Animal pollination dependence in Feijoa [Acca sellowiana (Berg) Burret] (Myrtaceae) and variation of pollinators between regions of Uruguay

Dependência da polinização animal em goiaba [Acca sellowiana (Berg) Burret] (Myrtaceae) e variação de polinizadores entre regiões do Uruguai

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ABSTRACT
The Feijoa "Guayabo del país" (*Acca sellowiana*) is a native Myrtaceae whose fruits have a great commercial potential for their nutritive properties. The objectives of this study were to determine the extent to which fruit production depends on biotic pollination and to identify potential pollinators in Uruguay. Two farms and three park, located in Salto and Canelones were used for the study. The percentage of fruit set in the branches excluding pollinators did not exceed 5.1%, whereas in the branches with free access of pollinators it varied between 10.4% (Salto) and 90.9% (park trees, Canelones). A total of 28 insect species and 3 Passeriforme species were identified on the flower varying with the region. *Apis mellifera* were present on all plantations with high frequency. Remarkably, 7 species of native bees and 3 species of native passerines were found only in the southern trees of the country. Some pollinators can be managed to improve production, of commercial quality, like honeybees or bumblebees.

Keywords: pollinator dependence, native fruits, production.

RESUMO
A goiaba "Guayabo del país" (Acca sellowiana) é uma Myrtaceae nativa cujos frutos apresentam grande potencial comercial por suas propriedades nutritivas. Os objetivos deste estudo foram determinar em que medida a produção de frutos depende da polinização biótica e identificar potenciais polinizadores no Uruguai. Duas fazendas e três parques, localizados em Salto e Canelones, foram utilizados para o estudo. A porcentagem de frutificação nos ramos excluindo polinizadores não ultrapassou 5,1%, enquanto nos ramos com livre acesso de polinizadores variou entre 10,4% (Salto) e 90,9% (árvores do parque, Canelones). Um total de 28 espécies de insetos e 3 espécies de Passeriformes foram identificados na flor variando com a região. Apis mellifera esteve presente em todos os plantios com alta frequência. Notavelmente, 7 espécies de abelhas nativas e 3 espécies de passeriformes nativos foram encontrados apenas nas árvores do sul do país. Alguns polinizadores podem ser manejados para melhorar a produção, de qualidade comercial, como abelhas ou zangões.

Palavras-chave: dependência de polinizadores, frutas nativas, produção.

1 INTRODUCTION

The fruits of native species with nutritional and sensory properties are commercially explored in Uruguay. Among them stands out the Feijoa "Guayabo del país" (*Acca sellowiana*) (Berg) Burret, synonym *Feijoa sellowiana* Berg., belonging to the family Myrtaceae, whose fruits have nutraceutical properties (Danner et al. 2010; Feippe et al. 2011, Bontempo et al. 2007; Beyhan et al. 2010; Weston 2010; Silveira et al. 2015; Rasekh et al 2020).

Acca sellowiana is a native species and therefore a lot of wild varieties is found throughout the territory, with different genotypic and phenotypic characteristics (Vignale & Bisio, 2005, Matias et al. 2020). In Uruguay, studies are being carried out to identify the different varieties with the

objective of producing them commercially (Lombardo et al. 2010; Feippe et al. 2011). Uruguayan varieties are internationally recognized for their value as a fruit and ornamental species, cultivated in different regions of the world (Amarante et al. 2008; Fischer et al. 2020). Thus, both in the Agricultural Research Institute (INIA) and the Faculty of Agronomy there are programs to study different varieties, trying to identify which are the most commercially suitable to reproduce them through clones. In this sense, it has been observed that there are varieties with higher fruit set than other, some that are self-compatible and others that are self-incompatible and even varieties that bloom at different times (Lombardo et al., 2010).

