Bee diversity in secondary forests and coffee plantations in a transition between foothills and highlands in the Guatemalan Pacific Coast

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Abstract

Background. Although conservation of pristine habitats is recognized in many countries as crucial for maintaining pollinator diversity, the contribution of secondary forest conservation is poorly recognized in the Latin American context, such as in Guatemala. San Lucas Tolimán (SLT) is a high-quality coffee production region from the Atitlan Province, which has the second highest deciduous forest cover in Guatemala and pristine forest is prioritized for conservation. In contrast, secondary forest protection is undetermined, since these forests are normally removed or strongly affected by coffee farming practices. This situation may affect the diversity of native pollinators, mainly bees, which usually rely on the secondary forest for food resources.

Methods. We conducted a study to investigate the importance of secondary forests around the SLT coffee plantations for pollinators. We compared bee diversity (richness, abundance and composition) in secondary forests of different age in coffee plantations with diverse farming techniques. Being the first study of pollinators in Guatemalan coffee plantations, we also recorded data for an entire year in order to describe bee seasonality.

Results. We found significant differences in bee diversity between the coffee plantations and secondary forests, while forest type was not as determinant for bee diversity. In the early dry season, secondary forests showed the greatest native bee diversity. During the late dry season, when the coffee was flowering, honeybees were dominant in the same plots. This study provides important management insights to support the conservation of pollinators, since our results offer guidelines to improve coffee production by increasing native pollinator diversity.

Key words: pollination, native bee diversity, Megachillidae, secondary forest, insect conservation

Introduction

Rates of land-use change in primary forests are increasing worldwide, threatening biodiversity (Montero-Castaño and Vilà, 2012). As a result, secondary forests become alternative habitats and resource providers that promote a faunal diversity more characteristic of primary forest (Peters et al., 2013; Taki et al. 2013). In tropical forests, one of the most threatened groups of fauna interacting in both primary and secondary forests are the pollinators (Winfree et al., 2011; Cariveau and Winfree 2015), a fact that highlights the importance of also conserving secondary forests (Taki et al. 2013, Winfree et al., 2011).

The conservation strategy in Guatemala for the past twenty-five years has been to preserve primary forest *in situ* (National Congress Decrees, 1989) by creating protected areas without management strategies to preserve the primary forest surroundings. As a result of this policy, the people in Guatemala are unaware of the role or ascribe little importance to secondary forest in terms of conserving biodiversity. In San Lucas Tolimán in Sololá, Guatemala, secondary forest is also underestimated; here, the traditional and conventional coffee farmers focus their conservation efforts on the pristine forest, mainly to ensure ecosystem services such as pollination and water provision. This is a good strategy for the conservation of native pollinators who nest in this forest (Jha and Dick, 2010; Klein et al., 2003; Klein et al., 2008; Rao and Stephen, 2010; Ricketts, 2004; Ricketts et al. 2008). However, many native pollinators also require secondary forest to obtain food resources throughout the year (Jha and Dick, 2010; Badano and Vergara, 2011; Klein et al., 2008; Kremen et al., 2004) and these are also important for maintaining biodiversity in general (Jules and Shahani, 2003; Kohler, 2007; Kremen et al., 2004; Kremen et al., 2007; Mandelik and Roll, 2009).

As some authors have suggested (Klein et al., 2007; Kremen et al., 2002, Ollerton et al., 2011; Winfree et al, 2007), among the vertebrate and invertebrate pollinators, bees are the most important pollination agents. Bees are responsible for pollinating nearly two thirds of crops worldwide (Brauman and Daily, 2008, Kremen et al., 2002); however, climate change and habitat fragmentation have endangered bee diversity and reduced bee populations, leading to a food crisis worldwide. Conservation of pollinators has therefore emerged as an issue of great importance (Abrol et al., 2012; Ollerton et al. 2011).

In recent years, Guatemala has become the seventh largest coffee producer in the world and it is the most important crop in the country in terms of the employment and foreign exchange that it produces (Anacafé, 2014). Sololá is a high-quality coffee region and crop fields are gaining territory at the expense of the forest due to the high demand for coffee produced in the region (Anacafé, 2014; Fischer and Victor, 2014).

At present in San Lucas Tolimán, the secondary vegetation commonly known as “monte” is normally cut down or treated with herbicides to prevent the secondary growth. Conventional farmers argue that by keeping the surroundings of the coffee clean they reduce the incidence of coffee pests, although there is no scientific evidence to support this belief. They also believe that by maintaining some primary forests they can guarantee pollinator diversity. On the other hand, the indigenous people who practice traditional farming are aware of the importance of secondary vegetation (Armas-Quiñonez Pers. Obs.). Through their traditional knowledge, they know that these represent a habitat for numerous important species. However, they also cut down all the secondary growth in common areas, arguing that it is for the safety of the children and for aesthetic purposes. In both cases, the secondary vegetation is removed. As a consequence, the area with secondary vegetation in the region could be insufficient to maintain the community of native bee pollinators, especially when the coffee is not in flower. In other words, the traditional and conventional coffee farmers are not aware of the critical importance of secondary forest to the preservation of the pollinators, a situation that must be addressed in Guatemala since it could lead to loss of biodiversity and subsequently to deficient coffee production (Philpott et al., 2008; Scheper et al., 2013; Steffan-Dewenter and Westphal, 2008).

In order to ensure production and obtain other income sources, coffee farmers of the Guatemalan highlands have introduced *Apis mellifera* hives into their farms. However, this practice may present some risk since it could have unknown impacts on native bee populations (Badano and Vergara, 2011; Garibaldi et al., 2011; Shavit et al., 2009; vanEngelsdorp and Meixner, 2010; Winfree et al., 2007), particularly in Guatemala where such interactions are poorly studied.

