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Africanized honeybee and its contribution to soybean yield in Brazil

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ABSTRACT

Cultivated areas of soybean in Brazil have been increasing with each harvest and the seed market is moving in the same direction, launching new varieties every year. To increase not only planted area but also productivity, it is necessary to use technologies and use integration systems. In this study, the effect of pollination by honeybees on two soybean varieties in two consecutive years was evaluated. Three treatments were established: cages with Apis mellifera honeybees, cages without bees and free areas for insect visitation. The results showed an increase 6.45% of soybean yield in areas with free access to insect visitation. While in the cages with the introduction of Africanized honeybee colonies the increase was 13.64%. These results indicate that even in the most recent cultivars, cross pollination can show productivity gains even in an autogamous species.

Keywords: Biotic pollination; Crop pollination; Glycine max; Pollination service; Transgenic soybean.

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Introduction

Currently, soybeans constitute the main commodity in the world, contributing mainly to oil production and for livestock feeding (Agarwal et al., 2013). In Brazil, soybean cultivation increased from 640 hectares in the 1940s to over 35 million hectares in 2018 (Gazzoni and Dall'agnol, 2018).

The growth of the area destined to crop was accompanied by the suppression of the natural vegetation. This deforestation often damages agriculture itself, since forests and natural areas harbor a large variety of pollinators (Blitzer et al., 2012). The pollination service is essential for the maintenance of ecosystems, since 90% of the world's flowering plants depend on pollination to reproduce (Ollerton et al., 2011).

Although some plant species are self-pollinating, such as soybeans, their flowers throughout evolution have maintained anatomical structures and floral resources compatible with structures developed by plants dependent on insect pollination. The most common structures to attract insect pollinators are nectar guides and floral nectaries (Palmer et al., 2009).

Since there are hundreds of soybean varieties and not all are attractive to honeybees (Alves et al., 2010), it is necessary to study the association of the main varieties planted with the honeybee's behavior, since these are the most efficient insects in the pollination of soybean (Chiari et al. al., 2008).

Based on this, the objective of this study was to estimate the contribution of the pollination service of Africanized honeybees to soybean yield. Evaluating whether the presence of colonies of *Apis mellifera* honeybees improves seed production in transgenic and conventional soybean varieties.

Material and methods

This research was carried out in the experimental area of Brazilian Agricultural Research Corporation (EMBRAPA), located in Londrina city (23° 08'47" S and 51° 19'11" W), which is situated in the North region of state of Paraná, Brazil. The study was conducted in November 2016 and 2017.

A completely randomized design was used with three treatments and four replications each. Three treatments were evaluated: covered soybean area with a colony of Africanized honeybees inside during the flowering; covered soybean area without a honeybee colony; and an uncovered soybean area, free for insect visitation. All treatments were evaluated with cultivars BRS 284 (conventional soybean) and BRS 1001 IPRO (transgenic soybean).

The area chosen for study were demarcated at random in the soybean field and the cages were put immediately before beginning of blossom. Pollination cages were made of nylon screen of 2 x 2 mm, supported by pipes of ¼ of inch of PVC tubes, forming cages of fours meter width, six meters length and two meters height in the highest part, on an area of 24 m^2.

Soybean yields for three treatments were obtained at harvesting, collected in the central area of each plot (12 m^2) to avoid any border effect. Samples of four linear meters were collected at random in each plot for counting the number of pods set per plant, number of pods containing one, two, three or four seeds and total number of seeds per plant. After that, these seeds were added to remaining plot harvested, and the yield of each plot was determined. The evaluation of mean weight of seeds was made through weights of 1000 seeds.

The data were statistically analyzed according to completely randomized design. Soybean yields were compared by analysis of variance and means compared by Tukey`s test (5%), using R software (R Development Core Team 2018).

Results and discussion

Table 1 shows soybean yield in the experimental area (kg/ha) in different treatments. There were significant differences between treatments. However, there was no difference (p>0.05) between conventional and transgenic varieties. Areas with Africanized honeybee colonies produced significantly (p<0.05) greater yield than areas covered without honeybee colony and uncovered area. Uncovered area also differed (p<0.05) to the covered area without honeybee.

Table 1. Mean seed yield (kg/ha) of soybean (*Glycine max*) and their respective standard error of mean, cv. BRS 284 (conventional) and BRS 1001 IPRO (transgenic), at the experimental station of Embrapa, in Londrina, state of Paraná

Treatment	Seed yield (kg/ha)
BRS 284	4,631.80 (± 557.74) a
BRS 1001 IPRO	4,541.02 (± 503.42) a
Covered area with honeybee colony	4,891.22 (± 394.34) a
Uncovered area	4,568.77 (± 538.84) b
Covered area without honeybee	4,299.24 (± 481.04) c

*Means followed by different lower-case letters, in the same column, differ at $p < 0.05$.

