Chapter

Diversity of Tephritidae and Agromyzidae (Diptera: Brachycera) in Flower Heads of Asteraceae in the Chaco

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Abstract

The Chaco is an international biome, connecting four countries: Paraguay (230,000 km^2), Bolivia (90,000 km^2), Argentina (520,000 km^2), and Brazil (Mato Grosso do Sul state (MS), with around 9,000 km^2 and in the middle of South America. Brazilian Chaco is restricted to Porto Murtinho region, MS. The daisies (Asteraceae) with near 24,000 species worldwide is characterized by herbs and shrubs that coevolved with several taxa of endophagous insects: dipterans Agromyzidae, Ceciidomyidae and Tephritidae; Coleoptera (Apionidae), Hemiptera (Miridae), Lepidoptera (Blastobasidae, Gelechiidae, Pterophoridae, Pyralidae, and Tortricidae) and the parasitoids of this endophagous insects, which found in the daisies's flower heads ideal conditions for food, breeding site and shelter. The Neotropical florivorous flies are the Agromyzinae (Agromyzidae), and Tephritinae (Tephritidae), which in their larval stage feed on Asteraceae inflorescences. To report the species of florivore flies, their host plants and parasitoids in flower heads of Asteraceae from the Brazilian Chaco, we sampled inflorescences of 25 species (± 500 flower heads/species) that were kept in containers to the emergence of the florivorous flies or their parasitoids sampled in the three phytophysiognomies. The adult insects after 48 hours of their emergence were fixed in 80% ethanol for later identification. A total 25 species of Asteraceae were evaluated in the Brazilian Chaco, being collected 17,000 flower heads. Nine tribes of two Asteraceae subfamilies were sampled, from which 15 species of florivorous flies were recovered. We found 5 genera with 9 of Tephritinae (Tephritidae), 6 species of *Melanagromyza* (Agromyzinae, Agromyzidae), and 104 parasitoids (Hymenoptera) of the florivorous flies.

Keywords: Florivory, *Melanagromyza* spp., Steppe Savannas, Tephritinae, weed biocontrol

1. Introduction

The florivorous flies are the most importante guild of insects feeding on daisis's flowers in Brazil. They belong to the subfamilies Agromyzinae (Agromyzidae), and Tephritinae (Tephritidae). Those flies, in their larval stage, feeds in Asteraceae inflorescences (flower heads).

Asteraceae is the bigest family of Angiospermae with around 24,000 species of plants worldwide, being characterized by herbs and shrubs that coevolved with several taxa of endophagous insects, which found in their flower heads ideal conditions for food, breeding site and shelter. As pointed out by Lewinsohn [1], several other taxa on Insecta has been reported on the Asteraceae flower heads, beyound florivore flies, such as Apionidae (Coleoptera), Miridae (Hemiptera), Blastobasidae, Gelechiidae, Pterophoridae, Pyralidae, and Tortricidae (Lepidoptera), as well their parasitoids, mainly Hymenoptera.

In Neotropical Region, the species of Asteraceae, depending on the biome evaluated, use to be the first or sencond plant Family in the rank of species richness, as pointed out for the Atlantic Forest on Espirito Santo state, Brazil [2]. The studies about the florivore insects are important for both: to expand scientific knowledge, as well as to solve environmental problems in food production systems. By now, at least 38 species and their subspecies of invasive Asteraceae have been the target of biological control programs around the world. About 21 species of Tephritidae have been manipulated to control Asteraceae species. The most significant biological control programs for such plants with florivorous flies are underway in Australia, Canada, USA, New Zealand and South Africa [3], and West Africa, where the stem-gall tephritid *Cecidocares connexa* (Macquart 1848) have been upplied on the biological supperession, against the invasive Asteraceae *Chromolaena odorata* (L.) R. M. King & H. Rob. [4]

Chaco is an international biome, distributed by four South American countries: Argentina, Paraguay, Bolivia and Brazil. The total area of this biome is spread over the four above countries: Paraguay (230,000 km^2), Bolivia (90,000 km^2), Argentina (520,000 km^2), and Brazil (Mato Grosso do Sul = MS, with around 9,000 km^2), in the center of South America. The word Chaco, derives from the indigenous term: "Quechua chaku" – that mean hunting territory or steppe savanna. The Brazilan Chaco is restricted to the municipality of Porto Murtinho, located in the south of the Pantanal, state of MS, where it is characterized as the humid Chaco. This sedimentary plain is covered by natural vegetation of Steppe Savanna and occupy the central region of South America, called Gran Chaco or simply Chaco. It's covered by the most extensive continuous dry forest in the American continent, and represents the only one subtropical dry forest of the world. The Chaco region, with approximately 1,000,000 km^2 (Gran Chaco), covers North Argentina (46%), West of Paraguay (32%), Southeast of Bolivia (15%), and a significant portion (7%) in the Midwest of Brazil (Porto Murtinho, Mato Grosso do Sul), separated from the biome matrix by the Paraguay river [5, 6].

