

FERNANDO MENDES BARBOSA

**BEEES INCREASE TOMATO CROP PRODUCTION AND ARE IMPACTED
BY BOTH LANDSCAPE CONTEXT AND FARM MANAGEMENT**

Tese apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Entomologia, para obtenção do título de *Doctor Scientiae*.

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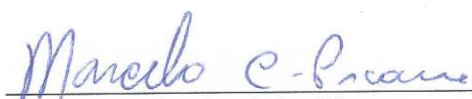
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Derly José Henriques da Silva



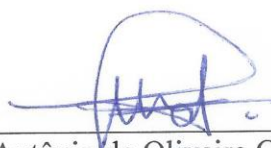
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“E Deus viu tudo o que havia feito, e tudo havia ficado muito bom...”

(Gn. 01:31 - NVI)

“Pois desde a criação do mundo os atributos invisíveis de Deus, seu eterno poder e sua natureza divina, têm sido vistos claramente, sendo compreendidos por meio das coisas criadas...”

(Rm. 01:20 - NVI)

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ABSTRACT

Barbosa, Fernando Mendes, D.Sc., Universidade Federal de Viçosa, July, 2018. **Bees increase tomato crop production and are impacted by both landscape context and farm management.** Adviser: Lucio Antônio de Oliveira Campos. Co-adviser: Weyder Cristiano Santana.

Most agricultural crops depend on animal-mediated pollination to some degree. Among insects, bees are the main pollinators, increasing yield and improving the quality of the agricultural production. On the other hand, the changes resulting from the intensification of the agriculture are pointed out as the main causes of the loss of diversity in pollinator communities, which may compromise the stability of food production. Given this, understanding how bees affect agricultural production and how changes resulting from intensification of agriculture impact the community of these pollinators is important to outline strategies that promote the increase and stability of the pollination services in agroecosystems. Thus, initially, I identified bees visiting the tomato flowers and the main characteristics of the identified species; I verified the effect of climatic variables and the period of day in which bees' activity took place in the tomato flower; and I evaluated fruit production under different types of pollination. Subsequently, I evaluated how the landscape configuration (landscape scale) and farm management (local scale) affect the diversity of bees in small tomato crops in the region of Zona Mata, Minas Gerais state, Brazil. Finally, I assessed the effect of the irrigation method on pollination and pollinators in a tomato crop. Overall, bees increase tomato production and respond negatively to loss of natural habitat and increased use of agrochemicals. In this study, an important step was taken in understanding how bees affect tomato production and how the community of these pollinators responds to changes resulting from the intensification of agriculture. Considering the current discussion involving expansion of agricultural activity and biodiversity conservation, the results of this study are particularly relevant for providing important data that can guide pollinator management plans.

RESUMO

Barbosa, Fernando Mendes, D.Sc., Universidade Federal de Viçosa, julho de 2018. **Abelhas aumentam a produção de tomate e são impactadas pelo contexto da paisagem e manejo da propriedade.** Orientador: Lucio Antônio de Oliveira Campos. Coorientador: Weyder Cristiano Santana.

A maioria dos cultivos agrícolas depende da polinização mediada por animais em algum grau. Entre os insetos, as abelhas são os principais polinizadores, aumentando a produtividade e melhorando a qualidade da produção agrícola. Por outro lado, as alterações decorrentes da intensificação da agricultura são apontadas como as principais causas da perda de diversidade nas comunidades de polinizadores, podendo comprometer a estabilidade da produção de alimentos. Desta forma, compreender como as abelhas afetam a produção agrícola e como as alterações decorrentes da intensificação da agricultura impactam a comunidade destes polinizadores é importante para delinear estratégias que promovam o aumento e estabilidade dos serviços de polinização em agroecossistemas. Assim, inicialmente, identifiquei abelhas visitando as flores do tomate e as principais características das espécies identificadas; verifiquei o efeito de variáveis climáticas e período do dia sobre a atividade das abelhas na flor do tomate; e avaliei a produção de frutos sob diferentes tipos de polinização. Em seguida, avaliei como a configuração da paisagem (escala de paisagem) e manejo da propriedade (escala local) afetam a diversidade de abelhas em pequenos cultivos de tomate na região da Zona Mata, estado de Minas Gerais, Brasil. Por último, verifiquei o efeito do método de irrigação sobre a polinização e polinizadores em um cultivo de tomate. De modo geral, as abelhas aumentam a produção de tomate e respondem negativamente à perda de hábitat natural e aumento do uso de agroquímicos. Neste estudo, um importante passo foi dado na compreensão de como as abelhas afetam a produção de tomate e como a comunidade deste polinizadores respondem às alterações decorrentes da intensificação da agricultura. Considerando a atual discussão envolvendo expansão da atividade agrícola e preservação da biodiversidade, os resultados deste estudo são particularmente relevantes por fornecer dados importantes que podem guiar planos de manejo de polinizadores.

GENERAL INTRODUCTION

Pollination is the transfer of pollen between the anthers and stigma of flowers that enables fertilization and reproduction (Potts et al., 2016). Although some crops with high production volume (rice, maize, and wheat) do not depend on biotic pollination, more than 75% of leading global foods depend on animal-mediated pollination to some degree (Klein et al., 2007). It is known that insects are important agents of biotic pollination (Ollerton et al., 2011) and that the bees represent the most important and specialized group of pollinators (Danforth et al., 2013) with more than 20,000 species described (Ascher and Pickering, 2018). Few species of bees including honey bees, stingless bees, bumble bees and some solitary species are managed, and most pollination services are conducted by wild bees. This group includes efficient pollinators that increase not only crop yield (Garibaldi et al., 2013), but the quality, shelf life, and commercial value of food (Klatt et al., 2013; Brittain et al., 2014). Thus, the pollination services of bees can be a limiting factor, introducing a yield gap in crops (Eeraerts et al., 2017).

In agroecosystems, short-term pollination can be provided by a few species that tend to persist in these environments due to species-specific traits (Ferreira et al., 2015; Martins et al., 2015). However, many studies have demonstrated that more diverse pollinator assemblages provide efficient and more stable pollination services than a single or few pollinator species (Hoehn et al., 2008; Garibaldi et al., 2013; Blitzer et al., 2016; Eeraerts et al., 2017). This efficiency and stability can be attributed to interspecific interactions that alter the behavior of pollinators, and functional complementarity takes place through niche partitioning among pollinators (Brittain et al., 2013; Blitzer et al., 2016).

In recent years, the decline of bee populations in many regions of the world has been reported (Potts et al., 2010; Burkle et al., 2013). This decline is often linked

to one or a combination of factors including land-use change and intensification, indiscriminate use of pesticides, climate change, proliferation of pests and pathogens, and introduction of exotic species (Potts et al., 2010; Goulson et al., 2015; Eeraerts et al., 2017). Most of these factors are related to the intensification of agriculture, characterized by the expansion of monocultures, the intensive use of off-farm inputs (fertilizers, pesticides, machinery), and the use of genetically modified crops (Kennedy et al., 2013; Deguines et al., 2014; Cusser et al., 2016). These changes cause isolation of natural habitats' populations and reduce food sources and nesting sites (Ferreira et al., 2013), negatively impacting pollinator communities (Connelly et al., 2015; Mogren et al., 2016; Zou et al., 2017). In addition, increasing the distance between agricultural areas and natural patches tends to decrease the diversity of pollinators and the level of crop pollination (Carvalho et al., 2010; Garibaldi et al., 2011; Geslin et al., 2016). On the other hand, the indiscriminate use of pesticides (insecticides, fungicides, and herbicides) can negatively affect pollinator populations by direct and indirect exposure, reducing pollination efficiency (van der Sluijs et al., 2013; Feltham et al., 2014; Park et al., 2015).

The response of bee communities in local (farm management) and regional scales (landscape context) varies according to species-specific traits (Tangtorwongsakul et al., 2017). Overall, wild bees and managed bees, especially honey bees, tend to present opposite responses regarding land use changes and intensification of farm management (Petersen and Nault, 2014; Eeraerts et al., 2017). The managed bee *Apis mellifera* is more opportunistic and generalist and thus appears to be less dependent on natural habitats than wild bees (Saturni et al., 2016). There are reports of a positive relationship between *A. mellifera* abundance and agriculture intensification (Marini et al., 2012; Petersen and Nault, 2014; Eeraerts et al., 2017). Due to their habits, such generalist species can survive in modified environments due

to foraging ability in almost all types of flowers (Ferreira et al., 2015). In addition, the introduction of hives or the existence of apiaries near the crops can increase the honey bees' abundance in these areas (Martins et al., 2015; Gibbs et al., 2016). On the other hand, agricultural landscapes may also favor some wild bee groups, mainly those of underground nesting that rely on bare soil to build their nests (Potts et al., 2005; Ferreira et al., 2015).

Despite potential conflicts of interest, biodiversity conservation and agricultural expansion may be concordant in farms where crops are managed properly (Tschardt et al., 2012). Both can be harmonized with the objective of maximizing agricultural yield and promoting ecosystem services preservation (Cusser et al., 2016). For example, small farms have shown to support a greater diversity of pollinators when compared to large farms, possibly due to the existence of small natural and semi-natural habitats between cultivated areas and crop diversity (Garibaldi et al., 2016; McKechnie et al., 2017; Zou et al., 2017). On the other hand, in areas with intensive agriculture, biodiversity loss can be minimized by adopting practices that maintain and restore food sources and nesting sites around crops (Bátary et al., 2011; Kovács-Hostyánszki et al., 2017). These practices include sown field margins, flower strips, hedgerows, and fallow fields (Korpela et al., 2013; Blaauw and Isaacs, 2014; Saunders et al., 2014; Kremen and M'Gonigle, 2015; M'Gonigle et al., 2015; Scheper et al., 2015; Williams et al., 2015; McKechnie et al., 2017).

With a worldwide harvest of over 177 million tons in 2016, tomato (*Solanum lycopersicum* L.) is the second most important vegetable crop in the world next to potato (FAOSTAT, 2018). Tomato crops are characterized by tillage, irrigation, and use of pesticides and fertilizers, ranging from small (<1 ha) to large areas (> 100 ha). Tomato flowers are hermaphrodite, nectar-free, and have fused anthers with poricidal dehiscence (forming a cone around the stigma). Pollen release depends on vibration

which is known as buzz pollination (De luca and Vallejo-Marin, 2013). Wind can induce self-pollination of tomato in open-field (Hanna, 1999), but bees can vibrate their flowers and efficiently realize pollination (Thorp, 2000). Despite being an autogamous plant, studies showed that pollination by bees benefits tomato (Bartelli and Nogueira-Ferreira, 2014; Deprá et al., 2014; Strange, 2015), resulting in a positive relationship between bee diversity increase and yield (Macias-Macias et al., 2009). Bee pollination increases the amount of pollen on the stigma (Silva-Neto et al., 2013), resulting in larger fruits and more seeds per fruit (Vergara and Fonseca-Buendía, 2012; Nunes-Silva et al., 2013; Ahmad et al., 2016). In addition, a recent study evaluated the impact of natural cover on sonicating bees (bees that vibrate the flower) in tomato crops and found that loss of natural habitat, overall, negatively impacts this group of bees (Franceschinelli et al., 2018).

Understanding the effect of pollination by bees on agricultural production and how communities of these pollinators and their services respond to changes in the landscape surrounding agricultural crops is important to outline strategies that promote the increase and conservation of these organisms (Eeraerts et al., 2017). It is also necessary to know how local factors, including agrochemical use and irrigation methods, affect the pollination and pollinators. Thus, in this thesis, I have investigated how pollination by bees affects the tomato production and how the communities of these pollinators responded to landscape changes and farm management. In Chapter 1, I identified bees visiting tomato flowers and their characteristics; I verified climate variables' effect and the period of day in which bee forage activity took place; and I evaluated the fruit production under different pollination types. In Chapter 2, I evaluated how the landscape configuration (landscape scale) and farm management strategy (local scale) affect the bees' diversity in small tomato crops in the Zona da

Mata region, Minas Gerais, Brazil. In Chapter 3, I assessed the irrigation method's effect on the pollination and pollinators' presence in the tomato crop.

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CHAPTER 1

Bee community in open-field tomato crop and pollination effect by wild bees on fruit production

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Abstract

Bees are important components of the pollinator community of most ecosystems, contributing to the production of crops. The knowledge of the bees' fauna associated with crops and the pollination effect by bees on the fruit production and quality is important to the design, planning, and execution of projects to conserve pollinator populations in agricultural systems. The objectives of this study were to identify bees visiting tomato flowers and their characteristics; verify the climate variable and the day period on bee forage activity; and to evaluate the fruit production under different pollination types. The study was conducted from July 2015 to September 2017 in the Cajuri, Coimbra, Januária and Manga municipalities, Minas Gerais state, Brazil. Eleven tomato fields were sampled. Fruit set and quality from different pollination methods were evaluated with the treatments: single visit (SV), open pollination (OP), mechanical pollination (MP), and control (self-pollination) (C). A total of 1,998 individuals distributed in Andrenidae, Apidae, and Halictidae families were collected, with greater abundance and species richness in Apidae. The tomato fruit mass was higher in the OP than in the SV and MP, which differed from the C. The high abundance of *Exomalopsis analis* and its occurrence in all fields, allied with its sonication behavior, indicate that this species is an important pollinator of the tomato. The tomato does not depend exclusively on bee pollination, but this improves its yield and quality, especially when performed by individuals of different species.

Keywords: agriculture, Apoidea, conservation, ecosystem, pollinators, yield

1. Introduction

Pollinators provide essential services for the reproduction of wild plants, and they increase yield of many food crops (Gallai et al., 2009; Holzschuh et al., 2012). More than 75% of the leading global food crop depends on animal-mediated pollination to some degree (Klein et al., 2007), and insects are the main biotic pollinators (Gallai et al., 2009). Among insects, wild bees are efficient pollinators of cultivated plants (Garibaldi et al., 2013), increasing the yield and improving the quality, shelf-life, and commercial value of foods (Klatt et al., 2013; Brittain et al., 2014).

Despite the importance of bees for agricultural production, the decline of their populations in many regions of the world has raised concerns about food production (Potts et al., 2005; Burkle et al., 2013). Intensification and change in land use, pesticide spraying, climate change, pest and pathogen proliferation, and introduced alien species are factors that, alone or combined, contribute to declining bee populations (Potts et al., 2005; Goulson et al., 2008; Eeraerts et al., 2017). Among these factors, the agricultural intensification is characterized by increased agrochemical input, tillage, monocultures, and loss and fragmentation of natural or semi-natural habitats (Kennedy et al., 2013; Deguines et al., 2014; Cusser et al., 2016). These changes cause isolation of natural populations and reduce food sources and nesting sites (Ferreira et al., 2013), negatively influencing pollinator communities (Connelly et al., 2015; Mogren et al., 2016).

Agricultural systems support a less diverse community of bees, and most ecosystem services tend to be provided by a few dominant species (Kleijn et al., 2015; Winfree et al., 2015; Zou et al., 2017). However, a more diverse pollinator community appeared to enhance pollination efficiency through spatial and temporal complementarity in its pollination behavior, suggesting that a single or a few efficient

species may not provide the same benefits as a rich community of functionally distinct species (Rader et al., 2016; Eeraerts et al., 2017).

The tomato is one of the most consumed vegetables, with a global production in 2016 of about 177 million tons (FAOSTAT, 2018). Tomato flowers are hermaphrodite, nectar-free, and have fused anthers with poricidal dehiscence (forming a cone around the stigma). Pollen release depends on vibration, which is known as buzz pollination (De luca and Vallejo-Marin, 2013). Wind can induce self-pollination of this plant in the field (Hanna, 1999), but bees can vibrate their flowers and efficiently realize pollination (Thorp, 2000). Despite being a self-compatible plant, pollination by bees has proven to increase tomato yield (Bartelli & Nogueira-Ferreira, 2014; Deprá et al., 2014; Strange, 2015).

The great diversity of the Brazilian territory, the increase in the demand of food, and the intensification of agriculture increase the importance of studying bees' fauna in agricultural environments considering that many crops depend on the pollination mediated by these insects. The biology and ecology of the bees associated with agricultural crops should be studied to design, plan, and execute projects to conserve pollinator populations. Thus, the objectives of this study were to identify bees (potential pollinators) visiting tomato flowers and their characteristics; verify the effect of the climate variables and day period on bees' foraging activity; and to evaluate the fruit quality resulting from different pollination types. Two hypotheses were considered: First, the bee community in tomato crops is composed of a few species accounting for most of the abundance; second, a single bee visit is sufficient to pollinate tomato flowers and to produce fruit with a heavier weight.

2. Material and methods

2.1. Study sites

The study was conducted from July 2015 to September 2017 in the municipalities of Cajuri and Coimbra (Zona da Mata region) and Januária and Manga (Norte de Minas region), Minas Gerais state, Brazil. Zona da Mata region is characterized by relatively heterogeneous landscapes with intensively managed coffee crop, annual crops, grasslands, forests, and natural habitats (predominantly Atlantic forest). The Norte de Minas landscape has small crops (<1 ha) including orchards, vegetables, pastures, and less than 10% of native vegetation, except for Manga with predominantly large cultivated areas (> 30 ha) with annual crop, pasture, pumpkins, and tomato. Information on the sites sampled are available in the supplementary data (Table S1).

