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Influence of Wild Bee Diversity on Canola Crop Yields

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Abstract

The foraging range of bees determines the spatial scale over which each species can provide pollination services. In agricultural ecosystems, productivity is related not only to the taxonomic diversity of bees per se, but also to the location of their nesting sites, which has impact on their flight range. Within this context, the present study sought to assess how wild bee assemblages affect the yield of *Brassica napus* at three different distances (25 m, 175 m and 325 m) from forest remnants in Southern Brazil. Pan traps were employed to sample bees, and data analysis was carried out using the Shannon diversity index and generalized linear models. We identified 11 species of native bees, both solitary and social, and high prevalence of the exotic species *Apis mellifera* L. Our findings show that canola crop yield was positively influenced by the diversity of bee species, demonstrating that native bees, not only *A. mellifera,* can greatly contribute to the productivity of canola crops. In addition, it was found that bee body size is significantly associated with flight distance travelled within the canola fields, and indicated a relationship with nesting sites. Thus, we hypothesize that higher canola yields are associated with increased presence of wild bee species, both social and solitary, and that maintenance of these pollinators is directly related to practices adopted in rural areas, whether within crop fields or in forest remnants used as nesting sites by wild bees.

Introduction

Pollinators play an important functional role within ecosystems by providing environmental services essential to the reproduction and survival of plants (Potts et al., 2016). Among insects, bees stand out as the major pollinators of angiosperms in the world (Ollerton et al., 2012). A full 33% of plants grown for human consumption is known to depend on bee-mediated cross-pollination (Klein et al., 2007). This beneficial relationship between bees and plants can be threatened in several ways. Studies on different crops have shown that many species and interconnected processes may collapse due to changes in adjacent habitats to crops, leading to decreased productivity and limiting yields (Vicens & Bosch, 2000).

Pollinator populations are subject to various effects of forest fragmentation, including agriculture, animal husbandry, and other anthropogenic pressures (Cresswell & Osborne, 2004). This gradual reduction of natural landscapes significantly reduces the flow of animals, pollen, and seeds (Samways, 1995). Consequently, habitat fragmentation may have negative impacts on richness and diversity of several functional groups, including pollinator insects (Kremen et al., 1993).

Assessment of insect diversity involves a variety of sampling methods designed to analyze the effects of the landscape on insect populations. Investigations on anthophilous insects have sought to ascertain their diversity in different crops and to identify potential pollinators (Westphal et al., 2008). Historically, such studies have used collection nets for sampling. However, since the last decade, the use of pan traps has increased due to their efficiency in capturing a wide range of floral visitors and the absence of collector bias (Westphal et al., 2008; Vrdoljak & Samways, 2012).

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Canola (*Brassica napus* L., var. *oleifera*), or the oilseed rape, is the third most widely grown oleaginous plant in the world. The oil extracted from its seeds is used for human consumption and as biodiesel (Marjanović-Jeromela et al., 2008). In Brazil, approximately 47,500 hectares of land, predominantly located in the South, are dedicated to the cultivation of canola (Conab, 2017). The massive flowering of canola is highly attractive to bees, as it provides abundant floral resources (nectar and pollen) (Holzschuh et al., 2016). Studies have shown that insect visitation improves the yield of grain crops, of which *Apis mellifera* L. is considered the main pollinator (Abrol, 2007; Rosa et al., 2010; Bommarco et al., 2012; Jauker et al., 2012; Halinski et al., 2015; Witter et al., 2015). In Canada, where canola originates, Sabbahi et al. (2005) introduced three *A. mellifera* hives per hectare in a canola field and found a 46% increase in seed yield. Studies have shown that the presence of bees in this crop can increase grain yield by up to 47% (Becker et al., 1992; Bommarco et al., 2012; Blochtein et al., 2014; Witter et al., 2015). However, despite the well-established importance of *A. mellifera* to canola yields, wild bees may also contribute significantly to a more efficient pollination of this crop (Garibaldi et al., 2013; Koh et al., 2016; Potts et al., 2016). The presence and diversity of these insects increases pollination and, consequently, the yield and market value of the crops (Sabbahi et al., 2005; Bommarco et al., 2012).