The Chaco Biome is home to a great diversity of environments: mountains, savannas, floodplains, swamps, wetlands and saltpans, with a great extension and diversity of flora and fauna. The ecosystems that are part of the Chaco have high rates of endemism and diversity of plant and animal species, compared to other arid, semi-arid and sub-humid environments. Nowadays, Chaco faces great anthropic pressure, due to the advances of agriculture that has increased, exploiting its phyto ecological regions. The high biodiversity of the Brazilian Chaco, and its rates of endemismo [5, 6], suggest that native fruit plants are potential hosts for some frugivorous and florivorous fly species there.

Tephritidae is the most important, and the second family in species richness (behind Cecidomyiidae) of the phytophagous Diptera, with around 5,000 species, being known mainly due the economic importance of their pest species upon fruit and vegetables [7]. But there are another face of these flies: those species that feed on plants, but don't cause economic loss. By other hand, they are of great economic importance due to their phytophagy, destroying seeds of undesirable Asteraceae species in agrosilvipastoral systems. These florivorous flies (Tephritinae), the bigest

Subfamily of Tephritidae - near 2,000 species, were subject of our research [8], and will be presented in this e-book chapter. We evaluated the diversity of its host Asteraceae, species richness and abundance of florivore's fly species in each species of Asteraceae evaluated, as well as the co-occurrence of other taxons of florivorous dipterans and their respective parasitoids in the Brazilian Chaco.

2. The mainly aims of this research

1. To Report the species of florivore flies (FF), their host plants, and their parasitoids, in flower heads of Asteraceae from the Brazilian Chaco. 2. To quantify abundance, species richness and diversity of the FF in three phytophysiognomies in the Brazilian Chaco.

3. Materials and methods

Sampling was carried out in the municipality of Porto Murtinho-MS, Brazil, in three different phytophysiognomies of the Brazilian Chaco. Flower heads of Asteraceae were collected at: Eldorado Farm (21 ° 42′20.9 "S 57 ° 47'45.6" W, altitude 82 m); Santa Carmem Farm (21 ° 50′23.2 "S 57 ° 49'13.6" W, altitude 78 m), and in a transect along the Highway MS-457 (altitude 124 m).

At the Eldorado farm, the predominant phytophysiognomy is of the type Wooded Steppe Savanna (WSS), and at the farm Santa Carmem the type is Park Steppe Savanna (PSS), subtype locally named *Carandazal*), with formation of Riparin Forest. Carandazal is characterized by the vegetation with predominance of the *Copernicia alba* Morong palm. In these two locations the transects were of approximately 1 km in open field, with a sampling effort of one person during 2 h. On Highway MS-457 trail, the predominant phytophysiognogmy is of the Grassy-Wood Savanna (GWS). The collections were carried out along the edges of this trial, starting from BR-267 (21 ° 44′51.4 "S 57 ° 33'44.1" W) towards the *Parque Municipal Cachoeira do APA* (= APA Waterfall Municipal Park). This last trail covered about 2 km.

The flower heads of all Asteraceae species found in the field were collected, preferably 200 in pre-anthesis per species. The flower heads were inserted in plastic bags, identifying the species of plant and the area. The collection carried out monthly and during from May 2017 to April 2018. Exsiccates from all Asteraceae evaluated were prepared.

The climate of the region in the Brazilian Chaco is considered tropical Aw by the Köppen classification, with hot and rainy summer and dry and milder winter. The rainfall varies between 1,100 mm and 1,800 mm. The vegetation in the Chaco is made up of shrubs, deciduous, microphiles and spinach. The soil is highly saline, with little water drainage, which is why in rainy season it generates temporary floods characteristic of the Pantanal.

