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Original article

The role of bee diversity in pollination and fruit set of yellow passion fruit (*Passiflora edulis* forma *flavicarpa*, Passifloraceae) crop in Central Brazil

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Abstract – Pollination is an important ecosystem service, especially to self-sterile crops as passion fruit, which depends on the large solitary bee for fruit set. We estimated the species richness of pollinators of yellow passion fruit in Central Brazil and examined whether there was any association with crop yield. We recorded 27 bee species on passion fruit flowers in commercial orchards in the region, from 2004 to 2007. Some 17 species were classified as pollinators (12 as effective and 5 as occasional pollinators). Species richness and frequency of pollinators were positively correlated with reproductive efficacy. Hand pollination substantially increased average fruit set (from 23.3% to 69.8%). Our results indicated that, although native pollinators still maintain economically viable natural fruit set in the region, pollination can be sustained and even enhanced by promoting conservation and management of bee diversity.

Cerrado / large bees / native pollinator / species richness / *Xylocopa*

1. INTRODUCTION

Crop pollination is an essential ecosystem service (Kearns et al. 1998; Kremen 2008), and 35% of global crop species depend on animal pollinators (Klein et al. 2007). In the last decades, the loss of pollinators and pollination services has led to increasing concern (Buchmann and Nabhan 1996; Kearns et al. 1998; Biesmeijer et al. 2006; Potts et al. 2010). Even if these concerns may prove to be somewhat exaggerated (Ghazoul 2005; Aizen et al. 2009), there is still little information on the status of wild pollinators around the world or on the pollination services they provide for many crop plants (Klein et al. 2007; Kremen et al. 2007).

The yellow passion fruit (*Passiflora edulis* Sims f. *flavicarpa* Deneger) is widely cultivated in Brazil and its fruits are consumed fresh or used in the juice industry (Silva 2005). Brazil is the main producer with more than 46,500 ha of planted area and a production of almost 665,000 tons/year (Agrianual Anuário da Agricultura Brasileira 2010). Furthermore, this crop is important to the development of sustainable farming systems in several areas, such as the Triângulo Mineiro region, which is the main industrial centre of passion fruit juice processing in Brazil (Silva 2005).

However, the yellow passion fruit depends on cross-pollination and requires either natural pollinators or active hand pollination for fruit production (Camillo 2003). The hand pollina-

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tion procedure is labour-intensive and increases the production costs in the Triângulo Mineiro region by approximately 15% (Silva 2005). Due to the relatively large flowers, pollinators of passion fruit are native species of large solitary bees, which depend both on the nectar, partly supplied by passion fruit orchards, and other food resources found on natural vegetation. Bees of the genera *Xylocopa*, *Centris*, *Epicharis*, *Eulaema* and *Bombus* have been recorded as pollinators of passion fruit in Brazil, and *Xylocopa* species are the most efficient natural pollinators (Corbet and Willmer 1980; Sazima and Sazima 1989; Camillo 2003) because of their large size (up to 4.5 cm long) and their behaviour during flower foraging (Camillo et al. 1986). *Xylocopa* bees are known popularly as carpenter bees due to their habit of nesting in wood (Camillo and Garófalo 1982). However, deforestation has caused a reduction of nesting places (dead wood) and natural food resources (pollen and nectar). This, combined with increased pesticides usage, has led to low densities of suitable native pollinators around orchards, which is currently one of the major problems faced by the yellow passion fruit industry (Camillo 2003). This is especially important in the Cerrado region, the Neotropical savannas in Central Brazil, nowadays a main agricultural frontier and where natural areas have been under increasing pressure of fragmentation and deforestation (Klink and Machado 2005).

Some recent reviews that associate land-use change and crop pollination services (Klein et al. 2007; Kremen et al. 2007; Ricketts et al. 2008; Garibaldi et al. 2011) call attention to land management practices aimed at conserving habitat conditions and landscape structure for the maintenance of sustainable pollination. In this context, passion fruit is pointed out as one of the nine crops in the world with clear evidence that wild pollinators contribute directly to production (Klein et al. 2007). Thus, the objectives of this study were to estimate the species richness of native pollinators of yellow passion fruit in Triângulo Mineiro region in Central Brazil, to examine whether the species richness and frequency of these pollinators correlates with fruit set yield and to discuss the sustainable use of pollination services for yellow passion fruit production.

