal solutions), decentralized solutions and changes in consumption. Using complementary mechanisms to apply these scenarios may minimize the negative impacts on biodiversity, especially those resulting from agriculture and energy production.
- **Investing in the conservation and restoration of biodiversity, ecosystems and their associated services shows potential for new social and economic development, as a source of job and income creation, and a reduction in poverty and socioeconomic inequity.** Equal use and access to natural capital are important elements to overcoming inequities in Brazil. They also guarantee the continuity of the multiple lifestyles and social and ecological systems that represent the cultural and ethnic diversity of the country. In addition, human well-being depends directly on the availability and access to ecosystem services (water, food, climate, culture) and biodiversity conservation creates business opportunities for industries including tourism, cosmetics, drug and food. However, there are still considerable obstacles to taking advantage of such opportunities. Manufacturing biodiversity-based products demands investments and a favourable business environment. Restoration, which is technically feasible in Brazil (see [https://www.bpbes.net.br/wp-content/uploads/2019/08/SPM\\_RestauracaoVF\\_ebook.pdf](https://www.bpbes.net.br/wp-content/uploads/2019/08/SPM_RestauracaoVF_ebook.pdf)), has demonstrated greater effectiveness in recovering ecosystem services than biodiversity. However it continues to be a costly process, ranging from R\$800 ( $\approx$  USD200) per hectare when natural regeneration is used, to R\$ 17,000 ( $\approx$ USD 4,250) per hectare in the case of seedling planting. Achieving 30% plant cover in the Atlantic Forest and maintaining the integrity of vertebrates essential to ecosystem functioning would cost around R\$ 445 million ( $\approx$ USD111 million), that is, less than 0.01% of annual GDP in Brazil, or 6.5% of agricultural subsidies.
- **Considering the ecological importance of connecting the landscape, recovery of native vegetation is recognized as an important strategy to mitigate the loss of biodiversity and restore ecosystem services, such as pollination and soil formation, which contribute to higher crop yields.** In more degraded biomes, enforcing the Native Vegetation Protection Law (New Forest Code) must provide for recovering native vegetation by implementing the Rural Environmental Registry (CAR) and the Environmental Regularization Program (PRA), with benefits for biodiversity and ecosystem services, including the production and conservation of water and carbon sequestration. It is estimated that enforcing the law would result in around 20 million hectares of restored area, enabling other gains for landowners, including higher yields and new green companies and jobs. Restoration planning should consider multiple functions and ecosystem services, such as planting species that are important for pollinators and contain active ingredients of interest to the pharmaceutical and cosmetic industries, as well as water body protection and soil stability services, in addition to cultural and spiritual services.
- **The land ownership issue and the relationship between agricultural production and conservation are central elements in land use planning, especially if the multiple ecosystem services, future demands and additional limitations caused by environmental changes are considered.** Managing agricultural production, using techniques that reduce carbon emissions and focus on existing deforested areas, has and will further increase the value of Brazilian agricultural products on the national and international market. Such adjustment in production activities will require policies and technologies that allow the occupation and increased yield of these lands, promoting techniques such as agroforestry systems and crop-livestock-forestry integration. An alternative measure would be to transform the agriculture funding system into an integrated model, focusing on the property as a whole and its production and growing system, instead of product-to-product funding as is standard practice today.



- **Connectivity should be considered when planning the management of biodiversity and ecosystem services.** Managing the natural resources of a particular area (farm, urban center, national park or geopolitical unit), has sometimes led to phenomena such as “leakage” or “spillover”, that is, transfer of the production area to less protected regions from the legal standpoint. These effects are due to “teleconnection” processes – remote associations between different locations, whereby what occurs in one place may affect another – that have been little investigated by scientists in Brazil. Thus, tools such as “a moratorium on certain products”, associated with the Rural Environmental Registry (CAR), may significantly decrease pressure on areas of native vegetation, but at the same time result in conversion in other ecosystems. For example, the success of the Soybean Moratorium in reducing deforestation in the Amazon was marred by the side effect of transferring deforestation to the Brazilian savanna (hereafter *Cerrado*), the neighboring biome. Containing these secondary impacts requires long-term planning and dynamic monitoring of land use.
- **Assessment of the effectiveness and efficacy of implemented policies is crucial to improving them and, in Brazil, there is a wide gap in this area, especially with respect to the scale of biodiversity and ecosystem services.** As an example, there are no studies about the effectiveness of protected areas for nature conservation and for providing direct and indirect socio-economic benefits. A combination of good governance, effective management and community engagement explains the success of some protected areas. Policies, such as Green Stipend (Bolsa Verde), National Plan for Agroecology and Organic Production (Planapo) and the National Benefit Sharing Program (PNRB), including the National Fund for Benefit, need to be reviewed regarding adequacy, degree of implementation and effectiveness.

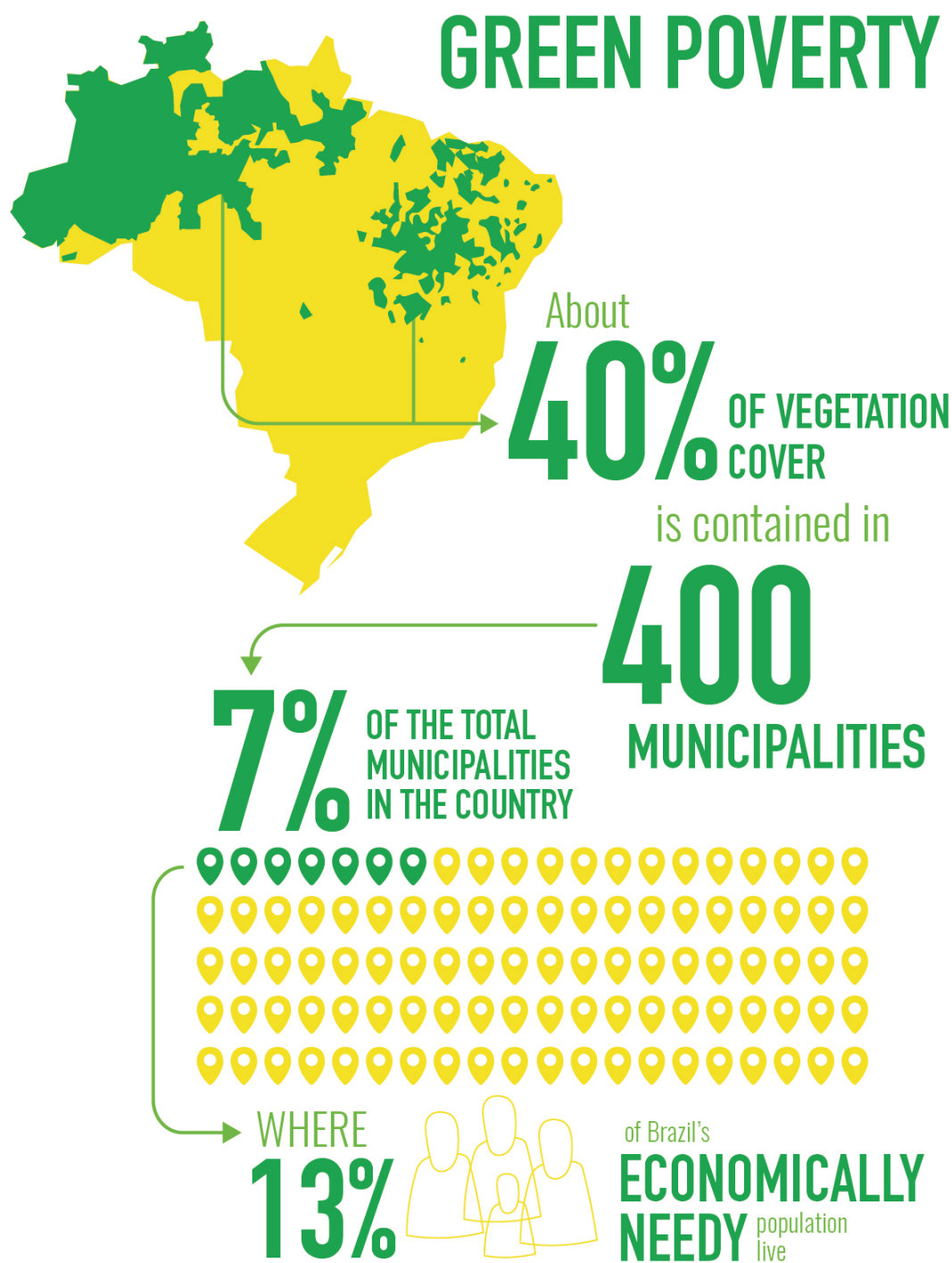
### 3. From risk to opportunity: the privilege of a megadiverse country

- **Up to 2030, land use change will continue to be the primary driver for the loss of biodiversity and ecosystem services and will be a key factor during most of the 21<sup>st</sup> century.** Despite the decline in annual habitat losses caused by deforestation in Brazilian biomes in the last decade, especially in *Amazonia*, the conversion of natural ecosystems remains high, particularly in the *Cerrado* (236,000 km<sup>2</sup> between 2000 and 2015) and *Caatinga* (45% of original cover). Even in the Atlantic Forest, the deforested area – around 29,000 hectares from 2015 to 2016 – is substantially higher than the area restored in the biome over the same time period. For example, in recent years, forest losses in the country were at least three times greater than the restoration promised area. Implementing land use planning with focuses in combining conservation and production is one of the main strategies

to preserve biodiversity and ecosystem services. However, only recognizing that the future of agricultural production depends on preserving ecosystem integrity will enable a definitive conciliation between socioeconomic growth and environmental conservation.

- **Food, water, climate and energy security depend on ecosystem services, such as pollination, water resource maintenance, climate regulation and control of disease vectors.** Of the 141 crops analyzed in the country, 85 depend on animal pollination. Around 80 families and 469 plant species are grown in agroforestry systems. More than 245 species of Brazilian flora are the source of cosmetics and pharmaceuticals and at least 36 native botanical species are registered as herbal medicines (see [https://www.bpb.es.net.br/wp-content/uploads/2019/03/BPBES\\_CompletoPolinizacao-2.pdf](https://www.bpb.es.net.br/wp-content/uploads/2019/03/BPBES_CompletoPolinizacao-2.pdf)). Over 40% of primary energy production in the country comes from renewable sources, and 2/3 of the electrical energy consumed originates in hydroelectric plants that depend on ecosystem integrity, especially the forests, to continue operating.
- **The concentrated poverty in municipalities with substantial native vegetation cover is a risk that may become a unique opportunity to reconcile conservation with human development.** Approximately 40% of vegetation coverage in Brazil is found in 400 municipalities (7% of the municipalities in the country) where 13% of the most economically underprivileged Brazilians live (Figure 2). Historically, replacing forests with agropastoral activities has not resulted in a significant increase in the human development index (HDI) of individuals living there, which exacerbates the rural exodus. The significant rise in income generation from conserving nature, such as the case of the Policy to Guarantee Minimum Prices for socio biodiversity products, applied to non-wood forest products extracted primarily by traditional populations and family farmers, will be essential to reconciling socioeconomic prosperity with natural resource conservation.
- **In order to address the risks of climate change, which are already impacting natural and social systems, the Ecosystem-based Adaptation Strategy stands out as a significant opportunity for Brazil.** Adapting to the adverse effects of climate change requires an innovative strategic approach, such as that based on ecosystems. With this tool, biodiversity management can improve the flow and quality of water and reduce vulnerability to natural disasters<sup>1</sup> and their consequent impacts, such as landslides and higher sea levels. These practices are less costly than alternatives based on building conventional infrastructure. Ecosystem-based adaptation conserves or restores natural resources, sequesters or stores carbon, and has the potential to reduce poverty.
- **The biological diversity of the country is also expressed in its immense cultural diversity. Incorporating indigenous and traditional knowledge about Brazilian biodiversity into day-to-day society is a golden opportunity for sustainable use of biodiversity and ecosystem services.**

<sup>1</sup> In the last two decades, Brazil has recorded 32,000 natural disasters such as droughts, gradual flooding, flash flooding, tornados, mass wasting, hailstorms and fluvial and coastal erosion.



**Figure 2.** The concentrated poverty in municipalities with substantial native vegetation cover is a risk that may become a unique opportunity to reconcile conservation with human development.

Brazil has more than 500 natural sites associated with multiple cultural manifestations. The country is home to 305 indigenous peoples speaking 274 languages, and dozens of other traditional populations, such as the *Caiçara* (traditional population on the south and southeast coast), *Quilombolas* (people of African origin living in hinterland settlements), rubber tappers, *Ribeirinhos* (riverside dwellers), babaçu coconut shellers, *Pantaneiros* (residents of the *Pantanal* wetlands) and *Vazanteiros* (island or riverbank dwellers), in

addition to historically receiving migratory flows from different parts of the world. These peoples possess vast knowledge of agrobiodiversity, fishing, fire management, natural medicine, among others of commercial, cultural and spiritual value.

- **Brazil has a wide variety of policy instruments and socioenvironmental governance options, as well as global commitments (ODS, Aichi, Paris) related to the possibility of a sustainable future. On the other hand, inefficient management control or lack of incentives to comply**

#### with the rules pose risks to consolidating this future.

After a period of stagnation, and even a reduction in the area protected by conservation units, 2018 saw a significant increase with the creation of four marine conservation units (two environmental protection areas and two national monuments), an environmental protection area and a national park in the *Caatinga* biome, and three extractive reserves in Maranhão state. The National System of Protected Areas covers more than 15% of Brazilian territory and 561 recognized or regulated indigenous lands, encompassing 12.2% of the country. However, protected areas with low enforcement rates, cancellation of financial conservation incentives to reduce poverty (such as the Green Stipend) and problems with the environmental licensing process of commercial undertakings reflects the lack of policies to ensure effective conservation and the sustainable use of biodiversity and associated ecosystems in the country. Planaveg<sup>2</sup>, the National Plan to Adapt to Climatic Changes and some items of the Native Vegetation Protection Law (LPVN)<sup>3</sup>, in addition to the National REDD+ Strategy (ENREDD+), among others, bring biodiversity to the forefront of discussions and are in line with the global goals adopted by Brazil, which represents an unprecedented opportunity to implement these policies.

#### 4. The role of science: dialogue and knowledge serving society

- **There is a lack of communication between science and society. This process needs to be improved by establishing an effective flow that makes communication inclusive and representative, reaching public and private decision makers.** In addition to credibility, science should present legitimacy and ability to produce relevant results (easy to understand and of collective and political interest) for society and decision makers. Synthesis Centers on Biodiversity and Ecosystem Services are an option for promoting these transdisciplinary studies, conducted in coproduction with other actors.
- **Public and private research funders in Brazil should be more active in promoting transdisciplinary science, which involves policy makers and other actors at its conception, to solve problems.** Open calls funding are essential in promoting basic research, but there is still a serious gap in promoting research aimed at addressing the problems and specific challenges faced by the Brazilian population. To bridge this gap, adequate scientific and technological policies are needed to address the challenges of sustainability. It is important to create a funding model and assess the impact of research aimed at generating support and contributions in order to improve

conservation policies and sustainable use of biodiversity and ecosystem services.

- **The last decade saw significant advances in the sharing and transparency of public data and information on biodiversity and ecosystem services that allow qualified decisions.** However, serious gaps remain, such as the lack of knowledge about less represented taxa and with different degrees of endemism, making it difficult to incorporate science into the decision process. Repatriations of knowledge on Brazilian biodiversity, associated with advances in compiling species lists, including endangered and invasive ones, and in understanding the functioning of natural systems, in addition to the use of free-access geospatial tools, are more common today, but still needs incentives.
- **Understanding the interactions between biodiversity, ecosystem services and human well-being is a pre-requisite for promoting the agenda of a number of multilateral environmental agreements and global goals. However, studies that assess these inter-relations remain scarce.** While in developed countries the proportion of studies that analyze the relation between biodiversity and ecosystem services and discuss human well-being is around 5%, in Brazil it is half that value. Analysis of more than 220 studies published in national and international journals shows that around 42% of investigations that explore the relation between biodiversity and ecosystem services in the country are focused on biological processes and more than 77% examined biodiversity from the native vegetation standpoint. The few studies that seek to value national biodiversity are concentrated primarily on material goods, pollination services and water and climate regulation. Little is known about the non-material values of biodiversity that generate well-being for the population, such as those related to ecotourism, scenic beauty and spiritual and cultural aspects.
- **Some biomes remain poorly studied in terms of land use changes and their impacts on biodiversity.** The *Caatinga*, *Pampa*, *Pantanal* and coastal and marine zones are systems that require more studies, especially the peculiarity in Brazil of the continued use of traditional agricultural practices. Taken together, these biomes account for 35% of the studies on the relation between biodiversity and ecosystem services in the country.
- **Scientific research and policy have different response times. Permanent efforts to integrate these two spheres of knowledge are needed to build confidence between policy makers and researchers.** It is important to explore the potential of science, technology and innovation to induce cooperation between government entities. It is also recommended that research funding agencies require a “summary for policy makers” as an output in their calls.

<sup>2</sup> National Plan of Native Vegetation Recovery = is the main implementation instrument of the National Policy for Native Vegetation Recovery (Proveg – Decree 8972 of 01/23/2017). Its objective is to broaden and strengthen public policies, financial incentives, markets, good agricultural practices and other native vegetation recovery measures.

<sup>3</sup> Native Vegetation Protection Law (LPVN – no. 12.651 of 05/25/2012) – substituted the Forest Code of 1965 and is in the regulatory phase at federal and state level, but the constitutionality of some of the changes is still being questioned.

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## Author Contributions

Carlos A. Joly: contributed to the concept and design of the study and to a critical revision, adding intellectual content.

Fabio R. Scarano: contributed to the concept and design of the study and to a critical revision, adding intellectual content.

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Andrea Larissa Boesing: contributed to data collection, data analysis and interpretation and to a critical revision, adding intellectual content.

Francisco Diogo Rocha Sousa: contributed to data collection, data analysis and interpretation and to a critical revision, adding intellectual content.

José Maurício Brandão Quintão: contributed to data collection, data analysis and interpretation and to a critical revision, adding intellectual content.

Leandra Regina Gonçalves: contributed to data collection, data analysis and interpretation and to a critical revision, adding intellectual content.

Maíra de Campos Gorgulho Padgurschi: contributed to data collection, data analysis and interpretation and to a critical revision, adding intellectual content.

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Paula Felício Drummond de Castro: contributed to manuscript preparation and to a critical revision, adding intellectual content.

Isabela Lima dos Santos: contributed to manuscript preparation and to a critical revision, adding intellectual content.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Infracommunities of bat flies (Diptera: Streblidae and Nycteribiidae) of bats (Mammalia: Chiroptera) in three conservation units in the State of Amapá, Brazil

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**Abstract:** Diptera belonging to the families Nycteribiidae and Streblidae are hematophagous ectoparasites, exclusively of bats. The Amapá State is located in the North Region of Brazil. The Amapá represents one of with the lowest rates of deforestation in the country, with approximately 80% of the territory preserved. Within the State, there are conservation units, Floresta Nacional do Amapá (FNA), Parque Nacional Montanhas do Tumucumaque (PNMT) and Reserva de desenvolvimento Sustentável do Rio Iratapuru (RDSI). These areas represent priority areas for the Conservation of Amazonian Biodiversity. Recently in Amapá studied on the diversity and distribution of bats, however, there are no records of associated arthropod ectoparasites. Therefore, the objective was to describe the pattern of association of Nycteribiidae and Streblidae species on bats in three protected areas of Amapá. Bats were collected through mist nets, where each host had its body visually revised and the Diptera collected were identified through a specific bibliography. One thousand and eighty- six individuals of bats flies were identified, distributed in 52 species (Streblidae = 49 and Nycteribiidae = three). Among the three infracommunities, the PNMT infracommunities was the largest found with abundance and richness of bats flies. All species found, represent new occurrences for the Amapá, being these, 18 new records for the North Region and seven new records for Brazil.

**Keywords:** Hippoboscoidea, Host-parasite relationship, Amazonia, Geographic distribution.

## Infracomunidades de moscas ectoparasitas (Diptera: Streblidae e Nycteribiidae) de morcegos (Mammalia: Chiroptera) em três unidades de conservação no Estado do Amapá, Brasil

**Resumo:** Dípteros pertencentes as famílias Nycteribiidae e Streblidae são ectoparasitos hematófagos, sendo exclusivamente de morcegos. O estado do Amapá localiza-se na região Norte do Brasil e representa um dos estados com as menores taxas de desmatamento do país, com aproximadamente 80% do território preservado. Dentro do Estado, existem as unidades de conservação, a Floresta Nacional do Amapá (FNA), o Parque Nacional Montanhas do Tumucumaque (PNMT) e a Reserva de Desenvolvimento Sustentável do Rio Iratapuru (RDS) que representam áreas prioritárias para a Conservação da Biodiversidade da Amazônia. Estudos sobre a diversidade e a distribuição de morcegos têm sido estudada recentemente no Amapá, no entanto, não existem registros de artrópodes ectoparasitos associados. Sendo assim, o objetivo foi descrever padrão de associação das espécies de Nycteribiidae e Streblidae sobre morcegos em três unidades de conservação do Amapá. Os morcegos foram coletados por meio de redes de neblina, onde cada hospedeiro teve seu corpo revisado visualmente e os dípteros coletados foram identificados através de bibliografia específica. Foram identificados 1086 indivíduos de moscas ectoparasitas, distribuídos em 52 espécies (Streblidae = 49 e Nycteribiidae = três). Entre as três infracomunidades, a infracomunidade de PNMT foi a maior encontrada tanto com relação a abundancia como em riqueza de dípteros ectoparasitas. Todas as espécies encontradas, representam novas ocorrências para o Estado do Amapá, sendo essas, 18 novos registros para a Região Norte e sete novos registros para o Brasil.

**Palavras-chave:** Hippoboscoidea; Interações parasito-hospedeiro; Amazônia; Distribuição geográfica.

## Introduction

Dipterans of the families Nycteribiidae and Streblidae are hematophagous ectoparasites, exclusively of bats (Wenzel et al. 1966, Marshall 1982). Nycteribiidae presents your distribution mainly in the Old World. In the American Continent, there are only two genera, *Basilia* Miranda-Ribeiro, 1903 with 53 species and *Hershkovitzia* Guimarães & D'Andretta, 1956 with four species (Graciolli et al. 2007, Graciolli & Dick 2009, Graciolli 2010). In Brazil were recorded 24 species of *Basilia* and two of *Hershkovitzia* (Graciolli 2019a). The Streblidae family occurs mainly in the New World. There are about 299 species described, with three subfamilies, Nycterophiliinae, Trichobiinae and Streblinae exclusive to the New World (Dick & Miller 2010, Dick et al. 2016).

Chiroptera represents the second largest mammal order, after Rodentia. They harbor exclusive ectoparasitic arthropod communities (Marshall 1982, Guerrero 1993). In Brazil, there are known 182 species in 69 genera of bats (Nogueira et al. 2018). In the North Region were recorded 144 bats in 63 genera (Reis et al. 2017), with 73 species recorded for Amapá State (Martins et al. 2011).

The Amapá State is located in the North Region of Brazil, on the delta of the Amazon River and making the transition to the highlands of the Guiana Shield. The State represents one of the lowest rates of deforestation in the North Region (INPE 2015). Furthermore, a large proportion of the State's territory (60%) is protected in 19 conservation units, divided into federal, state, municipal, full protection and sustainable use units (Drummond et al. 2008).

The diversity and distribution of bats have been studied recently in Amapá (Martins et al. 2006, 2011). However, there are no records of bat flies. Due to that, the objective of this work was to conduct a survey and describe the pattern of association of Nycteribiidae and Streblidae species on bats in three conservation units in the State of Amapá.

## Materials and Methods

The bat flies examined came from a rapid biological inventory of bats performed by Martins et al. (2006, 2011). The inventory followed the Rapid Assessment Program (RAP) developed by Conservation International.

### 1. Study area

Within existing protected areas in the Amapá State, Floresta Nacional do Amapá (FNA), Parque Nacional Montanhas do Tumucumaque (PNMT) and Reserva de Desenvolvimento Sustentável do Rio Iratapuru (RDSI) were chosen to be inventoried. They represent areas considered priorities for the conservation of the biodiversity of the Amazon (Capobianco et al. 2001). To select the collection sites locals, in each conservation area, were used pictures of a satellite to identify the principal vegetation formations. Prioritized regions with more than one environment, or a transition between them (Martins et al. 2006).

The FNA is located right in the center of the State and boasts an area of 4,120,000,000 m<sup>2</sup> approximately. The unit is accessed only inland waterway and feature predominantly vegetation of the upland forest, with some stains of flooded forests, "tabocais" (forest of bamboo) and rocky outcrop. The altitude varies from 80 to 450 m above sea level. This area was inventoried between four and 15th of August 2004, totalizing 21600 m<sup>2</sup>h (coordinates of collecting points: 01°18'07"N, 51°35'17"W; 01°06'37"N, 51°53'37"W).

The PNMT is located in the northwest region of the State, with an area of 38,700,000,000 m<sup>2</sup> approximately. It represents the largest continuous rainforest park in the world and therefore it is accessible only by air or waterways. The vegetation has a predominance of dense forests of mainland, igapó forest (dense ombrophylous alluvial forests), slopes forests and dried formations associated with rocky outcrop. The relief varies from 100 to 400 m above sea level. In this area, two expeditions were carried out. The first occurred between 16 and 25th of September 2004, and the second expedition occurred between 10 and 20th of January 2005. Totalizing in both expedition 37800 m<sup>2</sup>h (coordinates of collecting points: 01°35'45"N, 52°29'32"W; 02°11'36"N, 54°35'15"W; 03°12'59"N, 52°01'10"W; 01°23'13"N, 51°55'39"W; 01°50'41"N, 52°44'28"W).

The RDSI is located in the southwest of the State, with about of 8,060,000,000 m<sup>2</sup>. In addition, only accessible by waterways and features a predominance of an upland forest with a high concentration of Castanheiras-do-Brazil (*Bertholletia excelsa* Humb. & Bonpl., 1808) and therefore there are traditional populations who commercially exploit the resource. The relief varies from 100 to 500 m above sea level. This area was inventoried between 12 and 20th of November, totaling 16200 m<sup>2</sup>h (coordinates of collecting points: 00°16'35"N, 53°06'24"W; 00°18'36"S, 52°26'24"W; 0°34'45"N, 52°19'08.3"W).

### 2. Capture of host and bat flies

The details of the capture of bat were described in Martins et al. (2006, 2011). Each night, 10 mist-net (12 x 2.5) were opened from 18:00 to 24:00h. In each host had the body visually reviewed and the bat flies were removed with the help of tweezers and stored in Eppendorf type tubes containing ethanol 70%, and a label with the data collection. The identification of bat flies was made using the keys presented by Guimarães & D'Andretta (1956), Guimarães (1966, 1977) and Guerrero (1993, 1994a, b, 1995a, b, 1996). The bat flies identified were deposited in the Coleção Zoológica de Referência, Universidade Federal do Mato Grosso do Sul, Campo Grande.

### 3. Data analysis

The description of the infracommunities (community of parasite infrapopulations in a single host) (Bush et al. 1997) found in common in each conservation. It was analyzed only the hosts species with at least two infested bats. In each infracommunities, it was calculated the absolute number of occurrence and relative frequency (quotient between the absolute frequency of parasites and the total number of the population in each host). Information about all infracommunities are described in the supplementary material (Table S1).

## Results

At the total, it was found 207 infested bats for the three conservation units. Among the bats species, only 10 were in common for the tree localities. In FNA from 52 bats of 18 species were infested by 29 species of bat flies. In RDSI, we found 47 bats for 17 species infested by 28 species of bat flies. Finally, PNMT, 108 individuals of 25 species were infested by 41 species of bat flies.

In relation to the bat flies, were identified 1086 individuals, distributed in 50 species and two subspecies (Stebliidae = 49 and Nycteribiidae = 3). Nycteribiidae family was represented by three

species of *Basilia*. For Streblidae, *Trichobius dugesioides dugesioides* Wenzel, 1966 (n = 134) was the most abundant bat fly, follow of *Trichobius dugesioides phyllostomus* Guerrero, 1998 (n = 110), *Trichobius costalimai* Guimarães, 1938 (n = 98) and *Mastoptera minuta* (Costa Lima, 1921) (n = 90) (Tables 1, 2 and 3).

Four species of bats, *Pteronotus parnelli* (Gray, 1843), *Artibeus planirostris* (Spix, 1823), *Phyllostomus elongatus* (Geoffroy, 1810) and *Sturnira tildae* De la Torre, 1959, were found in all Conservation Units. We recorded a total of 39 composition of infracommunities bat flies

(FLONA = 11, PNMT = 16, RDSI = 12). *P. parnelli* presented the highest quantity of different parasites composition, with 11 infracommunities in all conservation's units. Despite this, the composition of the bat flies species infracommunities were different in each conservation unit. *Strebla consocia* Wenzel, 1976, *Trichobius dugesioides phyllostomus*, *Trichobius longipes* (Rudow, 1871) were in common for the three units. *Trichobius costalimai* and *Trichobius jobligi* Wenzel, 1966 were exclusive for PNMT and *Mastoptera minuta* and *Strebla galindoi* Wenzel, 1966 for RDSI (Table 4).

**Table 1.** Bats species, with a number of infested individuals. Their respective bat flies species and abundance in Floresta Nacional do Amapá Conservation Unit. Ni = number of individuals infested. N = number of individuals.

Family/Specie of host	Ni	Bat fly	N
<b>Mormoopidae</b>			
<i>Pteronotus parnelli</i> (Gray, 1843)	8	<i>Trichobius johnsonae</i> Wenzel, 1966	19
		<i>Trichobius caecus</i> Edwards, 1918	12
		<i>Nycterophilia parnelli</i> Wenzel, 1966	4
<b>Phyllostomidae</b>			
<i>Anoura geoffroyi</i> Gray, 1838	1	<i>Anastrebla modestini</i> Wenzel, 1966	1
<i>Artibeus planirostris</i> (Spix, 1823)	4	<i>Aspidoptera phyllostomatis</i> (Perty, 1833)	7
		<i>Megistopoda aranea</i> (Coquillett, 1899)	3
<i>Carollia brevicauda</i> (Schinz, 1821)	1	<i>Speiseria peytonae</i> Wenzel, 1976	1
		<i>Trichobius joblingi</i> Wenzel, 1966	1
<i>Carollia perspicillata</i> (Linnaeus, 1758)	3	<i>Strebla guajiro</i> (García & Casal, 1965)	4
		<i>Trichobius joblingi</i> Wenzel, 1966	3
<i>Chrotopterus auritus</i> (Peters, 1856)	1	<i>Strebla chrotopteri</i> Wenzel, 1976	1
<i>Desmodus rotundus</i> (Geoffroy, 1810)	1	<i>Strebla wiedemanni</i> (Kolenati, 1856)	2
<i>Glossophaga soricina</i> (Pallas, 1766)	1	<i>Trichobius uniformis</i> Curran, 1935	1
<i>Hsunycteris thomasi</i> (Allen, 1904)	4	<i>Trichobius lonchophyllae</i> Wenzel, 1966	26
		<i>Anastrebla</i> sp.	2
		<i>Trichobius</i> sp. (group dugesii)	1
<i>Lionycteris spurrelli</i> (Thomas, 1913)	1	<i>Trichobius lionycteridis</i> Wenzel, 1966	1
<i>Mimon bennettii</i> (Gray, 1838)	1	<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	3
<i>Mimon crenulatum</i> (Geoffroy, 1803)	3	<i>Basilia mimoni</i> Theodor & Peterson, 1964	2
		<i>Mastoptera minuta</i> (Costa Lima, 1921)	3
<i>Phyllostomus elongatus</i> (Geoffroy, 1810)	4	<i>Strebla consocia</i> Wenzel, 1976	9
		<i>Trichobius dugesioides phyllostomus</i> Guerrero, 1998	3
		<i>Trichobius longipes</i> (Rudow, 1871)	1
<i>Rhinophylla pumilio</i> Peters, 1865	1	<i>Neotrichobius delicatus</i> (Machado-Allisson, 1966)	1
<i>Sturnira tildae</i> De La Torre, 1959	2	<i>Megistopoda proxima</i> (Séguy, 1926)	4
		<i>Aspidoptera falcata</i> Wenzel, 1976	8
<i>Tonatia saurophila</i> Koopman & Williams, 1951	1	<i>Strebla galindoi</i> Wenzel, 1966	5
<i>Trachops cirrhosus</i> (Spix, 1823)	10	<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	46
		<i>Strebla mirabilis</i> (Waterhouse, 1879)	2
		<i>Speiseria magniocularis</i> Wenzel, 1976	3
		<i>Megistopoda aranea</i> (Coquillett, 1899)	1
<i>Trinycteris nicefori</i> (Sanborn, 1949)	5	<i>Strebla alvarezi</i> Wenzel, 1966	1
		<i>Strebla obtusa</i> Wenzel, 1976	18
		<i>Parastrebla handleyi</i> Wenzel, 1966	3

**Table 2.** Bats species, with a number of infested individuals. Their respective bat flies species and abundance in Parque Nacional Montanhas do Tumucumaque Conservation Unit. Ni = number of individuals infested. N = number of individuals.

Family/Specie of host	Ni	Bat fly	N
<b>Emballonuridae</b>			
<i>Rhynchonycteris naso</i> (Wied-Neuwied, 1820)	1	<i>Strebla asternalis</i> Wenzel, 1976	1
<i>Saccopteryx bilineata</i> (Temminck, 1838)	1	<i>Strebla asternalis</i> Wenzel, 1976	1
<b>Mormoopidae</b>			
<i>Pteronotus parnellii</i> (Gray, 1843)	7	<i>Trichobius johnsonae</i> Wenzel, 1966	18
		<i>Trichobius caecus</i> Edwards, 1918	12
		<i>Nycterophilina parnelli</i> Wenzel, 1966	2
<b>Noctilionidae</b>			
<i>Noctilio leporinus</i> (Linnaeus, 1758)	1	<i>Noctiliostrebla aitkeni</i> Wenzel, 1966	18
		<i>Paradyschiria lineata</i> Kessel, 1925	8
<b>Phyllostomidae</b>			
<i>Anoura geoffroyi</i> Gray, 1838	1	<i>Strebla curvata</i> Wenzel, 1976	1
		<i>Speiseria ambigua</i> Kessel, 1925	1
		<i>Trichobius tiptoni</i> Wenzel, 1976	1
<i>Artibeus obscurus</i> (Schinz, 1821)	1	<i>Neotrichobius bisetosus</i> Wenzel, 1976	1
<i>Artibeus planirostris</i> (Spix, 1823)	14	<i>Aspidoptera phyllostomatis</i> (Perty, 1833)	15
		<i>Megistopoda aranea</i> (Coquillett, 1899)	22
		<i>Metelasmus pseudopterus</i> Coquillett, 1907	2
<i>Carollia perspicillata</i> (Linnaeus, 1758)	1	<i>Strebla guajiro</i> (García & Casal, 1965)	1
		<i>Trichobius joblingi</i> Wenzel, 1966	2
<i>Chrotopterus auritus</i> (Peters, 1856)	5	<i>Strebla chrotopteri</i> Wenzel, 1976	14
		<i>Trichobius johnsonae</i> Wenzel, 1966	1
		<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	12
<i>Desmodus rotundus</i> (Geoffroy, 1810)	4	<i>Strebla wiedemanni</i> (Kolenati, 1856)	16
		<i>Trichobius parasiticus</i> Gervais, 1844	8
<i>Glossophaga soricina</i> (Pallas, 1766)	2	<i>Trichobius uniformis</i> Curran, 1935	2
		<i>Trichobius dugesii</i> Townsend, 1891	3
		<i>Strebla harderi</i> Wenzel, 1976	1
<i>Lophostoma brasiliense</i> Peters, 1866	1	<i>Mastoptera minuta</i> (Costa Lima, 1921)	7
		<i>Trichobius silvicolae</i> Wenzel, 1976	1
<i>Lophostoma silvicola</i> d'Orbigny, 1836	7	<i>Mastoptera minuta</i> (Costa Lima, 1921)	23
		<i>Trichobius silvicolae</i> Wenzel, 1976	13
		<i>Pseudostrebla riberoi</i> Costa Lima, 1921	1
<i>Mimon crenulatum</i> (Geoffroy, 1803)	4	<i>Basilina mimoni</i> Theodor & Peterson, 1964	13
<i>Phylloderma sternops</i> (Peters, 1865)	1	<i>Strebla christinae</i> Wenzel, 1966	2
<i>Phyllostomus discolor</i> (Wagner, 1843)	14	<i>Strebla hertigi</i> Wenzel, 1966	31
		<i>Trichobius costalimai</i> Guimarães, 1938	63
		<i>Trichobioides perspicillatus</i> (Pessôa & Galvão, 1936)	7
		<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	2
		<i>Strebla consocia</i> Wenzel, 1976	34
<i>Phyllostomus elongatus</i> (Geoffroy, 1810)	17	<i>Trichobius dugesioides phyllostomus</i> Guerrero, 1998	76
		<i>Trichobius longipes</i> (Rudow, 1871)	9
		<i>Trichobius costalimai</i> Guimarães, 1938	4
		<i>Trichobius joblingi</i> Wenzel, 1966	2
		<i>Trichobius</i> sp. <i>parasiticus</i> complex	10



Continuation Table 2.

Family/Specie of host	Ni	Bat fly	N
<i>Phyllostomus hastatus</i> (Pallas, 1767)	4	<i>Strebla consocia</i> Wenzel, 1976	4
		<i>Trichobius longipes</i> (Rudow, 1871)	8
<i>Rhinophylla pumilio</i> Peters, 1865	2	<i>Neotrichobius delicatus</i> (Machado-Allisson, 1966)	2
<i>Sturnira tildae</i> De La Torre, 1959	2	<i>Megistopoda proxima</i> (Séguy, 1926)	1
		<i>Megistopoda aranea</i> (Coquillett, 1899)	1
<i>Tonatia saurophila</i> Koopman & Williams, 1951	1	<i>Strebla galindoi</i> Wenzel, 1966	3
<i>Trachops cirrhosus</i> (Spix, 1823)	9	<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	50
		<i>Strebla mirabilis</i> (Waterhouse, 1879)	1
		<i>Speiseria magniocus</i> Wenzel, 1976	9
<i>Trinycteris nicefori</i> (Sanborn, 1949)	1	<i>Strebla obtusa</i> Wenzel, 1976	2
<b>Vespertilionidae</b>			
<i>Myotis albescens</i> (Geoffroy, 1806)	5	<i>Basilina dunni</i> Curran, 1935	14
<i>Myotis riparius</i> Handley, 1960	2	<i>Basilina anceps</i> Guimarães & Andretta, 1956	3

All species recorded in this work represent the first occurrence to the State of Amapá. Among these, 17 species and two genera of Streblidae are recorded for the first time in the North Region. This increased to 58 and 15 the number of species and genera respectively for the region. In relation to the Nycteribiidae, *Basilina anceps* Guimarães & D'Andretta, 1956 is registered for the first time for the North Region, increased to eight the number of species for the region. *Strebla asternalis* Wenzel, 1976, *Strebla matsoni* Wenzel, 1976, *Paradyschiria lineata* Kessel, 1925, *Speiseria peytonae*, Wenzel 1976, *Speiseria magniocus* Wenzel, 1976, *Trichobius lionycteridis* Wenzel, 1966 and *Trichobius macrophylli* Wenzel 1966 are recorded for the first time in the Brazil, increasing to 92 the number of Streblidae species for the country (Table 5).