The Asteraceae inflorescences collected were taken to the Laboratory of Systematic and Taxonomy of Tephritidae (LabTaxon), Universidade Federal da Grande Dourados (UFGD), where the flower heads were counted and placed in 500 ml plastic cups, with the juxtaposed openings forming a cage. The cups were attached with adhesive tape, forming a closed container that made it possible to contain and obtain species of florivorous flies and their parasitoids. After emergence of florivorous flies and/or their parasitoids, they remained alive for a period of 48 hours to acquire the chromatic pattern of the body and wings. They were subsequently conserved in 92% ethyl ethanol. After 20 days without any fly's emergence, inspections on the containers were stopped, and the remaining material discarded.

The exsiccates of the sampled Asteraceae were sent for identification of the species by Professor Dr. Jimi Naoki Nakajima (Universidade Federal de Uberlândia), specialist in Asteraceae. The duplicates of the identified Asteraceae were deposited at the CGMS Herbarium of the Biodiversity Museum-FCBA/UFGD in Dourados-MS, as the specimens voucher.

Statistical analyzes: Infestation rates were calculated using the equation: Number of FF/Number Flower heads x 100, being N the number FF (= number of Florivorous flies) divided by the Number of Flower heads (total of Flower heads/Asteraceae species). Were analysed the diversity in each phytohysiognomy. The diversity index (Shannon-Weaner) (H), Species Richness Index (Margalef), (Alpha), and Uniformity or Equitability Index (E) were calculated.

4. Results

Twenty-five species of Asteraceae of two subfamilies (Cichorioideae and Asteroideae), and 9 tribes (Vernonieae, Senecioneae, Astereae, Inuleae, Plucheae, Neurolaeneae, Heliantheae, Targeteae, Eupatorieae) were sampled in three phytophysiognomies: Grassy-Woody Steppe Savana, Park Steppe Savanna, and Wooded Steppe Savanna from the Brazilian Chaco at Porto Murtinho-MS, Brazil (**Table 1**).

A total 472 adults of florivorous flies from two families and 15 species (9 of Tephritidae and 6 of Agromyzidae) were recovered, being 163 agromyzids (*Melanagromyza*), and 309 tephritids from nine species were recovered from 15 species Asteraceae species. The Tephritid species belong to five genera: *Cecidochares* Bezzi 1910; *Dictyotrypeta* Hendel 1914; *Tomoplagia* Coquillett 1910; *Trupanea* Schrank 1795, and *Xanthaciura* Hendel 1914. We found nine morphospecies of Tephritinae. All specimens of Agromyzidae are grouped in a single genus: *Melanagromyza* Hendel 1920. The recovered parasitoids are (n = 104) Hymenoptera await identification by specialists. Herein, trophic interactions are reported between florivorous flies with 15 species of Asteraceae in the Brazilian Chaco (**Table 2**).

Among the 16 species of Asteraceae that host florivorous flies or their parasitoids, five were associated only with agromizids: *Bidens gardneri*, *Bidens pilosa*, *Acmella grisea*, *Wedelia brachycarpa* and *Cyrtocymura scorpioides*. Six hosted only tephritids: *Chromolaena ivifolia*, *Praxelis clematidea*, *Pectis odorata*, *Porophyllum angustissimum*, *Lepidaploa remotiflora* and *Lepidaploa remotiflora*. Four Asteraceae species hosted species of both florivorous flies (Tephritine and Agromyzinae): *Aspilia elata*, *Chromolaena margaritensis*, *Dimerostemma grazielae* and *Porophyllum ruderale*. Parasitoids emerged from the florivorous flies feeding on 11 Asteraceae species (**Table 2**).

Herein, for the first time we present the several associations of florivorous fly species with their Asteraceae (flower heads) host species in the Brazilian Chaco. The trophic interactions established here were the following: *Chromolaena margaritensis* was colonized by *Xanthaciura* sp.1; *Aspilia elata* by *Dictyotrypeta* sp.1, and *Dictyotrypeta* sp.2; *Dimerostemma grazielae* by *Trupanea* sp.1; *Pectis odorata* by *Trupanea* sp.1; *Lepidaploa remotiflora* by *Tomoplagia minattai*. From *Lessingianthus niederleinii* flower heads emerged adults of *Tomoplagia matzenbacheri*. From the flower heads of *Chromolaena ivifolia* occurred the highest abundance of florivorous flies (166). This Asteraceae species was infested only by species of tephritids. From *Aspilia elata* flower heads, were obtained greatest abundance of agromyzids (62) (**Table 2**).