Guava trees have two botanical varieties with different dispersal centers. The variety "Brazil" presents its center of dispersion in the mountain range of the northeast of the Brazilian states of Rio Grande do Sul and Santa Catarina. The variety "Uruguay" presents its center of dispersion in the southeast of the state of Rio Grande do Sul and the mountain range of the south and north of Uruguay. (Ducroquet et al. 2000; Thorp & Bieleski 2002). The guava presents hermaphrodite and longistillate flowers, with four green and tomentose sepals and 4 to 5 fleshy white petals, reddish on the underside, with numerous stamens and this flowers do not produce nectar (Ducroquet & Hickel 1997; Roig 1992; Ducroquet et al. 2000; Ramírez & Kallarackal 2017). It has been reported that cross-fertilization ensures a greater quantity and quality of fruit (Patterson 1990; Lombardo et al. 2010, Quintero 2012). The guava flower presents a tendency to protogyny, since the stigma becomes receptive 24 hours before the dehiscence of the anthers, and it can remain receptive up to 10 hours after it (Ducroquet et al. 2000; Lombardo et al. 2010). Ducroquet et al. (2000) propose that there are physiological barriers that make this species predominantly allogamous and Dettori and Palombi (2000) point out that there is an incompatibility barrier in the ovary. On the other hand, Finardi et al. (2002) evaluated the fructification of the guava tree and found self-incompatibility, suggesting the need for cross-pollination between genetically different plants for fruit production. Patterson (1990) also indicate that cross-fertilization ensures a greater quantity and quality of fruits. The most important pollinating agents are birds and insects (Ramírez & Kallarackal, 2017). The birds pollinate and eat the petals, preventing the proliferation of diseases (Quintero, 2012)

Ducroquet et al. (2000) estimated that about 25% fruit set may be due to entomophilic pollination. It has also been attributed much importance as efficient pollinating agents to several species of birds that visit the flowers to feed on their sweet petals (Pepone 1912; Ducroquet & Hickel 1997; Degenhardt et al. 2001). These authors propose, considering the architecture of the guava flowers, that the birds visit them to feed on their sweet petals, the head feathers are pollinated with pollen, and this is transported to other flowers when the bird is still looking for food. However, in

Uruguay, birds are not frequently seen in guava crops (Felipe García, personal communication). The wind can intervene in pollination, although it would not have a relevant role (Degenhardt et al. 2001). Currently most of the information available regarding pollination dependence comes from traditional crops (Klein et al. 2007; Quintero 2012). In Uruguay Lombardo et al. (2010) observed a great variety of insects on guava flowers, especially bumblebees, whose action as pollinators improves the size, weight and uniformity of the fruits. The aim of this study was to determine to what extent the production of fruits Guayabo (Feijoa) the country depends on biotic pollination and identify the biota in flowers as potential pollinators.

2 MATERIALS AND METHODS

Study sites

This study was carried out in two years. In 2009, a guava cultivation was carried out in the San Antonio Experimental Station of the Faculty of Agronomy – Salto, Uruguay (31°21′23″S 57°45′56″O) and in another crop located in INIA Las Brujas – Canelones, Uruguay (34°40´05" S 56°20´24"). Both crops consisted of juvenile trees, with the Faculty of Agronomy being the one with the most trees and the largest. In 2010 the studies were carried out in the cultivation of guavas of INIA Las Brujas and in three wild trees isolated from each other, located in private and municipal parks of the city of Sauce (Canelones).

Calendar of visits to the crops

In 2009 the guava cultivation of the Faculty of Agronomy was visited on November 13, 20 and 27, and INIA Las Brujas on November 7, 16 and 25 and December 2.

In 2010, the cultivation of guavas of INIA Las Brujas was visited on October 30 and November 6, 13, 20 and 27, and the trees of the city of Sauce on October 28 and 4, 11, 18 and 25 of November.

At each visit to the crops, the number of open flowers in the F2 and F3 stages was recorded, according to the classification of Ducroquet and Hickel (1991), to determine the rate of flowering in which the samplings were carried out

Dependence of the crop to biotic pollination

To determine the dependence of the crop on biotic pollination, the branches with flowers on different trees were covered with a mesh of white synthetic tulle with perforations of 1.5 mm to prevent the access of insects and birds. In 2009, 385 flowers of 5 trees of Faculty of Agronomy, 90 flowers of 6 trees of INIA Las Brujas and 605 flowers in 15 trees of parks and gardens of the city