2. MATERIALS AND METHODS

2.1. Study site

The study was conducted in commercial orchards located around Uberlândia and Araguari in Triângulo Mineiro region, State of Minas Gerais. The region is inside the Cerrado domain, the Neotropical savanna areas of Central Brazil. We studied four areas scattered in the region and <70 km from each other: Água Limpa Farm (19°05′10″ S, 48°21′15″ W, 0.5 ha of passion fruit orchard), Pissarão Farm (18°42′02″ S, 48°05′53″ W, 4 ha orchard), Três Irmãos Farm (19°02′22″ S, 48°33′05″ W, 1 ha orchard) and Campo Alegre Farm (18°59′06″ S, 48°14′26″ W, 4 ha orchard). All of the study areas were small farms (<50 ha) where the passion fruit was not the only crop or product.

Field observations were carried out during the passion fruit flowering periods from 2004 to 2007. There is a marked difference between seasons in the study region: a warmer rainy season from October to March and a cooler and mostly rainless dry season from April to September. The average monthly temperatures ranged from 18°C to 24°C and annual rainfall varied from 1,300 to 1,700 mm (Rosa et al. 1991).

2.2. Passion fruit floral biology

Yellow passion fruit has large perfect flowers with typical melittophilous characteristics. The flowers open after midday and close around 2200 hours (Sazima and Sazima 1989), but nocturnal visitors were never observed. If fertilisation does not occur, the flowers will wither and fall. Floral characteristics, such as the superior position of stigmas over anthers, protandry and delayed stigma positioning, make self-pollination difficult (Kavati 1998). Moreover, a complex self-incompatibility system results in a strict dependence on pollinators services (Rêgo et al. 2000; Suassuna et al. 2003). Yellow passion fruit flowering depends on at least 11 h of sunlight (Bruckner and Silva 2001), which coincides with the rainy season in the study region. Therefore, flowering begins from October and ceases until May, showing 7- to 10-day peaks of high flower production follow by 45–50 days with no or a few flowers scattered along the blooming period (Silva 2005).

2.3. Flower visitors

The orchards were visited weekly or biweekly during yellow passion fruit flowering. Observations of flower visitors were carried out between 1200 and 1700 hours (from the opening of flowers until the activity of visitors decreased) by walking along the vines and recording the visitors on open flowers for a total of 183 h. Observations were done on rainless days and within peaks of high flowering production. Sampling effort per area varied according to the difference in flowering periods and crop condition. Some bee visitors were collected and killed by using ethyl acetate for identification, but most often, it was possible to identify visitors directly in the field. We used the bee classification proposed by Silveira et al. (2002). Voucher specimens were incorporated to the Entomological Collection of the Federal University of Uberlândia.

We measured the thorax height of collected bees with callipers and used further behavioural observations to classify the visitors as pollinators, occasional pollinators, and thieves. Pollinators repeatedly made contact with reproductive structures in all their visits, while occasional pollinators were less frequent and sometimes failed to touch these structures. Thieves collected pollen or nectar without touching the stigma at all. Comparison of the thorax height of pollinators and thieves was done using ANOVA (Zar 1999). We considered visitor frequency as the number of visits observed on passion fruit flowers per sampling effort per day. We also estimated the occurrence frequency (OF) of visitors as the percentage of number of occurrences of each species by the total number of samples (Buschini 2006) and considered bees with OF ≥50% as highly frequent (HF), between 25% and 50% as frequent (F) and below 25% as less frequent (LF).

To compare data for species richness of flower visitor and pollinators of passion fruit among areas, we calculated the expected accumulation curves (sample-based accumulation curves sensu Gotelli and Colwell 2001) for each area using each observation section of 1 h as the sample unit. As the rarefaction curves assume that differences among samples are due only to random sampling effects (Gotelli and Colwell 2001), to evaluate the biases in the expected accumulation curves we used the 'patchiness simulation' in the EstimateS program to calculate sample-based rarefaction curves (see the EstimateS User's Guide in Colwell 2006). The resulting simulated curves were compared with those of empirical data. Because no marked difference was observed, we used the primary empirical data to create the rarefaction curves presented in the 'Results' section.

We estimated the values of species richness for flower visitors and pollinators for each area based on the distribution of uniques and duplicates (Colwell and Coddington 1994). We used the estimators Jackknife of first and second orders and the bias-corrected version of Chao 2. We avoided the Incidence-based Coverage Estimator because of the limit for rare or infrequent species needed (see the EstimateS User's Guide in Colwell 2006). These analyses were carried out using the EstimateS 8.2 software (Colwell 2006).

