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Hymenopteran Parasitoids Associated With Frugivorous Larvae in a Brazilian Caatinga-Cerrado Ecotone

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ABSTRACT The purpose of this study was to investigate native species of parasitoids of frugivorous larvae and their associations with host plants in commercial guava orchards and in typical native dry forests of a caatinga-cerrado ecotone in the State of Minas Gerais, Brazil. Nine species of parasitoids were associated with larvae of *Anastrepha* (Tephritidae) and *Neosilba* (Lonchaeidae) in fruit of *Psidium guajava* L. (Myrtaceae), *Ziziphus joazeiro* Mart. (Rhamnaceae), *Spondias tuberosa* Arruda (Anacardiaceae), *Spondias dulcis* Forst. (Anacardiaceae), *Syzygium cumini* (L.) Skeels (Myrtaceae), and *Randia armata* (Sw.) DC. (Rubiaceae). *Doryctobracon areolatus* was the most abundant species, obtained from puparia of *Anastrepha zenildae*, *An. sororcula*, *An. fraterculus*, *An. obliqua*, and *An. turpiniae*. This is the first report of *Asobara obliqua* in Brazil and of *As. anastrephae* and *Tropideucoila weldi* in dry forests of Minas Gerais State. The number of species of parasitoids was higher in areas with greater diversity of cultivated species and lower pesticide use. The forest fragments adjacent to the orchards served as shelter for parasitoids of frugivorous larvae.

KEY WORDS *Doryctobracon*, *Asobara*, *Aganaspis*, *Anastrepha*, *Neosilba*

Studies on tritrophic interactions involving frugivorous larvae, their parasitoids, and host fruits have been conducted basically on commercial crops, aiming to maximize parasitoid use to control fruit flies of economic concern (Aluja et al. 2003). Little is known about these flies and their associations in native areas or in areas with anthropic impact. Also, little attention has been attributed to the natural history and behavior of fruit flies in nature, therefore underestimating the complexity of fruit fly biology and ecology (Aluja 1999). Inventories of unaltered native environments are essential means of retrieving information to further current understanding on the relationships among parasitoids, frugivorous larvae and their host fruits.

The vegetation adjacent to agricultural areas has been studied to reveal the importance of these areas to supply food and alternative habitats for natural enemies. It is estimated that natural enemies increase in number, thus enhancing biological control, when native vegetation is present at the edges of a crop area or is associated with a plantation (Altieri et al. 2003). Studies have shown that the fragmentation of native habitats affects natural enemies more than it does phytophagous insects (Kruess and Tscharntke 2000).

In the northern State of Minas Gerais, Brazil, there is an ecosystem known as dry forest, whose native vegetation is part of the caatinga domain, with enclaves of cerrado. This ecotone shows the typical vegetation of a dry forest (Drummond et al. 2005), also known as Deciduous Seasonal Forest. Dry forests typically are tropical, occurring in patches within a larger expanse of cerrado or caatinga. Cerrado is one of the world's biodiversity hotspots. In the last 35 yr, >50% of its ≈2 million km^2 has been transformed into pasture and agricultural lands planted in cash crops. Cerrado has the richest flora among the world's savannas and high levels of endemism. Species richness of insects is equally high in cerrado (Klink and Machado 2005). Deforestation rates have very high in the caatinga too.

The potential use of fruit fly parasitoids for biological control has gained relevance within Integrated Pest Management programs, in which a crucial stage is to know the native fauna of natural enemies. Objectives of this study are to investigate native species of parasitoids and their associations with frugivorous larvae and respective host fruits, in commercial guava orchards and in an undisturbed environmental, and to determine if the undisturbed environmental is a shelter for these parasitoids in a caatinga-cerrado ecotone.