## Discussion

The richness of species found in this work confirms the information that the Brazilian Amazon presents a very large diversity of species of bat flies (Gracioli & Bernard 2002, Gracioli & Linardi 2002, Gracioli 2019a, 2019b). With the new occurrences of this work, the North Region remains with 58 species of Streblidae, with the same number in species to the Central-West Region. The Southeast Region has 53 species recorded, follow with Northeast with 48 and finally the South Region with 33 species (Barbier & Bernard 2017, Gracioli 2019b, Barbier et al. 2019). Despite this, the regions Central-West and Southeast have a greater amount in a number of genera of Streblidae, each with 18 genera (Gracioli 2019b). In the Northeast are found 14 genera, in the South 13 and finally, the North with the current work remains with 15 genera of Streblidae (Barbier & Bernard 2017, Gracioli 2019b, Barbier et al. 2019).

With the family Nycteribiidae this relationship is reversed. The Southeast represents the Region richer, with 18 species, following with the South Region with 13 species, Central-West with 11, the Northeast with four species and with the current work, the North Region remains with eight species of Nycteribiidae (Barbier & Bernard 2017, Gracioli 2019a, Barbier et al. 2019). Although the North and Northeast Regions not be as expressive in quantity of species, it represents the only regions that in addition the genus *Basilina*, also occurs the genus *Herskovitzia*

(*H. cabala* Peterson & Lacey, 1985, *H. inaequalis* Theodor, 1967 and *Herskovitzia* sp.) (Peterson & Lacey 1985, Gracioli 2001, Gracioli & Bernard 2002, Santos et al. 2013, Barbier & Bernard 2017, Gracioli 2019a).

Previously, Pará was the State with the highest number of species of Streblidae for the North Region of Brazil, with 28 species. Then stay Rondônia and Roraima with 14 species each and Amazonas with nine species (Gracioli 2019b). The high amount of species of bat flies found to Amapá (N = 52), demonstrates that much of the diversity species of bat flies for the region remains unknown (Gracioli & Linardi 2002, Gracioli & Bernard 2002, Gracioli 2019b).

The new registers for Brazil were already known for another country of the South and Central America. The species *S. asternalis* and *S. matsoni* where known only for Venezuela (Wenzel 1976, Guerrero 1994b, Frank et al. 2014). *S. magniocus* still be known for Venezuela, has also been found in Peru (Wenzel 1976, Guerrero 1994b, Frank et al. 2014). On the contrary, some species have a large distribution in South and Central America, but there was no occurrence for Brazil. Such as, *S. peytonae* which that is known for Bolivia, Colombia, Peru, Venezuela, Belize and Honduras (Wenzel 1976, Guerrero 1994b, Dick 2013, Frank et al. 2014, Dick et al. 2016) (Table 5).

In the present work, *Trichobioides perspicillatus* (Pessôa & Galvão, 1936) represents the first occurrence of genus and species for the North Region of Brazil. Previously the species still had occurrence for the Regions Northeast (Bahia, Maranhão, Pernambuco and Sergipe), Central-West (Distrito Federal and Mato Grosso do Sul) and Southeast (Minas Gerais) (Gracioli 2019b). Despite not have previously registered for the North Region, previous work has already pointed out that the species is found in the Amazonian ecoregion (Barbier & Bernard 2017). *Anastrebla modestini* Wenzel, 1966 also represents the first occurrence of genus/species for the North Region. Previously this species was described only for the regions South (Paraná, Santa Catarina and Rio Grande do Sul), Southeast (São Paulo, Rio de Janeiro and Minas Gerais) and Central-West (Distrito Federal and Goiás) (Gracioli 2019b). Its restricted distribution was probably the distribution of the host (*Anoura geoffroyi* Gray, 1838), which for North Region is known only for States of Amapá and Pará (Zortéa & Velazco 2017) (Table 5).

**Table 3.** Bats species, with a number of infested individuals. Their respective bat flies species and abundance in Reserva de Desenvolvimento Sustentável Rio Iratapuru Conservation Unit. Ni = number of individuals infested. N = number of individuals.

Family/Specie of host	Ni	Bat fly	N
<b>Mormoopidae</b>			
<i>Pteronotus parnellii</i> (Gray, 1843)	9	<i>Trichobius johnsonae</i> Wenzel, 1966	7
		<i>Trichobius caecus</i> Edwards, 1918	54
		<i>Nycterophilina parnelli</i> Wenzel, 1966	2
		<i>Mastoptera minuta</i> (Costa Lima, 1921)	1
		<i>Neotrichobius delicatus</i> (Machado-Allisson, 1966)	1
<i>Pteronotus</i> sp.	1	<i>Nycterophilina parnelli</i> Wenzel, 1966	4
<b>Phyllostomidae</b>			
<i>Artibeus lituratus</i> (Olfers, 1818)	2	<i>Megistopoda aranea</i> (Coquillett, 1899)	1
		<i>Paratrichobius longicrus</i> (Miranda-Ribeiro, 1907)	2
<i>Artibeus planirostris</i> (Spix, 1823)	5	<i>Aspidoptera phyllostomatis</i> (Perty, 1833)	7
		<i>Megistopoda aranea</i> (Coquillett, 1899)	2
<i>Carollia perspicillata</i> (Linnaeus, 1758)	1	<i>Trichobius joblingi</i> Wenzel, 1966	1
		<i>Speiseria ambigua</i> Kessel, 1925	1
<i>Chrotopterus auritus</i> (Peters, 1856)	2	<i>Strebla chrotopteri</i> Wenzel, 1976	3
		<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	2
<i>Lamproncyteris brachyotis</i> (Dobson, 1879)	1	<i>Strebla obtusa</i> Wenzel, 1976	5
<i>Lionycteris spurrelli</i> (Thomas, 1913)	1	<i>Trichobius lionycteridis</i> Wenzel, 1966	2
<i>Lophostoma silvicola</i> d'Orbigny, 1836	3	<i>Mastoptera minuta</i> (Costa Lima, 1921)	1
		<i>Trichobius silvicolae</i> Wenzel, 1976	3
		<i>Strebla galindoi</i> Wenzel, 1966	1
<i>Macrophyllum macrophyllum</i> (Schinz, 1821)	2	<i>Strebla matsoni</i> Wenzel, 1976	1
		<i>Trichobius macrophylli</i> Wenzel, 1966	8
<i>Micronycteris</i> sp.	1	<i>Strebla hoogstraali</i> Wenzel, 1966	6
<i>Phyllostomus discolor</i> (Wagner, 1843)	4	<i>Strebla hertigi</i> Wenzel, 1966	3
		<i>Trichobius costalimai</i> Guimarães, 1938	31
		<i>Trichobioides perspicillatus</i> (Pessoa & Galvão, 1936)	4
<i>Phyllostomus elongatus</i> (Geoffroy, 1810)	6	<i>Strebla consocia</i> Wenzel, 1976	14
		<i>Trichobius dugesioides phyllostomus</i> Guerrero, 1998	31
		<i>Trichobius longipes</i> (Rudow, 1871)	22
		<i>Strebla galindoi</i> Wenzel, 1966	5
		<i>Mastoptera minuta</i> (Costa Lima, 1921)	34
<i>Phyllostomus hastatus</i> (Pallas, 1767)	1	<i>Strebla consocia</i> Wenzel, 1976	1
		<i>Trichobius longipes</i> (Rudow, 1871)	9
		<i>Mastoptera minuta</i> (Costa Lima, 1921)	21
<i>Rhinophylla pumilio</i> Peters, 1865	2	<i>Neotrichobius delicatus</i> (Machado-Allisson, 1966)	3
<i>Sturnira tildae</i> De La Torre, 1959	2	<i>Megistopoda proxima</i> (Séguy, 1926)	4
		<i>Aspidoptera falcata</i> Wenzel, 1976	1
<i>Tonatia saurophila</i> Koopman & Williams, 1951	2	<i>Strebla galindoi</i> Wenzel, 1966	5
		<i>Trichobius silvicolae</i> Wenzel, 1976	1
<i>Trachops cirrhosus</i> (Spix, 1823)	1	<i>Trichobius dugesioides dugesioides</i> Wenzel, 1966	19
<b>Vespertilionidae</b>			
<i>Myotis albescens</i> (Geoffroy, 1806)	1	<i>Basilia dunni</i> Curran, 1935	2

**Table 4.** Bat species with their respective infracommunity. N = Absolute number of occurrence. AR% = Values of relative abundance.

Infracommunity FNA	N	AR%	Infracommunity PNMT	N	AR%	Infracommunity RDS	N	AR%
<b><i>Pteronotus parnellii</i> (N 8)</b>			<b><i>Pteronotus parnellii</i> (N 7)</b>			<b><i>Pteronotus parnellii</i> (N 9)</b>		
<i>T. johnsonae</i>	3	37%	<i>T. johnsonae</i>	5	71%	<i>T. caecus</i>	6	66%
<i>T. johnsonae</i> + <i>N. parnelli</i>	3	37%	<i>T. johnsonae</i> + <i>N. parnelli</i>	1	14%	<i>T. johnsonae</i>	1	11%
<i>T. caecus</i>	2	25%	<i>T. caecus</i>	1	14%	<i>T. caecus</i> + <i>M. minuta</i>	1	11%
						<i>N. delicatus</i> + <i>N. parnelli</i> + <i>T. johnsonae</i>	1	11%
<b><i>Artibeus planirostris</i> (N 4)</b>			<b><i>Artibeus planirostris</i> (N 14)</b>			<b><i>Artibeus planirostris</i> (N 5)</b>		
<i>A. phyllostomatis</i>	2	50%	<i>A. phyllostomatis</i>	7	50%	<i>A. phyllostomatis</i>	3	60%
<i>M. aranea</i>	1	25%	<i>M. aranea</i>	4	28%	<i>A. phyllostomatis</i> + <i>M. aranea</i>	2	40%
<i>A. phyllostomatis</i> + <i>M. aranea</i>	1	25%	<i>A. phyllostomatis</i> + <i>M. aranea</i> + <i>M. pseudopterus</i>	2	14%			
			<i>A. phyllostomatis</i> + <i>M. aranea</i>	1	7%			
<b><i>Phyllostomus elongatus</i> (N 4)</b>			<b><i>Phyllostomus elongatus</i> (N 17)</b>			<b><i>Phyllostomus elongatus</i> (N 6)</b>		
<i>S. consocia</i> + <i>T. dugesioides</i> <i>phyllostomus</i>	2	50%	<i>S. consocia</i> + <i>T. dugesioides</i> <i>phyllostomus</i>	7	41%	<i>T. dugesioides</i> <i>phyllostomus</i> + <i>S. consocia</i> + <i>T. longipes</i> + <i>M. minuta</i>	3	50%
<i>S. consocia</i>	1	25%	<i>T. dugesioides</i> <i>phyllostomus</i>	3	17%	<i>S. consocia</i> + <i>T. dugesioides</i> <i>phyllostomus</i>	1	16%
<i>T. longipes</i>	1	25%	<i>T. dugesioides</i> <i>phyllostomus</i> + <i>S. consocia</i> + <i>T. longipes</i>	3	17%	<i>T. dugesioides</i> <i>phyllostomus</i> + <i>S. galindoi</i> + <i>T. longipes</i>	1	16%
			<i>T. costalimai</i>	1	5%	<i>T. dugesioides</i> <i>phyllostomus</i>	1	16%
			<i>T. dugesioides</i> <i>phyllostomus</i> + <i>T. longipes</i>	1	5%			
			<i>S. consocia</i> + <i>T. longipes</i> + <i>Trichobius</i> sp. <i>parasiticus</i> complex	1	5%			
			<i>T. dugesioides</i> <i>phyllostomus</i> + <i>S. consocia</i> + <i>T. joblingi</i>	1	5%			
<b><i>Sturnira tildae</i> (N 2)</b>			<b><i>Sturnira tildae</i> (N 2)</b>			<b><i>Sturnira tildae</i> (N 2)</b>		
<i>M. proxima</i>	1	50%	<i>M. proxima</i>	1	50%	<i>M. proxima</i>	1	50%
<i>M. proxima</i> + <i>A. falcata</i>	1	50%	<i>M. aranea</i>	1	50%	<i>A. falcata</i>	1	50%

Some species have significantly increased its distribution in Brazil. Previously *T. lonchophyllae* Wenzel, 1966 and *Strebla harderi* Wenzel, 1976 were known only for the regions Central-West (Distrito Federal) and Southeast (Minas Gerais and Rio de Janeiro - *T. lonchophyllae*). *T. johnsonae* Wenzel, 1966, *B. anceps* and *Nycterophila parnelli* Wenzel, 1966 were recorded only in the regions Central-West (Distrito Federal, Goiás – *B. anceps*, Mato Grosso – *N. parnelli* and Mato Grosso do Sul – *T. johnsonae*). *Strebla hoogstraali* Wenzel, 1966 was known only for the State of Maranhão. The other new records for the North Region (*Strebla chrotopteri* Wenzel, 1976, *Trichobius tiptoni* Wenzel, 1976 and *Trichobius dugesii* Townsend, 1891) already had a wider distribution

along the other regions of Brazil (Graciolli 2019b). Although, the specie *S. curvata* Wenzel, 1976 already has an occurrence for the North Region, the identification is in dubious (Santos et al. 2012) (G. Graciolli personal communication) (Table 5).

In PNMT was found in greater richness and abundance of bat flies (41 species, N = 559), bat (25 species, N = 108) and consequently of infracommunities (N = 16) (Tables 2 and 4). This is probably the area have received two expeditions, while the others had only an expedition (Martins et al. 2006, 2011). The frequency of species richness in the infracommunities is similar than the frequency found by others works (Santos et al. 2013, Barbier & Graciolli 2016, Dornelles & Graciolli 2017).

**Table 5.** Registered of new occurrences for the Region North and for Brazil. (<sup>1</sup>Wenzel 1966, <sup>2</sup>Wenzel 1976, <sup>3</sup>Guerrero 1994b, <sup>4</sup>Dick 2013, <sup>5</sup>Frank et al. 2014, <sup>6</sup>Dick et al. 2016, <sup>7</sup>Graciolli 2019b).

Bat fly	New occurrence	Previous occurrence
<i>S. peytonae</i> Wenzel, 1976	New occurrence for Brazil	Bolivia <sup>3,4,5</sup> , Colombia <sup>5</sup> , Peru <sup>3,5,6</sup> , Venezuela <sup>2,3,5,6</sup> , Belize <sup>5</sup> , and Honduras <sup>4,5,6</sup>
<i>S. magniocularis</i> Wenzel, 1976	New occurrence for Brazil	Peru <sup>3</sup> and Venezuela <sup>2,3,5</sup>
<i>S. asternalis</i> Wenzel, 1976	New occurrence for Brazil	Venezuela <sup>2,3,5</sup>
<i>P. lineata</i> Kessel, 1925	New occurrence for Brazil	Venezuela <sup>2,3,5</sup> , Panamá <sup>5,6</sup> , French Guiana <sup>6</sup> and Peru <sup>6</sup>
<i>T. lionycteridis</i> Wenzel, 1966	New occurrence for Brazil	Belize <sup>5</sup> , Venezuela <sup>2,3,5</sup> , Panamá <sup>1,4,5</sup> and Peru <sup>6</sup>
<i>S. matsoni</i> Wenzel, 1976	New occurrence for Brazil	Venezuela <sup>2,3,5</sup>
<i>T. macrophylli</i> Wenzel, 1966	New occurrence for Brazil	Panamá <sup>1,3,5</sup> , Costa Rica <sup>5</sup> , Venezuela <sup>2</sup> and Bolivia <sup>3</sup>
<i>A. modestini</i> Wenzel, 1966	New occurrence of Genus for the Region North	South (Paraná, Santa Catarina and Rio Grande do Sul), Southeast (São Paulo, Rio de Janeiro and Minas Gerais) and Midwest (Distrito Federal and Goiás) <sup>7</sup>
<i>T. perspicillatus</i> (Pessôa & Galvão, 1936)	New occurrence of Genus for the Region North	Northeast (Bahia, Maranhão, Pernambuco and Sergipe), Midwest (Distrito Federal and Mato Grosso do Sul) and Southeast (Minas Gerais) <sup>7</sup>
<i>T. lonchophyllae</i> Wenzel, 1966	New occurrence for the Region North	Midwest (Distrito Federal) and Southeast (Minas Gerais and Rio de Janeiro) <sup>7</sup>
<i>T. tiptoni</i> Wenzel, 1979	New occurrence for the Region North	South (Paraná and Rio Grande do Sul), Southeast (Minas Gerais, Rio de Janeiro and São Paulo) and Midwest (Distrito Federal, Goiás and Mato Grosso do Sul) <sup>7</sup>
<i>S. chropteri</i> Wenzel, 1976	New occurrence for the Region North	Midwest (Distrito Federal and Mato Grosso do Sul), Southeast (Minas Gerais and São Paulo) and South (Paraná) <sup>7</sup>
<i>T. dugesii</i> Townsend, 1891	New occurrence for the Region North	South (Rio Grande do Sul), Southeast (Minas Gerais, Rio de Janeiro and São Paulo), Midwest (Distrito Federal, Goiás and Mato Grosso do Sul) and Northeast (Maranhão and Paraíba) <sup>7</sup>
<i>S. harderi</i> Wenzel, 1976	New occurrence for the Region North	Midwest (Distrito Federal) and Southeast (Minas Gerais) <sup>7</sup>
<i>B. anceps</i> Guimarães & D'Andretta, 1956	New occurrence for the Region North	Midwest (Distrito Federal and Goiás) <sup>7</sup>
<i>T. johnsonae</i> Wenzel, 1966	New occurrence for the Region North	Midwest (Distrito Federal and Mato Grosso do Sul) <sup>7</sup>
<i>N. parnelli</i> (Gray, 1843)	New occurrence for the Region North	Midwest (Mato Grosso and Distrito Federal) <sup>7</sup>
<i>S. hoogstraali</i> Wenzel, 1966	New occurrence for the Region North	Northeast (Maranhão) <sup>7</sup>

## Conclusion

This work represents the first record of bat flies (Streblidae and Nycteribiidae) for Amapá State. The number of species of bat flies up to 50 species and two subspecies for the State of Amapá, being these 18 new records for the North Region of the country and seven new records for Brazil. In addition, the infracommunities found in the three conservations units of Amapá are described.

## Supplementary material

The following online material is available for this article:

**Table S1** - All the infracommunities found in the three Conservation Units.

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## Author Contributions

Ana Carolina Moreira Martins: Contribution to data collection.

Gabriela Hrycyna: Contribution to manuscript preparation.

Gustavo Graciolli: Contribution to critical revision, adding intellectual content; Contribution to manuscript preparation.

## Conflicts of interest

The authors declares that they have no conflict of interest related to the publication of this manuscript.



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## Native parasitoids (Hymenoptera: Braconidae) of fruit flies (Diptera: Tephritidae) in Serra da Bodoquena National Park-MS, Brazil

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**Abstract:** Fruit flies are among the main groups of phytophagous insects. Their larvae, when feeding on the pulp or seeds of the host fruits, can cause rot and favor the entrance of pathogens. Thus, the objectives of this study was: (1) To know the trophic associations between species of parasitoids and fruit flies in the Parque Nacional da Serra da Bodoquena (PNSB)-MS, Brazil. (2) To inventory the parasitoid species associated with fruit flies in fruit trees in the Parque Nacional da Serra da Bodoquena. Monthly expeditions were carried out (From Jan 2017 to Feb 2018) in order to collect the fruits and obtain the fruit flies (Tephritidae) and their respective parasitoids. The collected fruits were transported to the laboratory of frugivorous insects (LIF) of the Universidade Federal da Grande Dourados (UFGD)-MS. The fruit fly larvae were daily collected and placed in transparent acrylic cups containing sterilized sand, where they remained till the emergence of adults and their parasitoids. Twenty-Three adult Tephritid parasitoids were recovered, represented by two species of Braconidae: *Doryctobracon areolatus* (18) and *Utetes anastrephae* (5), both infesting *Anastrepha* species: *Anastrepha fraterculus*, *A. sororcula* and *A. striata* in Myrtaceae: *Psidium guajava* and *Eugenia myrcianthes*. *Doryctobracon areolatus* and *Utetes anastrephae* are reported for the first time parasitizing *Anastrepha* species in *Eugenia myrcianthes*.

**Keywords:** Parasitism, Trophic Interactions, Biological Control, Conservation Unit, Natural Enemies.

## Parasitoides Nativos (Hymenoptera: Braconidae) de moscas das frutas (Diptera: Tephritidae) no Parque Nacional da Serra da Bodoquena-MS, Brasil

**Resumo:** As moscas-das-frutas estão entre os principais grupos de insetos fitófagos. Suas larvas, ao se alimentarem da polpa ou das sementes dos frutos hospedeiros, podem causar podridão e favorecer a entrada de patógenos. Sendo assim, os objetivos desse estudo foram: (1) Conhecer as associações tróficas entre as espécies de parasitoides e moscas das frutas no Parque Nacional da Serra da Bodoquena (PNSB). (2) Inventariar as espécies de parasitoides no PNSB. Foram realizadas expedições mensais de janeiro de 2017 a fevereiro de 2018 para as coletas de frutos e obtenção de moscas das frutas (Tephritidae) e seus respectivos parasitoides. Os frutos coletados foram transportados ao Laboratório de Insetos Frugívoros (LIF) da Universidade Federal da Grande Dourados-MS. As larvas das moscas das frutas foram diariamente recolhidas e inseridas em copos de acrílico transparentes contendo areia esterilizada onde permaneceram até a emergência dos adultos de tefritídeos e/ou seus parasitoides. Foram recuperados 23 adultos de parasitoides das larvas dos tefritídeos, representados por duas espécies de Braconidae: *Doryctobracon areolatus* (18) e *Utetes anastrephae* (5), ambos parasitando espécies de *Anastrepha*: *Anastrepha fraterculus*, *A. sororcula* e *A. striata* em Myrtaceae: *Psidium guajava* e *Eugenia myrcianthes*. *Doryctobracon areolatus* e *Utetes anastrephae* são relatados pela primeira vez parasitando espécies de *Anastrepha* em frutos de *Eugenia myrcianthes*.

**Palavras-chave:** Parasitismo, Interações Tróficas, Controle Biológico, Unidade de Conservação, Inimigos Naturais.

## Introduction

Fruit flies are among the main groups of phytophagous insects. Their larvae, when feeding on the pulp or seeds of the host fruits can cause rot and favor the entry of pathogens, causing the early fall of the fruits and making them unviable for *in natura* consumption or commercialization (Uchoa 2012).

Natural fruit fly enemies such as viruses, bacteria, fungi, protozoa, nematodes, predators, parasites and parasitoids are important biotic agents of mortality. They are usually present in natural environments and can be handled in labs or biofactories and employed in integrated management programs for species with pest status on vegetables and fruit trees (Stibicky 2004).

The hymenoptera parasitoids, mainly Braconidae, Figitidae and Pteromalidae, are considered the main regulators of fruit fly populations in the Neotropical region (Uchoa 2012). Researches on these trophic interactions have been intensified since the 1970, with advances in scientific investigations on ecology (Cappuccino 1995) and the natural history of fruit fly natural enemies.

There are two main guilds of parasitoids which attack fruit flies: those that parasitize juveniles (first two instars) and the ones that settle in third instar larvae and/or pre-pupae and emerge from the puparium at the end of this phase. The Tephritid larvae are parasitized still inside the fruits or when they leave their hosts to pupate in the ground. Normally, parasitoids oviposit endophytically in the host, reaching the frugivorous larvae inside the fruit. Upon hatching of the egg, the neonata parasitoid larva will feed and develop internally in the host tephritid larva. Then it will devour the whole pupal musculature, emerging through a hole produced by the imago jaws (Salles 1995; Cirelli & Dias 2003).

In Brazil, studies on fruit fly parasitoids are still scarce, mainly due to the fact that most of the researches are carried out by using traps which capture the adults of fruit fly species, so it is not possible to obtain the larvae of the tephritids and their emergent parasitoids. The main Braconidae species already inventoried in the country are: *Doryctobracon areolatus* (Szépligeti), *Utetes Anastrephae* (Viereck) and *Opius bellus* Gahan (Canal & Zucchi 2000, Costa et al. 2009).

The knowledge on Tephritid natural enemies in natural environments, especially in Integral Conservation Units (UCs), is scarce. As these parasitoids are not specific species, they help to regulate fruit fly populations in a natural way and can parasitize the larvae of several species that colonize a large diversity of fruit species, many of commercial value. In addition, these important biotic agents of mortality can be used for the biological control of Tephritids with pest status.

In Brazil, there are currently 73 national parks earmarked for the integral conservation of biodiversity and biotope: North (29), Northeast (8), Midwest (8), Southeast (13), South (15) (MMA, 2019). These units represent excellent sites for studies of biological diversity, since they have their fauna, flora and physical environment preserved. Thus, the objectives of this research were: To know the trophic associations between the parasitoid species and fruit flies in the Parque Nacional da Serra da Bodoquena (PNSB), Brazil; To inventory the parasitoid species associated with fruit flies in fruit trees in the PNSB.

## Material and Methods

### 1. Study area

According to Koppen, the climate in the Parque Nacional da Serra da Bodoquena (PNSB) (20°35'–21°30'S; 56°30'–57°W) is type AW, characteristic of wet Summer and dry Winter (between May and September), with annual rainfall varying between 1.000 and 1.700mm, reaching the lowest rainfall index in August. The maximum temperature in the dry period is 35°C and the average of the coldest month varies between 15 and 20°C (PCBAP 1997; Reys et al. 2005).

The predominant vegetation in the PNSB is the Deciduous Seasonal Forest, better known as dry forest, since it is associated with limestone rocks. Next to the water courses occurs the Deciduous Alluvial Seasonal Forest, which corresponds to the ciliary forest. There are also transition areas in the park with characteristics of both Cerrado and Seasonal forest (IBAMA 2000).

The PNSB is composed by two areas, comprising four municipalities (Jardim, Bonito, Bodoquena and Porto Murtinho) in Mato Grosso do Sul state. To the South (27.793ha) it is located in Porto Murtinho, Bonito and Jardim, and the fragment of the northern part (48.688ha) in Bodoquena and Bonito (PNSB 2013).

The PNSB region is a priority area for the biodiversity conservation in Mato Grosso do Sul, due to its unique characteristics. The park is formed by carbonate rocks which provide the occurrence of elastic features with numerous caves and grottos.

### Data collection and analysis

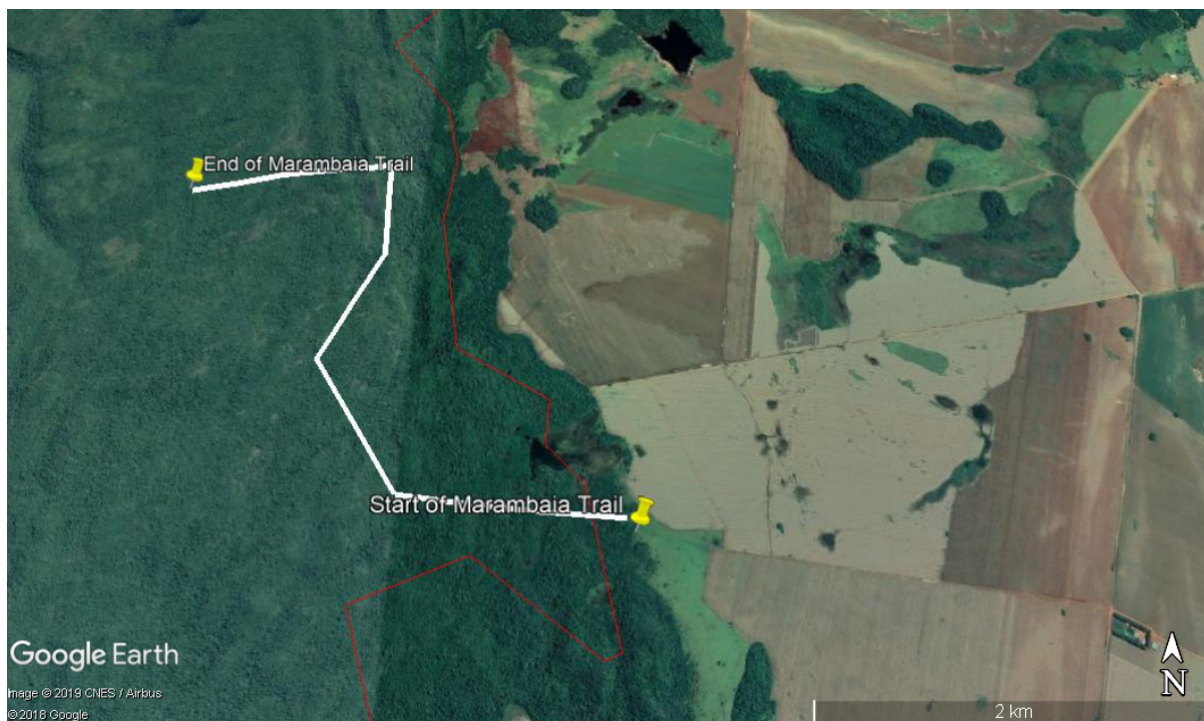
Monthly expeditions were carried out (from Jan 2017 to Feb 2018) in order to collect the fruits and obtain the fruit flies (Tephritidae) and their respective parasitoids in the PNSB. For this purpose, fruits that were found in both fruit trees and fallen to the ground were sampled, depending on their availability along the pre-established transects, each with 5m width (2,5m for each side).

These three trails are used two to four times a year to monitor the flora and fauna in the PNSB. The evaluated trails were: Santa Fé (21°30'22" S and 56°45'64" W), Marambaia (20°58'09" S and 56°42'36" W) and Catamarca (the spring and sink of the Perdido river (21°06'90" S and 56°42'36" W). (Figures. 1, 2 and 3).

All the sampled plants along the trails were exsicated and their species were identified by Dr. Zefa Valdivina Pereira, *Faculdade de Ciências Biológicas e Ambientais (FCBA)*, *Universidade Federal da Grande Dourados (UFGD)* and by Dr. Ângela Sartori, from the *Departamento de Biologia* of *Universidade Federal de Mato Grosso do Sul (UFMS)*, Campo Grande-MS.

The collected fruits were packed in cotton bags and transported to the *Laboratório de Insetos Frugívoros (LIF)* of the UFGD, Dourados-MS, where they were later transferred to wooden pallets allocated in plastic trays containing a fillet of water ( $\pm 1$  cm) on the bottom, according to Uchoa & Zucchi (1999) methodology for last instar larvae collection of fruit flies that leave the fruit to pupate.



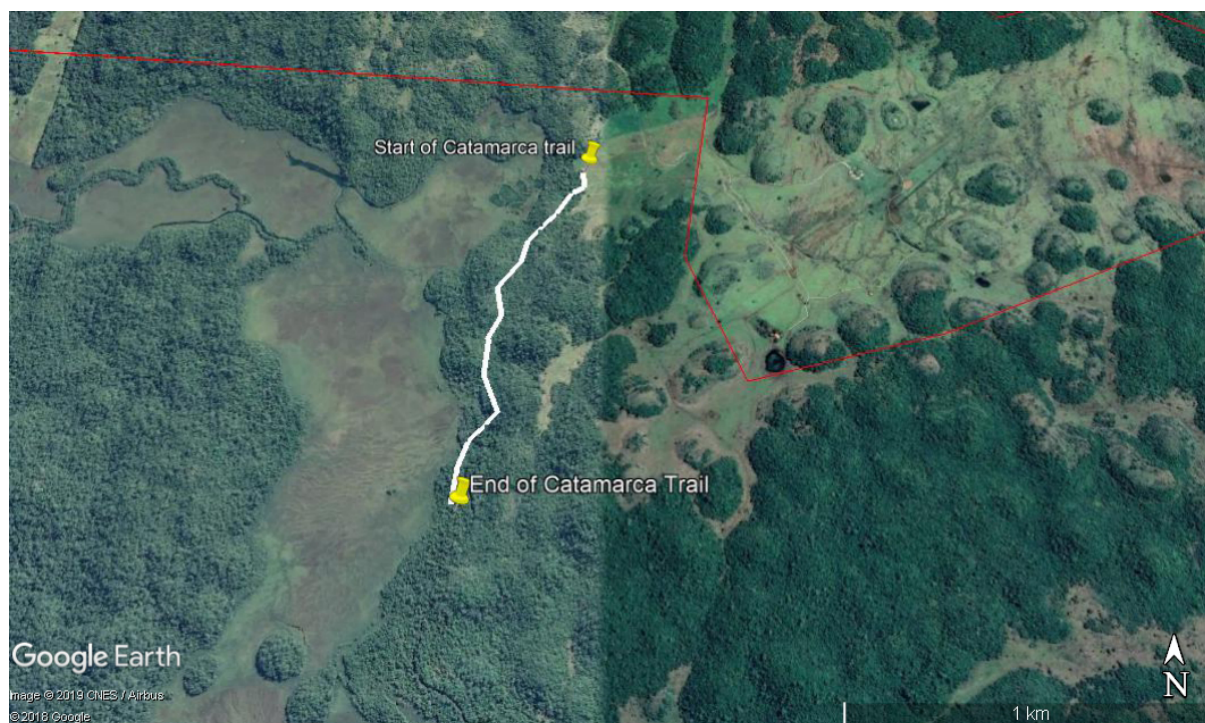


**Figure 1.** Marambaia Trail in the Parque Nacional da Serra da Bodoquena, Mato Grosso do Sul state, Brazil, used to monitor periodically the flora and fauna (Source: Google Earth, 2019).



**Figure 2.** Santa Fé Trail in the Parque Nacional da Serra da Bodoquena, Mato Grosso do Sul state, Brazil, periodically used to monitor the flora and fauna (Source: Google Earth, 2019).





**Figure 3.** Catamarca Trail in the Parque Nacional da Serra da Bodoquena, Mato Grosso do Sul state, Brazil, used to monitor periodically the flora and fauna (Source: Google Earth, 2019).

The larvae were daily collected from the trays and placed into clear acrylic cups containing sterilized sand, where they remained until the adult emergence of the fruit flies and/or their parasitoid species.

The identification of frugivorous Tephritid species was performed by the examination of female genitalia by Dr. Manoel Uchoa, in the laboratory using identification keys (Stone 1942, Steyskal 1977, Zucchi 2000).

The identification of the parasitoids emerged from the tephritid puparium was performed by Dr. Jorge Anderson Guimarães, EMBRAPA HORTALIÇAS (CNPq), Brasília-DF, using identification keys (Canal & Zucchi 2000; Guimarães et al. 2000, 2003, 2011).

The parasitism rates of fruit flies were calculated according to Uchoa et al. (2003), by the equation  $\%CP = N.R.P \times 100 / N.L3$ , where  $\% C.P$  = total percentage of parasitism rate, N.R.P = Number of recovered parasitoids and N.L3 = Number of third instar larvae from recovered the fruit flies.

## Results and Discussion

Twenty-three adult parasitoids were recovered from the tephritid larvae, represented by two Braconidae species: *Doryctobracon areolatus* (18) and *Utetes anastrephae* (5), both infesting *Anastrepha* species: *Anastrepha fraterculus* (Wiedmann), *A. sororcula* Zucchi and *A. striata* Schiner in Myrtaceae: *Psidium guajava* L. and *Eugenia myrcianthes* (Nied). (Table 1).

The higher abundance of parasitoids occurred in *E. myrcianthes*, with 10 individuals of *D. areolatus* and three of *U. anastrephae*. In *P. guajava*, eight adults of *D. areolatus* and two from *U. anastrephae*

were obtained. The highest parasitism index was obtained in *E. myrcianthes*, with 6.5% of parasitism, and in *P. guajava* the index was 6%, totaling 12.5 % of parasitized larvae (Table 1). The two parasitoid species found in this study (*D. areolatus* and *U. anastrephae*) are frequently obtained in researches on trophic interactions among fruit flies, host plants and their parasitoids (Uchoa et al. 2003, Marinho et al. 2009).

*Doryctobracon areolatus* is considered one of the most important native species of fruit fly parasitoids in the Neotropical region. It parasitizes several species of *Anastrepha* in South and Central America countries (Purcell 1998, Cancino & Montoya 2004). This species is considered promising for integrated pest management programs, aiming at the biocontrol of fruit flies (Uchoa et al. 2003, Uchoa 2012). Its largest abundance, reported in this study, is probably due to the wide geographic distribution of the species.

The high abundance of *D. areolatus* can be related to its largest ovipositor size ( $\pm 3.8\text{mm}$ ) in relation to *U. anastrephae* ( $\pm 1.6\text{mm}$ ). Probably this feature enables *D. areolatus* to oviposit in fruit fly larvae in a greater diversity of host fruit species with different sizes and thickness of epicarp and mesocarp (Sivinsky et al. 2009, Marinho et al. 2009).

*Utetes anastrephae*, as well as *D. areolatus*, is distributed throughout the neotropical region. This is the only species of *Utetes* genus recorded parasitizing *Anastrepha* species, with recorded occurrence in 20 Brazilian states and associated with 16 species of *Anastrepha* and *Ceratitis capitata* (Wiedmann) (Zucchi & Moraes 2008). The lowest abundance of *U. anastrephae* in relation to *D. areolatus* in the ecosystems has been frequently been reported in other researches (Torres et al. 2010; Falcão et al. 2012), results that are consistent with this study.

**Table 1.** Trophic interactions between fruit flies (Diptera: Tephritidae, host plants and parasitoids (Hymenoptera: Braconidae) in the Parque Nacional da Serra da Bodoquena-MS, Brazil (January 2017 and December 2018).

