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Nectarivore butterflies (Lepidoptera: Papilionoidea) from an Urban Forest Fragment in Manaus, Amazonas

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Abstract. Lepidoptera is a diverse and abundant group of insects, widely used in biodiversity and conservation studies, as it has great social appeal. From this perspective, this study was aimed to conduct a survey of flower – visiting butterfly species in Bosque da Ciência, at the Instituto Nacional de Pesquisas da Amazônia (INPA), an urban forest fragment open to public visitation in the State of Amazonas, Brazil. Thus, attractive plants for nectarivorous butterflies were observed, and the best location and conditions for viewing such diversity, as well as a guide for their identification was proposed. Forty-three species of flower – visiting butterflies were identified, among which *Anthoptus epictetus* (Fabricius), *Ascia monuste* (Linnaeus), *Anartia jatrophae* (Linnaeus), *Eurema albula* (Cramer) were observed to be more frequent. According to the species accumulation curve, the species richness of butterflies would be higher if greater collection effort were employed. The period of greatest flight activity and flower - visitation occurred from 9 am to 11 am. Of the six attractive plants identified, *Stachytarpheta cayennensis* (Rich.) and *Lantana camara* L. were the most visited, so we consider that these species are important for butterfly attractiveness. The butterflies of Bosque da Ciência are a representation of the nectarivores guild of the city of Manaus, Amazonas, and the establishment of an attractive butterfly garden in this place can contribute to the promotion of environmental education by stimulating the knowledge and awareness for biodiversity preservation.

Keywords: Amazon; biodiversity; floral visitors; forest fragments; species list.

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The urban forest fragments are remnants of natural vegetation surrounded by an urban matrix, sometimes transformed into woods and parks open to public visitation (Melo *et al.* 2011). They are considered essential environmental resources to guarantee the quality of life in urban centers (Kudo *et al.* 2016), important refuges for plants and animals not adapted to this matrix (Rodrigues *et al.* 1993; Silva *et al.* 2007), as some species of butterflies.

Butterflies, in particular, are animals used in environmental conservation campaigns, because they are flagship group due the great variability of colors and shapes (Freitas & Marini-Filho 2011), in addition to their relatively well-known taxonomy (Santos *et al.* 2016). They are diurnal insects and correspond to seven families of Lepidoptera: Hedylidae, Hesperiidae, Papilionidae, Pieridae, Lycaenidae, Riodinidae and Nymphalidae (Heikkilä *et al.* 2012).

Based on the feeding habit of adults, butterflies are divided into two guilds, nectarivores, when they feed on nectar or pollen from flowers, or frugivorous, when they feed on fermented fruit and plant sap (Brown Jr. & Freitas 1999). The nectarivores can be useful in education campaign, once they are frequent flower - visitors, it catches the attention of general public to such vegetation systems (Freitas *et al.* 2004) and bring grace and dynamism to the gardens, nurseries and woods, for having varied colors, designs and sizes easy to view (Freitas 2003).

Nectarivore butterflies are more attracted to colorful flowers, with edges of the corolla not too cut and smooth or flattened on the top of the petals, presence of much nectar, sometimes hidden in narrow tubes, which are the floral features for psychophilia (butterfly pollination) (Faegri & Pijl 1979). Most flowering plants visited by butterflies are common herbs in cities (Shapiro 2002).

The Bosque da Ciência at the Instituto Nacional de Pesquisas da Amazônia- INPA is an urban forest fragment in the State of Amazonas, Brazil, created for scientific dissemination and to raise awareness for preserving biodiversity (Oliveira & Oliveira 2009). Although Bosque da Ciência offers several tourist-educational attractions, it does not include certain elements of fauna within its visitation program, such as butterflies.

In order to highlight these elements of fauna and flora in the area of the Bosque da Ciência, and to offer subsidies for butterflies to be listed among the tourist-educational attractions offered by INPA, this study was aimed to record the flower – visiting butterfly species in Bosque da Ciência, and elaborate a guide to identify them in the field.

