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Butterflies richness (Lepidoptera, Papilionoidea) from Ilha Grande National Park, Paraná, Brazil

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Abstract
Insects of the Lepidoptera order play an important role in ecosystems, due to the various interactions established with plants, such as pollination, and with other animals. In addition, they constitute important models for biogeography and insect/plant interactions studies, which make them essential tools for the natural reserve management. Thus, the present study aimed to carry out an inventory of butterflies at Ilha Grande National Park, an important Conservation Unit in the Paraná State, South of Brazil. The sampling was carried out from February to November 2019, comprising three collection campaigns, during five days each and field effort of six people, totaling about 15 days and 60 hours, on six islands in the park. There were collected 147 individuals of 63 species, distributed in 6 families, with Nymphalidae presenting the greatest richness (45 species) and greatest abundance (93 individuals).

Keywords: Butterflies, richness, Ilha Grande national park

Introduction
Invertebrates constitute a functionally significant portion of terrestrial and aquatic biodiversity, being responsible for ecological processes essentials for the Earth's ecosystems maintenance [1, 2, 3]. However, despite their high diversity and ecological importance, these organisms have been neglected by biodiversity conservation policies [4], even though that aquatic invertebrates have a long history of use as water quality bioindicators [1].
Invertebrates can be good environmental and ecological bioindicators, as they are small, they are very sensitive to any environmental change, thus presenting quick responses and providing an indication of subtle changes in local environmental conditions [5]. In addition, they have short generation cycles, which results in rapid numerical responses to environmental variation [6]. However, the lack of systematic and biological knowledge about organisms with enormous richness of habits and habitats is still a major challenge for the biodiversity conservation [1, 7]. In this context, the relevance of carrying out fauna's inventory, especially of invertebrates such as insects, is evident.
The Lepidoptera order includes insects known as butterflies and moths [8]. These insects play an important role in ecosystems, due to the various interactions established with plants, such as pollination, and with other animals, as they represent important links in a lot of trophic networks [9]. There are 12,735 described species in Brazil, 3,479 of which are butterflies [10], gathered in the Papilionoidea superfamily, with seven families: Hesperiidae, Lycaenidae, Nymphalidae, Papilionidade, Pieridae, Riodinidae and Hedylidae [11].
In addition, they constitute important models for biogeography and insect / plant interactions studies, which make them essential tools for the natural reserve management [12]. The identification of these taxa is, therefore, fundamental for creation of Conservation Units, as well as for the management of the populations and habitats protected by these units [13].
The review of the specialized literature showed the existence of lepidopterans inventories in several regions in the Paraná State [14, 15, 16]. Inventories have also been found in urban parks [17] and in Conservation Units such as the Vila Velha State Park in Ponta Grossa [18] and at the Caiuá Ecological Station, Diamante do Norte Municipality [19], as well as endangered species records [20, 21, 22].

Considering all these informations about the butterfly richness in the Paraná State, and seeking to complement them, the present study aimed to carry out an inventory of butterflies at Ilha Grande National Park, an important conservation unit in the Paraná State, Brazil Southern.

Materials and Methods

The Ilha Grande National Park (53°41'09,2"W - 23°16'32"S and 54°16'21,7"W - 24°04'11,5"S), located on the border between Paraná and Mato Grosso do Sul States, encompasses a set of islands, islets and a floodplain stretch on the Paraná river. Historically, there was another conservation unit in its surrounding area, the Sete Quedas National Park, created in 1961 and extinguished in the 1980s, with the construction of Itaipu Hydroelectric and the consequent water reservoir formation [23]. Still in the same decade, the Ilha Grande Hydroelectric would also be built on the Paraná River stretch upstream of the Itaipu reservoir. This would eliminate the last stretch of the Paraná river free of dams. However, this construction was canceled due to popular and political pressure. From there, Municipal Environmental Protection Areas were created, and in 1994, the Ilha Grande Ecological Station, which subsequently, on September 30, 1997, gave rise to the Ilha Grande National Park [23].

