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Interaction Network and Niche Analysis of Natural Enemy Communities and their Host Bees (Hymenoptera: Apoidea) in fragments of Cerrado and Atlantic Forest

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Abstract

Natural enemies are important components of solitary bee communities that nest in preexisting cavities because they act as a relevant mortality factor and can regulate host population growth. Nevertheless, the natural enemy-host interaction remains poorly investigated. This research aimed to determine the composition of the community, the structure of the interaction network, and niche overlap and breadth of natural enemy species in areas of Cerrado (Brazilian savanna) and Semideciduous seasonal forest (Atlantic Forest) in the state of São Paulo, Brazil. Trap-nests made of black cardboard and bamboo canes were provided in the field and inspected monthly in each area, from August 2001 to July 2003 at Cerrado and from June 2006 to May 2008 at the Semideciduous seasonal forest. A modular structure in the interaction network was observed for both areas with the populations of natural enemies showing high degrees of specialization. This structure confers higher stability against disturbances than less specialized webs since these adversities spread more slowly through the network. The niche analysis showed low degrees of overlap for both, trophic and temporal, among the natural enemy populations.

Introduction

Of the approximately 20,000 known species of bees more than 85% are not social but solitary (Batra, 1984) and among the solitary bees, 5% of them are species that nest in preformed cavities (Krombein, 1967; Batra, 1984). Irrespective of being social or solitary, the bees form the most important group of pollinators providing a crucial ecosystem service through their role in the sexual reproduction of both wild plants and crops (Klein et al., 2007; Le Féon et al., 2013).

The abundance of many bee species has been declining for a variety of reasons, including agricultural intensification, which includes the loss of natural habitats, agricultural practices, floral resource availability and, increased pesticide and herbicide use (Potts et al., 2010). This decline of species diversity has been shown to result in productivity decrease in many ecosystems (Tilman et al., 2001) while the diverse pollinator communities provide more stable and productive ecosystem services (Rogers et al., 2014).

Among social species, the honey bees are the primarily managed pollinator in agriculture and although the increased use of them may mitigate the loss of pollination services caused by the decline of solitary bees, they cannot entirely substitute the contribution of solitary bees to crop pollination (Winfree et al., 2008; Garibaldi et al., 2013). Therefore, a better knowledge of factors driving the population dynamics of solitary bees is essential for the future conservation of suitable habitats and ecological interactions (Kearns et al., 1998). Population dynamics can be driven by resources (bottom-up) or by natural enemies (top-down). Bottom-up factors mainly act as density-independent factors that limit population growth while top-down factors, i.e., natural enemies, can regulate population dynamics by positive density-dependent parasitism or predation (Berryman, 2001).

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Therefore, natural enemies are important components associated with populations comprising the bee communities, responsible for many adult and immature deaths (Rocha-Filho et al., 2017). Despite this, while mutualistic interactions between plant-pollinator have received much attention, the antagonistic interaction between natural enemies and their host bees remain poorly investigated.

In general, natural enemies of solitary bees are insects belonging to three orders, Hymenoptera, Diptera and Coleoptera (Krombein, 1967). Depending on the resources used as food they can be classified as cleptoparasites or parasitoids; the first ones use the larval food provisioned by the host to feed their own immature, and the second ones employ the host immature as an alimentary resource to their offspring (Roulston & Goodell, 2011)

Among the bees used as hosts, the species that nest in preformed cavities comprise a relevant component of the trophic niche of several natural enemy species (Krombein, 1967). These bee species perform a high variety of nesting behavior, use different substrates to nest, distinct floral sources to provision their nests, and show different reproductive phenologies (Michener, 2007). This high diversity observed in this bee group relates with the variety of attack strategies, alimentary preferences, and phenology of many natural enemies and influence their niche breadth, once all these biological aspects represent one dimension of the multidimensional space proposed by Hutchinson (1957) for ecological niches. Therefore, to know the natural history of these natural enemies, their temporal niche, and hosts preferences is essential to understand the community structure and to propose methodologies for prevention of high mortalities rates of bees in conservational and management plans.

So, the understanding this poorly studied natural enemy-host system added with the great diversity observed in both groups, host bees, and their natural enemies, makes them a good model for interaction networks and ecological niche approach.

Another essential component of the community structure, the interaction network, complement the niche study because it represents the interaction strength, their stability and how these interactions are organized among the species of the community (Thébault & Fontaine, 2010; Pocock et al., 2016). Also, this approach is important not only to understand the structure of interactions but also to identify patterns of responses to environmental changes (Massol & Petit, 2013; Osorio et al., 2015).

This study aimed to investigate the composition and structure of the interaction networks in two communities of natural enemies and their hosts, species of solitary bees that nest in preexisting cavities. Also, this study investigates the diet breadth of each species of natural enemy and the overlap of trophic and temporal niches among these species.

Material and Methods

Study areas

The study was carried out in an area of Cerrado (Brazilian savanna) of the Santa Cecilia Farm (henceforth referred as SCF) (20°46’ S, 47°61’ W), municipality of Patrocínio Paulista, and in a remnant of Semideciduous seasonal forest (Atlantic Forest) of the Estação Ecológica dos Caetetus (henceforth referred as EEC) (22°26’ S, 49°44’ W), municipality of Gália and Alvinlândia, both areas of the state of São Paulo, Brazil.

The cerrado has an area of 98 ha that together with 49 ha of semideciduous seasonal forest form the protected area reserve of the SCF. Both areas, cerrado, and forest, are continuous and contiguous, and practically untouched for more than four decades (Teixeira et al., 2004). Our sampling was carried out in the cerrado area. According to Köppen (1948), the local climate is classified as Aw, with a cold and dry season from April to September and a hot and wet season from October to March. During the dry periods, the monthly temperature ranges from 17.4 °C to 22.3 °C and the precipitation from 0.2 to 122.9 mm, and during the hot seasons, the monthly temperature ranges from 21.1 to 23.3 °C and the precipitation from 29.3 to 363.8 mm. The sampling occurred from August 2001 to July 2003 in this area.

The EEC has an area of 2,178.84 ha. The local climate is classified as Cwa (Köppen, 1948), with temperatures below 18 ºC in winter and above 22 ºC in summer. The average annual precipitation was 1,700 mm (Tabanez et al., 2005). The vegetation consists mostly of the well-preserved forest, including seasonal semideciduous and gallery forests, immersed in a matrix of agricultural land, containing coffee, pastures, and rubber tree (*Hevea brasiliensis* L., Euphorbiaceae) crops (Tabanez et al., 2005). In this area, the sampling occurred from June 2006 to May 2008.

Data collection

The nests were obtained using the trap-nest methodology proposed by Camillo et al. (1995), where artificial cavities made of black cardboard and bamboo canes were provided in the field as nesting substrate. The cardboard tubes were 6 cm long × 0.6 cm diameter and 8 cm long × 0.8 cm diameter, with one end closed with the same material. These tubes were inserted into cavities drilled in wooden plates (length: 30 cm, height: 12 cm, thickness: 5.0 cm). The bamboo canes, with variable diameters (from 0.8 to 1.5 cm) and lengths (from 10 to 15 cm), were cut so that the nodal septum closed one end of the cane, and all sizes were not equally represented. The bamboo canes were inserted into three PVC tubes with a length of 25 cm and a diameter of 10 cm.