MATERIALS AND METHODS

Study area. The Bosque da Ciência corresponds to an area of approximately 13 hectares, located at Campus I of the Instituto Nacional de Pesquisas da Amazônia- INPA, in the East Zone of the city of Manaus (03º05'50" S and 59º59'13" W), State of Amazonas (Figure 1). The vegetation is typical of Amazon, ombrophylous "terra firme" forest (Luizão & Vasconcelos 2002). The climate is classified as 'Afi' under the Köppen system. The rainy season occurs from October/November to May, and the dry season is from July to September/October (Villar *et al.* 2008; Haghtalab *et al.* 2019)

Bosque da Ciência was created in 1994 to promote cultural and environmental education with a focus on biodiversity preservation (Bosque da Ciência 2020). It has fifteen tourist attractions, which are focal points for certain elements of both fauna and flora and in which the samplings were carried out: Ariranha, Peixe Boi, Casa da Ciência, Loja de Artesanato, Maloca, Ilha da Tanimbuca, Trilha Suspensa, Paiol da Cultura, Lago Amazônico, CEQUA, CasaEco, Chapéu de Palha, Casa de Rolo Resto, Viveiro dos Jacarés, Poraquê. Flowering plants (native or non-native) used by butterflies were found in four tourist attractions. Therefore, we primarily focused on these four attractions to obtain data from flower - visiting butterflies and verify the potential of each of these four tourist points to develop nectarivorous butterfly observation activities (Figure 1).

Selection of the botanical species. The searches for flowering plants occurred once a week, from May to August 2012 and May 2013, along the trails and at the tourist attraction of Bosque da Ciência. For the selection of the botanical species observed, it was analyzed whether they had flowers with characteristics of psychophilia syndrome (butterfly pollination). Each selected plant received a number to facilitate the records resulting from the observation. The botanical species were identified based on Ribeiro *et al.* (1999) and specialists.

Observation and identification of butterflies. The observations of flower-visiting butterflies were developed over nine months, from September 2012 to May 2013, after selection of botanical species. Each individual was observed for 45 minutes in the period of 8 am to 12 am (3 times per week) and in the period of 2 pm to 5 pm (1 time per week). It was counted as a visit, the number of times a butterfly inserted the proboscis in the flower, on each sampling occasion, that is, each time an individual plant was observed. At the time of the visit of the butterflies, photographic record was made with camera Cannon T3 Rebel - Lente 18-55 mm, noted: the species of the visiting butterfly, time, date and the plant visited.

The observations totaled 24,300 minutes, distributed in 540 sample occasions, on average nine times for each plant.

The butterflies were identified in the field or in the laboratory by means of photographic records, using Garwood & Lehman (2009), D'Abrera (2001) and were compared with identified specimens from the Invertebrate Collection-INPA. The systematic arrangement followed the proposal of Lamas (2004); Wahlbergh *et al.* (2014); Murillo-Ramos *et al.* (2008); Seraphim *et al.* (2018); Li *et al.* (2019). The photos were selected and organized in single plate that allowed the identification of butterflies in the field (Figure 2 and 3).

Data analysis. To verify the sufficiency of species richness of flower - visiting butterflies sampling over nine months, a species accumulation curve was made.