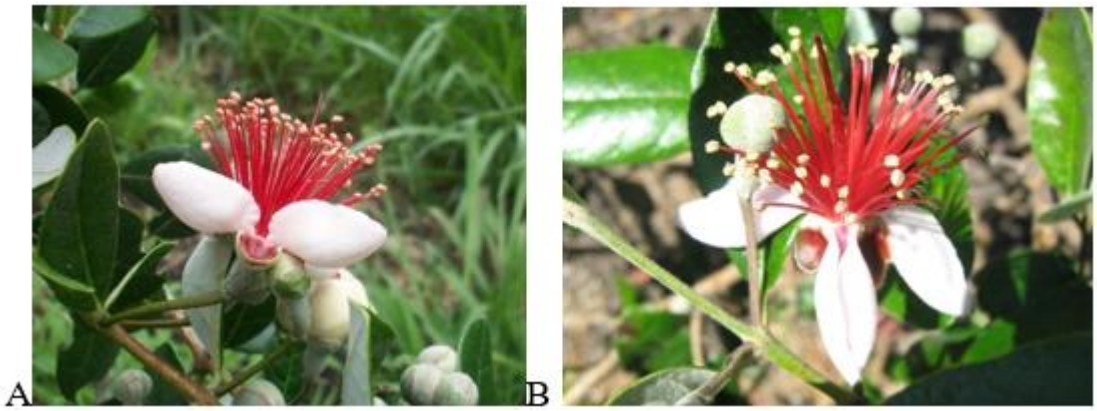
of Salto were covered. The tulle used allows the entry of wind and pollen grains carried by it, and sunlight enters without problems through the mesh which not influenced the anthesis. In the same trees where the flowers were enclosed, a branch with flowers was selected and marked to evaluate the fruiting with free pollination. The number of flowers with free access of insects in the branches marked in the plantations of Faculty of Agronomy, INIA Las Brujas and in the trees of the city of Salto was 193, 83 and 319, respectively. The enclosing of the flowers was done before the anthesis and it was found that the floral opening proceeded without problems under the mesh, discarding from the study those flowers that touched it and could be pollinated through it. In 2010 we proceeded in the same way as in the previous year covering a total of 75 flowers in 6 trees of INIA Las Brujas and 214 flowers in 3 wild trees of Sauce Parks. The number of flowers with free access of pollinators in INIA Las Brujas and in the wild trees was 76 and 210, respectively. After the flowering the amount of fruit formed in each of the treatments was recorded.

Floral visitors

In 2009, a survey was made of the fauna that was found on the floral structures in the guava crops of the Faculty of Agronomy and INIA Las Brujas. Only the flowers that were in the phenological stages F2 and F3 according to the classification of Ducroquet & Hickel (1991) were considered (Figure 1). The floral visitors were registered in four periods of the day: 07: 30-09: 00, 10: 00-11: 30, 13: 30-15: 00 and 15: 30-17: 00 hours.

In 2010 the observations were made in the cultivation of INIA Las Brujas and in three isolated trees in the city of Sauce. The observations were made in four periods of the day. In INIA Las Brujas: 07: 00-09: 00, 10: 00-11: 30, 13: 30-15: 00 and 15: 30-17: 30 hours, and in the three Sauce trees: 07: 00- 10:00, 10: 00-13: 00, 13: 30-16: 30 and 16: 30-19: 30 hours.

Figure 1. A- Phenological stages F2, B- Phenological stages F3



With the help of an entomological network, insects were collected for later classification in the laboratory. The identification of the different insects captured was done using the keys of Richards & Davies (1984) for orders of insects in general, Artigas and Hengst (1999) for dipterans, Arnett and Thomas (2001) and Arnett et al. (2002) for beetles, and Michener (2007) for bees. The presence of birds that interacted with flowers was also observed with binoculars. They were identified using the guide of birds of Uruguay of Azpiroz (2001).

Fruit setting

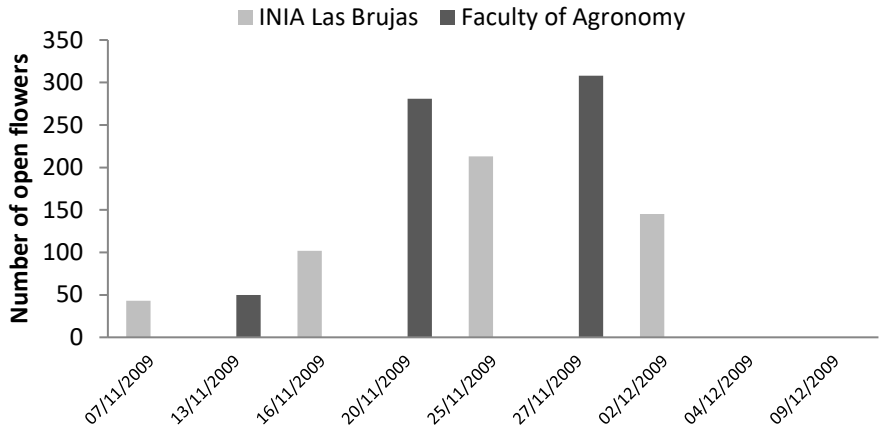
In the covered branches and those that could receive insects the fruit set percentage was determined. The percentage of fruit set in the two types of branches was compared using a 2 x 2 Chi-square test.

