Effects of forest fragmentation on Ficus adhatodifolia Schott ex Spreng phenology and on its interactions with wasps

Article *in* Tropical Ecology · November 2019

DOI: 10.1007/s42965-019-00049-6

CITATIONS
0

READS
72

4 authors, including:

Edmilson Bianchini
Universidade Estadual de Londrina
94 PUBLICATIONS **932** CITATIONS

SEE PROFILE

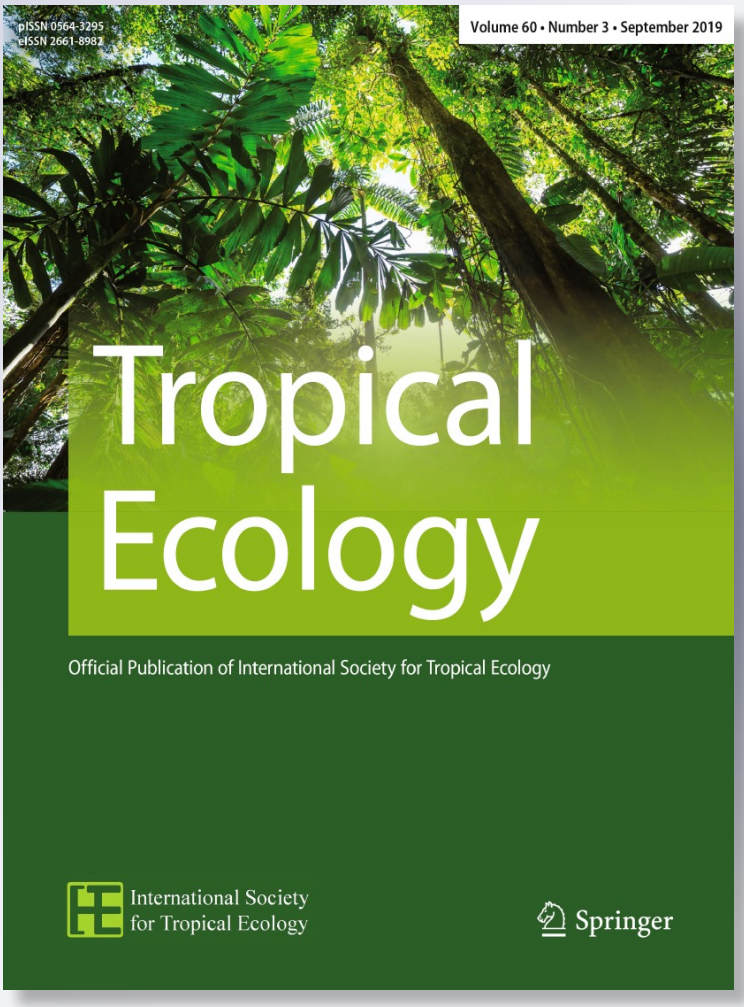
Effects of forest fragmentation on Ficus adhatodifolia *Schott ex Spreng phenology and on its interactions with wasps*

Luiz Fernando Ferreira Pol, Hugo Henrique Pires, José Eduardo Lahoz da Silva Ribeiro & Edmilson Bianchini

Tropical Ecology

ISSN 0564-3295
Volume 60
Number 3

Trop Ecol (2019) 60:462-471
DOI 10.1007/s42965-019-00049-6



Springer

Tropical Ecology (2019) 60:462–471
https://doi.org/10.1007/s42965-019-00049-6

International Society for Tropical Ecology

RESEARCH ARTICLE

Effects of forest fragmentation on *Ficus adhatodifolia* Schott ex Spreng phenology and on its interactions with wasps

Luiz Fernando Ferreira Pol[1] · Hugo Henrique Pires[1] · José Eduardo Lahoz da Silva Ribeiro[2] · Edmilson Bianchini[2]

Received: 12 December 2018 / Revised: 10 October 2019 / Accepted: 29 October 2019 / Published online: 26 November 2019

Abstract

Forest fragmentation can reduce population size, affecting the plant species and their biological interactions. We evaluated the effects of fragmentation on *Ficus adhatodifolia* Schott ex Spreng population, aiming to answer two questions: (1) is population size sufficient to maintain reproductive fig trees throughout the year? (2) Is the mean seed production per syconium higher in the larger forest fragments? Fifty-six trees of *F. adhatodifolia* were visited in a year, in five fragments and one urban area in southern Brazil. Mature syconia were collected and the seed and wasp production between areas were compared. Considering all forest remnants, we recorded syconia production throughout the year, but no fragment exhibited crop production in all months. The pollinator wasps *Tetrapus* sp. were found in almost all syconium, thus, indicating that *Ficus*-pollinator mutualism was not lost. Nevertheless, smaller fragments were not capable of maintaining reproductive individuals throughout the year, therefore, requiring pollinator wasps from other fragments. The size of fragments did not influence seed production [LRT P value = 0.1; LR stat (χ^2) = 4.64; n = 370]. Factors such as *Ficus* resilience and pollinating wasp migration may have contributed to the support of mutualism so far. Therefore, in highly fragmented landscapes such as the study region, the conservation of all forest remnants is essential to mutualistic interaction preservation. This procedure combined with the restoration of degraded areas determined by law are urgent and necessary for the conservation of regional biodiversity.

Keywords Fig · Mutualism · Neotropical forest · Pollinators · Population · Syconia

Introduction

Habitat fragmentation is one of the major threats to biodiversity at a global scale (Chapin et al. 2000) and Neotropical forests are fragmented especially due to timber harvesting and both urban and farming expansion (Tabarelli et al. 2004). When the total number of individuals is reduced in the forest, the abundance of reproductive plants is similarly reduced (Bruna et al. 2002) affecting not only those plants species but also their biological interactions (Kearns et al. 1998; Pardini et al. 2010; Figueroa et al. 2018).

Neotropical plant species maintain ecological interactions with several animal species, particularly with insects, through pollination (Bawa 1990; Ricklefs 2010). The interaction between figs (*Ficus*, Moraceae) and fig wasps (Agaoninae, Chalcidoidea) is perhaps the most specialized pollination mutualism known. Therefore, the presence of fig trees flowering throughout the year is essential for pollinator survival (Janzen 1979; Bronstein 1989; Weiblen 2002) and as a food source to fauna (David et al. 2012; Laurindo et al. 2019; Mackay and Gross 2019).