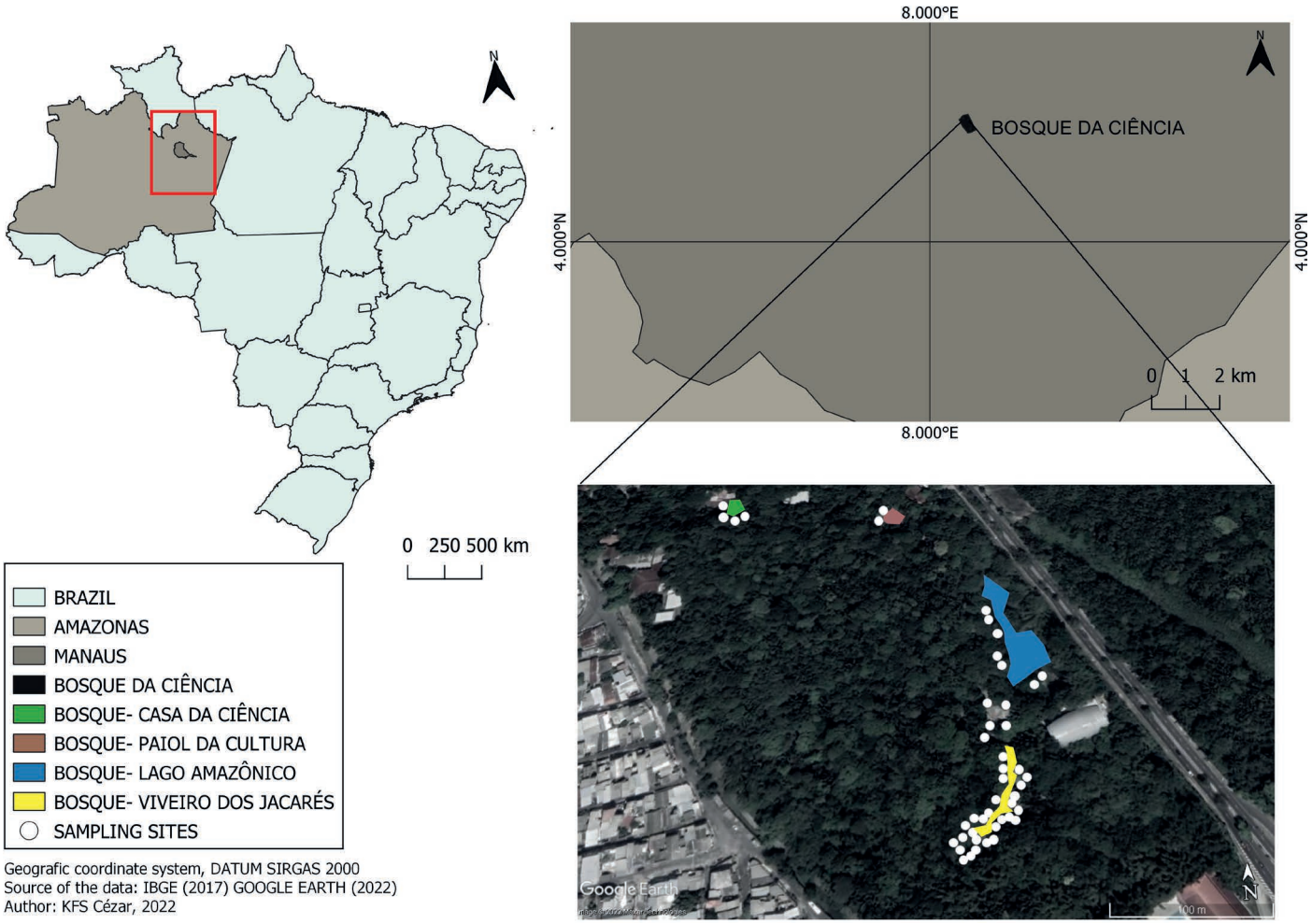

Figure 1. Observation points and the Bosque da Ciência- INPA, Manaus, Amazonas, Brazil. The white circles represent the plants used in the experiment.

To verify which period of the day had the highest rate of flower - visitation by the butterflies, the data of frequency of visitation by visiting butterfly species was made in relation to the visiting hour (from 8 am to 5 pm).

All analyses were performed in the statistical program R, version 3.2.3 (R Development Core Team 2015).

RESULTS AND DISCUSSION

In four of the fifteen tourist attractions of the Bosque da Ciência, 57 individuals of six botanical species visited by butterflies were registered: *Praxelis pauciflora* (Kunth) R. King, *Momordica charantia* L., *Warszewiczia coccinea* (Vahl), *Turnera ulmifolia* Sm., *Lantana camara* L., *Stachytarpheta cayennesis* (Rich.) (Table 1). These plants are common in open areas such as roadsides, so they are commonly found in urban areas.