The highest rates of infestation by florivorous flies in Asteraceae in the Brazilian Chaco were reported in the following species: *Acmella grisea* (34%); *Aspilia elata*

Asteraceae taxa (subfamilies, tribes and species)	Host status	Environments (Phytophysiognomies)
Cichorioideae		
Vernonieae *Cyrtocymura scorpioides* (Lam.) H. Robins.	Host	Grassy-Woody Steppe Savanna (GWSS)
Lepidaploa remotiflora (Rich.) H. Robins.	Host	Wooded Steppe Savanna (WSS) Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Lessingianthus niederleinii (Hieron.) H. Robins.	Host	Grassy-Woody Steppe Savanna (GWSS)
Lessingianthus rubricaulis (Bonpl.) H. Robins.	Nonhost	Grassy-Woody Steppe Savanna (GWSS)
Asteroideae		
Senecioneae *Erechtites hieracifolia* (L.) Raf.	Nonhost	Wooded Steppe Savanna (WSS)
Astereae *Conyza bonariensis* (L.) Cronquist	Nonhost	Wooded Steppe Savanna (WSS)[a]
Inuleae *Pluchea quitoc* DC.	Nonhost	Wooded Steppe Savanna (WSS) Grassy-Woody Steppe Savanna (GWSS)
Plucheae *Pterocaulon virgatum* (Lam.) DC.	Nonhost	Grassy-Woody Steppe Savanna (GWSS)
Neurolaeneae *Calea rupicola* Chodat	Nonhost	Grassy-Woody Steppe Savanna (GWSS)
Heliantheae *Acmella grisea* (Chodat) R.K. Jansen	Host	Grassy-Woody Steppe Savanna (GWSS)
Aspilia elata Pilg.	Host	Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Aspilia montevidensis (Spreng.) Kuntze	Nonhost	Grassy-woody Steppe Savanna (GWSS)
Bidens pilosa L.	Host	Grassy-Woody Steppe Savanna (GWSS)
Bidens gardneri Baker	Nonhost	Grassy-Woody Steppe Savanna (GWSS)
Dimerostemma grazielae H. Rob.	Host	Wooded Steppe Savanna (WSS) Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Wedelia brachycarpa Baker	Host	Wooded Steppe Savanna (WSS) Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Targeteae *Pectis odorata* Griseb.	Host	Wooded Steppe Savanna (WSS) Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Porophyllum angustissimum Gardner	Host	Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Porophyllum ruderale (Jacq.) Cass.	Host	Wooded Steppe Savanna (WSS) Park Steppe Savanna (PSS) Grassy-Woody Steppe Savanna (GWSS)
Eupatorieae *Austroeupatorium inulifolium* (Kunth) R.M. King & H. Rob.	Nonhost	Grassy-Woody Steppe Savanna (GWSS)[b]
Campuloclinium macrocephalum (Less.) DC.	Nonhost	Grassy-Woody Steppe Savanna (GWSS)
Chromolaena ivifolia (L.) R. M. King & H. Rob.	Host	Grassy-Woody Steppe Savanna (GWSS)

Asteraceae taxa (subfamilies, tribes and species)	Host status	Environments (Phytophysiognomies)
Chromolaena margaritensis (Hassl.) R. M. King & H. Rob.	Host	Wooded Steppe Savanna (WSS) Park Steppe Savanna (PSS)c Grassy-Wood Steppe Savanna (GWSP)
Praxelis clematidea (Griseb.) R. M. King & H. Rob.	Host	Wooded Steppe Savanna (WSS) Grassy-Woody Steppe Savanna (GWSS)
Urolepis hecatantha (DC.) R. M. King & H. Rob.	Nonhost	Wooded Steppe Savanna (WSS) Grassy-Woody Steppe Savanna (GWSS)

[a] *Wooded Steppe Savanna (WSS) = Fazenda Eldorado (= Eldorado Farm);*
[b] *Grassy-Woody Steppe Savanna (GWSS), Access to Rio Apa waterfall (Border with Paraguay), and Transition to the Brazilian Cerrado;*
[c] *Park Steppe Savanna (PSS) = Fazenda Santa Carmen (= Santa Carmen Farm), "Carandazal Phytophysiognogmy".*

Table 1.
Status of Asteraceae species for florivorous flies Tephritinae (Tephritidae, and/or Melanagromyza *Hendel 1920, Agromyzinae: Agromyzidae) in three Phytophysiognomiesof the Brazilian Chaco.*