3 RESULTS AND DISCUSSION

Evolution of the flowering in the calendar of visits

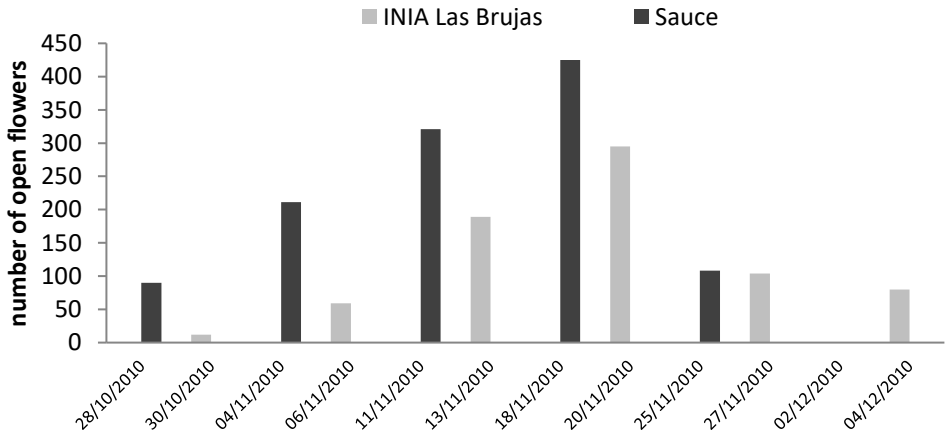
In 2009 the flowering in the cultivation of guava trees of the Faculty of Agronomy began the second week of November and ended abruptly on the end of this month. At the last visit (December 4) no flowers were found. In the cultivation of INIA Las Brujas, flowering began in the first week of November and ended in the first week of December. The peaks of flowering of both crops were coincident: 27 and 25 November for the Salto and Canelones crops, respectively (Figure 2). The longest duration of flowering in the INIA crop Las Brujas may be due to the lower number of pollinators (trees bloom asynchronously increasing the opening of flowers as they are pollinated) or environmental conditions more benevolent. In this sense, in the cultivation of Salto winds and rains were verified that affected many flowers. To this was added the presence of insects that fed on the floral structures.

Figure 2. Evolution of flowering in the guava crops of the Faculty of Agronomy (Salto) and INIA Las Brujas (Canelones) during the year 2009



In 2010 the flowering of the INIA Las Brujas crop began in the last week of October and some flowers were still present on the last visit of December 4. In the three trees of the city of Sauce, flowering began at the end of October and lasted until the end of November (Figure 3). The difference between the flowering periods of the trees of both sites may be due to a greater presence of pollinating agents in the trees of the city of Sauce (see below) or it may be different varieties of guava (Vignale and Bisio, 2005) .

Figure 3. Evolution of flowering in the crops of INIA Las Brujas and in the trees of the city of Sauce Canelones during the year 2010.



Dependence of guava trees to biotic pollination

It was found that the mesh of the branches did not interfere with the normal anthesis. A little before the fruit acquired consumption characteristics, the meshes were removed to determine the percentage of fruit set. The data obtained are shown in Table 1.

Table 1. Percentage of fruit setting in the experimental trees of guava

Year	Site	Pollinators access	N° plant	N° flowers	N° fruit	% fruit
2009	Faculty Agronomy	No	5	385	2	0,5
		Yes		193	20	10,4
		$Chi^2 = 28,16$; $P < 0,0001$				
	INIA Las Brujas	No	6	90	0	0
		Yes		83	21	25,3
		$Chi^2 = 18,39$; $P < 0,0001$				
2010	INIA Las Brujas	No	6	75	0	0
		Yes		76	42	55,3
		$Chi^2 = 32,06$; $P < 0,0001$				
	Trees Sauce	No	3	214	11	5,1
		Yes		210	191	90,9
		$Chi^2 = 120,47$; $P < 0,0001$				

The percentage of fruit setting in the branches with flowers unable to receive pollinators varied between 0 and 5.1%, while in the branches with flowers visited by insects varied between 10.4 and 90.9%. In the 4 trials, significant differences were found in the percentage of fruit set between the two treatments, showing that the fruit production in the guava trees has an important, albeit variable, dependence on entomophilous pollination.

