



Coffee cover surrounding forest patches negatively affect Euglossini bee communities

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Abstract – Orchid bees (Euglossini) are pollinators sensitive to landscape pressures related to agricultural land use, such as coffee farming. Coffee crops occupy a large land area in Brazil, and understanding the effects of coffee farming on bee communities is essential to pollinator conservation in modified landscapes. Here, we evaluated the Euglossini communities in forest patches surrounded by coffee crops in the Atlantic Forest. We hypothesized the negative effects of coffee cover (%) on euglossine richness and abundance. The euglossine males were sampled at a sampling point within forest patches of 15 landscapes in southeastern Brazil. A total of 1890 euglossine males in four genera and 14 species were sampled. *Eulaema nigrata* Lepeletier, 1841 was the dominant species (55.1%), followed by *Euglossa cordata* (Linnaeus, 1758) (25.5%). We found a new record for *Euglossa liopoda* Dressler, 1982, increasing the species' known range in the Atlantic Forest. The results showed that the euglossine richness and species abundance decreased in forest patches surrounded by a high coffee cover (%). These negative effects of coffee cover on the Euglossini communities are related to forest cover substitution by monocultures with low or no floral attractiveness for these bees. This study highlights that forest patches in agricultural landscapes sustain high levels of euglossine richness. Thus, we indicate the conservation importance of these Atlantic Forest patches for bee species requirements.

orchid bees / neotropical forest / coffee farming / agricultural landscape / pollinators

1. INTRODUCTION

Agricultural expansion and intensification are one of the main causes of habitat loss and fragmentation worldwide. These human activities affect the

landscape structure through changes in patch types and spatial arrangement of these elements in the landscape (i.e., landscape composition and configuration, respectively) (Fischer and Lindenmayer 2007; Fahrig 2017). Moreover, the appearance of anthropogenic matrices (i.e., areas of anthropic use) can increase isolation between habitat patches (Driscoll et al. 2013), influencing species maintenance and essential ecological services in modified

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landscapes, such as pollination by bees (Kremen et al. 2007; Cariveau and Winfree 2015; Martello et al. 2016).

The orchid bees (Hymenoptera: Apidae: Euglossini) are a Neotropical group with more than 240 species (Roubik and Hanson 2004; Moure et al. 2012). These bees are fundamental to the ecosystem's functioning through pollination, as well as are essential for pollination ecosystem services within agroecosystems (Roubik and Hanson 2004; Briggs et al. 2013). Different morphological and behavioral traits of Euglossini bees (e.g., long tongue, robust body, buzz behavior) make them important pollinators for many plant families (Cortopassi-Laurino et al. 2009; Rocha-Filho et al. 2012). Moreover, these bees have a broad flight range (Wikelski et al. 2010) and are pollen vectors of the plant species spatially distributed in the landscape (Janzen 1971).

For some reasons, Euglossini bees constitute important ecological indicators of environmental and landscape disturbances, including their forest dependence, wide geographic range, and easy sampling (Allen et al. 2019; Gonçalves and Faria 2021). These bees are quite sensitive to biotic and abiotic variable changes, such as phytophysiognomy type, altitude, temperature, and humidity (Aguiar and Gaglianone 2012; Sobreiro et al. 2019). Moreover, ecological attributes such as euglossine abundance show a high association with landscape structure (Allen et al. 2019; Carneiro et al. 2022), indicating that these bees are good ecological models for understanding bee responses to land-use changes (Brosi 2009; Allen et al. 2019; Gonçalves and Faria 2021). Several studies highlighted the influence of the landscape structure on Euglossini communities (e.g., Cândido et al. 2018; Allen et al. 2019; Opedal et al. 2020). These studies have indicated the influence of forest patch attributes and landscape context on euglossine richness and abundance (Brosi 2009; Carneiro et al. 2021; Sousa et al. 2022). For other bee groups and invertebrates — such as dung beetles, ants, wasps, and flies — anthropogenic matrices play an essential role in maintaining the richness and species abundance (Martello et al. 2016, 2022; Medeiros et al. 2021).