Species such as *L. camara*, *S. cayennesis*, *T. ulmifolia* are considered invasive plants (Arbo 2005; Fonseca *et al.* 2006; Sanders 2012). *Momordica charantia* is a cultivable plant, with food function and is borne spontaneously, due to sexual reproduction attraction many visitors including butterflies (Lenzi *et al.* 2005). *Warszewiczia coccinea* is an arboreal species measuring from 4 e 5 m high, has inflorescences surrounded by bracts that call attention of lepidopterans and hymenopterans (Ducan 2007).

We identified 43 species of flower - visiting butterflies (Figure 2 and 3), distributed in six families: Nymphalidae (12 spp.), Pieridae (nine spp.), Riodinidae (eight spp.), Lycaenidae (eight spp.), Hesperiidae (four spp.) and Papilionidae (two spp.) (Table 2). Due to the sampling period, form of butterfly sampling and the type of vegetation in the studied area, we did not record Hedylidae in Bosque da Ciência. Hedylidae butterflies have nocturnal habits, usually collected by light traps and are sensitive to landscape change, especially those of anthropic characters (Lourido *et al.* 2008). This is the first survey for this area, therefore, all species represent new records for Bosque da Ciência.

Some butterflies visited more than one plant, being common its record in many sample occasions and its record was common in many sample occasions, as is the case of *Anthoptus epictetus* (Fabricius), *Ascia monuste* (Linnaeus), *Anartia jatrophae* (Linnaeus) and *Eurema albula* (Cramer) (Table 2). As well as most of the plants observed in this work, these species of butterflies have preference for open areas and with a lot of solar incidences (Brown Jr. & Freitas 1999). Moreover, such species and butterflies can be considered as general flower-visitors, given that in plant-pollination network, the behavior of a visitor indicates its specificity or generalization in the use of food resources. Visitors foraging on different types of flowers can be considered generalists, while visitors foraging on a specific type of flower are considered specialists (Russel *et al.* 2017). For example, *A. epictetus* as well as other skippers are not very demanding about the plants they interact with, and use the nectar of many types of herbs (Cock 2010). *Ascia monuste* butterflies are known as cabbage pests and *E. albula* have been recorded to be consuming nectar from several flowers (Soares *et al.* 2012). In case of *A. jatrophae*, the territorialist behavior due to its host plant seems to influence the generalist habit of adults, as they need to use the flowers that are available around them (Lederhouse *et al.* 1992).

Other infrequent butterflies, which were recorded to have made only a single visit, are: *Appias drussilla* (Cramer), *Emesis lucinda* (Cramer), *Synargis calyce* (Felder & Felder) e *Danaus eresimus* (Cramer) (Table 2). The infrequent visits of *A. drussilla* and *D. eresimus* may be due to the fact that they are migratory butterflies and appear randomly in places that are close to their routes (Benyamini 2011; Jacinto-Padilla *et al.* 2017). *Emesis lucinda* and *S. calyce* belong to Riodinidae, this family has populations with low density and predominance of forest-dwelling species, few exist in open areas, for this reason, few species are recorded in regional lists (Nielsem & Salazar 2014).

The plants with the greatest diversity of visitor butterflies were *S. cayennensis* with 30 species and *L. camara* with 22 species (Table 2). Both species are considered to be exclusively pollinated by butterflies, the floral characteristics of both are generally used to represent the psychophilia pollination syndrome (Faegri & Pijl 1979). They are perennial shrub plants, with inflorescences with small and tubulous flowers (Ribeiro *et al.* 1999). *Stachytarpheta cayennesis* has blue flowers (Fonseca *et al.* 2006), and, in *L. camara* the color of the flowers varies between yellow, orange and pink (Sanders 2012). In addition, they have nectar with a high concentration of sucrose, a characteristic that attracts many butterflies (Cézar *et al.* 2022).