At SCF, three plates with 55 small tubes each, one plate with 20 large tubes, and nine sets of bamboo canes, each set containing 10 to 15 canes, were placed. The PVC tubes and the plates were hung from trees randomly chosen and positioned 1.80 m above the ground. At EEC, the trap-nests were set in seven collection sites; each site had available 40 small tubes and 40 large tubes, and 120 bamboo canes. The trap-nests were put on supports and kept at the study sites fixed at the height of 1.5 m from the ground. To protect the trap-nests from the sun and the rain, the canes were put into PVC tubes, and each plate received a small cover of hard plastic.

The traps were inspected monthly in each area, and completed nests were taken to the laboratory, replaced by empty tubes and kept at room temperature until the emergence of the adults. After the emergence, the nests were opened, and the contents were analyzed. Voucher specimens of bees and natural enemies were deposited in the Entomological Collection of the Departamento de Biologia da Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto, University of São Paulo.

Diet Breadth, Niche overlap, and Network Analysis

The interaction strength between natural enemies and their hosts was attributed based on the number of brood cells attacked by each parasite, once only one individual of natural enemy developed in each brood cell parasitized. The natural enemy *Melittobia* sp. (Hymenoptera: Eulophidae) was not used in the analysis of niche breadth and overlap, and in the network analysis because it was not possible to attribute the interaction strength due to the strategy of attack performed by this species.

The trophic niche breadth of the communities was calculated using three diversity indices for each natural enemy species: Richness (S), that represents the total number of bee species used as host; Shannon-Wiener diversity index (H´), that represents the relation between the number and abundance of each bee species used as host through the formula $-\sum_{i=1}^{s} p_i \, ln \, p_i$, where Σ is the total number of bee species used as host by the natural enemy, p_i is the proportion of the number of the bee species *i* used as host in relation of the total number of host used, and the *ln* is the natural logarithm; and Pielou's evenness index, that indicates if the host bees were explored in a uniform way (J´ = 1) or no (J´ = 0).

Trophic and temporal niche overlap degree among all natural enemies species in each community, SCF and EEC, were calculated in the software TimeOverlap version 1.0 (Castro-Arellano et al., 2010) using the Pianka (1973) and Czechanowski's indices (Feinsinger et al., 1981). Utilizing the Rosario algorithm, a null-model analysis based on 10,000 randomizations was performed to determine if the community overlap values found were different from that expected by chance using a two-tailed test and a significance level of 5%. To estimate the niche overlap, a matrix of absolute abundance of the number of brood cells attacked for each host species (trophic) or number of attacks observed monthly (temporal) for each natural enemy species were used.

Overlap degrees of trophic and temporal niches for each pair of natural enemies species were calculated with the Schoener index (1986) using the formula $NO_{ih} = 1 - 1/2 \sum k \left| p_{ik} - p_{hk} \right|$ implemented in the package spaa 0.2.2 for R version 3.4.3 (R Core Team, 2017), where *i* and *h* are the pair of natural enemy species compared, and *pik* and *phk* are the number of brood cell attacked for each host species *k* (trophic) or the total number of brood cells attacked in each month *k* (temporal).

In order to describe the natural enemies-hosts interactions, a weighted network was built for each sampling local (SCF and EEC) from a matrix of absolute abundance in the software Pajek version 5.03 (Mrvar and Batagelj, 2018) using the 'Kamada-Kawai free' method, in which natural enemy and host species were connected to one another. The degree of specialization in the diet of the populations was calculated in the software R version 3.4.2 (R Core Team 2017) measuring the $H_2´$ index (Blüthgen et al., 2006) for each community. The significance of this index was estimated with a Monte Carlo procedure using a null model (Patefield, 1981) with the generation of 1,000 random matrices.

The nestedness of each network was calculated in the software NODF version 2.0 based on the weighted metric WNODF (Almeida-Neto & Ulrich, 2011) . The significance of this index was estimated with a Monte Carlo procedure in which 1,000 random matrices were generated using a null model and the randomization algorithm rc (Ulrich & Gotelli, 2010; Almeida-Neto & Ulrich, 2011).

Weighted modularity was calculated for each network using the 'ComputeModules' function in a bipartite package for R software version 3.4.4.3 (R Core Team, 2017), which uses the QuanBioMo algorithm (Q) for quantitative data matrices (Dormann & Strauss, 2014). The number of steps taken for the analysis was 10 x 10^5. Modularity was tested against 1,000 null models using the method 'r2d', which yielded a score z_Q, equivalent to the z-score of a normal distribution; following the proposed by Dormann and Strauss (2014), values of z_Q above than 2 represents significant modularity. The modules formed by this analysis were represented in the network using different colors patterns.

To observe the 'network functional role'of each species of natural enemies and hosts, the 'czvalues' function based on species weighted (Bascompte et al., 2006) was used to calculated the connection (c-values) and participation values (z-values), indicating the contribution among and within modules, respectively (Olesen et al., 2007). Using the critical values showed by Olesen et al. (2007), 0.625 for c-value and 2.5 for z-value, the species were classified in specialists (low c and z), module hubs (low c and high z), connectors (high c and low z), and network hubs (high c and z). These three last classifications represent generalist organisms. Species with no z-score (= NA) were not shown in the graph.

Results

At SCF, 12 species of natural enemies attacked 49 nests of seven species of solitary bees. These natural enemies belong to four insect orders, Hymenoptera, Diptera, Coleoptera, and Neuroptera. According to the attack behavior, more parasitoid species than cleptoparasites were sampled (Table 1, Fig 1). During the period of study, we found asymmetry in the phenology of the natural enemy species, as in 2003 when only *Coelioxoides exulans* (Holmberg) was sampled (Fig 1).

The natural enemies with the highest number of hosts were *C. exulans* (4 species), *Leucospis cayennensis* Westwood (3 species), *Anthrax hylaios* Marston and *Anthrax oedipus* Fabricius (2 species). These parasites presented higher richness and diversity of host bees, and *C. exulans* attacked the highest number of brood cells (n = 29) (Tables 1 and 2).

Table 1. Natural enemies and their host bees that nested in trap-nests at Cerrado of the Santa Cecília Farm, SP, Brazil, from August 2001 to July 2003. a Parasitoid, b Cleptoparasite, NN = Number of attacked nests, NC = Number of attacked brood cells, and Code = Code of species used in the network.