Landscape composition is important for euglossine bees because it can provide complementary habitats or niche opportunities for some of these bee species (Hedström et al. 2006a; Aguiar et al. 2015; Carneiro et al. 2021). There is a remarkable decrease in euglossine richness and abundance in matrices such as forestry (e.g., commercial *Eucalyptus* plantations) and coffee crops compared to forest areas (Briggs et al. 2013; Aguiar et al. 2015). Furthermore, highly managed agroecosystems are less friendly to euglossine species exhibiting a higher dependence on forested habitats (Milet-Pinheiro and Schlindwein 2005; Aguiar et al. 2015), and among many factors, this probably results from higher input of agrochemicals, such as fertilizers, insecticides, fungicides, and herbicides.

Although these orchid bees show consistent responses to agricultural landscape attributes (Allen et al. 2019; Carneiro et al. 2021), the influence of coffee cover (%) on euglossine communities is poorly understood (Briggs et al. 2013). Some studies in Central America indicated that coffee farming systems (e.g., monoculture, polyculture) affect the Euglossini community's composition (Hedström et al. 2006a, b; Briggs et al. 2013). Brazil has been globally responsible for 34.9% (sd. 1.8%) of coffee production in the last 5 years (International Coffee Organization 2021). The cultivated area exceeds 2.2 million hectares, 1.9 million of them in southeastern Brazil (Conab 2021). Since Atlantic Forest originally covered this region, the expansion of coffee farming contributes to the high forest loss of this biodiversity hotspot. Thus, evaluating the influence of coffee cover (%) on euglossine communities can indicate the effects of forest cover loss related to agricultural expansion and intensification in tropical regions (Briggs et al. 2013; Dicks et al. 2021), and show paths to sustainable agriculture practices in modified landscapes combined with pollinator conservation.

Here, we evaluated the Euglossini community structure in 15 agricultural landscapes. We then analyzed how coffee cover (%) surrounding forest patches affects the euglossine richness and species abundance. Coffee species bloom a

few times a year, and although many bee species benefit from floral resources during this short flowering period (Hipótilo et al. 2018), the use of these resources by Euglossini species remains unknown. For this reason, and considering that coffee farming can negatively affect the Euglossini communities (Hedström et al. 2006a; Briggs et al. 2013), we expected a decrease in bee richness and abundance in landscapes with high coffee cover (%).

2. MATERIAL AND METHODS

2.1. Study area

We surveyed 15 agricultural landscapes in southeastern Brazil (Fig. 1), located in the municipalities of Varre-Sai (Rio de Janeiro state; L01–L06), Espera Feliz (Minas Gerais state; L07–L09), Guaçuí, and Alegre (Espírito Santo state; L10–L15). This region has altitudes varying between 500 and 2000 m. There are crops of *Coffea canephora* Pierre ex A. Froehner in the lower altitude regions and *Coffea arabica* Linnaeus in the higher altitude areas. We delimited the landscapes along a gradient of forest cover (Carneiro et al. 2021) and coffee cover (0.07–55.85%) (Fig. 1). Detailed information about the study area can be found in Carneiro et al. (2021).

2.2. Euglossini bee sampling

We selected a forest patch in each agricultural landscape, with a minimum size of 5.0 ha, and surrounded by coffee crops (Fig. 1). We sampled the euglossine males within the forest patch at least 200 m distant from the forest edge. The sampling was carried out during three days in the rainy season (November 2019 to March 2020) and two days in the dry season (August to September 2019, July 2020) (Carneiro et al. 2021). We used traps with five baits to collect the euglossine males: eucalyptol, eugenol, methyl cinnamate, methyl salicylate, and vanillin. The sampling method is detailed in Carneiro et al. (2021) and Carneiro et al. (2022). The specimens have been deposited in the entomological collection of the Experimental Ecology sector, Laboratório de Ciências Ambientais—LCA, Universidade Estadual do Norte Fluminense Darcy Ribeiro—UENF.