Lombardo et al. (2010) reported that the percentage of fruit set in branches excluding insects in cultivated trees varied between 0 and 8%, and in the branches with access to pollinators it reached 15%. The values found in this study are similar to those found by Lombardo et al. (2010) in the plantations of the Faculty of Agronomy and INIA Las Brujas in 2009, but significantly higher in INIA Las Brujas in 2010 and in the wild trees of Canelones (Table 1).

The percentage of fruit set in the meshed branches was higher in the Canelones wild trees (5.1%) than in the cultivated trees (0.5, 0 and 0%). This difference may be due to the fact that wild trees had more flowers than cultivated trees, which could increase the likelihood of pollen reaching flowers enclosed by wind or gravity. It is noteworthy that the Myrtaceae have many anthers with great pollen production.

In 2009 in the crops of the Faculty of Agronomy and INIA Las Brujas, the fruit set percentage in the branches with free access to pollinators was low (10.3 and 25.3%, respectively). This low performance could be explained, the low density of flowers in cultivated trees may

disadvantage guava trees compared to other botanical species in the environment in the competition for attracting insects.

In contrast, in the wild trees of Sauce the percentage of fruit set was higher (90.1%). In these cases, the high amount of flowers can benefit fruit set, both by attracting more insects and by increasing the chances that the released pollen reaches the flowers. The high value obtained in the three trees of the city of Sauce would indicate the good biological potential of guava trees to obtain fruits.

It is interesting the comparison of the results obtained in the cultivation of INIA Las Brujas in the years 2009 and 2010, where in a year it doubled the fruit set percentage (from 25.3 to 55.3%). This substantial improvement may be due to the fact that the growth of juvenile trees was associated with an increase in the number of flowers with a greater presence of insects. Thus, in addition to increasing the percentage of fruit set, the absolute amount of fruit per tree increases. A longitudinal study from a new crop where the number of flowers per tree is recorded, the presence of pollinating insects, the percentage of fruit set and total production would be an important input to study the economic profitability of guava crops.

Floral visitors

In 2009 the entomological fauna observed on the guava flowers of the Faculties of Agronomy and of INIA Las Brujas was determined, and in 2010 it was determined in the cultivation of INIA Las Brujas and in wild trees of the city of Willow. The groups identified, their abundance and their location in the flower are presented in Tables 2-5.

The different species and morpho-species found belong to the orders Hymenoptera, Coleoptera, Diptera and Passeriforme, confirming many differences between crops and between years. Of the 31 species found, 19 were observed in contact with the pistil and / or anthers of the flower.

Table 2. Insects, abundance and location in the flowers in the cultivation of guava from the Faculty of Agronomy in 2009

Faculty of Agronomy (2009)				
Order	Familia	Taxa	Abundance	About pistil and/or anthers
Coleoptera	Scarabaeidae	-	3	+
Coleoptera 1	-	-	2	-
Coleoptera	Coccinellidae	-	1	-
Coleoptera	Chrysomelidae	-	2	+
Diptera	Syrphiade	-	1	-
Diptera	Tabanidae	-	5	-
Diptera	Calliphoridae	-	1	-
Diptera 2	-	-	3	-
Diptera 3	-	-	95	+
Diptera 4	-	-	3	-
Diptera 5	-	-	19	-
Diptera 6	-	-	65	+
Diptera 7	-	-	2	-
Diptera 8	-	-	27	+
Hymenoptera	Formicidae 2	-	15	-
Hymenoptera	Sphecidae	*Sphex* sp.	122	-
Hymenoptera	Apidae	*Apis mellifera*	18	+

Table 3. Insects, abundance and location in the flowers in the guava cultivation of INIA Las Brujas in the year 2009.

INIA Las Brujas (2009)				
Order	Family	Taxa	Abundance	About pistil and/or anthers
Diptera 1	-	-	3	-
Hymenoptera	Formicidae 1	-	62	-
Hymenoptera	Apidae	*Apis mellifera*	13	+
Hymenoptera	Halictidae	-	7	+
Hymenoptera	Megachilidae	*Megachile* sp.	2	+
Hymenoptera	Halictidae	*Halictus* sp.	2	+

Table 4. Groups of insects, abundance and location in the flowers in the guava cultivation of INIA Las Brujas in the year 2010. Also included are the birds found.