Natural enemies		**Code**	**Host bees**	**Code**	**NN**	**NC**
Coleoptera (Meloidae)	*Nemognatha* sp.[b]	Ne_sp	*Centris analis*	Ce_an	1	1
			Tetrapedia rugulosa	Te_ru	3	3
Diptera (Bombyliidae)	*Anthrax aquilus*[a]	An_aq	*Tetrapedia diversipes*	Te_di	1	1
	Anthrax hylaios[a]	An_hy	*Tetrapedia diversipes*		1	1
			Tetrapedia rugulosa		3	3
	Anthrax oedipus[a]	An_oe	*Tetrapedia diversipes*		3	3
			Tetrapedia rugulosa		2	2
Hymenoptera (Apidae)	*Coelioxoides exulans*[b]	Cx_ex	*Tetrapedia diversipes*		3	3
			Tetrapedia curvitarsis	Te_cu	10	10
			Tetrapedia rugulosa		8	15
			Tetrapedia garofaloi	Te_ga	1	1
	Coelioxoides cf. *waltheriae*[b]	Cx_wa	*Tetrapedia* sp.	Te_sp	1	1
	Mesocheira bicolor[b]	Me_bi	*Centris tarsata*	Ce_ta	1	1
Hymenoptera (Eulophidae)*	*Melittobia* sp.*		*Anthodioctes* sp.		1	-
Hymenoptera (Leucospidae)	*Leucospis cayennensis*[a]	Le_ca	*Centris analis*		1	1
			Centris tarsata		2	2
			Tetrapedia diversipes		1	1
	Leucospis manaica[a]	Le_ma	*Tetrapedia rugulosa*		5	5
Hymenoptera (Vespoidea)	Mutillidae[a]	Muti	*Tetrapedia rugulosa*		1	1
Neuroptera	Mantispidae[a]	Mant	*Tetrapedia rugulosa*		1	1
Total					**49**	**56**

* Species not used in the analysis of this study (see Material and Methods)

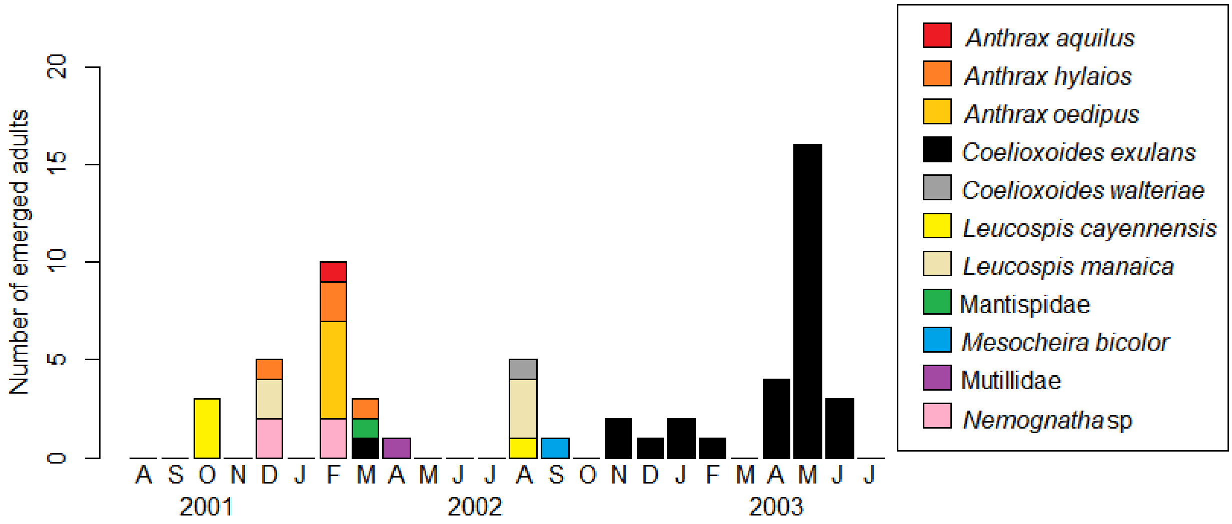

Fig 1. Phenology of the natural enemy species at Cerrado of the Santa Cecília Farm, SP, Brazil. The sampling period was from August 2001 to July 2003. Each adult emerged from one host brood cell.

Table 2. Diversity indices, Richness (S), Shannon diversity (H´) and Pielou's evenness (J´), for each natural enemy species at Cerrado of the Santa Cecília Farm and Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil. The sampling period at SCF was from August 2001 to July 2003, and from June 2006 to May 2008 at EEC.

Natural enemy		**SCF**			**EEC**		
		S	**H´**	**J´**	**S**	**H´**	**J´**
Coleoptera (Meloidae)	*Nemognatha* sp.	2	0.64	0.92	2	0.66	0.95
Diptera (Bombyliidae)	*Anthrax aquilus*	1	0	-			
	Anthrax hylaios	2	0.56	0.81			
	Anthrax oedipus	2	0.67	0.97			
	Anthrax sp.				2	0.26	0.37
Hymenoptera (Apidae)	*Coelioxoides exulans*	4	1.06	0.76	1	0	-
	Coelioxoides cf. *waltheriae*	1	0	-	3	0.69	0.63
	Coelioxoides sp.				2	0.69	1.00
	Mesocheira bicolor	1	0	-			
	Mesocheira sp.				2	0.60	0.86
Hymenoptera (Megachilidae)	*Coelioxys* sp.				4	1.21	0.88
Hymenoptera (Leucospidae)	*Leucospis cayennensis*	3	1.04	0.95			
	Leucospis manaica	1	0	-			
	Leucospis sp.				4	0.76	0.55
Hymenoptera (Vespoidea)	Mutillidae	1	0	-			
Lepidoptera	Pyralidae				3	0.96	0.87
Neuroptera	Mantispidae	1	0	-			

The network was modular, comprised of five modules, and not nested, with the community of natural enemies showing high specialization (Fig 2, Table 3).

The community of natural enemies showed low levels of trophic and temporal overlap. The paired analysis showed that only a few species of natural enemy overlap their trophic