2.3. Coffee cover quantification

We delimited the landscapes with 1500-m buffers from the bee sampling point, followed by vectorization and visual classification with satellite images in ArcGis software. From the

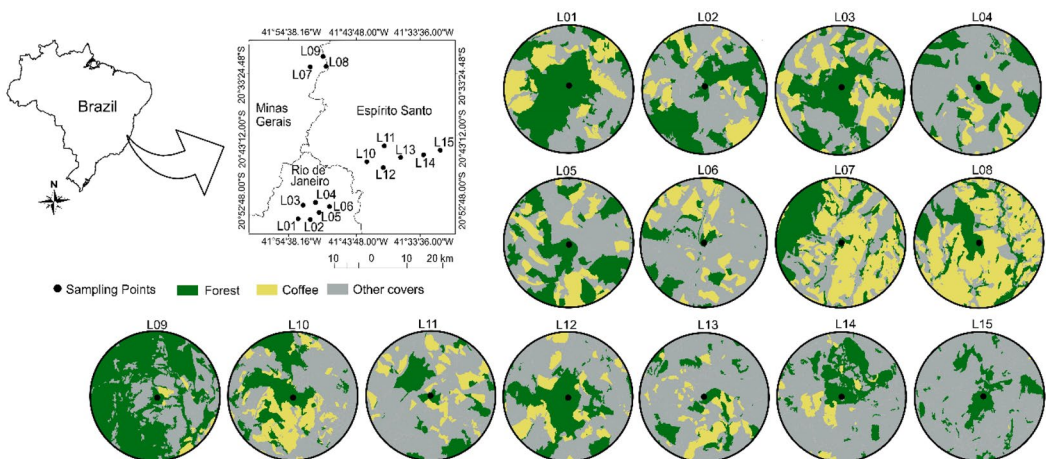


Figure 1. Geographical location of the Euglossini sampling points and spatial context of the 15 landscapes in coffee farming regions in southeastern Brazil.

rasterized map (5-m resolution) (Carneiro et al. 2021), we used the *lsm* function of the R landscapemetrics package (Hesselbarth et al. 2019) to quantify the coffee cover (%) in the landscape. For this, we used the PLAND landscape metric, which quantifies the class percentage in the landscape (McGarigal 2015). Because euglossine bees respond to landscape attributes in different spatial scales (Brosi 2009; Carneiro et al. 2022; Sousa et al. 2022), we used a multi-scale approach to measure the coffee cover (%) in different spatial scales, from 300 to 1500 m, with 50-m intervals. In this range are included the scales within which landscape attributes have been shown to influence euglossine richness and abundance (i.e., the scale of effect) (Brosi 2009; Cândido et al. 2018; Carneiro et al. 2021).

2.4. Data analysis

We evaluated the Euglossine community structure through the Shannon–Wiener diversity ($H' = -\sum p_i \ln p_i$), Berger–Parker dominance ($D = N_{\max}/N$), and Pielou uniformity ($J' = H'/\ln(S)$) indexes (Magurran 2004). We used rarefaction curves to correlate the number of sampled individuals with the species richness in each landscape. We plotted the rarefaction curves for each landscape group considering its geographic location (L01–L06; L07–L09; L10–L15, see Fig. 1) because there is a high variation in bee abundance between landscapes and to visualize the rarefaction curves better. These analyzes were performed with the Past 4.01 software (Hammer et al. 2001). The community composition similarity between the landscapes was quantified through Renkonen's index, recommended for small samples (Wolda 1981). We used Renkonen's values for an Unweighted Pair Group Method with Arithmetic Mean—UPGMA analysis. The UPGMA dendrogram was plotted from the small distances of Renkonen's similarity. We used a bootstrap with 1000 randomizations to quantify the stability and reliability of the clusters in the UPGMA dendrogram. Both Renkonen index and UPGMA were calculated

with the R R4eco package (available at: <http://github.com/wilsonfrantine/R4eco>).