Asteraceae Taxa	N° of Flower Heads	Agromyzidae	Tephritidae	* I.L.FF/F.H. (%)	Parasitoids (Hymenoptera)
Tribo					
Astereae *Conyza bonariensis*	50	0	0	0	0
Coreopsideae					
Bidens gardneri	128	3	0	2.34	0
Bidens pilosa	85	6	0	7.05	0
Eupatorieae					
Austroeupatorium inulifolium	2,500	0	0	0	0
Campuloclinium macrocephalum	211	0	0	0	0
Chromolaena ivifolia	1,100	0	166	15.09	21
Chromolaena margaritensis	2,299	12	18	1.30	5
Praxelis clematidea	424	0	1	2.12	38
Urolepis hecatantha	1,245	0	0	0	0
Heliantheae					
Acmella grisea	139	48	0	34.53	0
Aspilia elata	320	62	29	28.43	4
Aspilia montevidensis	73	0	0	0	0
Dimerostemma grazielae	1,019	23	3	2.55	12
Wedelia brachycarpa	256	3	0	1.17	0

Asteraceae Taxa	N° of Flower Heads	Agromyzidae	Tephritidae	* I.L.FF/F.H. (%)	Parasitoids (Hymenoptera)
Inuleae					
Pluchea quitoc	290	0	0	0	5
Pterocaulon alopecuroides	1,280	0	0	0	0
Neurolaeneae					
Calea rupicola	210	0	0	0	0
Senecioneae					
Erechtites hieracifolius	50	0	0	0	0
Tageteae					
Pectis odorata	1,123	0	17	1.51	2
Porophyllum angustissimum	925	0	16	1.72	4
Porophyllum ruderale	1,343	3	51	4.02	7
Vernonieae					
Cyrtocymura scorpioides	120	3	0	2.50	0
Lepidaploa remotiflora	995	0	2	0.20	2
Lessingianthus niederleinii	585	0	6	1.02	4
Lessingianthus rubricaulis	250	0	0	0	0
Total	17,000	163	309		104

[1]*I.L.FF/F.H. = Infestation Level of Florivorous Flies by the number of Flower heads in Asteraceae species.*

Table 2.
Abundance Florivorous flies and parasitoids (Hymenoptera) associated with flower heads of Asteraceae species in three phytophysionomies in the Brazilian Chaco (at Porto Murtinho-MS) (May 5, 2017 to April 5, 2018).

(28%), both from the tribe Heliantheae, and from *Chromolaena ivifolia* (15%) (Eupatorieae). The species of *Trupanea* (Tephritinae) showed the highest number of trophic interactions (6) with Asteraceae. *Trupanea* was also the only Tephritidae genus associated with more than one tribe (three). The Asteraceae species from the Tageteae and Vernonieae tribes, each presented trophic associations with only one tephritid genus: *Trupanea* and *Tomoplagia*, respectively (**Table 2**).

Praxelis clematidea was the host in which the higher abundance of parasitoids (38), and presented trophic interaction with only one specimen of *Xanthaciur*a sp.1. In the species of the tribes Eupatorieae and Heliantheae, ocurred higher florivore fly species richness, being obtained three species from each host plant. In the Heliantheae species were registered greatest abundance of Agromyzidae (136 adults), from which four of the six registered species of all the reared agromizids. In the species of Eupatorieae there was obtained higher abundance of tephritines (185), and 64 parasitoids (**Table 3**).

Characteristics of trophic interactions between florivorous flies and Asteraceae as a function of the Chaquean phytophysiognomies: Only the species *Trupanea* sp.1

Species of Asteraceae	Species of Tephritinae	Abundance
Tribe		
Eupatorieae		
Chromolaena ivifolia	*Cecidochares* sp.1	146
	Xanthaciura sp.2	20
Chromolaena margaritensis	*Xanthaciura* sp.1	17
Praxelis clematidea	*Xanthaciura* sp.1	1
Heliantheae		
Aspilia elata	*Dictyotrypeta* sp.1	23
	Dictyotrypeta sp.2	6
Dimerostemma grazielae	*Trupanea* sp.1	3
Tageteae		
Pectis odorata	*Trupanea* sp.1	17
Porophyllum angustissimum	*Trupanea* sp.2	16
Porophyllum ruderale	*Trupanea* sp.2	51
Vernonieae		
Lepidaploa remotiflora	*Tomoplagia minattai* Aczel 1955	2
Lessingianthus niederleinii	*Tomoplagia matzenbacheri* Prado, Norrbom & Lewinsohn, 2004	6

Table 3.
Species of Asteraceae infested by tephritine species (Tephritidae) and their absolute abundance in their flower heads sampled in the Brazilian Chaco (May 5, 2017 to April 5, 2018).