Botanical Family	Plant species	Weight (g)	Nº Fruits	Nº Larvae	Nº Adults	Fly species	Parasitoid species	% Parasitoids
Myrtaceae	<i>Psidium guajava</i>	1038	65	191	80	<i>Anastrepha sororcula</i> Zucchi (71)	<i>Doryctobracon areolatus</i> (8)	6
						<i>Anastrepha striata</i> Schiner (3)	<i>Utetes anastrephae</i> (2)	
						<i>Anastrepha fraterculus</i> (Wiedman) (1)		
						<i>Anastrepha</i> spp. (5)		
	<i>Eugenia myrcianthes</i>	779	133	197	145	<i>Anastrepha fraterculus</i> (45)	<i>Doryctobracon areolatus</i> (10)	6,5
						<i>Anastrepha sororcula</i> (9)	<i>Utetes anastrephae</i> (3)	
						<i>Anastrepha obliqua</i> (Macquart) (4)		
						<i>Anastrepha serpentina</i> (Wiedmann) (1)		
						<i>Anastrepha</i> spp. (85)		
						<i>Neosilba</i> sp. (6)		
Total		1,817	198	388	225		23	12,5

According to Hickel (2002), the pulp of the fruit can act as a barrier to parasitism, explaining the low abundance of *U. anastrephae* in relation to *D. aerolatus*. This pattern is also recorded in other inventories, such as Uchoa et al. (2003) and Taira et al. (2013). Since *Utetes anastrephae* ovipositor is shorter than that of *D. aerolatus*, it possibly attacks only larvae in smaller fruits with fine pericarp and a less thick endocarp.

Smaller fruits possibly favor parasitoid species with shorter ovipositor, since they facilitate the encounter of the larvae. On the other hand, parasitoids with long ovipositors can obtain larvae in fruits with pulps of different thicknesses, being able, in a way, to diminish the competition between the two species (*D. aerolatus* and *U. anastrephae*) by the host fruits (Marinho et al. 2009). In this research, the fruits infested by tephritids were medium in size (3 to 5 cm), which in a way could have favored the parasitoids with longer ovipositor, such as *D. aerolatus*.

According to Costa et al. (2009), the fruits that are collected from the ground are generally more parasitized than the ones taken directly from the trees. This probably happens due to the longer exposure time of the fruits in the field and also to the behavior of certain parasitoid species which more frequently parasitize larvae in fallen fruits, penetrating through openings previously made by other frugivorous insects or birds. This is another possible explanation for the reduced parasitism verified in this study, since most of the sampled fruits ( $\pm 70\%$ ) were collected directly from the fruit trees and not from the ground.

The low richness in parasitoid species is attributed to the low diversity of fruit flies collected in the study area. *Doryctobracon areolatus* and *Utetes anastrephae* are reported for the first time in *Eugenia myrcianthes* fruits. Researches on fruit fly parasitoids are still scarce in UCs, thus, this study contributes to broaden the knowledge about fruit fly natural enemies in conserved areas.

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## Author's Contributions

Luciano Brasil Martins de Almeida: Data collection and analysis; Preparation of the manuscript.

João Batista Coelho: Assistance in data collection.

Jorge Anderson Guimarães: Identification of parasitoid species.

Manoel Araécio Uchoa: Preparation of the manuscript and fruit fly identification.

## Conflict of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Diversity of Odonata (Insecta) in Seasonal Deciduous Forest fragments in southern Brazil (state of Rio Grande do Sul), with a new record for the state and comments on the seasonal distribution of the species

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**Abstract:** We present an Odonata (Insecta) check list of species occurring in a fragment of the Seasonal Deciduous Forest (Atlantic Forest biome) from the central region of the state of Rio Grande do Sul (RS), southern Brazil, along with a list of the odonate species recorded in this phytoecological region for the state. In addition, we provide comments on the seasonal distribution of the species occurring in the study area. Two streams and seven farm ponds located in the middle course of the Jacuí River basin were surveyed between December 2007 and February 2009. Overall, we recorded 49 species from 21 genera and six families. *Argia serva* Hagen in Selys, 1865 (Coenagrionidae) had its first occurrence record mentioned for the state, elevating to 183 the total number of Odonata species occurring in Rio Grande do Sul. The number of species recorded in the study area corresponds to ~26% of the known Odonata diversity in RS. Libellulidae was the most species-rich family (22 species, ~45% of the total), followed by Coenagrionidae (18 species, 37% of the total). The checklist for the Seasonal Deciduous Forest in RS indicated the occurrence of 83 species of Odonata in this phytoecological region (~45% of the known odonate species in the state). This elevated diversity could be related to the density of the vegetation structure. In the study area, 20 species were found in streams, and 45 in farm ponds. Species occurrence showed marked seasonal patterns in the study area, with 88% of the species recorded from summer to autumn, and no species detected in streams in the winter. Moreover, 70% of the species were recorded in either one or two seasons in farm ponds, while 65% occurred solely in one season in streams. This result indicates that the life cycle of Odonata in southern Brazil is strongly influenced by seasonal patterns in temperature.

**Keywords:** dragonflies, inventory, Atlantic Forest, streams, farm ponds.

## Diversidade de Odonata (Insecta) em fragmentos de Floresta Estacional Decidual no Sul do Brasil (estado do Rio Grande do Sul), com um novo registro para o estado e comentários sobre a distribuição sazonal das espécies

**Resumo:** Uma checklist das espécies de Odonata (Insecta) de um fragmento de Floresta Estacional Decidual (Bioma Mata Atlântica) localizado na região central do estado do Rio Grande do Sul (RS) é apresentada neste estudo, bem como uma checklist das espécies para esta região fitoecológica para o estado. Além disso, também é discutida a distribuição temporal da ocorrência das espécies na área de estudo. Dois riachos e sete reservatórios localizados no trecho médio da bacia do rio Jacuí foram estudados entre dezembro de 2007 e fevereiro de 2009. Ao todo, 49 espécies de 21 gêneros e seis famílias foram registradas. *Argia serva* Hagen in Selys, 1865 (Coenagrionidae) teve seu primeiro registro mencionado para o estado, elevando para 183 o número total de espécies de Odonata



ocorrentes no Rio Grande do Sul. O número de espécies registrado na área de estudo corresponde a cerca de 26% da diversidade de Odonata conhecida no RS. Libellulidae foi a família mais rica (22 espécies, ~45% do total), seguida por Coenagrionidae (18 espécies, 37% do total). A lista compilada de espécies para a Floresta Estacional Decidual no RS indicou a ocorrência de 83 espécies de Odonata nesta região fitoecológica (~45% da diversidade de Odonata conhecida do estado). Essa alta diversidade pode estar relacionada à densidade da estrutura da vegetação. Na área de estudo, 20 espécies foram encontradas em riachos e 45 em reservatórios. A distribuição temporal das espécies foi marcadamente sazonal na área de estudo, com 88% das espécies registradas do verão ao outono, e nenhuma espécie detectada em riachos no inverno. Além disso, 70% das espécies ocorreram em uma ou duas estações em reservatórios, enquanto 65% das espécies registradas em riachos foram detectadas somente em uma estação. Este resultado indica que o ciclo de vida das espécies de Odonata no extremo sul do Brasil é fortemente influenciado pelos padrões sazonais de temperatura.

**Palavras-chave:** libélulas, inventário, Mata Atlântica, riachos, reservatórios.

## Introduction

Odonata (dragonflies and damselflies) is a small order of aquatic insects with high importance to the ecological dynamics of freshwater ecosystems (Corbet 2004). Over 6300 species of the order are described worldwide (Schorr & Paulson 2019). The Neotropical Region holds an important portion of the global diversity of Odonata, with over 1700 resident species (von Ellenrieder 2009). However, in Brazil, the largest country of the region (covering ~50% of the area of South America), only 860 species from 15 families and 146 genera have been recorded to date (Pinto 2019). An extensive review on the diversity of Odonata in Brazil conducted in the early 2000s (De Marco & Vianna 2005) showed that studies had a remarkably uneven distribution in the country, with only ~1/3 of the territory adequately surveyed. Many studies have been carried out since then, and although the knowledge on the diversity of Odonata increased in few regions (Nóbrega & De Marco 2011, Juen & de Marco 2012, Koroiva et al. 2017, Dalzochio et al. 2018), no significant changes occurred in the original scenario. Thus, the knowledge of the spatial distribution of Odonata in Brazil remains poorly understood.

In the state of Rio Grande do Sul (RS; area of ~282000 km<sup>2</sup>), southernmost Brazil, many inventories and studies on the distribution of Odonata have been recently conducted (Renner et al. 2015, Renner et al. 2016a, b, Renner et al. 2017, Dalzochio et al. 2018). A recent compilation of the studies conducted in the state mentioned the occurrence of 182 species from nine families and 57 genera (Dalzochio et al. 2018), representing ~21% of the known diversity of Odonata in Brazil (Pinto 2019). Nevertheless, as in the rest of the Brazilian territory, many areas of the state remain poorly investigated (Dalzochio et al. 2018). The southernmost boundaries of the Atlantic Forest biome originally covered ~37% of the territory of RS (IBGE 2004). Although the Atlantic Forest is considered a hotspot of biodiversity in the Neotropical Region (Mittermeier et al. 2011), nowadays the remnants of this biome in RS are reduced to less than 10% of the original area (Rio Grande do Sul 2019). Previous studies described that Atlantic Forest remnants in Southern Brazil hold a great diversity of various insect groups (Ferro & Teston 2009, Iserhard et al. 2017). However, inventories of aquatic insects in the Atlantic Forest in RS are scarce, and, particularly for Odonata, large areas of the biome remain underrepresented regarding the knowledge of Odonata composition (Dalzochio et al. 2018).

The Atlantic Forest comprises several vegetation types (phytoecological regions) in the state of Rio Grande do Sul, ranging from Seasonal and Ombrophilous forests to highland grasslands matrices with patches of *Araucaria* forests (Oliveira-Filho & Fontes 2000, Cordeiro & Hasenack 2009, IBGE 2012) (Figure S1). Most inventories of Odonata conducted in the Atlantic Forest from RS have so far focused regions of the Highland Grasslands (*Campos de Cima da Serra*) (Kittel & Engels 2014, 2016, Renner et al. 2016a, b), while in the Seasonal Deciduous Forest phytoecological region, most of the current knowledge stem from inventories limited to a single basin (Consatti et al. 2014, Renner et al. 2015, Hanauer & Renner 2008, Renner et al. 2013) and sparse species occurrence records (see references in Dalzochio et al. 2018). Thus, the species composition of Odonata in this phytoecological region remains as one of the least studied in RS (Dalzochio et al. 2018). Furthermore, landscape structure features such as vegetation types are an important regional driver of the spatial distribution of Odonata (Paulson 2006, Kadoya et al. 2008). Phytoecological regions were found to play an important role in structuring the composition of many insect groups in RS (Ferro & Teston 2009, Siewert et al. 2014), and a recent study carried out in RS showed that the composition of Odonata largely changed among different vegetation types in the Pampa biome (Renner et al. 2019). However, no assessment of the differences in the composition of Odonata among the vegetation types of the Atlantic Forest biome in RS has been conducted yet.

Data on the biology of odonate species are also scarce in the subtropical regions of Brazil, greatly hindering the understanding of the patterns of diversity of the order. For instance, studies focusing on the life cycle and the temporal distribution of species of Odonata are rather incipient in southern Brazil. Previous studies have showed that several factors influence the life history of odonates, and climate is a strong driver of their life cycles (Corbet 2004). This is because seasonal environmental factors such as patterns in photoperiod, rainfall and temperature influence larval development and the activity patterns of adults, thus affecting the number of generations produced by odonate species across different climatic regions (Corbet 2004, Corbet et al. 2006). Data from tropical regions in Brazil showed that the life cycle of many odonates is linked to the wet-dry season (Vilela et al. 2016). However, while most of the Brazilian territory has tropical climate, the predominant climates in RS range from subtropical to subtemperate

(Maluf 2000), i.e., they are characterized by the absence of a dry season, but show remarkable cold winters and regularly distributed rainfall. It should therefore be expected that odonate species in RS have marked differences in their life cycles in relation to species inhabiting tropical regions from Brazil.

Habitat types (e.g., lentic and lotic) hold distinct compositions of odonate species (Renner et al. 2018). In addition, habitat type can influence the life cycle of odonate species. The review by Corbet et al. (2006) showed that important life-history traits of Odonata such as the length of the larval period and number of annual generations (voltinism) greatly changed between species from lentic and lotic ecosystems, likely due to the different ecological dynamics among habitat types, which in turn can influence certain traits related to larval growth in odonates (Córdoba-Aguilar 2008). However, studies assessing the life cycles and the seasonal distribution of Odonata in distinct habitat types are missing in southern Brazil.

In this context, we provided in this study a checklist of the species of Odonata occurring in a fragment of the Seasonal Deciduous Forest of the Atlantic Forest biome from southernmost Brazil (state of Rio Grande do Sul). We also compiled a checklist of Odonata species occurring in the Seasonal Deciduous Forest from Rio Grande do Sul, with comments on the diversity of Odonata in phytoecological regions in the state. Finally, we provided further information on the life cycle of the species inhabiting streams and farm ponds. In particular, we investigated seasonal patterns of species occurrence in the study area, in order to improve the knowledge of odonate biology in southern Brazil.

## Material and Methods

### 1. Study area

This study was conducted in the municipalities of Agudo and Ibarama (53°10'W; 29°10'S), central region of the state of Rio Grande do Sul (RS). The study area corresponds to the drainage area of the middle course of the Jacuí River basin (Figure 1), one of the most important watersheds of RS (~800 km long and area of ~71,000 km<sup>2</sup>) (Zamanillo et al. 1989). The Jacuí River basin runs along the transition between the Southern Brazilian Plateau (*Planalto Meridional Brasileiro*) and the Central Depression (*Depressão Central*) of RS, with altitudes ranging from 50 to 500 m (Pereira et al. 1989). The original vegetation in the study area is the Seasonal Deciduous Forest, in the southernmost boundaries of the Atlantic Forest biome (Durlo et al. 1982, Marcuzzo et al. 1998). Currently, few forest remnants and secondary-growth fragments distributed among small rural properties characterize the land use in the region (Rio Grande do Sul 2019), mostly because declivity hinders the practice of large agricultural activities (Marcuzzo et al. 1998). Nonetheless, the study area holds a large portion of the remnants of Seasonal Deciduous Forest in RS (Cordeiro & Hasenack 2009). The climate in the region is Cfa of Köppen, with average annual temperature ranging from 18 to 22 °C, while in winter periods the temperature is generally lower than 10 °C (Maluf 2000). Rainfall is regularly distributed throughout the year, with the annual precipitation ranging from 1500 to 1750 mm (Pereira et al. 1989).

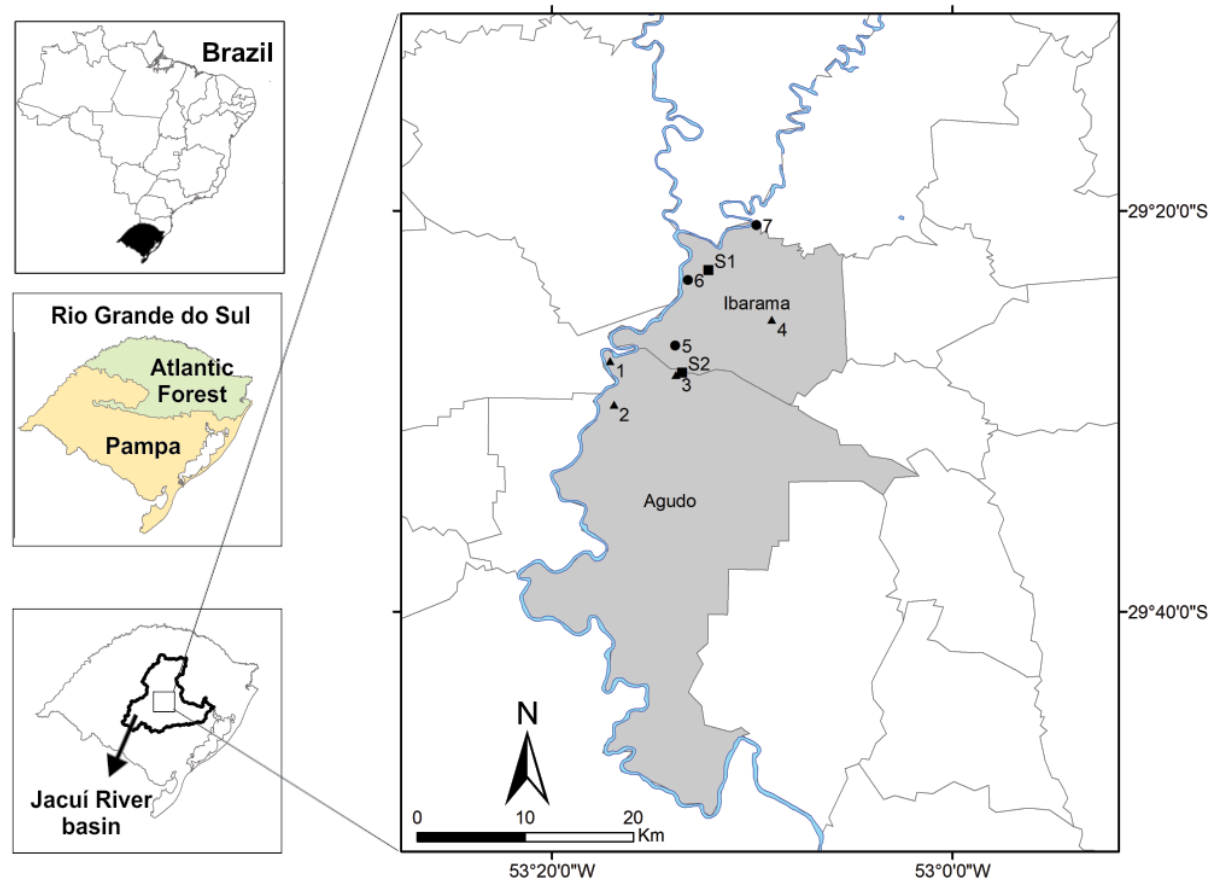
### 2. Data collection

Qualitative surveys of adult odonates were carried out monthly (from March 2008 to February 2009) at six sampling sites in the study area (four farm ponds and two streams contributors of the Jacuí River; Figure 1). The specimens were collected with a hand net in sunny days between 9:00 AM and 16:00 PM, periods of the day matching with the peak of activity of adults. Collection teams ranged from two to three persons, totaling a sampling effort of 1 hour by person. Collections were carried out along the margins of the farm ponds and stretches of ~50 m in the streams. All sites were previously assessed in studies focusing on the diversity and temporal distribution of larval assemblages of Odonata (Pires et al. 2013, 2014). The detailed description of the environment and the monthly-sampled data for each stream and farm pond studied are also available in the previously cited references. For the general inventory, records from surveys conducted between December 2007 and April 2008 in three additional farm ponds were also included in the study, adding up to nine sampling sites (Table 1; Figure 1). The collected specimens were fixed in situ with ethanol 70%. In the laboratory, specimens were determined to species level according to specialized literature (Garrison et al. 2006, Lencioni 2005, 2006). All the collected specimens were preserved in 85% ethanol and archived in the collection of the Science Museum (MCN) of the Vale do Taquari University (UNIVATES, Lajeado, Rio Grande do Sul, Brazil; collection ID from 2160 to 2371).

### 3. Data analyses and data compilation

We assessed the species richness in the study area according to the following methods: we first computed species accumulation curves for each habitat type (farm ponds and streams). The mean curve was generated after 500 random permutations of the samples (Gotelli & Colwell 2001). We also estimated the extrapolated richness in each habitat type using two incidence-based non-parametric richness estimators: first- and second-order Jackknife. Species accumulation curves and richness estimators were respectively calculated with the functions *specaccum* and *specpool* from the vegan package (Oksanen et al. 2018) in the R statistical environment (R Core Team 2018). The analyses of species richness were restricted to the subsets of the farm ponds and streams that were monthly sampled (from March 2008 to February 2009; streams: N = 24; farm ponds: N = 48).

In order to investigate and estimate the life cycles of species of Odonata in the study area, we took into consideration the observed number of collections of adult specimens along monthly date ranges broadly corresponding to the austral seasons: fall (March 2008 – May 2008); winter (June 2008 – August 2008); spring (September 2008 – November 2008), summer (December 2008 – February 2009). We thus used a combination of the number of observations and the consecutive occurrence of each odonate species as proxies of the number of generations produced within the sampling period and as primary sources for the estimations of the life cycles of each species. In this sense, species detected solely in one season were considered univoltine (i.e., species that likely had only one generation), while species collected over two or more consecutive seasons were considered bi- or multivoltine (i.e., species with two or more generations within a year).



**Figure 1.** Map of the study area indicating the location of the state of Rio Grande do Sul and the Jacuí River basin drainage area. At the right, the blue line represents the main course of the Jacuí River and the municipalities covered in the study (grey). Triangles (1 – 4): monthly-sampled farm ponds. Squares (S1 and S2): monthly-sampled streams (from March 2008 to February 2009). Circles (5 – 7): additional farm ponds sampled between December 2007 and January 2008.

**Table 1.** Geographic coordinates of the sampling sites. S1 and S2, see Figure 1.

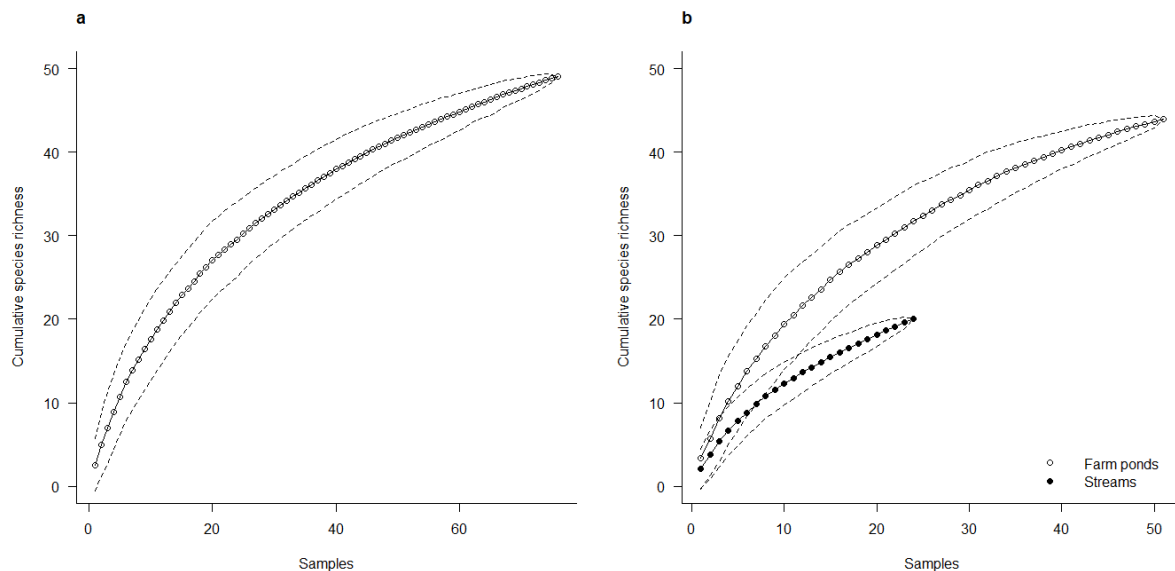
Sites sampled from March 2008 to February 2009	
Site	Coordinates
Lajeado da Gringa Stream (S1)	29°22'58 S 53°12'11 W
Lajeado do Gringo Stream (S2)	29°28'05 S 53°13'30 W
Farm Pond 1	29°27'30 S 53°17'30 W
Farm Pond 2	29°29'41 S 53°16'54 W
Farm Pond 3	29°28'6 S 53°13'23 W
Farm Pond 4	29°25'27 S 53° 9'1 W
Sites sampled between December 2007 and April 2008	
Farm Pond 5	29°26'44 S 53°16'50 W
Farm Pond 6	29°23'28 S 53°13'54 W
Farm Pond 7	29°20'12 S 53°14'02 W

Finally, we compiled the occurrence records of species of Odonata within the Seasonal Deciduous Forest phytocological region from the Atlantic Forest biome in RS. For this purpose, we interpolated the occurrence records of Odonata in RS from Dalzochio et al. (2018) with the original area of the Seasonal Deciduous Forest in the state (IBGE 2012) (Figure S1). We further compared the number of species recorded

in the Seasonal Deciduous Forest with the other phytocological regions from the Atlantic Forest in RS (including the number of exclusive and shared species among regions). In view of the restricted number of occurrences in the other phytocological regions, in this step we restricted the comparison to the Highland Grasslands.

## Results

We collected 626 specimens from six families, 21 genera and 49 species (including 32 specimens assigned to genus level only) in the study area. However, the species accumulation curve for all samples combined ( $N = 76$ ) did not reach the asymptote (Figure 2a), suggesting that it would increase with additional effort. The extrapolated richness calculated by each richness estimator ranged from 65.77 (first-order Jackknife; standard error = 4.94) to 73.68 (second-order Jackknife). Libellulidae was the most species-rich family (22 species from ten genera; 44.8% of the total), followed by Coenagrionidae (18 species from six genera; 36.5% of the total). The most species-rich genera were *Acanthagrion* Selys, 1876 (Coenagrionidae), *Lestes* Leach, 1815 (Lestidae) and *Micrathyria* Kirby, 1889 (Libellulidae) (five species each), followed by *Oxyagrion* Selys, 1876 (Coenagrionidae) and *Erythrodiplox* Brauer, 1868 (Libellulidae) (four species each) (Table 2).



**Figure 2.** Cumulative richness of species of Odonata in the middle course of the Jacuí River basin. (a) curve generated for the entire data set (N = 76 samplings). (b) curves based on the subsets of the species recorded in streams and farm ponds.

The richness and composition recorded in the study area differed between streams and farm ponds (Table 2). Twenty species occurred in streams, while 45 species occurred in farm ponds. In addition, species accumulation curves did not stabilize according to habitat type (Figure 2b). In streams, the extrapolated richness calculated by richness estimators ranged from 30.54 (standard error = 3.98; first-order Jackknife) to 37.99 (second-order Jackknife), while in farm ponds, it ranged from 58.7 (standard error = 4.92; first-order Jackknife) to 64.64 (second-order Jackknife). In other words, the richness of Odonata should be higher not only in the study area as a whole, but also in each habitat type.

In relation to the seasonal occurrence of the species in the study area, 88% of the species (N = 40) were observed between the spring and autumn. In the winter, no species were detected in streams, whereas only five species (*Acanthagrion cuyabae* Calvert, 1909, *Acanthagrion gracile* (Rambur, 1842), *Homeoura chelifera* (Selys, 1876), *Ischnura* sp. and *Lestes pictus* Hagen in Selys, 1862) were detected in farm ponds (Table 3). In farm ponds, the season with the highest number of observed species was summer (N = 28; 70%), followed by autumn (N = 24; 60%). In streams, autumn was the season with the highest number of species (N = 13; 65%), followed by spring (N = 11; 55%). The most frequent species in this environment were *Argia albistigma* Hagen in Selys, 1865, *Brechmorhoga nubecula* (Rambur, 1842) and *Hetaerina rosea* Selys, 1853. They occurred in all seasons except winter (Table 3).

The compilation of species records of Odonata showed that 83 species from 39 genera and seven families occur in the Seasonal Deciduous Forest remnants of RS (including the species recorded in this study; Table S1). Furthermore, a comparison of the species composition registered in the Seasonal Deciduous Forest with the studies conducted in the Highland Grasslands from RS (Kittel & Engels 2014, 2016; Renner et al. 2015, 2016a, b and references within Dalzochio et al. 2018) showed that 48 species were exclusive of the Seasonal Deciduous Forest, while 35 species were shared with the Highland Grasslands (Table S1).

## Discussion

### 1. Diversity of Odonata in the study area

The number of species recorded in the study area corresponds to ~26% of the species of Odonata mentioned up to the moment to RS (Dalzochio et al. 2018). This result indicates that the study area can harbor approximately one-third of the known diversity in RS. The richness observed in the study area was lower than that registered in inventories conducted in tropical Brazilian regions (Anjos-Santos & Costa 2006, Souza et al. 2013, Koroiva et al. 2017; 77, 57 and 111 species, respectively). This pattern was also valid after taking into account the habitat types assessed. The richness here observed for streams was lower than that observed for other streams in Brazil (Assis et al. 2005, Ferreira-Peruquetti & Fonseca-Gessner 2010, Juen et al. 2014; 29, 27 and 79 species, respectively). In relation to lentic habitats, (De Marco et al. 2014) detected 56 species in ponds from central Brazil (Cerrado biome). However, taking into account that our results stem from samplings conducted in just nine sites, the richness recorded here could be higher. In this sense, the inventory of odonate larvae previously conducted by Pires et al. (2013) revealed the occurrence of 30 genera in the study area, suggesting the existence of a higher diversity.

The diversity pattern of odonate families here observed was analogous to previous studies. The higher diversity of Libellulidae and Coenagrionidae (which added up to almost 80% of the species in the study area) was also detected in southeastern and central Brazil (Souza et al. 2013; Vilela et al. 2016; Koroiva et al. 2017; Barbosa et al. 2019; Borges et al. 2019) as well as in other studies conducted in Rio Grande do Sul (Pires et al. 2013; Renner et al. 2016a, b). These results corroborate the findings that Libellulidae and Coenagrionidae are the most speciose families of Odonata in the Neotropical Region (Kalkman et al. 2008).



**Table 2.** Composition of species of Odonata recorded in the study area according to habitat type.

	Family	Species	Habitat		Total	
			Farm ponds	Streams		
Anisoptera	Aeshnidae	<i>Rhionaeschna planaltica</i> (Calvert, 1952)	X		2	
	Gomphidae	<i>Phyllocycla propinqua</i> Belle, 1972		X	2	
		<i>Progomphus</i> sp.		X	4	
	Libellulidae	<i>Brechmorhoga nubecula</i> (Rambur, 1842)	X	X	34	
		<i>Dasythemis mincki</i> (Karsch, 1890)*	X		1	
		<i>Erythrodiplax atroterminata</i> Ris, 1911	X	X	11	
		<i>Erythrodiplax media</i> Borrer, 1942	X	X	113	
		<i>Erythrodiplax nigricans</i> (Rambur, 1842)	X		19	
		<i>Erythrodiplax paraguayensis</i> (Förster, 1904)	X		17	
		<i>Gynothemis venipunctata</i> Calvert, 1905	X		4	
		<i>Micrathyria hesperis</i> Ris, 1911	X		1	
		<i>Micrathyria hypodydima</i> Calvert, 1906	X		2	
		<i>Micrathyria ocellata</i> Martin, 1897	X		3	
		<i>Micrathyria pseudeximia</i> Westfall, 1992*	X		1	
		<i>Micrathyria stawianskii</i> Santos, 1953	X		3	
		<i>Oligoclada laetitia</i> Ris, 1911	X		2	
		<i>Orthemis attenuata</i> (Erichson in Schomburgk, 1848)	X		1	
		<i>Orthemis nodiplaga</i> Karsch, 1891	X		3	
		<i>Orthemis</i> sp. (1)*	X		2	
		<i>Orthemis</i> sp. (2)*	X		2	
		<i>Pantala flavescens</i> (Fabricius, 1798)	X		14	
		<i>Perithemis icteroptera</i> (Selys in Sagra, 1857)	X		6	
		<i>Perithemis mooma</i> Kirby, 1889	X	X	22	
	<i>Tramea binotata</i> (Rambur, 1842)	X		1		
	<i>Tramea cophysa</i> Hagen, 1867	X		3		
Zygoptera	Calopterygidae	<i>Hetaerina rosea</i> Selys, 1853	X	X	23	
	Coenagrionidae	<i>Acanthagrion ascendens</i> Calvert, 1909	X		1	
		<i>Acanthagrion cuyabae</i> Calvert, 1909	X		3	
		<i>Acanthagrion gracile</i> (Rambur, 1842)	X	X	81	
		<i>Acanthagrion lancea</i> Selys, 1876	X	X	18	
		<i>Acanthagrion</i> sp.	X		9	
		<i>Argia albistigma</i> Hagen in Selys, 1865	X	X	52	
		<i>Argia indocilis</i> Navás, 1934*	X	X	5	
		<i>Argia serva</i> Hagen in Selys, 1865***		X	4	
		<i>Argia</i> sp.		X	13	
		<i>Homeoura chelifera</i> (Selys, 1876)	X	X	12	
		<i>Ischnura caprelous</i> (Hagen, 1861)	X		2	
		<i>Ischnura fluviatilis</i> Selys, 1876	X	X	37	
		<i>Ischnura</i> sp.	X		2	
		<i>Oxyagrion hempeli</i> Calvert, 1909	X	X	3	
		<i>Oxyagrion pavidum</i> Hagen in Selys, 1876	X		5	
		<i>Oxyagrion simile</i> Costa, 1978	X	X	2	
		<i>Oxyagrion terminale</i> Selys, 1876	X	X	37	
		<i>Telebasis theodori</i> (Navás, 1934)	X		1	
		Lestidae	<i>Lestes bipupillatus</i> Calvert, 1909	X	X	20
			<i>Lestes dichrostigma</i> Calvert, 1909	X		1
	<i>Lestes forficula</i> Rambur, 1842		X	X	18	
	<i>Lestes minutus</i> Selys, 1862		X		1	
	<i>Lestes pictus</i> Hagen in Selys, 1862		X		3	
	Total			45	20	626

\* = species recorded in the additional farm ponds between December 2007 and April 2008. \*\*\* = first record for the state of Rio Grande do Sul.

**Table 3.** Occurrence of odonate species in the study area according to seasons and habitat type. Data refer to the monthly samplings conducted between March 2008 and February 2009. A = autumn (March 2008 - May 2008); W = winter (June 2008 - August 2008); Sp = spring (September 2008 – November 2008); Sm = summer (December 2008 – February 2009).

Season			A	W	Sp	Su	A	W	Sp	Su
Habitat type			Farm ponds				Streams			
Suborder	Family	Species								
Anisoptera	Aeshnidae	<i>Rhionaeschna planaltica</i> (Calvert, 1952)				X				
	Gomphidae	<i>Phyllocycla propinqua</i> Belle, 1972							X	
		<i>Progomphus</i> sp.							X	X
	Libellulidae	<i>Brechmorhoga nubecula</i> (Rambur, 1842)	X				X		X	X
		<i>Erythrodiplax atroterminata</i> Ris, 1911	X		X	X	X		X	
		<i>Erythrodiplax media</i> Borrer, 1942	X		X	X	X			
		<i>Erythrodiplax nigricans</i> (Rambur, 1842)	X			X				
		<i>Erythrodiplax paraguayensis</i> (Förster, 1904)	X							
		<i>Gynothemis venipunctata</i> Calvert, 1909	X			X				
		<i>Micrathyria hesperis</i> Ris, 1911	X							
		<i>Micrathyria hypodydima</i> Calvert, 1906				X				
		<i>Micrathyria ocellata</i> Martin, 1897	X							
		<i>Micrathyria stawiariskii</i> Santos, 1953	X		X	X				
		<i>Oligoclada laetitia</i> Ris, 1911	X			X				
		<i>Orthemis attenuata</i> (Erichson in Schomburgk, 1848)	X							
		<i>Orthemis nodiplaga</i> Karsch, 1891	X			X				
		<i>Pantala flavescens</i> (Fabricius, 1798)			X	X				
		<i>Perithemis icteroptera</i> (Selys in Sagra, 1857)			X	X				
		<i>Perithemis mooma</i> Kirby, 1889	X		X	X			X	
		<i>Tamea binotata</i> (Rambur, 1842)				X				
		<i>Tamea cophysa</i> Hagen, 1867			X	X				
Zygoptera	Calopterygidae	<i>Hetaerina rosea</i> Selys, 1853	X		X	X	X		X	X
	Coenagrionidae	<i>Acanthagrion ascendens</i> Calvert, 1909				X				
		<i>Acanthagrion cuyabae</i> Calvert, 1909		X						
		<i>Acanthagrion gracile</i> (Rambur, 1842)	X	X	X	X			X	X
		<i>Acanthagrion lancea</i> Selys, 1876	X		X	X	X			
		<i>Acanthagrion</i> sp.			X					
		<i>Argia albistigma</i> Hagen in Selys, 1865				X	X		X	X
		<i>Argia indocilis</i> Navás, 1934					X			
		<i>Argia serva</i> Hagen in Selys, 1865					X			
		<i>Argia</i> sp.					X		X	
		<i>Ischnura caprelous</i> (Hagen, 1861)			X	X				
		<i>Homeoura chelifera</i> (Selys, 1876)	X	X	X	X				X
		<i>Ischnura fluviatilis</i> Selys, 1876	X		X	X	X			
		<i>Ischnura</i> sp.		X						
		<i>Oxyagrion hempeli</i> Calvert, 1909				X			X	
		<i>Oxyagrion pavidum</i> Hagen in Selys, 1876	X		X	X				
		<i>Oxyagrion simile</i> Costa, 1978				X	X			
		<i>Oxyagrion terminale</i> Selys, 1876				X			X	
		<i>Telebasis theodori</i> (Navás, 1934)	X							
	Lestidae	<i>Lestes bipupillatus</i> Calvert, 1909	X		X	X	X			
		<i>Lestes dichrostigma</i> Calvert, 1909	X							
		<i>Lestes forficula</i> Rambur, 1842	X		X	X	X			
		<i>Lestes minutus</i> Selys, 1862	X							
		<i>Lestes pictus</i> Hagen in Selys, 1862		X						

The dominant genera in the study area also followed previously observed patterns. *Acanthagrion* and *Erythrodiplax* are among the most speciose genera in the Neotropical Region (Kalkman et al. 2008), and they have been generally reported as the most species-rich genera in inventories through the Brazilian territory (Souza et al. 2013; Vilela et al. 2016; Koroiva et al. 2017). *Lestes* is a common genus in lentic habitats, and it was reported as one of the dominant taxa in ponds in several studies conducted in subtropical South America (Maltchik et al. 2010, Pires et al. 2013).

## 2. New records

### 2.1. *Argia serva* Hagen in Selys, 1865 (Coenagrionidae)

*Argia serva* (Coenagrionidae) is recorded for the first time in RS. Within the South American genus *Argia* Rambur, 1842, *A. serva* is the species with the southernmost distribution range, restricted to the Southern Cone (Palacio et al. 2018). Previous records of the species include Argentina (Lencioni 2006; Palacio et al. 2018), Uruguay (von Ellenrieder et al. 2009) and the state of Santa Catarina in southern Brazil (Lencioni 2006). In our study, the four other specimens collected were found in a stream (Lajeado da Gringa), in autumn (April). Finally, based on the latest compilation of odonate species for RS (Dalzochio et al. 2018), the record of *A. serva* in the study area elevates the number of species of Odonata in the state to 183.