We classified the species into common and rare using an inflection point and Gaston's quartiles criteria (Gaston 1994; Siqueira et al. 2012). In these approaches, a relative abundance curve is used. From the curve shape change (i.e., inflection point), we visually defined the common species on the left side and rare species on the right side (Siqueira et al. 2012). Gaston's quartile criterion classifies the 25% most abundant species as common and the 25% less abundant species as rare. However, we also considered the species distributed in second and third quartiles to avoid singletons and doubletons being included in the rare species group (Siqueira et al. 2012). Then, we classified the common species as the 25% most abundant species and the 75% less abundant as rare species (Fig. S1).

We performed a multiscale analysis similar to Gestich et al. (2018) and Ahuatzin et al. (2021) to previously determine the scale of effect of the coffee cover (%) on the euglossine richness, abundance of common and rare species. The scale of effect is the spatial scale of a landscape attribute that best explains biological parameters (Miguet et al. 2016). For this, we used the *multift* R function (Huais 2018) with the R^2 selection criteria in a nested multiscale approach (Carneiro et al. 2022). The scale of effect was presented with the highest R^2 (Fig. S2, S3, S4). Afterward, we used General Linear Models with 5% significance to associate the euglossine community attributes with coffee cover (%) in the respective scales of effect. Before that, we log10 transformed the abundance variables to fit the normality assumptions. We used the *boxcox* function of the R MASS package for the model validation (Venables and Ripley 2002). These analyses were performed in the R 4.0.2 software (R Core Team 2020).

3. RESULTS

3.1. Euglossini community structure

We sampled 1890 euglossine males belonging to four genera and 14 species (Table 1). The

relative abundance in each landscape ranged from 0.3 to 20.6%. The bee richness ranged from four to nine species (Table 1). *Eulaema nigrita* Lepeletier, 1841 was sampled in all the 15 landscapes, with the highest relative abundance (> 50%) in 13 landscapes (excepted L08 = 14.2%; and L14 = 21.4%). *Euglossa cordata* (Linnaeus, 1758) was also sampled in all 15 landscapes and was the most abundant species (68.8%) in one of them. *Eulaema atleticana* Nemésio, 2009, *Eufriesea* gr. *auriceps* sp2, *Euglossa* cf. *leucotricha* Rebêlo and Moure, 1995, and *Euglossa liopoda* Dressler, 1982 were singletons in four landscapes (Table 1). Among the 14 species, three were classified as common and 11 species as rare (Table 1; Fig. S1). The highest richness of rare species ($S=6$) occurred in two landscapes, while one of the landscapes presented just one rare species ($S=1$) (Table 1). The highest species diversity was $H'=1.47$, and the lowest was $H'=0.8$ (Table 1). The highest species equitability was $J'=0.91$, and the lowest was $J'=0.44$, while the highest species dominance was $D=0.79$, and the lowest was $D=0.42$ (Table 1).

The rarefaction curves reached asymptote in four of 15 landscapes (Fig. 2a, c). We observed a curve slope change in the other rarefaction curves, although they have not stabilized, except for three curves, which were ascending (Fig. 2b). We found a higher similarity distance from Renkonen's index in the L08 community composition regarding the other landscapes (Fig. 3). The lower distance of the community similarity was between L06 and L013, with the higher reliability and repetition with the bootstrap method (Fig. 3).

3.2. Influence of coffee cover (%) on Euglossini bees

The scale of effect of coffee cover (%) on the euglossine communities was at smaller spatial scales (between 300 and 400 m) (Fig. S2, S3, S4). We found a negative and statistically significant effect of coffee cover on species richness ($R^2=0.34$; $p=0.020$), abundance of common ($R^2=0.70$; $p<0.001$) and rare species ($R^2=0.60$; $p=0.000$) (Fig. 4a–c).