The species richness was increasing during the execution of the work, but the curve of species accumulation shows that our sampling effort needed to be greater to reach the expected sampling of species richness for the area (Figure 4).

In the Amazon, the rainfall regime interferes with the richness of butterflies. The pattern found in other studies shows more species in the period of dry season (Oliveira *et al.* 2021; Cézar *et al.* 2022). Our study was developed mostly in the rainy season, although we found increasing results, the number of species would probably be higher if the observations were

Table 1. Flowering plants detected and observed in the tourist stations of the Bosque da Ciência- INPA (Individuals = individuals observed per plant).

Tourist attraction	Family	Plant species	Individuals
Viveiro dos Jacarés	Asteraceae	*Praxelis pauciflora* (Kunth) R. King & H. Rob	3
	Curcubitaceae	*Momordica charantia* L.	3
	Rubiaceae	*Warszewiczia coccínea* (Vahl) Klotzsch	1
	Turneraceae	*Turnera ulmifolia*L.	9
	Verbenaceae	*Lantana camara* L.	11
		Stachytarpheta cayennensis (Rich.)	8
Lago Amazônico	Asteraceae	*Praxelis pauciflora* (Kunth) R. King & H. Rob	2
	Turneraceae	*Turnera ulmifolia* L.	4
	Verbenaceae	*Stachytarpheta cayennensis* (Rich.)	1
Casa da Ciência	Rubiaceae	*Warszewiczia coccínea* (Vahl) Klotzsch	4
Paiol da Cultura	Verbenaceae	*Stachytarpheta cayennensis* (Rich.)	2
Total			**48**