### 3. Checklist of Odonata in the Seasonal Deciduous Forest and preliminary comments on the role of phytoecological regions in the distribution of Odonata in RS

The compilation of the species recorded in this study and the data from the literature indicate that ~45% of the known odonate species in RS occur in the Seasonal Deciduous Forest in RS. In relation to the role of phytoecological regions in the distribution of Odonata in southern Brazil, and in spite of the contingencies associated with the early stages of this assessment, our study provides initial evidence that odonate composition does differ between phytoecological regions from the Atlantic Forest in RS (Table S1). In particular, the Seasonal Deciduous Forest and the Highland Grasslands share a low number of species and show elevated species turnover (Table S1). Odonate composition can be highly sensitive to landscape structure features (Kadoya et al. 2008, Brasil et al. 2018). The review by Paulson (2006) described that factors associated with forest structure (e.g., tree density) can explain the occurrence of many species. In fact, vegetation types in other biomes from southern Brazil have been found to support distinct compositions of Odonata. For instance, a recent study detected significant differences in the composition of Odonata among vegetation types in southern Brazil (ecoregions within the Pampa biome; Renner et al. 2019). In RS, the Seasonal Deciduous Forest encompasses dense forested habitats, while the Highland Grasslands are open-like landscapes, characterized by patches of *Araucaria* forests distributed in a grassland matrix. In addition, the Highland Grasslands are located in areas of higher altitude (up to 1000 m) in RS (Oliveira-Filho & Fontes 2000, Cordeiro & Hasenack 2009, IBGE 2012), where temperature can reach low values. Previous studies showed that elevation gradients as well as forest and open landscapes can largely influence the regional richness and composition of Odonata (Novelo-Gutiérrez & Gómez-Anaya 2009, Renner et al. 2019). In this sense, such environmental differences in

vegetation type and elevation could have accounted for the differences in odonate composition here observed between the Seasonal Deciduous Forest and the Highland Grasslands.

### 4. Odonate assemblages in farm ponds and streams

Higher richness of Odonata was observed in lentic rather than lotic habitats, as shown in studies conducted in tropical (Cerrado biome: Vilela et al. 2016) and subtropical regions from Brazil, including studies with larval (Pires et al. 2013, 2014) and adult stages (Renner et al. 2016a, b). Odonate richness in lentic habitats is affected by several factors, such as the aquatic vegetation structure, which can provide perching and oviposition sites for adults (Kadoya et al. 2004, Rensburg & Turner 2009). The aquatic vegetation structure also provides shelter and refuges from predators for the larval stages (Rensburg & Turner 2009), allowing the development and completion of the life cycle and thus sustaining local populations. In this study, the farm ponds were largely covered by marginal and aquatic vegetation with different life forms along their perimeters (see original data in Pires et al. 2013; 2014), which likely provided suitable sites for the establishment of several species, thus increasing the richness in this habitat.

However, this pattern is not unequivocal in the literature. For instance, higher richness of Odonata was recorded in lotic rather than lentic habitats located in areas with intensive land use conversion, while the opposite pattern was observed in protected areas (Ferreira-Peruquetti & Fonseca-Gessner 2010). Renner et al. (2018) also detected higher average local richness of Odonata in streams located in more pristine areas in the Pampa biome in RS than in man-made lakes surrounded by converted landscapes. This suggests that odonate richness in each habitat (lentic or lotic) could be dependent on the surrounding land use. In this study, the similar surrounding land use in the sampling sites as well as the relative preserved condition of the study area as a whole likely did not affect odonate local richness.

The composition and number of exclusive species also varied between farm ponds and streams, and the general pattern of distribution of families and species according to habitat types was similar to those previously mentioned in the literature. Four species were exclusively found in streams (*Argia serva* and *Argia* sp. (Coenagrionidae); *Phyllocycla propinqua* Belle, 1972 and *Progomphus* sp. (Gomphidae)). Coenagrionidae represented the richest family (11 species), followed by Libellulidae (five species). The predominance of these families in streams is analogous to data from the Cerrado biome (Vilela et al. 2016) as well as from tropical and subtropical areas of the Atlantic Forest biome (Assis et al. 2005; Renner et al. 2016a, b). In relation to species distribution, Gomphidae species were exclusively found in streams, as observed in previous studies conducted in tropical regions from Brazil (Carvalho & Nessimian 2002, Assis et al. 2005). Additionally, larvae of most genera from this family were detected solely in streams in the study area (Pires et al. 2013). *Argia*, a typical dweller of streams (Lencioni 2006), was the richest genus in this habitat. In this study, although adults of some species of *Argia* were observed flying around farm ponds, their larvae were not found in lentic habitats (Pires et al. 2013). Similarly, Calopterygidae species were detected as adults in both habitats, although their larvae were exclusively found in lotic habitats, in accordance with the findings of previous studies (Carvalho & Nessimian 2002; Pires et al. 2013).

In farm ponds, all species of Libellulidae, which was the most diverse family in the study area (44.8% of the total), occurred exclusively in this habitat. Coenagrionidae species corresponded to 35.5% of the total number of species detected in farm ponds (16). In addition, all species of *Acanthagrion*, *Lestes* (five species each) and *Oxyagrion* (four species) occurred in farm ponds only (Table 2). Despite the predominance of several species of *Acanthagrion*, *Oxyagrion* and *Erythrodiplax* in farm ponds, as observed in other studies (Renner, Perico, et al. 2016, Renner, Périco, et al. 2016), *Micrathyria* was the richest genus (five species, along with *Lestes*) in this habitat. However, this result should not be considered surprising, as individuals of *Micrathyria* commonly occur in lentic ecosystems, both in larval and adult stages (Garrison et al. 2006). Larvae of this genus also predominated in farm ponds in the study area (Pires et al. 2013, 2014). In addition, 30 species were exclusive of farm ponds (Table 2). The higher number of exclusive species in farm ponds was also detected in other regions of southern Brazil (Renner et al. 2016b). Previous reviews showed that man-made farm ponds can increase the regional pool of species richness, mostly by acting as an additional habitat for generalist species (Ruggiero et al. 2008). In spite of the increase in the taxonomic richness, some authors detected that in a landscape context, the predominance of generalist species could potentially lead to communities more functionally homogenous (Olden 2006; DeVicor et al. 2008). Although studies on the functional structure of odonates in farm ponds are lacking to our knowledge, this could also potentially apply to the case of odonate communities in farm ponds.

##### 5. Comments on the seasonal distribution and life cycles of species of Odonata in the study area

Previous data on the temporal distribution of larval assemblages of Odonata in farm ponds from the study area (Pires et al. 2014) showed that abundance peaked in spring (from September to November), suggesting that eggs hatched from spring on (after temperature warming). Taking into account larval growth periods and posterior emergence, the detected pattern for larvae could be related to the highest number of adult species observed in the present study between summer and autumn. Synchronized emergence of odonate adults in warmer periods has been also described in species-specific studies conducted in Argentina (Muzón et al. 1990, Capítulo 2000). In this context, our results suggest the completion of one generation in farm ponds, which is supported by the detection of 70% of the species recorded in farm ponds (N = 28) in one or two consecutive seasons, mostly summer and autumn. In fact, few species (N = 10; 25%) were detected in three seasons. *Acanthagrion gracile* and *Homeoura chelifera* were the sole species occurring throughout all seasons in farm ponds. This result loosely suggests that species from these genera were able to complete more than one generation per year, as previously detected in other studies (von Ellenrieder 2000).

Corbet et al. (2006) stressed that most odonates inhabiting perennial streams tend to show slow life cycles, producing one or less generations per year. To our knowledge, no study has yet attempted at assessing the life cycle of odonates in subtropical Brazilian streams. Our results suggest a similar pattern, as the complete absence of odonates in winter and the elevated number of species (N = 13; 65%) that were solely detected in one season (mostly non-consecutive - spring and autumn).

In this context, our results are generally in accordance with the review on voltinism of Odonata by Corbet et al. (2006), which described that most species in subtropical regions show at least one generation per year.

All results combined, they indicate a strong seasonal pattern in the activity of adult odonate species in the study area. Several studies showed that the life cycle of Odonata is limited by the seasonality of climatic conditions. In this study, the colder temperatures and reduced photoperiod typical of the winter should have played a stronger role in this result. Temperature strongly drives egg development rates, influencing the synchronized emergence of adult odonates, as well as their activity patterns (Corbet 2004). However, our results are in opposition to studies on tropical regions of Brazil, which suggested the role of wet and dry seasons on seasonal occurrence of adults (Franco & Takeda 2002, Vilela et al. 2016). One of the characteristics of the temperate humid climate in RS is the absence of a marked dry season (Maluf 2000), and thus adult activity was more likely to be associated with temperature rather than rainfall in the region.

### Final remarks

Our study showed that the middle course of the Jacuí River basin harbors a significant part (potentially up to one third) of the known species diversity of Odonata of the state of Rio Grande do Sul. The range of habitat types assessed likely accounted for the elevated richness, as odonate composition differed between farm ponds and streams. The widespread distribution of farm ponds harboring high local richness of Odonata (as well as an elevated number of exclusive taxa) contributed to the high diversity observed in the study area, reinforcing the role of man-made habitats in affecting spatial patterns of freshwater diversity. In addition, our results are also likely related to the well-preserved environmental conditions and historic land use of the landscape in the study area. The high declivity of the study area hinders the conversion of large areas for anthropic land uses, contributing for the existence of relatively large fragments of preserved and/or secondary-growth forests in this region (Marcuzzo et al. 1998). This context contrasts with the remnants of the other phytoecological regions of the Atlantic Forest in the state, which have been largely converted (Cordeiro & Hasenack 2009).

Our results also represented the first attempt to highlight the role of the phytoecological regions in the distribution of Odonata in southern Brazil. The compilation of our results and of data from the literature showed that the Seasonal Deciduous Forest supports a large part of the known diversity of Odonata in RS. In a broader perspective, our results reinforce the need of conservation programs in the Atlantic Forest biome, as currently only ~7.5% of the original area of this biome is represented in Rio Grande do Sul (Rio Grande do Sul 2019). Finally, our study also provided information on the life cycle of Odonata species in a subtropical region from Brazil. In this sense, marked seasonal fluctuations in the occurrence of adult odonates were detected in the study area. Most species occurred from summer to autumn and were absent in the winter, suggesting a low number of generations. These results likely suggest a strong role of the seasonal variations in temperature and photoperiod, typical of subtropical climate, in the life cycle of Odonata in southern Brazil.



## Supplementary material

The following online material is available for this article:

Figure S1 - Occurrence records of odonate species within the original area of Seasonal Deciduous Forest in the state of Rio Grande do Sul.

Table S1 - List of odonate species occurring in areas of Seasonal Deciduous Forests in the state of Rio Grande do Sul, Brazil.

## Author Contributions

Mateus Marques Pires: contribution to data collection, species identification, data analysis and interpretation, drafting and intellectual content of the manuscript.

Carla Bender Kotzian: contribution to project conception, data collection and interpretation; contribution to critical revision and intellectual content of the manuscript.

Cleber Sganzerla and Marina Schmidt Dalzochio: contribution to species identification and drafting of the manuscript.

Gabriel Prass: contribution to species identification.

Eduardo Périco: contribution to critical revision and intellectual content of the manuscript.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Frugivorous butterflies from the Atlantic Forest in Southern Brazil (Lepidoptera: Nymphalidae)

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**Abstract:** This study aimed to present a list of the species of frugivorous butterflies occurring in Atlantic Forests, in the Conservation Units: National Forest of Chapecó (FLONA), Ecological Station of Mata Preta (ESEC) and State Park of Araucárias (PAEAR) and adjacent forest fragments, located in the western region of the state of Santa Catarina. Three samplings were conducted between December 2017 and March 2018, totaling 24 days of collection in each sampling area. Van Someren-Rydon traps were used to capture frugivorous butterflies. There were 4,231 frugivorous butterflies belonging to four subfamilies, 12 tribes and 49 species. In all, 37 species of frugivorous butterflies were sampled in FLONA and 29 in adjacent forest fragments. In ESEC, 29 species and 33 in adjacent forest fragments. In PAEAR, 33 species and 28 in adjacent forest fragments. Of the total species registered, 15 species are new records for the state of Santa Catarina and 11 are new records for the western region of the state. The most abundant species for FLONA were: *Manataria hercyna* (Hübner, 1821) and *Hermeuptychia* sp. In ESEC, were *Hermeuptychia* sp. and *Ypthimoides ordinaria* (Freitas, Kaminski & Mielke, 2012). In PAEAR, greater abundance of *Forsterinaria quantius* (Godart, 1824) and *Eryphanes reevesii* (Doubleday, 1849) were verified. For the adjacent forest fragments to Conservation Units, there was a greater abundance of *Hermeuptychia* sp., *Moneuptychia soter* (Butler, 1877), *Morpho epistrophus* (Fabricius, 1796) e *Forsterinaria quantius* (Godart, 1824). Satyrinae presented higher richness ( $S = 34$ ) and abundance (90.58%) in all areas sampled. The rarefaction and extrapolation curves for the Conservation Units and adjacent forest fragments showed a greater rise in the FLONA and PAEAR sampling units and their adjacent forest fragments. The estimated sampling coverage for Conservation Unit and forest fragments was above 97%. The richness calculated through the Jackknife 1 estimator, for the FLONA and PAEAR samplings, presented a value of 50.75 and 37.09, respectively. The fauna of frugivorous butterflies from this region, first investigated in areas of Conservation Units, showed to be expressive and well represented in the Atlantic Forest Biome, indicating its potential as a refuge for biodiversity.

**Keywords:** conservation, diversity, ecology, forest fragmentation, species richness.

## Borboletas frugívoras da Mata Atlântica no Sul do Brasil (Lepidoptera: Nymphalidae)

**Resumo:** O estudo teve como objetivo elaborar uma lista das espécies de borboletas frugívoras ocorrentes em florestas da Mata Atlântica, nas Unidades de Conservação: Floresta Nacional de Chapecó (FLONA), Estação Ecológica da Mata Preta (ESEC) e Parque Estadual das Araucárias (PAEAR) e fragmentos florestais adjacentes, localizados na Região Oeste de Santa Catarina. Foram realizadas três campanhas de coletas entre dezembro de 2017 e março de 2018, totalizando 24 dias de coletas em cada área amostral. Para a captura das borboletas frugívoras, foram utilizadas armadilhas Van Someren-Rydon. Foram registradas 4231 borboletas frugívoras pertencentes a quatro subfamílias, 12 tribos e 49 espécies. Foram amostradas 37 espécies de borboletas frugívoras na FLONA e 29 nos fragmentos florestais adjacentes. Na ESEC 29 espécies e 33 nos fragmentos florestais adjacentes. No PAEAR 33 espécies e 28 nos fragmentos florestais adjacentes. Do total de espécies registradas, 15 espécies de borboletas frugívoras são novos registros para o estado de Santa Catarina e 11 são novos registros para a região



oeste do estado. As espécies mais abundantes para a FLONA foram: *Manataria hercyna* (Hübner, 1821) e *Hermeuptychia* sp. Na ESEC, foram *Hermeuptychia* sp. e *Ypthimoides ordinaria* (Freitas, Kaminski & Mielke, 2012). No PAEAR verificou-se maior abundância das espécies *Forsterinaria quantius* (Godart, 1824) e *Eryphanes reevesii* (Doubleday, 1849). Para os fragmentos florestais adjacentes das Unidades de Conservação houve maior abundância das espécies: *Hermeuptychia* sp., *Moneuptychia soter* (Butler, 1877), *Morpho epistrophus* (Fabricius, 1796) e *Forsterinaria quantius* (Godart, 1824). Satyrinae apresentou maior riqueza ( $S=34$ ) e abundância (90,58%) de borboletas frugívoras em todas as áreas amostradas. As curvas de rarefação e extrapolação, para as Unidades de Conservação e fragmentos florestais adjacentes mostraram uma maior ascendência nas unidades amostrais da FLONA e PAEAR e seus fragmentos florestais adjacentes. A cobertura estimada de amostragem para as borboletas frugívoras, para as Unidades de Conservação e fragmentos florestais, ficou acima de 97%. A riqueza calculada através do estimador Jackknife 1, mostrou-se superior à riqueza observada, sendo que para as amostragens na FLONA e PAEAR, o estimador apresentou um valor de 50,75 e 37,09, respectivamente. A fauna de borboletas frugívoras da região, investigada pela primeira vez em áreas de Unidades de Conservação, mostrou-se bastante expressiva e bem representada no Bioma Mata Atlântica, indicando seu potencial como refúgio da biodiversidade.

**Palavras-chave:** conservação, diversidade, ecologia, fragmentação das florestas, riqueza de espécies.

## Introduction

The Atlantic Forest is among the five main hotspots in the world (Morelato & Haddad 2000; Conservação Internacional 2013), considered one of the most important biomes worldwide due to its high biodiversity, high number of endemic species and deforestation rate (Myers et al. 2000). This biome covered approximately 13% of the Brazilian territory (Cabral & Cesco 2008), currently there are less than 12% of the original area, and the remaining areas are represented by more than 245,000 fragments, of which more than 95% are smaller than 250 hectares (Ribeiro et al. 2009).

The western region of the state of Santa Catarina was severely deforested throughout the 20th century, mainly by the economic development after European colonization, characterized by logging (Cabral & Cesco 2008). As a consequence, large forest areas were gradually transformed into isolated fragments (Cerqueira et al. 2003, Cabral & Cesco 2008). It is notable that over the centuries the process of forest fragmentation has intensified due to human activities on a continuous basis, generating effects such as the expansion of agricultural areas, the formation of urban areas and the construction of roads (Haddad et al. 2015).

Vibrans et al. (2013) present data from the Forest Floristic Survey of Santa Catarina highlighting that the state covers three different phytophysiognomies that make up the Atlantic Forest: Dense Ombrophilous Forest, Mixed Ombrophilous Forest and Deciduous Seasonal Forest. The results show that the remaining native forest cover in the state is approximately 29%. In the west of Santa Catarina, the forest cover of the Deciduous Seasonal Forest is between 16 and 24% of the Mixed Ombrophilous Forest (Vibrans et al. 2013).

Due to the rapidity with which anthropic impacts occur, selecting species or assemblages of species to establish conservation and monitoring priorities is critical (Kremen 1992). The composition, richness and abundance of Lepidoptera in the environments can be indicators of the degree of environmental preservation, being its study of fundamental importance in the understanding of the ecological interactions between the different environments (Duarte et al. 2012).

Considering this, the importance of studying butterflies came with the progress of research on biodiversity conservation, given the sensitivity to environmental changes, the fragmentation and reduction of

natural areas (Freitas 2010). This is explained by the fact that butterflies are a large taxonomic group, faithful to their habitats, well known, quickly sampled and easily identified (Brown Jr. & Freitas 2000).

Lepidoptera correspond to approximately 26,000 species described throughout Brazil (Brown & Freitas 1999; Freitas & Almeida 2012), of which more than 3,250 are butterflies (Freitas & Marini-Filho 2011). In Brazil, strictly frugivorous butterflies are represented by four subfamilies of Nymphalidae: Satyrinae, Charaxinae, Biblidinae and some genera of Nymphalinae (Freitas et al. 2014). This guild comprises 50-75% of the Neotropical nymphalid fauna (Brown Jr. 2005), and because they are taxonomically and ecologically diverse in tropical environments, occur in all Brazilian biomes (Freitas et al. 2014).

Despite the diversity of habitats, there is little research concerning the order Lepidoptera in the state of Santa Catarina (Carneiro et al. 2008, Siewert et al. 2010a, Corso & Hernandez 2012, Belaver et al. 2012, Orlandim et al. 2016). An extensive study was conducted by Ferro et al. (2012) with the moths Arctiinae and Siewert et al. (2010b) with Sphingidae. The knowledge of the fauna of butterflies in the western region of the state began with the contribution of the ancient naturalist Fritz Plaumann, who left a collection of approximately 4,000 butterflies, deposited at the Fritz Plaumann Entomological Museum in the municipality of Seara (Lubenow 2016). Some recent surveys of Lepidoptera (considering butterflies and/or moths), using entomological nets, were conducted in the western region in Santa Catarina, highlighting the research done by Silva et al. (2011), Favretto (2012), Schmith et al. (2012), Favretto et al. (2013), Favretto & Santos (2014), Fanton & Sabedot-Bordin (2014), Favretto et al. (2015), Silva & Sabedot-Bordin (2015) and Colpani & Sabedot-Bordin (2018).

Although some researchers have made collections of butterflies in the state of Santa Catarina using an entomological net, the data currently available in publications can still be considered scarce (Piovesan et al. 2014). The lack of publications on frugivorous butterflies in Conservation Units and adjacent forest fragments, in the western region in Santa Catarina, is evidenced through the bibliographic search, using a trap with attractive bait as a collection technique. With the purpose of contributing to the knowledge about the fauna of butterflies of Santa Catarina and in order to provide subsidies for the conservation of species, the study aimed to elaborate a list of frugivorous butterfly species of the western region of the state.

## Material and Methods

### 1. Study areas

The study was conducted in two Federal Conservation Units: National Forest of Chapecó (FLONA) (27°06'24.8"S and 52°46'59.3"W) and Ecological Station of Mata Preta (ESEC) (26°30'57.31"S and 52°7'59.69"W) and a State Conservation Unit: State Park of Araucárias (PAEAR) (26°27'08"S and 52°33'56"W). All Conservation Units are fully protected and are located in the western region of the state of Santa Catarina, southern Brazil (Figure 1). The Conservation Units are inserted in areas that during the last decades suffered from intense pressure of forest exploitation and agricultural expansion. Around the Conservation Units, there are monocultures such as soybean and corn, grown by the conventional method and often occurring the use of transgenics (Apremavi 2009). All Conservation Units are inserted in the Atlantic Forest Biome with forest phytophysiognomies classified as Mixed Ombrophilous Forest with different successional stages (Dick et al. 2013).

FLONA is located in the municipalities of Chapecó and Guatambú, was created in 1968 and has an area of 1,590 hectares. Samplings of frugivorous butterflies were performed in fragment I of FLONA with an area of 1,287.54 hectares, located in Guatambú (ICMBio 2013).

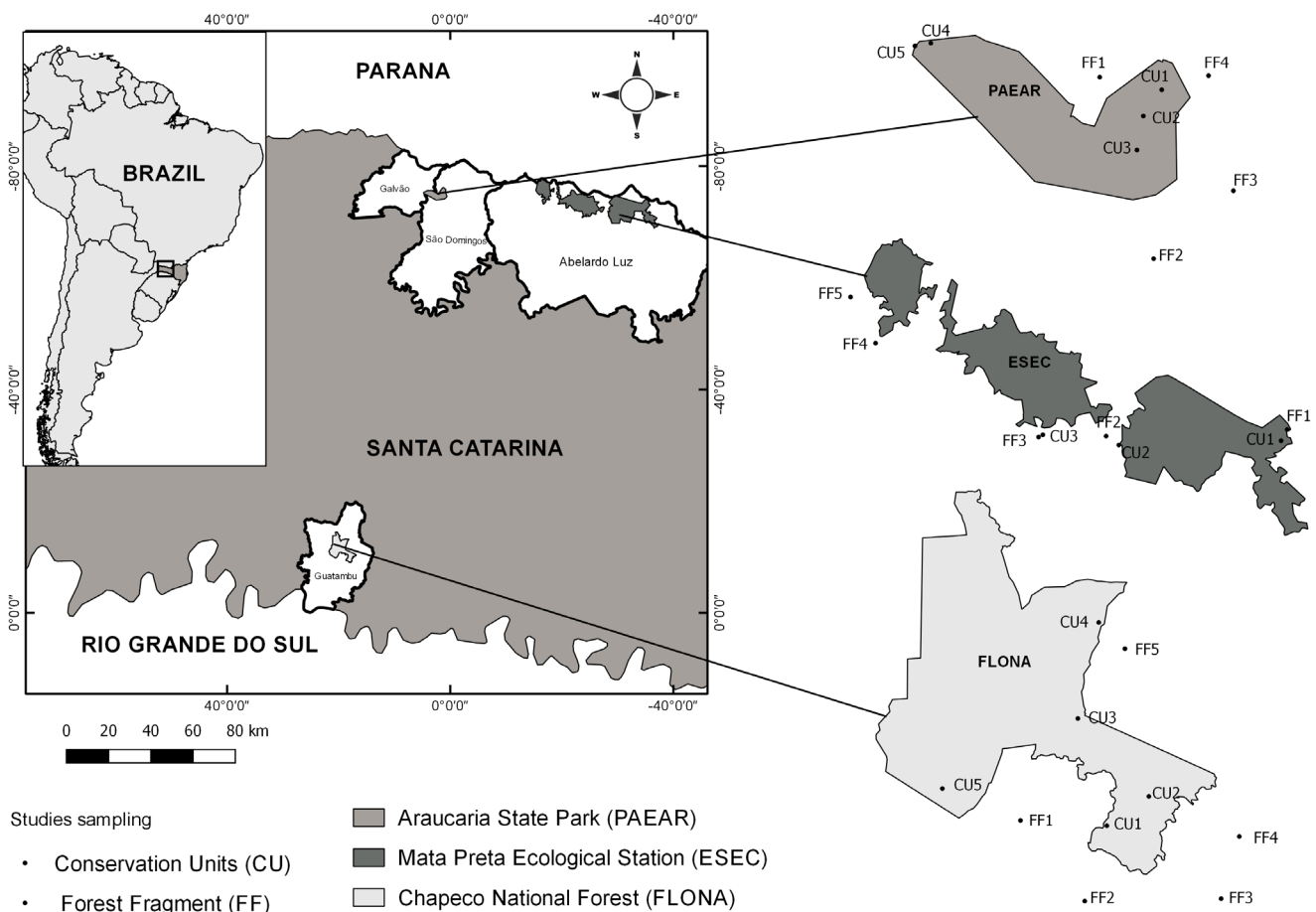
The ESEC of Mata Preta was established in 2005, has an area of 6,536 hectares and is located in the municipality of Abelardo Luz. Extensions of the ESEC of Mata Preta constitute private areas whose owners present legal proceedings in progress regarding the formation of the Conservation Unit (Apremavi 2009).

PAEAR was created in 2003 and covers an area of 612.5 hectares. PAEAR is located between the municipalities of São Domingos and Galvão. The creation of the park was a compensatory action established by the formation of the reservoir of the Quebra Queixo Hydroelectric Power Plant, located in the Chapecó River, in the municipalities of Ipuacu and São Domingos (Fatma 2016).

The climate of the western region of the state of Santa Catarina is cfa, subtropical humid, with abundant rainfall well distributed throughout the year. The average annual temperature is lower than 18°C and with average temperatures ranging from 13°C and 25°C (Alvares et al. 2014).

### 2. Sampling design

In order to collect the frugivorous butterflies, five sampling units (five for FLONA and PAEAR, three for ESEC) were defined inside the Conservation Units (CU) and a single sampling unit in each of the adjacent forest fragments of each CU. Five adjacent forest



**Figure 1.** Map of the study areas and sample units for collections of frugivorous butterflies in the municipalities of Guatambú, Abelardo Luz and São Domingos, Santa Catarina, from December 2017 to March 2018.

fragments were defined for FLONA and ESEC, and four for PAEAR (Figure 1). Adjacent forest fragments were different in size and had different distances from CU. Among the adjacent forest fragments there was a minimum distance of 250 meters (Santos et al. 2014). The sampling unit was formed by a linear transect. At the transect, the first trap was allocated at a distance of at least 50 meters from the edge (Uehara-Prado 2003). In each transect, there were a set of five traps for the capture of butterflies (Freitas et al. 2014), distanced from 30 to 50 meters (Santos et al. 2014) from each other, depending on the availability of places to hang them in the trees. The other transects of the sampling units with a minimum distance of 250 meters between them. A total of 135 traps were installed, 50 traps in FLONA/fragments (25/25); 40 traps in ESEC/fragments (15/25) and 45 in PAEAR/fragments (25/20).

Three samplings were conducted in each CU and respective adjacent forest fragments, from December 2017 to March 2018, totaling 24 days of collection. The traps were left active on the field for eight consecutive days, being inspected every 48 hours for removal of the captured frugivorous butterflies and bait replacement.

The procedure for the collection of frugivorous butterflies followed the protocol established by the National Lepidoptera Research and Conservation Network (RedeLep). Van Someren-Rydon traps were used to collect frugivorous butterflies. The traps were suspended in trees by ropes, at a height of approximately 1.5 m above the ground level (Uehara-Prado 2005). Each trap was supplied with a 50 mL plastic bottle containing an attractive bait. The bait used consisted of a mixture of sugarcane juice with well-ripe bananas at a proportion of 1/3, which was prepared 48 hours before the beginning of the sampling, time required to occur to the fermentation (Uehara-Prado 2003).

The collected frugivorous butterflies were sacrificed by thoracic pressure at the base of the wings and conditioned in properly identified entomological envelopes (Almeida et al. 1998). The collected butterflies were taken to the Laboratory of Entomology of the Universidade Comunitária da Região de Chapecó (UNOCHAPECÓ) in Santa Catarina, Brazil, for freezer storage and subsequent identification of the species. Species identification was carried out through specialized literature of Canals (2003), Lamas (2004), Wahlberg (2009) and using online identification guide (<http://butterfliesofamerica.com>). Identification was also performed with the help of experts.

Specimens of each sampled species were deposited in the reference collection of the Laboratory of Entomology of the Universidade Comunitária da Região de Chapecó (UNOCHAPECÓ), Chapecó, Santa Catarina; in the Zoological Collection of the Universidade do Vale do Rio dos Sinos (MZ UNISINOS), São Leopoldo, Rio Grande do Sul, Brazil.

The collections were performed under the license issued by ICMBio (ICMBio/SISBio Collection License 60789-1).

To represent the composition, richness and abundance of frugivorous butterflies in CU and adjacent forest fragments, the species were listed according to their respective taxa of subfamilies, tribe and genus in addition to the presence in each environment.

Richness estimates and sample coverage were performed using the rarefaction and extrapolation curve based on the Chao 1 estimator with 40 nodes and 500 randomizations. The analysis was run with the iNEXT software (Chao et al. 2016).

Sampling adequacy was verified from the Jackknife 1 species richness estimator for all sampled areas using the EstimateS 9.1 software (Colwell 2013), as suggested by Toti et al. (2000). We used EstimateS with 500 randomizations.

To illustrate butterflies species composition comparisons two ordinations were plotted (Non-metric MultiDimensional Scaling - nMDS) using a qualitative similarity index (Simpson). Qualitative analysis was performed using a species presence-absence matrix, using the Simpson index to build a dissimilarity matrix among sample units. To test for statistical differences among butterfly assemblage composition for the above similarity indexes, we applied two one-way ANOSIM (Analysis of Similarities) with Bonferroni correction, one for each factor. We also performed an analysis of similarity percentage (SIMPER) (Clarke & Warwick 1994), based on Bray-Curtis distance, in order to identify those taxa responsible for observed differences in species composition among forest classes, also quantifying their relative contribution for such differences. Composition analyses were developed using PAST (Paleontological Statistics 2.17c, Hammer & Harper 2009).

## Results

There were 4,231 frugivorous butterflies belonging to four subfamilies, 12 tribes and 49 species (Table 1). In total, 49 species of frugivorous butterflies (2,418 individuals) were sampled in FLONA (N=670, S=37) and adjacent forest fragments (N=1,748, S=29); 37 species (1,234 individuals) in ESEC (N=547, S=29) and adjacent forest fragments (N=687, S=33) and 35 species (579 individuals) in PAEAR (N=368, S=33) and adjacent forest fragments (N=211, S=28) (Table 1).

Of the total richness in FLONA and adjacent forest fragments, 25 species occurred in all sampled areas. In all, 12 species were sampled exclusively in the areas of CU and four in the adjacent forest fragments. In ESEC, 25 species were common to the areas sampled, four species were sampled exclusively in the areas of CU and eight occurred exclusively in the adjacent forest fragments. In turn, in PAEAR, 26 species were common to the sampled areas, another seven species were sampled exclusively in the CU areas and two occurred in the adjacent forest fragments (Table 1).

From the total of species, 15 species of frugivorous butterflies are new records for the state of Santa Catarina, namely 12 Satyrinae, two Biblidinae and one Charaxinae (Table 1). New species records for the state: *Zaretis strigosus* (Gmelin, 1790), *Cissia eous* (Butler, 1867), *Moneuptychia soter* (Butler, 1877) recorded in all sampled areas. *Forsterinaria necys* (Godart, 1824) was not recorded in the adjacent forest fragments of FLONA. *Carminda paeon* (Godart, 1804) was not sampled in PAEAR and adjacent forest fragments. *Callicore hydaspes* (Drury, 1782) was recorded only in FLONA and adjacent forest fragments; *Paryphthimoides poltys* (Prittwitz, 1865) was not sampled in ESEC, PAEAR and adjacent forest fragments. *Eunica tatila* (Herrich-Schäffer, 1855) and *Splendeuptychia libitina* (Butler, 1870), singleton species, occurring only in FLONA. *Catoblepia amphirhoe* (Hübner, 1825) was recorded in FLONA and adjacent forest fragments of PAEAR (doubleton) and *Pseudodebis euptychidia* (Butler, 1868) was sampled in FLONA, PAEAR and adjacent forest fragments. *Splendeuptychia ambra* (Weymer, 1911) registered in PAEAR. *Taygetis laches* (Fabricius, 1793) was not sampled in the adjacent forest fragments of PAEAR, FLONA

**Table 1.** List of species of frugivorous butterflies sampled with Van Someren-Rydon traps recorded in Conservation Units and adjacent forest fragments, western region of the state of Santa Catarina, Brazil, between December 2017 and March 2018. (ES) ESEC; (F) FLONE; (P) PAEAR; (T) Total butterflies in Conservation Units; (FES) ESEC Fragments; (FF) FLONA Fragments; (FP) PAEAR Fragments; (FT) Total butterflies in forest fragments; \*New records for Santa Catarina; \*\*New records for the western region of state of Santa Catarina.

TAXON	CONSERVATION UNITS				FOREST FRAGMENTS			
	ES	F	P	T	FES	FF	FP	FT
<b>Charaxinae (S=5)</b>								
<b>Preponini (S=2)</b>								
<i>Archaeoprepona chalciope</i> (Hübner, 1823)	-	1	8	9	1	-	2	3
<i>Archaeoprepona demophon</i> (Hübner, 1814)**	2	1	2	5	3	-	2	5
<b>Anaeni (S=3)</b>								
<i>Memphis acidalia victoria</i> (H. Druce, 1877)	1	2	4	7	4	5	-	9
<i>Memphis moruus stheno</i> (Prittwitz, 1865)	-	6	12	18	7	26	10	43
<i>Zaretis strigosus</i> (Gmelin, 1790)*	2	4	2	8	6	11	1	18
<b>Biblidinae (S=10)</b>								
<b>Biblidini (S=1)</b>								
<i>Biblis hyperia</i> (Cramer, 1779)	5	21	6	32	15	35	9	59
<b>Callicorini (S=1)</b>								
<i>Callicore hydaspes</i> (Drury, 1782)*	-	1	-	1	-	2	-	2
<b>Epicaliini (S=3)</b>								
<i>Cybdelis phaesyla</i> (Hübner, 1825)	1	1	-	2	3	1	-	4
<i>Eunica eburnea</i> Fruhstorfer, 1907	3	-	-	3	-	-	-	-
<i>Eunica tatila</i> (Herrich-Schäffer, 1855)*	-	1	-	1	-	-	-	-
<b>Epiphilini (S=3)</b>								
<i>Epiphile hubneri</i> (Hewitson, 1952)	11	2	1	14	12	3	-	15
<i>Epiphile oreia oreia</i> (Hübner, 1823)	13	18	1	32	9	-	2	11
<i>Temenis laothoe</i> Cramer, 1777	-	1	-	1	-	6	-	6
<b>Ageroniini (S=2)</b>								
<i>Hamadryas amphinome</i> (Linnaeus, 1767)	-	-	-	-	-	1	-	1
<i>Hamadryas epinome</i> (Felder & Felder, 1867)	3	1	1	5	14	5	7	26
<b>Satyrinae (S=33)</b>								
<b>Brassolini (S=8)</b>								
<i>Blepolenis bassus</i> (Felder & Felder, 1867)	-	-	-	-	1	-	-	1
<i>Blepolenis catharinae</i> (Stichel, 1902)**	-	-	-	-	4	-	-	4
<i>Caligo illioneus</i> (Cramer, 1776)	-	4	-	4	-	2	-	2
<i>Caligo martia</i> (Godart, 1824)	-	-	1	1	-	-	1	1
<i>Catoblepia amphirhoe</i> (Hübner, 1825)*	-	1	-	1	-	-	1	1
<i>Eryphanis reevesii</i> (Doubleday, 1849)	65	29	56	150	20	7	9	36
<i>Opoptera sulcius</i> (Staudinger, 1887)**	18	-	15	33	11	-	10	21
<i>Opsiphanes invirae</i> (Hübner, 1808)	2	2	1	5	1	-	1	2
<b>Satyrini (S=21)</b>								
<i>Carmina paeon</i> (Godart, 1804)*	7	1	-	8	12	3	-	15
<i>Cissia eous</i> (Butler, 1867)*	10	8	18	36	32	139	11	182
<i>Cissia phronius</i> (Godart, 1824)	1	6	7	14	6	64	8	78
<i>Euptychoides castrensis</i> (Schaus, 1902)**	19	13	1	33	15	72	-	87
<i>Forsterinaria necys</i> (Godart, 1824)*	6	2	20	28	1	-	2	3
<i>Forsterinaria quantius</i> (Godart, 1824)	21	43	65	129	9	6	23	38
<i>Godartiana muscosa</i> (Butler, 1870)**	1	-	6	7	5	1	2	8



Continuation Table 1.