Coffee farming in Guatemala is very heterogeneous in terms of farming techniques. Big farms have changed from traditional management to conventional and highly intensive farming techniques that usually include higher inputs of agrochemicals, mainly pesticides, or on rare occasions have changed to integrated pest management (IPM). At the same time, traditional farmers use multi-farming techniques, where small coffee plantations are cultivated using intercropping with several banana hybrids (*Musa x paradisiaca*), papaya (*Carica papaya*), macuy *(Solanum americanum*), besides other crops. Traditional farmers usually cannot afford agrochemicals (pesticides, herbicides and fertilizers) to spray on their fields, and this management could therefore be contributing more than conventional coffee cultivation to the maintenance of bee diversity (Schmitt, 2006; Schuepp et al., 2012). However, in Guatemala, there have been no studies published that address this issue.

This study was therefore conducted in order to investigate the importance of secondary forests in maintaining pollinator diversity around the San Lucas Tolimán coffee fields. To accomplish this goal, the study was designed to compare bee diversity in plots featuring different stages of secondary forest and in coffee fields managed under a range of farming practices.

Materials & Methods

Study sites

This study was conducted from March 2013 through February 2014 in San Lucas Tolimán foothills region. San Lucas Tolimán is located at the limit between the central highlands and costal lowlands in southeastern Guatemala, where many Kaqchikel indigenous people live. San Lucas Tolimán is bordered by two volcanoes, Atitlán and Tolimán that range from 800 to 3500 masl and produce a variety of microhabitats. According to the Villar classification, the primary vegetation of studied farms are within a subtropical humid forest, where broadleaf evergreen forest divides the mountain forest from the tropical humid savanna on the Pacific coast (Villar, 1998). These biotic and topographic differences give the area a dynamic ecotone with high precipitation (Villar, 1998; 2003). According to the Guatemalan National Council of Protected Areas (Consejo Nacional de Áreas Protegidas, 2013), the department of Sololá, where San Lucas Tolimán is located, has 35% forest cover, making it the most forest-covered department in Guatemala. This fact is associated with the high degree of community conservation but is also due to the presence of private farms that normally have their own forest reserves.

Sampling design

Sampling was conducted at three coffee farms with different management types (Table 1). The farms harvest *Coffea arabica*, cultivar "caturra". The three farms have secondary forests nearby. The names of these private farms must be withheld at the request of the owners.

In each of the studied farms, three plots of 60 m^2 were established. Each plot was categorized as early secondary growth, late secondary growth or coffee plantation. Early secondary growth was characterized by early secondary forest with up to one year of development, mainly herbaceous vegetation with few bushes and an abundant incidence of light. Late secondary growth was characterized by late secondary forest with two to three years of succession, with mainly shrub vegetation and luminosity slightly restricted below the high bushes. Coffee plots were selected in patches of shaded coffee in the selected farms. Plot characteristics are presented in Table 1.

Bee collection

Bees were sampled every month from March 2013 to February 2014 in each plot, covering the two Guatemalan climatic seasons established as dry or summer from November to April and rainy or winter from May to October (CONAP, 2008). During three days each month, five people searched for bees and flowering plants in all of the selected plots. For each flowering plant species in each plot, bee sampling was conducted for 40 minutes at different times between 8:00 to 12:00 p.m. The sampling schedule was done considering results of previous temperature and humidity monitoring where optimal conditions for bee activity in the area were established. Bee sampling was based on the direct search method on flowers, using net sweeping (Brosi et al., 2008; McGavin, 1997; McMullen, 1965). Bees captured from flowers were killed by freezing in individual containers. Native bee (no honey bee) specimens were mounted on insect pins labeled with field data and assigning a unique code for deposition in the "Colección de abejas nativas del

Centro de Estudios Conservacionistas de la Universidad de San Carlos de Guatemala". In contrast, honeybee specimens were sampled and recorded but not collected. Taxonomical keys were used for bee identification to genus or species (wherever possible) (Ayala 1999; Michener 2000) and with the collaboration of Mabel Vásquez of the expert Ricardo Ayala.

Data analysis

Seasonal richness and abundance. In order to analyze bee and plant richness and bee abundance though time we used ANOVAs with the two known seasons as explanatory variables. In addition, a Pearson correlation was performed to evaluate the relationship between bee and plant richness.

Then we grouped the bees into honeybees, native bees and stingless bees, and we analyzed abundance and richness data per bee group per season (explanatory variable) with ANOVAs. Also, Pearson correlations were performed in order to determine whether bee richness, particularly honeybee incidence, was correlated with other native bee groups over the study seasons.

Bees in coffee fields and the surrounding secondary forest. Bee abundance, richness and diversity were evaluated by comparing the secondary forests and coffee plantations, with the three different farming techniques (Table 1). For richness, we used the Chao1 index and Abundance-based Coverage Estimator (ACE), in order to take both rare and abundant bee species into account. The incidence of both rare and common bee species was estimated with the Chao2 and Incidence Covered Estimator (ICE) indices. Diversity was calculated by Shannon-Wiener (H). These indices were calculated using the EstimateS Program (Colwell, 2013). Bee abundance, richness, incidence and diversity estimation values (response variables) were compared with ANOVAs, taking into account the plot type and farming type (explanatory variables).

In addition, we built linear mixed-effect models to test the effect of vegetation (coffee plantation or secondary forest stage) with farming techniques (farms) on local bee abundance. Also, groups of bee abundance (honeybees, native bees, stingless bees and family bees) were made to evaluate with linear mixed-effect as a function of vegetation and of farming techniques (explanatory variables). A significant interaction term between explanatory variables in the models indicates and suggest how wild bees could been modulated. Models were performed in R Program (R Core Team, 2014) using the nlme library.