2.2. Sampling bees

Eleven fields were sampled in a period of 46 days. Bees visiting tomato flowers were collected from July to September 2015; March to May 2016; March to April 2017; and August to September 2017. In small fields (< 0.30 ha), we walked transects along all rows. In larger fields, we surveyed two areas (100 m long by 12 m wide) located in the edge of the crops. Each field was sampled between 8 a.m. and 2 p.m. for about 20-30 min/h, on dry and warm days (>20°C) with low wind speeds. Temperature and humidity were recorded every hour with a thermohygrometer. All bee visitors contacting the tomato flowers were sampled with a sweep net and subsequently killed with ethyl acetate in a killing jar. Bee samples per site and sampling period were pooled, sorted, pinned, labeled, and identified at species level. Voucher specimens were deposited at the Entomology Museum of the Universidade Federal de Viçosa (UFV), the bee collection of the Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto (FFCLRP-USP), and the taxonomic collections center of the Universidade Federal de Minas Gerais (UFMG). All bee species were classified according to visiting behavior in tomato flowers as milking (insertion behavior of the

proboscis in the cone formed by poricidal anthers to obtain pollen (Thorp 2000)), milking/robber (besides the insertion of the proboscis, the bees can damage the flower to obtain pollen), and sonicating (behavior of vibrating the flower with the indirect muscles of the thorax (Russell et al., 2016)) based on observations at the time of collection.

2.3. Pollination tests

Pollination tests were conducted in August and September 2015 in the agriculture sector of the Instituto Federal do Norte de Minas (IFNMG-Campus Januária). A crop with 350 plants of the H9559® hybrid was utilized. The plants were distributed in six rows and spaced 1 m between rows and 0.5 m between plants. Four treatments with four replications were performed in randomized blocks, totaling 16 plots. The treatments were: single visit (SV), open pollination (OP), mechanical pollination (MP), and control (C), in this case, spontaneous self pollination. Pollination tests were performed with flowers of the first three bunches of 10 plants per plot. The number of flowers per bunch ranged from five to 11. These flowers were wrapped with organza bags before anthesis in all treatments except open pollination. In the SV treatment, the flowers were observed from 9 a.m. and 2 p.m. During this period the organza bags were removed from the flower bunches, being bagged again after the first and only visit by bee. Bees of the genus *Augochloropsis* and *Centris* and the species *Bombus brevivillus*, *Exomalopsis analis*, and *Paratrigona lineata* were recorded in SV treatment, but, due to the loss of fruits by disease, only fruits pollinated by *Augochloropsis*, *Centris*, and *Paratrigona lineata* were compared. The results were pooled in the SV treatment because the fruit weight and fruit set between the genus were similar ($F_{2, 32} = 1.66$, $P = 0.2057$; $F_{2, 32} = 0.66$, $P = 0.5240$). In the OP, the flowers remained free for bee visits (we observed that the flowers in the OP received

more than three visits by bees, usually more than one species). In MP, the flowers were vibrated daily for 5 seconds, between 9 a.m. and 2 p.m. from anthesis to senescence with a Bitufo® electric toothbrush adapted to vibrate flowers (Palma et al., 2008). In the C treatment, the flowers remained bagged until the beginning of fruit development. The schedules used to apply the treatments were based on the period of receptivity of the stigma that is greater between 10:30 a.m. and 3:30 p.m. (Del Sarto et al., 2005). The first three bunches with ripe tomato fruit were harvested to evaluate the fruit set (%), fresh fruit mass (g), and seed numbers per fruit.

2.4. Statistical analyses

The relationship between temperature and relative humidity and the number of bees collected on tomato flowers was estimated by polynomial regression analysis to evaluate how climatic variables affect bee activity. An ANOVA ($\alpha= 0.05$) was conducted to evaluate whether there was variation in fruit set (%), fresh fruit mass (g), and seed numbers per fruit as a function of pollination treatments. In cases where ANOVA was significant, the Tukey test ($\alpha= 0.05$) was used for multiple comparisons. The response variable values were submitted to the Shapiro-Wilk and Bartlett tests to verify the normality and homogeneity of variance, respectively. Spearman correlation analysis ($\alpha= 0.05$) was performed between the seeds and the fruit mass numbers. All calculations and analyses were conducted in R (R Core Team, 2017).

3. Results

3.1. Bees' fauna

Seventy-seven and 63 sampling hours were spent in the Norte of Minas Gerais and Zona da Mata, respectively. In both regions, 1,998 individuals were collected during the 3 years. Fifty-six species and morpho-species of 21 genera and three

families were collected. Apidae represented 89.6% of the individuals collected, followed by Halictidae and Andrenidae, with 9.9 and 0.5%, respectively. The Andrenidae family was collected only in the Zona da Mata region (Table 1).

The species abundance and richness was higher in the Zona da Mata region, with 1,180 individuals of 36 species. In the North of Minas, 818 individuals from 29 species were collected. Among the species collected, only nine (16.07%) occurred in both regions. The species collected in both regions represented 60.56% of the bee community.

Table 1. Family, species, and abundance of bees collected in tomato crop in Norte de Minas (NM) and Zona da Mata (ZM), Minas Gerais state, Brazil in 2015 (015), 2016 (016), and 2017 (017) and total (T) individuals collected

Family/species	NM		ZM		T
	015	017	016	017	
Andrenidae					
<i>Oxaea flavescens</i> Klug, 1807	-	-	2	2	4
<i>Parapsaenythia serripes</i> (Ducke, 1908)	-	-	1	2	3
<i>Psaenythia</i> sp.	-	-	-	3	3
Apidae					
<i>Apis mellifera</i> Linnaeus, 1758	115	3	245	54	417
<i>Bombus (Fervidobombus) brevivillus</i> Franklin, 1913	28	74	-	-	102
<i>Bombus (Fervidobombus) morio</i> (Swederus, 1787)	-	-	10	19	29
<i>Bombus (Fervidobombus) pauloensis</i> Friese, 1913	-	-	4	-	4
<i>Centris (Centris) aenea</i> Lepeletier, 1841	-	-	1	1	2
<i>Centris (Hemisiella) tarsata</i> Smith, 1874	17	11	-	1	29
<i>Centris (Hemisiella) trigonoides</i> Lepeletier, 1841	41	23	-	-	64
<i>Centris (Trachina) fuscata</i> Lepeletier, 1841	2	4	-	-	6
<i>Euglossa</i> sp.	-	1	1	1	3
<i>Eulaema (Apeulaema) nigrita</i> Lepeletier, 1841	-	-	1	-	1
<i>Exomalopsis (Exomalopsis) analis</i> Spinola, 1853	74	35	301	295	705
<i>Exomalopsis (Exomalopsis) auropilosa</i> Spinola, 1853	-	-	32	75	107
<i>Exomalopsis (Exomalopsis) fernandoi</i> Moure, 1990	-	-	-	2	2
<i>Exomalopsis (Exomalopsis) fulvofasciata</i> Smith, 1879	-	-	2	-	2

<i>Exomalopsis (Exomalopsis) minor</i> Schrottky, 1910	-	-	1	8	9
<i>Exomalopsis (Exomalopsis) tomentosa</i> Friese, 1899	-	-	-	1	1
<i>Exomalopsis (Exomalopsis) ypirangensis</i> Schrottky, 1910	-	-	1	4	5
<i>Melipona (Eomelipona) asilvai</i> Moure, 1971	15	-	-	-	15
<i>Melipona (Eomelipona) bicolor</i> Lepeletier, 1836	-	-	-	1	1
<i>Melipona (Melikerria) quinquefasciata</i> Lepeletier, 1836	-	1	-	-	1
<i>Melipona (Melipona) quadrifasciata</i> Lepeletier, 1836	1	-	1	-	2
<i>Nannotrigona testaceicornis</i> (Lepeletier, 1836)	2	-	-	-	2
<i>Paratetrapedia</i> sp.	-	-	1	1	2
<i>Paratrigona lineata</i> (Lepeletier, 1836)	144	30	-	-	174
<i>Tetragonisca angustula</i> (Latreille, 1811)	-	-	5	-	5
<i>Thygater (Thygater) analis</i> (Lepeletier, 1841)	-	-	4	16	20
<i>Trigona hyalinata</i> (Lepeletier, 1836)	1	-	-	-	1
<i>Trigona recursa</i> Smith, 1863	4	36	-	-	40
<i>Trigona</i> sp.	4	-	-	-	4
<i>Trigona spinipes</i> (Fabricius, 1793)	12	3	12	3	30
<i>Xylocopa (Neoxylocopa) cearensis</i> Ducke, 1910	2	-	-	-	2
<i>Xylocopa (Neoxylocopa) suspecta</i> Moure & Camargo, 1988	-	-	-	1	1
<i>Xylocopa (Schonnherria) muscaria</i> (Fabricius, 1775)	1	-	-	1	2
Halictidae					
<i>Augochlora (Oxystoglossella)</i> sp.	1	-	-	-	1
<i>Augochlora (Oxystoglossella)</i> sp.2	-	1	-	-	1
<i>Augochloropsis</i> aff. <i>iris</i>	5	2	-	-	7
<i>Augochloropsis argentina</i> (Friese, 1908)	1	-	2	8	11
<i>Augochloropsis brachycephala</i> Moure, 1943	-	-	3	12	15
<i>Augochloropsis cleopatra</i> (Schrottky, 1902)	2	-	-	-	2
<i>Augochloropsis cupreola</i> (Cockerell, 1900)	-	-	-	5	5
<i>Augochloropsis illustris</i> (Vachal, 1903)	43	25	-	-	68
<i>Augochloropsis melanochaeta</i> Moure, 1950	-	1	-	-	1
<i>Augochloropsis smithiana</i> (Cockerell, 1900)	-	-	1	5	6
<i>Augochloropsis</i> sp.1	-	-	5	4	9
<i>Augochloropsis</i> sp.2	-	-	-	4	4
<i>Augochloropsis</i> sp.4	-	-	1	-	1
<i>Augochloropsis</i> sp.5	-	-	1	-	1
<i>Augochloropsis wallacei</i> (Cockerell, 1900)	-	-	-	2	2
<i>Dialictus</i> sp.	-	1	-	-	1
<i>Pseudaugochlora flammula</i> Almeida, 2008	5	-	-	-	5

<i>Pseudaugochlora graminea</i> (Fabricius, 1804)	1	1	5	4	11
<i>Pseudaugochlora indistincta</i> Almeida, 2008	-	-	2	-	2
<i>Pseudaugochlora pandora</i> (Smith, 1853)	21	24	-	-	45
Abundance	542	276	645	535	1998
Richness	24	18	26	28	56

Exomalopsis analis was the most abundant species, with 35.28% of the individuals collected, followed by the honeybee (*Apis mellifera*), with 20.87%. The five most abundant species represented 75.32% of the individuals. *Exomalopsis analis* was the only species collected in all fields, and 21 species (37.5%) occurred only in one of 11 fields. Most of the bees collected can vibrate the tomato flower.

3.2. Climate variables and flying activity

The relationship between climatic variables, temperature and relative humidity, and number of bees per flower was significant ($Y = -40.560 + 2.983X - 0.049X^2$, $R^2 = 0.75$ and $Y = -11.450 + 0.796X - 0.010X^2$, $R^2 = 0.46$). Bees visited the tomato flowers at temperatures between 23°C to 39°C and a relative humidity of 21% to 58%, especially in the morning. In both regions, the bee activity peaked between 9 a.m. and 11 a.m. for all bee groups except *Bombus*, with a peak at 8 a.m. (Figure 1). Bee visits to tomato flowers tend to decline after noon, with rare visits after 2 p.m.

3.3. Bee pollination

The tomato fruit set ($F_{3, 11} = 5.83$; $P = 0.0123$) was higher in the SV and MP treatments, followed by OP and the control (Figure 2). The fresh fruit mass was higher for OP than in the SV and MP, and it was lower in the control. The fruits of the OP were 33.5% heavier than the control ones. The seed numbers per fruit were similar in SV, OP, and MP, and they were lower in the control. Correlation between the seed numbers per fruit and fresh fruit mass was not significant ($r = 0.29$; $P = 0.074$).

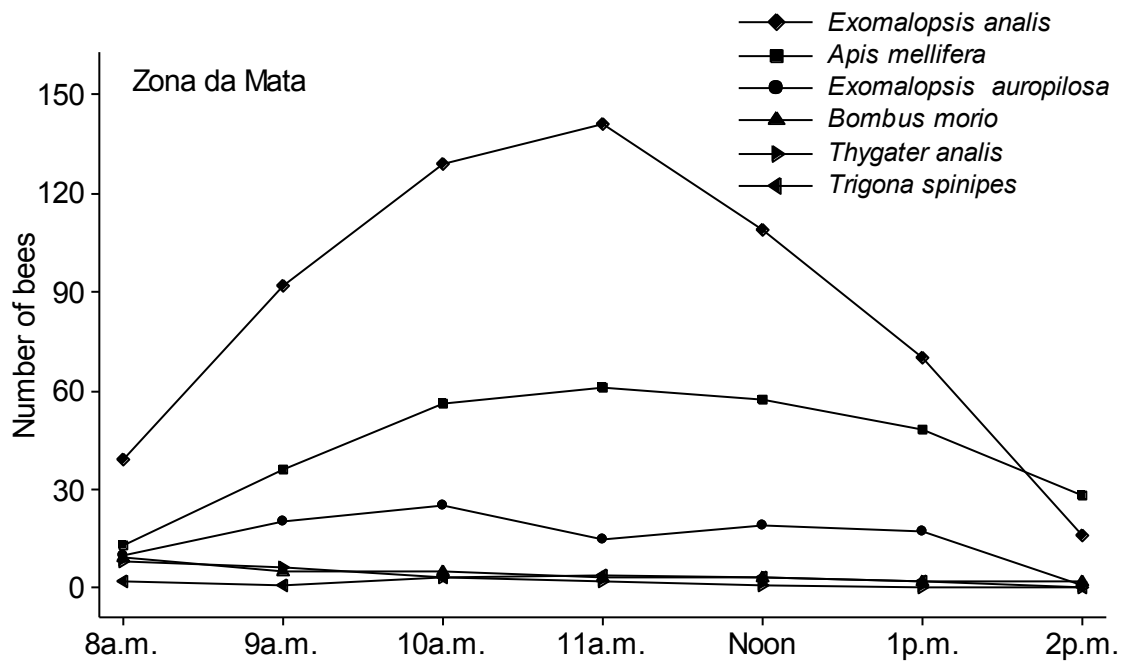
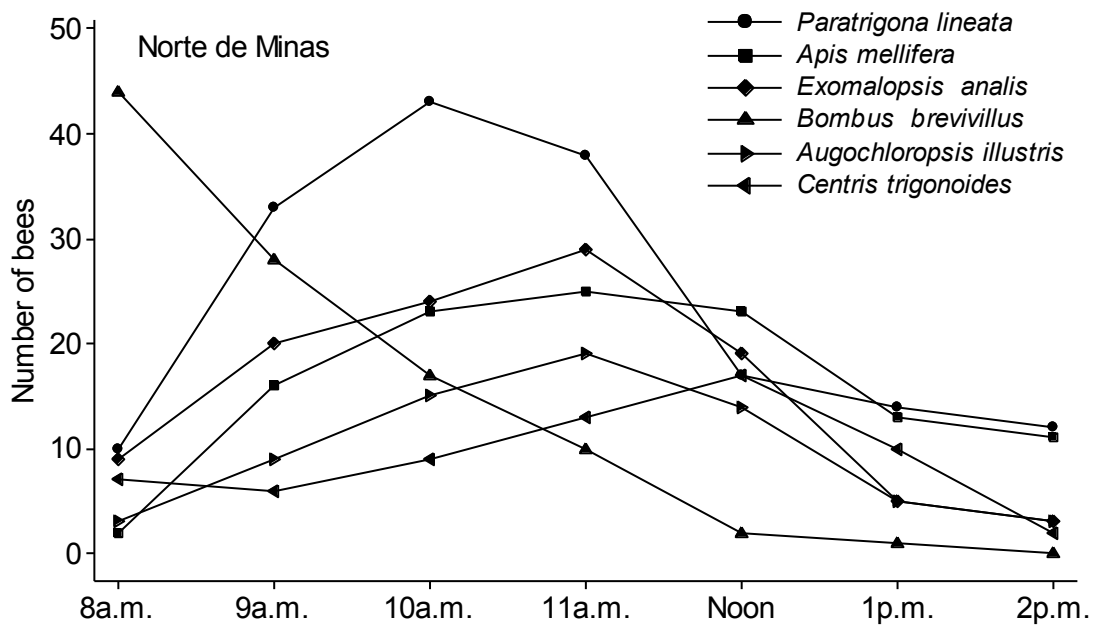


Figure 1. Activity of the main bee species on tomato flower during the day in Norte de Minas and Zona da Mata regions, Minas Gerais, Brazil.