TAXON	CONSERVATION UNITS				FOREST FRAGMENTS			
	ES	F	P	T	FES	FF	FP	FT
<i>Hermeuptychia</i> sp.**	189	114	28	331	141	494	22	657
<i>Moneuptychia soter</i> (Butler, 1877)*	51	32	15	98	100	369	22	491
<i>Pareuptychia ocirrhoe</i> (Fabricius, 1776)**	-	147	4	151	16	97	5	118
<i>Paryphthimoides poltys</i> (Prittwitz, 1865)*	-	4	-	4	3	53	-	56
<i>Pseudodebis euptychidia</i> (Butler, 1868)*	-	5	1	6	-	1	1	2
<i>Splendeuptychia ambra</i> (Weymer, 1911)*	-	-	1	1	-	-	-	-
<i>Splendeuptychia libitina</i> (Butler, 1870)*	-	1	-	1	-	-	-	-
<i>Taygetis acuta</i> Weymer, 1910**	-	2	31	33	2	-	7	9
<i>Taygetis laches</i> (Fabricius, 1793)*	2	-	4	6	-	3	-	3
<i>Taygetis ypthima</i> Hübner, 1821**	4	3	7	14	-	-	2	2
<i>Yphthimoides celmis</i> (Godart, 1824)*	1	-	11	12	-	19	6	25
<i>Yphthimoides ordinaria</i> Freitas, Kaminski & Mielke, 2012	78	9	9	96	90	203	18	311
<i>Yphthimoides straminea</i> (Butler, 1867) *	4	-	-	4	35	-	1	36
<i>Zischkaia pacarus</i> (Godart, 1824)**	-	1	1	2	-	-	-	-
<b>Melanitini (S=1)</b>								
<i>Manataria hercyna</i> (Hübner, 1821)	-	164	21	185	3	95	11	109
<b>Morphini (S=3)</b>								
<i>Morpho aega</i> (Hübner, 1822)	-	1	-	1	-	-	-	-
<i>Morpho epistrophus</i> (Fabricius, 1796)	23	-	-	23	93	-	-	93
<i>Morpho helenor</i> (Cramer, 1776)	2	10	2	14	11	5	-	16
<b>Nymphalinae (S=1)</b>								
<b>Coeini (S=1)</b>								
<i>Smyrna blomfieldia</i> (Fabricius, 1781)**	2	8	6	16	2	20	15	37
<b>N Total</b>	<b>547</b>	<b>670</b>	<b>368</b>	<b>1585</b>	<b>687</b>	<b>1748</b>	<b>211</b>	<b>2646</b>

and their adjacent forest fragments. *Yphthimoides straminea* (Butler, 1867) in the ESEC, FLONA and adjacent forest fragments of PAEAR. *Yphthimoides celmis* (Godart, 1824) did not occur in the FLONA and adjacent forest fragments of the ESEC (Table 1).

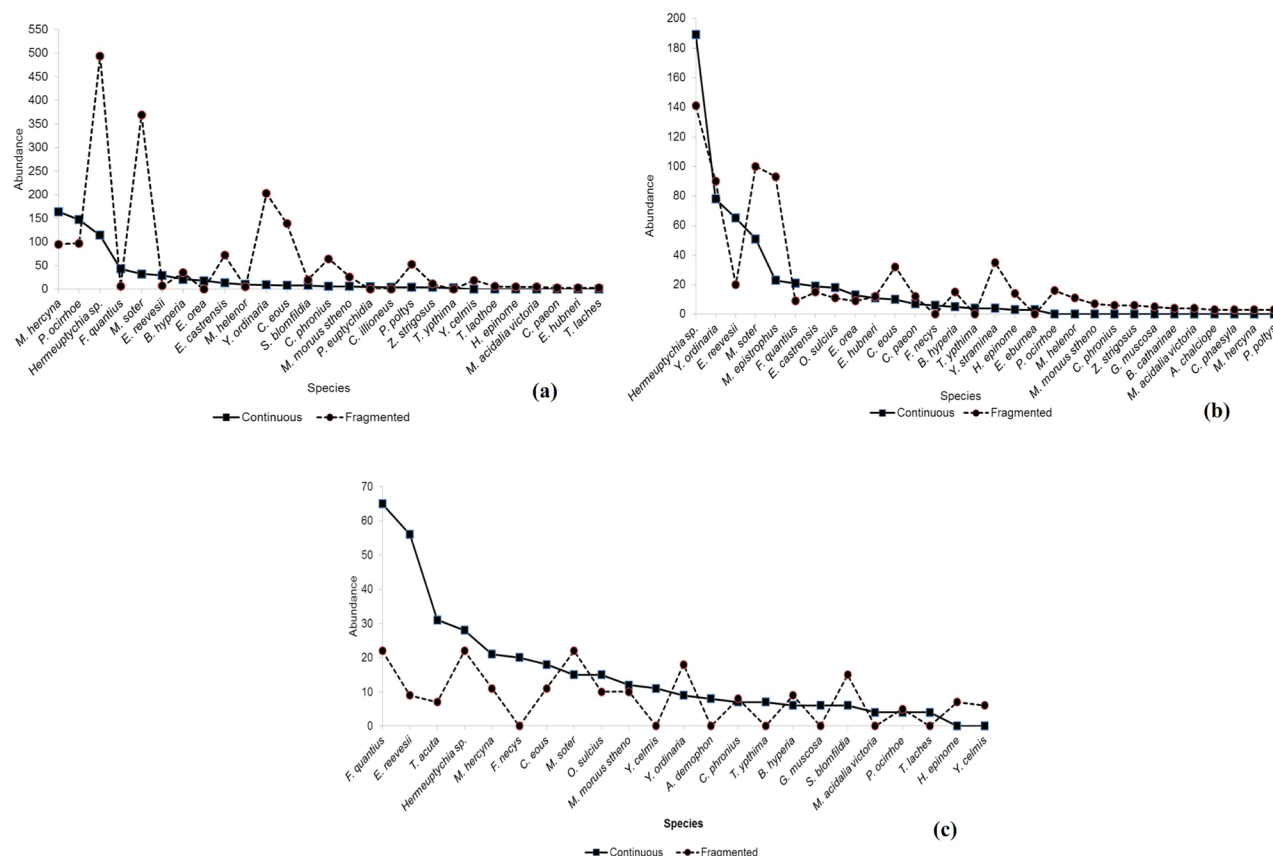
The study added 11 new records of frugivorous butterflies to the western region of the state of Santa Catarina, with eight Satyrinae, one Charaxinae and Nymphalinae (Table 1). *Hermeuptychia* sp., *Pareuptychia ocirrhoe* (Fabricius, 1776) and *Smyrna blomfieldia* (Fabricius, 1781) recorded in all areas sampled. *Archaeoprepona demophon* (Hübner, 1814) recorded in all areas sampled with the exception of the adjacent forest fragments of FLONA. *Blepopenis catharinae* (Stichel, 1902) sampled in ESEC. *Opoptera sulcius* (Staudinger, 1887) was not recorded in FLONA and adjacent forest fragments. *Euptychoides castrensis* (Schaus, 1902) did not occur in the adjacent forest fragments of PAEAR. *Godartiana muscosa* (Butler, 1870) was not registered in FLONA. *Taygetis acuta* (Weymer, 1910) was not recorded in ESEC and adjacent forest fragments of FLONA. *Taygetis yptima* (Hübner, 1821) was not recorded in the adjacent forest fragments of ESEC and FLONA and *Zischkaia pacarus* (Godart, 1824) was only sampled in FLONA (singleton) (Table 1).

The most abundant species in FLONA were *Manataria hercyna* (Hübner, 1821) with 164 individuals (24.48%), *P. ocirrhoe* (Fabricius, 1776) with 147 (21.94%) and *Hermeuptychia* sp. with 114 (17.01%)

(Table 1, Figure 2a). On the other hand, in the CU, 27 species of frugivorous butterflies totaled less than 10 individuals, being generally restricted to a single area sampled. Among the total fauna of frugivorous butterflies in the CU, 12 singletons were recorded: *A. demophon* (Hübner, 1814), *Archaeoprepona chalciope* (Hübner, 1814), *C. hydaspes* (Drury, 1782), *C. paeon* (Godart, 1804), *C. amphirhoe* (Hübner, 1825), *Cybdelis phaeosyla* (Hübner, 1825), *E. tatila* (Herrich-Schäffer, 1855), *Hamadryas epinome* (Felder & Felder, 1867), *Morpho aega* (Hübner, 1822), *S. libitina* (Butler, 1870), *Temenis laothoe* (Cramer, 1777) and *Z. pacarus* (Godart, 1824) and five doubletons: *Epiphile hubneri* (Hewitson, 1952), *F. necys* (Godart, 1824), *Memphis acidalia victoria* (H. Druce, 1877), *Opsiphanes invirae* (Hübner, 1808) and *T. acuta* (Weymer, 1911) (Table 1, Table 2).

In the adjacent forest fragments of FLONA, it was verified that the most abundant species were *Hermeuptychia* sp. with 494 individuals (28.26%), *M. soter* (Butler 1877) with 369 (21.11%) and *Y. ordinaria* (Freitas, Kaminski & Mielke 2012) with 203 (11.61%) (Table 1, Figure 2a). In these forest fragments, 15 species of frugivorous butterflies numbered less than 10 individuals and four singletons were recorded: *C. phaeosyla* (Hübner, 1825), *G. muscosa* (Butler, 1870), *Hamadryas amphinome* (Linnaeus, 1767) and *P. euptychidia* (Butler, 1868) and two doubletons: *C. hydaspes* (Drury, 1782) and *Caligo illioneus* (Cramer, 1776) (Table 1, Table 2).

## Frugivorous butterflies in Southern Brazil



**Figure 2.** Distribution of abundance of frugivorous butterfly species sampled with Van Someren-Rydon traps in Conservation Units and adjacent forest fragments, from December 2017 to March 2018, in the western region of Santa Catarina, Brazil. 2a = FLONA; 2b = ESEC; 2c = PAEAR.

**Table 2.** Species richness, abundance and Jackknife 1 species richness estimator for the guild of frugivorous butterflies sampled with Van Someren-Rydon traps, recorded in Conservation Units (CU) and adjacent forest fragments (FF), in the western region of the state of Santa Catarina, between December 2017 and March 2018.

Sampling areas	FLONA/CU	FLONA/FF	ESEC/CU	ESEC/FF	PAEAR/CU	PAEAR/FF
Species richness	37	29	29	33	33	28
Abundance	670	1748	547	687	368	211
Singletons	12	4	5	4	9	6
Doubletons	5	2	6	2	3	6
Jackknife 1	50.75±4.09	34.58±2.12	36.36±2.43	37.52±1.63	43.08±2.12	37.09±3.14

In ESEC, *Hermeuptychia* sp. with 189 individuals (34.55%), *Y. ordinaria* (Freitas, Kaminski & Mielke, 2012) with 78 (14.26%) and *Eryphanes reevesii* (Doubleday, 1849) with 65 (11.88%) were the most abundant species (Table 1, Figure 2b). In the CU, it was observed that 18 species of frugivorous butterflies totaled less than 10 individuals. Five species of singleton frugivorous butterflies were recorded: *C. phronius* (Godart, 1824), *C. phaesyia* (Hübner, 1825), *G. muscosa* (Butler, 1870), *M. acidalia victoria* (H. Druce, 1877) and *Ypthimoides celmis* (Godart, 1824), and six doubletons *A. chalciope* (Hübner, 1823), *Morpho helenor* (Cramer, 1776), *O. invirae* (Hübner, 1808), *S. bomfildia* (Fabricius, 1781), *T. laches* (Fabricius, 1793) and *Z. strigosus* (Gmelin, 1790) (Table 1, Table 2).

*Hermeuptychia* sp. with 141 individuals (20.52%), *M. soter* (Butler, 1877) with 100 (14.55%) and *Morpho epistrophus* (Fabricius, 1796) with 93 (13.53%) presented greater abundance in the adjacent forest fragments of ESEC (Table 1, Figure 2b). In these forest fragments, there were 18 species of frugivorous butterflies with less than 10 species, four

singletons: *A. demophon* (Hübner, 1814), *Blepolenis bassus* (Felder & Felder, 1867), *F. neceys* (Godart, 1824) and *O. invirae* (Hübner, 1808) and two doubletons: *S. bomfildia* (Fabricius, 1781) and *T. acuta* (Weyer, 1910) (Table 1, Table 2).

In PAEAR, there was a greater abundance of *Forsterinaria quantius* (Godart, 1824) with 65 individuals (17.66%), *E. reevesii* (Doubleday, 1849) with 56 (15.22%) and *T. acuta* Weyer, 1910 with 31 individuals (8.42%) (Table 1, Figure 2c). It was observed that in the CU, 22 species of frugivorous butterflies with less than 10 individuals were sampled. Also, nine species of singleton frugivorous butterflies were found: *Caligo martia* (Godart, 1824), *E. hubneri* (Hewitson, 1952), *Epiphile orea orea* (Hübner, 1823), *E. castrensis* (Schaus, 1902), *H. epinome* (Felder & Felder, 1867), *O. invirae* (Hübner, 1808), *P. euptychidia* (Butler, 1868), *Splendeuptychia ambra* (Weyer, 1911) and *Z. pacarus* (Godart, 1824) and three doubletons: *A. chalciope* (Hübner, 1823), *M. helenor* (Cramer, 1776) and *Z. strigosus* (Gmelin, 1790) (Table 1, Table 2).

The most abundant species in the adjacent forest fragments of PAEAR were *F. quantius* (Godart, 1824) com 23 individuals (10.90%), *Hermeuptychia* sp. and *M. soter* (Butler, 1877) with 22 (10.42%) (Table 1, Figure 2c). For the forest fragments, 19 species of frugivorous butterflies with less than 10 species, six singletons: *C. martia* (Godart, 1824), *C. amphirhoe* (Hübner, 1825), *O. invirae* (Hübner, 1808), *P. euptychidia* (Butler, 1868), *Ypthimoides straminea* (Butler, 1867) e *Z. strigosus* (Gmelin, 1790) and six doubletons: *A. demophon* (Hübner, 1814), *A. chalciope* (Hübner, 1823), *E. orea orea* (Hübner, 1823), *F. necys* (Godart, 1824), *G. muscosa* (Butler, 1870) and *T. ypthima* (Hübner, 1821) (Table 1, Table 2).

The general distribution of abundance showed a pattern of high dominance (Table 1, Figure 2a, Figure 2b, Figure 2c). The five most abundant species make up more than 70% of the individuals sampled in FLONA (Figure 2a) and ESEC (Figure 2b). The five most abundant species in the adjacent forest fragments of FLONA also presented the same pattern of dominance (Figure 2a). The five most abundant species in the adjacent forest fragments of ESEC corresponded to more than 60% of the individuals sampled (Figure 2b). On the other hand, in PAEAR the five most abundant species totaled more than 50% of the individuals sampled and in the adjacent forest fragments of this CU, 46% dominance was observed in the five most abundant species (Figure 2c).

Satyrinae presented higher species richness (69.38%) and abundance (90.58%) of frugivorous butterflies in all sampled areas (Table 1, Table 3), followed by Biblidinae (5.19%). Most of the captured frugivorous butterflies belong to the tribe Satyrini, being associated with all forest areas sampled (Table 1). It was found that in the adjacent forest fragments there was a higher percentage of Satyrinae when compared to CU (Table 3).

From the rarefaction and extrapolation curve, for the CU and respective forest fragments, based on the Chao 1 estimator (Figure 3), a total richness estimate was generated for the CU and adjacent forest fragments. The estimated sampling coverage for frugivorous butterflies in the CU and adjacent forest fragments was above 97%. Figure 3 shows that the richness approached an asymptote, indicating a greater rise in the sampling areas of FLONA, and the PAEAR and adjacent forest fragments.

The parameters of richness and abundance of frugivorous butterflies showed variations, mainly between the areas in the CU and their respective forest fragments. The number of species of frugivorous butterflies varied between 28 (PAEAR/FF) and 37 (FLONA/CU). The abundance in turn ranged from 211 (PAEAR/FF) to 1748 (FLONA/FF) butterflies (Table 2).

The Jackknife 1 richness estimator indicated that 72.9% of the frugivorous butterflies were sampled in FLONA and 83.7% for the

adjacent forest fragments. In ESEC, the estimator indicated a sampling of 79.8% and 87.9% for the adjacent forest fragments. In PAEAR, the analysis indicated that 76.6% of the frugivorous butterflies were sampled and in the adjacent forest fragments, 75.5% (Table 2). The expected richness, calculated through the Jackknife 1 estimator, was higher than the richness obtained in the samplings, and for the FLONA and PAEAR samplings, the estimator presented a high value (50.75 and 37.09) (Table 2).

Qualitative ANOSIM analyses indicated different species composition among Conservations Units (ANOSIM:  $R = 0.43$ ,  $p = 0.001$ , Figure 4a) and by forest fragments vs Conservation Units (ANOSIM:  $R = 0.10$ ,  $p = 0.001$ , Figure 4a). There is apparent founder effect in terms of species composition, since difference among sites (CU and FF) and between Conservation Units was found for Simpson index. Figure 4 illustrates a visual inspection of the nMDS scatterplots by species composition differences among Conservations Units (Figure 4a) and by forest fragments vs Conservation Units (Figure 4b).

The contribution of the most representative species in each environment to dissimilarity (SIMPER) between Conservation Units is presented in Table 4, and for different environments (CU vs FF) in Table 5. The most dominant species was *Hermeuptychia* sp. (overall contribution: 15.9%, for Conservation Units, Table 4; overall contribution: 15.5%, for CU vs FF, Table 5).

The species of greatest contribution to the dissimilarity among the CU belong to Satyrinae: *Hermeuptychia* sp. (15.9%), *M. hercyna* (9.4%) and *M. soter* (9%) (Table 4). These same species presented dissimilarity when analyzed between different environments, only *M. soter* (9.27%) contributed more than *M. hercyna* (8.53%) (Table 5).

## Discussion

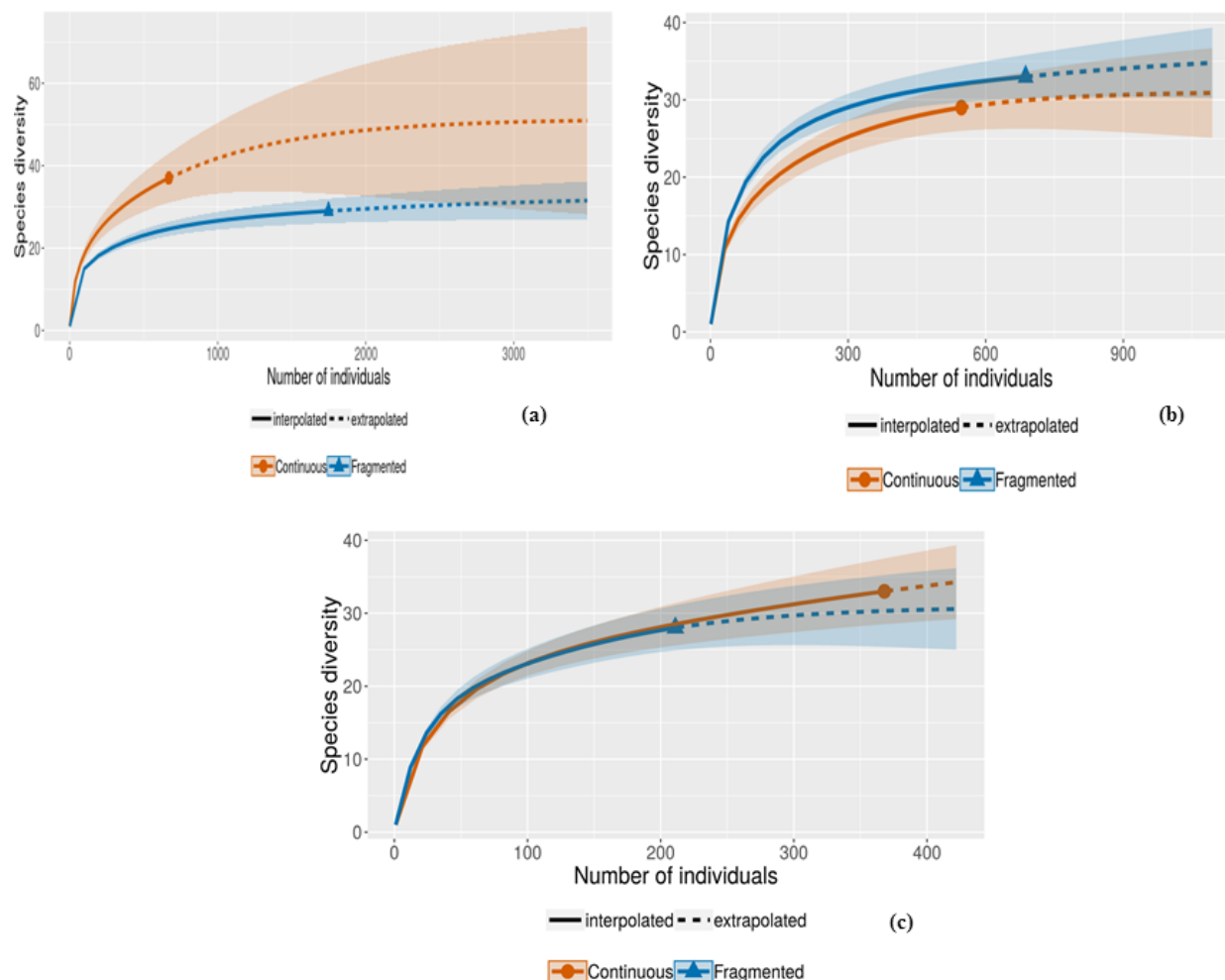
The total richness of frugivorous butterflies recorded in the present study was superior to what has been found for the state and western region of Santa Catarina [Carneiro et al. (2008) with 22; Corso & Hernandez (2012), with 20 species; Schmidt et al. (2012) with two, Piovesan et al. (2014) with 43, Fanton & Sabedot-Bordin (2014) with 14, Favretto et al. (2015), with 9, Silva & Sabedot-Bordin (2015) with 16 and Colpani & Sabedot-Bordin (2018) with 26]. The recent increase in the biodiversity studies on Lepidoptera is in the state of Santa Catarina highlighted, with surveys carried out in the municipalities of the Florianópolis, Joinville and Joaçaba (Orlandim et al. 2016).

Although the aforementioned studies were carried out with different sampling efforts and prevailing the use of entomological nets, and in certain cases in restricted and smaller areas, such comparisons indicate that the environmental heterogeneity of the sampling areas favors the maintenance of the frugivorous butterfly diversity.

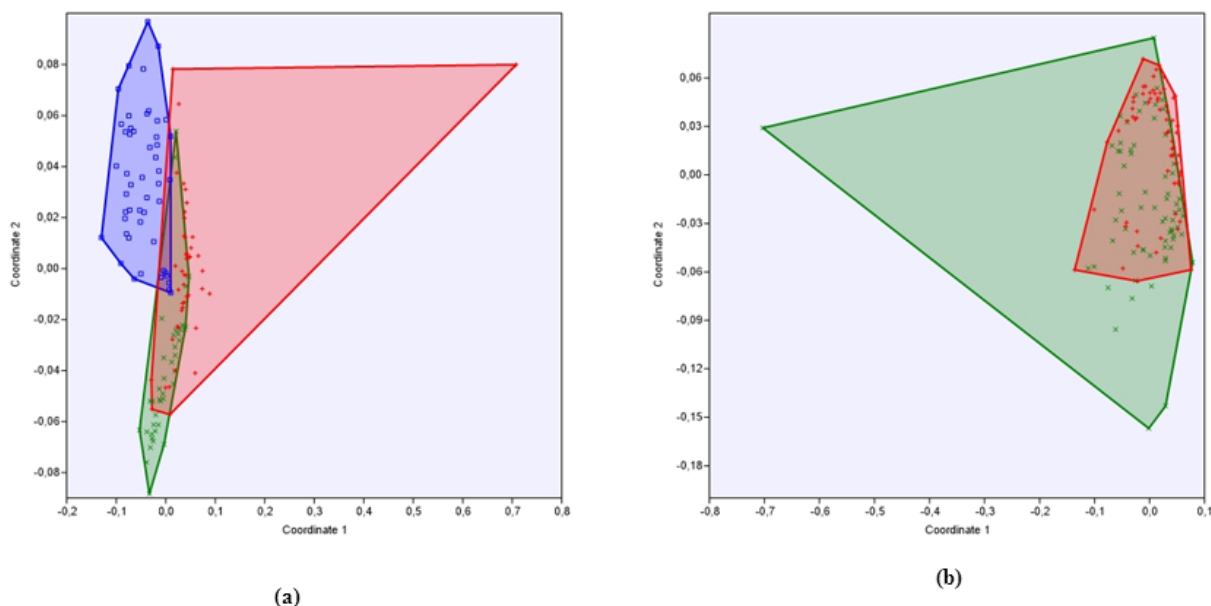
**Table 3.** Percentage of individuals of the subfamilies of frugivorous butterflies sampled with Van Someren-Rydon traps, recorded in Conservation Units and adjacent forest fragments in the western region of the state of Santa Catarina, Brazil, between December 2017 and March 2018. (F) FLONA; (ES) ESEC; (P) PAEAR; (T) Total butterflies in Conservation Units; (FF) FLONA Fragments; (FES) ESEC Fragments; (FP) PAEAR Fragments; (FT) Total butterflies in adjacent forest fragments.

Subfamilies	F	ES	P	T	FF	FES	FP	FT
Satyrinae	14.22	11.91	7.68	33.81	38.60	14.32	3.85	56.77
Biblidinae	1.09	0.85	0.21	2.15	1.25	1.37	0.42	3.04
Charaxinae	0.33	0.12	0.66	1.11	0.99	0.50	0.35	1.84
Nymphalinae	0.19	0.04	0.14	0.37	0.47	0.04	0.35	0.86

## Frugivorous butterflies in Southern Brazil



**Figure 3.** Richness estimates for rarefied and extrapolated sample for frugivorous butterflies sampled with Van Someren-Rydon traps in Conservation Units and adjacent forest fragments, western region of Santa Catarina, Brazil. 3a = FLONA; 3b = ESEC; 3c = PAEAR.



**Figure 4.** Ordination of butterflies faunal composition for Conservation Units (ESEC, FLONA, PAEAR), and for Conservation Units (CU) and adjacent forest fragments (FF) by Non-Metric MultiDimensional Scaling (nMDS), using qualitative similarity index in southern Brazil. (a) Conservation Units, ESEC, green; FLONA, blue; PAEAR, red. Simpson index. Stress = 0.439. (b) Environments, CU, red; and FF, green. Simpson index. Stress = 0.478.



**Table 4.** SIMPER analysis for ten butterflies species contributing more to dissimilarities between the Conservation Units (ESEC, FLONA, PAEAR) in Brazil (\*species percentage contribution to dissimilarity; # cumulative dissimilarity among three CU; † average species abundance in each CU).

Species	Contribution*	Cumulative % <sup>#</sup>	Mean abund. <sup>†</sup>	Mean abund. <sup>†</sup>	Mean abund. <sup>†</sup>
			ESEC	FLONA	PAEAR
<i>Hermeuptychia</i> sp.	15.89	15.89	8.25	12.2	1.11
<i>Manataria hercyna</i>	9.40	25.28	0.08	5.18	0.71
<i>Moneuptychia soter</i>	9.07	34.35	3.77	8.02	0.82
<i>Pareuptychia ocirrhoe</i>	8.82	43.17	0.4	4.88	0.2
<i>Ypthimoides ordinaria</i>	7.57	50.74	4.2	4.24	0.6
<i>Eryphanes reevesii</i>	5.82	56.56	2.13	0.72	1.44
<i>Forsterinaria quantius</i>	5.30	61.86	0.75	0.98	1.96
<i>Morpho epistrophus</i>	5.20	67.06	2.9	0	0
<i>Cissia eous</i>	3.82	70.87	1.05	2.94	0.64
<i>Biblis hyperia</i>	2.32	73.19	0.5	1.12	0.33
Overall average dissimilarity: 83.2					

**Table 5.** SIMPER analysis for ten butterflies species contributing more to dissimilarities between forestal fragments (FF) and Conservation Unit (CU) in Brazil (\*species percentage contribution to dissimilarity; # cumulative dissimilarity among FF and CU; † average species abundance in each).

Species	Contribution*	Cumulative % <sup>#</sup>	Mean abund. <sup>†</sup>	Mean abund. <sup>†</sup>
			FF	CU
<i>Hermeuptychia</i> sp.	15.54	15.54	9.39	5.09
<i>Moneuptychia soter</i>	9.27	24.81	7.01	1.51
<i>Manataria hercyna</i>	8.53	33.34	1.57	2.83
<i>Pareuptychia ocirrhoe</i>	7.69	41.03	1.69	2.32
<i>Ypthimoides ordinaria</i>	7.58	48.6	4.44	1.48
<i>Eryphanes reevesii</i>	6.65	55.26	0.51	2.31
<i>Forsterinaria quantius</i>	6.14	61.39	0.54	1.98
<i>Morpho epistrophus</i>	4.34	65.74	1.26	0.43
<i>Cissia eous</i>	4	69.73	2.6	0.55
<i>Biblis hyperia</i>	2.38	72.11	0.84	0.49
Overall average dissimilarity: 84.06				

In addition, surveys conducted using exclusively traps in fragments of Mixed Ombrophilous Forest in the state of Rio Grande do Sul, also disregarding differences in relation to sampling effort, identified a lower abundance of frugivorous butterflies compared to the present study [Graciotim & Morais (2016) with 31 species; Pedrotti et al. (2011) with 30 and Giovenardi et al. (2008) with 32].

Satyrinae concentrated the greatest richness and abundance of species in the 27 sampling units, a representativeness that was also found in other studies with butterflies in the state (Corso & Hernandez 2012, Carneiro et al. 2008, Schmidt et al. 2012, Piovesan et al. 2014, Fanton & Sabedot-Bordin 2014, Favretto et al. 2015, Silva & Sabedot-Bordin 2015 and Colpani & Sabedot-Bordin 2018). These results corroborate the observations of DeVries (1987) that the diversity of habitats in the neotropics would make the southern and southeastern regions of the Atlantic Forest of Brazil the largest in Satyrinae richness in the world, being considered the largest group within Nymphalidae (Lamas 2004).

The Neotropical Region is home to the greatest richness of satyrines of the world (D'abreria 1987). Satyrinae is a subfamily of wide diversity, biology and distribution, making up a third of all species of Nymphalidae

(Peña & Wahlberg 2008). Its main host plants are monocotyledons (DeVries 1987; Peña & Wahlberg 2008), abundant in clearings. The forest fragments sampled had trails and clearings, so it is believed that these spaces become conducive to the development and maintenance of this group, generating resources for both juveniles and adults.

Composing most of Satyrinae stands out the tribe Satyrini with more than 1,000 representatives among the almost 1,600 species of frugivorous butterflies in the Neotropical Region (Lamas 2004). In view of this representativeness, the high richness and abundance of Satyrini evidenced in the present study is not surprising. The high abundance of individuals obtained for the representatives of this tribe was associated with the areas that presented clearings. Bossart and Opuni-Frimpong (2009) point out that Satyrinae dynamics with grasses, which may become more numerous in environments with greater light penetration (for example, disturbed environments), can make this group an important biological indicator under the conditions of forest. According to Beccaloni et al. (2008), the host plants of Satyrini are composed mainly of grasses, host plants of caterpillars, thus the propagation of the tribe (Peña & Wahlberg 2008). In this way, high population densities could be expected for this group in the areas sampled.

An important Satyrinae species is *M. hercyna* (Hübner, 1821) because of its high abundance in FLONA (164 individuals). This species is considered rare in the northwest region of the state of Rio Grande do Sul (Biezanko 1960; Giovenardi et al. 2008) and Argentina (Nuñez-Bustos 2010). *Manataria hercyna* (Hübner, 1821) has crepuscular habitats, being found in dark and humid places (Nuñez-Bustos 2010) and is considered an indicator of an environment preserved in Atlantic Forest areas in southeastern Brazil (Brown Jr. & Freitas 2000). Other abundant Satyrinae in the sampled areas were *Hermeuptychia* sp., *M. soter* (Butler, 1877), *F. quantius* (Godart, 1824), *Y. ordinaria* (Freitas, Kaminski & Mielke, 2012) with generalist habitats (Brown Jr. 1992) and being commonly found (Morais et al. 2007; Nuñez-Bustos 2010). *Hermeuptychia* sp. is among the ten most abundant species in the state of Rio Grande do Sul (Morais et al. 2007).

Butterflies *Hermeuptychia* are widely distributed from the Southeastern United States to northern Argentina and present a large number of individuals and almost ubiquitous in most butterfly lists to sites in the Neotropics (Seraphim et al. 2014). All eight species recognized within *Hermeuptychia* are small and brown, with very similar interspecific species (Lamas 2004). Seraphim et al. (2014) indicates that external morphologies and intraspecific variable ocelli patterns make taxonomic identification based on difficult morphology. Part of this biodiversity is hidden in the form of cryptic species, which can be defined as a group of morphologically similar species usually identified under a single name (Bickford et al. 2007).

Although the characteristics of the surroundings of all the areas sampled were equivalent, it was verified in the sampling period the presence of extensive agricultural areas, being able to influence the composition of the fauna and favor the high dominance of some species of frugivorous butterflies. For Marin et al. (2009), the intensification of agriculture can affect the quality of the matrix and the persistence of species inhabiting the fragments of forest inserted in it, favoring the dominance of some species. Considering that lepidopterans are affected mainly by the impacts of agricultural activities (Bonebrake et al. 2010), for butterflies in particular, the way the matrix influences the fauna can vary between different species or assemblages, so in general, the disturbances favor the generalist species and negatively affect specialists (Littlewood et al. 2011).

The fact that more than 60% of the fauna sampled is composed of some species of frugivorous butterflies more abundant, characterizes the community with a high degree of dominance. Fragmentation and modification of environments tends to alter the natural balance in the diversity of different groups. Not all species respond in the same way and there may be different responses up to the level of specimens (Samways 2005). Nevertheless, what tend to happen is the decline of specialist species and the increase of populations of generalist species. Species more abundant in forest remnants could be considered resistant to fragmentation dynamics, while species that show a remarkable decrease in abundance could presumably suffer the negative effects in relation to fragmentation (Uehara-Prado et al. 2005).

The rarefaction and extrapolation curves for the CU and their forest fragments provide reliable responses, since there was a large sampling effort in the study. The estimated sampling coverage, above 97%, shows a good representation of the community of frugivorous butterflies in the region. For FLONA and PAEAR, it is observed that the richness approached an asymptote, indicating that a sample increase

would contribute with few additional species. According to Brown Jr. & Freitas (2000), in tropical environments, the curve rarely stabilizes.

The Jackknife 1 estimator shows that the richness of frugivorous butterflies in CU and forest fragments may be greater than the observed richness. Some studies show that richness of frugivorous species is higher in environments under stronger disturbances (Uehara-Prado et al. 2005), while others show that richness is lower in these environments and higher in more preserved environments (DeVries et al. 1997).

According to Bonebrake et al. (2010), butterfly communities are very variable between sites and between years, and are affected in the short term by differences in environmental/temporal conditions. Besides that, butterflies go out in search of host plants, food resources for adults, mating and overnight sites, reaching what is recognized as functional habitat (Marin et al. 2009). Therefore, it is important to establish relationships between the butterfly community and the vegetation structure, since both are closely related.

Studies emphasize how frugivorous butterflies are an excellent model for landscape characterization (Kremen 1992; Brown Jr. & Freitas 2000; Uehara et al. 2007), which corroborates, with the results of our study, where we can detect differences between the composition between different CU and between different environments.

Uehara et al. (2007) showed differences in the composition of butterfly species and in the distribution along different habitats in landscapes. Other studies have revealed differences in composition in response to habitat structural variables and associations of species of butterflies and their subfamilies to habitats with varying degrees of disturbance (Kremen 1992, Brown Jr. & Freitas 2000).

With the increasing reduction and modification of natural environments and the increasingly imminent threats to biodiversity, up-to-date studies on the ecology of species occupying a region, such as frugivorous butterflies, insects that can be very sensitive to environmental changes, are essential. Thus, the importance of surveys with a sampling methodology directed to the frugivorous butterflies is emphasized, allowing the adoption of actions contributing to the conservation of species.

The information generated through the present study contributes to the knowledge and characterization of the guild of frugivorous butterflies of the state. The fauna of frugivorous butterflies from the western region of the state of Santa Catarina, first investigated in areas of CU, has shown to be quite expressive and well represented in the Atlantic Forest Biome, indicating its potential as a refuge for biodiversity.

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## Author Contributions

Sandra Mara Sabedot Bordin: Contribution in data acquisition and identification of biological material; Contribution in the analysis and interpretation of data; Contribution in the preparation of the manuscript; Contribution in critical review, adding intellectual content.

Marcelo Monteiro: Contribution in data acquisition and identification of biological material.

Valéria Wesner Ferreira: Contribution in data acquisition and identification of biological material.

Junir Antonio Lutinski: Contribution in the analysis and interpretation of data; contribution in the critical review, adding intellectual content.

Everton Nei Lopes Rodrigues: Contribution in the analysis and interpretation of data; contribution in the critical review, adding intellectual content.

## Conflict of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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## Ichthyofauna of lotic environments in the Ivinhema river basin, upper Paraná river, Mato Grosso do Sul state, Brazil

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**Abstract:** In order to inventory the species richness and fish fauna composition along the Ivinhema river basin, 232 stretches of rivers and streams were sampled from 2000 to 2018, using a rectangular sieve, trawls, gillnets, cast nets, and electrofishing. A total of 141 species was caught in the basin, including seven orders and 35 families. For the recorded species, 42 are from the upper Paraná river, 51 from lower Paraná river, 27 from other basins, two from other continents, and the origin for 19 species are unknown/not reported. Six species are registered for the first time in the upper Paraná river basin: *Astyanax abramis*, *Moenkhausia oligolepis*, *Serrapinnus kriegi*, *Curimatopsis myersi*, *Pyxiloricaria menezesi*, and *Cichlasoma dimerus*. This is the first complete survey of the ichthyofauna in lotic environments along the whole Ivinhema river basin, containing relevant information for comparing studies and serving as reference data that can contribute to the management and implementation of politics for conservation of the basin. This survey also increases 24 species on the list of the most complete inventory in the upper Paraná river published in 2007.

**Keywords:** Fish communities, freshwater, survey.

## Ictiofauna de ambientes lóticos na bacia do rio Ivinhema, alto rio Paraná, Mato Grosso do Sul, Brasil

**Resumo:** Com o objetivo de inventariar a riqueza de espécies e a composição da ictiofauna ao longo da bacia do rio Ivinhema, 232 trechos de rios e riachos foram amostrados de 2000 a 2018, utilizando uma peneira retangular, rede de arrasto, rede de espera, tarrafas e pesca elétrica. Ao todo 141 espécies foram amostradas na bacia, incluindo oito ordens e 35 famílias. Para as espécies registradas, 42 são do alto rio Paraná, 51 do baixo rio Paraná, 27 de outras bacias, duas de outros continentes e para 19 espécies as origens são desconhecidas ou não reportadas. Seis espécies são registradas pela primeira vez na bacia alto rio Paraná: *Astyanax abramis*, *Moenkhausia oligolepis*, *Serrapinnus kriegi*, *Curimatopsis myersi*, *Pyxiloricaria menezesi* e *Cichlasoma dimerus*. Este é o primeiro levantamento da ictiofauna em ambientes lóticos para toda a bacia do rio Ivinhema, contendo informação relevante para estudos comparativos e servindo como dados de referências que podem contribuir para gerenciamento e implementação de políticas para conservação da bacia. Este levantamento também acrescenta 24 espécies na lista do inventário mais completo realizado no alto rio Paraná publicado em 2007.