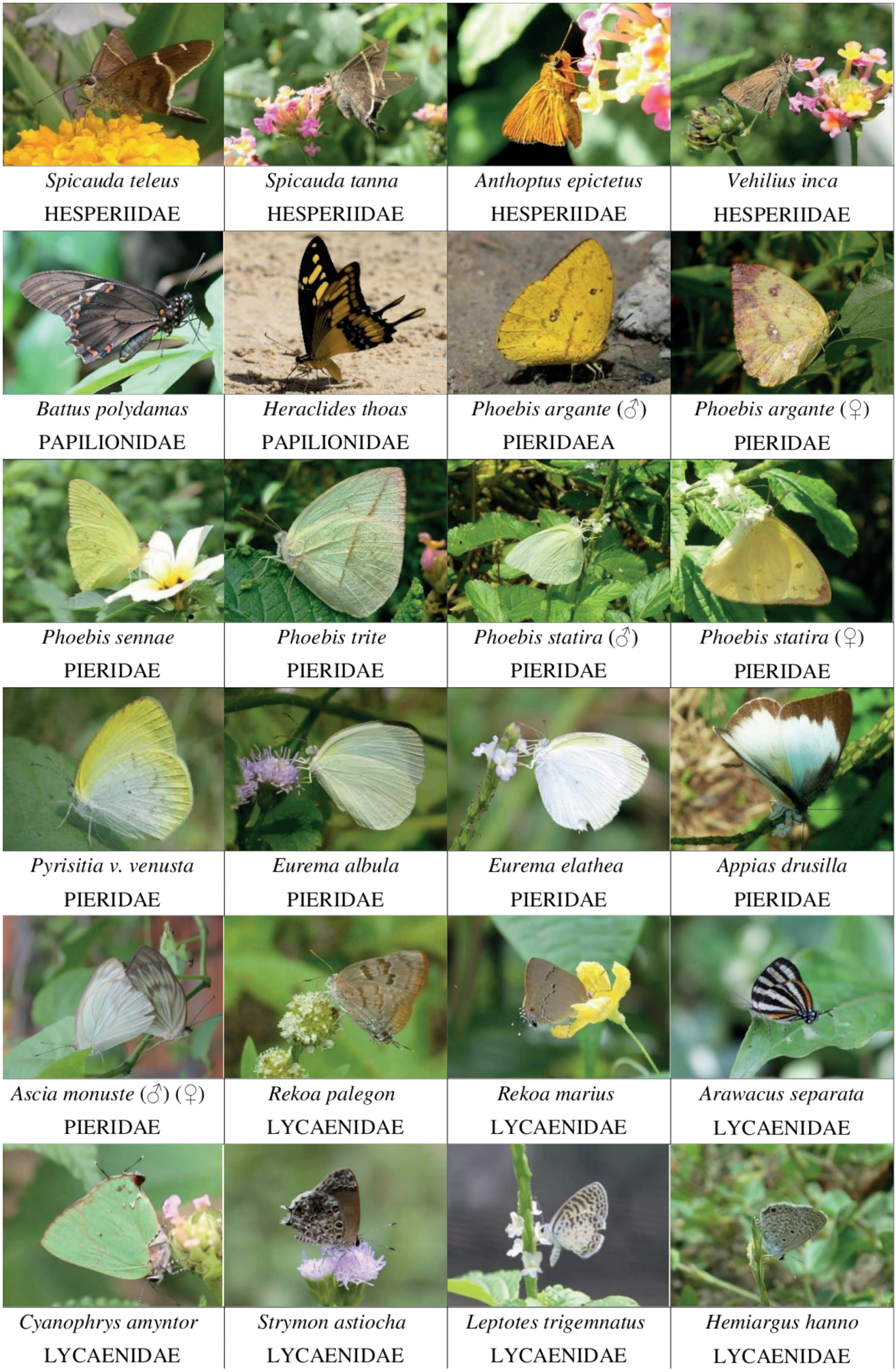

Figure 2. Identification guide of the nectarivore butterflies of the Bosque da Ciência- INPA: Hesperiidae, Papilionidae, Pieridae and Lycaenidae.

Figure 3. Identification guide of the nectarvore butterflies of the Bosque da Ciência- INPA: Riodinidae and Nymphalidae.

Table 2. Flower- visiting butterflies and frequency of visits in each plant in the Bosque da Ciência - INPA.

Butterflies (Taxa)	Plants (Family/specie)					
	Asteraceae *P. pauciflora*	**Curcubitaceae** *M. charantia*	**Rubiaceae** *W. coccínea*	**Turneraceae** *T. ulmifolia*	**Verbenaceae** *L. camara*	**Verbenaceae** *S. cayennensis*
Hesperiidae						
Spicauda teleus (Hübner)					5	
Spicauda tanna Evans					6	
Anthoptus epictetus (Fabricius)	3	1		3	11	9
Vehilius inca (Scudder)	2			3	4	4
Papilionidae						
Battus polydamas (Linnaeus)					2	
Heraclides thoas (Linnaeus)					4	
Pieridae						
Phoebis argante (Fabricius)					8	
Phoebis sennae (Linnaeus)				4	9	
Phoebis trite (Linnaeus)			7		5	4
Phoebis statira (Cramer)					4	3
Pyrisitia venusta venusta (Boisduval)	4			4		4
Eurema albula (Cramer)	6	4	3	6	4	7
Eurema elathea (Cramer)	5	4		5		6
Appias drusilla (Cramer)						1
Ascia monuste (Linnaeus)	4		3	4	5	6
Lycaenidae						
Rekoa palegon (Cramer)	3					
Rekoa marius (Lucas)			3			
Arawacus separata (Lathy)					1	3
Cyanophrys amyntor (Cramer)						3
Strymon astiocha (Prittwitzs)	4					6
Panthiades phaleros (Linnaeus)			3			
Leptotes trigemnatus (Butler)						4
Hemiargus hanno (Stoll)	4					4
Riodinidae						
Emesis lucinda (Cramer)			1			
Aricoris campestres (H. W. Bates)	1					5
Lemonias zygia Hübner						4
Synargis calyce (C. Felder & R. Felder)						1
Nymphidium azanoides Butler						3
Nymphidium caricae (Linnaeus)		3				5
Nymphidium lisimon (Stoll)		4				6
Stalachtis phlegia (Cramer)						2
Nymphalidae						
Danaus eresimus (Cramer)						1
Anartia amathea (Linnaeus)					3	3
Anartia jatrophae (Linnaeus)	4			4	4	6
Junonia evarete (Cramer)					3	5
Eresia clio (Linnaeus)	3					
Eresia eunice (Hübner)	3					
Ortilia liriope (Cramer)					2	3
Euptoeta hegesia (Cramer)				6	4	5
Agraulis vanillae (Linnaeus)					4	4
Dryas iulia (Fabricius)					3	4
Philaetria dido (Linnaeus)					2	
Heliconius sara (Fabricius)					6	6
Total	**46**	**16**	**20**	**39**	**99**	**127**