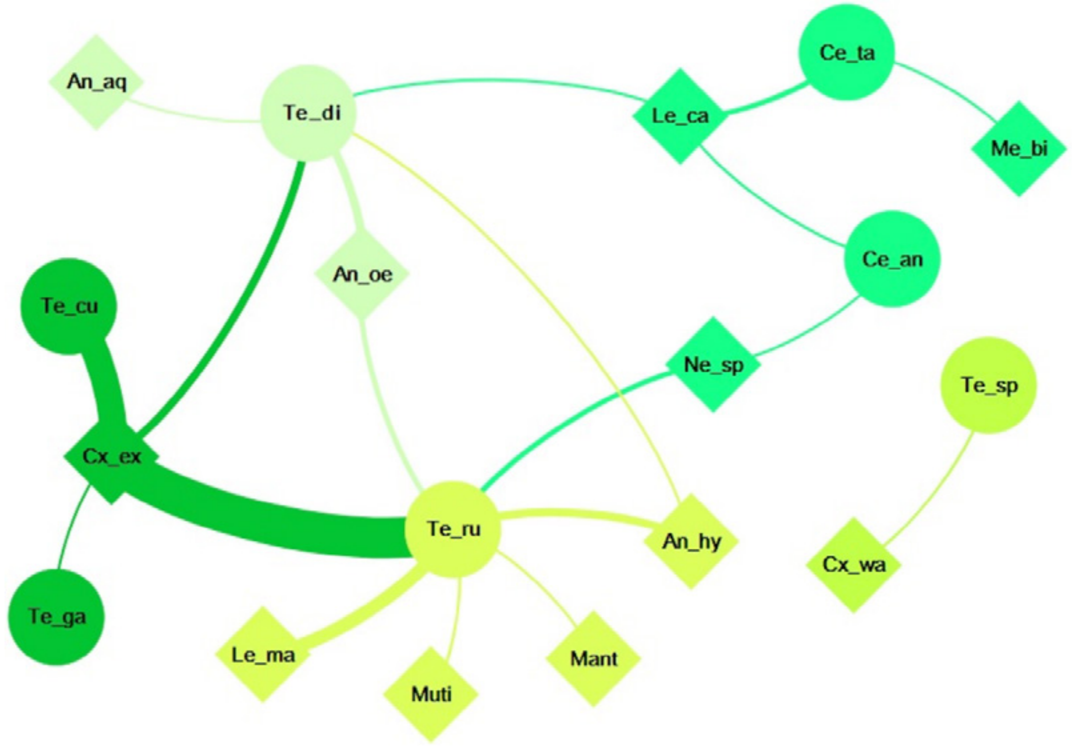

Fig 2. Interaction network between natural enemies and their hosts that nested in trap-nests at Cerrado of the Santa Cecília Farm, SP, Brazil, from August 2001 to July 2003. The circles represent the hosts, and the diamonds represent the natural enemy species. The thickness of the lines represents the interaction strength (number of parasitized brood cells). Each color pattern indicates a distinct module. Natural enemies and hosts' codes displayed in Table 1.

niches, highlighting the interaction observed between the bee fly *A. hylaios* and *Leucospis manaica* Roman (NOih = 0.75). Maximal values of trophic overlap (NOih = 1) were observed in natural enemies with rare occurrence, as for Mantispidae and Mutillidae. Paired temporal niche analysis did not show high overlap among the natural enemies populations, with similar phenology being observed only between few species, as for the cleptoparasite beetle *Nemognatha* sp. and the parasitoid *A. hylaios* (NOih = 0.75), and between the two bee fly species *Anthrax aquilus* Marston and *A. oedipus* (NOih = 1) (Tables 3 and 4).

For both, natural enemies and hosts, all species of the community performed a specialist behavior in the network, showing low levels in the interaction strength between (c-values) and within modules (z-values). The parasitoid *A. hylaios* showed the highest connectivity levels within the natural enemies species (c-value = 0.5), while the bee *Tetrapedia rugulosa* Friese showed the highest within the host species (c-value = 0.48). No species showed z-values above 1.3 (Fig 5, Supplementary Material 1).

Table 3. Weighted nestedness (WNODF), weighted modularity (Q) with the z-score (zQ), number of modules shaped (Mod), specialization degree (H2´), and trophic and temporal niche overlap metrics (Pianka and Czechanowski) calculated for the natural enemies at Cerrado of the Santa Cecília Farm (sampling from August 2001 to July 2003) and Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil (sampling from June 2006 to May 2008).

	Interaction Network					**Trophic niche overlap**		**Temporal niche overlap**	
	WNODF	**Q**	**Mod**	**z_Q**	**$H_2´$**	**Pianka**	**Czechanowski**	**Pianka**	**Czechanowski**
SCF	19.40^{NS}	0.32^{*}	5	3.72	0.85^{***}	0.37^{**}	0.29^{**}	0.14^{*}	0.11^{*}
EEC	26.33^{**}	0.31^{***}	4	11.41	0.63^{***}	0.46^{**}	0.37^{**}	0.18^{NS}	0.14^{NS}

Values significant for a * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, and ^{NS} for non significant values.

Table 4. Trophic (below) and temporal (above) niche overlap for each pair of natural enemy species at Cerrado of the Santa Cecília Farm, SP, Brazil, from August 2001 to July 2003. Highest values in bold. Natural enemies' codes displayed in Table 1.

	Ne_ sp	**Le_ca**	**Le_ma**	**Me_bi**	**Cx_ex**	**Mant**	**Muti**	**An_oe**	**An_hy**	**An_aq**	**Cx_wa**
Ne_sp	-	0	0.4	0	0	0	0	0.5	**0.75**	0.5	0
Le_ca	0.25	-	0.25	0	0	0	0	0	0	0	0.25
Le_ma	0.67	0	-	0	0	0	0	0	0.25	0	0.6
Me_bi	0	0.5	0	-	0	0	0	0	0	0	0
Cx_ex	0.52	0.1	0.52	0	-	0.03	0	0	0.03	0	0
Mant	0.67	0	**1**	0	0.52	-	0	0	0.25	0	0
Muti	0.67	0	**1**	0	0.52	**1**	-	0	0	0	0
An_oe	0.4	0.25	0.4	0	0.5	0.4	0.4	-	0.5	**1**	0
An_hy	0.67	0.25	**0.75**	0	0.62	**0.75**	**0.75**	0.65	-	0.5	0
An_aq	0	0.25	0	0	0.1	0	0	0.6	0.25	-	0
Cx_wa	0	0	0	0	0	0	0	0	0	0	-

At EEC, nine species of natural enemies attacked 148 nests of seven species of solitary bees. The natural enemies belong to four orders, Hymenoptera, Diptera, Coleoptera, and Lepidoptera. According to the attack behavior, we found a higher number of cleptoparasites than parasitoids (Table 2, Fig 3). Excepting Pyralidae, all natural enemy species were sampled in both years of study (Fig 3).

The natural enemies with the highest number of host bees were *Coelioxys* sp. (4 species), *Leucospis* sp. (4 species), *Coelioxoides* cf. *waltheriae* Ducke (3 species) and Pyralidae (3 species) (Tables 2 and 5, Fig 3). *Coelioxoides* cf. *waltheriae* and *Nemognatha* sp. were the cleptoparasites more frequent, attacking 66 and 56 cells respectively (Table 5).

The network presented a low degree of nestedness and a modular structure, with populations with a specialized diet, and four modules were formed in the network (Table 3*,* Fig 4).

The analysis showed trophic niche overlap among the natural enemy populations. Several species of natural enemies presented a high trophic overlap, as the interactions between *C. exulans* and *Anthrax* sp. (NOih = 0.93), *Nemognatha* sp. and *Coelioxys* sp. (NOih = 0.86), and *Nemognatha* sp. and *Coelioxoides* sp. (NOih = 0.86). The temporal analysis did not show overlap among the natural enemy populations, and in the paired analysis, the highest values were observed between *Coelioxys*. sp. and *Leucospis*. sp. (NOih = 0.5), and *Nemognatha* sp. and Pyralidae (NOih = 0.4) (Tables 3 and 6).