A natural environment must have enough *Ficus* trees (Wang et al. 2005) to maintain wasp populations (Pryanga 2004), otherwise, these population may decrease (Harris and Johnson 2004) or even become extinguished locally (Wang et al. 2005). Pollinator limitation of figs as a result of fragmentation may lead to lower seed production, recruitment and abundance of figs (Mawdsley et al. 1998). Harrison

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s42965-019-00049-6) contains supplementary material, which is available to authorized users.

✉ Edmilson Bianchini
bianchi@uel.br

[1] Programa de Pós Graduação em Ciências Biológicas, Universidade Estadual de Londrina, Caixa Postal 10011, Londrina, Paraná 86057-970, Brazil

[2] Departamento de Biologia Animal e Vegetal, Universidade Estadual de Londrina, Caixa Postal 10011, Londrina, Paraná 86057-970, Brazil

(2007) studied 313 syconium harvested from one fig individual has become very isolated from conspecifics through clearance of its specialized habitat. The author recorded that just 1% had been entered by a pollinator and no viable seeds were produced. The disruption of this mutualism may harm the local ecology (Bawa 1990), because other wasps, such as non-pollinating wasps (gallers, parasitoids, and hyperparasitoids), also need the syconium. Other organisms like mites (Walter 2000), fungi, nematodes (Baijnath and Ramcharun 1983; Pereira et al. 2000), protozoans, beetles, moths (Baijnath and Ramcharun 1983), ants (Compton and Robertson 1988; Dejean et al. 1997; Schatz and Hossaert-Mckey 2003; Schatz et al. 2008) and many vertebrates (Berg and Wiebes 1992; Laurindo et al. 2019; Mackay and Gross 2019) use this resource as well.

In Brazil, the Atlantic Forest biome suffered intense forest devastation and, currently, the landscape is usually composed of small forest fragments isolated from each other (Ribeiro et al. 2009). In these forest fragments, the population size of *Ficus* sp. may not be sufficient to maintain the population of its pollinator and the reproductive success of fig trees may depend on the immigration of wasps from other forest fragments. However, the distance between forest fragments, the quality of the matrix and the intensive use of pesticides in the region may be limiting factors for wasp immigration. Understanding the forest fragmentation effects on pollination and seed production in *Ficus* may contribute to the conservation of mutualistic species and all other organisms that benefit from this mutualism. This knowledge is essential to the forest management and reforestation projects success.

In this study, we evaluated the syconium production in *Ficus adhatodifolia* Schott ex Spreng and the production of seeds and wasps in the syconia, in fragments of different sizes. Two questions were addressed: (1) is population size sufficient to maintain reproductive fig trees throughout the year? (2) Is the mean seed production per syconium higher in the larger forest fragments?

Materials and methods

Studied species

Ficus adhatodifolia (subgenus Pharmacosycea) is found in South America's extra-Amazon region (Berg and Villavicencio 2004) and is morphologically similar to *F. insipida* Will. from the Amazon rainforest (Milton 1991). It is a monoecious, free-standing (Carauta and Diaz 2002) and evergreen species from northern Paraná State, Brazil (Bianchini et al. 2015), which is able to produce a large amount of syconia (hundreds were seen in the field) once or twice a year, in periods called crops. Besides that, it is relatively common for each individual to generate a few syconia outside the crop period. The reproductive phenology has been registered as asynchronous at the population level, with high-peak production between January and March (Bianchini et al. 2015). The syconium may be from 1 to 5 cm in diameter, with an interior that is usually reddish (personal observation) and a slightly prominent and superficial ostiole (Berg and Villavicencio 2004; Berg 2006).

There are five stages in syconium development (Galil and Eisikowitch 1968): A stage—from initial formation of the syconium until the maturity of pistillate flowers; B stage (female stage)—pistillate flowers anthesis, liberation of volatile substances and pollinator wasp arrival (founders); C stage—the galls containing the wasps and the achenes development, and the syconium cavity may be blocked; D stage (male stage)—staminate flowers anthesis, the male wasps complete their development, copulate with females and, subsequently, open a hole in the syconium wall; the female wasps hatch, become impregnated with pollen grains while walking through the flowers, and fly away surviving for at least 2 days (Jevanandam et al. 2013), looking for new receptive syconium; and E stage—the seeds complete their development and the syconium is now attractive to animal dispersers.

In accord to Bouček (1993), six wasp species are associated with *Ficus* subgenus *Pharmacosycea* which use the flower ovary to lay their eggs, preventing the formation of seeds on those flowers (Baijnath and Ramcharun 1983; Anstett et al. 1996). The pollinator wasp is a species of *Tetrapus* Mayr 1885 and other five non-pollinating wasp species belong to the genus *Critogaster* Mayr 1885 (Bouček 1993; Rasplus and Soldati 2006).

Study area

The study region belongs to the Atlantic Forest biome, particularly a Seasonal Semi-deciduous Forest (IBGE 2012). The landscape is highly fragmented and the forest fragments are mostly surrounded by agriculture and livestock. Five of these fragments were studied in the Londrina region, northern Paraná State, southern Brazil, as well as an urban area in Londrina city (Fig. 1).

The Godoy Forest State Park (PEMG) (23° 27′ 06″ E and 51° 15′ 09″ W) has 650 ha of well conserved forest, and when considering the surrounding forest remnants, it reached approximately 2800 ha of continuum forest (Vicente 2006). The Doralice Forest (MD) (23° 18′ 24″ S and 50° 58′ 39″ O) has 170 ha (Aguiar et al. 2003), with many lianas and some forest gaps close to the edge, but the interior of the forest is less altered, similarly to PEMG (Dias et al. 2002). The Ferraz Ecological Station (EEF) (23° 09′ 40″ S and 51° 09′ 54″ O) has nearly 130 ha, with a high number of exotic plants on the edge and on its trails, but with the interior