Finally, a cluster analysis with Euclidean distances with 100 bootstrap samples was performed, using the bee diversity data to look for similarities between the vegetation and farming techniques (farms) and to infer the importance of the coffee management for the bees. ANOVAs, Pearson correlations and cluster analysis were also performed using the R program (R Core Team, 2014).

Results

Seasonal richness and abundance. Over one year of study, 3,004 individual bees, belonging to 102 species (Appendix 1) and 100 species of flowering plants with visiting bees (Appendix 2) were recorded. Bee and plant richness inside plots were significantly correlated (t_{36}=7.82, p=2.8e^{-09}, cor=0.79) showing a close relationship between them (Fig. 1a).

The study area showed a sparse flowering season from March to October 2013 and high flowering in the early dry season from November 2013 to February 2014, closely reflecting the two climatic seasons in Guatemala. A few months after the lowest records of bee abundance (March to October 2013), the rainy season promoted vegetative growth in the secondary forest, and it was correlated with the highest values of flowering plant richness with a 21% increase (Fig. 2 and Appendix 2).

Bee abundance differed significantly between seasons ($F_{(1,36)}$=4.66, P=0.04), as did honeybee abundance ($F_{1,36}$=6.47, P=0.015). In contrast, plant richness ($F_{1,36}$=2.62, P=0.11), bee richness ($F_{1,36}$=1.36, P=0.25), stingless bee abundance ($F_{1,36}$=3.38, P=0.07) and native bee abundance ($F_{1,36}$=1.27, P=0.27) did not differ between seasons (See Appendix 3). There was an interesting finding in February 2014 (Fig. 2) when bee richness increased by 57% (from 28 to 53 species), presenting the highest value in the study, but out of phase with respect to the peak of local flowering plants (December 2013), coinciding with the beginning of drought, with the biggest drop of flowering (14%). In the following month when the coffee plants were flowering (March 2014), the greatest number of bees was recorded (437 records), but the native bee species began to decrease (10%).

Interestingly, honeybees and stingless bees were significantly and positively correlated (r=0.62, t_{36}=4.78, p=2.95e^{-05}, Fig. 1b), as were the honeybees and native bees (r=0.61, t_{36}=4.61, p=4.84e^{-05}, Fig. 1c). Highlighting that for the three groups of bees, the availability of floral resources is important for their activity.

Bees in coffee plantations and the surrounding secondary forest. The farms presented different bee richness and abundance. As is showed in Figure 3, farm 1 had the highest number of bees followed by farm 2 and farm 3. Farms 1 and 2 presented the highest bee abundance in plots with secondary forest, while in farm 3 the coffee plantations presented the highest abundance.

Bee richness mainly comprises five families (Fig. 3). Most of the bees are Apidae (84.8%), followed by Halictidae (8.9%), Andrenidae (2.6%), Megachilidae (2.3%) and Colletidae (1.5%). Species from the five bee families were registered in the three farms. On farm 3, 91% of captures belonged to Apidae, while farms 1 and 2 had a higher representation of Colletidae and Andrenidae. Bee family abundance per vegetation type (early secondary forests, late secondary forests and coffee plantations) showed a significant difference only for the Megachilidae family ($F_{2,4}$=7.005, p=0.049), particularly between early growth and coffee plantation (early growth with coffee: p=0.04; according to the Tukey multiple comparisons test, see Fig. 4a).

Total bee richness per farm and vegetation type showed no significant differences (p>0.05). Although differences were found between bee groups (Fig. 5), the stingless bees differed among farms ($F_{2,4}$=9.01, p=0.03), according to the Tukey multiple comparisons test, this difference was between farms 1 and 3 (p=0.03; Fig. 4b).

The diversity estimators Chao1, ACE, ICE (Fig. 6 and Appendix 4) calculated per farm and vegetation type did not present any significant difference (p>0.05). The Chao2 estimator showed significant differences among farms ($F_{2,4}$=7.2, p=0.047), between farms 1 and 3 (p=0.044; according to Tukey multiple comparisons test; Fig. 4c). The Shannon-Wiener diversity index (H) did not present significant differences among farms. In farms 1 and 2, the richness estimators gave higher values to plots with early secondary forest, while the highest estimate value on farm 3 was for the coffee plantation. The species accumulation curves showed stabilized curves that starting lie down in all plots (Fig. 7). Coffee plantations registered the lowest values in bee diversity, and accordingly, the richness estimators predicted the lowest number of species (Fig. 6 and 8). In farms 1 and 2, the coffee plantations registered most of the bee records during the coffee flowering season, otherwise, neither bee activity nor early growth vegetation were recorded because they clean adventive vegetation from coffee plantations, particularly in farm 2. On farm 3, the coffee was rarely cleaned in this manner, which contributed to the establishment of early secondary forest vegetation.

An important finding was the presence of a new species of bee genus *Rhathymus* Lepeletier & Serville, 1828, *Rhathymus atlitlanicus*, described in Ayala, Hinojosa-Díaz & Armas-Quiñonez (2019). Those bees were only found in secondary forest of farm 1 and farm 3 (Appendix 1).

With the linear mixed-effect models we found a significant interaction between farms (farming techniques) and vegetation type (coffee or secondary forest stage) for bee abundance (Table 2). We found a marked positive effect on total bee abundance and different bee groups in early secondary forests. However, in farm 3 a negative interaction of bee abundance without honey bee was found and a positive interaction with honey bee, suggesting an exclusive effect of each. Regarding particular bee families, a stronger and more positive effect was found for Apidae abundance in secondary forests. Halictidae and Andrenidae were more abundant in farm 2.