Considering that pollinators predominantly inhabit forest fragments adjacent to crop fields, and that bee body size is a predictor of their foraging range, they are expected to act on pollination at different scales (Greenleaf et al., 2007; Bommarco et al., 2012; Bailey et al., 2014; Wright et al., 2015). Therefore, investigations on potential pollinators of commercially important crops and their nesting habits are necessary not only for the development of strategies to support conservation and management of these species, but also to enable their more efficient use in crop pollination.

The present study was designed to evaluate whether the diversity and body size of bee species have influence on the productivity of canola fields located at varying distances from forest remnants in Southern Brazil. Therefore, we sought to answer the following questions: (1) Do the bee species present in flowering canola fields nest in nearby forest remnants? (2) Is bee body size related to distances between crop fields and adjacent forest remnants? (3) Does bee diversity increase canola crop yields?

Material and Methods

Study area

The study was conducted in four areas near and within *B. napus* fields (Hyola 420 cultivar), in the municipality of Esmeralda, Southern Brazil. The region is characterized by pasture land, forest fragments, and fields of annual crops (canola, soybean, maize, and wheat). The four investigated canola fields were 20 ha (field 1, Fig 1), 80 ha (2), 100 ha (3), and 80 ha (4) in size, located 2.5 to 23.5 km from each other. These fields are in the upper plateau of Serra do Nordeste region, with average annual temperatures of 14.4-16.8 °C, relative humidity of 76-83%, annual precipitation of 1,412-2,162 mm, and altitude of 944 m (Veloso et al., 1992). According to the Köppen classification, the region is considered to have Cfa – humid subtropical climate (Alvares et al., 2013), and its original vegetation is composed of Mixed Ombrophilous Forest and Grassland. There are neither apiaries nor meliponaries in the municipality.

Bee sampling

Bees were sampled with blue, yellow, and white pan traps, which remained exposed for 24 h per sampling (adapted from Westphal et al., 2008). These traps were laid out in plots, and consisted of five groups of three pots, keeping a distance of 15 m between groups and 3 m between pots, forming an equilateral triangle (adapted from FAO, 2010; Fig 1). The traps were placed within canola fields at three distances from the forest remnants: 25 m, 175 m, and 325 m, during flowering season (August-October), for a total of four samplings in 2010 (field 1 and 2) and seven in 2011 (fields 1 to 4).

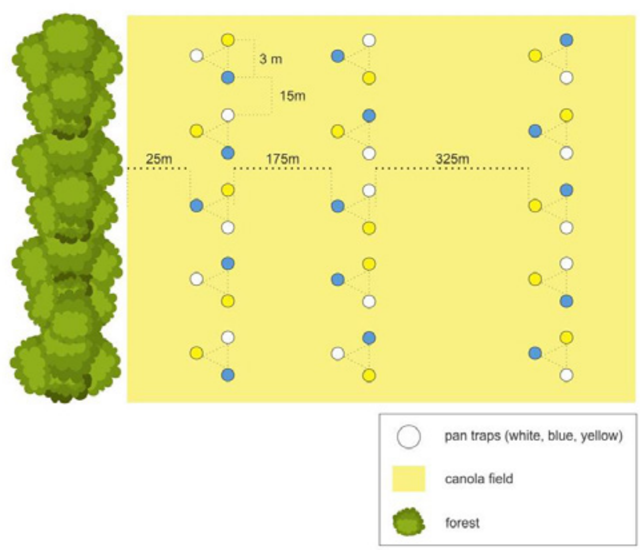

Fig 1. Schematic representation of pan traps'arrangement within the canola fields for bee sampling.

The bees were identified and deposited at the Bee Collection of the Museum of Science and Technology, at Pontifícia Universidade Católica do Rio Grande do Sul. After identifying the different species, we conducted a literature review to obtain relevant data on the characteristic nesting substrates and foraging range of each species (see Supplementary Material 1).

Bee size

To test for correlation between distance flown from the forest remnants and bee body size, we measured the intertegular distance, or span, of the collected specimens.

This morphological measurement is a robust estimator of bee body mass, size (Bullock, 1999), and flight range (Greenleaf et al., 2007; Wright et al., 2015). Measurements of intertegular distance were obtained using digital calliper only in female specimens due their role in brood care, therefore, return to their nests with provisions.