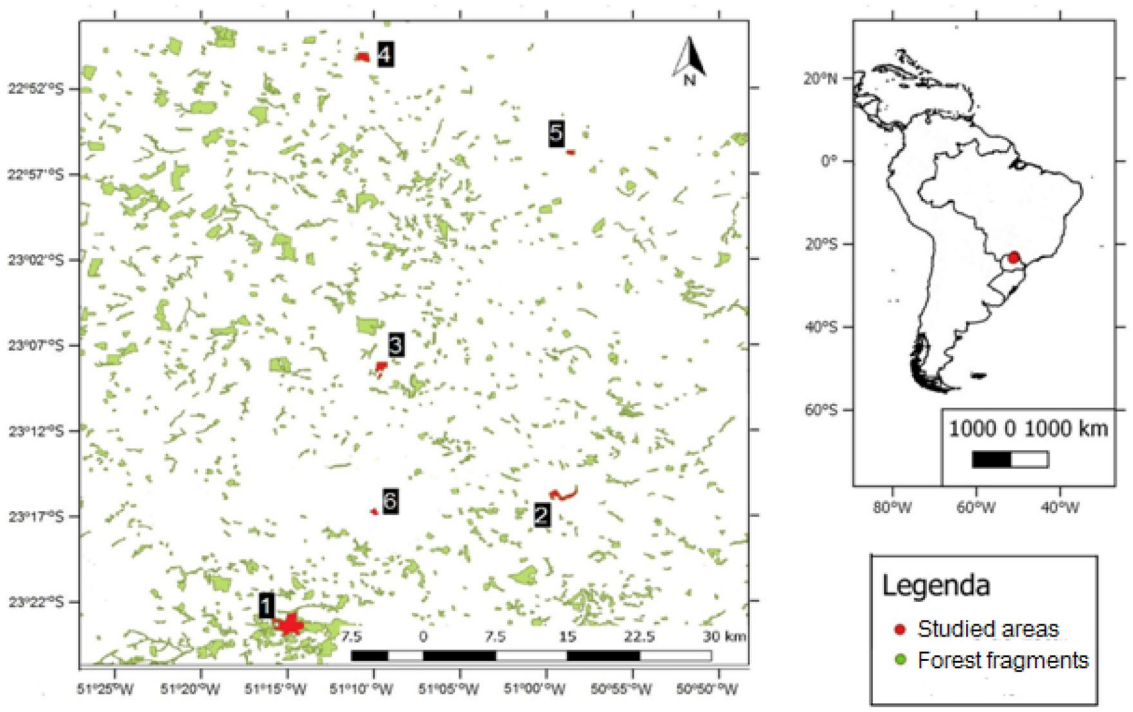


Fig. 1 Map of South America, with a zoom in the northern region of Paraná state, Brazil. The left image represent the forest fragments (green) and the studied fragments (red). 1—*Parque Estadual Mata dos Godoy*; 2—*Mata Doralice*; 3—*Estação Ecológica Ferraz*; 4—*Fazenda Alvorada*; 5—*Ibiaci*; 6—*Lago Igapó* Source: Quantum GIS 2.14 and Google Earth (colour figure online)

being less altered than the edge. The Alvorada Farm (FA) (22° 49′ 06″ S and 51° 11′ 11″ O) has an area of 128 ha and has suffered from selective logging, thus, presenting a large number of gaps and a lot of lianas, which hinders access to the interior of the fragment. The Ibiaci fragment (IB) (22° 55′ 21″ S and 50° 58′ 29″ O) has 45 ha, and presents many lianas and exotic plants. Igapó (IG) (23° 19′ 38.23″ S and 51° 09′ 50.37″ O) is not a forest fragment; it belongs instead to an urban landscape that surrounds Lake Igapó, in Londrina city. Exotic grass is predominant, as well as fruit and ornamental trees that belong to native or exotic species.

According to the Köppen classification, the climate is Cfa—humid subtropical mesothermal (Bianchini et al. 2003), with medium temperature variation, from 16 to 28 °C (Mendonça and Danni-Oliveira 2002), which creates light seasonality (Silva and Soares-Silva 2000; Bianchini et al. 2001). The higher rain and temperature rates occur in the summer, whereas the winter is colder and drier.

Data collection

Between August 2014 and July 2015, 56 individuals of *F. adhatodifolia* from area surrounding the trails were identified and monitored (PEMG—31; MD—10; EEF—7; FA—7; IB—1). The only individual found in the IB fragment did not reproduce during this period; for this reason, the fragment was not utilized in the posterior analyses. The fig trees from IG (3) were monitored from November 2014 to July 2015. The phenological data were grouped by month, even though additional visits were performed, following the assumption of Pereira et al. (2007). The syconium production and the reproductive stage were registered from each individual monitored. The B and D stages were used for measuring each *Ficus* tree production, using a rank of five points (based on Fournier 1974), which considers the percentage of the crown producing syconium: 0—no production; 1—up to 25%; 2—up to 50%; 3—up to 75%; 4—up to 100%. The monthly score by fragment was obtained by adding the scores of all fig trees sampled from that fragment.

The Google Earth Pro and Quantum Geographic Information System (QGIS) 2.14 software were used to count forest fragments in northern Paraná separating them in two groups, fragments with more than 50 ha and with less than 50 ha. It was also used to get more information about the chosen fragments for the field work. This area was delimited because IB, with less than 50 ha, had just one individual of *F. adhatodifolia* found, and as we saw in field, the shorter the fragment is, fewer individuals are found. The density of *F. adhatodifolia* in each fragment was estimated by linear density (Banack et al. 2002). The linear density was calculated by dividing the number of fig trees sampled in a trail by the total trail length (Banack et al. 2002), varying from 2.11 to

3.27 km. Individuals outside the trail were not considered for this analysis.

We collected 15–30 syconia in stage D per fig tree in five crops on the PEMG, MD and EEF. In the FA and IG, only 12–15 syconia in two crops were collected because of the low availability of syconia in these fragments. The collected syconia had to be still closed (when some resin close the ostiole) or with no galls opened. They were individualized in plastic bottle, capped with voil fabric and kept in the laboratory for 72 h for the wasps to emerge. Next, the material was placed in 70% alcohol and sealed until screening (Elias et al. 2007; Peng et al. 2010). All emerging wasps were counted, separated by sex, and only the females were identified on a specific level, using an identification key (Bouček 1993) and help from specialists from Laboratório de Ecologia Vegetal da Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto (LEV—USP). Furthermore, each syconium was cut into approximately four equal parts; in only one quarter, the unborn wasps and the achenes were counted. The total number of each was estimated by multiplying the value by four (Elias et al. 2007).

All animal species (adults or larvae) present in the syconium during stage D were collected and separated by fragments. Identification was made up to the lowest hierarchical level possible, with the aid of LEV—USP specialists.