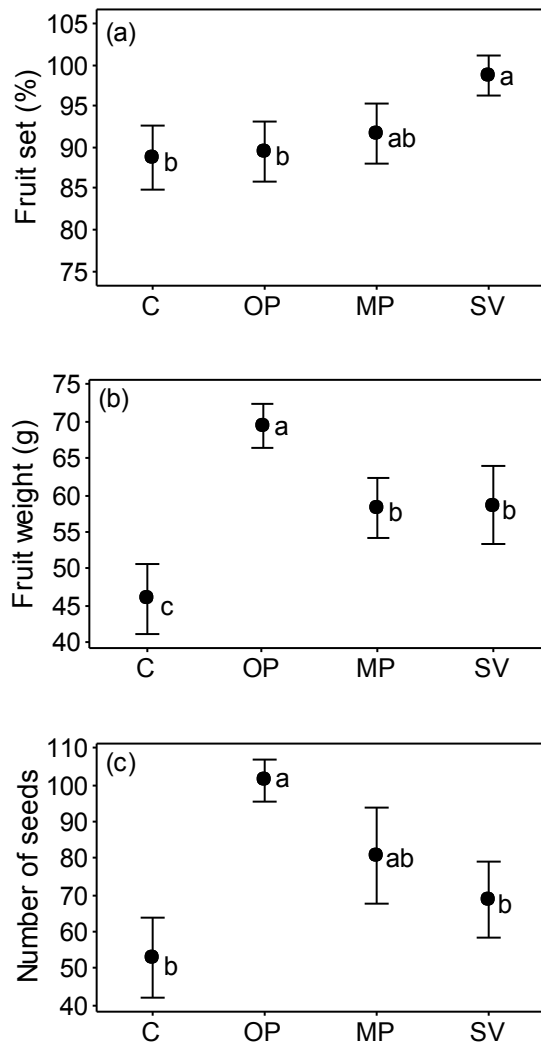


Figure 2. Fruit set (a), fruit weight in grams (b) and number of seeds per fruit (c) in function of control (C), open pollination (OP), mechanical pollination (MP), and single visit (SV). Means and standard error bars are shown for four experimental treatments. Bars followed by the same lowercase letter do not differ by the Tukey test ($\alpha= 0.05$).

4. Discussion

4.1. Bees' fauna

The tomato bee fauna is composed mainly of sonicating bees, with a wide variety of social classes and nesting habits, which is important for the pollination service stability in agricultural environments. The Apidae abundance may be related to

the great diversity of this group with diversified life histories. Species of this family have a range of social and foraging behavior, besides nesting habits (Danforth et al., 2013). These characteristics allow Apidae to explore diverse environments, including those with intensive agriculture (Ferreira et al., 2015). Conditions such as bare ground for nesting (Potts et al., 2005) and food sources in cultivated areas (Blitzer et al., 2012; Montero-Castaño et al., 2016) can support higher species numbers. This pattern was common in studies on bees' fauna in the tomato crop, with Apidae being dominant (Silva-Neto et al., 2013; Deprá et al., 2014; Santos et al., 2014).

The bees' greater abundance and richness in the Zona da Mata may be related to its more diverse landscape structure and composition compared to that of North of Minas Gerais, Brazil. Both regions are quite degraded, but the first has greater native forest patch numbers. These patches can provide food and nesting sources for bees, contributing to community diversification (Le Feon et al., 2010). On the other hand, nine species were common in both regions and represented 60.56% of the total abundance, indicating that the dominant fauna was practically similar in the two regions despite the differences in natural cover between them.

The higher abundance of *E. analis* and its presence in all sites sampled show that this species can be favored in agricultural environments, possibly due to the availability of bare ground which can benefit underground nesting species (Ferreira et al., 2015). On the other hand, intense soil disturbance in crop areas may affect underground species, but the possibility of nesting in the cultivated area, especially in carriers where the soil is compacted, may favor *E. analis* (Potts et al., 2005; Julier and T'ai, 2009). The high abundance of *Exomalopsis analis* and its occurrence in all sites, along with its sonication behavior, indicate that this species is an important pollinator of the tomato. This species was also the most abundant in other studies on communities of bees in the tomato fields of Goiás, Rio de Janeiro and Triângulo Mineiro (Silva-

Neto et al., 2013; Deprá et al., 2014; Santos et al., 2014), reinforcing its importance in the tomato crops.

The high numbers of honeybee individuals may be related to characteristics of this species such as nests with thousands of individuals and the ability to trace and recruit nestmates to many food sources through a sophisticated communication and labor system division. In agricultural environments, the mass flowering of other crops during the off-season of the tomato crop may favor this species maintenance in the area (Blitzer et al., 2012; Montero-Castaño et al., 2016). We were not aware of any apiaries near the study fields, so individuals may have been from feral nests. On the other hand, the honeybee is a less efficient pollinator than other bees because it does not sonicate flowers and obtains tomato pollen by inserting the proboscis into the anther cone (Dos Santos et al., 2009), a behavior known as milking. However, the large number of visits that this species performs per flower can compensate its relative inefficiency (Macias-Macias et al., 2009).

The predominance of sonicating bees in tomato crops can be explained by the efficiency of this group in extracting pollen from poricidal anthers, playing an important role in tomato pollination (Greenleaf and Kremen, 2006). On the other hand, non-sonicating bees may not be attracted to a tomato due to the absence of nectar in the flower of this plant and the difficulty in extracting pollen from its poricidal anther (King and Buchmann, 2003). However, in our study, the presence of non-sonicating bees in tomato crops may be related to the absence or low availability of other pollen sources besides the tomato in the areas sampled.

4.2. Climate variables and flying activity

The lower temperature may have favoured a greater *Bombus* activity early in the morning, since larger black bees may have difficulties with dehydration and

thermal regulation at high temperatures (Pereboom and Biesmeijer, 2003). Different foraging peaks per species indicate the possibility of temporal tomato pollen partition (Roubik, 1989). These differences may increase pollination efficiency due to complementarity between different bee species (Martins et al., 2015). In addition, the period of higher activity of the pollinators corresponds, in part, to the period of greater receptivity of the stigma in the tomato, which is from 10:30 a.m. to 3:30 p.m. (Del Sarto et al., 2005).

On the other hand, knowledge about the variation in flight activity of bees throughout the day could be used in the planning of pesticide spraying. This should be done in periods that do not coincide with those of greater activity for these insects to minimize their contact with the pesticides. These products should be applied after 2 p.m. to allow a longer interval between bee visits and spraying.

4.3. Bee pollination

The highest fruit set in the SV treatment indicates that a single bee visit was sufficient for a fruit set, which may be related to the pollen amount extracted from the flower in the first visit. *Bombus (Pyrobombus) impatiens* Cresson 1863 (Bombini: Apidae) removes 57.8% of the pollen from tomato anthers at its first visit, and this quantity is sufficient to fertilize the flower (Nunes-Silva et al., 2013). On the other hand, the absence of mechanical stimuli in flowers in the control may have led to lower pollen deposition on the stigma and, consequently, flower abortion, resulting in lower fruit production.

Multiple visits by different bee species in the OP may have allowed a greater pollen deposition on the stigma and, hence, the heavier fruit formation (Silva-Neto et al., 2013). The possibility of different pollinator species visiting the same flower may improve pollination services due to the behavioral particularities of each functional

group, complementing each other in time and space (Hoehn et al., 2008; Gagic et al., 2015; Martins et al., 2015). The lighter fresh fruit mass with MP demonstrates the lower efficiency of this method. MP was performed several times, but the vibration frequency of the equipment may not have been compatible with that required for efficient pollen release (Palma et al., 2008).

The similar seed numbers in tomato fruits in SV, OP, and MP treatments reinforces the lack of correlation between fruit mass and seed numbers. The fruit tomato development generally depends on pollination and fertilization, demonstrating the importance of seed formation for their growth (Shinozaki et al., 2015). However, it is possible that other factors have affected the relationship between fruit mass and number of seeds, including the possibility of genes being involved in forming parthenocarpic fruits (Mazzucato et al., 2015).

5. Conclusion

Our results support the general hypothesis that few species dominate bees' fauna in agricultural environments. However, contrary to our expectations, a single visit by a bee to the flower did not result in heavier fruits compared to open pollination when, supposedly, the flower was visited more than once by different species. Thus, we emphasize the importance of more diverse communities for the efficiency and stability of pollination services. Considering that the loss of diversity is associated with several factors, including intensification of agriculture, future studies on how the community of bees responds to landscape changes and farm management are needed.

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Appendix A. Supplementary data

Table S1. Species, family, visiting behavior, social class, and nest class of bees sampled in Norte de Minas and Zona da Mata, Minas Gerais state, Brazil

Species	Family	Visiting behavior	Norte de Minas		Zona da Mata		Total
			2015	2017	2016	2017	
<i>Apis mellifera</i> Linnaeus, 1758	Apidae	Milking	115	3	245	54	417
<i>Nannotrigona testaceicornis</i> (Lepeletier, 1836)	Apidae	Milking	2	-	-	-	2
<i>Tetragonisca angustula</i> (Latreille, 1811)	Apidae	Milking	-	-	5	-	5
<i>Trigona</i> sp.	Apidae	Milking/Robber	4	-	-	-	4
<i>Melipona (Eomalipona) asilvai</i> Moure, 1971	Apidae	Sonicating	15	-	-	-	15
<i>Melipona (Eomalipona) bicolor</i> Lepeletier, 1836	Apidae	Sonicating	-	-	-	1	1
<i>Melipona (Melipona) quadrifasciata</i> Lepeletier, 1836	Apidae	Sonicating	1	-	1	-	2
<i>Bombus (Fervidobombus) brevivillus</i> Franklin, 1913	Apidae	Sonicating	28	74	-	-	102
<i>Bombus (Fervidobombus) morio</i> (Swederus, 1787)	Apidae	Sonicating	-	-	10	19	29
<i>Bombus (Fervidobombus) pauloensis</i> Friese, 1913	Apidae	Sonicating	-	-	4	-	4
<i>Centris (Hemisiella) tarsata</i> Smith, 1874	Apidae	Sonicating	17	11	-	1	29
<i>Centris (Hemisiella) trigonoides</i> Lepeletier, 1841	Apidae	Sonicating	41	23	-	-	64
<i>Euglossa</i> sp.	Apidae	Sonicating	-	1	1	1	3
<i>Xylocopa (Neoxylocopa) cearensis</i> Ducke, 1910	Apidae	Sonicating	2	-	-	-	2
<i>Xylocopa (Neoxylocopa) suspecta</i> Moure & Camargo, 1988	Apidae	Sonicating	-	-	-	1	1
<i>Xylocopa (Schonnherria) muscaria</i> (Fabricius, 1775)	Apidae	Sonicating	1	-	-	1	2
<i>Trigona hyalinata</i> (Lepeletier, 1836)	Apidae	Milking/Robber	1	-	-	-	1

<i>Trigona spinipes</i> (Fabricius, 1793)	Apidae	Milking/Robber	12	3	12	3	30
<i>Oxaea flavescens</i> Klug, 1807	Andrenidae	Sonicating	-	-	2	2	4
<i>Parapsaenythia serripes</i> (Ducke, 1908)	Andrenidae	Sonicating	-	-	1	2	3
<i>Psaenythia</i> sp.	Andrenidae	Sonicating	-	-	-	3	3
<i>Exomalopsis (Exomalopsis) analis</i> Spinola, 1853	Apidae	Sonicating	74	35	301	295	705
<i>Exomalopsis (Exomalopsis) auropilosa</i> Spinola, 1853	Apidae	Sonicating	-	-	32	75	107
<i>Exomalopsis (Exomalopsis) fernandoi</i> Moure, 1990	Apidae	Sonicating	-	-	-	2	2
<i>Exomalopsis (Exomalopsis) fulvofasciata</i> Smith, 1879	Apidae	Sonicating	-	-	2	-	2
<i>Exomalopsis (Exomalopsis) minor</i> Schrottky, 1910	Apidae	Sonicating	-	-	1	8	9
<i>Exomalopsis (Exomalopsis) tomentosa</i> Friese, 1899	Apidae	Sonicating	-	-	-	1	1
<i>Exomalopsis (Exomalopsis) ypirangensis</i> Schrottky, 1910	Apidae	Sonicating	-	-	1	4	5
<i>Centris (Centris) aenea</i> Lepeletier, 1841	Apidae	Sonicating	-	-	1	1	2
<i>Centris (Trachina) fuscata</i> Lepeletier, 1841	Apidae	Sonicating	2	4	-	-	6
<i>Paratetrapedia</i> sp.	Apidae	Sonicating	-	-	1	1	2
<i>Thygater (Thygater) analis</i> (Lepeletier, 1841)	Apidae	Sonicating	-	-	4	16	20
<i>Dialictus</i> sp.	Halictidae	Milking	-	1	-	-	1
<i>Augochlora (Oxystoglossella)</i> sp.	Halictidae	Sonicating	1	-	-	-	1
<i>Augochlora (Oxystoglossella)</i> sp.2	Halictidae	Sonicating	-	1	-	-	1
<i>Augochloropsis</i> aff. <i>iris</i>	Halictidae	Sonicating	5	2	-	-	7
<i>Augochloropsis argentina</i> (Friese, 1908)	Halictidae	Sonicating	1	-	2	8	11
<i>Augochloropsis brachycephala</i> Moure, 1943	Halictidae	Sonicating	-	-	3	12	15
<i>Augochloropsis cleopatra</i> (Schrottky, 1902)	Halictidae	Sonicating	2	-	-	-	2

<i>Augochloropsis cupreola</i> (Cockerell, 1900)	Halictidae	Sonicating	-	-	-	5	5
<i>Augochloropsis illustris</i> (Vachal, 1903)	Halictidae	Sonicating	43	25	-	-	68
<i>Augochloropsis melanochaeta</i> Moure, 1950	Halictidae	Sonicating	-	1	-	-	1
<i>Augochloropsis smithiana</i> (Cockerell, 1900)	Halictidae	Sonicating	-	-	1	5	6
<i>Augochloropsis</i> sp.1	Halictidae	Sonicating	-	-	5	4	9
<i>Augochloropsis</i> sp.2	Halictidae	Sonicating	-	-	-	4	4
<i>Augochloropsis</i> sp.4	Halictidae	Sonicating	-	-	1	-	1
<i>Augochloropsis</i> sp.5	Halictidae	Sonicating	-	-	1	-	1
<i>Augochloropsis wallacei</i> (Cockerell, 1900)	Halictidae	Sonicating	-	-	-	2	2
<i>Pseudaugochlora flammula</i> Almeida, 2008	Halictidae	Sonicating	5	-	-	-	5
<i>Pseudaugochlora graminea</i> (Fabricius, 1804)	Halictidae	Sonicating	1	1	5	4	11
<i>Pseudaugochlora indistincta</i> Almeida, 2008	Halictidae	Sonicating	-	-	2	-	2
<i>Pseudaugochlora pandora</i> (Smith, 1853)	Halictidae	Sonicating	21	24	-	-	45
<i>Paratrigona lineata</i> (Lepelletier, 1836)	Apidae	Milking/Robber	144	30	-	-	174
<i>Trigona recursa</i> Smith, 1863	Apidae	Milking/Robber	4	36	-	-	40
<i>Eulaema (Apeulaema) nigrita</i> Lepelletier, 1841	Apidae	Sonicating	-	-	1	-	1
<i>Melipona (Melikerria) quinquefasciata</i> Lepelletier, 1836	Apidae	Sonicating	-	1	-	-	1
Abundance			542	276	645	535	1998
Richness			24	18	26	28	56

Table S2. Characteristics of the sites and tomato crops used for the sampling of bees in Norte de Minas and Zona da Mata, Minas Gerais, Brazil

	Municipality			
	Cajuri	Coimbra	Januária	Manga
Region	Zona da Mata	Zona da Mata	Norte de Minas	Norte de Minas
Natural cover	Atlantic Rain Forest*	Atlantic Rain Forest	Savanna/SDTF**	Savanna/SDTF
Climate type (Köepen)	Cwa	Cwa	Aw	Aw
Sampling year	2016	2016/2017	2015/2017	2015
Number of fields sampled	1	5	4	1
Size of tomato crop (ha)	0.3	0.2-0.4	0.3-2.0	30
Variety	Aguamiel®	Aguamiel®	H9553®/H9992®	H9553®
Tomato growing	Indeterminate	Indeterminate	Determinate	Determinate
Spacing (m)	0.5 x 1.2	0.5 x 1.2	0.5 x 1.0	0.3 x 1.2
Irrigation system	Drip	Drip	Sprinkler/Drip	Center pivot
Farming practices	Conventional	Conventional	Conventional	Conventional

*Atlantic Rain Forest = Brazilian Atlantic forest; **Savanna = Brazilian Cerrado/SDTF (Seasonally dry tropical forest) = Brazilian Caatinga [Based on Pennington and Lavin (2015) modified from Olson et al. (2001)].

Chapter 2

Impact of landscape configuration and farm management on bee communities in tomato fields

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Abstract

Bees are important components of the pollinators' fauna, providing essential services for the reproduction of wild plants and production of many food crops. Paradoxically, wild bees are strongly affected by agricultural intensification at both local and landscape scales. Increasing the distance to natural habitats, loss of natural habitats, and increased use of agrochemicals are hypothesized to negatively affect the community of bees. Here, we evaluate the effect of landscape configuration and farm management on bee communities in tomato crops. Bees were sampled in 11 tomato fields in Cajuri and Coimbra, Minas Gerais state, Brazil. For each site, the percentage of land use class was calculated within a buffer around the tomato field with a radius of 500 and 1,000 meters. Additionally, we conducted face-to-face interviews with farmers and an *in situ* survey to assess the field management used on each site. We used generalized linear mixed effects models to analyze variation in the wild bee richness, wild bee abundance, honey bee abundance, and functional guild among study sites. Overall, while wild bees responded negatively to natural habitat loss and farm management intensification, honeybees responded positively to these variables. Our results support a more general hypothesis that both farm management (local) and landscape scales are important in structuring the community of bees in agricultural environments and provide important data that can guide pollinator management plans.