Materials and Methods

Fruit was sampled in guava orchards and fragments of dry forest vegetation in areas near the cities of Jaíba, Janaúba, and Matias Cardoso of the State of Minas Gerais, Brazil (Table 1). The mean temperature in the

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Table 1. Location, geographical coordinates, and characteristics of the areas sampled (guava orchards adjacent to fragments of native forest)

Cities	Areas	Geographical coordinates	Management of guava orchards	Fruit diversity in nearby	Agricultural systems of guava orchards	Fruit diversity in the forest fragment
Jaíba	Area 1	15° 6′1.9″ S; 43° 58′56.5″ W	Management min., with cultural practices and no chemical control	*Citrus* spp., *Cocos nucifera*, *Cucurbita* spp., *Manihot esculenta*, *Musa* spp., *Spondias mombi*	Smallholder farming	*Ziziphus joazeiro*, *Syzygium cumini*, *Spondias dulcis*
Jaíba	Area 2	15° 08′26.4″ S; 44° 02′04.7″ W	Without management and without cultural practices	*Musa* spp.	Smallholder farming	*Ziziphus joazeiro*, *Randia armata*
Janaúba	Area 3	15° 49′52.8″ S; 43° 16′20.1″ W	Management min., with cultural practices and no chemical control	*Citrus* spp., *Cocos nucifera*, *Musa* spp.	Experimental orchard	*Ziziphus joazeiro*, *Spondias tuberosa*
Matias Cardoso	Area 4	15° 1′56.3″ S; 43° 50′20.8″ W	Intensive management, with cultural practices and chemical control	*Annona* sp., *Mangifera indica*	Commercial Orchard	*Spondias tuberosa*

region ranges from 21 to 25°C, with total annual rainfall of 700–1,200 mm (rainy period in October-April and dry period in May–September) and relative humidity ranging from 60 to 70% (Antunes 1994).

Collections from guava, *Psidium guajava* L. (Myrtaceae), were made in areas of smallholder farmers, private company and an experimental area. Other host fruit species, including juá, *Ziziphus joazeiro* Mart. (Rhamnaceae); Brazil plum or umbu, *Spondias tuberosa* Arruda (Anacardiaceae); tahitian apple or cajá, *Spondias dulcis* Forst. (Anacardiaceae); jambolan, *Syzygium cumini* (L.) Skeels (Myrtaceae); and indigoberry, *Randia armata* (Sw.) DC. (Rubiaceae) were collected in fragments of native forest (Table 1). Guava orchards were adjacent to fragments of native forest. The orchards were cultivated with the variety 'Paluma', with different types of management (in terms of culture treatment and use of chemical pesticides) and different levels of plant diversity in the surrounding areas (Table 1).

Fruit collection and processing procedures followed methods commonly used (e.g., Aluja et al. 2003). Ripe or ripening fruits were collected randomly from trees canopies and as recently fallen fruit on the ground (if the fruit was undamaged). Fruit was collected every 2 wk, from March 2005 to March 2006. The sample sizes were dependent of fruit availability. Fruit was kept in plastic trays containing a layer of vermiculite and covered with voil. Puparia were individually placed in glass flasks containing vermiculite, until the flies or the parasitoids emerged. The specimens were fixed in 70% ethanol. Association between a fly species and a parasitoid species was considered when a single species of fruit fly and of parasitoid emerged in the same flask.

Voucher specimens were deposited at the Departamento de Entomologia e Acarologia da Escola Superior de Agricultura Luiz de Queiroz (ESALQ/USP, in Piracicaba, São Paulo, Brazil), at Universidade Estadual de Montes Claros (UNIMONTES, in Janaúba, Minas Gerais, Brazil), and at the Departamento de Ecologia e Biologia Evolutiva, Universidade Federal de São Carlos (UFSCar, in São Carlos, São Paulo, Brazil).

Results and Discussion

Fruit was collected from six host plants: *P. guajava; Z. joazeiro; S. tuberosa; S. dulcis; S. cumini* and *R. armata.* In total, 544 parasitoids emerged (475 braconids, 68 figitids, and 1 pteromalid) from frugivorous larvae in three species of fruit (*P. guajava*, *S. tuberosa*, and *Z. joazeiro*). Four *Anastrepha* species and three *Neosilba* species (*N. pendula*, *N. zadolicha*, and *Neosilba* sp.) were reared (Table 2).

Infestation levels (pupae per gram of fruit) were variable. The highest level of infestation of *Anastrepha* and Lonchaeidae was observed in *Z. joazeiro* in area 3. The lowest level of infestation of *Anastrepha* was in *P. guajava* in area 4, and the lowest level of infestation of Lonchaeidae was in *S. tuberosa* in area 3 (Table 3).