Data analysis

To describe the species distribution inside the syconium, the frequency of occurrence was used (number of infested syconia × 100/number of syconia collected). In order to verify a possible interference of the size of fragments on the seed and wasp development, the fragments were separated into three groups of different sizes: large (Parque Estadual Mata dos Godoy, 680 ha); medium (Mata Doralice, Estação Ecológica Ferraz and Fazenda Alvorada, between 170 and 128 ha); Small (Igapó, not a forest fragment, with some sparse trees). Subsequently, the GLM (Generalized Linear Models) Negative Binomial was used because the data was over-dispersed (residual deviance much higher than degrees of freedom), and this test is the most suitable for over-dispersed data (Demétrio et al. 2014). We used the mean number of seeds and wasps per syconium, with the mean number of *Tetrapus* and *Critogaster* as co-variables. The result was compared with a null model to verify whether the size of the area influenced the results. The relation between pollinator wasps and non-pollinator wasps with seeds is observed in the co-variable results of GLM. All analyses were performed with the R Studio 3.1.3 program using the packages bbmle, bestglm, glmmML, hnp, languageR, lattice, lme4, MASS, MuMln, nlme, pscl, mgcv for hypotheses ii.

Results

Syconia production

Ficus adhatodifolia presented asynchronous reproduction in the population (Table 1, Fig. 2), but when fig trees were analyzed individually, reproduction was usually synchronous within the same crop, in northern Paraná. The syconia production occurred throughout the year, with a maximum of three stage D crops per individual in this period, whilst others had no stage D crops (Table 1, Fig. 2). The average

Table 1 Total scores for the phenology ranking on stage B (white bars) and D (black bars) in five Atlantic rainforest fragments, in the Londrina region, PR, southern Brazil, over a 1-year period (2014/2015)

	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul
PEMG	iiiiiiii	I	I	Ii		I	iiiiiiiii	iiiiiiii	Ii			i
PEMG	▮	▮	▮					▮		▮		
MD	I		Iiiiii		Ii			Iiii	Iii		I	
MD		▮		▮		▮				▮		
EEF			Iiii	I	Iiii						Ii	
EEF					▮	▮						▮
FA	I						I	Ii	I			
FA									▮	▮		
IG				Ii					I			ii
IG					▮							

The white bar for PEMG in February represents the highest ranking score obtained: 20 points

PEMG Parque Estadual Mata dos Godoy, *MD Mata Doralice*, *EEF—Estação Ecológica Ferraz*, *FA Fazenda Alvorada*, *IG Lago Igapó*

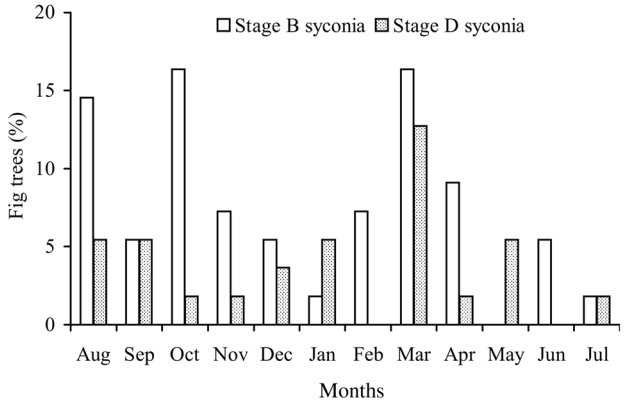


Fig. 2 Percentage of fig trees with stage B and stage D syconia, within a year, in five fragments of Atlantic rainforest in Londrina region, PR, southern Brazil

number of crops initiated per individual (stage A) was (mean ± SE) 1.25 ± 0.12 (N = 56). Abortion was common in all fragments, usually with more than 50% of each crop lost. For this reason, the average number of crops per fig tree that reached stage B was 1.00 ± 0.03 (N = 56), and stage D was 0.52 ± 0.03 (N = 56).

No fragment exhibited crop production in all months, based on the sampled fig trees (Table 1). Considering all fragments, the highest percentage of fig trees receiving pollinator wasps was recorded in October and March (16.4%) and in March (12.7%) was recorded the highest percentage of fig trees releasing wasps (Fig. 2). There were no *F. adhatodifolia* attracting pollinators in May, whereas no emerging wasps were reported on the sampled fig trees in February and June (Fig. 2).

PEMG and MD had the greatest range of months (10 months) with syconia on the B or D stages in at least one tree, with a peak syconia production in August, February and March for PEMG and October for MD. EEF had a range of 6 months, with a peak syconia production in October and December, FA (5 months) with a peak syconia production in March and IG (4 months) with a peak syconia production in November and July (Table 1). In rare cases, some trees had stages that lasted over a month, especially in stages A and C. In just one particular tree, the B stage was observed lasting over a month, and even so that crop did not developed any further. PEMG, besides having a larger forest area, has a high linear density of *F. adhatodifolia* (9.0 fig tree km^{-1}), followed by MD (3.36) and EEF (3.13).

Seeds, wasps and other invertebrates

Seventeen species of non-pollinating invertebrates were found in the syconia of *F. adhatodifolia*, including five morphotypes of larvae and 12 species of adult animals (Table 2).

Table 2 Occurrence of non-pollinating invertebrate species (adult or larvae) associated with the stage D syconia of *Ficus adhatodifolia*, in Atlantic rainforest fragments from Londrina region, southern Brazil

Invertebrate species	PEMG	MD	EEF	FA	IG
Critogaster nuda	X	X	X	X	X
C. singularis	X	X	X	X	X
C. piliventris	X	X	X	X	X
Staphylinidae	X	X	X	X	X
C. flavescens	X	X	X		X
Ceratopus sp.	X	X	X	X	
Diptera 1	X	X			
Ficicola sp.	X				
Diptera 2	X				
Diptera 3	X				
Larvae					
Coleoptera[a]	X	X	X	X	X
Morphotype 1	X	X		X	
Morphotype 2	X				
Morphotype 3	X				
Morphotype 4	X				

PEMG Parque Estadual Mata dos Godoy, *MD Mata Doralice*, *EEF Estação Ecológica Ferraz*, *FA Fazenda Alvorada*, *IG Lago Igapó*

[a]The Coleoptera larvae are probably *Ceratopus* sp. (based on Capinera 2008). Thus, the absence of mature beetles in IG samples does not represent the actual absence of this insect in the area

From the 12 species, only mite and ant species were not collected. The beetle larvae from Curculionidae family (*Ceratopus* sp.) develop by consuming all of the syconium tissue. The PEMG fragment had the highest number of non-pollinating invertebrate species interacting with *F. adhatodifolia*, while IG and FA had fewer recorded species.