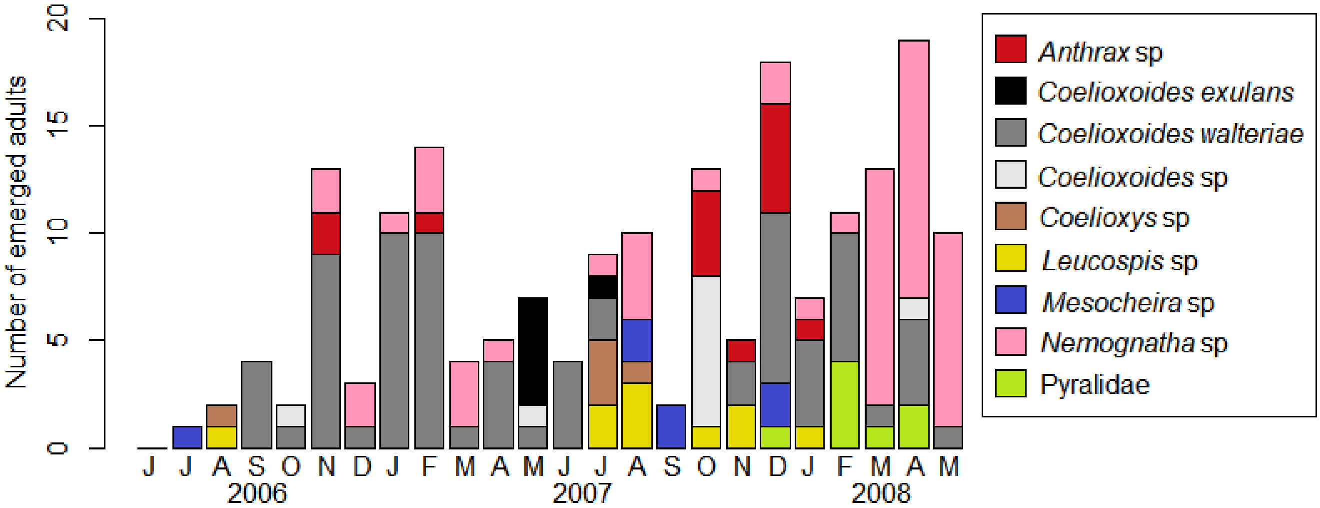

Fig 3. Phenology of the natural enemy species at the Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil, from June 2006 to May 2008. Each adult emerged from one host brood cell.

As observed for SCF, all species of natural enemies and hosts exhibited a specialized behavior at EEC, with low c and z-values. The cleptoparasite *Nemognatha* sp. and the host *Tetrapedia diversipes* Klug showed the highest c-values, closed to 0.6, and no species showed z-values above 1.2 (Fig 5, Supplementary Material 1).

Both communities were composed by similar groups of parasites and hosts. *Tetrapedia* species were the most abundant host, while *Coelioxoides* species were the most abundant natural enemy sampled. Besides, *Centris* and *Mesocheira* bees, *Nemognatha* beetles, *Leucospis* wasps and *Anthrax* bee-flies were also important components of the two communities.

Table 5. Natural enemies and their host bees that nested in trap-nests at the Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil, from June 2006 to May 2008. a Parasitoid, b Cleptoparasite, c Nest destroyer, NN = Number of attacked nests, NC = Number of attacked cells, and Code = Code of species used in the network.

Natural enemies		**Code**	**Host bees**	**Code**	**NN**	**NC**
Coleoptera (Meloidae)	*Nemognatha* sp.[b]	Ne_sp	*Tetrapedia diversipes*	Te_di	17	20
			Tetrapedia sp.	Te_sp	30	35
Diptera (Bombyliidae)	*Anthrax* sp.[a]	An_sp	*Tetrapedia diversipes*		12	13
			Tetrapedia sp.		1	1
Hymenoptera (Apidae)	*Coelioxoides exulans*[b]	Cx_ex	*Tetrapedia diversipes*		4	6
	Coelioxoides cf. *waltheriae*[b]	Cx_wa	*Tetrapedia diversipes*		43	50
			Tetrapedia rugulosa	Te_ru	2	4
			Tetrapedia sp.		9	12
	Coelioxoides sp.[b]	Cx_sp	*Tetrapedia diversipes*		3	4
			Tetrapedia sp.		3	4
	Mesocheira sp.[b]	Me_sp	*Centris tarsata*	Ce_ta	3	5
			Centris sp.2	Ce_sp2	1	2
Hymenoptera (Megachilidae)	*Coelioxys* sp.[b]	Co_sp	*Centris analis*	Ce_an	1	1
			Centris tarsata		1	1
			Centris sp.	Ce_sp	1	1
			Centris sp.2		1	3
Hymenoptera (Leucospidae)	*Leucospis* sp.[a]	Le_sp	*Centris analis*		1	1
			Centris sp.		1	1
			Tetrapedia diversipes		6	6
Lepidoptera	Pyralidae[c]	Pyra	*Centris analis*		1	1
			Tetrapedia diversipes		2	2
			Tetrapedia sp.		5	5
Total					**148**	**178**

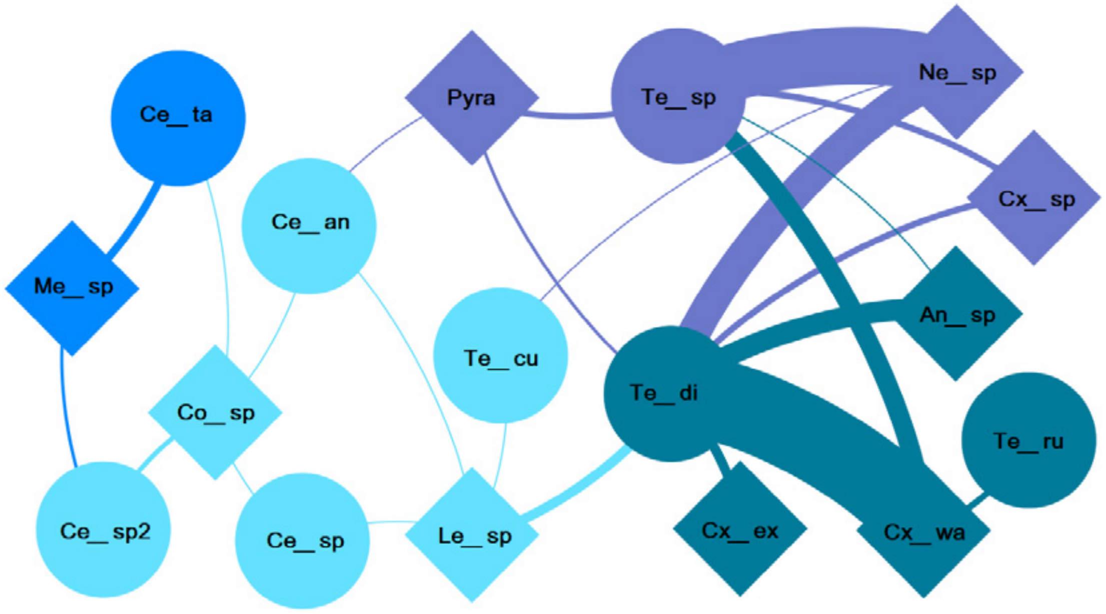

Fig 4. Interactions network between natural enemies and their hosts that nested in trap-nests at the Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil, from June 2006 to May 2008. The circles represent the hosts, and the diamonds represent the natural enemy species. The thickness of the lines represents the interaction strength (number of parasitized brood cells). Each color pattern indicates a distinct module. Natural enemies and hosts' codes displayed in Table 5.