Nine parasitoids species, all native of Neotropical region, were recovered from frugivorous larvae. These were *Doryctobracon areolatus* (Szépligeti, 1911); *D. brasiliensis* (Szépligeti, 1911); *Utetes anastrephae* (Viereck 1913); *Opius bellus* Gahan, 1930 (Braconidae, Opiinae); *Asobara anastrephae* (Muesebeck 1958); and *Asobara obliqua* (Papp 1969) (Braconidae, Alysiinae); *Aganaspis pelleranoi* (Brèthes, 1924); *Tropideucoila weldi* Lima, 1940 (Figitidae, Eucoilinae); and *Pachycrepoideus vindemmiae* (Rondani, 1875) (Pteromalidae) (Table 2).

Six parasitoid species were associated with tephritids and lonchaeids (Table 4). *Aganaspis pelleranoi* and *D. areolatus* parasitized the highest number of fruit fly species, whereas *As. obliqua*, *O. bellus*, and *P. vindemmiae* were obtained from just one unique fruit fly host.

Doryctobracon areolatus was the most abundant parasitoid and associated with *An. obliqua*, *An. sororcula*, and *An. zenildae* (Table 4). It was collected in both ecosystems (guava orchards and dry forest fragments), confirming the abundance of this parasitoid in the northern of Minas Gerais as observed by Alvarenga et al. (2009), and its potential relevance for biological control programs. This parasitoid stands out in various regions of Brazil for its abundance and frequency (Zucchi 2008).

The other braconids were restricted to a smaller number of fruit hosts. *Doryctobracon brasiliensis* was found only on guava. *Utetes anastrephae* parasitized

Table 2. Parasitoid and their association with fruit flies in orchards of guava and fragments of dry forest in Minas Gerais, Brazil, from March 2005 to March 2006

Areas families	Parasitoid species	Parasitoids (no.)	Parasitoid species (%)	Host flies	Host plants
Area 1–smallholder farming					
Braconidae	*Doryctobracon areolatus*	1	0.18	*An. zenildae*	*Psidium guajava*
		28	5.15	*An. zenildae*	*Ziziphus joazeiro*
		1	0.18	*An. Sororcula*	*Psidium guajava*
		102	18.75	*An. zenildae; An. sororcula*	*Psidium guajava*
		12	2.21	*An. zenildae; An. fraterculus*	*Psidium guajava*
		34	6.25	*An. zenildae; An. sororcula; An. fraterculus*	*Psidium guajava*
		6	1.10	*An. zenildae; An. obliqua; An. fraterculus*	*Psidium guajava*
		8	1.47	*An. zenildae; An. obliqua; An. turpiniae*	*Psidium guajava*
	Doryctobracon brasiliensis	2	0.37	*An. zenildae; An. sororcula*	*Psidium guajava*
		2	0.37	*An. zenildae; An. fraterculus*	*Psidium guajava*
		1	0.18	*An. zenildae; An. sororcula; An. fraterculus*	*Psidium guajava*
		3	0.55	*An. zenildae; An. obliqua; An. fraterculus*	*Psidium guajava*
	Utetes anastrephae	1	0.18	*An. zenildae*	*Ziziphus joazeiro*
	Asobara anastrephae	1	0.18	*An. zenildae*	*Psidium guajava*
		4	0.74	*An. zenildae*	*Ziziphus joazeiro*
		1	0.18	*An. sororcula*	*Psidium guajava*
		15	2.76	*An. zenildae; An. sororcula*	*Psidium guajava*
Figitidae	*Aganaspis pelleranoi*	13	2.39	*An. zenildae*	*Psidium guajava*
		1	0.18	*An. zenildae; An. sororcula*	*Ziziphus joazeiro*
		46	8.46	*An. zenildae; An. sororcula*	*Psidium guajava*
		1	0.18	*An. zenildae; An. fraterculus*	*Psidium guajava*
		1	0.18	*An. zenildae; An. fraterculus; An.sororcula*	*Psidium guajava*
		2	0.37	*An. zenildae; An. obliqua; An. fraterculus*	*Psidium guajava*
	Tropideucoila weldi	1	0.18	*An. fraterculus; An. zenildae; An. sororcula*	*Psidium guajava*
Area 2–smallholder farming					
Braconidae	*Doryctobracon areolatus*	177	32.54	*An. zenildae*	*Ziziphus joazeiro*
		6	1.10	*An. zenildae*	*Psidium guajava*
		1	0.18	*An. sororcula*	*Psidium guajava*
		1	0.18	*An. zenildae; An. sororcula*	*Ziziphus joazeiro*
	Utetes anastrephae	26	4.78	*An. zenildae*	*Ziziphus joazeiro*
		1	0.18	*An. zenildae; An. sororcula*	*Ziziphus joazeiro*
	Asobara anastrephae	4	0.74	*An. zenildae*	*Ziziphus joazeiro*
		1	0.18	*An. sororcula*	*Psidium guajava*
		10	1.84	*An. zenildae*	*Psidium guajava*
Figitidae	*Aganaspis pelleranoi*	1	0.18	*An. zenildae*	*Psidium guajava*
		1	0.18	*N. pendula; Neosilba* sp.	*Psidium guajava*
Area 3–experimental orchard					
Braconidae	*Doryctobracon areolatus*	15	2.76	*An. zenildae*	*Ziziphus joazeiro*
		5	0.92	*An. obliqua*	*Spondias tuberosa*
	Utetes anastrephae	3	0.55	*An. zenildae*	*Ziziphus joazeiro*
	Asobara obliqua	1	0.18	*N. pendula; Neosilba* sp.	*Ziziphus joazeiro*
	Opius bellus	2	0.37	*Anastrepha* sp.	*Ziziphus joazeiro*
Figitidae	*Aganaspis pelleranoi*	1	0.18	*N. zadolicha; N. pendula; Neosilba* sp.	*Ziziphus joazeiro*
Area 4–commercial orchard					
Pteromalidae	*Pachycrepoideus vindemmiae*	1	0.18	*Neosilba* sp.	*Psidium guajava*