4. DISCUSSION

We described the Euglossini communities in 15 landscapes associated with coffee farming in southeastern Brazil. The observed species richness ($S=14$) was similar to that recorded in nearby areas of the semideciduous Atlantic Forest, such as southern Minas Gerais state (Silveira 2014; $S=14$) and northern Rio de Janeiro state (Aguiar et al. 2014; $S=11$ species). *Euglossa liopoda* has its first record below 20° S latitude, expanding the species' geographical distribution about 250 km south of its southernmost records in the Atlantic Forest (Neves and Viana 1997; Nemésio 2009; Medeiros et al. 2017). *Eulaema atleticana* was recorded in one out of 15 landscapes, and this species has been observed as a singleton in the region (Aguiar and Gaglianone 2012). The northern portion of Rio de Janeiro state may be the current southern limit of this bee in the Atlantic Forest (Aguiar and Gaglianone 2012).

Eulaema nigrita and *Euglossa cordata* showed the highest relative abundances among the sampled species. These species occur in different ecosystems of the Atlantic Forest (Sofia and Suzuki 2004; Aguiar and Gaglianone 2012; Medeiros et al. 2017), Amazon Forest (Moure et al. 2012; Cândido et al. 2018), Brazilian Cerrado, and Seasonally Dry Tropical Forest (Martins et al. 2018; Carneiro et al. 2018; Sousa et al. 2022). Given this environmental plasticity, these two euglossine species were found at high abundance in open and perturbed landscapes (Tonhasca et al. 2002; Aguiar and Gaglianone 2008; Silva and De Marco 2014; Aguiar et al. 2015). On the other hand, *Euglossa clausi* Nemésio and Engel (2012), sampled in seven landscapes, is an endemic species of the Atlantic Forest (Nemésio and Engel 2012). This bee has been pointed out as a potential bioindicator of conserved ecosystems (Ramalho et al. 2009; Aguiar et al. 2015). Then, some agricultural landscapes with higher forest cover may be more friendly to euglossine populations with a forest dependence (Carneiro et al. 2022).

Table 1 Composition of the Euglossini bee communities in 15 landscapes in southeastern Brazil. The species abundance in each landscape is presented as relative abundance (%). ^cCommon species, ^rRare species

	L01	L02	L03	L04	L05	L06	L07	L08	L09	L10	L11	L12	L13	L14	L15
<i>Eulaema nigrita</i> Lepeletier ^c	75.78	55.55	72.22	79.22	65.62	65.48	54.54	14.28	54.54	62.61	51.92	77.89	65.45	21.48	51.37
<i>Eulaema cingulata</i> (Fabricius) ^r	2.24	1.38	–	–	5.20	11.50	–	14.28	–	1.86	5.76	2.10	6.36	0.76	1.22
<i>Eulaema atlantica</i> Nemésio ^r	–	–	–	1.29	–	–	–	–	–	–	–	–	–	–	–
<i>Eufriesea</i> gr. <i>auriceps</i> sp1 ^r	0.44	0.69	1.58	–	1.04	–	–	–	–	–	–	–	–	–	–
<i>Eufriesea</i> gr. <i>auriceps</i> sp2 ^r	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
<i>Exaerete smaragdina</i> (Guérin-Ménéville) ^r	–	–	–	–	1.04	–	–	–	–	0.93	1.92	–	–	–	0.30
<i>Euglossa cordata</i> (Linnaeus) ^c	8.52	22.91	12.69	5.19	8.33	13.27	9.09	14.28	9.09	19.60	23.07	6.31	14.54	68.79	18.34
<i>Euglossa fimbriata</i> Moure ^c	6.72	7.63	3.17	7.79	10.41	4.42	9.09	–	9.09	4.67	5.76	1.05	10.00	5.88	19.57
<i>Euglossa securigera</i> Dressler ^r	2.69	9.02	4.76	5.19	2.08	5.30	27.27	14.28	9.09	4.67	5.76	4.21	3.63	2.55	9.17
<i>Euglossa pleosticta</i> Dressler ^r	–	–	–	–	1.04	–	–	–	–	–	1.92	–	–	0.51	–
<i>Euglossa clausi</i> Nemésio and Engel ^r	3.13	1.38	3.96	1.29	5.20	–	–	–	–	5.60	–	8.42	–	–	–
<i>Euglossa</i> cf. <i>leucotricha</i> Rebêlo and Moure ^r	–	0.69	–	–	–	–	–	–	–	–	–	–	–	–	–
<i>Euglossa liopoda</i> Dressler ^r	–	–	0.79	–	–	–	–	–	–	–	–	–	–	–	–
<i>Euglossa truncata</i> Rebêlo and Moure ^r	0.44	0.69	0.79	–	–	–	–	42.85	18.18	–	3.84	–	–	–	–
Relative abundance (%)	11.79	7.62	6.66	4.07	5.07	5.97	0.58	0.37	0.58	5.66	2.75	5.02	5.82	20.70	17.30
Richness	8	9	8	6	9	5	4	5	5	7	8	6	5	6	6
Diversity (<i>H'</i>)	0.94	1.30	1.02	0.80	1.25	1.08	1.12	1.47	1.29	1.17	1.45	0.84	1.08	0.91	1.26
Equitability (<i>J'</i>)	0.45	0.59	0.49	0.44	0.56	0.67	0.80	0.91	0.80	0.60	0.69	0.46	0.67	0.50	0.70
Dominance (<i>D</i>)	0.75	0.55	0.72	0.79	0.65	0.65	0.54	0.42	0.54	0.62	0.51	0.77	0.65	0.68	0.51