Keywords: agriculture, biodiversity, conservation, ecosystem, land-use, pollinator

1. Introduction

Pollination is an essential service for the sexual reproduction of wild plant species, also increasing the yield of several crops (Gallai et al., 2009; Bommarco et al., 2012). More than 75% of the leading global food crops depend on animal-mediated pollination (Klein et al., 2007), and insects, especially bees, are the main biotic pollinators (Gallai et al., 2009). Wild bees are efficient pollinators of cultivated plants (Garibaldi et al., 2013), improving the yield, quality, shelf-life, and commercial value of food (Klatt et al., 2013; Brittain et al., 2014); yet, a decline in bees' population in many world regions has raised concerns about the yield and stability of food production (Potts et al., 2010; Burkle et al., 2013). Several reasons have been raised for their decline, including intensification and land use change, indiscriminate use of pesticides, climate change, proliferation of pests and pathogens, and introduction of exotic species (Potts et al., 2010; Goulson et al., 2015).

Agricultural intensification is characterized by increased agrochemical input, tillage, large crops, and loss and fragmentation of natural habitats (Kennedy et al., 2013; Deguines et al., 2014). These changes cause isolation of natural populations and reduce food sources and nesting sites (Ferreira et al., 2013), negatively affecting pollinator communities (Connelly et al., 2015; Mogren et al., 2016). It is known that placing crop areas in proximity with natural or semi-natural vegetation is important to increase the diversity of pollinators and level pollination in crops (Carvalho et al., 2010; Geslin et al., 2016). On the other hand, indiscriminate use of pesticides (insecticides, fungicides and herbicides) can negatively affect pollinator populations by direct and indirect exposure, reducing pollination efficiency (van der Sluijs et al., 2013; Feltham et al., 2014; Park et al., 2015).

Wild bees and honey bees tend to present opposite responses regarding the loss

of natural habitat and pesticide use. The managed bee *Apis mellifera* is more opportunistic and generalist and thus appears to be less dependent on natural habitats than wild bees (Saturni et al., 2016). There are reports of a positive relationship between *A. mellifera* abundance and agriculture intensification (Marini et al., 2012; Petersen and Nault, 2014; Eeraerts et al., 2017). Due to their habits, especially the ability to forage in almost all types of flowers, such generalist species can survive in modified environments (Ferreira et al., 2015). In addition, the introduction of hives or the existence of apiaries near the crops can increase honey bees' abundance in these areas (Martins et al., 2015; Gibbs et al., 2016). Agricultural landscapes may also favor some wild bee groups, mainly underground nesting groups that rely on bare soil to build their nests (Potts et al., 2005; Ferreira et al., 2015). Due to the ability of some few wild bee species to persist in highly modified anthropogenic landscapes (Tangtorwongsakul et al., 2018), the natural community of bees tends to shift its composition to a few dominant species (Kleijn et al., 2015; Mogren et al., 2016) which should then provide the majority of pollination services (Winfree et al., 2015; Cusser et al., 2016). However, this less diverse bees' community of both species richness and functional traits (Hoehn et al., 2008; Gagic et al., 2014; Martins et al., 2015) may not be able to provide stable pollination services over space and time (Rader et al., 2015; Cusser et al., 2016).

Tomato is one of the most consumed vegetables in the world and in Brazil, with crop areas ranging from small (< 1 ha) to large (> 100 ha). Tomato crops are grown intensively, being characterized by tillage, irrigation and use of pesticides and fertilizers. Despite being an autogamous plant, studies showed that pollination by bees benefits tomato (Bartelli and Nogueira-Ferreira, 2014; Deprá et al., 2014), with a positive relationship between bee diversity and yield (Macias-Macias et al., 2009). Bee pollination increases the amount of pollen on the stigma (Silva-Neto et al., 2013),

resulting in larger fruits and more seeds per fruit (Vergara and Fonseca-Buendía, 2012; Nunes-Silva et al., 2013). However, the loss of natural habitat in the landscape surrounding the tomato crops may negatively affect sonicating bees (Franceschinelli et al., 2017).

Understanding how communities of pollinators and their services respond to changes in the landscape surrounding agricultural crops is important to outlining strategies that promote the increase and conservation of these organisms (Eeraerts et al., 2017). It is also necessary to know how local factors affect the pollinators. In this study, we aim to evaluate how the landscape configuration (landscape scale) and farm management strategy (local scale) affect the bees' diversity in small tomato crops in the Zona da Mata region, Minas Gerais, Brazil. We tested the following hypotheses stating that bees' diversity should positively respond to (i) proximity with natural habitats and (ii) landscape configuration with more natural areas; and negatively respond to (iii) farm management intensification. We expect that with the increasing proximity and percentage of natural and semi-natural areas, more bee species will be found in the tomato patches. We also expect that bees' species richness should decline with increasing use of insecticides, fungicides, and fertilizers.

2. Material and methods

2.1. Study sites

The study was carried out in the Coimbra and Cajuri municipalities, Minas Gerais state, southeastern Brazil, within the Atlantic Forest domain. The region is characterized by relatively heterogeneous landscapes consisting of intensively managed coffee crop, annual crops, grasslands, and forests and natural habitat, predominantly Atlantic Forest. The climatic type of the region is the Cwa, Koppen classification, with a dry winter and a rainy summer. Eleven farms producing tomato

were selected for this study (Fig. 1). Tomato areas ranged from 0.16 to 1.86 ha (0.72 ± 0.63). The distance between the studied tomato fields ranged from 2 km to 9 km. Additional information is available in Table S1 and S2 of the supplementary data.

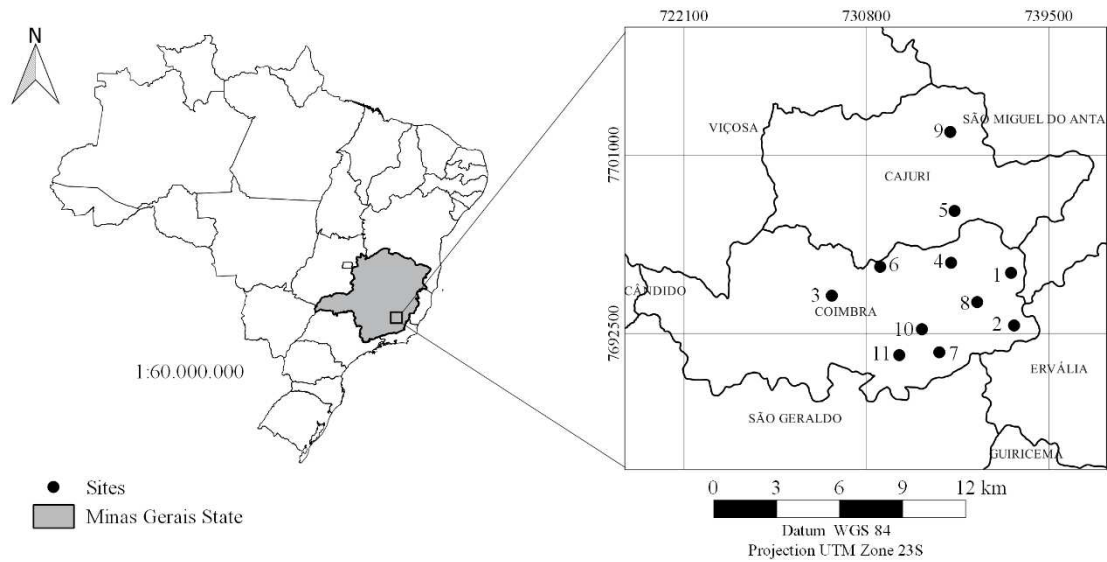


Figure 1. Eleven study sites located in Cajuri and Coimbra, Zona da Mata region, Minas Gerais state, Brazil.

2.2. Landscape configuration

Eleven fields of tomato crop were selected along a gradient of surrounding landscape complexity. A GIS (Geographic Information System) was used to map the land use in the surrounding landscape of each site. We allocated the land use class in the 500- and 1,000-meter buffer zone around the tomato fields based on orbital images of the Sentinel 2A satellite (Fig. 2).

The Sentinel 2A satellite acquires the reflected radiation in 13 spectral bands, of which only three were used because they have a more detailed spatial resolution. The screen classification (selection of land use around tomato fields) was performed with bands 4 (red), 3 (green) and 2 (blue) to obtain a natural color composition (RGB-432) with 10 m of resolution. An *in situ* survey was carried out in all tomato areas to classify

land uses in detail. The Sentinel 2A image used corresponds to the designation T23KQT of December 30, 2016.

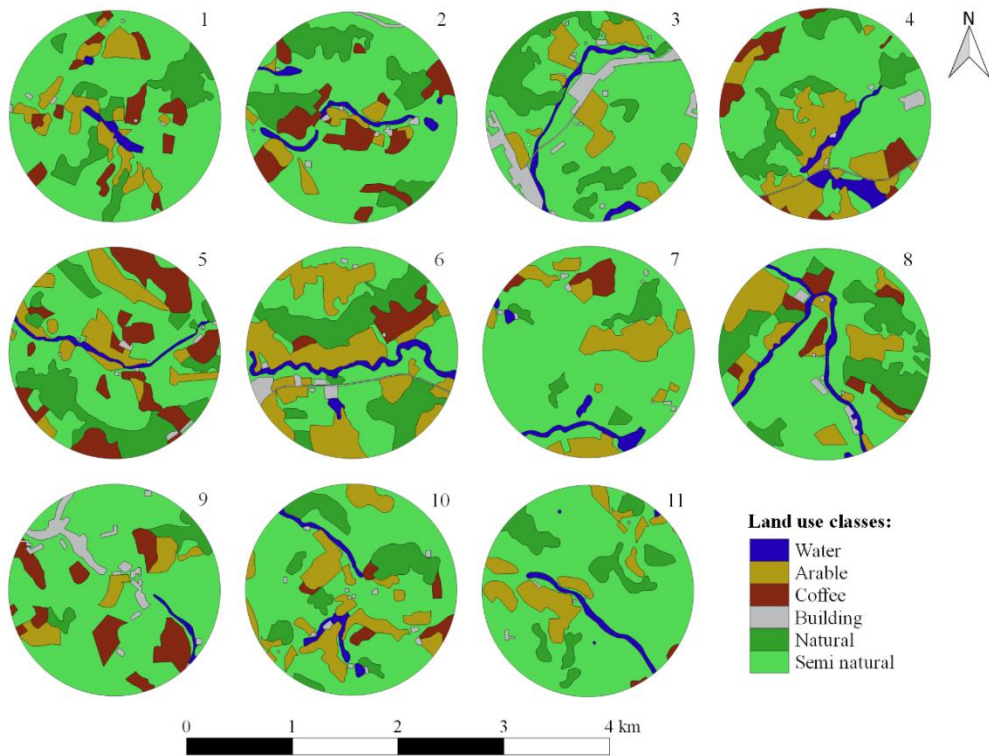
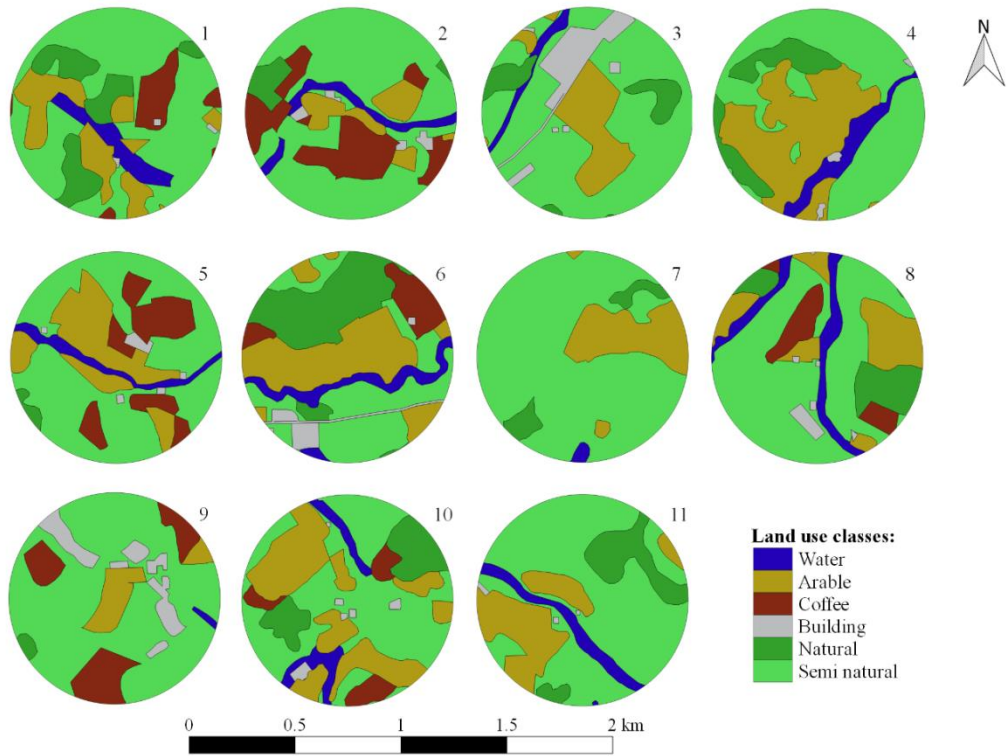


Figure 2. 500- and 1000-m radius circle around each tomato crop with land use proportion. The land-use classes include water, arable, coffee, building, natural, and semi-natural.

For each site, the percentage of land use class was calculated within a buffer around the tomato field with a radius of 500 and 1000 meters using QGIS (QGIS DEVELOPMENT TEAM, 2017). The following land use classes were used: natural habitat (Atlantic Forest), semi natural habitat (cultivated forest and pastures), coffee (coffee fields), arable (bean, maize, and vegetable), water (lagoon, lake, river, and floodplain), and building (masonry building, road, and urban).

2.3. *Farm management*

We conducted face-to-face interviews with farmers and an *in situ* survey to assess the field management used on each site. We collected information on eight variables: crop area size, tomato variety, input of leaf fertilizers, insecticides, fungicides, herbicides, number of sprays per week, and reuse of the area. All farms perform the conventional management of pests, disease, and weeds. The amount of pesticides and frequency of spraying ranged between the sites. The variable use of insecticide and fungicide was based on the number of commercial products, active ingredients, and chemical groups. The use of fertilizers took into account the number of commercial products and the amount of nutrients supplied. Insecticides, fungicides, and leaf fertilizers used in tomato crops are presented in Table S5.

2.4. *Sampling bees*

A total of 33 days was spent sampling all 11 sites between March 10 and April 21, 2017. We used this period for sampling because of the heavy rainfall before March and cold after April (<19°C), which reduces the flight activity of bees. In small fields

(< 0.30 ha), we walked transects along all the rows. In larger fields, we surveyed two areas that were 100 m long by 12 m wide located at the edge of each crop. Each field was sampled between 8 a.m. and 1 p.m. during a period of 30 min/h, on dry and warm days (>20°C) with low wind speeds. Surveys were repeated three times on each site. All bee visitors that came into contact with the tomato flowers were sampled with a sweep net, and they were subsequently killed with ethyl acetate in a killing jar. Bee samples of each site and sampling period were pooled, sorted, pinned, labeled, and subsequently identified to species level. Voucher specimens were deposited at the Entomological Museum of the Universidade Federal de Viçosa.

All bee species were categorized based on a compilation of ecological and life histories from the existing literature (Michener, 2007) and body size (Table S3). We considered the following traits to delineate functional guild: nesting class, social class, and body size (based on inter-tegula distance). Nesting class was categorized as above-ground cavity, carpenter, exposed, miners, and soil cavity. Social class was categorized as communal, facultatively social (It is not a distinct social category, but rather indicates a species' capacity for plasticity in its social organization), highly eusocial, primitively eusocial, and solitary. The concepts about the degree of sociability were based on Michener (2007) as well as other studies referenced in the supplementary data. Considering the lack of data on the social behavior of many species, our classification for some species was based on the predominant social behavior in the family, subfamily, or genus. The inter-tegula distance (ITD) was classified as small (< 2 mm), medium (2.1 - 4.0 mm), large (4.1 - 6.0 mm), and extra-large (> 6.1 mm) (Table S3 and S4).

2.5. Statistical analyses

Data from the three different bee sampling rounds were analyzed separately for each site. To assess the impact of landscape configuration and farm management on bee species richness and abundance we used generalized linear mixed models (GLMM, Bolker et al., 2009). We used a negative binomial error distribution (considering overdispersion in our count data; O'Hara and Kotze, 2010) with a log link function. Initially, the following land use classes and farm management were considered as explanatory variables: nearest natural habitat (NNH), natural habitat proportion (natural), semi-natural habitat proportion (semi-natural), coffee field proportion (coffee), arable soil proportion (arable), water proportion (water), building proportion (building), tomato crop size (area), tomato variety (cultivar), irrigation method (irrigation), insecticide input (insecticide), fungicide input (fungicide), fertilizer input (fertilizer), number of reuse of tomato crop area (reuse), and number of sprays per week (spraying).

We tested the correlation between the explanatory variables by calculating Pearson correlation coefficients and only considered models that did not include highly correlated variables. As most of the land use classes are highly correlated (Table S6), a principal component analysis (PCA) was conducted for each buffer. A "landscape metrics" variable was generated from axis 1 and 2 of the PCA for each buffer. Axis 1 and 2 retained 72.68% and 17.91% of the variation in a 1000-meter radius and 69.87% and 18.94% in a 500-meter radius, respectively (Table S7). Axis 1 and 2 of the PCA were highly correlated with both 500- and 1000-meter scales, which were also self-correlated. For simplicity, we retained in the model only the axis of the PCA of 1000 m. Semi-natural habitat and coffee were the highest weight variables in axis 1 (PCA1) and axis 2 (PCA2), respectively. All farm management variables were included in the models except insecticide input and fungicide input, which were correlated with each other ($r = 0.392$; $P < 0.05$). Insecticide input was correlated with fertilizer input ($r = -$

0.382; $P < 0.05$) and spraying ($r = 0.412$; $P < 0.05$). The following explanatory variables were included in the models as fixed factors: area, cultivar, fertilizers, irrigation, NNH, PCA1 (1000), PCA2 (1000), reuse, and spraying. In all models, we included the sampling site identity as a random factor.