Finally, the cluster analysis of bee diversity per vegetation type (Fig. 9) presents a significant pattern that groups the study plots into two significant aggregations. The strongest aggregation with 97% confidence is composed by the early secondary forests from farm 1 and farm 3 and the coffee plots of farm 3 (the farm that allows secondary forest plants into the coffee plantation). The other aggregation with 97% confidence, is composed by the coffee and late secondary forests of all three farms. Within this group there are significant differences among the plots of farm 1, where coffee is harvested following traditional practices, and on the other side of the same group, is the late secondary forest linked with the coffee of farm 2. Cluster analysis shows that the early secondary forest in farm 2 is 77% different to the rest of the plots.

Discussion

The effect of flowering seasonality on bees. The bees recorded over the whole year in secondary forest and coffee fields could give an insight into the synchronization that exists between bees and phenology of flowering plants, due to the periods of highest bee abundance matching with flowering periods and the correlation between these parameters throughout the study. This correlation support previous findings (Brosi, 2009, Banks et.al, 2014, De Marco and Monteiro, 2004, Taki et al., 2013) about the great importance of secondary forests in the provision of resources to bees. This seasonality can have some important consequences for bee diversity but also for coffee production, since the highest abundances of native bees were observed during secondary forest flowering (March 2014), close to the coffee flowering that varies according to the first rains. Early flowering of coffee could coincide with secondary forest flowering, which would cause native bees to interact with the high honeybee abundance possibly giving rise to a saturated environment of pollinators for the coffee and secondary forest. However, the asynchrony of pollinators and flowering periods in coffee fields must be treated with care and taken into account in crop management as a priority for improved of coffee production (Boreux et. al., 2013).

When investigating the relationship between honeybee and native bee abundance, we found a positive correlation between them, as other studies in Mexican coffee plantations have found (Badano and Vergara, 2011). This fact suggests that honeybees may not yet have saturated this ecosystem, and that, according to Banks et al. (2013), the contribution of pollinators from nearby forests to the coffee plantations is still high. This notion can be supported by the fact that honeybees only maintain high populations during coffee flowering, otherwise, their cultured populations are kept to a minimum in the area. However, in the linear mixed-effect model in farm 3, where honey bee were more abundant, a very strong negative effect was found for native bee abundance. In this farm honey bees may be displacing native bees (Thomson, 2016).

Secondary forests as a coffee pollinator enhancers

The data of bee abundance, richness and diversity supports the importance of secondary forests for bee pollinators (Arnan 2010, Carvalheiro 2012, Banks et al. 2013, Brosi 2008; Boreux et al. 2013), not only to maintain bee diversity, but also to improve coffee production in the Pacific Coast Foothills of Guatemala. The significant mixed effect model remarks how the first stage of secondary forest -early growth- interact positively with bee abundance (Table 2). Meanwhile, the presence of stingless bees in the secondary forests and also in the flowering coffee plants, suggests that these bees, which depend exclusively on the forest for nesting, also depend on secondary forests for the acquisition of essential resources (Winfree 2010, Vanddeler 2006).

Secondary forests showed that they can maintain and provide resources for the native bees during periods when the coffee is not flowering. The farmers that use conventional agriculture need to place greater emphasis on preserving secondary forests, rather than only pristine forests (Blanque 2006). Pollination tests are required in order to compare the pollination efficiency of native bees and honeybees in these plantations. In this way, it would be possible to more definitively establish the importance of preserving native bees and the places where they obtain resources, such as the secondary forest for coffee production (Winfree 2010).

Effect of farm management

We found important differences in bee diversity between farms. Farm 1 shows the highest diversity values, suggesting that, despite the lack of high technology, traditional knowledge remains effective in preserving native bee diversity, especially for the stingless bees. The conventional farm with high-intensive management (farm 2) keeps the coffee plantations cleared of early secondary forest but maintains surrounding secondary vegetation on the farm; therefore, the presence of *Tephrosia* spp. could function as a provision plant for native bee species. A positive interaction between farm 2 and bee family abundance can suggest the importance of nearby forest, since the farm 2 manage their own forest reserve, providing the bees with a complex landscape that may enhance bee resource acquisition, as suggested previously by different authors (Carrié *et. al.* 2016, Winfree, et al., 2009).

Also, it is evident that the conventional farming with low-intensity management and a disturbed surrounding forest (farm 3) has the lowest bee diversity and, that it is the early secondary forest left in the coffee fields for a short time period, that function as resource provider for the surrounding bee diversity. This could explain why, in the cluster analysis (Fig. 9), the coffee of farm 3 was grouped together with the early secondary forest of farm 1 and 2.

None of the studied farms use honeybee breeding to pollinate coffee. Some neighboring farms, however, do have managed bee hives and it is possible that the honey bees recorded during the study came from those neighboring farms. The farm with the closest neighboring honeybee hives was farm 3, which showed the lowest bee abundance, richness and diversity values as well as the highest honeybee abundance. This fact is highlighted in the linear mixed-effect model where farm 3 showed positive interaction with honey bee abundance but negative interaction with other bees. On the other hand, this farm (3) also shows a poor surrounding forest management providing few resources for native bees, contrary of farm 1 that promotes this mixed-landscape (Carrié *et. al.*2016).

Another factor to consider that may affect bee diversity is the use of chemicals in the farms: the quantity of insecticides used for pest control inside (such as that used to control the Mediterranean fruit fly), but also those used outside farms in neighboring crops cultivated near coffee (Brittain et al. 2010; Schmitt 2009; Schuepp 2012).

Conclusions

In coffee plantations the presence of secondary forest in the early growth stage, show a significant positive effect on bees, and taking into account the importance of bees in pollination this is a natural way to increase the pollinators. Regarding farm management, this study can be used to apply certain strategies that would benefit native bee diversity around the country. Through incorporating some traditional farming techniques into conventional coffee field management, such as letting the surrounding secondary forests grow at least in the early dry season, it can be demonstrated that these provide floral resources for native bees that will also visit the coffee during its flowering stage. Our results also support the value of traditional farming, which in this study demonstrated a high diversity of native bees, capable of pollinating small coffee plots and thus saving the cost of maintaining honeybee hives for pollinating small crops.