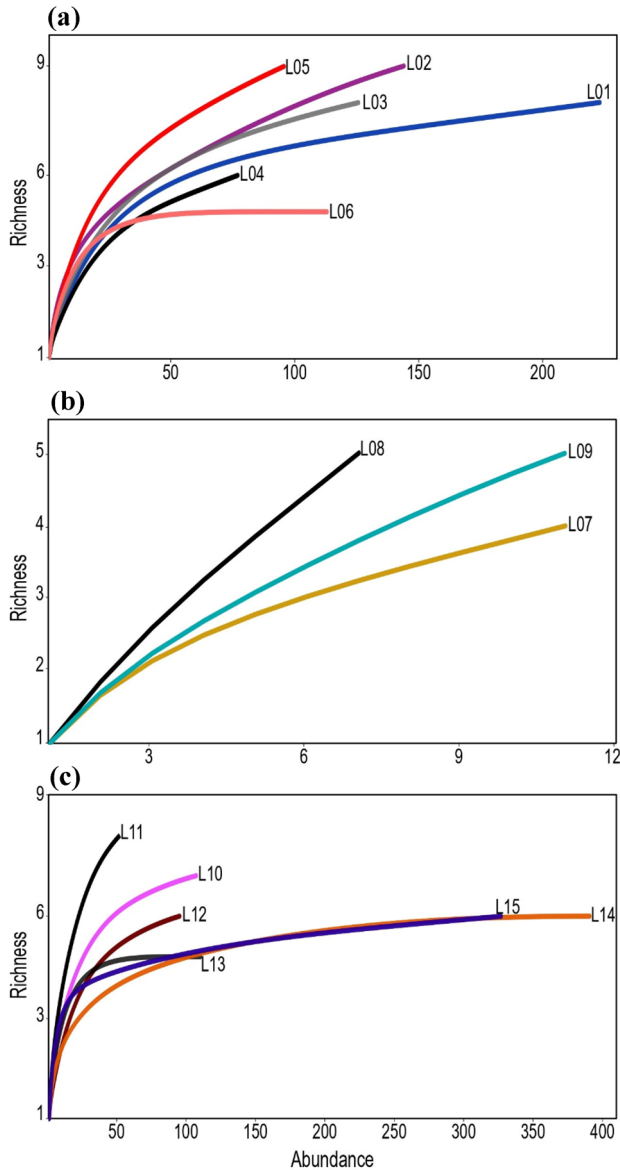


Figure 2. Accumulation curves of the Euglossini bee communities in (a) six landscapes (L01–L06) in Rio de Janeiro state, (b) three landscapes (L07–L09) in Minas Gerais state, and (c) six landscapes (L10–L15) in Espírito Santo state.