2.4. Quantification and analysis of natural pollination

We used hand pollination tests to evaluate natural pollination efficacy and fruit production (fruit set). Tests were done on different days for each area, and we considered each day of pollination treatment as a sampling unit. Hand cross-pollinations were done after the first hour of anthesis, using pollen from different individuals. Both hand-pollinated flowers and flowers left for natural pollination were tagged at the pedicel with threads of different colours. Flowers used for pollination treatments were from different plants along the orchard lines. Fruit set from each treatment was verified 15 days after pollination. Some pistils of hand (n=17) and natural (n=58) pollination flowers were collected approximately 24 h later and fixed in 70% alcohol in order to observe pollen tube growth under fluorescence microscopy (Martin 1959). Pollination quality was estimated using the percentage of ovules penetrated by pollen tubes. The Mann–Whitney test was applied to verify differences in pollen tube-penetrated ovules between pollination treatments (Zar 1999).

We used the χ^2 test to verify differences in fruit set between hand cross-pollination and natural pollination treatments for each area. We compared fruit set from hand cross-pollination to verify differences in crop management among areas and natural fruit set to compare pollination activity. General differences in hand-pollinated and natural fruit set among areas were also tested using ANOVA, but due to the required samples size, only data for Água Limpa, Campo Alegre and Pissarão areas were used in this case. For the statistical analyses, we calculated the reproductive efficacy (natural pollination fruit set/cross-pollination fruit set; sensu Ruiz and Arroyo 1978) for each observation session. The reproductive efficacy can be used as an estimate of fruit/seed set under optimum pollination; therefore, it provides a direct indication of pollinator efficiency for obligate cross-pollinating plants (Ruiz and Arroyo 1978) and it is less influenced by nutritional or physiological differences among orchards.

To verify any correlation between pollinator parameters (species richness and frequency) and passion fruit crop yield (reproductive efficacy), we used each observation session (sampled day) and fruit set results from pollination treatments done the same day or subsequent day in the same area. We used a total of 19 sessions, 8 for the Agua Limpa farm, 6 for Campo Alegre farm, 3 for Três Irmãos farm and 2 for Pissarão farm. We used the Pearson correlation coefficient with Bonferroni correction. We log transformed data before the statistical analyses to match the assumptions. Since we used data from different areas, we analysed the effect of variable area on reproductive efficacy and pollinator variables using a general linear model (as in Hoehn et al. 2008) with reproductive efficacy as dependent variable and we found no area effect (result not shown).

3. RESULTS

3.1. Size and behaviour of flower visitors and pollinators

We observed 3,411 bees belonging to 27 species visiting yellow passion fruit flowers. Of these species, 12 were classified as effective pollinators, 5 as occasional pollinator and 10 as thieves (Table I).

There were significant differences in thorax height between effective pollinators, occasional pollinators and thieves (ANOVA, F=366.31, df=114, P<0.01) (Figure 1). Pollinators (species in Table I) were larger than 1.2 cm body length and their thorax height ranged from 0.8 to 1.1 cm, which led to contact with the stigma and anthers in every visit. Occasional pollinators (species in Table I) thorax height ranged from 0.6 to 0.7 cm and they failed to touch the anthers or stigma during some of their visits. All pollinator species collected nectar and they were never observed collecting pollen on passion fruit flowers. These bees landed on the flower corona, proceeded towards the base of the androgynophore and introduced the mouthparts to reach the nectar chamber. During the visits, bees touched anthers or stigmas with the upper part of their thorax and carried pollen from one flower to another. The most frequent pollinator species were *Xylocopa* (*Megaxylocopa*) *frontalis*, *X.* (*Neoxylocopa*) *suspecta* and *Centris* (*Ptilotopus*) *scopipes*, all classified as effective pollinators (Table I).

Thieves were significantly smaller than the pollinators (<1.2 cm in length and <0.5 cm in thorax height) (Figure 1). They were observed collecting pollen directly on anthers of the same flower or less often collecting nectar, but in any case, they rarely touched the stigma (less than once every 100 visits). Among the small bee visitors observed in passion fruit flowers, *Apis mellifera* was the most frequent (Table I) and a major pollen thief. In addition, *Trigona* species, including *T. hyalinata*, *T. spinipes* and *Trigona* sp. 1, were considered the main nectar robbers because they usually perforated the nectar chamber.