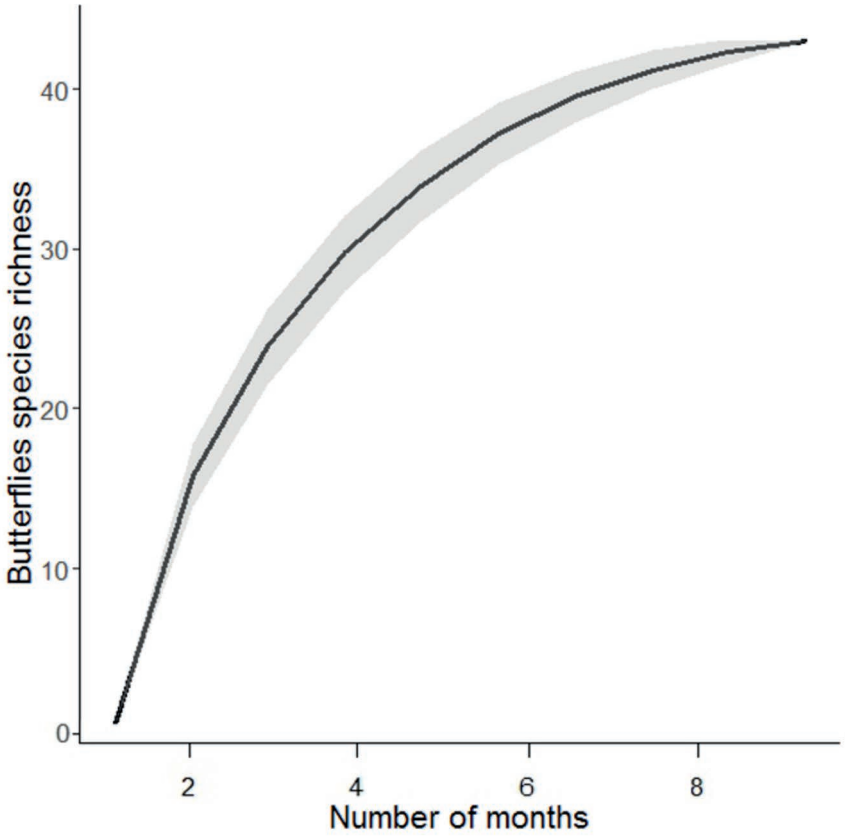

Figure 4. Richness accumulation curve of nectarivore butterflies species over the months of study sampling at the Bosque da Ciência-INPA, Manaus, Amazonas, Brazil.