Florivorous Flies	Wooded Steppe Savanna	Park Steppe Savanna	Grassy-woody Steppe Savanna
Cecidochares sp. 1	0	0	146
Dictyotrypeta sp.1	0	23	0
Dictyotrypeta sp.2	0	6	0
Tomoplagia minattai	0	0	2
Tomoplagia matzenbacheri	0	0	6
Trupanea sp.1	3	16	1
Trupanea sp.2	31	33	3
Xanthaciura sp.1	8	7	3
Xanthaciura sp.2	0	0	20
Melanagromyza spp. (6 spp.)	3	81	87
Totals	45	166	268
Parasitoid Species Richness (S)	4	6	8
	19	50	27

Table 4.
Abundance of endophagous insects associated with Asteraceae chapters in three phytophysiognomies in the Brazilian Chaco (Porto Murtinho-MS) (May 5, 2017 to April 5, 2018).

and *Xanthaciura* sp.1 occurred simultaneously in the three phytophysiognomies. The species of *Dictyotrypeta* spp. were specific to PSS and *Cecidochares* sp.1 from GWSS. The two species of *Tomoplagia* recorded in this study, *Tomoplagia matzenbacheri* and *Tomoplagia minattai*, were specific to GWSS, as well as *Xanthaciura* sp.2.

None of the species of tephritids reported for WSS was specific to this phytophysiognomy (**Table 4**).

The phytophysiognomy with highest species richness (S = 6), and abundance of Tephritinae (268) was GWSS. The highest abundance of parasitoids (50) was found in the phytophysionomy Park Steppe Savanna (PSS). PSS presented the highest diversity by the Shannon (H) index, despite it haven't the greatest species richness (S). Probably this is due to the fact that this index takes into account the homogeneity of species in the environment. In the Grassy-Wood Steppe Savana (GWSS) the highest value of diversity was registered for the Margalef index. This index consider the sample size, which can be explained by the high abundance of the florivore fly *Cecidochares* sp.1 in this phytophysiognomy (**Tables 4** and **5**).

Applying the rarefaction method in the samples ("curve of collector"), it was possible to obtain estimates of the species richness of Tephritinae from the Chaquenha community. It indicated that the samples were insufficient to reach the asymptote of curve of collector. This means that the sampling effort was not enough to detect all species of florivorous flies present in the Brazilian Chaco. Due to the mosaic of phytophysiognomies in this biome, the few sampling points proved to be insufficient, to estimate the total species richness of florivorous flies in the Brazilian Chaco, even doing repetitions in the four year seasons (**Figure 1**).

Phytophysiognomies	H	ALFA	E	S
Wooded Steppe Savanna (WSS)	0.7285	0.5351	0.6631	3
Park Steppe Savanna (PSS)	1.4163	0.9077	0.88	5
Grassy-Woody Savana (GWS)	1.3084	0.9077	0.7302	7
Three combined Phytophysionomies	1.6927	1.4063	0.7302	9

Table 5.
Indices of Shannon (H), Margalef (A), equitability (E) and species richness (S) for Tephritinae (Diptera: Tephritidae) associated with flower heads of Asteraceae in three phytophysionomies from the Brazilian Chaco (Porto Murtinho -MS) (May 5, 2017 to April 5, 2018).