We used generalized linear mixed effects models to analyze variation in the wild bee richness, wild bee abundance, honey bee abundance, and functional guild among study sites (Table 1 and 2). For each response variable, we selected the set of models based on Akaike information criterion with a second-order correction for small sample sizes (AICc), and we also calculated the Akaike weight (w) for each model. Models with ΔAICc smaller than 2 were considered equally plausible, and model averaging was used to determine the effect of each explanatory variable in the response variable (Burnham and Anderson, 2004).

All calculations and analyses were conducted in R (R Core Team, 2018) with car package (Fox et al., 2011), ggplot2 package (Wickham et al., 2016), lme4 package (Bates et al., 2014), MuMIn package (Barton, 2016), psych package (Revelle and Revelle, 2015), Rmisc package (Hope, 2013), and vegan package (Oksanen et al., 2015).

3. Results

3.1. Bees' community

We collected 1,932 bees distributed in three families (Andrenidae, Apidae and Halictidae), 21 genera, and 40 species and morpho-species (Table S3). *Exomalopsis analis* was most abundant and dominant in all sampling sites, representing 66.67% of all collected individuals, followed by *Apis mellifera* (honey bee) and *Exomalopsis auropilosa*, with 11.18% and 5.85%, respectively. Thirty percent of the species were sampled only once and was comprised of single individuals.

Table 1. Most parsimonious models of wild bee richness, wild bee abundance, honey bee abundance, and functional guild for the effects of landscape configuration farm management on the bee community in 11 tomato crops. Models were selected based on AICc and only the three top ranked models are presented; df is degrees of freedom; logLik is the log likelihood of the model; R² is the explained deviance; Δ AICc is the difference in AICc between a model and the model with the lowest AICc: differences between 0-2 = substantial support, 4-7 = considerably less support, >10 = essentially no support (Burnham & Anderson, 2002), and the weight represents a degree of belief in the model (Bolker, 2008).

Response variable/model	Model No	df	LogLik	R ²	AICc	Δ AICc	Weight
Wild bee richness							
Richness ~ cultivar + NNH + PCA2 + spraying	1	7	-68.99	0.63	156.5	0.00	0.493
Richness ~ cultivar + NNH + PCA2 + spraying	2	8	-68.74	0.64	159.5	3.01	0.109
Richness ~ cultivar + NNH + PCA1 + PCA2 + spraying	3	8	-68.74	0.64	159.5	3.01	0.109
Wild bee abundance							
Abundance ~ irrigation + PCA1 + PCA2 + spraying	1	7	-135.38	0.70	289.2	0.00	0.167
Abundance ~ irrigation + PCA2 + spraying	2	7	-135.38	0.70	289.2	0.00	0.167
Abundance ~ irrigation + PCA2 + spraying	3	7	-135.38	0.70	289.2	0.00	0.167
Honey bee abundance							
Abundance ~ reuse + spraying	1	6	-88.55	0.49	192.3	0.00	0.172
Abundance ~ PCA1 + reuse + spraying	2	6	-88.55	0.49	192.3	0.00	0.172
Abundance ~ reuse + spraying	3	6	-88.55	0.49	192.3	0.00	0.172
Functional guild							
Functional ~ irrigation + spraying	1	5	-65.00	0.24	142.2	0.00	0.267
Functional ~ area + irrigation + spraying	2	6	-63.91	0.28	143.1	0.82	0.177
Functional ~ cultivar + NNH + PCA2 + spraying	3	7	-62.36	0.35	143.2	0.97	0.164

Table 2. Generalized linear mixed-effects models showing the effects of area (tomato size crop), cultivar (tomato variety: aguamiel vs. totally), fertilizers (tomato crop fertilizer input), irrigation (irrigation method: drip vs. manual), NNH (nearest natural habitat), PCA1 (axis 1 of use land class in a 1000-m buffer), PCA2 (axis 2 of use land class in a 1000-m buffer), reuse (number of re-use of tomato crop area) and spraying (number of sprays per week) on the wild bee richness, wild bee abundance, honey bee abundance and functional guild on the 11 tomato crops. Significance levels are indicated by asterisks (* P < 0.5, ** P < 0.01, *** P < 0.001).

	Wild bee richness			Wild bee abundance			Honey bee abundance			Guild functional		
	Estimate	SE	P	Estimate	SE	P	Estimate	SE	P	Estimate	SE	P
(Intercept)	2.16	0.14	<0.001***	4.16	0.10	<0.001***	1.88	0.20	<0.001***	1.82	0.13	<0.001***
Area	-0.13	0.15	0.413	-0.05	0.09	0.589	-0.12	0.20	0.572	-0.14	0.11	0.220
Cultivar	-0.70	0.23	0.004**	-0.26	0.19	0.187	-0.27	0.35	0.447	-0.16	0.34	0.644
Fertilizers	0.02	0.12	0.854	0.23	0.07	0.001**	-0.47	0.15	0.002**	-0.05	0.10	0.605
Irrigation	0.03	0.28	0.920	-0.59	0.17	<0.001***	-0.69	0.38	0.080	-0.48	0.25	0.068
NNH	0.35	0.11	0.002**	0.15	0.08	0.080	-0.09	0.15	0.578	0.18	0.12	0.146
PCA1	0.02	0.12	0.854	0.23	0.07	0.001**	-0.47	0.15	0.003**	-0.05	0.10	0.605
PCA2	-0.38	0.12	0.003**	-0.24	0.09	0.009**	-0.04	0.16	0.803	-0.20	0.13	0.119
Reuse	-0.01	0.12	0.918	-0.05	0.09	0.617	-0.43	0.22	0.060	0.01	0.13	0.982
Spraying	-0.30	0.10	0.004**	-0.34	0.08	<0.001***	0.29	0.18	0.130	-0.21	0.12	0.082

The community was dominated by miners accounting for 72.50% of the total richness and 82.71% of the abundance. Facultatively social bees represented most of the species, but communal bees were more abundant (74.33%). Most species were a small size, representing 79% and 45% of the total abundance and richness, respectively.

3.2. Landscape configuration impact on bee community

The impact of the landscape configuration and farm management were tested in a single model but are presented in different sections in the results to facilitate understanding. Overall, the landscape cover (500- and 1000-meter radius) affected the bee community. The most parsimonious model showed that species richness of wild bees was negatively influenced by PCA2 ($\beta = -0.38$; $P = 0.003$) and positively influenced by NNH ($\beta = 0.35$; $P = 0.002$) (Table 2). The abundance of wild bees was negatively impacted by PCA2 ($\beta = -0.24$; $P = 0.009$) and positively influenced by PCA1 ($\beta = 0.23$; $P = 0.001$). Honey bee abundance was negatively associated with PCA1 ($\beta = -0.47$; $P = 0.003$).

Regarding functional guild groups where we used nesting habit, sociality, and size traits, we grouped the 40 species into 17 functional guilds (Table S4). There was no significant effect of land use on functional guild.

3.4. Farm management impact on bee community

Agricultural practices affected the bee community. Wild bees' richness was negatively associated with the Tottaly® cultivar ($\beta = -0.70$; $P = 0.004$) and spraying ($\beta = -0.30$; $P = 0.004$). Wild bee abundance was negatively impacted by manual irrigation ($\beta = -0.59$; $P < 0.001$) and spraying ($\beta = -0.34$; $P < 0.001$). Fertilizers had a positive effect on wild bee abundance ($\beta = 0.23$, $P = 0.001$) and a negative effect on

honeybee abundance ($\beta = -0.47$; $P = 0.002$). Farm management did not significantly affect the functional guilds. We calculated the weighted importance of all variables, and the three most important predictor variables were plotted for each of the response variables (Figure 3).

4. Discussion

In this study, we uncovered results showing that the community of bees was impacted at both local (farm management) and regional scales. Overall, while wild bees responded negatively to natural habitat loss and farm management intensification, honeybees responded positively to these variables. It has been reported that the impact of agricultural intensification on the bees' community in small farms is stronger on a local scale (Happe et al., 2008; Tangtorwongsakul et al., 2018). However, here we present results that support a more general hypothesis that both farm management (local) and landscape scales are important in structuring the community of bees in agricultural environments.

Contrary to our expectations, wild bee richness responded positively to the increase of NNH (nearest natural habitat), and a possible explanation was the high correlation found between NNH and the proportion of natural habitat. Several studies have shown that the abundance and richness of bees in agricultural areas tend to decrease with the increasing distance to natural and semi-natural habitats and that these sites play an important role as a refuge for pollinators (Carvalho et al., 2010; Joshi et al., 2016). However, bee community response to the proximity to natural habitats depends on the landscape configuration and composition (Jha and Kremen, 2013; Joshi et al., 2016). In our case, the increase of the proportion of natural area counteracts the distance to natural areas and enhances the abundance and richness of wild bees. Moreover, the NNH was less than 330 m in most of the sites, an accessible distance to

most bees (medium and small size), considering that the flight distance depends on the body size (Greeleaf et al., 2007; Zurbuchen et al., 2010).

The negative relationship between proportion of coffee crop (PCA2) and the richness and abundance of wild bees may be related to the high technological level adopted in this crop, characterized by the increase in the agrochemicals used in place of traditional methods of production (Jezzer et al., 2018). Intensification in the coffee crop tends to affect the community of bees and the maintenance of ecosystem services (Saturni et al., 2017) because it is associated with loss of nesting sites and low diversity of food sources (Hipólito et al., 2018). Based on our results, we suggest that management practices in adjacent crops can shift the responses of the community of bees in the target crop, in our case, the tomato. Thus, it is important to consider the management practices of the main non-target crops in the landscape when evaluating the community of bees in agricultural environments.

The increase in the abundance of wild bees in response to the reduction of semi-natural cover and the increase in natural cover (PCA1) reinforces the importance of natural areas as food resources and nesting sites for insects, which can ensure the presence of pollinators in agricultural areas (Cusser et al., 2016). For example, *Exomalpsis analis*, the most abundant species in this study, is likely to be favored in intensive agriculture environments due to its underground nesting habit that depends on the availability of bare soil (Potts et al., 2010; Ferreira et al., 2015). However, although *E. analis* does not depend on natural areas for nesting, this species depends on the food sources present in these areas.

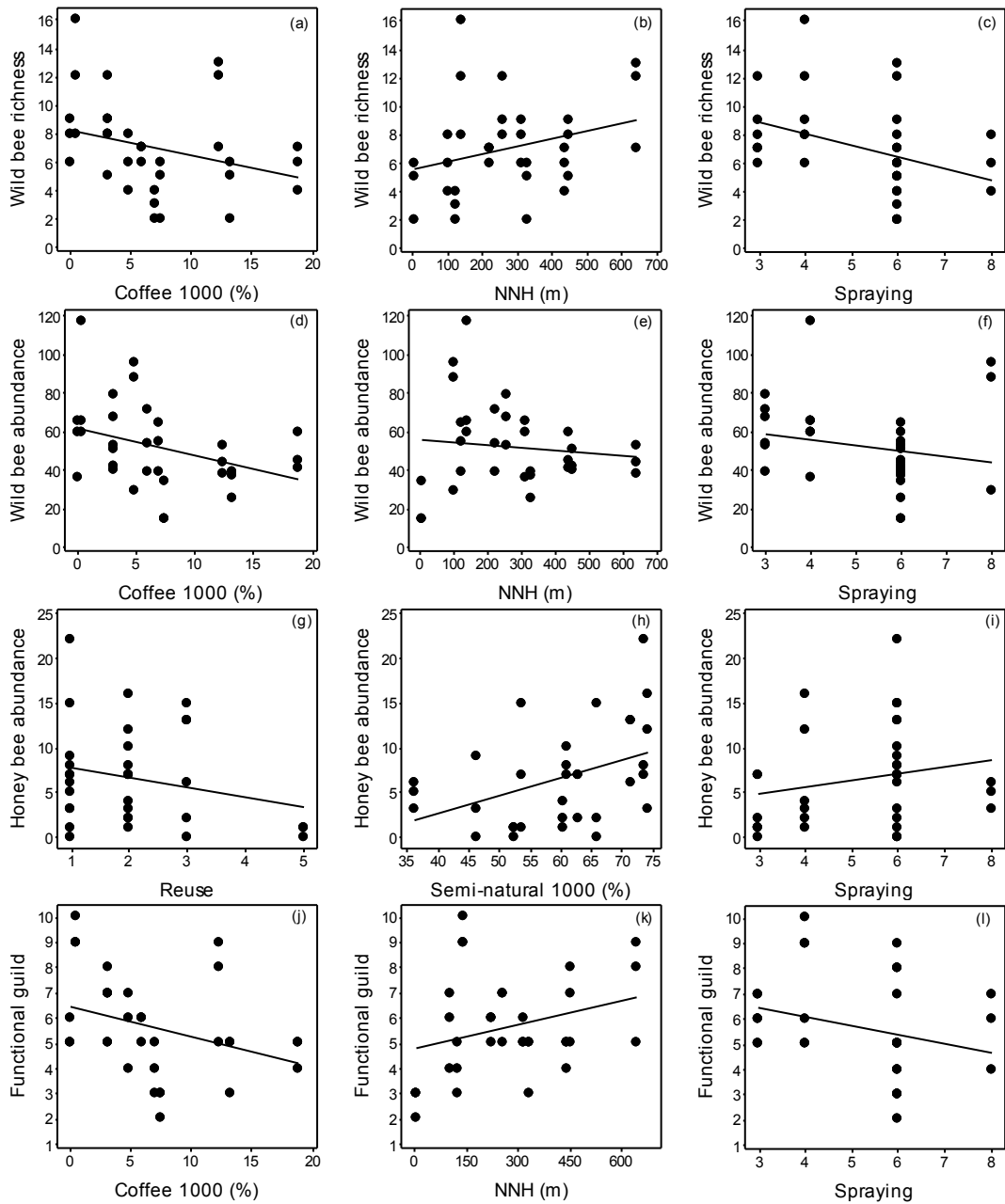


Figure 3. Relationship between the more important landscape and farm management variables and wild bee richness (a-c), wild bee abundance (d-f), honeybee abundance (g-i) and functional guild (j-l). Coffee 1000 (% of coffee land use in a 1000-m buffer), NNH (nearest natural habitat in meters), reuse (number of re-use of tomato crop area), semi-natural 1000 (% of semi-natural land use in a 1000-m buffer) and spraying (number of sprays per week).

On the other hand, the opposite response of the honeybee may be related to the opportunistic and generalist behavior of this species, making it less dependent on natural habitat than wild bees (Ferreira et al., 2006; Saturni et al., 2016). Semi-natural habitats were represented by eucalyptus forest and pastures and can be important food sources for the honeybee, favoring their presence in these areas. Our conclusions are supported by other studies that found a positive relationship between honeybee abundance and agricultural intensification (Marini et al., 2012; Petersen and Nault, 2014; Eeraerts et al., 2017). In addition, the introduction of hives or the presence of apiaries near the crop areas can increase the abundance of the honey bee (Martins et al., 2015; Gibbs et al., 2016), although none of the cases have been observed during our sampling, suggesting that individuals were from feral nests.

A possible explanation for the lack of effect of landscape configuration and farm management on the functional guild may be related to the low functional diversity of the bees' community in this region, since only a group, including small bees and miners, represented about 75% of the total abundance. However, classification of variables by order of importance showed that coffee, NNH, and spraying were the most important variables for a functional guild, with a response pattern similar to the richness of wild bees. Higher functional diversity can improve pollination services through resource use complementarity and has been argued to be the best index to evaluate a community of pollinators (Hoenh et al., 2008; Gagic et al., 2015; Martins et al., 2015; Blitzer et al., 2016). Although we were not able to find any significant relationships, we advocate the importance of considering functional diversity as an index to evaluate the response of the bee community to landscape changes, since different groups of bees tend to respond differently to these changes (Tangtorwongsakul et al., 2018).

The negative impact of the Tottaly® cultivar on bee richness may be related to some variation in the flower that has made it less attractive than the Aguamiel® cultivar. In agricultural systems, flower characteristics may vary depending on the cultivar, including the pollen availability (Malligher and Prassfka, 2017a), emission of volatile compounds (Mas et al., 2018; Twindle et al., 2018), and morphological characters (Portlas et al., 2018) that may affect the attractiveness of pollinators. Several studies have shown that variations of floral traits in different cultivars affect pollinator activity and may have an impact on crop yield (Courcelles et al., 2013; Carruthers et al., 2017; Mallinger and Prassfka, 2017b). Considering the importance of the variations in the floral characteristics to the attractiveness of the pollinators, it would be crucial for these traits to be included as targets in the current genetic breeding programs, mainly of crops highly dependent on biotic pollination (Palmer et al., 2009; Bailes et al., 2015; Mas et al., 2018).