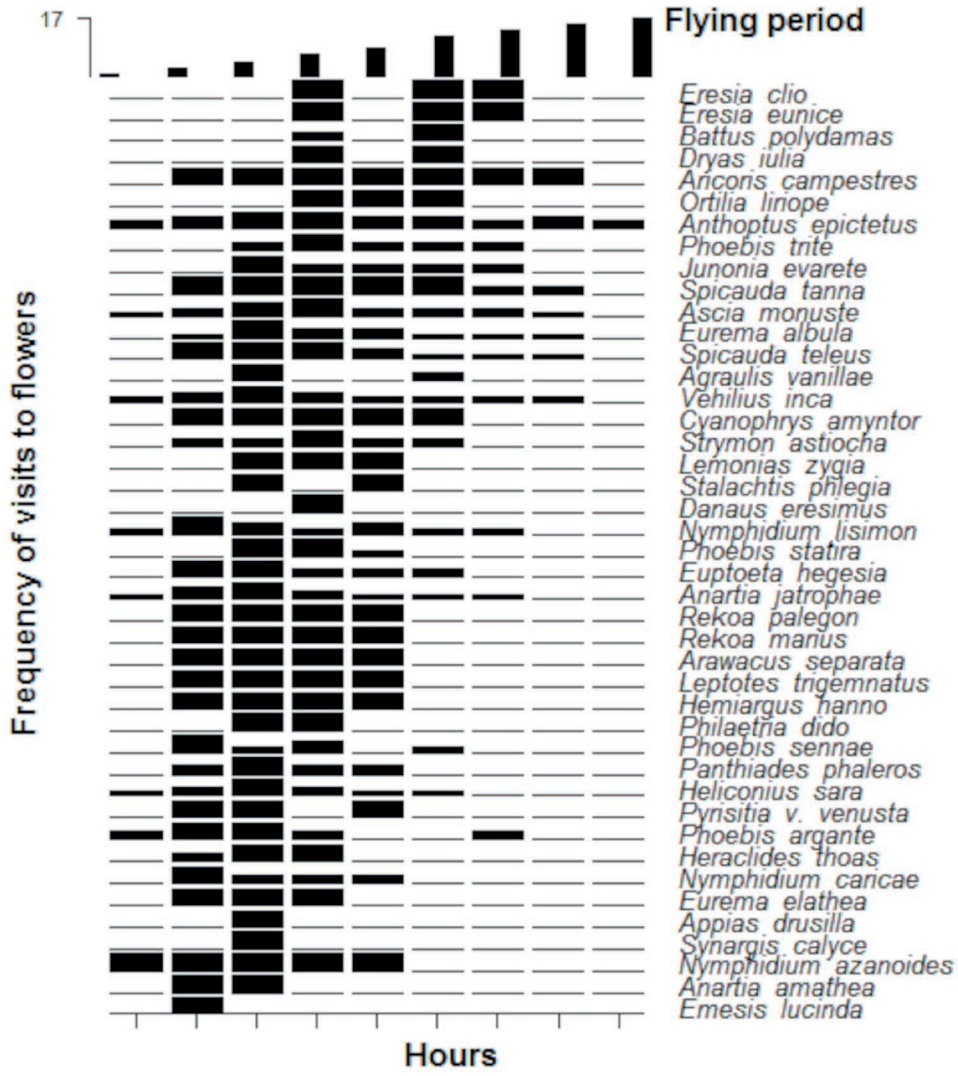

Figure 5. Time of visitation of butterflies to flowers, recorded from September 2012 to May 2013 in the Bosque da Ciência- INPA.

extended to the following months, into the dry season.

It was observed that from 9 am to 11 am there was higher activity of butterflies, the demand for food resources (visit the flowers), so most of the observations occurred at this time (Figure 5). This pattern of butterfly activity follows naturally since most species of the group are diurnal and only few have twilight habits (Freitas 2003).

Based on our observations, the most suitable points for the implantation of butterfly gardens are open areas with high solar incidence. Inside Bosque da Ciência, the most favorable place is the surrounding the "Viveiro dos Jacarés", which presented a greater abundance of plants with flowers and butterflies.

Plants such as *S. cayennensis* and *L. camara* are highly explored as nectar sources by butterflies. So, we suggest that they should be considered important elements within the attraction gardens. The most frequently expected visitors are *A. epictetus*, *A. monuste*, *A. jatrophae* and *E. albula*.

The butterflies of Bosque da Ciência are a representation of the nectarivores diversity in the urban area of Manaus. The floral attractions in the woods, contributes to the appearance of this butterfly guild on site. The establishment of an attractive butterfly garden can contribute to the promotion of environmental education, thereby achieving one of the objectives of Bosque da Ciência, which has been created to stimulate knowledge and awareness of biodiversity preservation among general public.

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