Tetrapus sp. and seeds of *F. adhatodifolia* were found in more than 95% of the syconia sampled, being the most abundant organism and structure in the syconia. For the three most common non-pollinating wasp species, MD presented the lowest frequency, while PEMG presented the highest frequency for *C. singularis* and *C. piliventris*, and FA for *C. nuda* (Fig. 3). Regarding the other non-pollinator wasps, *C. flavescens* Müller was present in less than 0.5% of all syconia, although it was absent only in the FA area. The pollinator (Fig. S1) and non-pollinator wasps (Figs. S2–S5) can be observed in supplementary material.

The size of forest fragments did not influence seed production (question [ii]) (Table 3), even in the IG area, the seed production by syconium was maintained (Fig. 4). Comparing the tested model (Table 3) with the null model, it was observed that there was no significant difference [LRT P value = 0.1; LR stat (χ^2) = 4.64; N = 370], showing that the number of seeds produced did not differ with fragment size (Fig. 4).

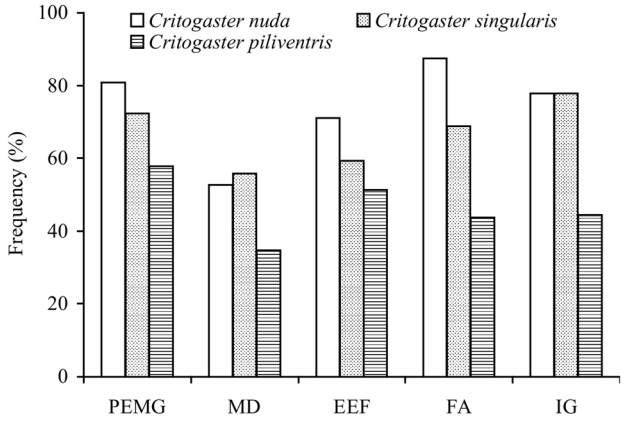


Fig. 3 Frequency of non-pollinating wasps in the syconia of *Ficus adhatodifolia* sampled in five Atlantic forest fragments from Londrina region, PR, southern Brazil. *PEMG Parque Estadual Mata dos Godoy*, *MD Mata Doralice*, *EEF Estação Ecológica Ferraz*, *FA Fazenda Alvorada*, *IG Lago Igapó*

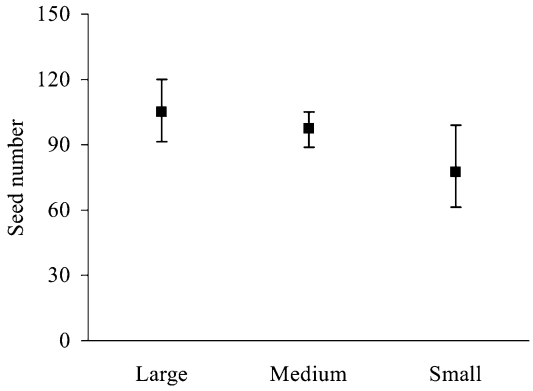


Fig. 4 Mean number of seeds in the syconia of *Ficus adhatodifolia* sampled in Atlantic rainforest fragments from Londrina region, PR, southern Brazil. Large—*Parque Estadual Mata dos Godoy*, medium—*Mata Doralice, Estação Ecológica Ferraz and Fazenda Alvorada*; small—*Lago Igapó*. Vertical bars indicate 95% confidence intervals

Table 3 Influence of the Atlantic rainforest fragment size from Londrina region, PR, southern Brazil, and the co-variables *Tetrapus* and *Critogaster*, over the mean number of seeds produced by syconium, in *Ficus adhatodifolia*, using generalized linear models

	Seeds			
	EC	SE	*Z*	*P*
Large[a]	4.55	0.09	50.30	< 0.001
Medium	− 0.13	0.08	− 1.65	0.10
Small	− 0.25	0.14	− 1.76	0.08
Tetrapus	0.002	< 0.001	3.62	< 0.001
Critogaster	− 0.001	0.14	− 0.71	0.48

Large—*Parque Estadual Mata dos Godoy*; medium—*Mata Doralice, Estação Ecológica Ferraz* and *Fazenda Alvorada*; small—*Lago Igapó*

EC estimated coefficient, *SE* standard error

[a]"Large" was used as the intercept. This fragment was used as the control parameter as it was the closest to a natural environment

Pollinator wasps influenced seed production (*P* value < 0.001), because we observed that the more pollinator wasps are born from a syconium, the more seeds are also produced (*Z*: + 3.62) (Table 3). The number of non-pollinator wasps had no significant alteration in seed production (*P* value = 0.48) (Table 3).

Discussion

Syconia production

The asynchronous reproduction in the *Ficus adhatodifolia* population (Table 1, Fig. 2) corroborates the results of Bianchini et al. (2015). This has been expected for fig trees (Janzen 1979; Herre 1996; Nazareno and Carvalho 2009) and is also required for the maintenance of plant-pollinator mutualism. However, while there has been high syconia production in some months, others have not presented any crop, suggesting that this asynchrony is not perfect. The unproductive months have also shown that this asynchrony is not sufficient, among the studied fig trees, for the maintenance of mutualism. The synchronous reproduction within the same crop is common in other monoecious fig trees (Bronstein 1992), but was not expected for this subgenus (Berg 2006).

The high abortion rate is common in Neotropical figs (Janzen 1979; Bronstein 1988; Korine et al. 2000) and we observed that many aborted B-stage syconia did not have founder wasps. This may suggest that the number of founder wasps was not sufficient to pollinate all of the syconia on the fig trees (Wang et al. 2014), but the lack of founder wasps can also be result of the syconium morphology of this particular *Ficus* species, where the ostiole remains opened for longer periods or may never close (personal observation).

The high abortion rate and low linear density of fig trees at FA and IG fragments (1.8 fig tree km^{-1} and 0.9 fig tree km^{-1}, respectively), made it difficult to sample these areas, not reaching the numbers of syconia collected in other fragments. Regions without a minimum population size of fig trees are not able to maintain wasp population, which may lead to the local extinction of these species (Chen et al. 2004). This did not occur in the fragments larger than 50 ha studied in Northern Paraná (question [i]), not even in IG, where the pollinator wasps were born from almost all syconia that reached stage D. The lack of syconia in many months in these two areas shows the need for the continuous migration of wasps between fragments. Fortunately, fig trees from Pharmacosycea subgenus usually germinate

in disturbed areas such as gaps and forest edge (Banack et al. 2002) helped by bats that are important *Ficus* seed dispersers and tolerant to forest fragmentation (Laurindo et al. 2019). Furthermore, wasps can travel up to many kilometers looking for syconia (Nason et al. 1998; Nazareno and Carvalho 2009; Heer et al. 2015). Therefore, our results suggest that if forest fragments continue to exist even scattered throughout the landscape, may be this specific mutualism will not disappear. In accord to Nazareno and Carvalho (2009) even widely isolated remnants of forests may be important for the maintenance of diversity genetic of figs and their pollinators.