Productivity of canola crops

To assess the productivity of canola crops, once siliques had matured, 20 plants were harvested and spaced 1.5 m apart, forming 225 m^2 plots in each field, totaling three harvest plots per field. These plants were used to analyze seed production under free visitation of insects at three different distances from forest remnants: 25, 175, and 325 m. The number of seeds per plant was counted manually with the aid of a vacuum seed counter (ERICKSEN De Leo), and seed weight measured on an analytical balance (AUY200). The mean seed weight per plant was obtained from the sum of the individual weight of each plant divided by the total number of plants.

Statistical analysis

To assess species abundance and richness, we used the Shannon diversity index, which responds more sensitively to changes in the importance of rarer species (Peet, 1974). The *diversity* function of the "vegan" package in R was used for this purpose (Oksanen et al., 2016). We then constructed a Gaussian family (link = log) generalized linear model (GLM) to assess whether mean canola grain weight (yield) could be explained by bee diversity. GLM fit was assessed by the pseudo-R^2 statistic, using the *rsquared* function in R package "piecewiseSEM" (Lefcheck, 2015).

The influence of bee size (intertegular distance) on the distance from the forest remnants (25, 175, 325 m) was assessed using a generalized linear mixed model (GLMM) with negative binomial family estimated by maximum likelihood (Laplace approximation). This error distribution family was selected after checking for overdispersion with a Poisson-distributed GLMM. In addition to the distance variable, nesting sites (Supplementary Material 1) and interaction between species and nests were used as predictor variables (fixed effects) for the GLMM. Fields and pan trap colors were regarded as random effects. Analysis was conducted using function *glmer.nb* from package "lme4" (Bates et al., 2015). Finally, bee species collected in each of the four canola fields, and stratified by social vs. solitary, were plotted as a function of distance from the corresponding forest remnants. All analyses were carried out in R (R Core Team, 2018).

We computed the accumulation curve of species and its respective interpolation/extrapolation of Hill number based on sample size and abundance of individuals (bees per distance) which values of diversity (q=0) were estimated from Chao1 (Chao et al., 2014) after 2,000 replications. For this, we used the individual-based abundance data using the function *iNEXT* from package of same name (Hsieh et al., 2016). We also calculated the extrapolated species richness in our data matrix in order to estimate the number of unobserved species. We used the function *specpool* from "vegan". The extrapolated richness was calculated with chao equation, that weights the number of individuals observed in a single sampling.

We also performed a diversity profile according to diversity order from Hill number (Hill, 1973). Such analysis was developed to widely evaluate bee diversity on canola fields. These values determine the sensibility of relative abundance of species by sites (Chao et al., 2014), here assumed as distances from forest fragments. Thus, we employed the bee community data matrix to generate the diversity profile using the function *renyi* (hill=TRUE) from package "vegan"(Oksanen et al., 2016).

The similarity percentages, *i.e.* the pairwise comparisons of groups of sampling units and the average contributions of each bee species to the average overall Bray-Curtis dissimilarity, was estimated after 2,000 replications using the function *simper* from "vegan". This function displays the most important species for each pair of groups (distances from forest fragments). As a result, these species must contribute at least to 70% of differences between groups.

We also used bee community data to carry out a non-metric multidimensional scaling (NMDS). This ordination was employed to observe the dissimilarity on bee composition related to distance from forest fragments (25 m, 175 m, and 375 m). However, before performing this analysis, we standardized our community data using the function *decostand* (method = "total"). Consequently, we applied the dissimilarity index of Bray–Curtis (proper to abundance data) using the function *vegdist* to implement NMDS. Thus, we used two functions from "vegan" (*metaMDS* and *stressplot*) to fit NMDS and to find goodness of fit measure for points in NMDS. Stress values lesser than 0.2 are considered adequate.

Finally, we used our dissimilarity object provided by the function *vegdist* to fit a permanova type II in function *adonis.II* from package "RVAideMemoire" (Hervé, 2015). Subsequently, we performed pairwise comparisons between group levels (distance from fragments) using the function *pairwise.perm.manova* from the same package with 2,000 permutations and statistic test by Pillai method and p-value adjustment after "fdr".

Results

Bee diversity within canola fields

A total of 314 bees were collected from the four sampling areas, representing 11 native and one exotic species (*A. mellifera).* There are no managed colonies of any species of bee in the city, thus all collected bees were feral. The exotic species *A. mellifera* was the most abundant, accounting for 58% of all collected specimens (Supplementary Material 1).