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Figure 1

Bee richness and plant richness

Pearson correlations between: (a) Bee richness and plant richness ($r=0.79$, $p=2.8e^{-09}$). (b) Honeybee and stingless bee abundance ($r=0.62$, $p=2.95e^{-05}$). (c) Honeybee and native bee abundance ($r=0.61$, $p=4.84e^{-05}$)

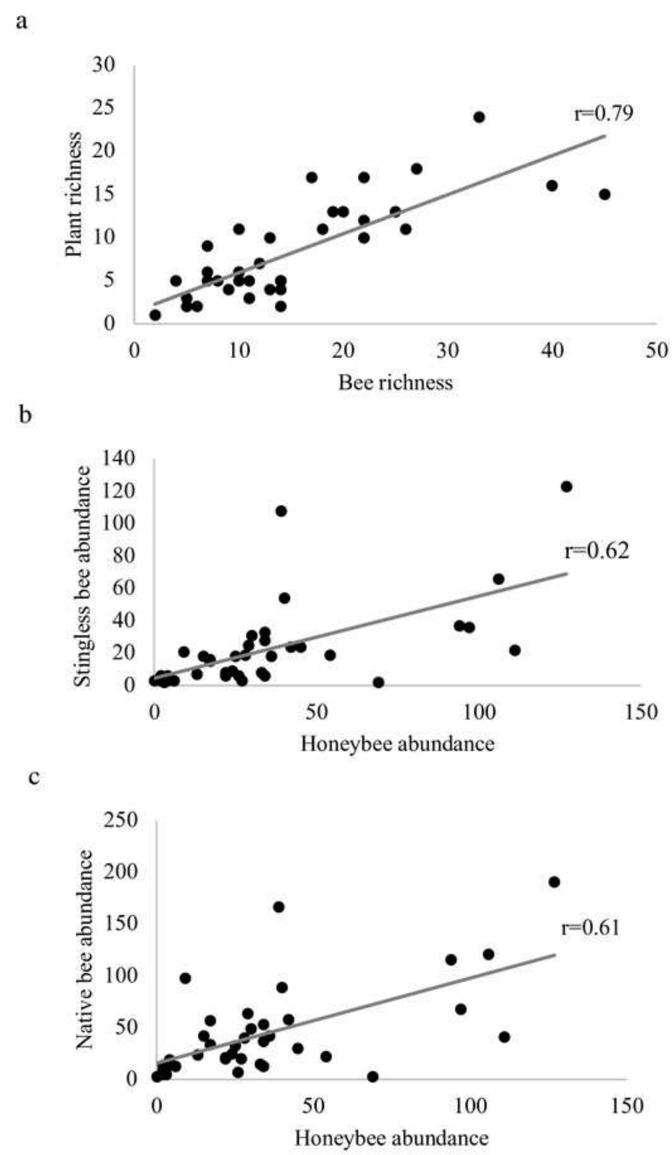


Figure 1. Pearson correlations between: (a) Bee richness and plant richness ($r=0.79$, $p=2.8e^{-09}$). (b) Honeybee and stingless bee abundance ($r=0.62$, $p=2.95e^{-05}$). (c) Honeybee and native bee abundance ($r=0.61$, $p=4.84e^{-05}$).

Figure 2

Bee and flower through time

Variation of bee and flowering plant richness through time, showing the two growing seasons in the region.

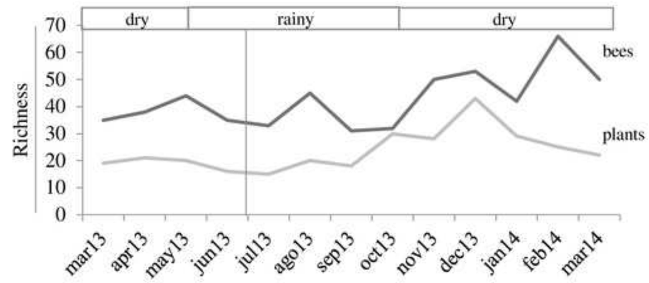
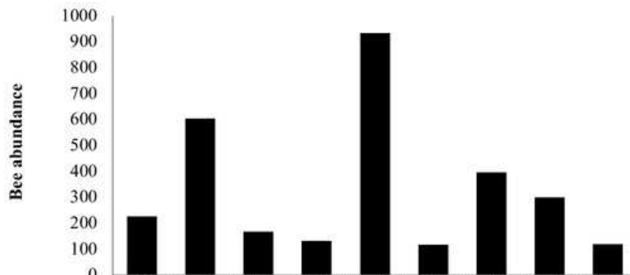


Figure 2. Variation of bee and flowering plant richness in months in which the samplings were conducted and the two seasons

Figure 3

Total bee abundance per farm and vegetation type

Total bee abundance per farm and vegetation type. Cumulative abundance records per farm and plot are shown in the upper part of the figure. The lower table shows the total abundance of records per bee family



	FARM1 coffee	FARM1 early	FARM1 late	FARM2 coffee	FARM2 early	FARM2 late	FARM3 coffee	FARM3 early	FARM3 late
■ Megachilidae 2.3%	7	20	3	0	21	3	0	10	4
■ Halictidae 8.9%	16	35	6	6	159	6	29	3	7
■ Colletidae 1.5%	9	4	4	0	23	2	2	0	1
■ Apidae 84.8%	192	546	155	123	681	105	349	288	108
■ Andrenidae 2.6%	4	0	1	3	51	2	16	0	0

Figure 3. Total bee abundance per farm and type plots. Cumulative abundance records per farm and plot are shown in the upper part of the figure. The lower table shows the total abundance of records per bee family.