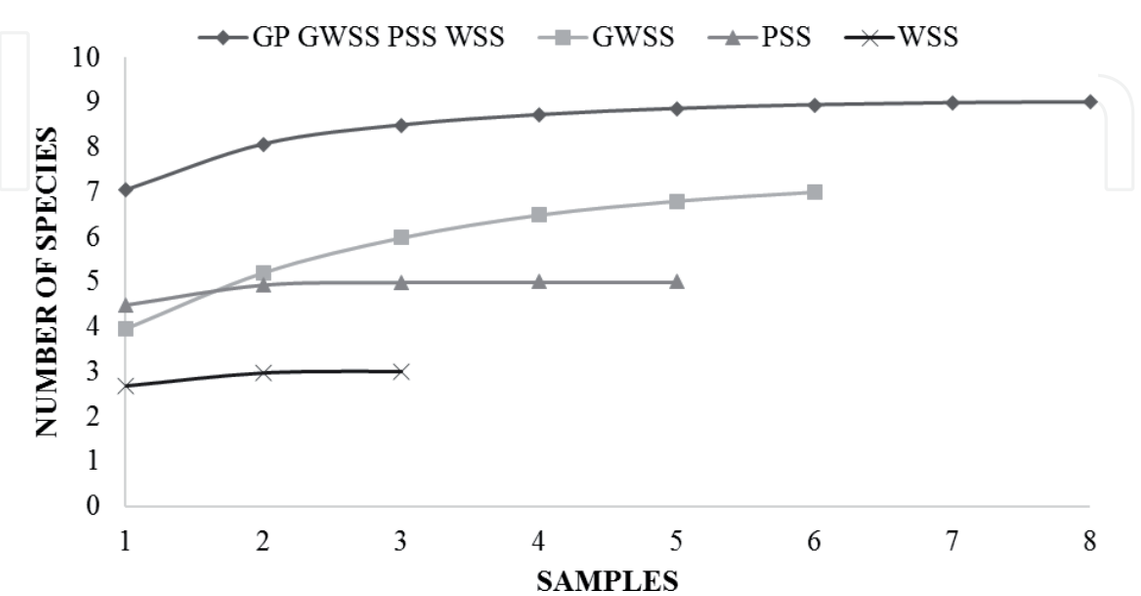

Figure 1.
Curve of species accumulation of Tephritinae (Diptera: Tephritidae) in three phytophysiognomies: General Panorama (GP), Grassy-Woody Steppe Savanna (GWS), Park Speppe Savanna (PSS), and Wood Speppe Savanna (WSS), in the Brazilian Chaco (Porto Murtinho-MS) (May 5, 2017 to April 5, 2018).

5. Discussion

The Brazilian Chaco presented a lower florivorous species richness than other neighboring Neotropical biomes already evaluated. Eighteen genera of Tephritinae (Tephritidae) and 80 species occurring in Brazil were reported by Prado [9]. In the Cerrado Biome, 12 genera are listed [10], and 10 genera have already been cataloged in the Atlantic Forest [11].

This research represents the first inventory of florivorous flies (Tephritidae and Agromyzidae) feeding in Asteraceae flower heads in the Chaco. Associations of *Melanagromyza* species (Agromyzidae) with Asteraceae species are reported for the Cerrado Biome [10], but there was yet no data for the Brazilian Chaco.

Due to the lack of keys for several taxa of Neotropical Tephritinae, many publications were unable to perform specific identification of florivorous flies. Species of *Cecidochares*, *Dicyotrypeta*, *Trupanea* and *Xanthaciura* are constantly only morphospecified.

In the Brazilian Chaco, the abundance of tephritids was higher than that of agromyzids, as well as the species richness (9 Tephritinea spp. against 6 probable species of *Melanagromyza* spp. (Agromyzinae, Agromyzidae). This pattern was also repeated in the Cerrado Biome [10]. According to several authors in the Neotropical Region, the most frequent and abundant species of florivore Diptera are the tephritines (Tephritidae). Herein, tephritines and agromyzines presented similar frequencies. Therefore, this pattern is not well understood and needs further studies in the Chaco biome to clarify its patterns of co-occurrence.

The Brazilian Chaco presented 16 trophic interactions between 15 species of florivorous flies and their Asteraceae species. In the Cerrado Biome, 49 species of Asteraceae are reported as hosts of florivore dipterans [10]. Herein, *Chromolaena margaritensis* hosted *Xanthaciura* sp.1 and *Melanagromyza* sp. 6. These are the first records for such associations worldwide. The flower heads of *C. margaritensis* has already been reported as a host for *Cecidochares* species in South Brazil [12].

Tomoplagia species were specific to the Vernonieae tribe. This relationship is already known [9]. In this research, *Trupanea jonesi* was the most generalist species, infesting two distinct asterace tribes: Heliantheae and Tageteae. The polyphagy of *T. jonesi* is well known, more than 100 associations with their host plants have been established [13].

The low diversity of florivorous flies recorded in the Brazilian Chaco can be explained by the low diversity of Asteraceae species there or by insufficient sampling effort. Probably, the species richness of florivorous flies is positively correlated with the species richness of sampled Asteraceae in the Chaco. Biomes richer in Asteraceae show a greater diversity of these trophic interactions. The Cerrado presents a high diversity of Asteraceae and associated species of Tephritinae, as has pointed out by other inventories [9, 11, 12].

Tomoplagia matzenbacheri and *Tomoplagia minattai* found in this research are new records for the state of Mato Groso do Sul (MS). The only previous work developed in the state of MS with florivorous flies was carried out by Uchoa, Wachter-Serapião & Roque [14] in the Cerrado, a fragment of the Atlantic Forest and an agroecosystem (orchard).