Community similarities were stable in most landscapes, except for two of them (L08 and L14). This result may be related to the landscapes' shared dominance of *E. nigrata*, but these two landscapes showed a dominance change from *E. nigrata* to *Euglossa truncata* and *E. cordata*, respectively. While the low total abundance

(L08, $N=7$) was associated with the high altitudes (> 1200 m), the proximity of a urbanized matrix (L14) seems to be an important factor. *E. cordata* has been sampled in urban areas, exhibiting an exceptional adaptation in such environments through time (Rocha-Filho et al. 2020). On the other hand, similar sites present

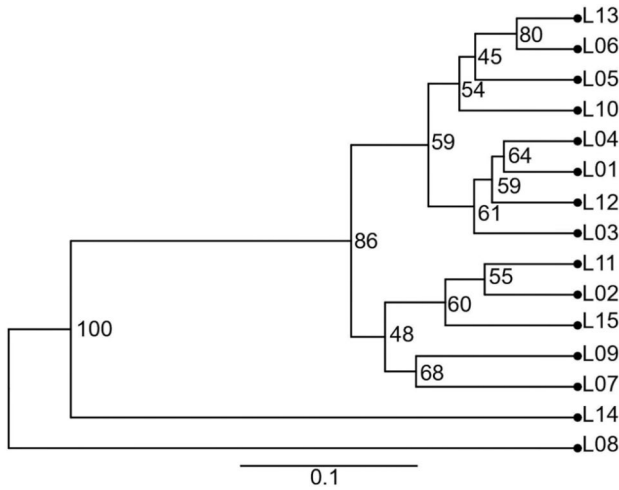


Figure 3. Dendrogram of the Renkonen's similarity index generated from Unweighted Pair Group Method with Arithmetic Mean—UPGMA. The numbers in the nodes represent repetition from 1000 randomizations by bootstrap.

the landscape attributes shaping the community composition. The higher community similarity was between landscapes spatially distant (L06 and L13, ~30 km), but they share a low forest cover (11.5% and 12.5%, respectively) which may have influenced this result, as observed for other landscape components elsewhere (Cândido et al. 2018). These two landscapes also shared the same species composition and asymptotic rarefaction curves with few individuals ($N < 50$), indicating a low euglossine richness in both

landscapes. In some scenarios, human-modified landscapes may favor euglossine species with higher environmental tolerance and wide spatial distribution (e.g., *E. cordata*, *E. nigrita*) (Cândido et al. 2018; Carneiro et al. 2021). Therefore, the landscape composition seems to shape the similarity among our sampling sites, with factors such as human land modifications driving these community patterns. In addition, within agroecosystem-dominated landscapes of Brazilian Cerrado, natural forest cover and

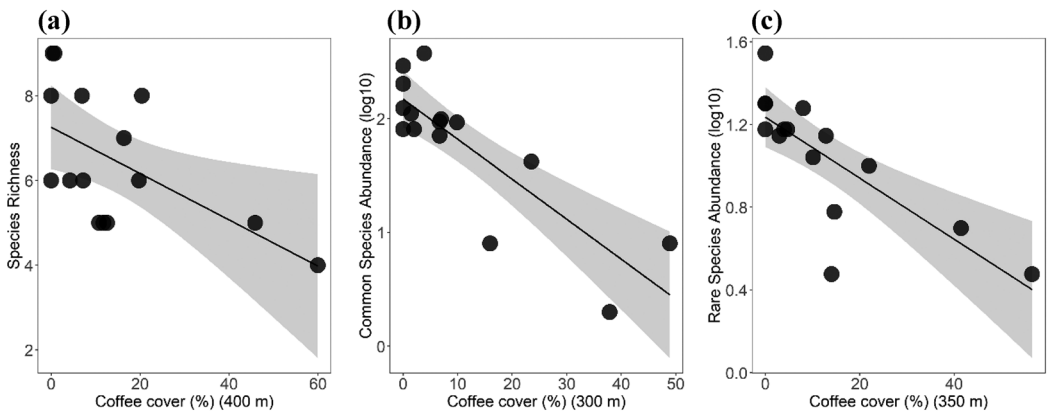


Figure 4. Influence of coffee cover (%) on species richness (a), abundance of common (b), and rare species (c) of Euglossini bee communities.

fragmentation per se were the best predictors for Euglossini species richness (Sousa et al. 2022). However, in that study, the authors did not explore the effect of the two most common anthropogenic matrices (agriculture and pasture).