The phytophysiognomy of the study area includes Seasonal Semideciduous Forest and Ombrophilous Forest, belonging to the Atlantic Forest domain, being considered at the macro-regional level as a transitional ecotone between Paraná's Semideciduous Seasonal Forest and Mato Grosso do Sul State's Cerrado (Brazilian Savannah) [24, 25, 26]. It is estimated that in the last century, 83.41% of the Paraná State territory had forest cover [27]. However, this forest cover had been losing space to different human activities, such as intensive agriculture and livestock. The Seasonal Semideciduous Forest, predominant phytophysiognomy in the study area, also suffered with the advance of areas with secondary vegetation and it is estimated that less than 5% of its total area remains today [28]. Its forest formation is characterized by arboreal elements (perennial or deciduous) that reach up to 30 meters in height, without formation of a continuous top cover, with thick trunks that have a long stem, usually ended by broad upward shearing [28].

The sampling of biological material was carried out from February to November 2019, comprising three collection campaigns, during five days each and field effort of six people, totaling about 15 days and 60 hours, on six islands in the park (Figure 01). Between the 1970s and 1995, human activities on these islands resulted in the transformation of native vegetation cover into pasture and cultivation areas. In 1997, with the creation of the Ilha Grande National Park, the process of eviction the islands, due to the construction project for the Ilha Grande hydroelectric, was intensified. Although this project was interrupted, this process gave rise to a series of socio-environmental conflicts. Thus, only from the 2000s, with the consolidation of the park's eviction and conservation actions, there was the beginning of the degraded areas natural regeneration [29, 30].

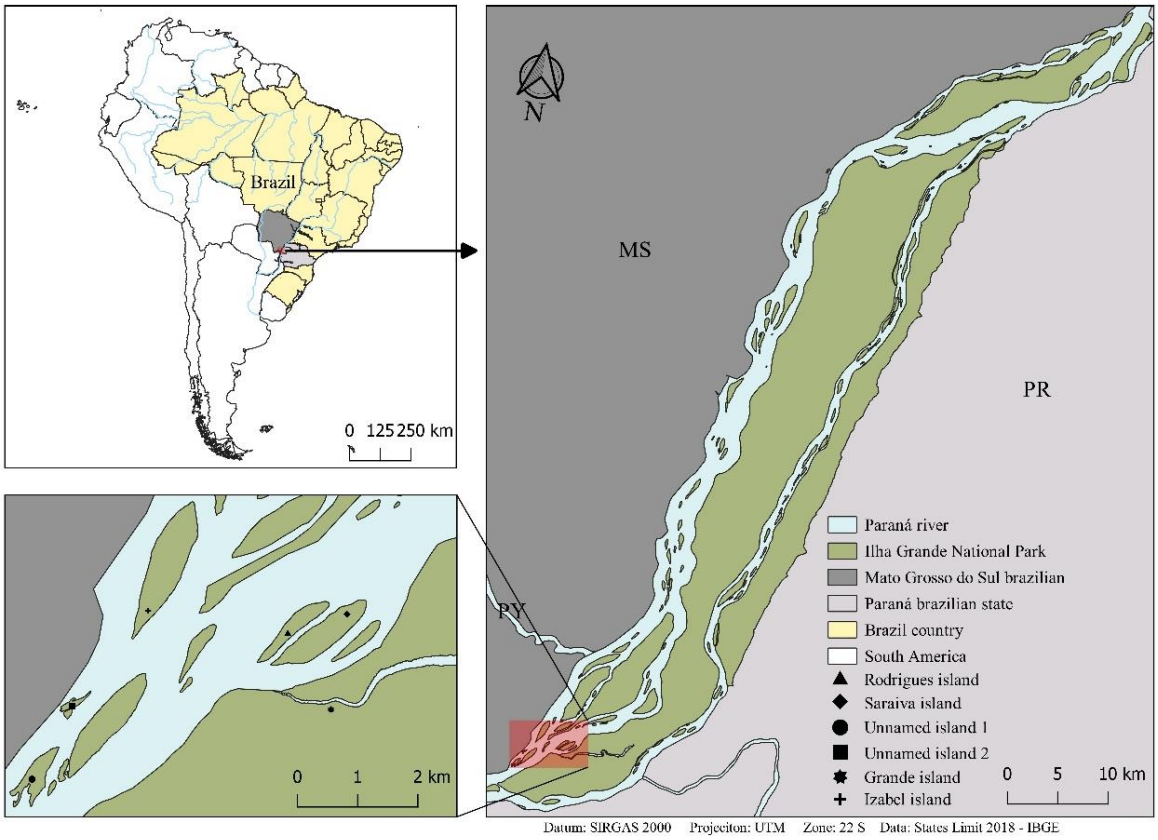

Fig 1: Islands where the collection campaigns were carried out, Ilha Grande National Park, Paraná, Southern Brazil.