The number of bee species observed visiting the yellow passion fruit flowers and the estimates for each area are presented in Table II. Our results were higher than that found in other areas in Brazil (Table III). The largest species richness was recorded in Água Limpa farm (n=22) and the smallest in Pissarão farm (n=15),

Table I. Floral visitors on *P. edulis* f. *flavicarpa* orchards in Triângulo Mineiro region, State of Minas Gerais, Central Brazil.

Bee visitors	Resource	Relative frequency	OF	Observed areas
Effective pollinators		**36.1%**		
Acanthopus excellens Schrottky, 1902	Nectar	0.5%	LF	AL,TI
Bombus (*Fervidobombus*) *pauloensis* Friese, 1913	Nectar	2.7%	F	AL,CA,PF,TI
Centris (*Ptilotopus*) *denudans* Lepeletier,1841	Nectar	0.3%	LF	AL
Centris (*Ptilotopus*) *scopipes* Friese, 1899	Nectar	4.8%	HF	AL,CA,PF,TI
Centris (*Ptilotopus*) *sponsa* Smith, 1854	Nectar	0.03%	LF	CA
Centris (*Trachina*) *longimana* Fabricius, 1804	Nectar	1.2%	LF	AL,CA,PF,TI
Epicharis (*Epicharana*) *flava* (Friese, 1900)	Nectar	0.5%	LF	AL,CA,PF,TI
Eulaema (*Apeulaema*) *nigrita* Lepeletier, 1841	Nectar	0.3%	LF	AL,PF,TI
Xylocopa (*Megaxylocopa*) *frontalis* (Olivier, 1789)	Nectar	12.8%	HF	AL,CA,PF,TI
Xylocopa (*Neoxylocopa*) *grisescens* Lepeletier, 1841	Nectar	1.3%	F	AL,PF,TI
Xylocopa (*Neoxylocopa*) *hirsutissima* Maidl, 1912	Nectar	0.2%	LF	AL,CA
Xylocopa (*Neoxylocopa*) *suspecta* Moure & Camargo, 1988	Nectar	11.5%	HF	AL,CA,PF,TI
Occasional pollinators		**1.2%**		
Centris (*Centris*) *flavifrons* (Fabricius, 1775)	Nectar	0.1%	LF	AL
Centris (*Xanthemisia*) *lutea* Friese, 1899	Nectar	0.2%	LF	AL,CA
Epicharis (*Epicharis*) *bicolor* Smith, 1874	Nectar	0.4%	LF	AL,PF
Oxaea austera Gertäcker, 1867	Nectar	0.4%	LF	AL,PF
Oxaea flavescens Klug, 1807	Nectar	0.1%	LF	AL,PF,TI
Thieves/robbers		**63.2%**		
Apis mellifera Linnaeus, 1758	Pollen/nectar	36.4%	HF	AL,CA,PF,TI
Augochlora sp.	Pollen/nectar	0.1%	LF	AL,PF
Augochloropsis sp.	Pollen/nectar	0.03%	LF	TI
Frieseomelitta varia (Lepeletier, 1836)[a]	Pollen/nectar	26.7%[a]	LF	AL,CA
Paratrigona lineata (Lepeletier, 1836)[a]	Pollen/nectar		LF	CA,TI
Scaptotrigona sp1[a]	Pollen/nectar		LF	CA
Tetragonisca angustula Latreille, 1836[a]	Pollen/nectar		LF	AL,TI
Trigona hyalinata (Lepeletier, 1836)[a]	Pollen/nectar		LF	AL,CA,PF,TI
Trigona sp1[a]	Pollen/nectar		LF	CA
Trigona spinipes (Fabricius, 1793)[a]	Pollen/nectar		HF	AL,CA,PF,TI

Occurrence frequency (OF): *HF* high frequency, *F* frequent, *LF* low frequency; areas: *AL* Água Limpa farm, *CA* Campo Alegre farm, *PF* Pissarão farm, *TI* Três Irmãos farm

[a] The relative frequency of some species were shown together

but when we considered pollinator species richness, the highest value was also for Água Limpa farm (n=16) but the lowest was for Campo Alegre farm (n=9). The data were similar to the estimated pollinator species richness (Table II), but the rarefaction curves of species richness (Figure 2a) did not reach a clear asymptote for any of the areas. Nevertheless, when pooling together data for all areas, the rarefaction curves and richness estimators of pollinators coincided with the ones obtained for the Água Limpa orchard (Figure 2b).