Figure 4

Bee diversity differences

Tukey Multiple Comparison test, significant results. (a) Differences between landscape type plots according to Megachilidae bee family abundance. (b) Differences between farms according to the Chao2 richness estimator. (c) Differences among farms and stingless bee abundance. Different upper-case letters denote significant differences between evaluated groups.

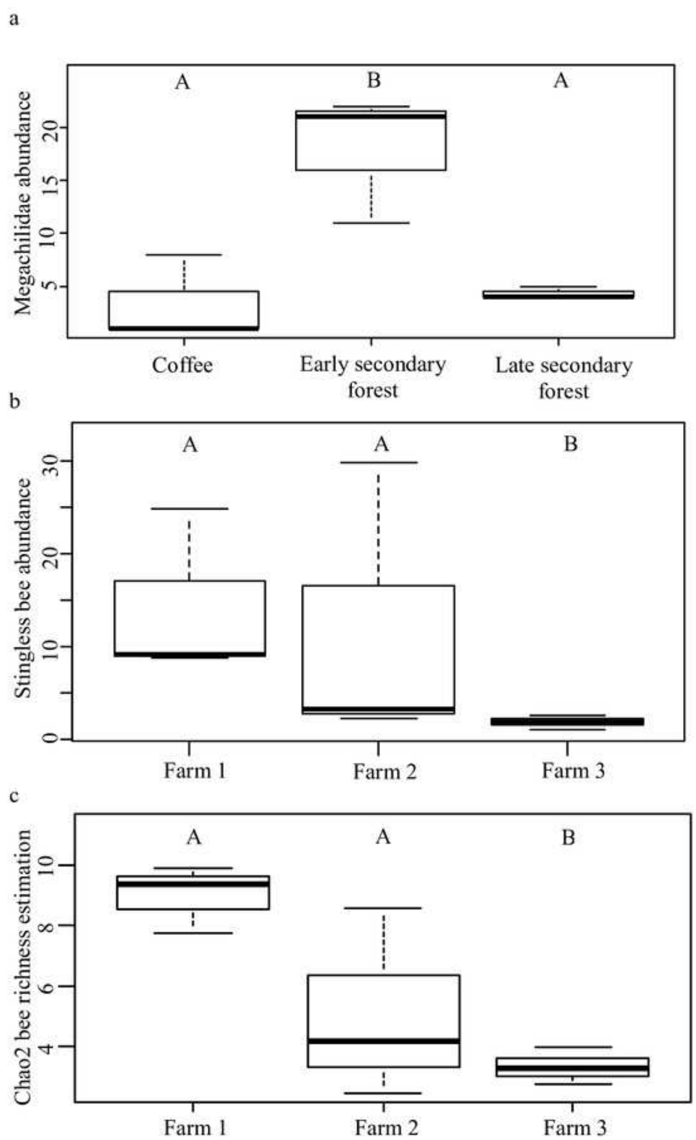


Figure 4. Tukey Multiple Comparison test, significant results. (a) Differences between landscape type plots according to Megachilidae bee family abundance. (b) Differences between farms according to the Chao2 richness estimator. (c) Differences among farms and stingless bee abundance. Different upper-case letters denote significant differences between evaluated groups.

Figure 5(on next page)

Bee abundance per farm and vegetation type

Bee abundance per farm and vegetation type. Honeybee abundance is shown in black, native bee abundance in light grey and stingless bee abundance in dark grey.

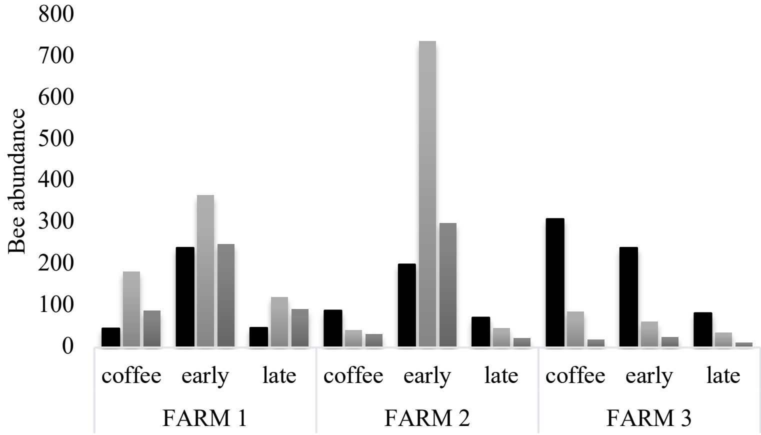


Figure 5. Bee abundance per farm and landscape type plots. Honeybee abundance is shown in black, native bee abundance in light grey and stingless bee abundance in dark grey.

Figure 6

Bee richness and diversity estimators per farm and vegetation type

Bee species richness per farm and vegetation type. S represents the bee richness found, ACE represents the abundance-base coverage richness estimator (S_{ACE}), ICE represents the incidence coverage estimator (S_{ICE}), Chao1 richness estimator (S_{Chao1}), and Chao2 incidence estimator (S_{Chao2}).