The collection of individuals had been made through an active search with an entomological network. After collection, these were sacrificed by chest pressure and individually stored in entomological envelopes with the location, date and collector informations, for future identification. The specimens were included in the UNICAMP Butterfly Laboratory collection, São Paulo State and in the IFSULDEMINAS Zoology laboratory, Campus Inconfidentes, Minas Gerais State. The study was authorized by ICMBio through the license SISBIO 65047-1.

Five non-parametric richness estimators were used (*ICE*, *Chao 2*, *Jacknife 1*, *Jacknife 2* e *Boostrap*) to calculate the park's lepidopters richness, which were chosen because they are considered efficient to estimate the richness [31] and because they are based on the species presence. Therefore, estimators based on abundance were discarded. All calculations were performed using the EstimateS 9.1.0 software [32], with data submitted to 1000 randomizations. In addition, collection efficiency was measured, which consists of comparing observed and expected richness, whose ratio is calculated by estimators that indicate whether sampling was efficient [33].

Results and Discussion

There had been collected 147 individuals of 63 species, distributed in 6 families (Table 1) with Nymphalidae presenting the greatest richness (45 species) and greatest abundance (93 individuals) while the Lycaenidae family had the lowest richness (1 species) and the lowest abundance (2 individuals). The richness average estimates ranged from 67 to 166 species (Table 2 and Figure 2), the lowest collection efficiency (Table 2) was measured according to the ICE estimator (37%) and the highest was Bootstrap (94%).

The low species richness observed may be a collection methodology reflection, since in the present study, attractive traps Someren-Rydon model were not used [34], which are more efficient for catching frugivorous canopy butterflies, Nymphalidae family [35, 36, 37]. The collection efficiency in this study ranged from 37% to 68% (Table 2), which indicates that it was low, since for a sampling to be considered efficient the values must be greater than 70% [33].

The richness estimators calculated between 67 and 166 species in the collection site, although the extreme values presented by the *ICE* and *Bootstrap* estimators are common in the scientific literature. The *ICE* estimator tends to overestimate richness, especially in studies with smaller samples and the *Bootstrap* estimator has a tendency to underestimate the species richness [38]. Another factor that may have contributed to the low richness of this study is the presence of a high canopy in the sampled islands. This may have promoted restrictions for the active search methodology, so that the species sampling may have been insufficient.

Table 1: Butterfly families and species (number of individuals per species) collected in the Ilha Grande National Park, Paraná, Southern Brazil

Nymphalidae
Actinote sp. (1)
Adelpha iphicleola leucates (Fruhstorfer, 1915) (1)
Aeria olena (Weymer, 1875) (1)
Agraulis vanillae maculosa (Stichel, (1908) (1)
Anartia amathea roeselia (Eschscholtz, 1821) (1)
Anartia jatrophae jatrophae (Linnaeus, 1763) (1)
Biblis hyperia nectanabis (Fruhstorfer, 1909) (3)
Caligo illioneus (Cramer, 1775) (1)
Charaxinae sp. (1)
Chlosyne lacinia saundersi (Doubleday, 1847) (1)
Colobura dirce dirce (Linnaeus, 1758) (1)
Dryas iulia alcionea (Cramer, 1779) (2)
Danaus eresimus plexaure (Godart, 1819) (1)
Dryadula phaetusa (Linnaeus, 1758) (2)
Dynamine coenus coenus (Fabricius, 1793) (2)
Epityches eupompe (Geyer, 1832) (1)
Eryphanis automedon amphimedon (C. Felder & R. Felder, 1867) (1)
Euptoieta hegesia meridiania (Stichel, 1938) (1)
Fountainea ryphea phidile (Geyer, 1837) (4)
Hamadryas februa februa (Hübner, 1823) (1)
Heliconius erato phyllis (Fabricius, 1775) (9)
Heliconius ethilla narcaea (Godart, 1819) (1)
Hermeuptychia sp. (1)
Historis odius dios (Lamas, 1995) (1)
Hypna clytemnestra huebneri (Butler, 1866) (1)
Junonia evarete evarete (Cramer, 1779) (2)
Mcclungia cymo salonina (Hewitson, 1855) (1)
Mechanitis lysimnia lysimnia (Fabricius, 1793) (2)
Myscelia orsis (Drury, 1782) (1)
Nica flavilla flavilla (Godart, 1824) (1)
Paryphthimoides poltys (Prittwitz, 1865) (1)
Paulogramma pygas (Godart, 1824) (1)
Pareuptychia ocirrhoe interjecta (R.F. d'Almeida, 1952) (2)
Pareuptychia summandosa (Gosse, 1880) (1)
Posttaygetis penelea (Cramer, 1777) (1)
Pseudodebis ypthima (Hübner, 1821) (1)
Sais rosalia (Cramer, 1779) (7)
Siproeta stelenes meridionalis (Fruhstorfer, 1909) (1)
Taygetis laches marginata (Staudinger, [1887]) (2)
Tithorea harmonia pseudethra (Butler, 1873) (25)
Vanessa braziliensis (Moore, 1883) (1)
Zaretis strigosus (Gmelin, 1790) (1)