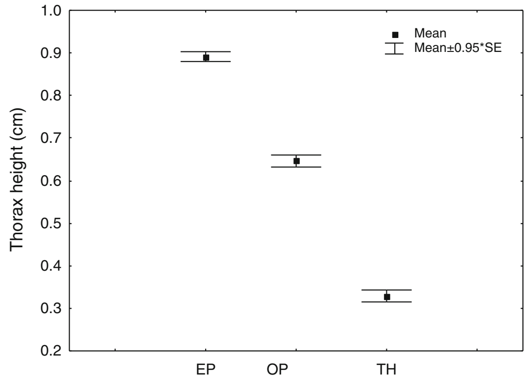

Figure 1. Average (±SE) thorax height of effective pollinators (*EF*), occasional pollinators (*OP*) and thieves (*TH*) of *P. edulis* f. *flavicarpa* observed in orchards in Triângulo Mineiro region, State of Minas Gerais, Central Brazil.

3.2. Pollination tests

There were significant differences between the pollination treatments, with much larger fruit production after hand cross-pollination in all orchards (Table IV). But when the general results for the areas were compared, there was no difference either for hand cross-pollination, natural fruit set or reproductive efficacy among Água Limpa, Campo Alegre and Pissarão farm areas (ANOVA test F=0.045, df=2, P=0.956; F=0.172, df=2, P=0.843; and F=1.726, df=2, P=0.210, respectively), and pairwise comparison between all areas using χ^2 did not show any important difference (data not shown). In any case, natural fruit set and reproductive efficacy values were much higher than those found for other passion fruit cultivation areas in Brazil (Table III).

Table II. Estimates of species richness for flower visitors and pollinators of *P. edulis* f. *flavicarpa* in orchards of the Triângulo Mineiro region, State of Minas Gerais, Central Brazil.

Study areas	Species richness estimation Jack 1 (mean±SD)	Jack 2 (mean)	Chao 2 (mean±SD)	Observed richness Visitors
Água Limpa farm	27±2.1	32	32±10.2	22
Campo Alegre farm	19±1.6	19	18±1.4	16
Pissarão farm	20±1.8	22	17±3.0	15
Três Irmãos farm	20±1.7	22	19±11.7	16
Total	31±2.0	34	30±4.1	27
	Pollinator richness estimation			Observed richness
	Jack 1 (mean±SD)	Jack 2 (mean)	Chao 2 (mean±SD)	Pollinators
Água Limpa farm	18±1.4	20	17±2.3	16
Campo Alegre farm	11±1.3	11	9±0.9	9
Pissarão farm	15±1.7	16	12±2.1	11
Três Irmãos farm	13±1.5	15	11±2.5	10
Total	19±1.4	21	18±2.3	17

Table III. Species richness of bees visiting flowers of *P. edulis* f. *flavicarpa* and fruit production in other studies areas in Brazil

Study area in Brazil	Source	V/P	NP %	HCP %	RE
Campinas, SP	Sazima and Sazima 1989	6/3	6/25[a]	53	0.12/0.49
Cordeirópolis, SP	Bruckner and Silva 2001	ni	3.6	85.7	0.04
Holambra, SP	Camillo 2003	ni	3.3/25[b]	ni	ni
Jaboticabal, SP	Ruggiero 1973	ni	20	75.4	0.27
Presidente Prudente, SP	Kavati 1998	ni	7.5	50.8	0.15
Votuporanga, SP	Bruckner and Silva 2001	ni	12	79	0.15
Londrina, Assaí, PR	Oliveira et al. 2005	ni	12	84	0.14
Morretes, Londrina, PR	Melo et al. 2006	11/5	ni	ni	ni
São Luís do Curu, CE	Freitas and Oliveira-Filho 2003	ni	13/25[b]	ni	ni
Juazeiro, BA	Bos et al. 2007	ni	16	51.6	0.31
Campos, RJ	Benevides et al. 2009	10/7	ni	ni	ni
Araguari, MG	Leone 1990	13/9	ni	ni	ni
Uberlândia, Araguari, MG	Present study	27/17	23.3	69.8	0.38

V/P number of species of bee visitors and pollinators, *NP* fruit set from natural pollination, *HCP* fruit set from hand cross-pollination, *RE* reproductive efficacy, *ni* not informed