Despite the apparent predominant role of *A. mellifera* bees in the canola fields, native bee species also warrant attention, whether because some of them were also present in all distances or because their behavior makes them as efficient as *A. mellifera* for pollination of this crop. In this regard, one native species stood out with a result very similar to *A. mellifera*: *Trigona spinipes* (Fabricius), which was collected in all distances except for 325 m in field 1 and 25 m in field 3 (Fig 2). This is an interesting finding because, *T. spinipes* is smaller than *A. mellifera* (intertegular distance 1.79 ± 0.11 mm vs. 3.15 ± 0.03 mm).

We have found at least 12 bee species visiting canola fields in Southern Brazil. However, as chao estimator suggests, if more bees are sampled the probability of finding more bee species is higher (chao = 21 ± 9) (Fig 3). As data indicate, it is more likely to find more bee species nearer (25 m) than far (175 m, 375 m) from forest fragments. It demonstrates the positive impact of having such ecosystem, as they provide cavities suitable for nesting and floral resources for potential pollinators.

Diversity profile of Hill number indicates that diversity of bee species (richness, q = 0) near forest fragments (25 m) is higher than at larger distances (175 m, 375 m). As such, our results suggest that canola fields may largely benefit from bee species provided by natural vegetation around crops. On the other hand, if we observe the Shannon-Weiver index (q = 1) it is possible that both distances (175 m, 375 m) are similar, while at 25 m this diversity index remains higher than at longer distances from forest fragments (Fig 4).

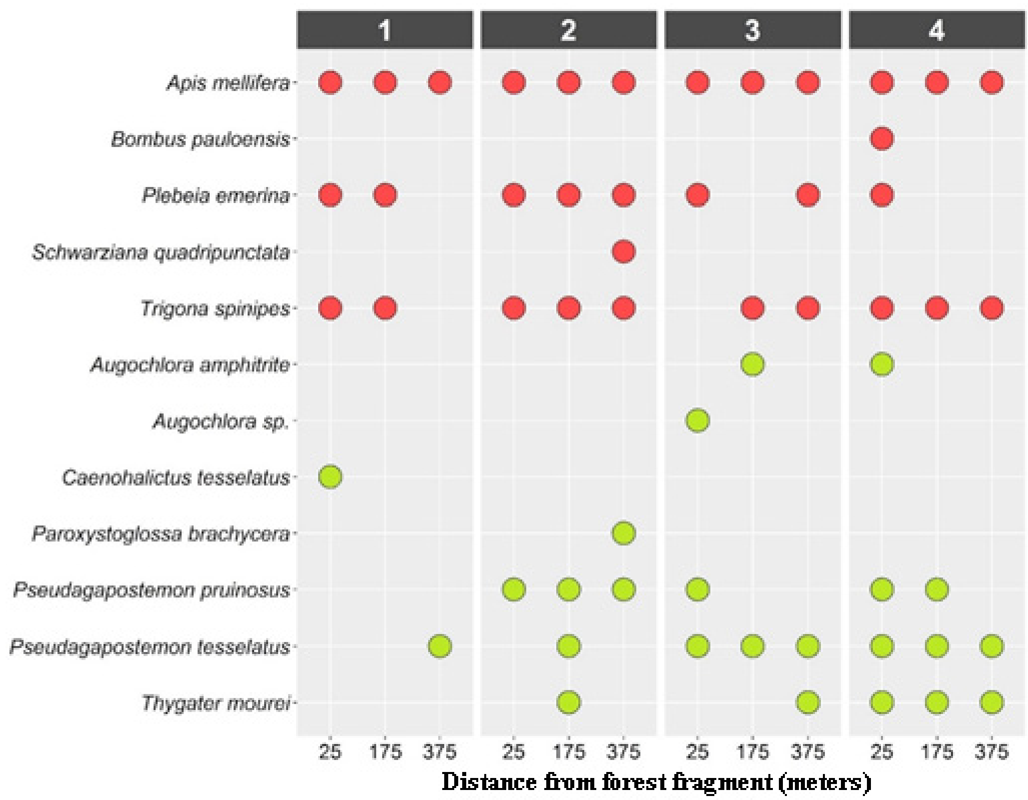

Fig 2. Location of bees collected within four canola fields at three distances from forest remnants (25 m, 175 m, 325 m). Red circles represent social species, while green circles represent solitary species.