**Palavras-chave:** Comunidades de peixes, água doce, inventário.

## Introduction

The upper Paraná river basin, with 367 fish species, being 317 described and 50 not formally described (Langeani et al. 2007), is located entirely within the territory of Brazil and extends from the municipalities of Guaira and Mundo Novo, upstream to the ancient waterfall known as Sete Quedas, which is currently submerged by the Itaipu Reservoir, and encompasses 891,000 km<sup>2</sup> or 10.5% of the area of Brazil (Agostinho et al. 2003). This portion of the Paraná river basin is the most studied regarding Brazilian freshwater fish (Agostinho et al. 2007), but for some regions the knowledge is still insufficient. Most of those studies were performed in the Brazilian Southeast (Paraná and São Paulo states) and despite of increasing researches, there have been only a few studies in the Midwest Brazilian (region where upper Paraná is inserted, including Mato Grosso do Sul and Goiás states and Federal District) (Aquino et al. 2009).

In the particular case of the Ivinhema river basin, a tributary practically free of impoundments and located in the western portion of upper Paraná river basin, some studies have been conducted to determine the environmental features that influence fish species in some few headwater streams (Súarez et al. 2007, Súarez & Lima-Junior, 2009), that influence spacial and temporal variation of fish in streams of lower Ivinhema (Súarez 2008a) or the fish richness in streams of the whole Ivinhema basin (Súarez et al. 2011). Fish eggs and larvae distribution related to environmental factors were also studied in the upper Ivinhema and its river tributaries (Nascimento & Nakatani 2006) and fishfauna associated to macrophyte mats were described in the lower Ivinhema (Bulla et al. 2011). In the latter, in Súarez (2008b) and in Súarez & Lima-Junior (2009) a small list of 28, 46 and 64 fish species is presented, respectively, and restricted to the lower Ivinhema river. The richness of 111 fish species was reported in Súarez et al. (2011), but no list of fish species was presented. In the current study, more streams and rivers stretches were sampled and then we described a larger fish richness and composition along the whole Ivinhema river basin, including streams and the main river tributaries.

Given that the upper Paraná river basin is located in a densely populated area that is also one of the most industrialized regions in Brazil (Thomaz et al. 2004), it is not surprising that this area has experienced continuous environmental alteration, that may have a substantial influence on fish communities and their habitats (Bryce et al. 1999, Holt et al. 2006). Therefore, studies describing fish community composition and occurrence in the western part of the upper Paraná river basin are needed, where lotic environments, free from impoundment, still exist. This is particularly important in the state of Mato Grosso do Sul, which has experienced over the last decade great expansion of sugar-cane cultivation and alcohol production (Domingues & Thomaz Junior 2012). Studies, like the present work, would be very important in obtain information useful in the minimization and mitigation of the impacts of anthropic alteration and facilitate management policies for conservation, specifically for the fish fauna.

It is in this context that the present study was conducted, using data from field sampling, with the aim of inventorying the richness and composition of fish species in the Ivinhema river basin.

## Material and Methods

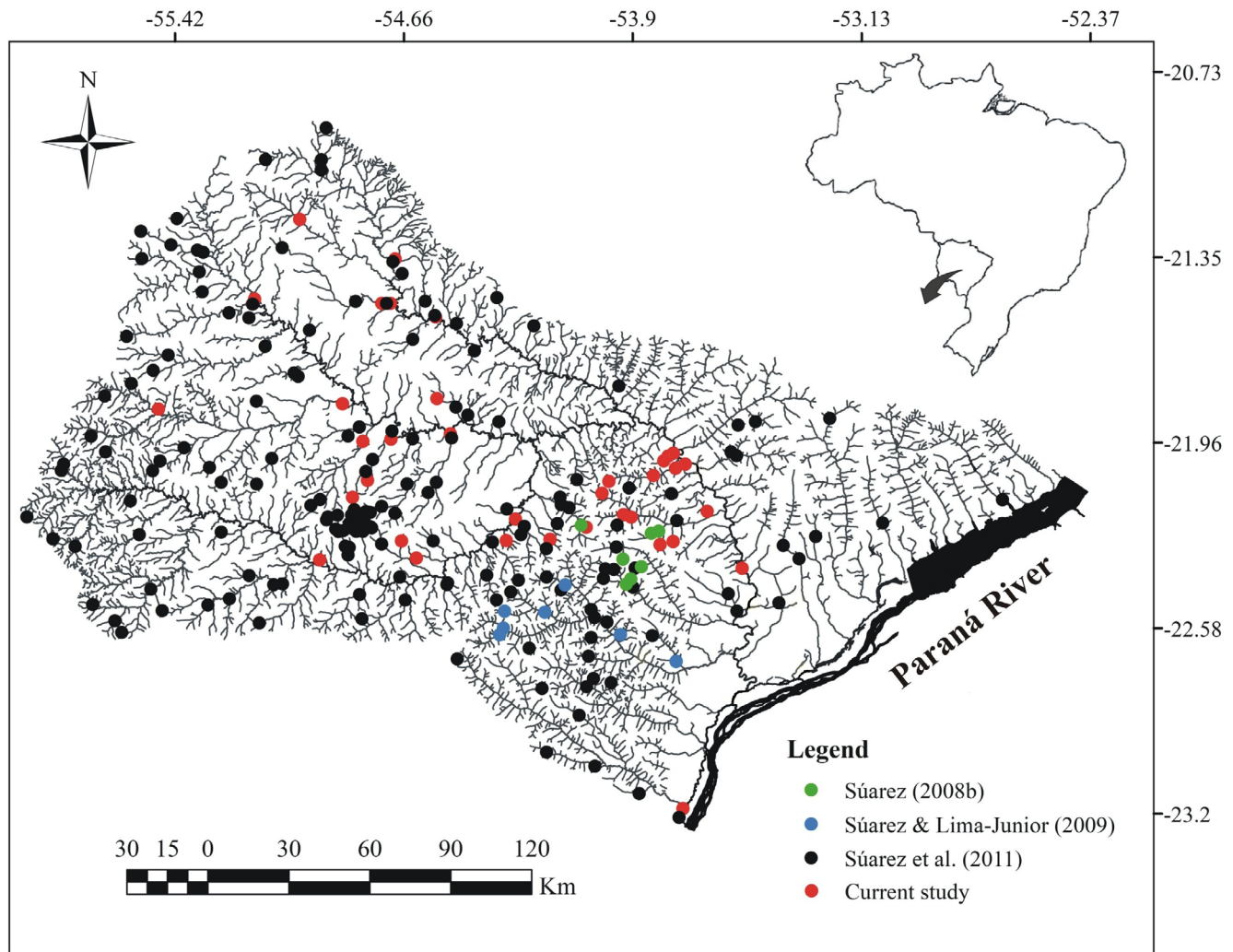
The Ivinhema river is the main tributary of the Paraná river in the western portion of the state of Mato Grosso do Sul. The mouth of this river is in the stretch of the Paraná river below Engenheiro Sérgio Motta dam but above Itaipu hydroelectric dam, which is the only stretch of the Paraná free of impoundments (Agostinho et al. 2000), including the Ivinhema river basin. The headwater is the Dourados river (area = 9,900 km<sup>2</sup>) at 700 m of altitude, with the contribution of other sub-basins including Santa Maria river (area = 2,700 km<sup>2</sup>), Brilhante river (area = 8,878 km<sup>2</sup>), Vacaria river (area = 6,500 km<sup>2</sup>), and the Ivinhema river (18,045 km<sup>2</sup>), forming the Ivinhema river basin, with a total area of 46,023 km<sup>2</sup>. Samplings were performed along 232 stretches of rivers and streams, throughout the Ivinhema river basin from January 2000 to June 2018, being that seven stretches were sampled in Súarez (2008b), seven in Súarez & Lima-Junior (2009), 186 in Súarez (2011) and 32 in the current study (Figure 1).

Fish were sampled from an approximately 100 m extension of each stream: 40 throws of a 1.2 x 0.8 m (2 mm mesh size) rectangular sieve; 10 trawlings (1.5 x 5 m; 2 mm mesh size); 12 hours of exposition to a sequence of 14 gillnets (10 x 1.5 m, with mesh sizes between adjacent knots: 1.5, 2.0, 3.0, 4.0, 5.0, 6.0, 7.0, 8.0, 9.0, 10.0 cm) and three cast nets (height 1.5 to 3 m, diameter 3 to 6 m, and mesh sizes between adjacent knots: 1.5, 3.0, 6.0 cm). Also, electrofishing was employed in small streams, with approximately 100-m-long hauls. In rivers, fish were also captured with a series of 14 gillnets and cast nets in each stretch, whereas along the banks of the rivers the rectangular sieve and trawls were used mainly to sample small sized fish.

Fish were anesthetized with Eugenol solution, euthanized and, in the sequence, fixed in 10% formalin and preserved in 70% ethanol. The euthanasia followed all the ethic criterion established by the Universidade Estadual de Mato Grosso do Sul, along the accomplishment of all the projects that originated the current data. The collections of specimens were authorized by the Brazilian Environment Institute (IBAMA), through special license number # 13458-115900-1, issued by SISBIO. Taxonomic identifications followed Graça & Pavanelli (2007) and current taxonomic reviews. Some specimens were deposited in zoological collections: Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura da Universidade Estadual de Maringá (NUP/UEM), Museu de Zoologia da Universidade de São Paulo (MZUSP), Museu da Universidade Federal do Rio Grande do Sul (UFRGS), Museu de Zoologia da Universidade Estadual de Londrina (MZUEL) and Coleção de Peixes da Universidade estadual de Mato Grosso do Sul (CPUEMS).

For those species that we were not able to present voucher specimens, we inserted the information of other references that had already reported the occurrence of the species in the Ivinhema river basin or upper Paraná river basin.

After the list was compiled, in order to confirm species names, possible synonyms and their basin of occurrence, we confronted it with information available on 'Check List of the Freshwater Fishes of South and Central America' (Reis et al. 2003), 'Catálogo das Espécies de Peixes de Água Doce do Brasil' (Buckup et al. 2007), 'Catalog of Fishes' (Eschmeyer et al. 2018) and 'Fishbase' (Froese & Pauly 2018).



**Figure 1.** Sampled stretches of rivers and streams that comprise the Ivinhema river basin, in the upper Paraná river basin (between January 2000 and June 2018), Mato Grosso do Sul State, Brazil.

The geographic origin for fish species followed Langeani et al. (2007) as autochthonous (native), natural species from the upper Paraná; allochthonous, species from other Neotropical regions and introduced in the upper Paraná; and exotic, species from other continents. For some species not reported in Langeani et al. (2007), the origin was defined according to reviews for the species or the remarks of Ota et al. (2018).

The fish fauna composition based in size was analyzed through the maximum size of each species (standard length for fish that has caudal fin, total length for fish that has only this information available and for Gymnotiformes or Synbranchiformes, and the total disc length for stingrays). This information were gathered from FishBase or directly measured on the specimen in the case of species is not described, not inserted in the database or the sampled specimen was longer than the reported in the FishBase. The size classes were obtained by Sturges formula ( $K = 1 + 3.32 \log n$ , where  $K$  = number of classes; and,  $n$  = the size sample), which furnishes the number of classes for the total sampling, based on the range of total fish size divided by the number of classes.

## Results

A total of 141 species was caught in this study. From this, eight species are without voucher numbers, but we inserted references that already reported these species in the Ivinhema river basin, and only one species we did not find any reference reporting its occurrence in the basin (Table 1). All species are distributed in 7 orders and 35 families. Characiformes and Siluriformes were the most representative orders with 65 (46.1%) and 50 species (35.5%), respectively, and Characidae and Loricariidae were the most abundant families with 28 (19.8%) and 17 species (12%), respectively. Although four species were identified at genus level, they were considered different species from those that are already described and reported for the upper Paraná basin.

After verification for synonymies and species that were redescribed, our study also contributed to increase the fish inventory furnished by Langeani et al. (2007), since 24 species were not reported on that list, and six of these species are the first reporting (species with asterisk) in the upper Paraná river: *Phalloceros harpagos*,



**Table 1.** Fish species caught between January 2000 and June 2018 in the Ivinhema river basin and its sub-basins (Santa Maria river – S; Dourados river – D; Vacaria river – V; Brilhante river – B; Ivinhema river – I), upper Paraná river basin. Voucher numbers, size (standard length, total length\*, or total disc length\*\* in centimeters) and species origin.

Species	Voucher or Reference	Size (cm)	sub-basin	origin
<b>MYLIOBATIFORMES</b>				
Potamotrygonidae				
<i>Potamotrygon falkneri</i> Castex & Maciel, 1963	Agostinho et al. 1997, Lonardon et al. 2006, Lacerda et al. 2008	78**	B,I	allochthonous
<i>Potamotrygon motoro</i> (Müller & Henle, 1841)	Agostinho et al. 1997, Lonardon et al. 2006, Lacerda et al. 2008	50**	B	allochthonous
<b>CHARACIFORMES</b>				
Acestrotrichidae				
<i>Acestrotrichus lacustris</i> (Lütken, 1875)	CPUEMS118	27	S,D,V,B,I	autochthonous*
Anostomidae				
<i>Leporellus vittatus</i> (Valenciennes, 1850)	#	30	S,V	autochthonous
<i>Leporinus amblyrhynchus</i> Garavento & Britski, 1987	CPUEMS18	27.4	I	autochthonous
<i>Leporinus friderici</i> (Bloch, 1794)	NUP9375	40	S,D,V,B,I	autochthonous*
<i>Leporinus lacustris</i> Campos, 1945	NUP9387	20.3	D,B,I	autochthonous
<i>Leporinus octofasciatus</i> Steindachner, 1915	CPUEMS424	31.2	S,I	autochthonous*
<i>Leporinus striatus</i> Kner, 1859	CPUEMS290	25	S,D,V,B	autochthonous
<i>Megaleporinus macrocephalus</i> (Garavento & Britski, 1988)*	CPUEMS754	60	S,V,B,I	allochthonous
<i>Megaleporinus piavussu</i> Britski, Birindelli & Garavento, 2012*	NUP9371	40	D,I	autochthonous*
<i>Megaleporinus obtusidens</i> (Valenciennes, 1837)*	CPUEMS301	39.8	I	autochthonous*
<i>Schizodon borellii</i> (Boulenger, 1900)	CPUEMS705	30	S,D,V,I	autochthonous*
Bryconidae				
<i>Salminus brasiliensis</i> (Cuvier, 1816)	CPUEMS707	100	S,D,V,B,I	autochthonous*
<i>Salminus hilarii</i> Valenciennes, 1850	NUP9374	50	S,D,V,B,I	autochthonous*
Characidae				
<i>Aphyocharax anisitsi</i> Eigenmann & Kennedy, 1903	CPUEMS261	5.5	I	allochthonous
<i>Aphyocharax dentatus</i> Eigenmann & Kennedy 1903	NUP9944	7.2	S,V,I	autochthonous*
<i>Aphyocharax</i> sp.	NUP9361	5.3	S,D,V,B,I	unknown
<i>Astyanax abramis</i> (Jenyns, 1842)	MZUEL14444	14	I	autochthonous
<i>Astyanax biotae</i> Castro & Vari, 2004	CPUEMS537	5.2	---	autochthonous
<i>Astyanax bockmanni</i> Vari & Castro, 2007	Suárez 2008b	6.7	S,D,I	autochthonous*
<i>Astyanax fasciatus</i> (Cuvier, 1819)	CPUEMS116	12	S,D,V,B,I	autochthonous*
<i>Astyanax lacustris</i> (Lütken, 1875)*	NUP9378	10.6	S,D,V,B,I	autochthonous
<i>Astyanax paranae</i> Eigenmann, 1914	CPUEMS523	11.3	D,V,B,I	autochthonous
<i>Galeocharax knerii</i> (Steindachner, 1879)	CPUEMS280	33.1	D,V,B,I	autochthonous
<i>Hyphessobrycon anisitsi</i> (Eigenmann, 1907)	CPUEMS170	6	D,B,I	autochthonous*
<i>Hyphessobrycon eques</i> (Steindachner, 1882)	NUP9314	4	S,D,I	autochthonous*
<i>Knodus moenkhausii</i> (Eigenmann & Kennedy, 1903)	CPUEMS522	4.8	---	allochthonous
<i>Moenkhausia bonita</i> Benine, Castro & Sabino, 2004	NUP9317	4.4	D,I	unknown
<i>Moenkhausia forestii</i> Benine, Mariguela & Oliveira, 2009	NUP9318	3.8	S,D,V,B,I	autochthonous*
<i>Moenkhausia intermedia</i> Eigenmann, 1908	CPUEMS232	8	S,D,B,I	autochthonous*
<i>Moenkhausia oligolepis</i> (Günther, 1864) Glass tetra	CPUEMS531	10	---	unknown
<i>Moenkhausia sanctaefilomenae</i> (Steindachner, 1907)	NUP9936	7	S,D,V,B,I	autochthonous*

Continuation Table 1.

Species	Voucher or Reference	Size (cm)	sub-basin	origin
<i>Oligosarcus pinto</i> Campos, 1945	NUP9315	8.4	S,D,V,B,I	autochthonous
<i>Piabarchus stramineus</i> Eigenmann, 1908*	CPUEMS119	5.6	S,D,V,B,I	autochthonous
<i>Piabina argentea</i> Reinhardt, 1867	NUP9323	7.3	S,D,V,B,I	autochthonous*
<i>Psellogrammus kennedyi</i> (Eigenmann, 1903)	NUP9321	5.9	S,D,V,B,I	autochthonous*
<i>Roeboides descavadensis</i> Fowler, 1932	NUP9363	8.9	S,D,V,B,I	allochthonous
<i>Serrapinnus calliurus</i> (Boulenger, 1900)	MZUEL14483	4.4	I	autochthonous*
<i>Serrapinnus heterodon</i> (Eigenmann, 1915)	CPUEMS208	4.1	V,B	autochthonous
<i>Serrapinnus kriegi</i> (Schindler, 1937)	MZUEL14348	2.4	I	unknown
<i>Serrapinnus notomelas</i> (Eigenmann, 1915)	NUP9937	3.6	S,D,V,B,I	autochthonous
<i>Serrapinnus</i> sp.	CPUEMS16	3.2	D,V,B,I	unknown
Crenuchidae				
<i>Characidium gomesi</i> Travassos, 1956	CPUEMS248	6.5	D,V,B,I	autochthonous
<i>Characidium lagosantense</i> Travassos, 1947	CPUEMS257	4.1	I	unknown
<i>Characidium laterale</i> (Boulenger, 1845)	MZUEL14344	3.5	I	autochthonous
<i>Characidium zebra</i> Eigenmann, 1909	NUP9353	7.4	S,D,V,B,I	autochthonous*
Curimatidae				
<i>Curimatopsis myersi</i> Vari, 1982	MZUEL14516	4.4	I	unknown
<i>Cyphocharax modestus</i> (Fernández-Yépez, 1948)	NUP9327	16.2	S,D,V,B,I	autochthonous
<i>Cyphocharax vanderi</i> (Britski, 1980)	MZUEL14496	6.8	I	autochthonous
<i>Steindachnerina brevipinna</i> (Eigenmann & Eigenmann, 1889)	NUP9365	10.9	S,D,V,B,I	allochthonous
<i>Steindachnerina insculpta</i> (Fernández-Yépez, 1948)	NUP9358	10.6	D,V,B,I	autochthonous
Cynodontidae				
<i>Rhaphiodon vulpinus</i> Spix & Agassiz, 1829	Agostinho et al. 1997, Reynalte-Tataje et al. 2011	80	B,I	autochthonous*
Erythrinidae				
<i>Erythrinus erythrinus</i> (Bloch & Schneider, 1801)	CPUEMS542	20	I	allochthonous
<i>Hoplerythrinus unitaeniatus</i> (Agassiz, 1829)	NUP9376	25	D,V,B,I	allochthonous
<i>Hoplias misionera</i> Rosso, Mabragaña, González-Castro Delpiani, Avigliano, Schenone, Días de Astarloa, 2016*	MUZEL14495	49	S,D,V,B,I	autochthonous*
Hemiodontidae				
<i>Hemiodus orthonops</i> Eigenmann & Kennedy, 1903	CPUEMS275	25	S,I	allochthonous
Lebiasinidae				
<i>Pyrrhulina australis</i> Eigenmann & Kennedy, 1903	NUP9356	5	S,D,V,B,I	autochthonous*
Parodontidae				
<i>Apareiodon affinis</i> (Steindachner, 1879)	Agostinho et al. 1997	17	D	autochthonous*
<i>Apareiodon ibitiensis</i> Campos, 1944	MZUSP85418	11.3	S,D,B,I	autochthonous
<i>Apareiodon piracicabae</i> (Eigenmann, 1907)	NUP9934	12	S,D	autochthonous
<i>Parodon nasus</i> Kner, 1859	CPUEMS138	12.7	S,D,V,B,I	autochthonous*
Prochilodontidae				
<i>Prochilodus lineatus</i> (Valenciennes, 1836)	CPUEMS595	59.2	S,D,V,B,I	autochthonous*
Serrasalminidae				
<i>Metynnis lippincottianus</i> (Cope, 1870)	NUP9380	14.9	I	unknown
<i>Myloplus tiete</i> (Eigenmann & Norris, 1900)*	NUP9364	16.3	I	autochthonous
<i>Serrasalmus maculatus</i> Kner, 1858	CPUEMS521	20.2	S,D,V,B,I	autochthonous*
<i>Serrasalmus marginatus</i> Valenciennes, 1837	CPUEMS708	27	S,D,B,I	autochthonous

Continuation Table 1.

Species	Voucher or Reference	Size (cm)	sub-basin	origin
<b>GYMNOTIFORMES</b>				
<b>Apteronotidae</b>				
<i>Apteronotus albifrons</i> (Linnaeus, 1766)	NUP9386	50*	D,I	allochthonous
<b>Gymnotidae</b>				
<i>Gymnotus inaequilabiatus</i> (Valenciennes, 1839)	CPUEMS697	60*	D,V,I	allochthonous
<i>Gymnotus omarorum</i> Richer-de-Forges, Crampton & Albert, 2009	MUZEL14328	25.4*	I	unknown
<i>Gymnotus pantanal</i> Fernandes, Albert, Daniel-Silva, Lopes, Crampton & Almeida-Toledo, 2005	MUZEL14486	25.1*	I	autochthonous*
<i>Gymnotus paraguensis</i> Albert & Crampton, 2003	CPUEMS179	24*	D,I	allochthonous
<i>Gymnotus sylvius</i> Albert & Fernandes-Matioli, 1999	NUP9389	36*	S,D,V,B,I	autochthonous*
<b>Hypopomidae</b>				
<i>Brachyhypopomus gauderio</i> Giora & Malabarba, 2009*	NUP9390	18.6*	B,I	allochthonous
<b>Rhamphichthyidae</b>				
<i>Gymnorhamphichthys britskii</i> Carvalho, Ramos & Albert, 2011*	NUP9384	11.5	I	autochthonous*
<b>Sternopygidae</b>				
<i>Eigenmannia trilineata</i> López & Castello, 1966	NUP9381	25*	S,D,B,I	autochthonous*
<i>Eigenmannia virescens</i> (Valenciennes, 1847)	CPUEMS284	35.8*	S,B,I	autochthonous*
<i>Sternopygus macrurus</i> (Bloch & Schneider, 1801)	NUP9388	141*	S,D,B,I	autochthonous*
<b>SILURIFORMES</b>				
<b>Aspredinidae</b>				
<i>Bunocephalus larai</i> Ihering, 1930	NUP9352	5.6	S,B,I	autochthonous
<b>Auchenipteridae</b>				
<i>Auchenipterus osteomystax</i> (Miranda Ribeiro, 1918)	CPUEMS709	27	B,I	autochthonous*
<i>Tatia neivai</i> (Ihering, 1930)	NUP9313	8.2	D,B,I	autochthonous
<i>Trachelyopterus coriaceus</i> Valenciennes, 1840	MZUEL14503	18*	I	unknown
<i>Trachelyopterus galeatus</i> (Linnaeus, 1766)	NUP9377	22	D,V,B,I	autochthonous*
<b>Callichthyidae</b>				
<i>Callichthys callichthys</i> (Linnaeus, 1758)	NUP9359	17	V,B,I	autochthonous*
<i>Corydoras aeneus</i> (Gill, 1858)	NUP9362	7.5	S,D,V,B,I	autochthonous*
<i>Hoplosternum littorale</i> (Hancock, 1828)	NUP9368	15.8	B,I	autochthonous*
<i>Leptoplosternum pectorale</i> (Boulenger, 1895)	MUZEL14322	6	I	unknown
<b>Cetopsidae</b>				
<i>Cetopsis gobioides</i> Kner, 1858	NUP9367	10.9	B,I	autochthonous*
<b>Clariidae</b>				
<i>Clarias gariepinus</i> (Burchell, 1822)	CPUEMS594	32.9	D,V	exotic
<b>Doradidae</b>				
<i>Ossancora eigenmanni</i> (Boulenger, 1895)	CPUEMS704	15.2	S,I	allochthonous
<i>Platydoras armatulus</i> (Valenciennes, 1840)	Agostinho et al. 1997	43	B	unknown
<i>Pterodoras granulosus</i> (Valenciennes, 1821)	CPUEMS273	60	B,I	allochthonous
<i>Trachydoras paraguayensis</i> (Eigenmann & Ward, 1907)	CPUEMS224	19.2	D,B,I	allochthonous
<b>Heptapteridae</b>				
<i>Cetopsorhamdia iheringi</i> Schubart & Gomes, 1959	NUP9348	11.1	D,V,I	autochthonous
<i>Imparfinis borodini</i> Mees & Cala, 1989	CPUEMS701	15.7	B	autochthonous
<i>Imparfinis mirini</i> Haseman, 1911	NUP9355	8.5	D,B,I	autochthonous
<i>Imparfinis schubarti</i> (Gomes, 1956)	NUP9348	10.6	D,V,I	autochthonous

Continuation Table 1.

Species	Voucher or Reference	Size (cm)	sub-basin	origin
<i>Phenacorhamdia tenebrosa</i> (Schubart, 1964)	MZUSP85419	6.7	S,D,V,B,I	autochthonous
<i>Pimelodella avanhandavae</i> Eigenmann, 1917	NU9372	12.5	S,D,V,B,I	autochthonous
<i>Pimelodella gracilis</i> (Valenciennes, 1835)	NUP9346	18	S,D,V,B,I	autochthonous*
<i>Pimelodella taenioptera</i> Miranda-Ribeiro, 1914	CPUEMS820	12.1	D,V,B	allochthonous
<i>Rhamdia quelen</i> (Quoy & Gaimard, 1824)	MZUEL14499	38.7	D,V,B,I	autochthonous*
Loricariidae				
<i>Ancistrus</i> sp.	NUP9328	11	V,B	unknown
<i>Curculionichthys insperatus</i> Britski & Garavello, 2003*	MZUSP85735	3	S,D,V,B,I	autochthonous
<i>Farlowella hahni</i> Meinken, 1937*	NUP9396	22.5	D,B,I	autochthonous*
<i>Hisonotus francirochai</i> (Ihering, 1928)	MUZEL14355	3.6	I	autochthonous
<i>Hypostomus albopunctatus</i> (Regan, 1908)	CPUEMS268	40	S,V,B	autochthonous
<i>Hypostomus ancistroides</i> (Ihering, 1911)	MUZEL14450	21	S,D,V,B,I	autochthonous
<i>Hypostomus cochliodon</i> Kner, 1854	CPUEMS228	23	S,D,V,B,I	allochthonous
<i>Hypostomus iheringii</i> (Regan, 1908)	NUP9354	11.6	S,D,V,B,I	autochthonous
<i>Hypostomus regani</i> (Ihering, 1905)	CPUEMS712	41	S,D,I	autochthonous
<i>Hypostomus strigaticeps</i> (Regan, 1908)	NUP9340	16	S,D,V,B,I	autochthonous
<i>Loricariichthys platymetopon</i> Isbrücker & Nijssen, 1979	NUPP9338	30	S,D,V,B,I	allochthonous
<i>Otothyropsis marapoama</i> Ribeiro, Carvalho & Melo, 2005	NUP9395	3.6	S,D,I	autochthonous
<i>Otothyropsis polyodon</i> Caleagri, Lehmann A. & Reis, 2013	NUP9392	4.2	S,D,V,B,I	unknown
<i>Pterygoplichthys ambrosetti</i> Eigenmann & Kennedy, 1903	CPUEMS229	39.5	B,I	autochthonous*
<i>Pyxiloricaria menezesi</i> Isbrücker & Nijssen, 1984	NUP9382	14	S,D,V,B,I	unknown
<i>Rineloricaria lanceolata</i> (Günther, 1868)	NUP9336	9.5	S,D,B,I	unknown
<i>Rineloricaria</i> sp.	NUP9342	8.6	S,D,I	unknown
Pimelodidae				
<i>Hemisorubim platyrhynchos</i> (Valenciennes, 1840)	CPUEMS272	52.5	D,V,B,I	autochthonous*
<i>Pimelodus maculatus</i> La Cépède, 1803	CPUEMS281	36	S,D,B,I	autochthonous*
<i>Pimelodus ornatus</i> Kner, 1858	CPUEMS235	38.5	D	allochthonous
<i>Pseudoplatystoma corruscans</i> (Spix & Agassiz, 1829)	Agostinho et al. 1997, Suárez & Lima-Junior 2009, Reynalte-Tataje et al. 2011, Vaini et al. 2016	114	D,V,B,I	autochthonous*
<i>Sorubim lima</i> (Bloch & Schneider, 1801)	CPUEMS713	50.5	B,I	allochthonous
Pseudopimelodidae				
<i>Pseudopimelodus mangurus</i> (Valenciennes, 1835)	Suárez 2008b, Suárez & Lima-Junior 2009	34.5	I	autochthonous*
Scoloplacidae				
<i>Scoloplax empousa</i> Schaefer, Weitzman & Britski, 1989	NUP9347	2	I	unknown
Trichomycteridae				
<i>Paravandellia oxyptera</i> Miranda Ribeiro, 1912	CPUEMS66	2.8	D	autochthonous*
<i>Trichomycterus davisii</i> (Haseman, 1911)*	UFRGS11158	8	D	autochthonous
Cynolebiidae				
<i>Melanorivulus ivinhemensis</i> Volcan, Severo-Neto & Lanes, 2018*	NUP9350	3	D,I	autochthonous
CYPRINODONTIFORMES				
Poeciliidae				
<i>Phalloceros harpagos</i> Lucinda, 2008	NUP9360	3.4	D,B,I	autochthonous*
<i>Poecillia reticulata</i> Peters, 1859	NUP9379	5	D,V,B	allochthonous



Continuation Table 1.

Species	Voucher or Reference	Size (cm)	sub-basin	origin
SYNBRANCHIFORMES				
Synbranchidae				
<i>Synbranchus marmoratus</i> Bloch, 1795	NUP9357	150*	S,D,V,B,I	autochthonous*
CICHLIFORMES				
Cichlidae				
<i>Apistogramma commbrae</i> (Regan, 1906)	CPUEMS133	3.3	I	allochthonous
<i>Astronotus crassipinnis</i> Heckel, 1840	CPUEMS541	24	V,B	allochthonous
<i>Chaetobranchopsis australis</i> Eigenmann & Ward, 1907	NUP9366	12	I	allochthonous
<i>Cichlasoma dimerus</i> (Heckel, 1840)	MZUEL14512	11.7	I	unknown
<i>Cichlasoma paranaense</i> Kullander, 1983	NUP9385	7.8	S,D,V,B,I	autochthonous
<i>Coptodon rendalli</i> (Boulenger, 1897)*	NUP9316	16.6	D,B,I	exotic
<i>Crenicichla britskii</i> Kullander, 1982	MZUSP85416	14.5	S,D,V,B,I	autochthonous
<i>Gymnogeophagus setequedas</i> Reis; Malabarba & Pavanelli, 1992	CPUEMS598	9.8	D	autochthonous*
<i>Laetacara araguaiae</i> Ottoni & Costa 2009*	MUZEL14324	6	I	allochthonous

<sup>1</sup> Species name with asterisk indicates that the deposit was made with the synonym species and it changed according to the new identification.

<sup>2</sup> Acronym refers to the following collections: Universidade Estadual de Maringá (NUP), Coleção de Peixes - Universidade Estadual Paulista/Campus de São José do Rio Preto (DZSJRP), Museu de Zoologia da Universidade de São Paulo (MZUSP), Museu de Ciências e Tecnologia da PUCRS (MCP), Universidade Federal do Rio Grande do Sul (UFRGS) and Museu de Zoologia da Universidade Estadual de Londrina (MZUEL), Coleção de Peixes da Universidade estadual de Mato Grosso do Sul (CPUEMS).

<sup>3</sup> Autochthonous: species from upper Paraná, asterisk indicates the type locality outside upper Paraná; Allochthonous: species from other Neotropical basins; Exotic: species from other continents and Unknown species no information about on the origin.

<sup>4</sup> Voucher number indicates species deposited by this work; References were used when it was not possible to deposit voucher specimens, but the species was already reported in the Ivinhema river basin; hastag, the only species without deposited specimens in the Ivinhema river basin, but reported by Langeani et al. (2007) in the upper Paraná river basin.

SÚAREZ, Y.R. & LIMA-JÚNIOR, S.E. 2009 Spatial and temporal variation in stream fish assemblages of Guirai River Basin, Upper Paraná Basin. *Biota Neotrop.* 9(1):101-111 <http://www.biotaneotropica.org.br/v9n1/en/abstract?article+bn01709012009> (ultimo acesso em 21/01/2019)

VAINI, J.O., CRISPIM, B.A., SILVA, D.B.S., BENITES, C., RUSSO, M. R. & GRISOLIA, A. B. 2016. Genetic variability of pure *Pseudoplatystoma corruscans* and *Pseudoplatystoma reticulatum* individuals in the Paraná and Paraguay River basins. *Fish. Sci.* 82:605-611.

*Moenkhausia forestii*, *Brachyhypopomus gauderio*, *Laetacara araguaiae*, *Gymnotus omarorum*, *Gymnorhamphichthys britskii*, *Otothyropsis polyodon*, *Melanorivulus ivinhemensis*, *Astyanax abramis*\*, *Moenkhausia oligolepis*\*, *Serrapinnus kriegi*\*, *Curimatopsis myersi*\*, *Pyxiloricaria menezesi*\*, *Cichlasoma dimerus*\*, *Characidium lagosantense*, *Rineloricaria lanceolata*, *Moenkhausia bonita*, *Metynnis lippincottianus*, *Farlowella hahni*, *Trichomycterus davisi*, *Apistogramma commbrae*, *Chaetobranchopsis australis*, *Psellogrammus kennedyi*, and *Serrapinnus calliurus*. Some species may be visualized in Figure 2.

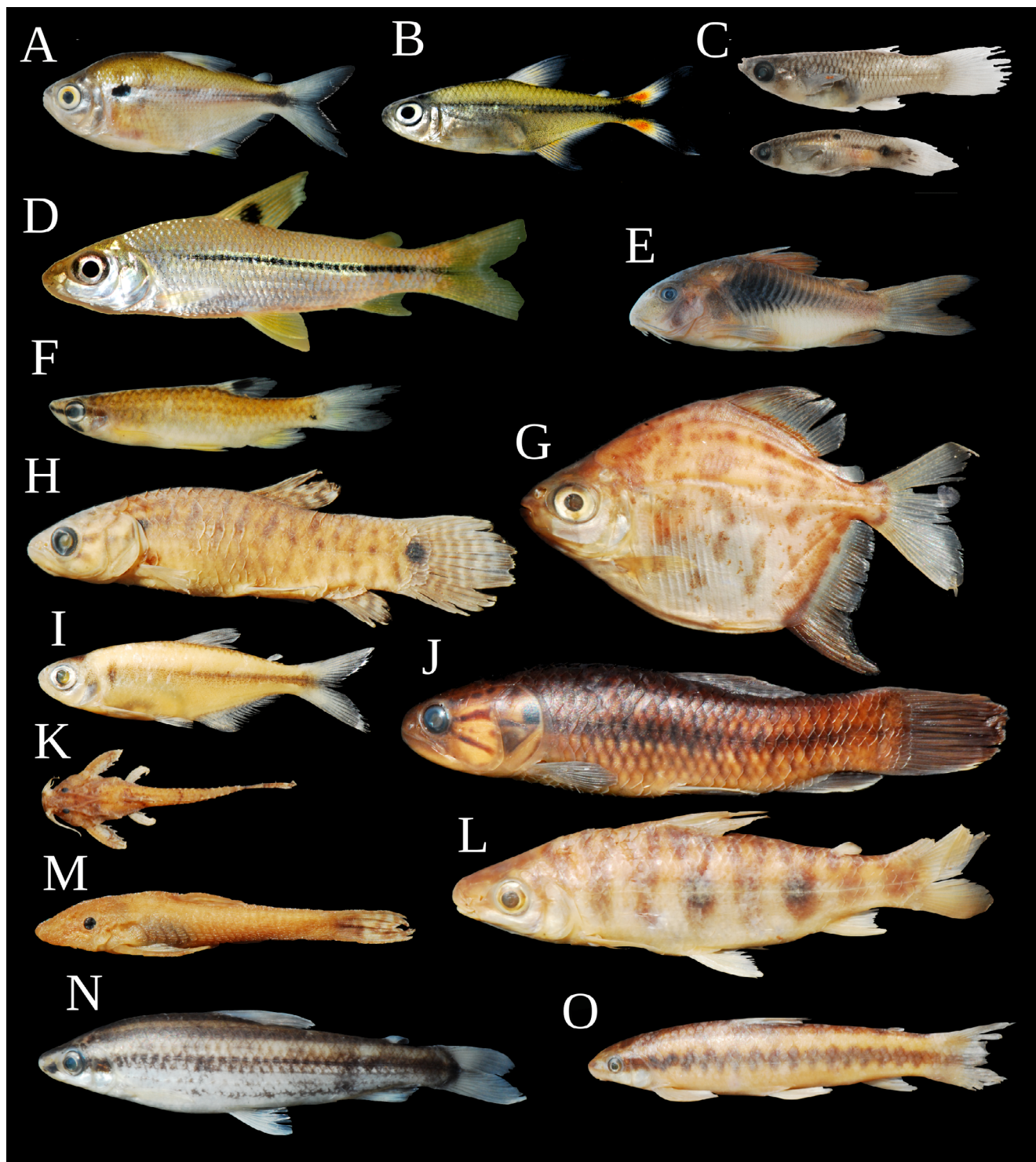
Regarding to geographic origin, 93 species were autochthonous from the Paraná river (42 from upper Paraná and 51 from lower Paraná), 27 were allochthonous and two were exotics (Table 1). It was not possible to define the origin of 19 species, mainly because nothing is reported about them or because the species are yet to be described.

The length of fish species varied from two to 150 cm. Small-sized fish were mostly common (63.8%, 90 species) in size class until 20.7 cm (Figure 3), fish of medium size were in the second size class 39.4 cm (19.9%, 28 species) and the other 16 species (16.3%) were considered large size, above 39.5 cm and until 150 cm.