[a]The presence and absence of *Trigona* sp., respectively

[b] Before and after introducing *Xylocopa*'s nests in the area, respectively

Pollen tube growth and ovule penetration analysis showed a much higher number of penetrated ovules (U=118.5, P<0.001) in hand cross-pollinated pistils than in open-pollinated pistils. In pistils that received the hand cross-pollination treatment, 68.8±1.68% of the ovules were penetrated by pollen tubes (n=17 flowers), while only 27.8±3.24% ovules were penetrated in pistils exposed to natural pollination (n=58 flowers). The reproductive efficacy correlated with species richness of all pollinators visiting yellow passion fruit flowers (Pearson coefficient with Bonferroni correction: r=0.55, P=0.015; Figure 3a) and with species richness of effective pollinators (r=0.53, P=0.020; Figure 3b), but no significant association was observed with occasional pollinators alone (r=0.39, P=0.100) or thieves (r=−0.08, P=0.741). Considering the frequency of visitors, we also found a positive correlation between reproductive efficacy and the frequency of pollinators as a whole (r=0.66, P=0.002; Figure 3c), as well as with frequency of effective pollinators (r=0.66, P=0.002; Figure 3d), but no significant result was found with occasional pollinators or thieves frequency (r=0.38, P=0.106 and r=0.08, P=0.730, respectively).

4. DISCUSSION

This paper supports the idea that bee species richness influenced the fruit set of yellow passion fruit and that such diversity of pollinators added to their frequency on flowers and seemed to maintain a much higher reproductive efficacy than in other cultivation areas in Brazil. Although both pollinator density and species diversity have been showed to be important to ensure pollination services and crop yields, the mechanisms linking biodiversity and pollination need further exploration (Klein et al. 2007; Kremen et al. 2007).

Despite possible methodological differences between compared surveys, our results indicated that bee diversity around these Central Brazil orchards is higher than in other parts of the country. But it is still a subsample of the

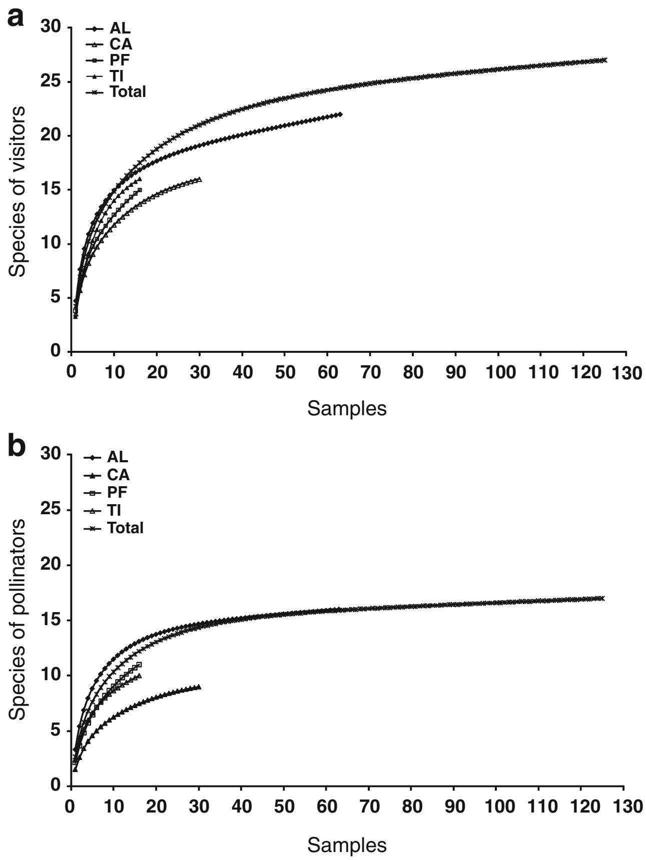

Figure 2. Sample rarefaction curves of bee species visiting flowers of *P. edulis* f. *flavicarpa* in commercial orchards in Triângulo Mineiro region, Minas Gerais, Central Brazil. **a** Flower visitors, **b** pollinators.

possible bee fauna associated with passion fruit pollination, as indicated by rarefaction curves. Compared with the other Brazilian biomes, the Cerrado has a relatively rich bee fauna with a

Table IV. Percentage of fruit set and reproductive efficacy (natural fruit set/hand cross-pollination fruit set) after pollination treatments in *P. edulis* f. *flavicarpa* in Triângulo Mineiro region, State of Minas Gerais, Central Brazil.