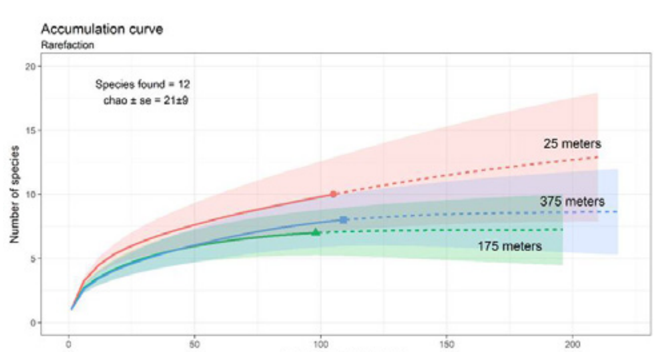

Fig 3. Accumulation curve of species: number of species found versus number of expected species. Bee diversity on canola fields in Southern Brazil. Interpolation (solid lines) and extrapolation (dashed lines) of Hill number with order q. Note: *shadow* means confidence intervals (95%).

The similarity percentages suggest that species of social bees (*Plebeia emerina* (Friese), *A. mellifera*, *T. spinipes*) greatly contribute for discriminating between different set of distances from forest fragments (Table 1). Thus, even though the smallest social bee (*P. emerina*) contributes to discriminate the nearest distance (25 meters), other larger social bees are relevant at longer distances, as *A. mellifera* and *T. spinipes*. On the other hand, solitary bees are also important to canola crops. For example, *Pseudagapostemon tesselatus* Cure appears as a relevant species aiding to discriminate all sets of distances (Table 1). These results strongly suggest that canola crops may benefit from bees provided by wild vegetation and bee species inhabiting the soil within fields.

Table 1. Cumulative contributions of most influential species to the differentiate set of distances from forest fragments around canola (*Brassica napus*) fields in Southern Brazil.

25 - 175 meters	25 - 375 meters	175 - 375 meters
Plebeia emerina	*Apis mellifera*	*Apis mellifera*
0.23	0.26	0.34
Pseudagapostemon tesselatus	*Plebeia emerina*	*Pseudagapostemon tesselatus*
0.45	0.48	0.65
Apis mellifera	*Pseudagapostemon tesselatus*	*Trigona spinipes*
0.65	0.69	0.79
Trigona spinipes	*Trigona spinipes*	
0.80	0.80	

The NMDS ordination (stress = 0.13) indicates that bee composition nearer to forest fragments is significantly different than at larger distances (Fig 5, Table 2). We suggest that vegetation provides substrates as tree cavities to support social bees like *P. emerina* and *A. mellifera*, and aerial nesting areas for *T. spinipes*. Further, data show that wider distances from wild vegetation statistically modifies the diversity of bees, suggesting that either larger bees with higher foraging range may visit the canola crops or solitary bees that nest on soil are also visiting canola flowers in Southern Brazil.

Table 2. Cumulative contributions of bee community to different sets of distances from forest fragments around canola (*Brassica napus*) fields in Southern Brazil.

Permanova type II tests Response: bee community data					
	Sum Sq	**Mean Sq**	**d.f.**	**F**	***p***
Distances	0.92	0.92	1	2.57	0.04
Residuals	0.46	0.03	13		
	0.55		14		
Pairwise comparisons between distances from forest fragments (p-values)					
	25	**175**			
175	0.02	-			
375	0.04	0.59			

Canola Yield

Regarding bee diversity and canola grain yield, we found a Shannon diversity index ranging from 0.78 in field 1 to 1.61 in field 4 (Fig 6). Our results demonstrate that bee diversity measured by the Shannon index significantly affected canola yield (Gaussian GLM, F = 615, $p < 0.001$, pseudo-$R^2 = 0.99$) (Fig 6).