Wasps and other invertebrates

The three most common non-pollinator wasps were also sampled in more than a half of the analyzed stage D syconia. The high frequency of the occurrence of these wasps (Fig. 3) showed that they are, together with *Tetrapus* sp., common wasp species in the *F. adhatodifolia* syconium (Bouček 1993; Berg and Villavicencio 2004), and they are resistant to the forest fragmentation in Northern Paraná. However, the low availability of syconia in IG and FA may have reduced the survival of some gallers with limited displacement (Table 2). Therefore, the analysis of these results suggests that there were no major infestations by non-pollinating wasps in smaller and more isolated forest fragments, unlike that observed by Wang et al. (2005). This is important information that contributes to the maintenance of mutualism in the fragmented landscape of the study region. The occurrence of *Ficicola* sp. in the PEMG represents the first record of this genus in *F. adhatodifolia*. This non-pollinating wasp genus is found in *Ficus* subgenus *Urostigma* (Farache, personal note). *Ficus citrifolia* Mill. and *F. eximia* Schott are possible hosts in Northern Paraná. The only *Ficicola* sp. individual recorded in this study cannot be considered a new species in the interaction with *F. adhatodifolia*. It is more likely to be a rare or accidental case of reproduction in an uncommon place, despite the compatibility and gall formation.

Seed and wasp production

Tetrapus sp. and seeds of *F. adhatodifolia* were found in almost all syconia sampled, which confirms the reproduction success of both species. Syconia lacking pollinator wasps were considered overexploited by non-pollinator wasps, plus pollinator wasp reproduction failure (e.g. a syconium sampled at EEF had 0 pollinator wasps, 112 non-pollinator wasps and, 40 seeds), while seedless syconia were considered plant reproduction failure (e.g. syconium sampled at MD had 162 pollinators wasps, 23 non-pollinators and, 0 seeds), because the founder wasp penetrated the syconium, but it might have had no pollen attached to the body or pollen which was not compatible.

As pollinator wasps can reach long distances (Ahmed et al. 2009), the size of forest fragments did not influence seed production (question [ii]) (Table 3), suggesting that the pollen combined from some founder wasps may be sufficient to pollinate most syconium flowers. Syconia of *F. adhatodifolia* must have more flowers than one or two founder wasps can exploit ovipositing, because we observed that the more pollinator wasps are born from a syconium, the more seeds are produced (Table 3).

The non-pollinator wasps presenting no influence in seed production was similar to results observed in *F. glabrata* and *F. insipida* (West et al. 1996), but differed from those observed in *F. andicola* Standl., where non-pollinator wasps negatively influenced the number of seeds (Cardona et al. 2013).

In the fragmented landscape of Northern Paraná, it is possible that *F. adhatodifolia* occurs in small and altered fragments, such as IB fragment (shorter than 50 ha), which represents the majority of fragments in the region (approximately 79.5%, according to measurements with help from Google Earth Pro and QGIS 2.14). These fragments should be the ones that are most dependent on pollinator wasp migration over longer distances.

The shortest distance between IG and PEMG, if compared to the other studied fragments, may be fundamental for that mutualism maintenance in IG. The greater the distance from a fig tree in stage B from a fig tree in stage D, the more difficult it is for the wasps to achieve their goal. Pollinator wasps are capable of flying long distances, sometimes over 160 km (Ahmed et al. 2009), although they usually fly between 5.8 and 14.2 km (Nason et al. 1998), taking advantage of air currents. The PEMG distances to the other studied fragments, except IG, are greater than 26 km (Google Earth Pro), suggesting that the pollinator wasps from PEMG may find some difficulties in migrating to these fragments.

As PEMG has a larger forest area and has a high linear density of *F. adhatodifolia*, certainly, there are many more trees of *F. adhatodifolia* in this fragment, in addition to those sampled. Therefore, it is likely that continuous syconia production occurs there, and it might be the main exporter of pollinator and non-pollinator wasps for the neighboring fragments, assuming a major role in the maintenance of mutualism in northern Parana state.

During the sampling, we also observed that many individuals were producing less than 10 syconia (usually one or two identified in a whole tree) out of the crop period. This strategy may be another factor that contributes to the maintenance of wasp populations and, consequently, tree populations as well. On the other hand, a mean production of only 0.52 crops per year on stage D demands large populations of this species, in order to avoid dependence on the

few syconia produced outside the crop period. For complete reproductive success, the pollinator population must remain stable, the seed dispersal animals must perform their role, the seeds must germinate and the seedlings must establish where they have been deposited.

Considering that (1) pollinating wasps were sampled in almost all the syconia sampled in the study region; (2) that there was no difference in seed production between forest fragments; (3) that there were no major infestations by non-pollinating wasps in the smaller and more isolated forest fragments, we suggest that the forest fragments were sufficient to maintain the fig-wasps-fig trees interaction. Therefore, for the conservation of these species it is essential that the forest remnants be conserved. It is also necessary to highlight the importance of further studies on fragmentation, using plant species pollinated by animals with limited displacement.

This study highlights the importance of ecological data into population studies, allowing a better understanding of species biology and in turn increasing the efficacy of forest management and reforestation projects, as well as species conservation.

Conclusions

It is possible that *F. adhatodifolia* individuals isolated in the rural landscape may play the role of wasp bridges between fragments, as observed for other *Ficus* species in the region. *Ficus adhatodifolia* usually germinate in disturbed areas such as gaps and forest edge. Therefore, the forest fragmentation does not necessarily exclude *F. adhatodifolia* from the disturbed environment, but it does not mean that mutualism is safe, because fig tree populations can decrease. In this study we observed that the smaller the fragments, the fewer *F. adhatodifolia* individuals are observed. However, even small and isolated forest remnants, such as IB and IG, can contribute to the conservation of mutualistic species and other associated species. Therefore, in highly fragmented landscapes such as the study region, the conservation of all forest remnants is essential. Restoration of degraded areas and the establishment of ecological corridors between forest remnants will allow the conservation of biodiversity. These actions are urgent and necessary.