The Brazilian Chaco presented 25 species of Asteraceae, which apparently represents 92% of the species cataloged for this Biome [15]. However, many of the species recorded in this study do not appear in the floristic inventories of the Brazilian Chaco and vice-versa. New floristic studies must answer if there is a subsampling or if it is the result of the divergent interpretation among different authors about the characterization of phytophysiognomies truly Chaqueans.

Herein, *Dimerostemma grazielae* was host to florivorous flies in the Brazilian Chaco. This species of Asteraceae is considered rare in Brazil [16]. *Calea*

rupicola that was recorded on the APA trail (GWS) (=APA Waterfall Municipal Park = *Parque Municipal Cachoeira do Apa*) is considered endemic to the state of Mato Grosso do Sul, also reported in the Pantanal region [15]. *C. rupicola* has not been associated with florivorous flies in this research. In the state of Mato Grosso do Sul there are, at least, 32 endemic species of Asteraceae.

The Wood Steppe Savanna (WSS) presented pioneer plants such as *Conyza bonariensis* and *Erechtites hieracifolius*, indicating that environment as the most degraded. A conjecture of elements is related to the low diversity in this place: deconfiguration of the flora by human action, reducing the species richness of Asteraceae, and as well as, some punctual flooding during the summer period, caused by anthropic action.

In this study, the occurrence of *Pectis odorata* was reported in the Brazilian Chaco. This plant was common in flooded areas, mainly in the WSS. *Pectis gardneri*, presents adaptations for the common water deficit in the Brazilian Chaco, as pointed out by Antunes [17]. New floristic studies can answer if there are also adaptations to constant flooding, in species of Asteraceae and other taxa in the Brazilian Chaco.

Herein, we added new data about biodiversity of Tephritinae (Tephritidae) and Agromyzinae (Agromyzidae), presenting essential information to fight for environmental preservation, as well as contribute to the catalog of flora and fauna from Chaco. Human-caused phenomena, such as global warming and habitat destruction, have increasingly threatened the planet's biodiversity. Endemic species are the most susceptible to disappear, due to their ecological sensitivity. As there is a dependence on florivorous flies for their host Asteraceae, a relationship extremely species-specific, both taxa have a greater chance of co-extinction. The savannas are at high risk of species extinction, due to the loss of habitat being much greater than the conservation efforts by human community and political authorities.

Finally, in the Brazilian Chaco: *Cecidochares* sp.1 was the most abundant species of florivorous fly upon the Asteraceae flower heads. A species of this same genus, *Cecidochares connexa* (Tephritinae) has been successful employed for the biological control of an exotic Asteraceae in Ghana, West Africa (*Chromolaena odorata*). This plant species was introduced on Africa by decade of 1930 and to humid regions of tropical Asia, with negative impacts on agriculture and regional biodiversity [4].

Trupanea species were the most polyphagous in the Chaco. From the Heliantheae tribe of Asteraceae we recovered the highest abundance of *Melanagromyza* spp. (Agromyzinae, Agromyzidae). From the species Eupatorieae tribe were reared highest abundances of tephritids and their parasitoids (Hymenoptera). Park Steppe Savanna (PSS) was the phytophysiognomy among the three evaluated that presented the highest diversity (H′) of Tephritinae, probably, due to the heterogeneity of this phytophysiognomies in the Brazilian Chaco.

6. Conclusions and perspectives

1. In the Brazilian Chaco do occur at least 15 florivore fly species, nine of Tephritinae (Tephritidae), from five different genera, and six species of *Melanagromyza* (Agromyzinae, Agromyzidae);

2. The Grassy-Woody Savanna is the Chaquean phytophysiognomy that harbored higher species richness;

3. Some florivore fly (Tephritinae and Agromyzinae) species needs to better studied to employ in programs of biological control for invasive Asteraceae in the Neotropical Region.

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Conflict of interest

The authors declare no conflict of interest.

Recommendations

More sampling points are needed to represent the real diversity of florivorous flies in the Brazilian Chaco, as well as, to quantify and qualify the endophagous insect interactions with Asteraceae on the Chaco Biome (Argentina, Bolivia and Paraguay).

To have a better understanding and a refined quantifications and qualification on the relationships between florivorous fly species, their natural enemies and Asteraceae, is important individualize capitulum samples to obtain the infesting insects or their respective parasitoids.

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