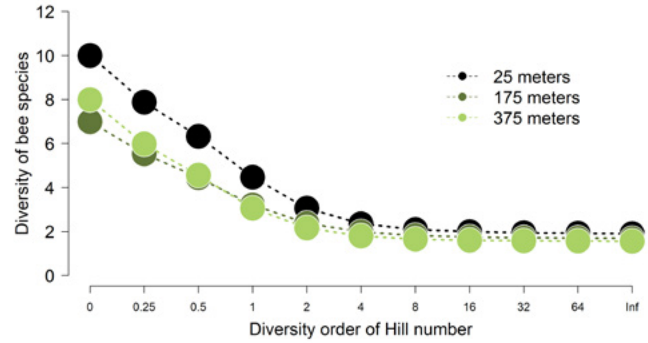

Fig 4. Diversity profile of Hill number: bee diversity based on three distances from forest fragments on canola (*Brassica napus*) crops in Southern Brazil. For moving x-axis to left the rare species become more important, while to opposite side (right) there is more weight to equability of bee species. Some diversity indexes can be extracted from x-axis as: a) 0 = species richness; b) Shannon index; c) Simpson index; inf = Berger-Parker index.

Bee body size and foraging range (distance from forest remnants)

Regarding body size, the largest forager sampled in this study was *Bombus pauloensis* Friese (intertegular distance: 4.87 mm – Fig 7). However, only one individual was collected 25 m from a forest fragment, possibly because the canola crop is not so attractive to this species. The stingless bees *T. spinipes*, *Schwarziana quadripunctata* Lepeletier, and *P. emerina*, all of which have smaller intertegular spans, were found in nearly all sampled distances, except *S. quadripunctata*, which was found only at 325 m from forest remnants. It is worth noting that *P. emerina* is 2.6 times smaller than *A. mellifera* (intertegular distance: 1.19 mm and 3.15 mm, respectively), and that *T. spinipes* (1.79 mm) is 1.76 times smaller than *A. mellifera*. *Thygater mourei* Urban was collected at all distances in field 4, 175 m in field 2, and 325 m in field 3. *Caenohalictus essellates* (Moure) was the only species found solely at 25 m from the sampled forest fragment (Fig 2).

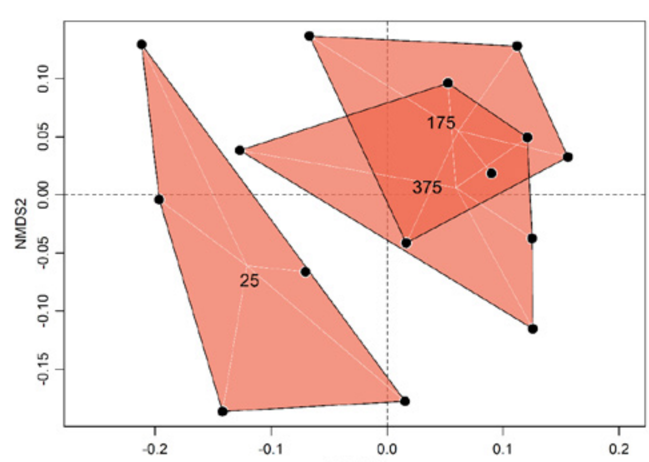

Fig 5. Non-metric multidimensional scaling: bee composition visiting canola crops in Southern Brazil. Note: *numbers inside polygons* indicate the set of distances from wild vegetation; *points* means sample units; *White lines* are spider bodies calculated as centroids (averages) of polygons.

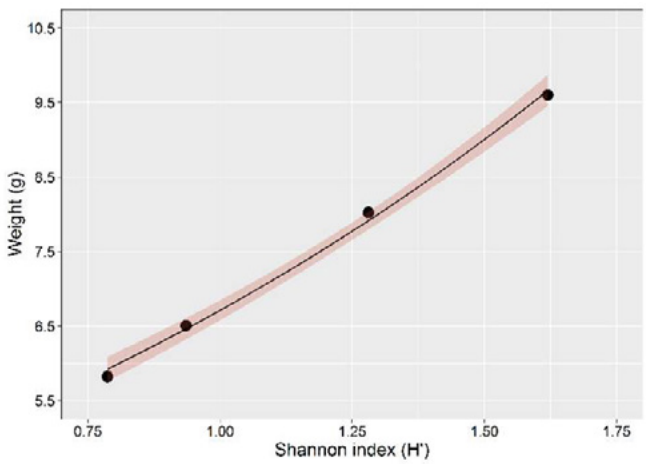

Fig 6. Relationship between canola grain yield (seed weight) and bee diversity (Shannon index, H'). Note: Points denote observed values; line denotes the predicted model (Gaussian family GLM); pink-shaded area denotes the 95% confidence interval. Model fit: pseudo-$R^2 = 0.99$.