The increase of the number of sprays per week negatively affected the abundance and richness of wild bees, and this can be explained by the high correlation between this variable and the use of insecticides. Pyrethroids, organophosphates, and neonicotinoids were the most used insecticide groups, and this may have negatively impacted the community of pollinators since these products are highly toxic to bees (Sena-Fernandes et al., 2008; Solomon et al., 2014). Recently, the number of studies evaluating the impact of pesticides on bees has increased, especially with neonicotinoids and managed species such as honey bees (Sena-Fernandes et al., 2008; Christen and Fent, 2017; Wood et al., 2018), bumble bees (Larson et al., 2013; Felthan et al., 2014; Potts et al., 2018), stingless bees (Dornelles et al., 2017; Moreira et al., 2018), and leafcutting bees (Artz et al., 2015; Piccolomini et al., 2018), demonstrating that the toxicity varies depending on the species and the group of insecticide (Arena and Sgolastra, 2014). Although honey bee-based toxicity may be predictive of field

effects on the wild bees' community, understanding the risks to wild bees requires more detailed studies on non-social bees (Park et al., 2015). However, despite the scarcity of knowledge in this area, the adoption of some management strategies related to the best spraying time and the use of selective insecticides could help to preserve the pollinators.

The negative impact of manual irrigation on wild bees' abundance may be related to the lower efficiency of this method compared to dripping. Directly, irrigation can affect pollinators through the influence of soil moisture on underground nesting species (Cane, 2008) or indirectly, when it affects the attractiveness of flowers and the quality of floral rewards (Julier and T'ai, 2009; Gillespie et al., 2015; Sardinãs et al., 2017). Here, it is possible that the community has been indirectly affected by the effect of the irrigation on floral characteristics and quality of rewards (Burkle and Runyon, 2016; Waser and Price, 2016; Gallagher and Campbell, 2017), since in both methods the water distribution is well located, which would hamper direct effects on nesting. In addition, there is a greater uniformity and efficiency of water distribution in drip irrigation (Baiamonte et al., 2015), which may have contributed to a higher physiological performance of the tomato and, consequently, greater attractiveness of the flowers.

The increased use of fertilizers positively impacted the abundance of wild bees and negatively impacted honey bee abundance. A possible explanation may be related to the pollen collection behavior of the bees in the tomato flower. Most wild bees perform flower vibration (poricidal dehiscence) to release the pollen hidden in the cone (anthers set), collecting the pollen deposited in their bodies (King and Buchmann, 2003) in a relatively short period of time (a few seconds). On the other hand, honey bees do not vibrate the flower and spend a relatively long time (> 30 seconds) manipulating the flower with its proboscis to obtain pollen (personal observation),

which could increase the oral exposure of this species to the foliar fertilizers used. Despite increasing concern about the increased use of agrochemicals, little is known about the impact of fertilizers on bees. A study evaluated the lethal and sublethal potential of two leaf fertilizers on a stingless bee species and found that copper sulfate fertilizer was highly toxic through oral exposure and that other micronutrients may have a sublethal effect (Rodrigues et al., 2016). Only one copper sulfate fertilizer was registered in this study. However, this same element was present in most compound fertilizers, usually associated with other micronutrients, which may explain the negative effects on the honey bee.

5. Implications and concluding remarks

The incompatibility between agricultural intensification and preservation of biodiversity has been the subject of many discussions recently; however, a scenario where both are harmonized is possible (Vieira et al., 2018). Faced with this issue, a major challenge has been the maintenance of natural areas in agricultural environments as a way to support a greater diversity. In Brazil, the Forest Code (FC) is the key piece of legislation regulating land use and management on private properties, requiring the preservation of natural areas in Legal Reserve and Areas of Permanent Preservation (Federal Law 12.727, October 17, 2012). Information about how much of the natural vegetation would be needed to maintain greater biodiversity in different contexts is scarce; however, some studies argue that the requirements of the current FC are not enough (Soares-Filho et al., 2014; Brancalion et al., 2016; Viera et al., 2018). Concerned about the loss of biodiversity in agricultural environments, many studies have proposed practices for the preservation of pollinators that include restoration of natural areas or the establishment of areas rich in flowers such as planting flowers at the edges of crops, wildflower strips, hedgerows, and fallow fields (Korpela et al.,

2013; Blaaw and Isaacs, 2014; Kremen and M’Gonigle, 2015; M’Gonigle et al., 2015; Scheper et al., 2015; Williams et al., 2015; McKechnie et al., 2017). However, Vieira et al. (2018) argued that the best way to conserve biodiversity in the short term is to avoid deforestation. On the other hand, the costs of establishment and maintenance of those habitats might discourage farmers from adopting these practices, but potential productivity gains from increased biodiversity could offset these costs.

Chemical control of pests and preservation of pollinators, especially bees, can be harmonized when pesticides are used correctly. For this purpose, the adoption of integrated pest management (IPM) would be an important initial step. This system, besides being efficient, takes into account economic, ecological, and social criteria. The use of IPM in tomato crops tends to reduce the frequency of spraying, preserving non-target organisms (Picanço et al., 2007) among which could be the pollinators. Considering the IPM principles, the following practices could be adopted to minimize the impact of insecticides on the bee population: use efficient and selective insecticides; application of pesticides at hours of lower bee activity (at the end of the day or early morning); and avoid spraying during the blooming stage of the crop.

Our results are particularly relevant considering the traditional discussion between agricultural intensification and biodiversity preservation. In this study, we provided important data that can guide pollinator management plans. Given the impact of both landscape context and agricultural practices on bees’ communities, we emphasize the importance of approaching both scales when designing management plans.

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Appendix A. Supplementary data

Table S1. Site, farmer, area (tomato size crop), cultivar (tomato variety: aguamiel vs. totally), irrigation (irrigation method: drip vs. manual), reuse (number of re-use of tomato crop area), spraying (number of sprays per week), insecticide (tomato crop insecticide input), fungicide (tomato crop fungicide input) and fertilizers (tomato crop fertilizer input) of 11 tomato fields in Zona da Mata region, Minas Gerais state, Brazil

Site	Farmer	Area (ha)	Cultivar	Irrigation	Reuse	Spraying	Insecticide			Fungicide			Fertilizer	
							CP*	AI**	CG***	CP	AI	CG	CP	NNP****
1	I	0.36	Aguamiel	Manual	3	6	10	10	9	9	10	7	7	10
2	II	0.56	Aguamiel	Drip	2	6	13	12	10	12	12	10	12	12
3	III	1.01	Totally	Manual	2	4	10	9	8	9	10	8	8	13
4	IV	1.86	Totally	Drip	1	6	19	18	14	10	11	9	5	5
5	V	1.78	Aguamiel	Drip	1	6	16	16	13	11	11	10	9	11
6	VI	0.6	Aguamiel	Drip	1	8	25	21	13	15	14	12	2	5
7	VII	1.29	Totally	Drip	1	6	15	13	11	11	8	6	4	5
8	VIII	0.16	Totally	Manual	5	3	18	16	12	12	12	8	7	12
9	IX	0.36	Aguamiel	Drip	3	6	8	8	7	6	7	6	6	6
10	X	0.18	Totally	Manual	2	3	11	11	9	16	16	14	7	12
11	XI	0.28	Aguamiel	Drip	2	4	13	12	11	11	11	8	3	3

*Number of commercial product; **Number of active ingredient; ***Number of chemical group; ****Number of nutrients provided.

Table S2. Site, Farmer, NNH (nearest natural habitat in meters), natural (natural habitat proportion), semi-natural (semi-natural habitat proportion), coffee (coffee field proportion), arable (arable soil proportion), water (water proportion), and building (building proportion) in 500- and 1000-meter radius

Site	Farmer	NNH (m)*	Buffer 500 m (%)						Buffer 1000 m (%)					
			Natural	Semi	Coffee	Arable	Water	Building	Natural	Semi	Coffee	Arable	Water	Building
1	I	5	13.64	55.77	6.95	17.20	6.02	0.41	12.75	65.82	7.37	11.97	1.70	0.37
2	II	331	3.97	59.74	19.95	9.56	5.23	1.54	16.03	61.08	13.19	5.44	3.10	1.15
3	III	314	7.24	63.81	0.00	15.85	2.60	10.47	15.68	60.41	0.00	11.89	3.19	8.83
4	IV	125	10.53	46.52	0.00	35.50	6.72	0.71	12.06	53.45	6.91	20.86	5.02	1.69
5	V	438	1.11	56.89	14.90	20.94	4.74	1.44	18.10	46.32	18.78	14.03	2.06	0.88
6	VI	103	23.17	30.24	7.63	27.49	7.44	4.02	21.40	36.26	4.85	30.18	4.36	2.93
7	VII	450	4.32	78.41	0.00	16.53	0.75	0.00	5.16	73.41	3.09	14.67	3.12	0.53
8	VIII	222	9.82	56.24	7.46	16.02	8.56	1.91	19.97	52.31	5.92	15.78	4.69	1.31
9	IX	643	0.93	70.59	12.58	7.04	0.47	8.38	3.94	71.23	12.32	5.88	0.77	5.84
10	X	256	11.32	46.61	5.90	29.35	5.42	1.41	12.99	62.71	3.01	17.39	2.92	0.96
11	XI	140	11.08	63.61	0.00	19.06	5.91	0.33	11.33	74.00	0.38	11.57	2.46	0.25

Table S3. Species, abundance, samples number (SN), nest (nesting class), social (social class), ITD (inter-tegula distance), size (size class) and functional guild (FG) of the bees collected in Cajuri and Coimbra, Minas Gerais state, Brazil

Species	Abundance	SN	Nest	Social	ITD (mm)	Size	FG
<i>Acamptopoeum prinii</i> (Holmberg, 1884)	1	1	Miner	Sol	2.10	Md	N
<i>Apis mellifera</i> Linnaeus, 1758	216	30	AGC	HE	2.80	Md	E
<i>Augochlora</i> sp.1	1	1	Miner	FS	1.54	Sm	K
<i>Augochloropsis argentina</i> (Friese, 1908)	14	9	Miner	FS	1.63	Sm	K
<i>Augochloropsis brachycephala</i> Moure, 1943	19	13	Miner	FS	1.70	Sm	K
<i>Augochloropsis cleopatra</i> (Schrottky, 1902)	10	6	Miner	FS	2.18	Md	J
<i>Augochloropsis cupreola</i> (Cockerell, 1900)	13	9	Miner	FS	1.90	Sm	K
<i>Augochloropsis illustris</i> (Vachal, 1903)	1	1	Miner	FS	1.48	Sm	K
<i>Augochloropsis smithiana</i> (Cockerell, 1900)	8	6	Miner	FS	2.27	Md	J
<i>Augochloropsis</i> sp.1	26	12	Miner	FS	2.28	Md	J
<i>Augochloropsis</i> sp.2	10	9	Miner	FS	2.06	Md	J
<i>Augochloropsis</i> sp.3	1	1	Miner	FS	2.12	Md	J
<i>Augochloropsis wallacei</i> (Cockerell, 1900)	4	4	Miner	FS	1.65	Sm	K
<i>Bombus (Fervidobombus) morio</i> (Swederus, 1787)	53	17	AGC	PE	5.03	Lg	I
<i>Bombus (Fervidobombus) pauloensis</i> Friese, 1913	8	6	AGC	PE	3.80	Md	H
<i>Centris (Centris) aenea</i> Lepeletier, 1841	1	1	Miner	Sol	5.54	Lg	M
<i>Centris (Hemisiella) tarsata</i> Smith, 1874	4	4	AGC	Sol	3.40	Md	L
<i>Dialictus travassosi</i> (Moure, 1940)	1	1	Miner	FS	1.09	Sm	K

<i>Euglossa</i> sp.	2	2	AGC	Sol	3.57	Md	L
<i>Eulaema (Apeulaema) nigrata</i> Lepeletier, 1841	5	3	Soil	Com	5.90	Lg	D
<i>Exomalopsis (Exomalopsis) analis</i> Spinola, 1853	1288	33	Miner	Com	1.91	Sm	A
<i>Exomalopsis (Exomalopsis) auropilosa</i> Spinola, 1853	113	20	Miner	Com	1.67	Sm	A
<i>Exomalopsis (Exomalopsis) fernandoi</i> Moure, 1990	6	6	Miner	Com	1.76	Sm	A
<i>Exomalopsis (Exomalopsis) fulvofasciata</i> Smith, 1879	3	2	Miner	Com	2.37	Md	B
<i>Exomalopsis (Exomalopsis) minor</i> Schrottky, 1910	9	7	Miner	Com	1.64	Sm	A
<i>Exomalopsis (Exomalopsis) tomentosa</i> Friese, 1899	2	2	Miner	Com	1.67	Sm	A
<i>Exomalopsis (Exomalopsis) ypirangensis</i> Schrottky, 1910	6	4	Miner	Com	1.60	Sm	A
<i>Melipona (Eomelipona) bicolor</i> Lepeletier, 1836	1	1	AGC	HE	3.15	Md	E
<i>Melissoptila</i> sp.	1	1	Miner	Sol	1.60	Sm	O
<i>Oxaea flavescens</i> Klug, 1807	4	3	Miner	Com	5.19	Lg	C
<i>Parapsaenythia serripes</i> (Ducke, 1908)	6	5	Miner	Sol	1.90	Sm	O
<i>Paratetrapedia</i> sp.	1	1	Miner	Sol	1.88	Sm	O
<i>Psaenythia</i> sp.	4	3	Miner	Sol	2.50	Md	N
<i>Pseudaugochlora callaina</i> Almeida, 2008	1	1	Miner	FS	2.26	Md	J
<i>Pseudaugochlora graminea</i> (Fabricius, 1804)	13	10	Miner	FS	2.46	Md	J
<i>Schwarziana quadripunctata</i> (Lepeletier, 1836)	1	1	Soil	HE	1.70	Sm	G
<i>Thygater (Thygater) analis</i> (Lepeletier, 1841)	31	15	Miner	Sol	3.31	Md	N
<i>Trigona spinipes</i> (Fabricius, 1793)	41	8	Exp	HE	1.72	Sm	F
<i>Xylocopa (Neoxylocopa) suspecta</i> Moure & Camargo, 1988	2	2	Carp	FS	6.65	EL	P
<i>Xylocopa (Schonherria) muscaria</i> (Fabricius, 1775)	1	1	Carp	FS	5.17	Lg	Q

Nesting class: **AGC** = above-ground cavity (bees build their nests in different pre-existing cavity types above the ground); **Carp** = carpenter (bees make holes in the wood to build their nests); **Exp** = exposed (bees construct exposed nests of plant fiber); **Miner** = miners (bees dig holes in the ground to build their nests); **Soil** = soil cavity (bees build their nests in pre-existing cavities in the ground). Social class: **Com** = communal (when two or more females use the same nest, but each makes and provisions her own cells and lays an egg in each of them); **FS** = facultatively social (species exhibit behavioral plasticity in response to changes in ecological conditions); **HE** = highly eusocial (bees always living in colonies and new colonies are established socially by groups or swarms); **PE** = primitively eusocial (bees may live in small colonies, mostly started by single females working as solitary individuals performing all necessary functions of nest construction, foraging, provisioning cells or feeding larvae progressively, and laying eggs); **Sol** = solitary (bees construct their own nest and provide food for the offspring with the help of other bees and usually die or leave before the offspring mature). Size class: **Sm** = small (< 2 mm); **Md** = medium (2.1 - 4.0 mm); **Lg** = large (4.1 - 6.0 mm); **EL** = extra-large (> 6.1 mm). Nesting class and social class were based on the following studies: Michner and Kerfoot (1967); Rozen Jr. (1974, 1984); Silva et al. (2001); Coelho et al. (2002); Aguiar and Gaglianone (2003); Aguiar and Garófalo (2004); Michener (2007); Schwarz et al. (2007); Cardinal and Danforth (2011); Gibbs et al. (2012); Danforth et al. (2013); Oliveira et al. (2015); Rehan and Toth (2015); Harpur et al. (2017); Shell and Rehan (2017); Toth and Rehan (2017).

Table S4. Functional guild based in nest class, social class, size, and size class

Functional guild	Nest class	Social class	Size (mm)	Size class
A	Miner	Communal	< 2	Small
B	Miner	Communal	2.1 - 4.0	Medium
C	Miner	Communal	4.1 - 6.0	Large
D	Soil cavity	Communal	4.1 - 6.0	Large
E	Above-Ground Cavity	Highly Eusocial	2.1 - 4.0	Medium
F	Exposed	Highly Eusocial	< 2	Small
G	Soil cavity	Highly Eusocial	< 2	Small
H	Above-Ground Cavity	Primitively Eusocial	2.1 - 4.0	Medium
I	Above-Ground Cavity	Primitively Eusocial	4.1 - 6.0	Large
J	Miner	Facultatively social*	2.1 - 4.0	Medium
K	Miner	Facultatively social*	< 2	Small
L	Above-Ground Cavity	Solitary	2.1 - 4.0	Medium
M	Miner	Solitary	4.1 - 6.0	Large
N	Miner	Solitary	2.1 - 4.0	Medium
O	Miner	Solitary	< 2	Small
P	Carpenter	Facultatively social*	> 6.1	Extra-large
Q	Carpenter	Facultatively social*	4.1 - 6.0	Large

*Facultatively social is not a distinct social category, but rather indicates a species' capacity for plasticity in its social organization.