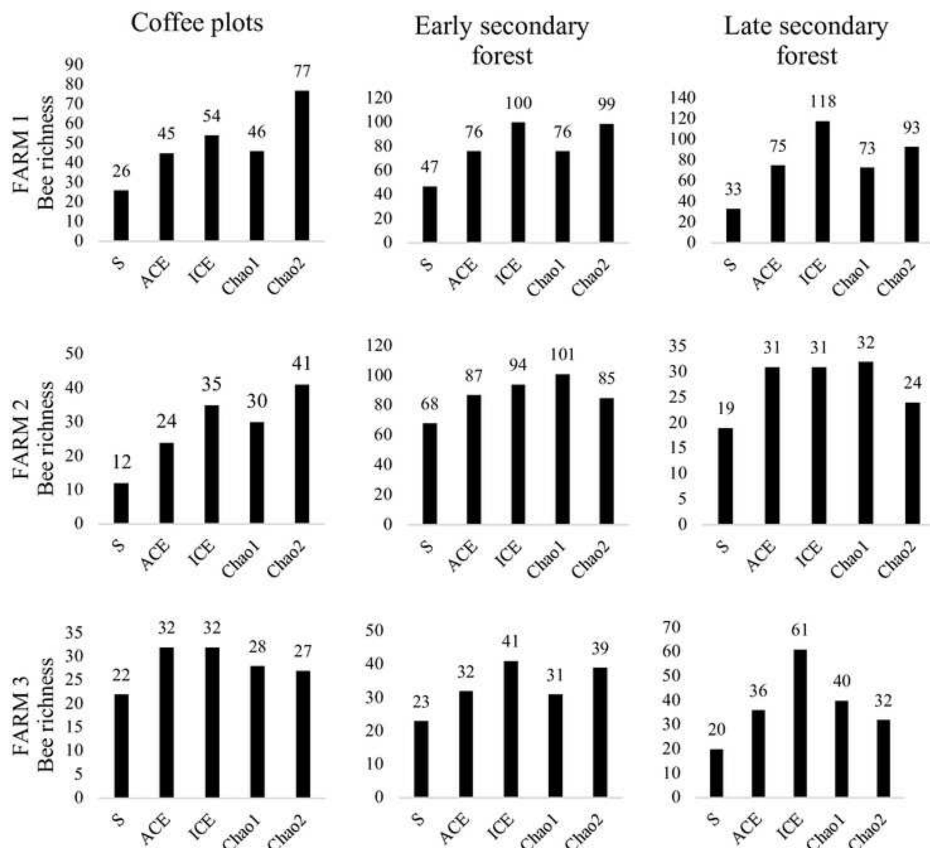


Figure 6. Bee species richness per farm and landscape type plots. (B) Bee richness. (ACE) Abundance-base coverage richness estimator -S_{ACE-}. (ICE) Incidence coverage estimator -S_{ICE}-. (Chao1) Richness estimator -S_{Chao1}-. (Chao2) Incidence estimator -S_{Chao2}-.

Figure 7

Bee species accumulation curves

Species accumulation curves for bee richness obtained in the study per farm and vegetation type

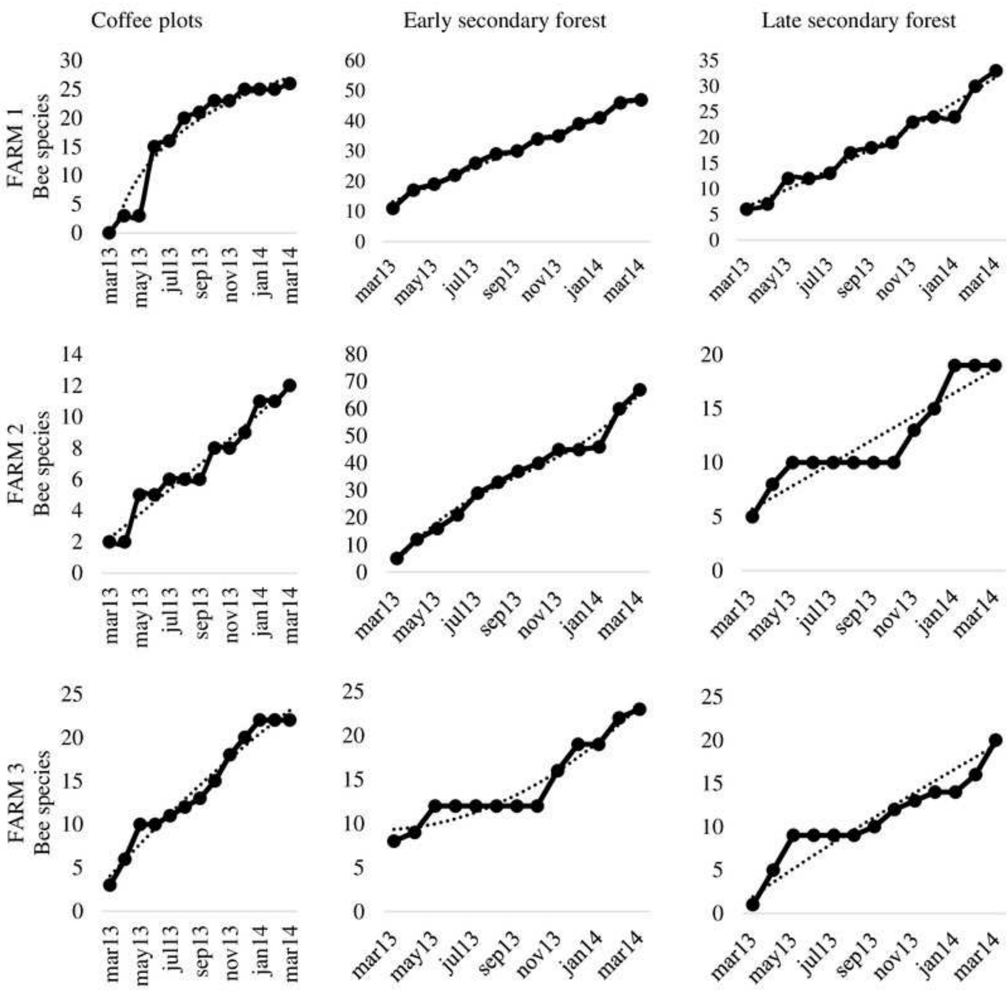


Figure 7. Species accumulation curves of bee richness obtained in the study per farm and type plots.

Figure 8

Bee richness and abundance registered per vegetation type plots.