Study area	Fruit set by treatment % (*n*)		Reproductive efficacy
	Hand cross-pollination	Natural pollination	
Água Limpa farm	71.6 (213)^{a}	22.5 (389)^{b}	0.33
Pissarão farm	58.2 (32)^{a}	33.8 (35)^{b}	0.58
Três Irmãos farm	85.0 (54)^{a}	21.3 (51)^{b}	0.25
Campo Alegre farm	64.4 (119)^{a}	15.6 (101)^{b}	0.24

In each line, different letters indicate significant differences (χ^2, $P<0.05$)

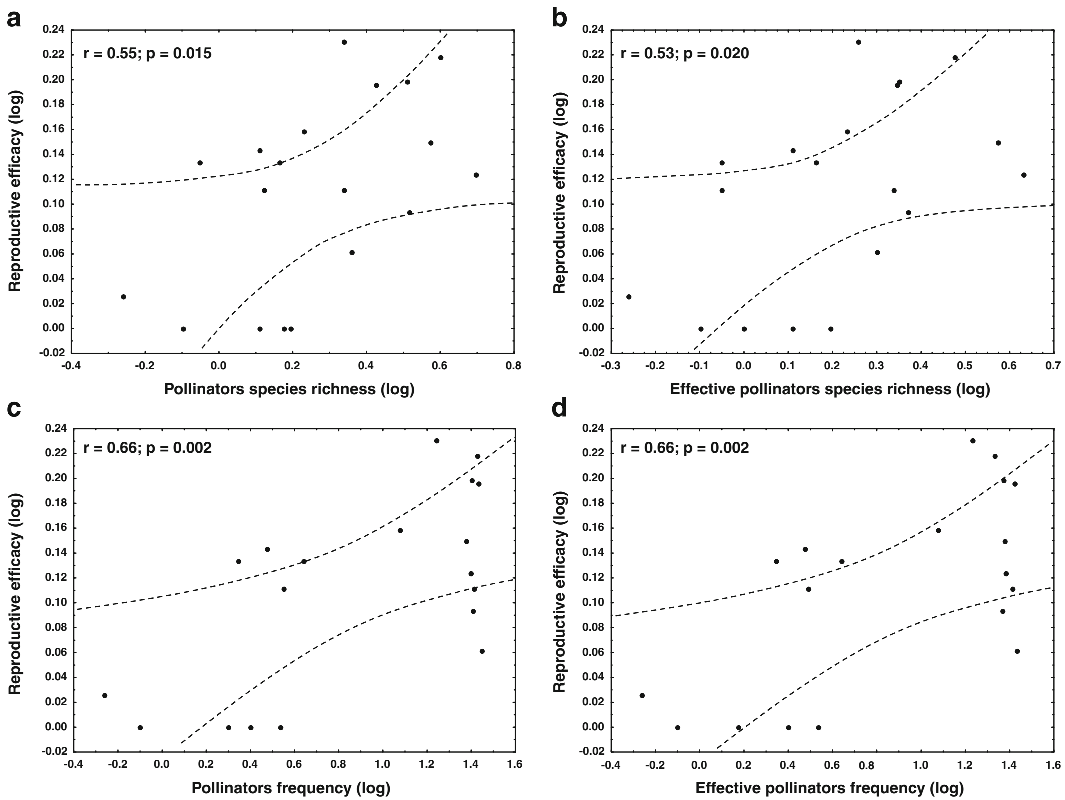

Figure 3. Significant correlations between visitors of passion fruit flowers and reproductive efficacy at different sample sessions. **a** Pollinator species richness, **b** effective pollinator species richness, **c** pollinator frequency, and **d** effective pollinator frequency. *Dashed lines* represent the confidence interval.

high percentage of rare species, relatively low population densities and wide variation in the composition of local faunas apparently related to floristic variation (Silveira and Campos 1995).

The abundance of pollinators has been proved to increase fruit set in other passion fruit orchard and the lack of them has been pointed as one of the main restriction to productivity in many areas in Brazil (Camillo 2003). Moreover, the frequency of pollinators had a positive correlation with fruit set of yellow passion fruit in orchards located in Southeastern Brazil (Benevides et al. 2009), but no study has reported a species richness as high as that found here.