Table S5. List of agrochemicals utilized in 11 tomato fields in Cajuri and Coimbra, Minas Gerais state, Brazil, by commercial name, common name, chemical group, manufacturer and farmer (number of farmers that used the commercial product)

Commercial name (®)	Common name	Chemical group	Manufacturer	Farmer
Insecticide				
Abamectin Nortox	Abamectin	Avermectins	Nortox	1
Abamex	Abamectin	Avermectins	Nufarm	1
Actara 250 WG	Thiamethoxam	Neonicotinoids	Syngenta	2
Agritoato	Dimethoate	Organophosphates	Nufarm	1
Akito	<i>beta</i> -Cypermethrin	Pyrethroids	Arysta	1
Applaud 250	Buprofezin	Buprofezins	Arysta	6
Assist	Mineral oil	Aliphatic hydrocarbons	Basf	1
Atabron 50 EC	Chlorfluazuron	Benzoylureas	Arysta	4
Bac control WP	Bacillus thuringiensis	Biological	Vectorcontrol	7
Bazuka	Methomyl	Carbamates	FMC	4
Belt	Flubendiamide	Diamides	Bayer	1
Benevia	Cyantraniliprole	Diamides	Du pont	4
Capataz BR	Chlorpyrifos	Organophosphates	Ouro fino	2
Certero	Triflumuron	Benzoylureas	Bayer	2
Connect	Imidacloprid/ <i>beta</i> -Cyfluthrin	Neonicotinoids/Pyrethroids	Bayer	2
Cyptrin 250 EC	Cypermethrin	Pyrethroids	Nufarm	1
Danimen 300 EC	Fenpropathrin	Pyrethroids	Iharabras	7
Decis 25 EC	Deltamethrin	Pyrethroids	Bayer	1
Delegate	Spinetoram	Epinosins	Dow agrosociences	4
Dimexion	Dimethoate	Organophosphates	Cheminova	3

Dimilin	Diflubenzuron	Benzoylureas	Basf	1
Dipel	Bacillus thuringiensis	Biological	FMC	7
Evidence 700 WG	Imidacloprid	Neonicotinoids	Bayer	1
Fastac 100	<i>alpha</i> -Cypermethrin	Pyrethroids	Basf	7
Granary	Imidacloprid	Neonicotinoids	FMC	1
Haiten	Phenol ether	Nonylphenols	Arysta	2
Imunit	<i>alpha</i> -Cypermethrin/Teflubenzuron	Pyrethroids/Benzoylureas	Basf	1
Karate Zeon 50 EC	<i>lambda</i> -Cyhalothrin	Pyrethroids	Syngenta	1
Klorpan 480 EC	Chlorpyrifos	Organophosphates	Nufarm	5
Kraft 36 EC	Abamectin	Avermectins	Cheminova	9
Lannate BR	Methomyl	Carbamates	Du pont	3
Larvin 800 WG	Thiodicarb	Carbamates	Bayer	2
Lorsban 480 BR	Chlorpyrifos	Organophosphates	Dow agrosiences	1
Milbeknock	Milbemectin	Milbemycins	Iharabras	1
Mirza 480 SC	Triflumuron	Benzoylureas	FMC	1
Mospilan	Acetamiprid	Neonicotinoids	Sipcam	7
Natur'l óleo	Vegetable oil	Fatty acids esters	Stoller	1
Nomolt 150	Teflubenzuron	Benzoylureas	Basf	4
Nufos 480	Chlorpyrifos	Organophosphates	Cheminova	2
Oberon	Spiromesifen	Tetronic and Tetramic acid derivatives	Bayer	5
Orthene 750 BR	Acephate	Organophosphates	Arysta	3
Pirate	Chlorfenapyr	Pyrroles	Basf	7
Polytrin	Profenofos/Cypermethrin	Organophosphates/Pyrethroids	Syngenta	1
Premio	Chlorantraniliprole	Diamides	Du pont	7

Provado 200 SC	Imidacloprid	Neonicotinoids	Bayer	2
Rumo	Indoxacarb	Oxadiazines	FMC	1
Safety	Etofenprox	Pyrethroids	Arysta	1
Thiobel 500	Cartap	Nereistoxin analogues	Arysta	3
Trigard 750 WP	Cyromazine	Cyromazines	Syngenta	10
Turbo	<i>beta</i> -Cyfluthrin	Pyrethroids	Bayer	2
Veget'oil	Vegetable oil	Fatty acids esters	Oxiquímica	3
Warrant	Imidacloprid	Neonicotinoids	Bayer	1
Fungicide				
Academic	Cymoxanil /Mancozeb	Cyanoacetamides/Dithiocarbamates	FMC	7
Cabrio Top	Metiram /Pyraclostrobin	Dithiocarbamates/methoxycarbamates	Basf	6
Cantus	Boscalid	Carboxamides	Basf	2
Carbendazim Nortox	Carbendazim	Benzimidazoles	Nortox	1
Cercobin 700 WP	Thiophanate-methyl	Thiophanates	Sipcam	5
Cimox WP Helm	Cymoxanil /Mancozeb	Cyanoacetamides/Dithiocarbamates	Scam SPA	6
Cobre Atar BR	Cuprous oxide	Inorganic	Syngenta	4
Consento	Propamocarb/Fenamidone	Carbamates/Imidazolinones	Bayer	5
Curzate BR	Cymoxanil /Mancozeb	Cyanoacetamides/Dithiocarbamates	Dow agrosiences	5
Daconil BR	Chlorothalonil	Chloronitriles	Arysta	5
Emzeb 800 WP	Mancozeb	Dithiocarbamates	Arysta	4
Forum	Dimethomorph	Cinnamic acid	Basf	3
Frownicide 500 SC	Fluazinam	2,6-dinitroanilines	Syngenta	2
Galben-M	Benalaxyl /Mancozeb	Acylalanines/Dithiocarbamates	FMC	1
Impact 125 SC	Flutriafol	Triazoles	Cheminova	1

Kasumin	Kasugamycin	Hexopyranosyl antibiotic	Arysta	5
Kocide WDG Bioactive	Copper hydroxide	Inorganic	Du pont	3
Kumuluf DF	Sulphur	Inorganic	Basf	2
Manzate WG	Mancozeb	Dithiocarbamates	Dow agrosiences	6
Metil Tiofam	Thiophanate-methyl	Thiophanates	Sipcam	1
Midas BR	Famoxadone/Mancozeb	Oxazolidine-diones/Dithiocarbamates	Sipcam	1
Mythos	Pyrimethanil	AnilinoPyrimidines	Bayer	2
Orthocide 500	Captan	Phthalimides	Arysta	7
Polyram DF	Metiram	Dithiocarbamates	Basf	8
Proplant	Propamocarb	Carbamates	Chimac	3
Recop	Copper oxychloride	Inorganic	Atar	8
Revus	Mandipropamid	Mandelic acid amides	Syngenta	5
Ridomil Gold MZ	Metalaxyl-M/Mancozeb	Acylalanines/Dithiocarbamates	Dow agrosiences	5
Score	Difenoconazole	Triazoles	Syngenta	1
Sumiguard 500 WP	Procymidone	Dicarboximides	Arysta	2
Sumilex 500 PM	Procymidone	Dicarboximides	Sipcam	4
Supera	Copper hydroxide	Inorganic	Sipcam	2
Fertilizer				
Acadian	N-P-K	Inorganic	Acadian	1
Amino Plant	N	Inorganic	Amino Plant	1
Betagrif Crystal	Mineral	Inorganic	Crystal	2
Biozyme TF	Mg-B-Cu-Fe-Mn-Mo-Zn	Inorganic	Arysta	1
Ca EDTA	Ca	Inorganic	Procure	2
Cal Super	N-P-K-Ca	Inorganic	Agrichem	1

Codamax	N-P	Inorganic	Proplanta	1
Copper Crop	Cu	Inorganic	Alltech	2
Crop Set	N-K	Inorganic	Alltech	1
Crystal Spores	Mineral	Inorganic	Crystal	1
Cupratil	Cu-Zn	Inorganic	Kifol	1
Ecolife	N-C	Inorganic	Quinabra	1
Fera avant	N-P-K	Inorganic	Avant	1
Flow Boro	B	Inorganic	No information	1
Hortumus	K	Inorganic	Arvensis	2
Hydrofert	N-P-K	Inorganic	Biolchim	1
K-Fol	Ca-B	Inorganic	GBM	1
K-Tionic	N-K	Inorganic	GBM	1
Librel Ca	Ca	Inorganic	Ciba	1
Micros Comnagro	Mg-B-Cu-Fe-Mn-Mo-Zn	Inorganic	Comnagro	1
Niphokam	N-P-K	Inorganic	Quimifol	4
Nutrioxi	K-S-Mg-B-Mn-Mo-Zn	Inorganic	Nutrioxi	1
P-Count-K	P-K	Inorganic	No information	2
Primefol Boro Fit	B	Inorganic	Quimifol	1
Quimifol Boro L	B	Inorganic	Quimifol	2
Quimifol Cálcio F	Ca	Inorganic	Quimifol	2
Quimifol Florada	Ca-B	Inorganic	Quimifol	1
Quimifol P30W	N-P-Mg	Inorganic	Quimifol	1
Quimifol pHFós	N-P	Inorganic	Quimifol	2
Qumifol Titanium	N	Inorganic	Quimifol	1

Raizal	N-P-K	Inorganic	GBM	1
Redufol Max	N	Inorganic	Ubyfol	2
Restorer H	N-P-K-B-Cu-Fe-Mn-Mo-Zn-C	Inorganic	Biolchim	1
Rizamina 420	N-P-K-Mg-S-B-Cu-Fe-Zn	Inorganic	Biolchim	1
Spray Dunger	N-P-K-Mg-Ca-S-B-Cu-Fe-Mn-Mo-Zn	Inorganic	Biolchim	3
Sprintalga TS	N-Mo-C	Inorganic	Biolchim	1
Stimulate	Hormone	Inorganic	Stoller	1
Sumo-K	Mn-C	Inorganic	Apex	1
Torped Gold	N-P-K-B-Cu-Fe-Mn-Mo-Zn-C	Inorganic	Oxiquímica	1
Triamin Plus	B-Cu-Mn-Mo-Zn	Inorganic	Arvensis	1
Triunfo Flex	N-P-K	Inorganic	Produquímica	1
Ubyfol Disperse Ultra	N-P	Inorganic	Ubyfol	1
Ubyfol K 50 + S	K-S	Inorganic	Ubyfol	1
Ubyfol Kymon	N-K	Inorganic	Ubyfol	2
Ubyfol MS-2	B-Cu-Fe-Mn-Mo-Zn	Inorganic	Ubyfol	1
Ubyfol MS-Boro	B	Inorganic	Ubyfol	3
Ubyfol N-32	N	Inorganic	Ubyfol	1
Ubyfol S-CaB	Ca-S-B	Inorganic	Ubyfol	3
Ubyphos + K	K-P	Inorganic	Ubyfol	2
VitaliK	Ca-Mn-Cu-Zn	Inorganic	Arysta	1

Table S6. Pearson correlation coefficient of land use (500- and 1000-m buffer) and farm management. NNH = nearest natural habitat, N = natural, S = semi-natural, C = coffee, A = arable, W = water, B = building, Area = tomato size crop, reuse = number of re-use of tomato crop area, SW = number of sprays per week, I = insecticide, F = fungicida, Ft = fertilizers. Significance levels are indicated by asterisks (* 0.5, ** 0.01, *** <0.001)

	N500	S500	C500	A500	W500	B500	N1000	S1000	C1000	A1000	W1000	B1000	Area	Reuse	SW	I	F	Ft
NNH	-0.822 ***	0.622 ***	0.362 *	-0.552 ***	-0.792 ***	0.422 *	-0.492 **	0.432 *	0.432 *	-0.512 **	-0.482 **	0.362 *	0.162	-0.032	0.012	-0.382 *	-0.372 *	0.182
N500	-	-0.782 ***	-0.342	0.562 ***	0.652 ***	-0.152	0.482 **	-0.452 **	-0.512 **	0.782 ***	0.492 **	-0.142	-0.302	-0.052	0.172	0.512 **	0.532 **	-0.432 *
S500		-	-0.072	-0.702 ***	-0.752 ***	0.122	-0.692 ***	0.792 ***	0.022	-0.772 ***	-0.512 **	0.132	0.042	0.162	-0.232	-0.642 ***	-0.752 ***	0.102
C500			-	-0.452 **	0.042	0.022	0.252	-0.252	0.842 ***	-0.362 *	-0.332	-0.112	-0.152	0.152	0.282	-0.072	0.122	0.552 ***
A500				-	0.512 **	-0.362 *	0.282	-0.472 **	-0.262	0.802 ***	0.592 ***	-0.272	0.372*	-0.462 **	0.042	0.612 ***	0.582 ***	-0.332
W500					-	-0.452 **	0.762 ***	-0.592 ***	-0.062	0.512 **	0.612 ***	-0.452 **	-0.192	0.242	-0.122	0.612 ***	0.562 ***	0.022
B500						-	-0.042	-0.042	-0.092	-0.182	-0.212	0.982 ***	-0.092	0.132	-0.022	-0.362 *	-0.262	0.162
N1000							-	-0.832 ***	0.092	0.472 **	0.532 **	-0.052	-0.032	0.092	-0.022	0.572 ***	0.582 ***	0.332
S1000								-	-0.282	-0.702 ***	-0.552 ***	-0.032	-0.282	0.172	-0.352 *	-0.782 ***	-0.512 **	-0.092
C1000									-	-0.302	-0.342	-0.172	0.312	-0.042	0.412 *	0.072	-0.132	0.402 *
A1000										-	0.672 ***	-0.132	0.202	-0.352 *	0.282	0.812 ***	0.562 ***	-0.482 **
W1000											-	-0.112	0.232	-0.072	-0.062	0.742 ***	0.412 *	-0.132
B1000												-	0.022	0.062	-0.032	-0.332	-0.292	0.142
Area													-	-0.672 ***	0.382 *	0.392 *	-0.202	-0.052
Reuse														-	-0.542 **	-0.322	-0.202	0.322
SW															-	0.412 *	-0.142	-0.342
I																-	0.392 *	-0.382 *
F																	-	0.122

Table S7. PCA of land use classes in 500* and 1000** buffer

	PC1*	PC2*	PC3*	PC4*	PC5*	PC6*
Standard deviation	15.0333	7.8268	4.72919	3.40825	1.48214	0.004479
Proportion of Variance	0.6987	0.1894	0.06915	0.03591	0.00679	0.000000
Cumulative Proportion	0.6987	0.8881	0.95729	0.99321	1.00000	1.000000

	PC1**	PC2**	PC3**	PC4**	PC5**	PC6**
Standard deviation	13.3137	6.6089	3.76516	2.82230	0.88999	0.01528
Proportion of Variance	0.7268	0.1791	0.05813	0.03266	0.00325	0.00000
Cumulative Proportion	0.7268	0.9060	0.96409	0.99675	1.00000	1.00000

Table S8. Weights of land use classes on axis 1 and 2

Land use classe	PC1 (500)	PC2 (500)	PC1 (1000)	PC2 (1000)
Natural	-0.8298312	0.1405198	0.84221096	0.05824804
Semi-Natural	0.9740183	0.2194439	-0.99369823	-0.09693891
Coffee	0.1310007	-0.9621100	0.19349271	0.93837691
Arable	-0.8245255	0.4362158	0.76372800	-0.56784473
Water	-0.7498488	-0.1526132	0.59754875	-0.45178008
Buldging	0.2143510	-0.1473108	-0.01674611	-0.02961944

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CHAPTER 3

Effects of irrigation method on pollination and pollinators in an open-field tomato crop

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Abstract

Tomato is one of the highest water-demanding crops, and the use of different irrigation systems and different water management strategies may affect crop yield. Despite the fact that tomato is a self-fertilizing (autogamous) plant, research has shown that bees ("buzz pollination") improve yield and fruit quality. Here, we assess the effect of irrigation method on bee visitation and pollination on flowers of the tomato crop. The study was conducted from July to October 2017 in Minas Gerais state, Brazil, using 6 plots with 150 tomato plants. Three plots received water by sprinkler irrigation and the other 3 by drip irrigation, in a randomized complete block design. Bees were sampled from plants watered with both irrigation systems to evaluate bee abundance and richness. Fruit set and fruit weight were compared between irrigation systems. Bee assemblage abundance and species richness were not affected by the irrigation method, and the mechanical impact of the sprinkler did not provide the same level of bee pollination. As pollination and pollinators were not negatively affected, the best irrigation method should take into account agronomic aspects such as financial viability, and water use efficiency. The drip irrigation system seems to be the most effective because it exhibits these characteristics, even though it may have a higher cost.

Keywords: Apidae, bees, drip irrigation, species richness, sprinkler irrigation, tomato

1. Introduction

Tomato is one of the world's most important and widespread crops, with a global production in 2016 of about 177 million tons (FAOSTAT, 2018). Tomato production depends on several factors including water management and environmental factors, such as crop variety, soil type, soil moisture, temperature, humidity, and rainfall. Considering that this crop has a high water requirement, especially in the blooming and fruit development stages, irrigation is necessary to achieve production quality (Cantore et al., 2016).

Tomato irrigation can be carried out using sprinkler, drip, or furrow irrigation systems. The sprinkler irrigation system is characterized by wetting the entire plant and is performed with sprinklers or a center pivot. On the other hand, drip irrigation is characterized by delivering water directly to the plant root zone, which can be done with drip emitters. This latter method has high efficiency of water application (Gerçek et al., 2017), and plays an important role in water conservation as well as allowing the application of fertilizers via an irrigation system (Lu et al., 2016). Efficient water use is one of the main concerns today, especially in arid and semi-arid regions. Thus, different irrigation systems and management strategies have been introduced to the tomato crop to improve water use efficiency in irrigation (Al Ghobari et al., 2016; Gerçek et al., 2017). However, irrigation practice changes may influence other factors that affect crop yield, such as pollinators and pollination.