Table 6. Trophic (below) and temporal (above) niche overlap for each pair of natural enemy species at the Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil, from June 2006 to May 2008. Highest values in bold. Natural enemies' code displayed in Table 1.

	Le_sp	**Co_sp**	**Pyra**	**Me_sp**	**Cx_wa**	**Cx_sp**	**An_sp**	**Cx_ex**	**Ne_sp**
Le_sp	-	**0.5**	0	0.29	0.11	0.1	0.24	0.17	0.13
Co_sp	0.22	-	0	0.2	0.03	0	0	0.17	0.09
Pyra	0.4	0.14	-	0.13	0.26	0.1	0.13	0	**0.4**
Me_sp	0	0.45	0	-	0.11	0	0.29	0	0.11
Cx_wa	0.67	0	0.47	0	-	0.08	**0.39**	0.04	0.33
Cx_sp	0.5	0	**0.79**	0	0.68	-	0.29	0.1	0.12
An_sp	0.67	0	0.36	0	**0.83**	0.57	-	0	0.17
Cx_ex	0.67	0	0.29	0	**0.76**	0.5	**0.93**	-	0.02
Ne_sp	0.38	0	**0.86**	0	0.54	**0.86**	0,.43	0.36	-

Discussion

The communities here studied were composed of similar families and genera of natural enemies and hosts, and the natural enemies showed identical richness and diversity values of hosts used. It was also observed a modular network with high levels of specialization in the interactions between its component members, natural enemies, and their hosts.

The modular structure is expected in antagonistic interactions where the predators, or natural enemies in this study, tend to optimize their attacks, and the preys, or hosts, tend to optimize their defenses, in an arms race, increasing the module formation in the web (Thébault & Fontaine, 2010). However, unlike what is reported here, high specialization degrees are not always observed in this network structure (Araujo et al., 2018). Organisms with preferences in the diet are more likely to occur in habitats structurally more diverse and poorly isolated (Pereira-Peixoto et al., 2016; Araujo et al., 2018). Both areas sampled in this study are surrounded by urban and agricultural landscapes, and despite the low diet breadth and the specialized performance observed in the network, the majority of the natural enemy species were reported attacking several host species, characterizing a generalist organism in an ecological concept.

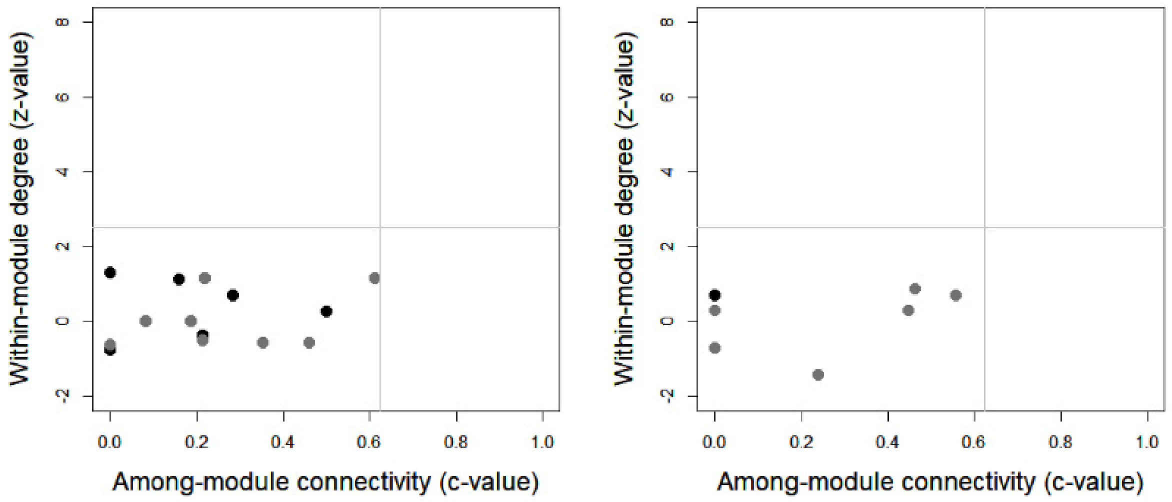

Fig 5. Connection (c-value) and participation (z-value) values for natural enemies (left) and hosts species (right) in the network of Cerrado of the Santa Cecília Farm, SP, Brazil (black points) (sampling from August 2001 to July 2003) and of the Semideciduous seasonal forest of the Estação Ecológica dos Caetetus, SP, Brazil (gray points) (sampling from June 2006 to May 2008). Following Olesen et al. (2007), the critical values (gray lines) for c is 0.625, and for z is 2.5.

According to Michener (2007), cleptoparasite bees usually attack nests of related lineages. This association between host-cleptoparasite would explain the attacks of the species of the genus *Coelioxoides* for the nests of their sister genus *Tetrapedia*, as had been reported by several authors (Aguiar et al., 2005; Gazola & Garófalo, 2009; Rocha-Filho et al., 2017).

The high abundance of *Coelioxoides* parasites observed in both communities is related with the high abundance of their available host, *Tetrapedia* bees, once parasites must synchronize their life cycles with their host's life cycles (Wcislo, 1987).

Among other species that attack closed phylogenetical taxon are the cuckoo bees of the genus *Coelioxys*, cleptoparasites of many Megachilidae bees (Krombein, 1967). However, differently of the Tetrapediini parasites and as observed in this study, *Coelioxys* exhibited a less specialized behavior, parasitizing nests of others bee groups, as the species of the genus *Centris*. This association with nests of the oil-collecting bees has also been reported by other authors such as Morato et al. (1999), Aguiar and Martins (2002), Gazola and Garófalo (2009), Araujo et al. (2018), Oliveira and Gonçalves (2017), Rocha-Filho et al. (2017) and Araujo et al. (2018).

Species analyzed in this study and that are the natural enemies of many bees and wasps species, as *Anthrax* and *Leucospis* (Krombein, 1967), showed a restrict diet breath due to the limited temporal niche. The same situation was observed for others parasites sampled, as the Pyralidae moth and the mantisflies, who were reported parasitizing many insects groups (Buys, 2008; Hook et al., 2010), but in this study, they behave as a more specialized organism in the network analysis, attacking few host species.

These aspects of the trophic and temporal niche contributed to the formation of a modular structure in the interaction network. It is worth mentioning that many of the natural enemies analyzed in this study use other groups as hosts, like eusocial bees, ground-nesting bees, and other insects; thus, in the present study was only evaluated a part of the trophic niche of some natural enemies. As other studies showed a modular pattern for this system even considering a broader niche breath for these natural enemies (Araújo et al. 2018), we expected that our network structure maintain the modularity even adding others groups of host.