Pieridae
Dismorphia amphione astynome (Dalman, 1823) (1)
Eurema agave pallida (Chavannes, 1850) (4)
Enantia lina psamathe (Fabricius, 1793) (1)
Leucochimona icare matatha (Hewitson, 1873) (1)
Nymphidium sp. (1)
Phoebis argante argante (Fabricius, 1775) (1)
Pyrisitia leuce leuce (Boisduval, 1836) (14)

Papilionidae
Heraclides anchisiades capys (Hübner, [1809]) (1)
Parides neophilus eurybates (Gray, 1853) (12)
Parides panthonus castilhoi (d'Almeida, 1967) (3)
Papilioninae sp. (1)
Parides sp. (1)

Hesperiidae
Battus polydamas polydamas (Linnaeus, 1758 (3)
Fountainea ryphea phidile (Geyer, 1837) (4)
Urbanus sp. (1)
Xenophanes tryxus (Stoll, 1780) (1)
Mylon maimon (Fabricius, 1775) (1)

Lycaenidae
Theritas hemon (Cramer, 1775) (2)

Riodinidae
Arawacus separata (Lathy, 1926) (1)
*Nymphidium lisimon attenuatum (*Stichel, 1929) (1)

Table 2: Estimated average richness of butterfly species and collection efficiency, obtained based on five estimators: *ICE*, *Chao* 2, *Jacknife* 1, *Jacknife* 2 and *Bootstrap*, for the three sampled periods at Ilha Grande National Park, Paraná, Southern Brazil. The values indicate the estimated average richness ± standard deviation.

	Estimator				
	ICE	**Chao 2**	**Jackknife 1**	**Jackknife 2**	**Bootstrap**
ER[1]	166,55	130,33±3,13	103,36±4,35	97,27±4,03	67,03±5,09
CE[2]	37%	48%	61%	68%	94%

1 = Estimated richness; 2 = Collection efficiency.

The Nymphalidae family showed the highest richness species in the present study, probably because the species occur in different ecosystems [39] as well as due to their great diversity of food niches [40]. This pattern was also registered in other studies carried out on phytophysiognomies in the Atlantic Forest, Minas Gerais State's domain, in which Silva *et al*. (2007) [41], Andrade & Teixeira (2017) [42] and Vieira *et al*. (2020) [37] recorded 46,1%, 73,56% and 59% species of this family, respectively, in Semideciduous Forest. While Oliveira *et al*. (2018) [43] recorded 57% of nymphalids in Mixed Forest. However, in other studies conducted in the Paraná State the results were different. Garcia-Salik *et al*. (2014) [44] in a study conducted in Caiuá Ecological Station, Diamante do Norte municipality, extreme Northwest of Paraná, observed greater representativeness for the Hesperiidae family, followed by Nymphalidae, Lycaenidae, Riodinidae, Pieridae and Papilionidae in decreasing order of species abundance. This result was similar to other studies carried out in the Conservation Units in that state, such as in the Iguaçu National Park [45], in Yacutinga Private Reserve [46] and in Osununú Private Reserve [47].