Acknowledgements The authors thank the Conselho Nacional de Desenvolvimento Científico e Tecnológico, Coordenação de Aperfeiçoamento de Pessoal de Nível Superior and Fundação Araucária for the PELD-MANP Grant (process number 441540/2016-3). This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES)—Brazil—Finance Code 001 (research grants and fellowship to LFFP). The authors also thank all the help received from Laboratório de Biologia Reprodutiva de *Ficus*, from Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto—USP, especially to Rodrigo A. S. Pereira and Fernando Farache.

References

Aguiar LM, Reis NR, Ludwig G, Rocha VJ (2003) Dieta, área de vida, vocalizações e estimativas populacionais de *Alouatta guariba* em um remanescente florestal no norte do estado do Paraná [Diet, living area, vocalization and populational estimate of *Alouatta guariba* in a forest remaining from north of Parana state]. Neotrop Primates 11:78–86

Ahmed S, Compton SG, Butlin RK, Gilmartin PM (2009) Wind-borne insects mediate directional pollen transfer between desert fig trees 160 km apart. PNAS 106:20342–20347

Anstett MC, Kjellberg F, Bronstein JL (1996) Waiting for wasps: consequences for the pollination dynamics of *Ficus pertusa* L. J Biogeogr 23:459–466

Baijnath H, Ramcharun S (1983) Aspects of pollination and floral development in *Ficus capensis* Thunb. (Moraceae). Bothalia 14:883–888

Banack SA, Horn MH, Gawlicka A (2002) Disperser vs. establishment-limited distribution of a riparian fig tree (*Ficus insipida*) in a Costa Rican tropical rain forest. Biotropica 34:232–243

Bawa KS (1990) Plant-pollinator interactions in tropical rain forests. Annu Rev Ecol Syst 21:399–422

Berg CC (2006) The subdivision of *Ficus* subgenus *Pharmacosycea* section Pharmacosycea (Moraceae). Blumea 51:147–151

Berg CC, Villavicencio X (2004) Taxonomic studies on *Ficus* (Moraceae) in the West Indies, extra-Amazonian, Brazil, and Bolivia. Ilicifolia 4, University of Bergen, Bergen

Berg CC, Wiebes JT (1992) African fig trees and fig wasps. Koninklijke Nederlandse Akademie van Wetenschappen, Amsterdam

Bianchini E, Pimenta JA, Santos FAM (2001) Spatial and temporal variation in the canopy cover in a tropical semi-deciduous forest. Braz Arch Biol Technol 44:269–276

Bianchini E, Popolo RS, Dias MC, Pimenta JA (2003) Diversidade e estrutura de espécies arbóreas em área alagável do município de Londrina, sul do Brasil [Diversity and structure of arboreal species in wetlands from Londrina county, south of Brazil]. Acta Bot Bras 17:405–419

Bianchini E, Emmerick JM, Messetti AVL, Pimenta JA (2015) Phenology of two *Ficus* species in seasonal semi-deciduous forest in Southern Brazil. Braz J Biol 75:206–214

Bouček Z (1993) The genera of chalcidoid wasps from *Ficus* fruit in the new world. J Nat Hist 27:173–217

Bronstein JL (1988) Predators of fig wasps. Biotropica 20:215–219

Bronstein JL (1989) A mutualism at the edge of its range. Experientia 45:622–637

Bronstein JL (1992) Seed predators as mutualists: ecology and evolution of the fig/pollinator interaction. In: Bernays E (ed) Insect–plant interactions IV. CRC Press Inc., Boca Raton, pp 1–44

Bruna EM, Nardy O, Strauss SY, Harrison S (2002) Experimental assessment of *Heliconia* acuminate growth in a fragmented Amazonian landscape. J Ecol 90:639–649

Capinera JL (2008) Encyclopedia of enthomology. Springer Science, Dordrecht

Carauta JPP, Diaz BE (2002) Figueiras no Brasil [Fig trees in Brazil]. Editora UFRJ, Rio de Janeiro

Cardona W, Kattan G, Ulloa PC (2013) Non-pollinating fig wasps decrease pollinator and seed production in *Ficus andicola* (Moraceae). Biotropica 45:203–212

Chapin FS, Zavaleta ES, Eviner VT, Naylor RL, Vitousek PM, Reynolds HL, Hooper DU, Lavorel S, Sala OE, Hobbie SE, Mack MC (2000) Consequences of changing biodiversity. Nature 405:234–242

Chen Y-R, Wu W-J, Chou L-S (2004) Synchronization of fig (*Ficus microcarpa* L.) abundance and pollinator (*Eupristina verticillata*:

Agaoninae) population dynamics in northern Taiwan. J Nat Taiwan Mus 57:23–35
Compton SG, Robertson HG (1988) Complex interactions between mutualisms: ants tending homopterans protect fig seeds and pollinators. Ecology 69:1302–1305
David JP, Murugan BS, Manakadan R (2012) Seasonality in fruiting of fig and non-fig species in a tropical dry evergreen forest in Sriharikota Island, southern India. Trop Ecol 53:1–13
Dejean A, Bourgoin T, Gibernau M (1997) Ant species that protect figs against other ants: result of territoriality induced by a mutualistic homopteran. Ecoscience 4:446–453
Demétrio CGB, Hinde J, Moral RA (2014) Models for overdispersed data in entomology. In: Ferreira CP, Godoy WAC (eds) Ecological modelling applied to entomology. Springer, Berlin, pp 219–259
Dias MC, Vieira AOS, Paiva MRC (2002) Florística e fitossociologia das espécies arbóreas das florestas da bacia do rio Tibagi [Floristic and phytosociology of the forests arboreal species from Tibagi river basin]. In: Medri ME, Bianchini E, Shibatta AO, Pimenta JA (eds) A bacia do rio Tibagi [The Tibagi river basin]. Edição dos Editores, Londrina, pp 109–124
Elias LG, Farache FHA, Pereira RAS (2007) Efeito de vespas não-polinizadoras sobre o mutualismo *Ficus*-vespas de figos [Efect of non-pollinating wasps about *Ficus*-fig wasps mutualism]. Iheringia Ser Zool 97:253–256
Figueroa CC, Niemeyer HM, Cabrera-Brandt M, Briones LM, Lavandero B, Zuniga-Reinoso A, Ramirez CC (2018) Forest fragmentation may endanger a plant-insect interaction: the case of the highly specialist native aphid *Neuquenaphis staryi* in Chile. Insect Conserv Diver 11:352–362
Fournier LA (1974) Un método cuantitativo para la medición de características fenológicas en árboles [A quantitative method for measurement of phenological characteristics in trees]. Turrialba 24:422–423
Galil J, Eisikowitch D (1968) On the pollination ecology of *Ficus sycomorus* in east Africa. Ecology 49:259–269
Harris F, Johnson SD (2004) The consequences of habitat fragmentation for plant–pollinator mutualisms. Int J Trop Insect Sci 24:29–43
Harrison RD (2007) Maintenance of specificity in an isolated fig. Biotropica 39:275–277
Heer K, Kalko EKV, Albrecht L, Garcia-Villacorta R, Staeps FC, Herre EA, Dick WC (2015) Spatial scales of genetic structure in free-standing and strangler figs (*Ficus*, Moraceae) inhabiting neotropical forests. PLoS One 10:e0133581
Herre EA (1996) An overview of studies on a community of Panamanian figs. J Biogeogr 23:593–607
IBGE-Instituto Brasileiro de Geografia e Estatística (2012) Manual técnico da vegetação brasileira [Technical manual of Brazilian vegetation]. IBGE, Rio de Janeiro
Janzen D (1979) How many parents do the wasps from a fig have? Biotropica 11:127–129
Jevanandam N, Goh GR, Corlett RT (2013) Climate warming and the potential extinction of fig wasps, the obligate pollinators of figs. Biol Lett 9:1–4
Kearns CA, Inouye DW, Waser N (1998) Endangered mutualisms: the conservation of plant–pollinator interactions. Annu Rev Ecol Syst 29:83–112
Korine C, Kalko EKV, Herre EA (2000) Fruit characteristics and factors affecting fruit removal in a Panamanian community of strangler figs. Oecologia 123:560–568
Laurindo RS, Novaes RLM, Vizentin-Bugoni J, Gregorin R (2019) The effects of habitat loss on bat-fruit networks. Biodivers Conserv 28:589–601
Mackay KD, Gross CL (2019) Climate change threatens a fig-frugivore mutualism at its drier, western range margin. Proc Linn Soc N S W 141:1–17
Mawdsley NA, Compton SG, Whittaker RJ (1998) Population persistence, pollination mutualisms, and figs in fragmented tropical landscapes. Conserv Biol 12:1416–1420
Mendonça FA, Danni-Oliveira IM (2002) Dinâmica atmosférica e tipos climáticos predominantes da bacia do rio Tibagi [Atmospheric dynamics and prevailing climatic types of Tibagi river basin]. In: Medri ME, Bianchini E, Shibatta AO, Pimenta JA (eds) A bacia do Rio Tibagi [The Tibagi river basin]. Edição dos Editores, Londrina, pp 63–66
Milton K (1991) Leaf change and fruit production in six neotropical Moraceae species. J Ecol 79:1–26
Nason JD, Herre EA, Hamrick JL (1998) The breeding structure of a tropical keystone plant resource. Nature 391:685–687
Nazareno AG, Carvalho D (2009) What the reasons for no inbreeding and high genetic diversity of the neotropical fig tree *Ficus arpazusa*? Conserv Genet 10:1789–1793
Pardini R, Bueno AA, Gardner TA, Prado PI, Metzger JP (2010) Beyond the fragmentation threshold hypothesis: regime shifts in biodiversity across fragmented landscapes. PLoS One 5:0013666
Peng Y-Q, Compton SG, Yang D-R (2010) The reproductive success of *Ficus altissima* and its pollinator in a strongly seasonal environment: Xishuangbanna, Southwestern China. Plant Ecol 209:227–236
Pereira RAS, Semir J, Menezes AO Jr (2000) Pollination and other biotic interactions in figs of *Ficus eximia* Schott (Moraceae). Rev Bras Bot 23:217–224
Pereira RAS, Rodrigues E, Menezes AO Jr (2007) Phenological patterns of *Ficus citrifolia* (Moraceae) in a seasonal humid-subtropical region in Southern Brazil. Plant Ecol 188:265–275
Pryanga A (2004) Spatial dynamics of mutualistic interactions. J Anim Ecol 73:128–142
Rasplus JY, Soldati L (2006) Familia Agaonidae [Agaonidae family]. In: Fernández F, Sharkey MJ (eds) Introducción a los hymenoptera de la región neotropical [Introduction to hymenoptera from neotropical region]. Sociedad Colombiana de Entomologia & Universidad Nacional de Colômbia, Bogotá, pp 683–698
Ribeiro MC, Metzger JP, Martensen AC, Ponzoni FJ, Hirota MM (2009) The Brazilian Atlantic Forest: how much is left, and how is the remaining forest distributed? Implications for conservation. Biol Conserv 142:1141–1153
Ricklefs RE (2010) A economia da natureza [The nature's economy], 6a edn. Guanabara Koogan, Rio de Janeiro
Schatz B, Hossaert-McKey M (2003) Interactions of the ant *Crematogaster scutellaris* with the fig/fig wasp mutualism. Ecol Entomol 28:359–368
Schatz B, Kjellberg F, Nyawa S, Hossaert-McKey M (2008) Fig wasps: a staple food for ants on *Ficus*. Biotropica 40:190–195
Silva FC, Soares-Silva LH (2000) Arboreal flora of the Godoy Forest State Park, Londrina, PR, Brazil. Edinb J Bot 57:107–120
Tabarelli M, Silva JMC, Gascon C (2004) Forest fragmentation, synergisms and the impoverishment of Neotropical forests. Biodivers Conserv 13:1419–1425
Vicente RF (2006) O Parque Estadual Mata dos Godoy [The Godoy forest state park]. In: Torezan JMD (ed) Ecologia do Parque Estadual Mata dos Godoy [Ecology of Godoy forest state park]. Itedes, Londrina, pp 13–18
Walter DE (2000) First record of a fig mite from the Australian region: *Paratarsonemella giblindavisi*sp.n. (Acari: Tarsonemidae). Aust J Entomol 39:229–232
Wang R-W, Yang C-Y, Zhao G-F, Yang J-X (2005) Fragmentation effects on diversity of wasp community and its impact on fig/

fig wasp interaction in *Ficus racemosa* L. J Integr Plant Biol 47:20–26

Wang R-W, Dunn DW, Sun BF (2014) Discriminative host sanctions in a fig-wasp mutualism. Ecology 95:1384–1393

Weiblen GD (2002) How to be a fig wasp. Annu Rev Entomol 47:299–330

West SA, Herre EA, Windsor DM, Green PRS (1996) The ecology and evolution of the New World non-pollinating fig wasp communities. J Biogeogr 23:447–458