only larvae in juá, which has adequate characteristics (small size; thin epicarp, and mesocarp) for the short ovipositor of this parasitoid (e.g., Sivinski et al. 1997, Carvalho 2005). Among the Alysiinae, we highlight the first record of *As. obliqua* in *Neosilba* sp. on juá in Brazil. *Asobara anastrephae* parasitizes *An. obliqua* on mango in the State of Tocantins (Bomfim et al. 2007), *An. striata* on guava and cajá and *An. antunesi* on cajá in the State of Amapá (Silva et al. 2007), and it was recorded in Bahia (Bomfim et al. 2010). This is the first record of *As. anastrephae* in the state of Minas Gerais.

Table 3. Degree of infestation of *Anastrepha* and Lonchaeidae on fruit in the orchards and in the dry forest in Minas Gerais, Brazil

Host fruit	Total no. of pupae of *Anastrepha*	Number of pupae of *Anastrepha* per g of fruit	Total no. of pupae of Lonchaeidae	Number of pupae of Lonchaeidae/g of fruit
Area 1–smallholder farming				
Psidium guajava	4,680	2.128	163	0.309
Ziziphus joazeiro	202	0.158	55	0.034
Area 2–smallholder farming				
Psidium guajava	122	0.039	54	0.017
Ziziphus joazeiro	1,903	0.172	428	0.055
Area 3–experimental orchard				
Ziziphus joazeiro	212	3.068	317	3.282
Spondias tuberosa	298	0.107	0	0
Area 4–commercial orchard				
Psidium guajava	0	0	3	0.002

Table 4. Tritrophic relation between host fruits, frugivorous larvae, and their parasitoid in orchards of guava and in the fragments of dry forest in Minas Gerais, Brazil

Host plants	Host flies	Parasitoid species
Psidium guajava	*Anastrepha zenildae*	*Aganaspis pelleranoi*
Psidium guajava	*Anastrepha sororcula*	*Asobara anastrephae*
Psidium guajava	*Anastrepha zenildae*	*Asobara anastrephae*
Psidium guajava	*Anastrepha sororcula*	*Doryctobracon areolatus*
Psidium guajava	*Anastrepha zenildae*	*Doryctobracon areolatus*
Psidium guajava	*Neosilba* sp.	*Pachycrepoideus vindemmiae*
Spondias tuberosa	*Anastrepha obliqua*	*Doryctobracon areolatus*
Ziziphus joazeiro	*Anastrepha zenildae*	*Asobara anastrephae*
Ziziphus joazeiro	*Anastrepha zenildae*	*Doryctobracon areolatus*
Ziziphus joazeiro	*Anastrepha* sp.[a]	*Opius bellus*
Ziziphus joazeiro	*Anastrepha zenildae*	*Utetes anastrephae*

[a] Male.