Hence, probably the most important finding of this study is the apparent role of species richness of pollinators for the natural pollination efficacy of yellow passion fruit crop. Species-richer bee assemblages provide more stable pollination service to other crops (Garibaldi et al. 2011) such as watermelon (Kremen 2008), sunflower (Greenleaf and Kremen 2006) and coffee (Klein et al. 2003; Ricketts 2004; Steffan-Dewenter et al. 2006). But what would be the mechanism linking bee diversity and higher fruit set? Higher fruit set in passion fruit seemed to be the result of a higher efficiency in ovules penetration by pollen tubes after cross-pollinations. As the species is self-incompatible and self pollen tube growth is arrested in the style (Rêgo et al. 2000), reduced ovule penetration after natural pollination may be the result of a small amount of pollen deposited on the stigma but also of incompatible pollen tube arresting before ovule penetration. Since pollen tube growth and ovule penetration is a function of the amount of cross pollen, it is possible that pollen heterogeneity and the amount of cross pollen arriving on the stigma may be improved by the different foraging and visiting behaviours of a greater array of large bee species. In addition, the presence of multiple bee species may influence the movement patterns between flowers (Greenleaf and Kremen 2006).

The conservation status of the areas surrounding orchards was not examined in detail, but a preliminary broad assessment of the environmental quality suggests that conservation of natural environments may explain bee species richness. The apparently most disturbed Campo Alegre farm, the nearest to the urban area (around 8.3 km), had the lowest richness of pollinators and lowest fruit set. Bee species richness increases with the amount of semi-natural habitats in the landscape and shows the negative impact of agricultural intensification (Le Féon et al. 2010). Higher species richness closer to natural or semi-natural areas may result in pollinator communities more stable over space and time (Garibaldi et al. 2011). Furthermore, many studies have correlated higher pollinators richness in agroecosystems and pollination services availability with the proximity of natural areas (e.g. Greenleaf and Kremen 2006; Hoehn et al. 2008; Ricketts et al. 2008; Winfree et al. 2011) including the stability of pollination services (Garibaldi et al. 2011). Since passion fruit do not provide pollen for the large bee pollinators and even nectar offer is seasonal, surrounding natural vegetation is vital for persistence of these bees which are active all year round (Camillo and Garófalo 1982).

Although we did not find a significant effect of thieves species richness on reproductive efficacy, *A. mellifera* and species of *Trigona* have been considered pollen and nectar thieves of the passion fruit (Sazima and Sazima 1989; Leone 1990; Camillo 2003). The Africanized honey bee, *A. mellifera*, is an important pollinator for many crops (e.g. Delaplane and Mayer 2000), but is a poor pollinator of passion fruit and quickly depletes available pollen (Camillo 2003). Since thieves in passion fruit flower were bees of social habits with large colonies, it may explain their high visiting frequency. In contrast, most species of pollinators were solitary bees with relatively small populations, such as *Xylocopa* and *Centris* (*Ptilotopus*) species, which may represent <3% of the bees commonly observed visiting flowers in Brazil (Andena et al. 2005).

Natural pollination seems to be enough to maintain yellow passion fruit production at economically viable levels, which is around 20% (Silva 2005). Although fruit set values after hand cross-pollination were much higher, hand pollination has also a higher cost for the producer and tends to become more difficult to perform due the lack of trained workforce in an increasingly urbanised region (Klink and Moreira 2002). Since at least 67% of the Brazilian Cerrado biome had been converted to intensive human use by the early 2000s (Myers et al. 2000), this intensive land use has a great impact on nest substrates and natural floral resources for the large bee species that pollinate yellow passion fruit.

Natural pollination can be optimised by supplying nest substrate as nesting boxes (Freitas and Oliveira-Filho 2003) or bamboo canes (Camillo 2003). But pollinators would still need a continuous supply of floral resource in order to survive around passion fruit orchards. In any case, our results suggested that conservation efforts should focus also on the maintenance of bee diversity, since it has an important role on fruit set and sustainable production of passion fruit. This crop should be viewed as a flag agroecosystem where productivity depends on bee diversity and natural landscape and where good management practices would favour both production and conservation.

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Le rôle de la diversité des abeilles dans la pollinisation et la fructification des fruits de la passion jaune (*Passiflora edulis* forma *flavicarpa*, Passifloraceae) cultivés dans le centre du Brésil.

Cerrado / abeilles/pollinisateurs autochtones / richesse en espèces / *Xylocopa*

Die Rolle der Bienendiversität für Bestäubung und Fruchtansatz im Anbau der gelben Passionsfrucht (*Passiflora edulis* forma *flavicarpa*, Passifloraceae) in Zentralbrasilien.

Cerrado / große Bienen/einheimische Bestäuber / Artenreichtum / *Xylocopa*

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