The low similarity of the current study with the other studies in Paraná mentioned above, which reported Hesperiidae as the family of greatest richness and abundance, may reflect some factors, such as the fast flight and small size, typical of this family, which consequently makes its sampling difficult, unlike Nymphalidae, which has conspicuous and more easily registered species [16].

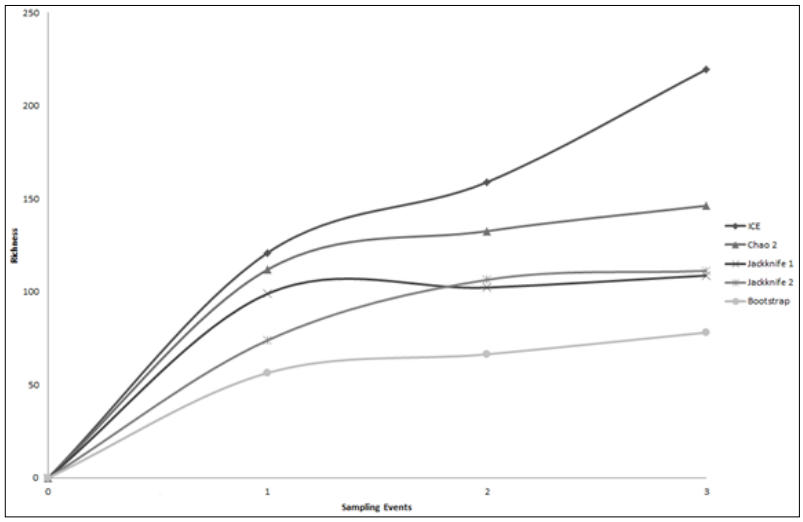

Fig 2: Butterfly richness estimates by sampled period (February, April and November 2019, numbers 1, 2 and 3, respectively) in the Ilha Grande National Park, Paraná.

The most abundant species in this study, with 25 individuals collected, was *Tithorea harmonia pseudethra* (Butler, 1873) (Figure 3), subfamily Danainae, Nymphalidae. *Tithorea harmonia pseudethra* is a common butterfly in several types of environments, such as river edges, riparian forests and also in the disturbed environments such as orchards and gardens [12], which would explain its abundance in the study area which has a recent history of landscape changes.

Fig 3: *Tithorea harmonia pseudethra* (Butler, 1873) collected at Ilha Grande National Park, Paraná, Brazil. Image: Ricardo Costa

Highlight for the subspecies *Parides panthonus castilhoi* (Papilionidae: Troidini) (Figure 4), which is critically endangered, but which was observed in this study. The species *Parides panthonus castilhoi* d'Almeida, 1967, is possibly the most critically endangered Brazilian papilionidae [22, 48, 49, 50]. An individual of this species was initially collected at Ilha Grande National Park [22], and three other specimens were recorded in the present study, and until then there was only one record for the São Paulo State and another for Mato Grosso do Sul State [22].

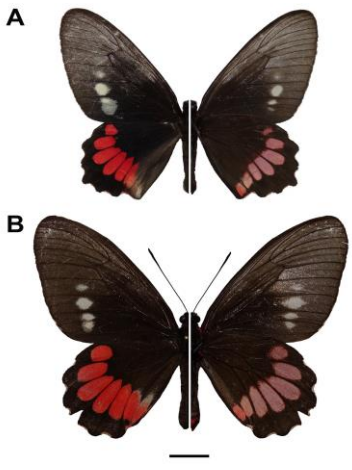

Fig 4: Butterfly *Parides panthonus castilho*i collected at Ilha Grande National Park; A, Male; B, Female. Left side = dorsal view; Right side = ventral view. Scale = 1 cm. Image: Augusto Henrique Batista Rosa

Conclusion

It is observed that studies on the butterfly diversity are essential in any region of Brazil, due to the great national biodiversity and the impact of the accelerated rhythm of natural environments anthropization. This is evident because we found the subspecies *Parides panthonus castilhoi* (Papilionidae: Troidini) in the present study. Therefore, further studies and greater sampling effort are suggested for the butterfly registration at Ilha Grande National Park.

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