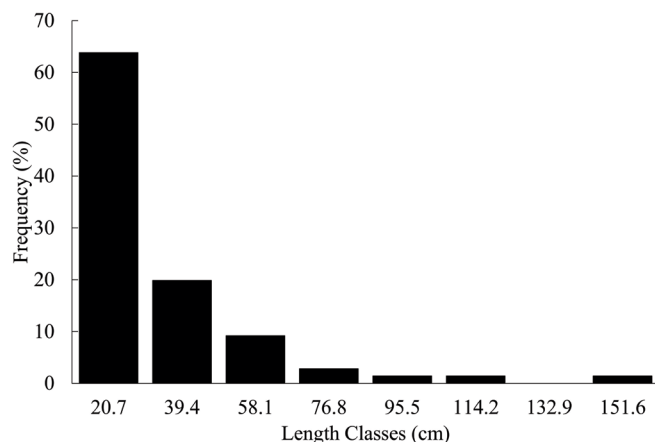
## Discussion

For the upper Paraná river basin it was reported 317 described fish species by Langeani et al. (2007). Although, since it was published this list may be increased, in function of the new descriptions, our study has 117 identified species (35.6%) in common, it can be said that the Ivinhema river basin, covering only 5% of the total area of the upper Paraná, possesses an expressive fish fauna, one third of the species.

From 24 species not reported on the inventory of Langeani et al. (2007), eight were described after the publication: *P. harpagos*, *M. forestii*, *B. gauderio*, *L. araguaiae*, *G. omarorum*, *G. britskii*, *O. polyodon*, and *Melanorivulus ivinhemensis* (Lucinda 2008, Benine et al. 2009, Giora & Malabarba 2009, Ottoni & Costa 2009, Richerde-Forges et al. 2009, Carvalho et al. 2011, and Calegari et al. 2013, Volcan et al. 2018, respectively). *Phallocerus harpagos*, *M. forestii*, *G. britskii*, *O. polyodon*, and *M. ivinhemensis* were already described with specimens sampled in the upper Paraná river basin. It is noteworthy that *M. ivinhemensis* was described recently and is considered endemic in marginal areas of small first order streams and wetlands in the Ivinhema river (Ota et al. 2018). *Brachyhypopomus gauderio* was described from



**Figure 2.** Representative specimens for some fish species reported in the Ivinhema river basin, in the upper Paraná river basin, Mato Grosso do Sul State, Brazil. A) *Astyanas lacustris*; B) *Hemigrammus marginatus*; C) (Female and male) *Poecilia reticulata*; D) *Steindachnerina brevipinna*; E) *Corydoras aeneus*; F) *Pyrrhulina australis*; G) *Myloplus tiete*; H) *Erythrinus erythrinus*; I) *Knodus moenkhausii*; J) *Hoplerethrinus unitaeniatus*; K) *Scoloplax empousa*; L) *Leporinus octofasciatus*; M) *Otothyropsis polyodon*; N) *Leporinus striatus*; O) *Paradon nasus*.



**Figure 3.** Frequency of fish by size classes along the Ivinhema river basin, upper Paraná river basin, Mato Grosso do Sul State, Brazil.

Paraguay and Uruguay basins and Brazilian southeastern coast (Giora & Malabarba 2009), but according to Ota et al. (2018) this species was identified as *B. cf. pinnicaudatus* (Graça & Pavanelli 2007) and its occurrence in the upper Paraná might be associated to introduction as a live bait by anglers. *Laetacara araguaiaiae* was described from the Verde and Araguaia rivers (Otoni & Costa 2009), but as a non-native species, it was reported the upper Paraná floodplain, probably introduced by the aquarium trade (Ota et al. 2018), and now we expanded its occurrence to the river Ivinhema.

For six species, the current study had extended their occurrence to the upper Paraná river: *Astyanax bimaculatus* subgroup was revised by Lucena & Soares (2016) and they re-described *A. abramis* as a valid species from Paraguay, Lower Paraná and La Plata river basin. *Moenkhausia oligolepis* was reported originally only in the Guianas and Amazon river basins, but Benine et al (2009) with a detailed revision of this species reported its occurrence in Paraguay river. For *S. kriegi*, *C. myersi*, *P. menezesi* and *C. dimerus* and the other above-mentioned species, there was not found any report for these six species in the upper Paraná, thus these are the first reports.

Ten species not reported in Langeani et al. (2007) had their taxonomic status revised or occurrence extended. *Characidium lagosantense* was re-described and reported in the Mogi-Guaçu river by Silveira (2008). The taxonomic revision of *Rineloricaria* species was carried out in the Paraguay river (Vera-Alcaráz et al. 2012), but *R. lanceolata* occurrence was also recorded in the Ivinhema river in 2011 and reported by Froehlich et al. (2017). *Moenkhausia bonita* was described in 2004 in the Paraguay river (Benine et al. 2004), but its occurrence in the Ivinhema river was registered in 2006 and reported by Froehlich et al. (2017) and Ota et al. (2018). *Metynnis lippincottianus* was reported in the Ivinhema river by Suárez (2011) and Froehlich et al. (2017). It is interesting to point out that *M. lippincottianus* was not reported by Langeani et al. (2007) and it was adverted that it had been identified in the upper Paraná as *M. maculatus* (Graça & Pavanelli 2007). However, as an original Amazonian fish, two populations of this species were reported in two tributary streams of Ilha Solteira dam, suggesting these populations were installed locally (Froehlich et al. 2017) and now our study reported this species in many streams in the Ivinhema river. Its occurrence can be associated with the releasing of specimens for restocking or with the aquarium trade (Ota 2015).

*Farlowella hahni*, despite not being reported by Langeani et al. (2007), it was reported in the upper Paraná by Graça & Pavanelli (2007) as *F. amazonum*, but Ota et al. (2018) comparing the characters, described for the genus by Retzer & Page (1996), decided that the species would match more with the description of *F. hahni*. This species was reported before in the lower Paraná river and its occurrence in the upper portion can be associated to the inundation of the Sete Quedas Falls after the filling of Itaipu Reservoir (Ota et al. 2018). *Trichomycterus davisii*, first identified as *Trichomycterus* sp. (Graça & Pavanelli 2007), has now been assigned as *T. davisii* belonging to a species complex (Ota et al. 2018). *Apistogramma commbrae* and *C. australis* may have been introduced accidentally by aquarists or even after the construction of the Itaipu dam, since these species originally occurred only in the lower Paraná river and were firstly reported in the upper Paraná (Ivinhema river) by Suárez et al. (2011). *Psellogrammus kennedyi*, also reported for this latest author, and *S. calliurus* are considered a not-native species in the upper Paraná and had their occurrence associated with the function of a fish ladder that connects downstream portion (lower Paraná) to upstream portion (upper Paraná) in the Itaipu Dam (Ota et al. 2018).

Whereas Langeani et al. (2007) estimated at least 50 more species to be described, our inventory shows that the true richness of the Ivinhema river basin may be even greater insofar as specific identification was not possible for four species (four species at genus level). According to taxonomists, we probably have four new species: *Serrapinnus* sp., *Ancistrus* sp., and *Rineloricaria* sp. (personal communication). Thus, even without considering other species with uncertain taxonomic definitions, 2.8% of the sampled species are poorly or completely unknown to science.

The greater representation of the orders Characiformes and Siluriformes is a frequent pattern in Neotropical waters (Castro et al. 2004). Both orders accounted for 81.6% of the sampled species, which is consistent with that found by Langeani et al. (2007). This pattern is not surprising since both orders are the most representative of South America freshwater fish (Lowe-McConnell 1999). On the other hand, the greater representation of the families Characidae and Loricariidae reflects a recurrent pattern in Brazilian continental waters (Reis et al. 2003, Buckup et al. 2007).

Although the majority of fish reported in the Ivinhema river basin was autochthonous (29.8%) in the upper Paraná and autochthonous in the lower Paraná (36.2%), summing 66%, there was a substantial occurrence of allochthonous species (20%). The two exotic species recorded (1.4%), *Clarias gariepinus* and *Coptodon rendalli*, were clearly introduced into the basin through pisciculture (Langeani et al. 2007). This is the first report of *C. gariepinus* in the Dourados and Vacaria rivers, expanding the knowledge about their occurrence in tributaries of upper Paraná, which is considered to be highly adaptable and capable of negatively affecting the native fish fauna (Alves et al. 1999). During sampling, local fishermen and riverine communities reported that this species is frequently caught in these sub-basins and in abundance in the Dourados river.

Even though we do not have knowledge about the origin of 13.5% of the species, a point that is very concerning in this inventory is that only 29.8% of species may be considered native from the upper Paraná, the remaining fishes (56.7%) are from the lower Paraná, other basins or other continents. When the subject is homogenization and species



invasion, it is important to consider not only allochthonous and exotic species. Introduced species from different parts of the basin may be more problematic than species from other basins or continents (Vitule 2009). In Paraná river, even the species that overcame the barrier from the lower to the upper parts, because of the impoundment caused by Itaipu Dam, may cause some troubles for upper communities, such as pointed by Taylor et al. (1984): hybridization, genetic losses, trophic alterations, parasites or diseases introduction. Those alterations in local communities may result in extinction of native species, biodiversity losses or biota homogenization (Vitule 2009).

The streams of the present study exhibited greater species richness (112) than streams of other works in upper Paraná, such as Castro et al. (2003, 2004), Gubiani et al. (2006) e Gomiero & Braga (2006), which reported 52, 64, 62 and 48 species, respectively. In spite of these researches have used different methodologies and our samples were carried out in more stretches and in a greater area, consequently reflecting on difference catch effort and species richness, the rivers of the Paraná river basin in the state of São Paulo have experienced the influence of a series of impoundments for hydroelectric reservoirs, a process that affects sub-basin connections, thereby preventing recolonization and, thus, influencing species richness. On the other hand, the Ivinhema river basin, in the western portion of the upper Paraná, does not contain significant impoundments, with only two small hydropower plants in the upper portion of the Dourados river (São João I and II), or any other barriers, thus maintaining the potential for greater connectivity among streams, facilitating species flux and even recolonization by species from the Paraná river floodplain.

The most common species sampled were *Astyanax lacustris*, *Serrapinnus notomelas*, *Piabarchus stramineus*, and *Hypostomus ancistroides*. These small-sized species have great feeding plasticity and are known for their capacity to adapt to and colonize different water bodies. These species were reported throughout the Ivinhema river basin and are widely distributed in the upper Paraná river (Castro et al. 2004, Gubiani et al. 2006, Teresa & Casatti 2010, Viana et al. 2013).

A wide range of standard length was registered for the sampled fish (2 to 150 cm), however, it was evidenced that the fish fauna of Ivinhema river Basin is composed basically of small-sized species, about 63.8% smaller than 20.7 cm. Despite of a greater number of samples in streams, and this may influence in a higher richness of small-sized fish (Castro 1999, Castro et al. 2003, 2004, Couto & Aquino 2011), this may be considered a pattern for the basin, because the large-sized species were sampled mainly below the intermediate altitude (430m) and in larger rivers, in areas located in tributaries such as Dourados, Brilhante and Ivinhema rivers. Moreover, 65% of 310 species analyzed in the upper Paraná river were small-sized ( $\leq 20.8$  cm) (Langeagni et al., 2007) and 50% of South-American freshwater fish are small-sized (Castro 1999). According to this last work, this pattern may be a result of the high hydrological variability in streams; we believe that this pattern act more intensively structuring fish assemblages locally, where the water levels may vary between rainy and dry periods until five meters (personal observation), selecting generalist and resilient small-sized fish that are more prone to establish in these areas.

In conclusion, the current study is particularly significant because it helps reduce the lack of information on fish communities of an ecosystem that has been kept relatively free from dams and

impoundments, from its headwaters to its confluence with the Paraná river basin. These data are also important for understanding the distribution of fish among sub-basins, adding new species records for the Ivinhema river basin, and the upper Paraná basin as a whole, serving as reference data for each region and contributing to the management, decision-making, and implementation of politics to conserve the basin as a whole. Furthermore, these data should prove useful as a reference for monitoring the fish fauna of the basin, which has experienced great alterations to its landscapes over the last decade due to the expansion of sugarcane crops and also may be suffering some pressure by a high percentage of non-native species in the upper Paraná.

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## Author Contributions

Wagner Vicentin: Substantial contribution in the concept and design of the study; contribution to data collection; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Fabiane Silva Ferreira: Substantial contribution in the concept and design of the study; contribution to data collection; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

Yzel Rondon Suárez: Substantial contribution in the concept and design of the study; contribution to data collection; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intellectual content.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.



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## Arctiini Leach, [1815] (Lepidoptera, Erebidae, Arctiinae) of the Brazilian Amazon. IV – Subtribe Euchromiina Butler, 1876

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**Abstract:** The Euchromiina moths comprise 735 species in the neotropics. Here we provide a list of Euchromiina species from the Brazilian Amazon. The list was produced from specimens deposited in the most important Brazilian collections and from literature data. Nearly 30% (219) of the neotropics Euchromiina species were recorded, including 13 new occurrences for the Brazilian Amazon. Santarém and Belém were the municipalities with the highest number of species records, with 96 and 74, respectively. Although the number of Euchromiina records is high in the Amazon, this value is underestimated because the fauna was never sampled in the vast majority of the biome. This is a worrying scenario because the Amazon has the world highest absolute rate of forest reduction. In order to suggest efficient conservation policies for the Amazon Arctiinae fauna, it is urgent to intensify the sampling effort in this biome.

**Keywords:** Amazon, Tiger moths, Inventory, Noctuoidea, Wasp Moths.

## Arctiini Leach, [1815] (Lepidoptera, Erebidae, Arctiinae) da Amazônia Brasileira. IV – Subtribo Euchromiina Butler, 1876

**Resumo:** As mariposas Euchromiina compreendem 735 espécies na região Neotropical. Neste trabalho, nós apresentamos uma lista das espécies de Euchromiina que ocorrem na Amazônia brasileira. A lista foi produzida através de observação de espécimes depositados nas mais importantes coleções brasileiras e também através de dados da literatura. Aproximadamente 30% (219) das espécies de Euchromiina que ocorrem na região Neotropical foram registradas, incluindo 13 novas ocorrências para a Amazônia brasileira. Santarém e Belém foram os municípios com maior número de espécies registradas, com 96 e 74, respectivamente. Embora o número de registros de Euchromiina seja considerado alto na Amazônia, este valor é subestimado visto que a fauna nunca foi amostrada na imensa maioria do bioma. Este cenário é preocupante, pois a Amazônia possui a maior taxa absoluta de redução florestal do mundo. A fim de podermos sugerir políticas de conservação eficientes para a fauna de Arctiinae da Amazônia, é urgente que se intensifique o esforço de coleta neste bioma.

**Palavras-chave:** Amazônia, mariposas-tigre, Inventário, Noctuoidea, mariposas-vespa.

## Introduction

Tropical rainforests support some of the highest species richness of the Earth and correspond to 60% of the hotspots defined by Myers et al. (2000). The Amazon represents 40% of the tropical rainforest areas (Aragão et al. 2014). It covers part of nine countries in South America, with 69% of this area in Brazil (Vieira et al. 2008). However, the original area of the Brazilian Amazon has been reduced by 20% (INPE 2015).

Lepidoptera inventories in the Brazilian Amazon are scarce (Santos et al. 2008). Although there have been scientific publications on Amazonian Lepidoptera since the 19th century, many earlier inventories in the Brazilian Amazon do not provide an accurate description of the collection sites, which significantly restricts the use of their data (Casagrande et al. 2012). Several recent inventories also have problems, such as being made in a short period of time and/or poorly distributed in space (usually along major rivers or near urban centers).

Arctiinae was worldwide Lepidoptera taxa (Heppner 1991). With approximately 11,000 species, the tiger moths are divided in four tribes (Zahiri et al. 2012), but only Arctiini and Lithosiini occurred in the Neotropics. Arctiini are distributed in seven subtribes: Arctiina, Callimorphina, Spilosomina, Phaegopterina, Pericopina, Ctenuchina and Euchromiina (Weller et al. 2009, Vincent & Laguerre 2014).

The Euchromiina moths are found mainly in the neotropics and comprises approximately 68 genera and 735 species (Weller et al. 2009). Several species have nocturnal activity, but some species are exclusively diurnal (Hagmann 1938). Many species of Euchromiina form mimetic rings with butterflies, beetles, and especially wasps (Simmons 2009). Their wings can have areas or be totally transparent, closely resembling the wings of Hymenoptera. Moreover, some species simulate the petiole of Hymenoptera by reducing of abdomen sclerites and increasing thorax sclerites (Simmons 2009). Males of several species have modified scales below the second abdominal segment that can be released in dangerous situations or during mating (Yack 2004). Some Euchromiina use ultrasound in courtship instead of or in combination with feromonal cues (Sanderford et al. 1998). Several Euchromiina larvae are brilliantly coloured (Weller et al. 2009) and feed on several plant families. Adults of some species are pharmacophagous on pyrrolizidine alkaloids (Conner & Jordan 2009).

We present a list of Euchromina moths occurring in the Brazilian Amazon. We make the species list mainly from specimens deposited in the most important Brazilian collections and also from literature data. This study is a continuation of Teston & Ferro (2016a, b) and Teston et al. (2019) and aims to increase knowledge of the diversity of Arctiinae in the Amazon region.

## Materials and Methods

We intensively searched the literature and examined specimens from entomological collections of the Instituto Nacional de Pesquisas na Amazônia (INPA; Manaus), Museu Paraense Emílio Goeldi (MPEG; Belém), Coleção Becker (VOB; Camacan), Coleção Entomológica Padre Jesus Santiago Moure of the Universidade Federal do Paraná (DZUP; Curitiba), Fundação Instituto Oswaldo Cruz (FIOC; Rio de Janeiro), Museu de Zoologia of the Universidade de São Paulo (MZUSP; São Paulo), Museu Nacional of the Universidade Federal do Rio de Janeiro (MNRJ; Rio de Janeiro), and Laboratório de Estudos

de Lepidópteros Neotropicais (LELN) of the Universidade Federal do Oeste do Pará (UFOPA; Santarém). To identify the species, we used literature (Hampson 1898, 1914, Seitz 1919-1925) and specimens deposited in the visited collections. The systematic organization to generic level follows Hampson (1898) and Weller et al. (2000), with updates of the generic names according Watson et al. (1995) and corrections of Pinheiro & Duarte (2013), Pinheiro & Gaal-Haszler (2015) and Pinheiro (2016).

The geographical coordinates of the localities in the Brazilian Amazon with Arctiinae records were obtained from the Geo Loc tool of “Species Link date & tools” (<http://splink.cria.org.br/geoloc>) and Google Earth (<https://earth.google.com/web/>). The list is organized alphabetically. Species and records without precise location data, and those from locations that belong to more than one biome (e.g., Cerrado and Amazon) were not included in the list.

## Results

Our research generated a list of 219 Euchromiina species (Table 1), including 13 new occurrences for the Brazilian Amazon (indicated by “NEW”). Eighteen species appear as new records for the municipalities and their respective States (indicated by “AMZ”). In total, 71 (9.2%) Amazonian municipalities had Euchromiina species records (Table 2 and Figure 1). Santarém (PA), Belém (PA), and São Félix do Xingu (PA) were the municipalities with the highest number of species, with 96, 74, and 47, respectively.

We recorded 49 genera, of which 19 were monospecific. The genera *Cosmosoma* Hübner, [1823] presented the highest number of species (27), followed by *Saurita* Herrich-Schäffer, [1855] (19) and *Leucotmemis* Butler, 1876 (12). The species with the highest number of locality records were *Belemnina eryx* (Fabricius, 1775) and *Isanthrene porphyria* (Walker, 1854) (with 15), followed by *Orcynia calcarata* (Walker, 1854) (14), *Cosmosoma telephus* (Walker, 1854) and *Histioea proserpina* (Hübner, 1827) (13). Ninety-one species (41.4%) occurred in only one locality (Table 1).

## Discussion

The number of Euchromiina species recorded for the Brazilian Amazon was high. It corresponds to 29.8% of neotropics Euchromiina fauna (735, Weller et al. 2009) and was more than two times higher than the Mexican (112, Hernández-Baz et al. 2013), the Brazilian Cerrado (76, Ferro et al. 2010), and Rio Grande do Sul Brazilian State (60, Ferro & Teston 2009) Euchromiina richness.

The explanation of Santarém and Belém are the Amazon municipalities with the highest number of Euchromiina records probably are related to the proximity and access to the collection sites, which allow a larger sample effort. Belém is the capital of the state of Pará and has an important museum (Museu Paraense Emílio Goeldi, founded in 1866) and other research institutions, as well as easy access to sampling sites. Santarém was widely sampled by H. Zerny and G. Hagmann (Zerny 1931, Hagmann 1938). Hagmann resided in Santarém and Zerny spent a season collecting moths there. Belém was also the second municipality with the highest number of Phaegopterina, Pericopina and Ctenuchina records (Teston & Ferro 2016a, b, Teston et al. 2019).



**Table 1.** Euchromiina (Erebidae, Arctiinae, Arctiini) species of the Brazilian Amazon. The record column shows the Brazilian state in abbreviated form followed by municipality. The name of the locality is enclosed in braces and the author of the first record is in parentheses. \* New record. States abbreviations: AC = Acre, AM = Amazonas, AP = Amapá, MA = Maranhão, MT = Mato Grosso, PA = Pará, RO = Rondônia and RR = Roraima.

Species	Record
1. <i>Abnormipterus abnormis</i> (Hampson, 1898)	AM, [Boa Vista do Ramos] {Massauari} (Hampson 1898)
2. <i>Autochlores bijuncta</i> (Walker, 1856)	AM, Tefê {Ega} (Walker [1865]); PA, [Belém] (Walker 1856)
3. <i>Autochlores caunus</i> (Cramer, [1779])	AM, São Paulo [de Olivença] (Zerny 1931); PA, Santarém {Taperinha} (Zerny 1931)
4. <i>Autochlores collocata</i> (Walker, [1865])	AM, Benjamin Constant*, Tefê {Ega} (Walker [1865]); MA, Açailândia*; RO, Cacaúlândia*
5. <i>Autochlores completa</i> (Walker, 1854)	AM, [Eirunepé] {Juruá river, São Felipe} (Zerny 1931), São Paulo de Olivença*; PA, [Belém] (Walker 1854a)
6. <i>Autochlores crinopoda</i> Kaye, 1918 <sup>NEW</sup>	AM, Benjamin Constant*, Maués*; PA, Novo Progresso {Cachimbo}*,
7. <i>Autochlores ectomelaena</i> Hampson, 1898	AM, Itamar[a]t[i] (Rothschild 1931), upper Amazonas [river] (Hagmann 1938); PA, Santarém {Taperinha} (Hagmann 1938)
8. <i>Autochlores enagrus</i> (Cramer, [1779])	AM, Itacoatiara*, Tefê {Ega} (Hampson 1898); PA, Óbidos*, Santarém*, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
9. <i>Autochlores proterva</i> (Draudt, 1916)	PA, Belterra {National Forest of Tapajós} (Freitas 2014)
10. <i>Autochlores simplex</i> (Walker, 1856)	AM, Tefê (Zerny 1931); PA, [Belém] (Walker 1856), Santarém {Mojú} (Zerny 1931)
11. <i>Autochlores solimões</i> Schaus, 1924	AM, [Tefê] {Mouth of Rio Tefê and Rio Solimões} (Schaus 1924)
12. <i>Belemnia eryx</i> (Fabricius, 1775)	AM, Borba*, Fonte Boa (Rothschild 1910), Humaitá (Rothschild 1910), São Paulo de Olivença*, Tefê (Rothschild 1910); MA, Açailândia*; MT, Sinop*; PA, Itaituba*, Parauapebas {Serra Norte, Carajás}*, Prainha (Butler 1878), Santarém*, Tapajós [river] (Hampson 1901); RO, Ariquemes*, Cacaúlândia*, Pimenta Bueno*, Porto Velho {Aliança} (Rothschild 1910)
13. <i>Belemnia inaurata</i> (Sulzer, 1776) <sup>AMZ</sup>	AM, Manicoré*, São Gabriel da Cachoeira*, Tefê*; AP, Serra do Navio*; MT, Sinop*; PA, Belém*, Santarém*
14. <i>Belemnia ochriplaga</i> Hampson, 1901	AM, Fonte Boa (Rothschild 1910), Humaitá (Rothschild 1910), Manaus*, Tefê (Rothschild 1910); PA, [Belém] (Hampson 1901), Belterra {National Forest of Tapajós} (Freitas 2014), Ju[r]ut[i] (Rothschild 1910), Prainha (Hampson 1901), Santarém*; RO, Porto Velho {Aliança} (Rothschild 1910)
15. <i>Belemniastis eucyane</i> (R. Felder, 1875)	RO, Porto Velho {Aliança} (Hampson 1920)
16. <i>Belemniastis whiteleyi</i> (Druce, 1888) <sup>NEW</sup>	AM, Benjamin Constant*
17. <i>Bodosa tina</i> (Walker, 1854)	AM, Maués (Machado Filho & Rêgo Barros 1969), Tefê {Ega} (Machado Filho & Rêgo Barros 1969); PA, Óbidos (Machado Filho & Rêgo Barros 1969), Santarém {Taperinha} (Machado Filho & Rêgo Barros 1969)
18. <i>Calonotos acutipennis</i> Zerny, 1931	PA, Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931)
19. <i>Calonotos aequimaculatus</i> Zerny, 1931	PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
20. <i>Calonotos angustipennis</i> Zerny, 1931	PA, Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
21. <i>Calonotos chalcipleura</i> Hampson, 1898	AM, Barcelos {Moura}*, Manaus*; AP, Serra do Navio*; MA, Açailândia*; MT, Aripuanã*; PA, Belém*, Novo Progresso {Cachimbo}*, Parauapebas {Serra Norte, Carajás}*, Óbidos*, Santarém {Taperinha} (Zerny 1931); RO, Cacaúlândia*
22. <i>Calonotos helymus</i> (Cramer, [1775])	AP, Serra do Navio*; PA, Santarém (Valente et al. 2018)
23. <i>Calonotos hoffmannsi</i> (Rothschild, 1911)	PA, Itaituba to Óbidos (Rothschild 1911), Itaituba (Hampson 1914)
24. <i>Calonotos longipennis</i> Rothschild, 1911	PA, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
25. <i>Calonotos phlegmon</i> (Cramer, [1775])	AM, Manaus*, São Paulo [de Olivença] (Zerny 1931); AP, Serra do Navio*; MA, Açailândia*; MT, Aripuanã*; PA, [Belém] (Hampson 1898), Capitão Poço*, Marabá*, Novo Progresso {Cachimbo}*, Parauapebas {Serra Norte, Carajás}*, Óbidos*; RO, Cacaúlândia*, Jarú*

Continuation Table 1.

Species	Record
26. <i>Calonotos tiburtus</i> (Cramer, [1779]) <sup>NEW</sup>	AM, Manaus*
27. <i>Calonotos triplaga</i> (Hampson, 1909)	AM, Amazons (Hampson 1898), Manaus (Hampson 1909); AP, Serra do Navio*; MA, Açailândia*; PA, Altamira {Monte Santo} (Teston & Delfina 2010) and {Serra do Pardo National Park} (Teston & Correa 2015), Oriximiná {Rio Cuminá}*; Parauapebas {Serra Norte, Carajás}*; Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
28. <i>Calonotos tripunctata</i> Druce, 1898	PA, Belterra {National Forest of Tapajós} (Freitas 2014)
29. <i>Chrostosoma dhamis</i> Schaus, 1928	PA, [Belém] (Schaus 1928)
30. <i>Chrostosoma dolens</i> (Walker, 1854)	PA, [Belém] (Walker 1854a)
31. <i>Chrostosoma haematica</i> (Perty, 1834)	AM, Amazon river (Perty 1834), Benjamin Constant*, São Paulo de Olivença*, Tefé {Ega} (Walker [1865]); AP, Serra do Navio*; PA, Belém*, Viséu*
32. <i>Chrostosoma tricolor</i> (R. Felder, 1874)	AM, Amazon river (R. Felder 1874)
33. <i>Cosmosoma achemon</i> (Fabricius, 1781)	AM, Benjamin Constant*, Manaus*; MA, Açailândia*; MT, Aripuanã*; PA, [Belém] (Druce 1897), Óbidos*, Prainha (Butler 1878), Santarém (Hampson 1898) {Taperinha} (Zerny 1931); RO, Porto Velho*
34. <i>Cosmosoma ada</i> (Herrich-Schäffer, [1855])	[AM], Amazonas (Zerny 1912)
35. <i>Cosmosoma admota</i> (Herrich-Schäffer, [1854])	PA, Belterra {National Forest of Tapajós} (Freitas 2014), Santarém (Valente et al. 2018), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
36. <i>Cosmosoma anoxanthia</i> Druce, 1905	PA (Hagmann 1938)
37. <i>Cosmosoma auge</i> (Linnaeus, 1758)	AM, Manicoré*; MA, Açailândia*; PA, Santarém {Mojú and Taperinha} (Zerny 1931) RO, Candeias do Jamari*
38. <i>Cosmosoma batesii</i> (Butler, 1876)	PA, [Belém] (Butler 1876), Santarém {Taperinha} (Zerny 1931)
39. <i>Cosmosoma centralis</i> (Walker, 1854) <sup>AMZ</sup>	MA, Açailândia*; PA, Belém*; RO, Cacaúlândia*
40. <i>Cosmosoma consolata</i> (Walker, 1856)	PA, Belém*, Altamira {Monte Santo} (Delfina & Teston 2013), Parauapebas {Serra Norte, Carajás}*; São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*, Porto Velho*
41. <i>Cosmosoma contracta</i> (Walker, 1856)	[AC], Upper Juruá river (Zerny 1931); AM, [Lábrea] {Rio Purus, Huitanaã} (Bryk 1953), Valley of Amazon [river] (Walker 1856); PA, Santarém {Taperinha} (Zerny 1931)
42. <i>Cosmosoma corvica</i> (Dognin, 1910)	PA, Santarém {Taperinha} (Zerny 1931), Altamira {Monte Santo} (Delfina & Teston 2013)
43. <i>Cosmosoma durca</i> Schaus, 1896 <sup>AMZ</sup>	MA, Açailândia*
44. <i>Cosmosoma festiva</i> Walker, 1854	AM, Parintins {Villa Nova} (Hampson 1914); MA, Açailândia*
45. <i>Cosmosoma klagesi</i> Rothschild, 1910	PA, [Belém] (Hampson 1914), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
46. <i>Cosmosoma melathoracia</i> Kaye, 1901	PA, Santarém {Taperinha} (Zerny 1931)
47. <i>Cosmosoma metallescens</i> (Ménétriés, 1857)	AM, [Autazes] {Rio Autaz} (Bryk 1953), [Eirunepé] {Juruá river, São Felipe} (Zerny 1931), Parintins {Villa Nova} (Hampson 1898); PA, Altamira {51°BIS} (Teston et al. 2012) and {Monte Santo} (Delfina & Teston 2013) and {Serra do Pardo National Park} (Teston & Correa 2015), [Belém] (Butler 1876), Belterra {National Forest of Tapajós} (Freitas 2014), Marabá*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
48. <i>Cosmosoma nelea</i> Möschler, 1877	PA, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
49. <i>Cosmosoma pheres</i> (Stoll, [1782]) <sup>AMZ</sup>	AM, Manaus*; MA, Açailândia*; PA, Belém*; RO, Cacaúlândia*
50. <i>Cosmosoma rasera</i> Jones, 1914 <sup>AMZ</sup>	MA, Açailândia*; PA, Marabá*, Novo Progresso {Cachimbo}*; RO, Candeias do Jamari*
51. <i>Cosmosoma remota</i> (Walker, 1854) <sup>AMZ</sup>	AM, Manicoré*, Santa Isabel do Rio Negro*; MA, Açailândia*; MT, Aripuanã*; PA, Belém*, Capitão Poço*, Parauapebas {Serra Norte, Carajás}*; Santarém*, Viséu*; RO, Cacaúlândia*

Continuation Table 1.

Species	Record
52. <i>Cosmosoma seraphina</i> (Herrich-Schäffer, [1854])	PA, [Belém] (Herrich-Schäffer [1854]), Novo Progresso {Cachimbo}*; RO, Porto Velho*
53. <i>Cosmosoma stibosticta</i> (Butler, 1876)	PA, Altamira {Monte Santo} (Teston & Delfina 2010)
54. <i>Cosmosoma subflamma</i> (Walker, 1854)	AM, Amazonas (Zerny 1931), Manaus*, Rio Urubu*; PA, Altamira {Monte Santo} (Teston & Delfina 2010), Belém*, Bragança*, Itaituba*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
55. <i>Cosmosoma telephus</i> (Walker, 1854)	AM, [Eirunepé] {Juruá river, São Felipe} (Zerny 1931); AP, Serra do Navio*; MA, Açailândia*; MT, Aripuanã*, Sinop*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Belém*, Novo Progresso {Cachimbo}*, Parauapebas {Serra Norte, Carajás}*, Peixe Boi*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
56. <i>Cosmosoma tengyra</i> (Walker, 1854)	PA, [Belém] (Hampson 1898)
57. <i>Cosmosoma teuthras</i> (Walker, 1854)	MA, Açailândia*; MT, Aripuanã*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Parauapebas {Serra Norte, Carajás}*, Santarém (Butler 1876) {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
58. <i>Cosmosoma thoracica</i> Schaus, 1905	PA, Santarém {Taperinha} (Zerny 1931), Altamira {Monte Santo} (Teston & Delfina 2010), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
59. <i>Cosmosoma xanthocera</i> Hampson, 1898	AM, [Boa Vista do Ramos] {Massauari} (Hampson 1898)
60. <i>Diospage rhebus</i> (Cramer, 1779)	AM, Amazons (Rothschild 1910), Benjamin Constant*, Fonte Boa (Rothschild 1910), Manicoré*, São Paulo de Olivença (Hampson 1901), Tefé (Rothschild 1910); PA, Óbidos*, Santarém*; RO, Cacaúlândia*, Porto Velho {Aliança} (Rothschild 1910)
61. <i>Dixophlebia quadristrigata</i> (Walker, [1865])	AM, Tefé {Ega} (Walker [1865]); PA, Santarém {Taperinha} (Zerny 1931)
62. <i>Dycladia correbioides</i> (C. Felder, 1869)	[RR], [Caracará] {Rio Branco} (Bryk 1953)
63. <i>Dycladia lucetius</i> (Stoll, [1781])	AM, Fonte Boa*, Manaus*, Rio Urubu*; MA, Açailândia*; PA, Altamira {Monte Santo} (Teston & Delfina 2010) {51°BIS} (Teston et al. 2012), Belém*, Belterra {National Forest of Tapajós} (Freitas 2014), Chaves (Hampson 1898), Santarém (Valente et al. 2018), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015), Soure*
64. <i>Dycladia transacta</i> (Walker, 1856)	PA, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
65. <i>Erruca cruenta</i> (Perty, 1834)	AM, Amazon river (Perty 1834)
66. <i>Erruca erythrarchos</i> (Walker, 1854) <sup>NEW</sup>	AM, Benjamin Constant*; MA, Açailândia*; PA, Marabá*
67. <i>Gymnelia felderi</i> Rothschild, 1931	[AM], Amazons (Rothschild 1931)
68. <i>Gymnelia scita</i> (Walker, 1856)	AM, Tefé {Ega} (Walker [1865])
69. <i>Gymnelia simillimum</i> (Rothschild, 1911)	AM, Amazon river (Rothschild 1911)
70. <i>Gymnelia vesparia</i> (Perty, 1834)	AM, Amazon river (Perty 1834), Parintins {Villa Nova} (Walker 1856)
71. <i>Gymnelia villia</i> (Druce, 1906) <sup>NEW</sup>	RO, Cacaúlândia*
72. <i>Gymnelia xanthogastra</i> (Perty, 1834)	AM, São Paulo de Olivença*
73. <i>Histioea amazonica</i> Butler, 1876	AM, Benjamin Constant (Rio Javari) (Machado Filho & Rêgo Barros 1973), [Beruri] {Re[d]e[n][ção] on Rio Purus} (Butler 1878), Borba (Machado Filho & Rêgo Barros 1973), [Ipixuna] {Lago Cerrado on Rio Juruá} (Butler 1878), Manaus (Zerny 1931), Parintins {Villa Nova} (Butler 1876), São Paulo [de Olivença] {Solimões [river]} (Bryk 1953), Tabatinga (Butler 1878), Tefé {Ega} (Butler 1876); PA, Óbidos (Machado Filho & Rêgo Barros 1973), Santarém {Tapará} (Zerny 1931)
74. <i>Histioea bellatrix</i> (Walker, 1854)	AM, Manicoré {Rio Madeira} (Machado Filho & Rêgo Barros 1973)
75. <i>Histioea cepheus</i> (Cramer, 1779)	AM, Benjamin Constant*, [Maués] {Rio Para[u]ar[i]} (Machado Filho & Rêgo Barros 1973), São Paulo de Olivença (Machado Filho & Rêgo Barros 1973), Tefé (Machado Filho & Rêgo Barros 1973); AP, Serra do Navio (ICOMI) (Machado Filho & Rêgo Barros 1973); PA, [Almeirim] {Rio Paru}*; Altamira {51°BIS} (Teston et al. 2012), Marabá*, Óbidos*, Oriximiná {Rio Cuminá}*, Parauapebas {Serra Norte, Carajás}*, Santarém {Taperinha} (Draudt 1931)

Continuation Table 1.