Under an individual and populational perspective, the niche specialization can favor the optimizing in the cost-benefit relation, but also can increase the chance of extinction in face to environmental changes (Biesmeijer et al., 2006). However, in a perspective of the community, the modular structure and the low connections for the nodes observed in our study indicate that the host-natural enemy interactions at SCF and EEC would present a high persistence and resilience against disturbances (Thébault & Fontaine, 2010).

Although the sampling effort, years and phytophysiognomy were different, the communities of SCF and EEC were structured by similar groups of natural enemies and host bees. Likewise, other authors reported similar composition in distinct Brazilian areas (Aguiar & Martins, 2002; Gazola & Garófalo, 2009; Mesquita & Augusto, 2011; Oliveira & Gonçalves, 2017; Rocha-Filho et al., 2017; Araújo et al., 2018), which evidences a closed relationship between these parasites species and their host bees. Evolutionary history and phylogenetic diversity are factors that strongly influence these ecological interactions (Michener, 2007; Staab et al., 2016; Andreazzi et al., 2017).

In conclusion, this study described the diversity of natural enemies and the network interaction structure in the community. Preferences and seasonality strongly influenced the richness and abundance of these parasites and their interactions with each other and with their hosts. Another point, the similarity observed among communities and time evidence the close relationship between these parasite species and their hosts. Our study contributes to a better understanding of the complex and poorly studied natural enemy-host interaction, and provides relevant information on the diet breadth of these important components of the communities.

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Supplementary Material

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References

Andreazzi, C.S., Thompson, J.N., & Guimarães Jr, P.R. (2017). Network structure and selection asymmetry drive coevolution in species-rich antagonistic interactions. The American Naturalist, 190: 99-115. doi: 10.1086/692110.

Aguiar, A.J.C. & Martins, C.F. (2002). Abelhas e vespas solitárias em ninhos-amadilha na Reserva Biológica Guaribas (Mamanguape, Paraíba, Brasil). Revista Brasileira de Zoologia, 19: 101-116. doi: 10.1590/S0101-81752002000500005.

Aguiar, C.M.L., Garófalo, C.A. & Almeida, G.F. (2005). Trap-nesting bees (Hymenopera, Apoidea) in areas of dry semideciduous forest and Caatinga, Bahia, Brazil. Revista Brasileira de Zoologia, 22: 1030-1038. doi: 10.1590/S0101-81752005000400031.

Almeida-Neto, M. & Ulrich, W. (2011). A straightforward computational approach for measuring nestedness using quantitative matrices. Environmental Modelling & Software, 2: 173-178. doi:10.1016/j.envsoft.2010.08.003.

Araujo, G.J.; Fagundes, R. & Itabaiana, Y.A. (2018). Trap-nesting Hymenoptera and their network with parasites in recovered Riparian forests Brazil. Neotropical Entomology, 2: 1-11. doi: 10.1007/s13744-017-0504-4.

Bascompte, J., Jordano, P. & Olesen, J.M. (2006). Asymmetric coevolutionary networks facilitate biodiversity maintenance. Science, 312:431-433. doi: 10.1126/science.1123412.

Batra, S.W.T. (1984). Solitary bees. Scientific American, 250: 120-127. doi: 10.1038/scientificamerican0284-120

Berryman, A.A. (2001). Functional web analysis: detecting the structure of population dynamics from multi-species time series. Basic and Applied Ecology, 2: 311-321. doi: 10.1078/1439-1791-00069.

Biesmeijer, J.C., Roberts, S.P.M., Reemer, M., Ohlemüller, R., Edwards, M., Peeters, T., Schaffers, A.P., Potts, S.G., Kleukers, R., Thomas, C.D., Settele, J. & Kunin, W.E. (2006). Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. Science, 313: 351-354. doi: 10.1126/science.1127863.

Blüthgen, N., Menzel, F. & Blüthgen, N. (2006). Measuring specialization in species interaction networks. Ecology, 6: 1-12. doi: 10.1186/1472-6785-6-9.

Buys, S.C. (2008). Observations on the biology of *Anchieta fumosella* (Westwood 1867) (Neuroptera: Mantispidae) from Brazil. Tropical Zoology, 21: 91-95.

Camillo, E., Garófalo, C.A., Serrano, J.C. & Muccillo, G. (1995). Diversidade e abundância sazonal de abelhas e vespas solitárias em ninhos-armadilha (Hymenoptera, Apocrita, Aculeata). Revista Brasileira de Entomologia, 39: 459-70.

Castro-Arellano, I., Lacher, T. Jr., Willig, M.R. & Rangel, T.F. (2010). Assessment of assemblage-wide temporal niche segregation using null models. Methods in Ecology and Evolution, 1: 311-318. doi: 10.1111/j.2041-210X.2010.00031.x.

Dormann, C.F. & Strauss, R. (2014). A method for detecting modules in quantitative bipartite networks. Methods in Ecology and Evolution, 5: 90-98. doi: 10.1111/2041-210X.12139.

Feinsinger, P., Spears, E.E. & Poole, R.W. (1981). A simple measure of niche breadth. Ecology, 62: 27-32. doi: 10.2307/1936664.

Garibaldi, L.A., Steffan-Dewenter, I., Winfree, R., Aizen, M.A., Bommarco, R., Cunningham, S.A. & Klein, A.M. (2013). Wild pollinators enhance fruit set of crops regardless of honey bee abundance. Science, 339: 1608–1611. doi: 10.1126/science.1230200

Gazola, A.L. & Garófalo, C.A. (2009). Trap-nesting bees (Hymenoptera: Apoidea) in forest fragments of the State of São Paulo, Brazil. Genetics and Molecular Research, 8: 607-622.

Hook, A.W., Oswald, J.D. & Neff, J.L. (2010). *Plega hagenella* (Neuroptera: Mantispidae) parasitism of *Hylaeus* (*Hylaeopsis*) sp. (Hymenoptera: Colletidae) reusing nests of *Trypoxylon manni* (Hymenoptera: Crabronidae) in Trinidad. Journal of Hymenoptera Research, 19: 77-83.

Hutchinson, G.E. (1957). Concluding remarks. Cold Spring Harbor Symposia on Quantitative Biology, 22: 415-427. doi:

10.1101/SQB.1957.022.01.039.

Kearns, C.A., Inouye, D.W. & Waser, N.M. (1998). Endangered mutualisms: The conservation of plant-pollinator interactions. Annual Review of Ecology, Evolution and Systematic, 29, 83–112. doi: 10.1146/annurev.ecolsys.29.1.83.

Köppen, W.P. (1948). Climatologia. Fundo de Cultura Econômica. Cidade do México, Buenos Aires: 479 p.

Klein, A.M., Vaissière, B.E., Cane, J.H., Steffan-Dewenter, I., Cunningham, S.A., Kremen, C. & Tscharntke, T. (2007). Importance of pollinators in changing landscapes for world crops. Proceedings of the Royal Society B: Biological Science, 274: 303–313. doi: 10.1098/rspb.2006.3721.