Seventy-five per cent of global crops benefit from animal pollination to some degree (Klein et al., 2007). Wild bees are the most efficient pollinators of cultivated plants (Garibaldi et al. 2013), improving yield, quality, shelf life, and food commercial value (Bartomeus et al., 2014; Brittain et al., 2014). Among these crops, the tomato has been studied, both in the open-field and greenhouse. The tomato is autogamous plant; however, its flower requires external stimulus for the release of pollen and

fertilization, because its anthers are poricidal (the pollen is released via small apical pores or slits). Although the wind may promote self-pollination in tomato (Thorp, 2000), studies have shown that sonicating bees (buzz pollination syndrome) improve yield and fruit quality (Greenleaf and Kremen, 2006; Bartelli and Nogueira-Ferreira, 2014; Deprá et al., 2014).

Bees can be impacted directly when the irrigation affects nesting, mainly of underground and solitary bee species (Cane, 2008), or indirectly, when the flower's attractiveness is affected by wetting or soil water availability (Gillespie et al., 2015; Sardinãs et al., 2017). Here, we believe that the irrigation system may affect tomato pollination in two ways: when sprinkling is used, the impact of water droplets on the plant may lead to the release of pollen from the anthers sufficient for fertilization and formation of heavier fruits, or, indirectly, affecting the presence of pollinators in the crops, mainly wild bees (Gallagher and Campbell 2017).

Considering that bees benefit the tomato and that the crop is carried out under different watering systems including sprinkler irrigation and drip irrigation, this work seeks to determine if the irrigation method affects pollinator presence and pollination in the tomato crop. Two hypotheses were formulated: a) the impact of water droplets from the sprinkling system is sufficient to allow the same level of pollination performed by bees, and b) abundance and richness of the bee community in the dripping system is greater than that in sprinkling.

2. Material and Methods

2.1. Study site

This study was conducted from July to October 2017 at the Instituto Federal do Norte de Minas - Campus Januária (15.4877777S, 44.3619444W), Minas Gerais state, Brazil, in a region characterized by transition areas between the Cerrado

(Brazilian Savannah) and the Caatinga (Dry Forest). The climatic type of the region is Aw according to the classification of Köepen, with a dry winter and rainy summer.

2.2. Tomato crop

Six plots (8 x 10 m), each with 150 tomato plants per plot, were used. Tomato hybrid H9553® (Heinz Seeds) was grown in 1 m spacing between rows and 0.5 m between plants. Fertilization was carried out according to the soil analysis recommendations. Spraying of insecticides, fungicides, and leaf fertilizers was performed twice per week during the tomato crop cycle, except for the blooming stage.

2.3. Irrigation systems

Three plots were irrigated by sprinkling and the other 3 by dripping; the plot arrangement was a randomized complete block design. In the sprinkler treatment, we used Fabrimar "Pingo Set 30"® sectoral sprinklers with a flow rate of 0.5 m³/h (red nozzle), with an operating pressure of 0.196 MPa, and a 90° operating angle. In the drip treatment, Azud ASR R70® adjustable drippers were used, with a flow rate of 0.07 m³/h and an operating pressure of 0.098 MPa. For this treatment, one dripper per plant was inserted in polyethylene tubing. The main pipes of the two irrigation systems were independent, and irrigations were performed daily after 2 p.m. following management recommendations in order to maintain equivalent soil moisture levels according to the crop water requirements and soil type.

2.4. Sampling bees

Bees visiting tomato flowers were collected in Aug and Sep 2017. We walked transects along all rows of each plot, and each plot was sampled between 8 a.m. and 1 p.m. for 5 min/h, on dry and warm days (> 20 °C) with low wind speeds. Temperature and humidity were recorded every hour with a TFA® Wireless weather station Diva

Plus. All bees that contacted the tomato flowers were sampled with a sweep net (20 cm in diameter) and were subsequently killed with ethyl acetate in a killing jar. Bee samples of each plot and sampling period were pooled, sorted, pinned, labeled, and subsequently identified according to species level by professor Eduardo Andrade Botelho de Almeida (FFCLRP-USP). Voucher specimens were deposited at the Entomological Museum of the Universidade Federal de Viçosa (UFV) and in the bee collection of the Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto (FFCLRP-USP).

2.5. Pollination tests

Three pollination treatments were installed in each plot to evaluate if the irrigation system influenced pollination: bagged (control), open pollination, and open + mechanical pollination. These treatments were carried out in the 6 central rows of each plot. In each row, 3 plants were randomly selected for each treatment, totaling 18 plants per treatment. The first flower cluster emitted by of each plant was selected for the application of treatments. In the bagged treatment, the flower cluster was completely wrapped with an organza bag (10 x 15 cm) before the first flower's anthesis and was removed after the senescence of the last flower. In the open pollination treatment, the flowers remained unbagged for visitation by bees. In the treatment of open + mechanical pollination, the flowers were vibrated daily for 5 seconds (Palma et al., 2008) with a vibrator made with an electric toothbrush, and they remained unbagged for visitation by bees. The vibration (open + mechanical pollination) was performed from 11:00 a.m. to 12:00 p.m., from anthesis to flower senescence. In all treatments, the fruit set was evaluated in all clusters identified. The first 4 fruits (basal) of each cluster were weighed at the time of harvest to evaluate individual fruit weight. The tomato yield was evaluated in the 10 central plants of the penultimate row of each

plot as possible support for the discussion. A screen was installed around the plots to block the wind and minimize its influence on pollination. For this purpose, was used a shading screen (black) of 1.5 m of high, installed at a distance of 4 m from plot margins.

2.6. Statistical analyses

Values of all the response variables were analyzed with Shapiro-Wilk and Bartlett tests to verify the normality and homogeneity of variances, respectively. The Mann-Whitney U test was used to determine if there were differences in bee abundance and bee species richness between the irrigation systems. Fruit set and fruit weight of the pollination treatments were analyzed by ANOVA ($\alpha = 0.05$) and, when significant, with the Tukey HSD test ($\alpha = 0.05$). An unpaired t -test ($\alpha = 0.05$) was used to test whether there were differences in fruit set and fruit weight between the 2 irrigation systems. In addition, the unpaired t -test ($\alpha = 0.05$) was used to evaluate whether there was a difference in mean productivity between the two irrigation systems. The analyses were conducted in R (R-Core-Team, 2017).

3. Results

Two hundred and seventy-three bees of 18 species were collected (Table 1). One hundred and forty-two individuals of 12 species were collected in the sprinkling system, along with 134 individuals of 16 species in the dripping system. The abundance and species richness did not differ between the irrigation systems ($W = 40$; $P > 0.05$ and $W = 26$; $P > 0.05$, respectively). There were also no differences in the abundance of the principal genera collected (*Augochloropsis*, *Bombus*, *Centris*, and *Exomalopsis*).

Table 1. Family, species, abundance, and richness of bees collected in the tomato crop in sprinkler and drip irrigation systems

Family/Species	Sprinkler	Drip	Total
Apidae			
<i>Apis mellifera</i> Linnaeus, 1758	3	-	3
<i>Bombus (Fervidobombus) brevivillus</i> Franklin, 1913	51	23	74
<i>Centris (Hemisiella) tarsata</i> Smith, 1874	5	6	11
<i>Centris (Hemisiella) trigonoides</i> Lepeletier, 1841	12	11	23
<i>Centris (Trachina) fuscata</i> Lepeletier, 1841	2	2	4
<i>Euglossa</i> sp.	1	-	1
<i>Exomalopsis (Exomalopsis) analis</i> Spinola, 1853	15	20	35
<i>Melipona (Melikerria) quinquefasciata</i> Lepeletier, 1836	-	1	1
<i>Trigona recursa</i> Smith, 1863	10	26	36
<i>Trigona spinipes</i> (Fabricius, 1793)	2	1	3
Halictidae			
<i>Augochlora (Oxystoglossella) sp.2</i>	-	1	1
<i>Augochloropsis aff. iris</i>	-	2	2
<i>Augochloropsis illustris</i> (Vachal, 1903)	15	10	25
<i>Augochloropsis melanochaeta</i> Moure, 1950	-	1	1
<i>Dialictus</i> sp.	-	1	1
<i>Pseudaugochlora graminea</i> (Fabricius, 1804)	-	1	1
<i>Pseudaugochlora pandora</i> (Smith, 1853)	11	13	24
Abundance	142	134	276
Richness	12	16	18

Fruit set did not vary with pollination types in the sprinkler treatment ($F = 0.09$; $df = 115$; $P > 0.05$) or in the drip treatment ($F = 0.84$; $df = 115$; $P > 0.05$). Fruit weight was lower in bagged pollination and did not differ between the open pollination and

open + mechanical pollination in both irrigation systems ($F = 169.33$; $df = 313$; $P < 0.001$ and $F = 167.63$; $df = 313$; $P < 0.001$).

Fruit set did not vary with irrigation systems (Fig. 1A). On the other hand, fruit weight was higher in the drip irrigation system in the three types of pollination (Fig. 1B). The yield was higher in the drip irrigation system, with an average of 2.82 kg per plant versus 2.15 kg of the sprinkler irrigation system ($t = -2.37$; $df = 58$; $P < 0.05$).

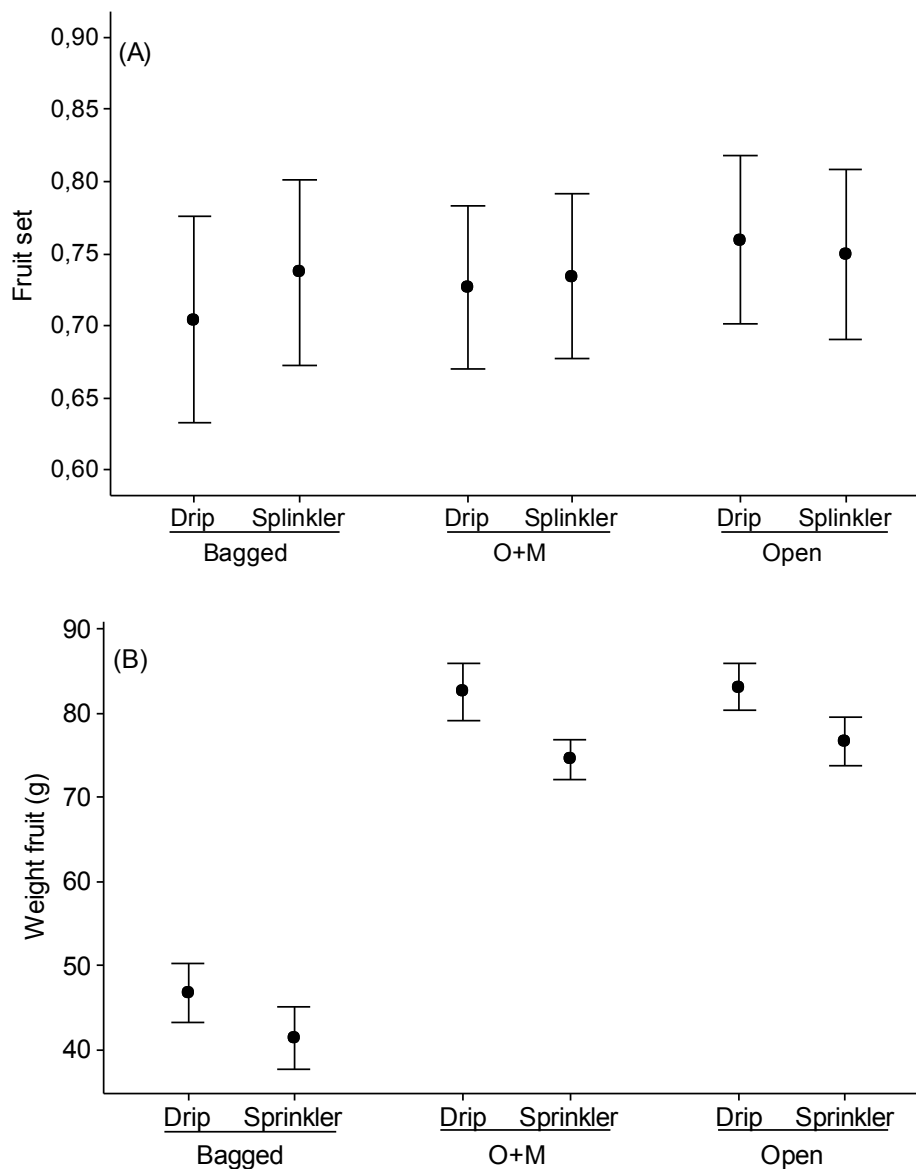


Fig. 1. Fruit set (A) and weight fruit (B) in relation to type of irrigation and type of pollination. O + M = open + mechanical pollination.

4. Discussion

The irrigation method did not affect the abundance and richness of bee species foraging on the tomato flowers, and this may be related to the nesting behavior of the individuals collected. Irrigation seems to have a greater influence on solitary bees that nest underground, and that build their nests inside irrigated crop areas (Julier and T'Ai, 2009). Moisture has the potential to saturate nests, causing larval death by drowning, or favoring the development of fungi, the main cause of mortality of young bees in species that nest underground (Batra, 1984; Polidori et al., 2010). On the other hand, solitary bees have developed different strategies (e.g., the lining of brood cell by hydrophobic material) to deal with excess moisture in the soil, and in some cases bees can survive for several months in flooded places (Roubik and Michener, 1980; Pietsch et al., 2016). Additionally, the moisture provided by the irrigation system may facilitate soil digging by bees (Cane, 2008; Julier and T'Ai, 2009). In our study, the great abundance of eusocial or communal bees, represented by the genera *Bombus*, *Exomalopsis*, *Paratrigona* and *Trigona*, suggests that most of the individuals collected came from nests outside the crop area. This leads us to believe that the irrigation system did not directly affect the bee community.

Irrigation may indirectly affect pollinators because it changes flower attractiveness (Gillespie et al., 2015; Burkle and Runyon, 2016). Additionally, nectar is an important reward that determines the foraging behavior of bees (Pike et al., 2016). However, there is no nectar production by the tomato flower (Thorp, 2000), so pollen is the principal reward. Pollen production and other important characteristics such as flower morphology and floral volatile emission may be affected by soil water availability (Waser and Price, 2016; Gallagher and Campbell, 2017). The lack of significant difference in bee abundance and richness between the two irrigation systems leads us to believe that there were no variations in the floral characteristics of

the two systems or, if there were variations, they did not influence bee visitation. In addition, the bees collected in our study are similar to bees' fauna described in other studies on the tomato crop (Silva-Neto et al., 2013; Deprá et al., 2014; Santos et al., 2014), mainly the dominant species or genera, which reinforces our conclusions about irrigation effect in the community of bees.

The absence of effects due to pollination on fruit set indicates that the irrigation method does not affect this characteristic. It is possible that the mechanical stimulus caused by wind offset any interference with pollination due to irrigation, although the effect of the wind was minimized by the installation of a screen around the crop. On the other hand, the lower fruit weight in the bagged (control) treatment in the sprinkler irrigation treatment indicates that the external stimulus caused by the sprinkler water droplets was not sufficient to reach the pollination level performed by the bees. Open pollination, possibly performed by the bees sampled in the crop, did not differ from the open + mechanical pollination, and this result is an indicator that there was no pollination deficit in the crop.

The higher weight of fruits in the drip irrigation system may be related to the effect of irrigation on the physiology of the tomato. In drip irrigation there is better distribution of water, which can affect the development of the root and consequently the absorption and assimilation of nutrients that directly influence the fruit mass (Nangare et al., 2016). Irrigation could affect fruit mass through pollination in two ways. First, irrigation could directly induce pollination due to the impact of the water droplets of the sprinkler on the flower, causing release of the pollen and consequently pollination. This hypothesis would be confirmed if the bagged (control) treatment of the sprinkler irrigation system had produced fruits with equal or greater weight to those that underwent the treatments of open pollination and open + mechanical pollination, but this did not occur. Second, irrigation could affect the presence of the pollinators

and consequently the pollination, but the results showed that the pollinators were not affected either.

Our results demonstrate the importance of bees to improve tomato yield. This is based on the observation that, contrary to our expectations, the mechanical impact of the sprinkler did not provide the same level of bee pollination. On the other hand, it is possible that high-flow sprinklers present results contrary to our conclusions because they produce larger droplets and could have a greater mechanical impact on the tomato flowers. However, high flow systems are not common in tomato production. Because pollination and pollinators were not affected, the choice of irrigation system should take into account agronomics criteria such as financial viability, and water use efficiency. The drip irrigation system seems to be the most effective because it demonstrates these characteristics even though it may be more expensive (Baiaomonte et al., 2015; Lu et al., 2016).

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GENERAL CONCLUSIONS

In this thesis, I took an important step towards understanding how bees affect the tomato production and how communities of these pollinators respond to the landscape configuration and farm management. The results support the general hypothesis that few species dominate bees' fauna in agricultural environments. Despite the fact that tomato is an autogamous plant, bee-mediated pollination increases fruit weight. However, a single visit by a bee to the flower did not result in heavier fruits compared to open pollination when, supposedly, the flower was visited more than once by different species. Thus, I emphasize the importance of more diverse communities for greater efficiency and stability of pollination services.

Bees' community was impacted by both farm management and landscape configuration. Overall, while wild bees responded negatively to natural habitat loss and farm management intensification, honeybees responded positively to these variables. The results are particularly relevant considering the traditional discussion between agricultural intensification and biodiversity preservation and can guide pollinator management plans.

An additional evaluation of the impact of two irrigation systems on pollination showed that this management practice was not significant. The mechanical impact of the sprinkler did not provide the same level of bee pollination. As pollination and pollinators were not affected, other criteria such as agronomics, financial viability, and water use efficiency should be taken into account when choosing an irrigation system.