We demonstrated a significant difference in body size among bees sampled at each of the three distances from the forest edge (negative binomial GLMM, $\chi^2 = 20.07$, $p < 0.001$, Fig 7). Furthermore, we found significant differences between the nesting sites of the collected species (negative binomial GLMM, $\chi^2 = 2.25$, $p < 0.001$), and in the interaction of individual size and nesting substrates of the sampled species (negative binomial GLMM, $\chi^2 = 6.25$, $p = 0.012$).

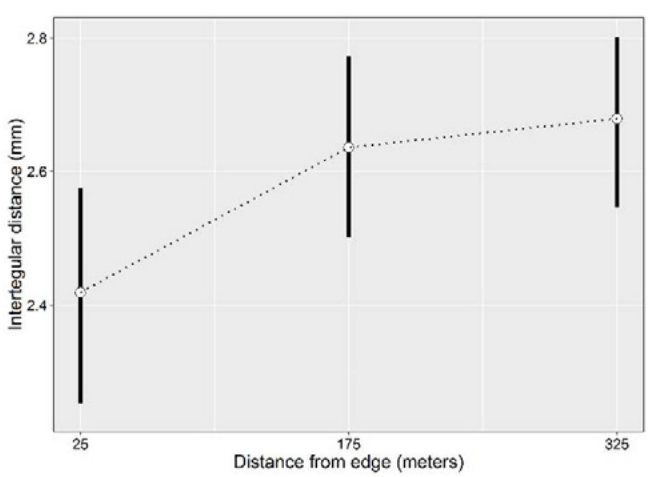

Fig 7. Intertegular distance of bees in relation to distance from the sampled forest remnant (25 m, 175 m, 325 m). Points denote the mean; error bars denote the 95% confidence interval. All measures of dispersion estimated after 999 replications (bootstrap).

Discussion

Bee diversity

The predominance of *A. mellifera* may be related to several factors, including the presence of wild nests of this species in the region, the large population of individuals in nests, the effective recruitment behavior and the species plasticity regarding nesting substrates (Beekman & Ratnieks, 2000). Furthermore, food scarcity is common in the region during winter, making massive canola blooms attractive for bee species. In addition to this predominance, *A. mellifera* was the only species found in all sampled distances. This finding was expected, as similar studies suggested that this species can fly distances greater than 10 km (Beekman & Ratnieks, 2000), which far exceeds the distance assessed in our experiment (< 500 m).

Although the estimated flight range of *T. spinipes* (800 m; Araújo et al., 2004) is much shorter than that expected for *A. mellifera* (10 km; Beekman & Ratnieks, 2000), the high population density of their nests (180,000 individuals; Jaffé et al., 2014) greater than that of *A. mellifera* and their efficient recruitment habits accomplished through chemical signaling, *i.e.*, odor-trail recruitment communication (Nieh et al., 2004), may have contributed to the presence of this species in practically all distances. Therefore, the native bee species *T. spinipes* stands out as a promising alternative to *A. mellifera* to improve cross-pollination in canola crops, particularly because their individuals remain longer on canola flowers when compared to *A. mellifera* (D'Ávila & Marchini, 2005).

The yield results found highlight the importance of native bee species in addition to *A. mellifera* in canola fields in Southern Brazil. Furthermore, pollination by bees in agricultural areas increases not only the quantity but also the quality of grain (Ricketts et al., 2008) and, consequently, the biofuel productivity (Durán et al., 2010). Therefore, taxonomic identification of species and better understanding of their nesting habits provide inputs for preservation of the diversity of bee populations in canola-growing areas and to the strengthening of sustainable farming models.

Another species of native social bee that stood out in this study was *P. emerina*. This species was collected in all fields, except at 325 m in field 1 and 175 m and 325 m in field 4. Witter et al. (2015) showed that this species is also an efficient pollinator of canola, contributing to grain yield with a 75% rate of fruit set; there was no difference in pollination efficiency between visits of *P. emerina* and *A. mellifera*. Accordingly, other authors have pointed out the efficiency of native bees as pollinators of this crop (Garibaldi et al., 2013; Potts et al., 2016).