5. CONCLUDING REMARKS

We confirmed the expectation that increased coffee cover (%) negatively affects euglossine richness and abundance. The coffee plants provide floral resources (i.e., nectar and pollen) for many bee species (Hipólito et al. 2018). However, these floral resources are unattractive for euglossine species (Briggs et al. 2013; Hipólito et al. 2018; Hautequestt unpubl. data). These orchid bees can explore floral resources in pioneer plants at coffee crops (Hautequestt unpubl. data), but these plants are withdrawn through high-intensity management. Moreover, some coffee plantations receive loads of agrochemicals throughout the year, such as fertilizer and herbicides. These factors may act synergistically, resulting in the negative effects of coffee cover on Euglossini bees.

The coffee cover in the spatial scales evaluated was negatively correlated with forest cover (Fig. S5). Then, the negative effects of coffee cover on Euglossini communities are linked to forest cover loss, especially in the focal forest patches. This indirectly corroborates other observations showing lower euglossine richness and abundance in smaller forest patches (Brosi 2009; Nemésio and Silveira 2010). Forest cover (%) also played an important role in maintaining species richness and abundance within modified landscapes of Brazilian Cerrado and Atlantic Forest (Sousa et al. 2022; Carneiro et al. 2022). In addition, fragmentation per se had a negative effect on these bees (Sousa et al. 2022). It is important to emphasize that the landscape heterogeneity on larger spatial scales in these agricultural areas positively influences the euglossine richness and abundance (Carneiro et al. 2021, 2022). On these spatial extensions, the negative effects of coffee cover may be diluted by a

compositional heterogeneity associated to patch types functionally important for euglossine species (Aguiar et al. 2015; Carneiro et al. 2021). Our results show that — if extensive coffee areas replace this landscape heterogeneity — the local negative effects of the coffee cover can be spatially amplified (Briggs et al. 2013). Therefore, it is essential to maintain the high spatial heterogeneity in these landscapes for euglossine species requirements. This can be achieved through agricultural practices that small coffee producers carry out on their properties, such as coffee crops interspersed by many land uses, such as agroforestry and pastures, preferentially pastures interspersed by shrub vegetation and scattered trees.

In this study, we observed rare species' occurrences and expanded the geographical distribution of a rare orchid bee species for the Atlantic Forest hotspot. Furthermore, we showed that a high coffee cover (%) drives euglossine communities to lower richness and species abundance. These coffee areas have negatively affected even superabundant species known for higher tolerance to environmental disturbances. Our findings could be extrapolated to other monoculture crops without any floral resources for Euglossini bees (e.g., Milet-Pinheiro and Schlindwein 2005), especially in systems with low environmental heterogeneity. Because the forest patches are in private areas and surrounded by agricultural environments, we suggest increasing the connectivity of these forest patches as a reasonable management measure to maintain euglossine communities' stables in such landscapes.

SUPPLEMENTARY INFORMATION

The online version contains supplementary material available at <https://doi.org/10.1007/s13592-022-00952-3>.

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AUTHOR CONTRIBUTION

LSC, MCG, and WMA designed the study, idealized the hypothesis, and wrote the manuscript; SHS contributed to the data discussion; LSC, MCR, and WFS analyzed the data; GARM confirmed the taxonomic identifications. The manuscript was revised and approved by all authors.

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DATA AVAILABILITY

Additional data is available in the supplementary material.

CODE AVAILABILITY

Not applicable.

DECLARATIONS

Ethics approval The bee sampling was authorized by Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis-IBAMA (N° 71013–3).

Consent to participate Not applicable.

Consent for publication Not applicable.

Conflict of interest Not applicable.

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