INIA Las Brujas (2010)				
Order	Family	Taxa	Abundance	About pistil and/or anthers
Diptera 3	-	-	1	+
Diptera 4	-	-	1	+
Hymenoptera	*Halictidae*	-	10	-
Hymenoptera	Apidae	*Apis mellifera*	35	+
Hymenoptera	Megachilidae	*Megachile* sp.	6	+
Hymenoptera	Halictidae	*Halictus* sp.	5	+
Hymenoptera	Apidae	*Bombus pauloensis*	5	+
Hymenoptera	Apidae	*Xylocopa augusti*	3	+
Hymenoptera	Vespidae	*Polybia scutellaris*	9	+
Hymenoptera	Formicidae 1	-	47	-
Diptera 1	-	-	5	-
Diptera	Syrphiade	-	27	-
Passeriformes	Turdidae	*Turdus rufiventris*	3	+
Passeriformes	Thraupidae	*Thraupis bonariensis*	3	+
Passeriformes	Mimidae	*Mimus saturninus*	2	+

Table 5. Groups of insects, abundance and location in the flowers in wild guava trees of the city of Sauce. The found birds are also included.

Wild trees of Sauce (2010)				
Order	Family	Taxa	Abundance	About pistil and/or anthers
Diptera 3	-	-	23	+
Diptera 4	-	-	20	+
Hymenoptera	*Halictidae*	*Halictidae*	21	+
Hymenoptera	Apidae	*Apis mellifera*	109	+
Hymenoptera	Megachilidae	*Megachile* sp.	36	+
Hymenoptera	Halictidae	*Halictus* sp.	34	+
Hymenoptera	Apidae	*Bombus pauloensis*	16	+
Hymenoptera	Apidae	*Bombus bellicosus*	5	+
Hymenoptera	Apidae	*Xylocopa augusti*	21	+
Hymenoptera	Apidae	*Xylocopa violaceae*	8	+
Hymenoptera	Vespidae	*Polybia scutellaris*	25	+
Hymenoptera	Formicidae 1	-	72	-
Diptera 1	-	-	62	-

Coloptera	Chrysomelidae	-	11	-
Diptera	Syrphiade	-	34	-
Passeriformes	Turdidae	*Turdus rufiventris*	15	+
Passeriformes	Thraupidae	*Thraupis bonariensis*	28	+
Passeriformes	Mimidae	*Mimus saturninus*	16	+

In the cultivation of Faculty of Agriculture in the north, fauna found in the flowers shown in Table 2. It is evident the presence of 10 species of Diptera, where the most frequent three were found on the pistil and/ or anthers The remaining Diptera species were located on the fleshy petals of the flowers, where they feed on sugary secretions.

The most abundant species was a wasp of the genus *Sphex*. This wasp feeds on the sweet petals of the guava tree, breaking them with its mouthparts. The wounds left by the wasps on the petals are what the various species of flies take advantage of to feed themselves. It was observed that when the *Sphex* wasps are present Diptera away from them and await their withdrawal sustained anthers and pistil of flowers and moving continuously. Thus, in the presence of wasps diptera may constitute potential pollinating agents of the guava. It is necessary to study in depth the wasp-diptera relationship and the consequences it may have on the pollination of guava trees. Honeybees were also present in the guava flowers, but no native bee species were recorded.

In the cultivation of INIA Las Brujas, few insect species were identified on the flowers, with the Formicidae ant being the most abundant (Table 3). They also found three species of native bees of different families and honeybees that is exotic, the latter being in greater proportion. The presence of native bees, absent in the farm of Salto, is very important since it is a group of insects that, due to their eating habits and pecking behavior, is recognized as an efficient pollinator (Michener, 1974). The four species of bees were the only insects present in the flowers that were in contact with the reproductive structures.

In this way, the situation may arise that the guava of both crops studied in 2009, located in two very distant geographical areas, are pollinated by different species, without preventing similar fruit set percentages. Observations of insects on guava flowers carried out in 2010 were characterized by finding greater diversity than in the previous year (Tables 4 and 5). Thus, considering only the insects that can be pollinating agents for being in contact with the reproductive structures of the flowers, 11 species were identified in the INIA Las Brujas crop and 14 in the wild willow trees. These values contrast with the 6 species found in the Faculty of Agronomy and the 4 in INIA Las Brujas in the previous year (Tables 2 and 3).