On the other hand, regarding the occurrence of solitary bee species at different sampled distances, a notable finding was the genus *Pseudagapostemon*, with species *P. tesselatus* and *Pseudagapostemon pruinosus* Moure and Sakagami. Solitary bees, especially Halictidae, were found mainly at the 175 m and 325 m distances. It bears noting that these bees nest in soil (Supplementary Material 1), including within areas in which annual crops are grown (Eickwort & Sakagami, 1979; Dalmazzo & Roig-Alsina, 2012; R. H., personal observation), as observed in the canola fields by the authors during this study. Given this, we consider that these bees may contribute to the pollination of this crop, irrespective of distance from forest remnants.

Supplemental contributions to pollination and importance of canola for bees

Studies have shown a 12 to 47% increase in canola crop yield secondary to cross-pollination with *A. mellifera* and other native bee species (Rosa et al., 2010; Bommarco et al., 2012; Garibaldi et al., 2013; Blochtein et al., 2014; Witter et al., 2015; Koh et al., 2016; Potts et al., 2016). Forest fragments and other semi-natural areas play a relevant role in wild bees nesting by facilitating their permanence in these sites. No-till farming, widely adopted in the investigated area for annual crops (including canola), may facilitate nesting of the solitary bee species recorded in this study (Morandin et al., 2007). Therefore, solitary and social bees play complementary roles in canola fields in Southern Brazil, influencing pollination processes and increasing grain yield.

Another important factor that contributed to bee species abundance and richness in canola fields was that this crop blossoms in the winter, a period of food scarcity, and its massive flowering provides copious and readily available pollen and nectar. Considering that bees need to visit a large number of flowers to meet their individual needs, those of the brood and those of the colony (Corbett et al., 1991), their significant presence in this crop is a good predictor of higher canola yields.

Foraging range of bees

The intertegular distance proved to be an efficient estimator of foraging range based on mathematical equations, providing additional knowledge on their position regarding this scale and corroborating the findings of Araújo et al. (2004).

Analysis showed that individual bees with larger intertegular span reached longer foraging distances than smaller bees (except *B. pauloensis*). These individuals tend to forage from plants closer to their nests, especially when there is an abundance of floral resources as in canola fields (Morandin et al., 2007). In stingless bees, body size may act as a limiting factor for maximum flight range, making many species feed near their nests (Araújo et al., 2004). However, although *P. emerina* has a small intertegular span, its specimens were found at all distances, probably as they are able to fly farther than 325 m (Araújo et al., 2004).

Data on the foraging range of bees provided important information to help us understand the scale at which bee populations respond to the landscape (Greenleaf et al., 2007), as well as to determine the spatial scale in which each bee can provide pollination services (Kremen, 2005). Changes such as specialization of foraging behavior to search for specific floral resources, spatial orientation methods, location and abundance of food resources as well as the availability of nesting sites, may influence the spacial scale occupied by bees (Araújo et al., 2004).

In some regions of Southern Brazil, beekeepers take honeybee hives into canola fields, exploiting its abundant pollen resources, and stimulating the growth of their bee colonies during periods of food scarcity (winter) by improving the number of individuals and keeping a “strong colony” for spring. This period is fundamental for multiplication of hives and for introducing pollination in different crops (Blochtein et al., 2014). Since a few years, these benefits for beekeepers allied to the increased quality and quantity of crop production led farmers and beekeepers to start a promising partnership in Brazil. Furthermore, in the future, beekeepers of native bees (meliponicultors) can contribute substantially to the improvement of canola yields (Garibaldi et al., 2013).

We have demonstrated that, in Southern Brazil, the diversity of bee assemblages has a positive influence on canola productivity. We also obtained evidence of the importance of forest remnants and canola fields as nesting sites for bees. In both landscapes, the practices adopted by farmers are fundamental to ensure the protection of bees’ nests, whether underground or arboreal. Therefore, the preservation of wild bees – social and solitary – can greatly contribute to the productivity of canola fields depending directly on the practices adopted in rural areas. Such preservation will reflect on the yields of this crop and protect the biodiversity of pollinators in the vicinity of canola fields.

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Supplementary Material

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Authors’ Contribution

R Halinski conceived the study, wrote the manuscript and analyzed the data; TG Kaehler performed analyses of morphological measurements; CF dos Santos conducted data analysis; and B Blochtein reviewed the presented research process and findings.

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