Bee richness and abundance registered per vegetation type plots. Black bars show farm 1 data, light grey bars show farm 2 data and dark grey bars show farm 3 data. The total richness and abundance values are shown in the numbers above the bars

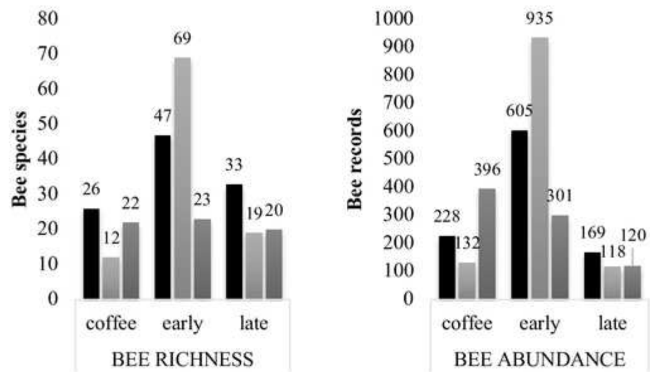


Figure 8. Bee richness and abundance registered per landscape type plots. Black bars show farm 1 data, light grey bars show farm 2 data and dark grey bars show farm 3 data. The total richness and abundance values are shown in the numbers above the bars.

Figure 9

Dendrogram showing differences in bee species communities

Dendrogram showing total bee species abundance recorded between plots of the three farms (cluster analysis calculated in R using Vegan and Pvclust packages and Euclidean distance). The*p*-value is shown at the top of each edge, presented as a percentage value of confidence, in which a value of 95 or higher represents a significant supported data aggregation. In the red is shown the two significantly groups found.

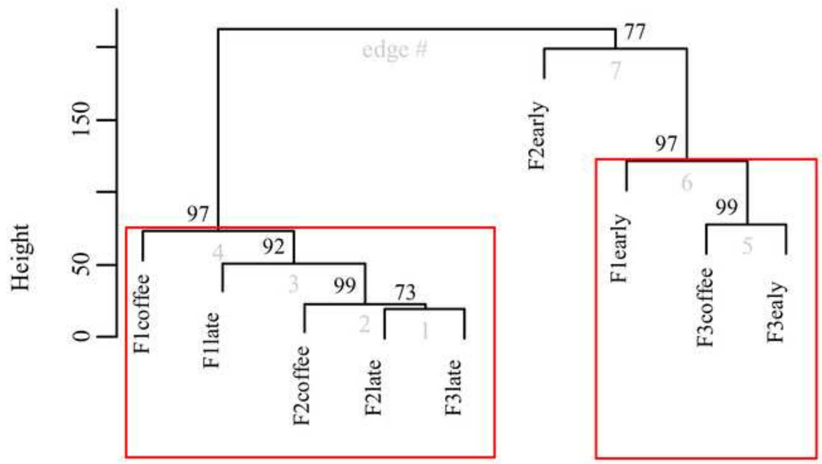


Figure 9. Cluster dendrogram of bee species abundances recorded between plots of the three farms (cluster analysis calculated in R using Vegan and Pvclust packages and Euclidean distance). The *p*-value is shown at the top of each edge, presented as a percentage value of confidence, in which a value of 95 or higher represents a significant supported data aggregation. In the red is shown the two significantly groups found.

Table 1(on next page)

Description of plots for the three studied sites.

In each farm vegetation type, coffee farming management and adjacent forest management is described

1 **Table 1.** Description of plots for the three studied sites.

Coffee plantation farms	Type of plots	Coffee farming techniques	Adjacent forest management
1	**Early:** short height secondary forest **Late:** medium height secondary forest **Coffee:** traditional coffee field, shaded with native species	Traditional farming: traditional methods of pest removal, rare use of chemical fertilizers and pesticides.	Community forest management. The community regulates and controls the use of the forest, creating a preserved forest.
2	**Early:** short height secondary forest **Late:** medium height secondary forest **Coffee:** shaded coffee with *Grevillea robusta* and *Inga* sp.	Conventional with high intensity farming practices: controlled production, integrated pest control with minimum use of pesticides and herbicides.	Private reserve. Forest with low human disturbance and no access granted to the local people, creating in a preserved forest.
3	**Early:** short height secondary forest **Late:** medium height secondary forest **Coffee:** shaded coffee with *Grevillea robusta*.	Conventional with low intensity farming practices: uncontrolled production and pest control, occasional use of pesticides and herbicides.	Private reserve. Forest with human intervention and low control of access for local people, producing a disturbed forest.

2

Table 2 (on next page)

Significant interactions found in generalized linear mixed-effect model

Significant interactions found in generalized linear mixed-effect model assessing the effect of Farm and vegetation type on total bee abundance an abundance per bee group, with and without honey bees.

Table 2. Significant interactions found in generalized linear mixed-effect model.

Response variable	Variables (farm.landscape)	Estimate	SE	p-value
TOTAL BEE ABUNDANCE	Early secondary forest	**28.41**	5.40	0.000
BEE GROUPS				
Honey bee abundance	Farm3	8.65	3.08	0.0062
	Early secondary forest	6.24	2.99	0.0400
Bee abundance without honey bee	Farm3	-12.61	4.2	0.0042
	Early secondary forest	**22.11**	4.17	0.0000
Stingless bee abundance	Early secondary forest	**17.33**	4.89	0.0001
Social bee abundance	Early secondary forest	**17.52**	4.04	0.0000
BEE FAMILIES				
Andrenidae abundance	Farm2	1.483	0.551	0.0085
Apidae abundance	Early secondary forest	**22.222**	4.36	0.0000
Halictidae abundance	Farm2	3.329	1.012	0.0014
	Early secondary forest	3.83	0.974	0.0002
Megachilidae	Early secondary forest	1.147	0.34	0.0011