In INIA Las Brujas identified, in addition to the three species of native bees found in the same crop the previous year, two new species of native bees: *Bombus pauloensis* and *Xylocopa*

augusti. In the wild trees of Sauce the new species of native bees found were four: *Bombus bellicosus*, *B. pauloensis*, *Xylocopa violaceae* and *X. augusti*.

The observed bumblebees were queens that had emerged from hibernation, as workers are usually found from January (Salvarrey et al., 2017). Both *B. pauloensis* and *B. bellicosus* are very abundant species in Uruguay, the first distributed throughout the territory and the second south of the Rio Negro (Santos et al., 2017). The presence of bumblebees in the guava flowers had been reported by Lombardo et al. (2010).

The two species of *Xylocopa* found had already been recorded for the south of the country by Santos et al, (2020). This group formed by large insects, although it was present with few individuals in the flowers, can be important pollinators of the guava trees.

The highest insect richness was recorded in the three wild trees of the city of Sauce. It is possible that these large trees, with more flowers attract more and more diverse insects, basically aphid, than younger trees. Perhaps the insects find in these large trees, with more flowers, a more profitable offer of pollen and nectar, or less competition with other pollinating insects. This would result in a better pollination of the flowers with an increase in the percentage of fruit set, as verified in these trees (91%, Table 1).

Unlike what happened in 2009, in 2010 birds (Passeriformes) were observed interacting with guava flowers. The "zorzales" (*Turdus rufiventris*), "naranjeros" (*Thraupis bonariensis*) and "calandrias" (*Mimus saturninus*) were spotted at different times feeding from the flowers, as described by Anderson (2003). *Turdus rufiventris* and *T. bonariensis* were clearly observed feeding on the petals of the flower. On the other hand, with the *M. saturninus* there remains the doubt of whether it is the petals, or the insects associated with the flower that attracts them. Anyway, any of the birds mentioned causes an intense movement between the branches and flowers that will surely release large amounts of pollen. By looking at Table 1 and Table 2, we see there is higher fruit set in sites and seasons where birds were floral visitors. This suggests that birds could be more efficient pollinators. Although, in subsequent investigations, observations should also be made at night, to rule out nocturnal pollinators. Cordeiro et al, 2019 propose that several species of Myrtaceae in America have nocturnal pollinators, although the studied species have different flower phenology (Cordeito et al, 2016).

4 CONCLUSION

This study showed that the production of the fruits of the guava trees depend significantly on the presence of pollinators. It is difficult to qualify the degree of dependence of this crop

according to the classification carried out by Klein et al, 2007. Other studies must be carried out considering the great variability of genetic material that is available in the country.

The honeybees were present in all the crops observed with a remarkable abundance. This opens the possibility of pollinating the guayabo tree (Feijoa tree) by placing colonies of bees in the vicinity of the crops. A specific study would be needed to determine how much tree production increases when the number of bees in the crop increases, to justify the hiring of pollination services to beekeepers.

Different species of native bees were present in the observations made in the department of Canelones, especially in the year 2010, although they were not detected in Salto. Among these are the bumblebees of the genus *Bombus*. Although during the flowering period it is only possible to see queens emerging from hibernation, there is the possibility of pollinating the crop with workers if bumblebees can be artificially bred by de-seasonalizing the natural cycle. In this sense, in Uruguay there are already incipient results of breeding of native bumblebees (Salvarrey et al., 2013) that generate expectation that the breeding technique of these valuable pollinators can be mastered and produce colonies on a larger scale. The increase in other native bee species could be increased by favoring nesting sites and avoiding the use of insecticides harmful to bees (Shepherd et al., 2003).

In the culture of Salto an interesting relation was observed between a wasp of the genus *Sphex* and several species of Diptera, where these are located in the anthers of the flowers when the wasps are feeding on the petals. The role these Diptera may have in pollination is unknown, but the fact that the fruit set percentage in Salto was important even when no aphid was observed, except *A. mellifera*, suggests that Diptera can be pollinating agents.

Finally, the role that birds can play in pollination is unclear. The results of this study do not provide information in this regard, especially when they were only present in the trees of the town of Sauce and in a single year, more research should be addressed in this regard.

Uruguay has been developing lines of research at INIA and Faculty of Agronomy to study the commercial potential of guava trees (Vignale and Bisio, 2005, Lombardo et al., 2010, Feippe et al., 2011). This study makes it clear that the pollination of trees is a major issue for fruit production.

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