Species	Record
76. <i>Histioea glaucozona</i> Druce, 1898	[AM], Amazons (Druce 1898b)
77. <i>Histioea hoffmannsi</i> Rothschild, 1911	AM, Humaitá {Rio Madeira} (Rothschild 1911)
78. <i>Histioea paraensis</i> Machado Filho & Rêgo Barros, 1971	PA, Óbidos (Machado Filho & Rêgo Barros 1971)
79. <i>Histioea proserpina</i> (Hübner, 1827)	AC, Porto Walter {Alto Juruá} (Machado Filho & Rêgo Barros 1971), Xapuri (Machado Filho & Rêgo Barros 1971); AM, Amazons (Hampson 1898), [Autazes] {Rio Autaz} (Bryk 1953), Benjamin Constant {Rio Javari} (Machado Filho & Rêgo Barros 1971), [Eirunepé] {Juruá river, São Felipe} (Zerny 1931), Manaus (Machado Filho & Rêgo Barros 1971), Maués*, Rio Negro (Zerny 1931), São Paulo de Olivença (Machado Filho & Rêgo Barros 1971), São Gabriel da Cachoeira*, Tefé {Egas} (Machado Filho & Rêgo Barros 1971); PA, Óbidos (Machado Filho & Rêgo Barros 1971), Oriximiná {Rio Cuminá}* , Santarém {Taperinha} (Zerny 1931)
80. <i>Homoeocera stictosoma</i> Druce, 1898	PA, Almeirim {Jari} (Hawes et al. 2009)
81. <i>Hyda basillutea</i> (Walker, 1854)	AM, Santa Isabel do Rio Negro*; AP, Mazagão*; MT (Zerny 1931), Sinop*; PA, Belém*, Igarapé Açú*, Ourém*, Prainha (Butler 1878), Santarém {Taperinha} (Zerny 1931), Tucuruí*
82. <i>Hypatia delecta</i> (Butler, 1876)	PA, [Belém] (Butler 1876)
83. <i>Hypatia melaleuca</i> (Walker, 1854)	PA, [Belém] (Walker 1854a); RO, Cacaúlândia*
84. <i>Hypocharis albicincta</i> Cerda, 2008	PA, Belterra {National Forest of Tapajós} (Freitas 2014)
85. <i>Hypocharis clusia</i> (Druce, 1897)	AM, São Paulo de Olivença*; PA (Hagmann 1938)
86. <i>Isanthrene aterrima</i> (Walker, [1865])	AM, Parintins*, Tefé {Ega} (Walker [1865]); AP, Serra do Navio*; RO, Cacaúlândia*
87. <i>Isanthrene melas</i> (Cramer, [1775])	AM, Benjamin Constant*, [Eirunepé] {Juruá river, Matto Pyri} (Zerny 1931), Parintins {Villa Nova} (Hampson 1898), São Paulo de Olivença*; PA, [Belém] (Walker 1854a), Óbidos (Zerny 1931), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
88. <i>Isanthrene notipennis</i> (Butler, 1876)	AM, Parintins {Villa Nova} (Butler 1876); MA, Açailândia*; PA, Óbidos*, Santarém*
89. <i>Isanthrene porphyria</i> (Walker, 1854)	AM, Benjamin Constant*, [Eirunepé] {Juruá river, São Felipe and Matto Pyri} (Zerny 1931), Manicoré*, Parintins {Villa Nova} (Hampson 1898), São Paulo de Olivença*, Tefé {Ega} (Hampson 1898); AP, Serra do Navio*; MA, Açailândia*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), [Belém] (Hampson 1898), Paragominas*, Santarém*, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*, Jarú*
90. <i>Isanthrene profusa</i> Hampson, 1898	AM, Benjamin Constant*, Tefé {Ega} (Hampson 1898); PA, Altamira {Monte Santo} (Teston & Delfina 2010) and {51°BIS} (Teston et al. 2012) and {Serra do Pardo National Park} (Teston & Correa 2015), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
91. <i>Isanthrene varia</i> (Walker, 1854)	AM, São Paulo de Olivença*; MT, Sinop*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), [Belém] (Walker 1854a), Santarém {Mojú and Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Porto Velho*
92. <i>Isanthrene vespiformes</i> (Butler, 1876)	AM, Parintins {Villa Nova} (Butler 1876); PA, Itaituba*, Óbidos (Zerny 1931), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
93. <i>Lepidoneiva erubescens</i> (Butler, 1876)	AM (Bryk 1953); MT, Sinop*; PA, Novo Progresso {Cachimbo}*
94. <i>Leucotmemis climacina</i> (Butler, 1876) <sup>NEW</sup>	MA, Açailândia*; RO, Cacaúlândia*
95. <i>Leucotmemis dorsalis</i> (Walker, 1854)	AM, Manaus (Bryk 1953); MA, Açailândia*; PA, Santarém (Walker 1854a); RO, Cacaúlândia*
96. <i>Leucotmemis emergens</i> (Walker, [1865])	AM, Manaus (Bryk 1953), Tefé {Ega} (Walker [1865])
97. <i>Leucotmemis felderi</i> (Rothschild, 1911)	AM, Amazon river (Rothschild 1911)
98. <i>Leucotmemis flavidior</i> Gaede, 1926	PA, [Belém] (Gaede 1926)



Continuation Table 1.

Species	Record
99. <i>Leucotmemis intersecta</i> (Walker, [1865])	AM, Tefé {Ega} (Walker [1865]); PA (Hagmann 1938)
100. <i>Leucotmemis margaripha</i> (Butler, 1876)	PA, [Belém] (Butler 1876), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
101. <i>Leucotmemis nexa</i> (Herrich-Schäffer, [1854])	AM, Parintins {Villa Nova} (Butler 1876); AP, Serra do Navio*; MT, Sinop*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Belterra {National Forest of Tapajós} (Freitas 2014), Óbidos (Zerny 1931), Santarém (Herrich-Schäffer [1854]) {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
102. <i>Leucotmemis ornatula</i> (Walker, 1854)	PA, [Belém] (Walker 1854a)
103. <i>Leucotmemis tenthredoides</i> (Walker, 1856)	AM, Rio Juruá (Butler 1878), Santa Isabel do Rio Negro*, Tefé {Ega} (Butler 1876), Tonantins (Bryk 1953), Valley of the Amazons [river] (Walker 1856); PA, Belém*, Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931); RO, Porto Velho*
104. <i>Leucotmemis torrida</i> (Walker, 1854)	PA, Altamira {Monte Santo} (Teston & Delfina 2010), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931), Tapajós [river] (Walker 1854a)
105. <i>Leucotmemis varipes</i> (Walker, 1854)	AM, Manaus (Butler 1878); PA, [Belém] (Walker 1854a), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931); RO, Cacaúlândia*
106. <i>Loxophlebia cinctata</i> Hampson, 1905	AM, Barcelos {Moura and Tomar}*; Manaus*; MT, Sinop*; PA, Santarém {Taperinha} (Zerny 1931); RO, Cacaúlândia*
107. <i>Loxophlebia crocata</i> (Herrich-Schäffer, [1854]) <sup>AMZ</sup>	MA, Açailândia*; PA, Capitão Poço*
108. <i>Loxophlebia crumata</i> Dognin, 1911	PA (Hagmann 1938)
109. <i>Loxophlebia diaphana</i> (Sepp, [1848])	PA, [Belém] (Walker 1854a), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
110. <i>Loxophlebia imitata</i> (Druce, 1884) <sup>AMZ</sup>	MA, Açailândia*
111. <i>Loxophlebia picta</i> (Walker, 1854)	AM, Santa Isabel do Rio Negro*; MA, Açailândia*; PA, [Belém] (Walker 1854a), Capitão Poço*, Novo Progresso {Cachimbo}*; Santarém (Walker 1854a) {Taperinha} (Zerny 1931); RO, Cacaúlândia*, Porto Velho*
112. <i>Loxophlebia postflavia</i> Druce, 1898	PA, Santarém {Taperinha} (Zerny 1931)
113. <i>Loxophlebia pyrgion</i> (Druce, 1884)	PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
114. <i>Loxophlebia roseipectus</i> Rothschild, 1931	AM, [Itacoatiara] {Lower Amazons, junction with Rio Madeira} (Rothschild 1931)
115. <i>Loxophlebia semiaurantia</i> Rothschild, 1931	PA, [Belém] (Rothschild 1931)
116. <i>Loxophlebia triangulifera</i> (R. Felder, 1874)	[AM], Amazons (Hampson 1898); PA, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
117. <i>Macrocneme adonis</i> Druce, 1884	AM, Barcelos {Moura}*; Borba*, Manaus*, São Paulo de Olivença*; PA (Hagmann 1938), Belém*, Belterra {National Forest of Tapajós} (Freitas 2014), Santarém (Valente et al. 2018), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
118. <i>Macrocneme chrysis</i> (Guérin-Méneville, [1844])	AM, Rio Purus (Bryk 1953); PA, Santarém {Taperinha} (Zerny 1931)
119. <i>Macrocneme lades</i> (Cramer, [1775])	AM, Manicoré*; PA, Altamira {Monte Santo} (Teston & Delfina 2010) and {Serra do Pardo National Park} (Teston & Correa 2015), [Belém] (Hampson 1898), Belterra {National Forest of Tapajós} (Freitas 2014), Capitão Poço*, Santarém (Valente et al. 2018), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
120. <i>Macrocneme leucostigma</i> (Perty, 1834)	PA, Santarém {Taperinha} (Zerny 1931)
121. <i>Macrocneme maja</i> (Fabricius, 1787)	AM, Rio Negro (Zerny 1931); PA, [Belém] (Hampson 1898), Santarém {Taperinha} (Zerny 1931)

Continuation Table 1.

Species	Record
122. <i>Macrocneme thyra</i> Möschler, 1883	AM, São Paulo de Olivença*, Tefé (Bryk 1953); PA, [Belém] (Hampson 1898), Belterra {National Forest of Tapajós} (Freitas 2014)
123. <i>Macrocneme thyridia</i> Hampson, 1898	AM (Travassos Filho 1940); PA, Santarém (Valente et al. 2018)
124. <i>Macrocneme verdivittata</i> (Klages, 1906) <sup>AMZ</sup>	AM, Barcelos {Moura}*; PA, Novo Progresso {Cachimbo}*, Parauapebas {Serra Norte, Carajás}*
125. <i>Macrocneme vidua</i> (Bryk, 1953)	[RR], [Caracarái] {Rio B[r]anco} (Bryk 1953)
126. <i>Macrocneme zongonata</i> Dietz, 1994	PA, Santarém (Valente et al. 2018)
127. <i>Mesothen desperata</i> (Walker, 1856)	AM, Valley of [river] Amazon (Walker 1856); PA (Hagmann 1938), Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém*, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
128. <i>Mesothen endoleuca</i> Druce, 1905	PA, Santarém {Taperinha} (Zerny 1931)
129. <i>Mesothen inconspicua</i> (Kaye, 1911) <sup>AMZ</sup>	RO, Cacaúlândia*; PA, Capitão Poço*
130. <i>Mesothen pyrrha</i> Schaus, 1889	PA, Santarém {Taperinha} (Zerny 1931)
131. <i>Metaloba argante</i> (Druce, 1897)	MA, Açailândia*; PA, Altamira {Monte Santo} (Teston & Delfina 2010) and {Serra do Pardo National Park} (Teston & Correa 2015), Belterra {National Forest of Tapajós} (Freitas 2014), Novo Progresso {Cachimbo}*, Parauapebas {Serra Norte, Carajás}*, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
132. <i>Metamya chrysonota</i> (Hampson, 1898)	PA, Itaituba (Hampson 1898)
133. <i>Metamya intersecta</i> (Hampson, 1898)	PA, [Belém] (Hampson 1898)
134. <i>Metamya picta</i> (Druce, 1898) <sup>AMZ</sup>	AM, Manicoré*; PA, Santarém*
135. <i>Methysia notabilis</i> (Walker, 1854)	PA, [Belém] (Walker 1854a), Santarém*
136. <i>Micragyrta diminuta</i> (Walker, 1854)	AM, Borba*, Tefé {Ega} (Walker [1865]); PA, [Belém] (Walker 1854b), Santarém {Taperinha} (Zerny 1931)
137. <i>Mimagyrta abdominalis</i> (Rothschild, 1912)	AM, Humaitá (Rothschild 1912); PA, [Belém] (Zerny 1931)
138. <i>Mimagyrta pampa</i> (Druce, 1893)	AM, Tefé (Hampson 1898)
139. <i>Mystrocne atavia</i> Hampson, 1898	AM, Fonte Boa (Hampson 1898)
140. <i>Mystrocne varipes</i> (Walker, 1854)	AM, Santa Isabel do Rio Negro*, Tefé*; PA, Anajás*, [Belém] (Walker 1854a), Breves*, Cametá (Zerny 1931)
141. <i>Nyridela chalciope</i> (Hübner, 1827)	AM, Manaus*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Belém*, Capitão Poço*, Novo Progresso {Cachimbo}*, Óbidos*, Parauapebas {Serra Norte, Carajás}*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
142. <i>Orcynia calcarata</i> (Walker, 1854)	AM, Manaus*, Novo Aripuanã*, Tefé*; AP, Serra do Navio*; MA, Açailândia*; PA, Almeirim {Jari} (Hawes et al. 2009), Belém {Utinga} (Pereira 1958), Marabá*, Novo Progresso {Cachimbo} (Pereira 1958), Parauapebas {Serra Norte, Carajás}*, Santarém (Walker 1854a) {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*, Jarú*
143. <i>Pezaptera sordida</i> (Walker, 1856)	AM, Manaus (Zerny 1931), Parintins {Villa Nova} (Hampson 1898), Tefé {Ega} (Walker [1865]); PA, Santarém (Walker 1856)
144. <i>Phaeosphesia opaca</i> (Walker, 1856)	MA, Açailândia*; PA, [Belém] {Pará, Valley of the Amazon [river]} (Walker 1856), Santarém*
145. <i>Pheia admirabilis</i> Bryk, 1953	AM, [São Gabriel da Cachoeira] {Taracua} (Bryk, 1953)
146. <i>Pheia albisigna</i> (Walker, 1854)	AM, Tefé (Hampson 1898); MT, Sinop*; PA, Novo Progresso {Cachimbo}*, Parauapebas {Serra Norte, Carajás}*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*, Porto Velho*; [RR], [Caracarái] {Rio Branco} (Bryk 1953)
147. <i>Pheia elegans</i> (Druce, 1884) <sup>AMZ</sup>	PA, Novo Progresso {Cachimbo}*

Continuation Table 1.

Species	Record
148. <i>Pheia gaudens</i> (Walker, 1856)	PA, [Belém] (Walker 1856), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
149. <i>Pheia haemapleura</i> Hampson, 1914	PA (Hagmann 1938)
150. <i>Pheia haematosticta</i> Jones, 1908 <sup>AMZ</sup>	PA, Novo Progresso {Cachimbo}* , Santarém*; RO, Cacaúlândia*
151. <i>Pheia serpens</i> Kaye, 1918	AM, Itacoatiara {Serpa} (Kaye 1918); PA, Santarém {Taperinha} (Zerny 1931)
152. <i>Pheia sperans</i> (Walker, 1856)	[AM], Valley of the Amazon [river] (Walker 1856)
153. <i>Pheia taperinhae</i> Dognin, 1923	PA, Santarém {Taperinha} (Dognin 1923)
154. <i>Pheia utica</i> (Druce, 1889)	PA, Santarém (Valente et al. 2018)
155. <i>Phoenicoprocta corvica</i> (Dognin, 1910)	PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém (Valente et al. 2018), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
156. <i>Phoenicoprocta haemorrhoidalis</i> (Fabricius, 1775) <sup>AMZ</sup>	MA, Açailândia*; PA, Capitão Poço*
157. <i>Phoenicoprocta insperata</i> (Walker, 1856)	AM, [Atalaia do Norte] {Braga on Rio Javary} (Butler 1877), Santa Isabel do Rio Negro*, São Paulo de Olivença*; PA, [Belém] (Walker 1856), Santarém {Taperinha} (Zerny 1931)
158. <i>Phoenicoprocta sanguinea</i> (Walker, 1854) <sup>NEW</sup>	AM, Manaus*, São Paulo de Olivença*; AP, Serra do Navio*; PA, Belém*, Santarém*
159. <i>Phoenicoprocta vacillans</i> (Walker, 1856)	AM, Benjamin Constant*, Manaus*, [São Gabriel da Cachoeira] {Taracua} (Bryk, 1953), São Paulo de Olivença*, Tefé*; AP, Serra do Navio*; PA, Altamira {Monte Santo} (Teston & Delfina 2010), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha and Moju} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Porto Velho*
160. <i>Pleurosoma angustata</i> (Moeschler, 1878)	PA, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
161. <i>Poecilosoma chrysis</i> Hübner, 1823	AM, Tefé {Ega} (Walker [1865]); PA, Belém*, Breves (Zerny 1931), Marabá*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaúlândia*
162. <i>Poecilosoma eone</i> (Hübner, 1827)	AM, [Eirunepé] {Juruá river, São Felipe} (Zerny 1931), Itacoatiara (Zerny 1931), Manaus (Zerny 1931), Tefé {Ega} (Walker 1854a); PA, Altamira {51°BIS} (Teston et al. 2012) and {Serra do Pardo National Park} (Teston & Correa 2015), Santarém (Walker 1854a) {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
163. <i>Poecilosoma marginatum</i> (Walker, 1856)	AM, Valley of the Amazon [river] (Walker 1856)
164. <i>Poecilosoma nigerrima</i> (Walker, [1865])	AM, Tefé {Ega} (Walker [1865])
165. <i>Poliopastea anthracina</i> (Klages, 1906)	PA, Altamira {Monte Santo} (Delfina & Teston 2013), Belém*, Belterra {National Forest of Tapajós} (Freitas 2014), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
166. <i>Poliopastea coelebs</i> Bryk, 1953	AM, Amazonas [river] (Bryk 1953)
167. <i>Poliopastea errans</i> (Hübner, [1819])	PA, [Belém] (Butler 1876)
168. <i>Poliopastea esmeralda</i> (Butler, 1876)	AM, Tefé {Ega} (Butler 1876)
169. <i>Poliopastea indistincta</i> (Butler, 1876)	AM, Alto Amazonas (Hagmann 1938); PA, [Belém] (Butler 1876); RO, Cacaúlândia*
170. <i>Poliopastea plumbea</i> Hampson, 1898	AM, Parintins (Hampson 1898), Tabatinga (Hampson 1898); PA, Altamira {Monte Santo} (Teston & Delfina 2010) and {Serra do Pardo National Park} (Teston & Correa 2015), Belém*, Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
171. <i>Poliopastea vittata</i> (Walker, 1854)	PA, [Belém] (Walker 1854a), Santarém {Taperinha and Moju} (Zerny 1931)
172. <i>Pompiliodes aliena</i> (Walker, 1854)	AM, Manaus (Hampson 1898), Tefé {Ega} (Hampson 1898); MA, Açailândia*; PA, [Belém] (Walker 1854a), Santarém {Taperinha} (Zerny 1931); RO, Cacaúlândia*
173. <i>Pompiliodes postica</i> (Walker, 1856)	PA, Santarém (Walker 1856) {Taperinha} (Zerny 1931)

Continuation Table 1.

Species	Record
174. <i>Pompiliodes tenebrosa</i> (Walker, 1854)	PA, [Belém] (Walker 1854a)
175. <i>Pompilopsis tarsalis</i> (Walker, 1854)	AM, Manicoré {Rio Madeira} (Zerny 1931); PA, [Belém] (Walker 1854a), Santarém {Taperinha} (Zerny 1931)
176. <i>Pseudomya picta</i> Schaus, 1894	PA, São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
177. <i>Psoloptera leucosticta</i> (Hubner, 1827)	MA, Açailândia*; PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), [Belém] (Hampson 1898), Marabá*, Santarém (Hampson 1898), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaulândia*, Jarú*
178. <i>Psoloptera leucotmemica</i> Bryk, 1953	AM, [Autazes] {Rio Autaz, Cururú} (Bryk 1953)
179. <i>Psoloptera melini</i> Bryk, 1953	AM, Manaus (Bryk 1953)
180. <i>Psoloptera thoracica</i> (Walker, 1854)	AM, Benjamin Constant*, Borba*, São Paulo de Olivença*, Tefé {Ega} (Walker 1854a); RO, Porto Velho*
181. <i>Rhynchopyga discalpa</i> Kaye, 1918	PA, Santarém (Valente et al. 2018)
182. <i>Rhynchopyga meisteri</i> (Berg, 1883)	PA, Tapajós [river] (Hampson 1898)
183. <i>Rhynchopyga pimpinella</i> Bryk, 1953	AM, Manaus (Bryk 1953)
184. <i>Sarosa acutior</i> (R. Felder, 1869)	AM, Amazonas [river] (R. Felder 1869), Benjamin Constant*; MA, Açailândia*; PA, Belterra {National Forest of Tapajós} (Freitas 2014), Capitão Poço*, Marabá*, Novo Progresso {Cachimbo}*, Santarém {Taperinha and Moju} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); RO, Cacaulândia*, Jarú*, Porto Velho*
185. <i>Sarosa ignicornis</i> Hampson, 1914	PA, Santarém {Taperinha} (Zerny 1931)
186. <i>Sarosa mora</i> Schaus, 1911 <sup>NEW</sup>	AP, Porto Grande*, Serra do Navio*
187. <i>Sarosa pompilina</i> Butler, 1876 <sup>AMZ</sup>	AM, Benjamin Constant*; PA, Novo Progresso {Cachimbo}*
188. <i>Saurita attenuata</i> Hampson, 1905	PA, Altamira {Serra do Pardo National Park} (Teston & Correa 2015), [Oriximiná] {Rio Trombetas, Rapid of Porteira} (Butler 1877), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)
189. <i>Saurita biradiata</i> (R. Felder, 1869)	AM, Amazonas [river] (R. Felder 1869)
190. <i>Saurita cassandra</i> (Linnaeus, 1758)	AM, Benjamin Constant*, Fonte Boa*; PA, Altamira {Monte Santo} (Teston & Delfina 2010), [Belém] (Zerny 1931), Belterra {National Forest of Tapajós} (Freitas 2014), Benevides*, Breves*, Capitão Poço*, Gurupá*, Marabá*, Ourém*, Santarém (Valente et al. 2018), Tucuruí*; RO, Porto Velho*
191. <i>Saurita concisa</i> (Walker, 1854)	PA, [Belém] (Walker 1854a), Belterra {National Forest of Tapajós} (Freitas 2014), Santarém (Valente et al. 2018)
192. <i>Saurita concisina</i> Bryk, 1953	AM, Rio Purús (Bryk 1953)
193. <i>Saurita cryptoleuca</i> (Walker, 1854)	PA, [Belém] (Walker 1854a)
194. <i>Saurita fumosa</i> (Schaus, 1912) <sup>NEW</sup>	AM, Benjamin Constant*
195. <i>Saurita fusca</i> Dognin, 1923	PA, [Vitória do Xingu] {Ponte Nova, Rio Xingu} (Dognin 1923)
196. <i>Saurita intricata</i> (Walker, 1854) <sup>NEW</sup>	RO, Cacaulândia*
197. <i>Saurita lacteata</i> (Butler, 1877)	AM, Rio Jutai (Butler 1877); PA, Santarém {Taperinha} (Zerny 1931)
198. <i>Saurita lasiphlebia</i> Dognin, 1906	AP, Serra do Navio*; PA, Bragança (Oberthür 1912)
199. <i>Saurita melanifera</i> Kaye, 1911	AM, Rio Purús (Bryk 1953)
200. <i>Saurita pebasa</i> (Kaye, 1918)	PA, Belterra {National Forest of Tapajós} (Freitas 2014)
201. <i>Saurita sericea</i> (Herrich-Schäffer, [1854]) <sup>AMZ</sup>	MA, Açailândia*
202. <i>Saurita temenus</i> (Stoll, [1781])	AM, Manaus (Bryk 1953), Rio Purús (Bryk 1953), São Gabriel [da Cachoeira] (Bryk 1953); PA, Altamira {Monte Santo} (Teston & Delfina 2010), [Belém] (Walker 1854a), Santarém {Taperinha} (Zerny 1931), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015); [RR], [Caracarái] {Rio B[r]anco} (Bryk 1953)
203. <i>Saurita tipulina</i> (Hübner, [1812])	PA, Altamira {51°BIS} (Teston et al. 2012) {Monte Santo} (Delfina & Teston 2013), [Belém] (Walker 1854a), Belterra {National Forest of Tapajós} (Freitas 2014), São Félix do Xingu {Serra do Pardo National Park} (Teston & Correa 2015)



Continuation Table 1.

Species	Record
204. <i>Saurita triangulifera</i> (Druce, 1898)	[AM], Amazons (Druce 1898a)
205. <i>Saurita tristissima</i> (Perty, 1834)	AM, [Boa Vista do Ramos] {Massauari} (Zerny 1931); PA, [Belém] (Walker 1854a)
206. <i>Saurita vindonissa</i> (Druce, 1883) <sup>AMZ</sup>	RO, Cacaulândia*, Jarú*
207. <i>Sauritina dubiosa</i> Schaus, 1905	PA, [Belém] (Hampson 1914)
208. <i>Sphecopops artacta</i> (Walker, [1865])	AM, Tefê {Ega} (Walker [1865])
209. <i>Sphecosoma adominalis</i> Schaus, 1905	MA, Açailândia*; PA (Hagmann 1938), Belém*, Santarém*; RO, Cacaulândia*
210. <i>Sphecosoma albipalpe</i> Draudt, 1915	AM, Amazonas (Draudt 1915)
211. <i>Sphecosoma cognata</i> (Walker, 1856)	AM, Valley of the Amazon [river] (Walker 1856)
212. <i>Sphecosoma mathani</i> Rothschild, 1911	AM, Tefê (Rothschild 1911)
213. <i>Sphecosoma melissa</i> Schaus, 1896 <sup>AMZ</sup>	PA, Santarém*
214. <i>Sphecosoma nigriceps</i> Hampson, 1903 <sup>NEW</sup>	AM, Manaus*; PA, Capitão Poço*
215. <i>Sphecosoma rufipes</i> Rothschild, 1911 <sup>NEW</sup>	AM, Manaus*; PA, Belém*; RO, Cacaulândia*
216. <i>Sphecosoma testacea</i> (Walker, 1854)	AM, Itacoatiara*, Santa Isabel do Rio Negro*; PA, [Belém] (Hampson 1898), Santarém*
217. <i>Syntomeida austera</i> Dognin, 1902 <sup>NEW</sup>	PA, Belém*, Oriximiná {Cuminá river}*; RO, Cacaulândia*
218. <i>Syntomeida melanthus</i> (Cramer, [1779])	PA (Hagmann 1938), Belém*
219. <i>Syntomeida sintomoides</i> (Boisduval, 1836)	PA, [Igarapé-Miri] {estuary of Tocantins [river]} (Zerny 1931)

<sup>AMZ</sup> Species recorded for states within the Amazon biome by Ferro and Diniz (2010), but without precise location and biome information, and Amazon biome by Ferro and Diniz (2007). So these species are new records for the municipalities. <sup>NEW</sup> New record for the Brazilian Amazon.

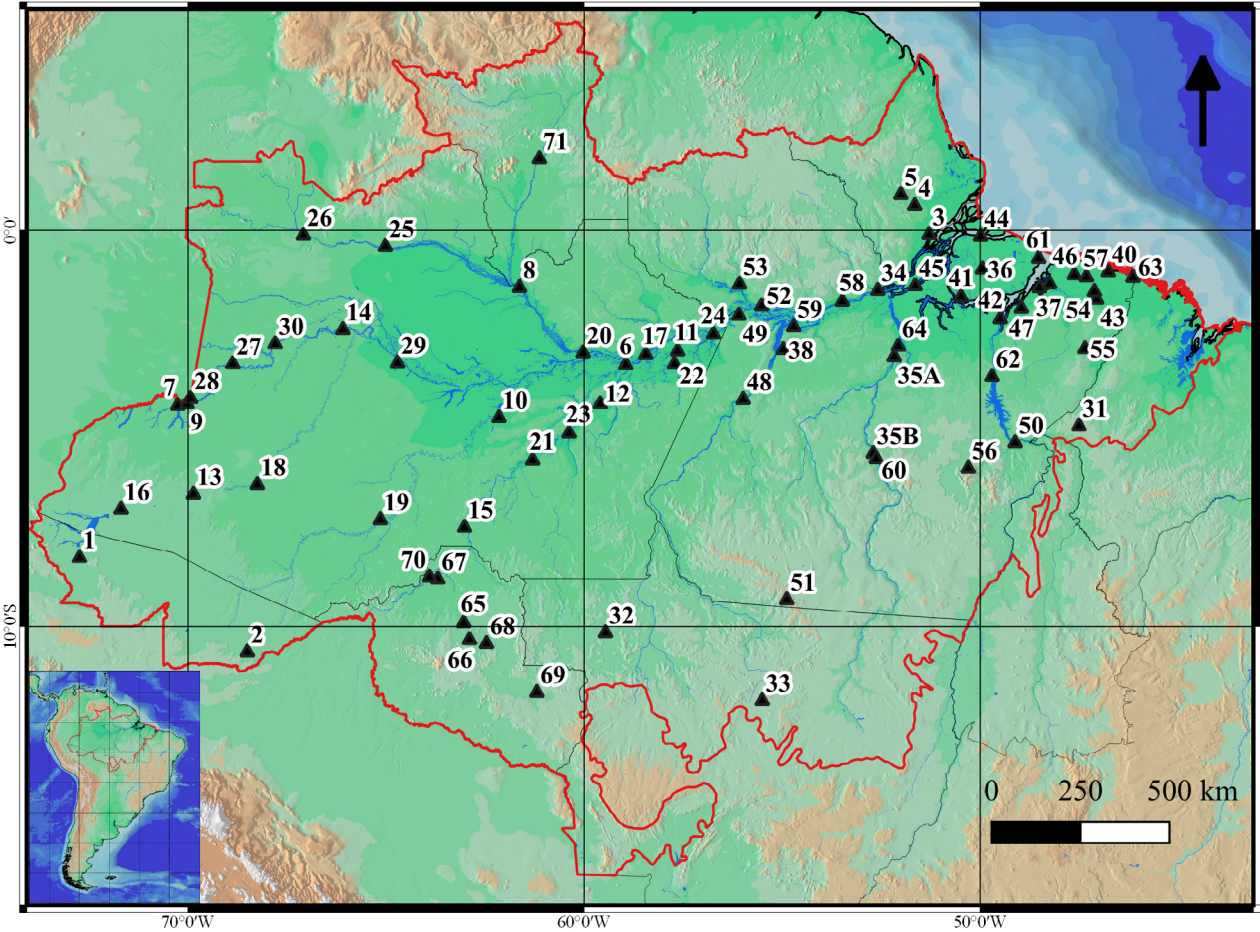


Figure 1. Geographic distribution of Euchromiina species records in the Brazillian Amazon. The numbers refer to the municipalities of Table 2.

**Table 2.** Geographic coordinates of municipalities and richness of the Euchromiina species (Erebidae, Arctiinae, Arctini) in the Brazilian Amazon Biome.

Nº	State	Municipality	Richness	Latitude	Longitude
1	AC	Porto Walter	1	08°16'07"S	72°44'37"W
2	AC	Xapuri	1	10°39'06"S	68°30'15"W
3	AP	Mazagão	1	00°06'55"S	51°17'21"W
4	AP	Porto Grande	1	00°37'01"N	51°38'60"W
5	AP	Serra do Navio	16	00°53'45"N	52°00'07"W
6	AM	Autazes	3	03°24'02"S	58°57'00"W
7	AM	Atalaia do Norte	1	04°25'46"S	70°15'41"W
8	AM	Barcelos	4	01°27'01"S	61°37'59"W
9	AM	Benjamin Constant	19	04°22'60"S	70°01'52"W
10	AM	Beruri	1	04°44'09"S	62°09'01"W
11	AM	Boa Vista do Ramos	3	03°03'19"S	57°38'20"W
12	AM	Borba	5	04°23'17"S	59°35'37"W
13	AM	Eirunepé	7	06°40'01"S	69°52'00"W
14	AM	Fonte Boa	6	02°30'51"S	66°05'30"W
15	AM	Humaitá	4	07°30'23"S	63°01'14"W
16	AM	Ipixuna	1	07°03'03"S	71°41'41"W
17	AM	Itacoatiara	5	03°08'36"S	58°26'39"W
18	AM	Itamarati	1	06°25'31"S	68°15'12"W
19	AM	Lábrea	1	07°18'51"S	65°08'40"W
20	AM	Manaus	28	03°06'07"S	60°01'30"W
21	AM	Manicoré	9	05°48'34"S	61°18'00"W
22	AM	Maués	4	03°23'01"S	57°43'07"W
23	AM	Novo Aripuanã	1	05°07'15"S	60°22'47"W
24	AM	Parintins	12	02°37'42"S	56°44'08"W
25	AM	Santa Isabel do Rio Negro	7	00°24'51"S	65°01'08"W
26	AM	São Gabriel da Cachoeira	5	00°07'50"S	67°05'20"W
27	AM	São Paulo de Olivença	20	03°22'42"S	68°52'20"W
28	AM	Tabatinga	2	04°15'10"S	69°56'17"W
29	AM	Tefé	39	03°21'16"S	64°42'40"W
30	AM	Tonantins	1	02°52'24"S	67°48'08"W
31	MA	Açailândia	33	04°56'49"S	47°30'17"W
32	MT	Aripuanã	6	10°10'01"S	59°27'33"W
33	MT	Sinop	9	11°52'51"S	55°30'08"W
34	PA	Almeirim	3	01°31'24"S	52°34'54"W
35A	PA	Altamira <sup>†</sup>	29	03°11'55"S	52°10'15"W
35B	PA	Altamira {Serra do Pardo National Park} <sup>†</sup>	13	05°38'21"S	52°41'52"W
36	PA	Anajás	1	00°59'13"S	49°56'23"W
37	PA	Belém	74	01°27'21"S	48°30'15"W
38	PA	Belterra {National Forest of Tapajós} <sup>†</sup>	28	03°01'05"S	54°58'10"W
39	PA	Benevides	1	01°21'42"S	48°14'40"W
40	PA	Bragança	2	01°03'13"S	46°45'56"W
41	PA	Breves	3	01°40'56"S	50°28'49"W
42	PA	Cametá	1	02°14'40"S	49°29'45"W
43	PA	Capitão Poço	11	01°44'48"S	47°03'33"W
44	PA	Chaves	1	00°09'36"S	49°59'18"W

Continuation Table 2.

Nº	State	Municipality	Richness	Latitude	Longitude
45	PA	Gurupá	1	01°24'18"S	51°38'23"W
46	PA	Igarapé Açú	1	01°07'45"S	47°37'11"W
47	PA	Igarapé-Miri	1	01°58'31"S	48°57'34"W
48	PA	Itaituba	5	04°16'35"S	55°59'01"W
49	PA	Juruti	1	02°09'08"S	56°05'31"W
50	PA	Marabá	9	05°22'07"S	49°07'04"W
51	PA	Novo Progresso {Cachimbo}	17	09°19'60"S	54°52'59"W
52	PA	Óbidos	15	01°55'04"S	55°31'04"W
53	PA	Oriximiná	5	01°21'60"S	56°04'44"W
54	PA	Ourém	2	01°33'07"S	47°06'52"W
55	PA	Paragominas	1	02°59'45"S	47°21'10"W
56	PA	Parauapebas	14	06°00'56"S	50°17'51"W
57	PA	Peixe Boi	1	01°10'60"S	47°18'59"W
58	PA	Prainha	4	01°47'60"S	53°28'47"W
59	PA	Santarém	96	02°26'36"S	54°42'29"W
60	PA	São Félix do Xingu {Serra do Pardo National Park} <sup>†</sup>	47	05°46'26"S	52°37'13"W
61	PA	Soure	1	00°43'01"S	48°31'24"W
62	PA	Tucuruí	2	03°42'01"S	49°42'00"W
63	PA	Viseu	2	01°11'49"S	46°08'23"W
64	PA	Vitória do Xingu	1	02°55'60"S	52°03'59"W
65	RO	Ariquemes	1	09°54'48"S	63°02'26"W
66	RO	Cacaulândia	39	10°20'21"S	62°53'43"W
67	RO	Candeias do Jamari	2	08°48'35"S	63°41'44"W
68	RO	Jarú	6	10°26'20"S	62°27'58"W
69	RO	Pimenta Bueno	1	11°40'21"S	61°11'37"W
70	RO	Porto Velho	15	08°45'43"S	63°54'13"W
71	RR	Caracarái	4	01°47'60"N	61°07'50"W

Geographic coordinates of municipality marked with <sup>†</sup> are the citations referred, other obtained by Google Earth or Geo Loc tool (see Materials and Methods).

The Euchromiina richness of Santarém and Belém were the largest recorded in Brazilian sites. For example, in Salesópolis was recorded 47 Euchromiina species (Ferro & Diniz 2007) and in Joinville 42 species (Ferro et al. 2012). These two last sites were located in the Atlantic Forest biome and were intensively sampled. In the Cerrado sites, were recorded a much lower number of Euchromiina species, ranging from 11 (Scherrer et al. 2013) to 30 (Moreno & Ferro 2016). Moreover, the number of Euchromiina species of a single Amazon site (Santarém, 96) was higher than the entire Cerrado Euchromiina fauna (Ferro et al. 2010). It may indicate that this taxon is more diverse in rain-forest than in xeric environments. Studies with Arctiini in the Altamira (Pará Brazilian State) showed that there are 2 times more Euchromiina species in Amazonian forest sites (64, Teston & Correa 2015) than in Cerrado sites (32, Valente et al. 2018) and 4 times more Euchromiina species in Amazonian forest sites than altered sites (pasture and orchard) (16, Delfina & Teston 2013). Martins et al. (2017) also obtained 2.5 times more butterfly species in sites of Amazonian forest than in Cerrado sites in the Maranhão Brazilian State.

Teston et al. (2019) have reported 847 Arctiinae species in the Brazilian Amazon. Due to the new records obtained in our study (13), there was an increase in richness for the biome, generating a total of 860 tiger moth Amazon species. As observed for the subtribes Phaegopterina (Teston & Ferro 2016a), Pericopina (Teston & Ferro 2016b) and Ctenuchina (Teston et al. 2019), the number of Amazonian Euchromiina species is underestimated because the fauna was never sampled in the vast majority of the biome (less than 10% of the municipalities were sampled). Even the sites already sampled need to be studied in the long-term (at least 1 year of sampling) because it is known that tiger moths respond to climate/seasonal changes (Kitching et al. 2000, Hilt et al. 2007, Scherrer et al. 2013, Ferro et al. 2014) and that Lepidoptera richness is higher in long-term surveys than in short-term surveys (Ferro & Diniz 2007, Moreno & Ferro 2016, Martins et al. 2017). Moreover, studies that sample Amazonian Lepidoptera in different types and strata of vegetation are rare. However, it is known that tiger moths respond to vegetation changes (Kitching et al. 2000, Ferro & Diniz 2007, Ferro & Romanowski 2012) and some genera of Euchromiina (*Macrocneme*

and *Poliopastea*) are canopy flyers (Brehm 2009). In order to suggest efficient conservation policies for the Amazon Arctiinae fauna, it is urgent to intensify the sampling effort in this biome, both spatially and temporally. Finally, in addition to reduce species distribution gaps, it is also necessary to invest in studies on the taxonomy of *Euchromiina* in order to decrease the bias of the Linnean shortfall (Whittaker et al. 2005) on diversity patterns since the taxonomy of this taxon is still complicated: there are many descriptions based on only one individual, many species to be described, and many synonyms to elucidate.

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## Author Contributions

José Augusto Teston: Substantial contribution in the concept and design of the study.

Viviane Gianluppi Ferro: Substantial contribution in the concept and design of the study.

## Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

## Data availability

The data are deposited in the respective collections mentioned in the Material and Methods.

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