Both varieties had a high percentage of self-pollination, producing 4,299.24 kg/ha in the covered areas without honeybees and other insect pollinators. This value is 28.98% higher than the national average of 3,382.77 kg/ha (CONAB, 2018). However, cross pollination promoted by pollinating insects resulted in a significant increase in productivity. In open areas for free visitation of insects the production was 6.45% higher than in the covered areas without honeybees. While in the areas covered with honeybees the increase in seed yield was 13.45%.

Bettler et al. (2018) observed that when climatic conditions are favorable, the visitation of *Apis mellifera* honeybees in the soybean area increase the number of seeds per area. Similar result was found by Chiari et al. (2008), where the presence of honeybees increased soybean seed yield by more than 37%. The increase in seed yield probably occurred due to the better distribution of the pollen grains in the female organs of the flower, due to the movement of the honeybee when coming into contact with these structures at the moment of collecting the nectar (Milfont et al., 2013). Probably the highest productivity in the areas covered with Africanized honeybee colonies was higher than in the free areas for visitation due to the greater competition of honeybees for the flowers, since they were housed in a smaller area.

Regarding the number of pods per plant, Table 2 shows that in the presence of pollinator insects, the plants presented a higher number of pods with three and four seed ($p<0.05$), which also contributed to the increase of productivity in these areas when compared to the areas covered without honeybees. This result can be considered an indirect measure of the flower abortion rate, since when there was cross pollination mediated by the pollinator insects, the plants were able to set larger numbers of pods and with more seeds inside them.

Table 2. Mean of total pod set and number of pods with 1, 2, 3 or 4 seeds in a soybean (*Glycine max*) cv. BRS 284 and BRS 1001 IPRO, under three pollination treatments, at the experimental station of Embrapa, in Londrina, state of Paraná

Treatments	Total pods	Pods with 1 seed	Pods with 2 seed	Pods with 3 seeds	Pods with 4 seeds
Covered with honeybee	74.28±3.14a	14.14±0.85a	22.39±2.74a	34.51±1.23a	2.25±0.47a
Uncovered area	59.88±5.91b	10.3±1.12b	19.79±2.04a	28.66±1.79b	1.13±0.51b
Covered without-honeybee	53.14±3.55b	8.1±0.94b	18.81±1.83a	26.18±1.88b	0.95±0.39b

*Means followed by different lower-case letters, in the same column, differ at $p < 0.05$.

Although self-pollination is a beneficial reproductive mechanism for soybeans, there may be faults in the distribution of viable pollen grains in the female organ of the flowers and thus the set of three or four seed pods occurs in a smaller quantity (Milfont et al. 2013), or even increases the number of flowers aborted by the plant.

Monasterolo et al. (2015) observed that bees of the family Halictidae, after *Apis mellifera*, were the most frequent in visits to soybean flowers. A

similar result was found by Santos et al. (2013), in which bees of the family *Apidae*, *Halictidae* and *Megachilidae* were the most frequent visitors of soybean in Uruguay. Hence the importance of maintaining native vegetation close to agricultural cultivation areas, since many cultivated crops can benefit from interaction with pollinators. Zelaya et al. (2017) verified an increase of 37% in seed yield in the growing areas that were located closer to the forest.

The highest seed yield observed in this study is not related to seed weight (Table 3). Since there was no statistical difference between cultivars and nor among treatments ($p > 0.05$). Variation in seed weight due to insect pollination in soybean is much less frequent than seed yield per unit area (Bettler et al., 2018).

Table 3. Mean weight of 1000 seeds and their respective standard error of mean, cv. BRS 284 (conventional) and BRS 1001 IPRO (transgenic), at the experimental station of Embrapa, in Londrina, state of Paraná

Treatment	Weight of 1000 seeds (g)
BRS 284	14.83 (± 0.74) a
BRS 1001 IPRO	14.39 (± 0.42) a
Covered area with honeybee colony	14.27 (± 0.34) a
Uncovered area	14.55 (± 0.66) a
Covered area without honeybee	15.01 (± 0.92) a

*Means followed by different lower-case letters, in the same column, differ at $p < 0.05$.

Possibly, soybean only formed pods capable of being nourished until complete seed formation, since no statistical difference was observed in the seed weight between treatments. The lowest number of pods with three and four seeds per pod formed in the treatment without honeybees may have occurred due to the inability of the soybean, in this condition, to nourish all the seeds that would be formed in larger pods. Thus, there was lower production of pods with higher number of seeds, however the smaller pods formed produced seeds of the same weight as the other treatments, demonstrating that the lack of cross pollination did not affect seed quality.

Conclusion

The results indicate that even though it is a species able to perform self-pollination efficiently, there are innumerable benefits to soybean cv. BRS 284 and BRS 1001 IPRO when cross pollination is carried out by native insects and, mainly, in the presence of Africanized honeybees. Although native insects promote increased seed yield in these varieties, the pollination service of *Apis mellifera* honeybees present the highest productivity, as well as a larger number of plants with pods containing three and four seeds per pod.

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