Aganaspis pelleranoi was the unique figitid associated exclusively with fruit fly and lance fly larvae. This parasitoid is common and widely distributed in Brazil (Guimarães et al. 2004). The figitid *T. weldi* parasitized *An. fraterculus*, *An. zenildae*, and *An. sororcula*, as well as drosophilids in guava. This species previously had been associated only with *N. pendula* (Lonchaeidae) (Guimarães et al. 2003). *Pachycrepoideus vindemmiae* was associated with *Neosilba* sp. (first report in a lonchaeid species) in guava (Table 4).

Fruit fly research and monitoring programs have been implemented in the irrigated perimeters of Jaíba and Gorutuba, in the northern of Minas Gerais state, since 1994 (Canal-Daza 1997, Alvarenga et al. 2009). Besides parasitoid species discussed herein, *D. fluminensis* (Costa Lima 1938) and *U. anastrephae; O. bellus* and *Diachasmimorpha longicaudata* (Ashmead) (introduced species) also were recorded in this region.

We observed differences in species composition of parasitoids between guava orchard and dry forest. *Asobara obliqua*, *O. bellus*, and *U. anastrephae* were collected exclusively in forest areas, whereas *D. brasiliensis*, *P. vindemmiae*, and *T. weldi* were found only in orchards (Table 5). However, *Ag. pelleranoi*, *As. anastrephae* and *D. areolatus* were collected in both environments. The latter species was the most abundant in both, but with higher numbers in forest sites.

Higher number of species of parasitoids was found in areas with greater diversity of crops and lower pesticide use, namely area 1 (six species), area 3 (five species) and, area 2 (four species). In areas 1 and 2, the orchards were subject to minimal management, consisting basically of routine pruning. These two areas also presented higher number of fruit trees in neighboring areas. The characteristics of these areas propitiated a constant succession of fruit flies in the orchards, which was reflected in the higher number of species of parasitoids. Guava production in these areas is extensive and many fruit are left on the ground. Area 3 was different from the other two because its orchard was not for commercial purposes and contained a smaller number of guava trees. Area 4 was subject to intensive, systematic management, with few fruit on the ground, and featuring intensive use of chemical pesticides applied every 15 d to the guava orchard and surrounding orchards. Consequently, just one species of parasitoid was collected in area 4 (Table 2).

Table 5. Composition of parasitoids in the orchards and in the dry forest and in Minas Gerais, Brazil

Orchard	Forest
Aganaspis pelleranoi	*Aganaspis pelleranoi*
Asobara anastrephae	*Asobara anastrephae*
Doryctobracon areolatus	*Asobara obliqua*
Doryctobracon brasiliensis	*Doryctobracon areolatus*
Pachycrepoideus vindemmiae	*Opius bellus*
Tropideucoila weldi	*Utetes anastrephae*

The natural parasitism of fruit flies is affected mainly by the host fruit, host flies, and location (Canal-Daza and Zucchi 2000). It would be interesting to emphasize that in northern Minas Gerais, the parasitoid species that occur in unperturbed environments (dry forests) are practically the same of those found in commercial orchards in other Brazilian regions.

The neighboring vegetation fragments with greater availability of native fruit also contributed to higher number of species. This can be exemplified by areas 1, 2, and 3. In these areas, the dry forest fragments were less affected and had more food resources available to the fruit flies. In area 3, there was higher number of native fruit fly host plants, and consequently in this area the variety of parasitoid species was high. The fragment in area 4 was deeply degraded, with low availability of native fruits. This was reflected as lower incidence of parasitoids in this area. Therefore, our data reveal that the forest fragments adjacent to the orchards served as shelter for fruit fly parasitoids in a caatinga-cerrado ecotone. Moreover, we report for first time *As. obliqua* in Brazil and *As. anastrephae* and *T. weldi* in dry forests of Minas Gerais State.

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