Krombein, K.V. (1967). Trap-nesting wasps and bees: life histories, nests and associates. Washington: Smithsonian Press, 510 p.

Le Féon, V., Burel, F., Chifflet, R., Henry, M., Ricroch, A.E., Vaissière, B.E. & Baudry, J. (2013). Solitary bee abundance and species richness in dynamic agricultural landscapes. Agriculture, Ecosystems and Environment, 166: 94-101. doi: 10.1016/j.agee.2011.06.020.

Massol, F., & Petit, S. (2013). Interaction networks in agricultural landscape mosaics. In Advances in Ecological Research, 49: 291-338. doi: 10.1016/B978-0-12-420002-9.00005-6.

Mesquita, T.M.S. & Augusto, S.C. (2011). Diversity of trap-nesting bees and their natural enemies in the Brazilian savanna. Tropical Zoology, 24: 127-144

Michener, C.D. (2007). The Bees of the World. 2nd Edition. Baltimore: The John Hopkins University Press, 953 p

Morato, E.F., Garcia, M.V.B. & Campos, L.A.O. (1999). Biologia de *Centris* Fabricius (Hymenoptera, Anthoporidae, Centridini) em matas contínuas e fragmentos na Amazônia Central. Revista Brasileira de Zoologia, 16: 1213-1222. doi: 10.1590/S0101-81751999000400029.

Mrvar, A. & Batagelj, V. (2018). Pajek and Pajek-XXL, Programs for Analysis and Visualization of Very Large Networks, Reference Manual. 112 p

Oliveira, P.S. & Gonçalves, R.B. (2017). Trap-nesting bees and wasps (Hymenoptera, Aculeata) in a Semidecidual Seasonal Forest fragment, southern Brazil. Papéis Avulsos de Zoologia, 57:149-156. doi: 10.11606/0031-1049.2017.57.13.

Olesen, J.M., Bascompte, J., Dupont, Y.L. & Jordano, P. (2007). The modularity of pollination networks. Proceedings of the National Academy of Sciences, 104: 19891-19896. doi: 10.1073/pnas.0706375104.

Osorio, S., Arnan, X., Bassols, E., Vicens, N. & Bosch, J. (2015). Local and landscape effects in a host–parasitoid interaction network along a forest–cropland gradient. Ecological Applications, 25: 1869-1879. doi: 10.1890/14-2476.1.

Patefield, W.M. (1981). Algorithm AS159. An efficient method of generating r x c tables with given row and column totals. Journal of Applied Statistics, 30: 91-97.

Pereira-Peixoto, M.H., Pufal, G., Staab, M., Martins, C.F. & Klein, A.M. (2016). Diversity and specificity of host-natural enemy interactions in an urban-rural interface. Ecological Entomology, 41: 241-252. doi: 10.1111/een.12291.

Pianka, E.R. (1973). The structure of lizard communities. Annual Review of Ecology and Systematics, 4:53-74. doi: 10.1073/pnas.71.5.2141.

Pocock, M.J.O., Evans, D.M., Fontaine, C., Harvey, M., Julliard, R., McLaughlin, Ó., Silvertown, J., Tamaddoni-Nezhad, A., White, P.C.L.& Bohan, D.A. (2016). The Visualization of Ecological Networks, and Their Use as a Tool for Engagement, Advocacy and Management. In G. Woodward & D.A. Bohan (Eds.), Ecosystem Services: From Biodiversity to Society (pp. 41-85). Academic Press.

Potts, S.G., Biesmeijer, J.C., Kremen, C., Neumann, P., Schweiger, O. & Kunin, W.E. (2010). Global pollinator declines: Trends, impacts and drivers. Trends in Ecology & Evolution, 25: 345–353. doi: 10.1016/j.tree.2010.01.007.

R Core Team. (2017). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Rocha-Filho, L.C., Rabelo, L.S., Augusto, S.C. & Garófalo, C.A. (2017). Cavity-nesting bees and wasps (Hymenoptera: Aculeata) in a semi-deciduous Atlantic forest fragment immersed in a matrix of agricultural land. Journal of Insect Conservation, 21: 727-736. doi: 10.1007/s10841-017-0016-x.

Rogers, S.R., Tarpy, D.R. & Burrack, H.J. (2014). Bee species diversity enhances productivity and stability in a perennial crop. PlosOne, 9: e97307. doi: 10.1371/journal.pone.0097307.

Roulston, T.H. & Goodell, K. (2011). The role of resources and risks in regulating wild bee populations. Annual Review of Entomology, 56: 293-312. doi: 10.1146/annurev-ento-120 709-144802.

Schoener, T.W. (1986). Resource partitioning. In: J. Kikkawa & D.J. Anderson (Eds.), Community ecology pattern and process, (pp. 91-126). London: Blackwell Scientific.

Staab, M., Bruelheide, H., Durka, W., Michalski, S., Purschke, O.,Zhu, C.D., & Klein, A.M. (2016). Tree phylogenetic diversity promotes host–parasitoid interactions. Proceedings of the Royal Society B, 283:20160275. doi: 10.1098/rspb.2016.0275.

Tabanez, M.F., Durigan, G., Keuroghlian, A., Barbosa, A.F., Freitas, C.A., Silva, C.E.F. & Mattos, I.F.A. (2005). Plano de manejo da Estação Ecológica dos Caetetus. Instituto Florestal Série Registros, 29: 1-104.

Thébault, E. & Fontaine, C. (2010). Stability of Ecological Communities and the Architecture of Mutualistic and Trophic

Networks, Science 1: 329- 853. doi: 10.1126/science.1188321.

Teixeira, M.I.J.G., Araújo, A.R.B., Valeri, S.V. & Rodrigues, R.R. (2004). Florística e fitossociologia de área de cerrado S.S. no município de Patrocínio Paulista, nordeste do Estado de São Paulo. Bragantia, 63: 1-11.

Tilman, D., Reich, P.B., Knops, J., Wedin, D., Mielke, T. & Lehman, C. (2001). Diversity and productivity in a long-term grassland experiment. Science, 294: 843-845. doi: 10.1126/science.1057544.

Ulrich, W. & Gotelli, N.J. (2010). Null model analysis of species associations using abundance data. Ecology, 91: 3384-3397. doi: 10.1890/09-2157.1.

Wcislo, W.T. (1987). The roles of seasonality, host synchrony, and behaviour in the evolutions and distributions of nest parasites in the Hymenoptera (Insecta), with special reference to bees (Apoidea). Biological Review, 62: 515-543. doi: 10.1111/j.1469-185X.1987.tb01640.x.

Winfree, R., Williams, N.M., Gaines, H., Ascher, J.S. & Kremen, C. (2008). Wild bee pollinators provide the majority of crop visitation across land-use gradients in New Jersey and Pennsylvania, USA. Journal of Applied Ecology, 45: 793-802. doi: 10.1111/j.1365-2664.2007.01418.x