DOI: 10.5433/1679-0359.2025v46n3p757

Survival and flight capacity of *Apis mellifera* L. (Hymenoptera: Apidae) after exposure to insecticide residues on melon leaves

Sobrevivência e capacidade de voo de *Apis mellifera* L. (Hymenoptera: Apidae) após exposição a resíduos de inseticidas em folhas de meloeiro

Carlos Henrique Peixoto de Barros[1]; Ewerton Marinho da Costa[2]; Anderson Bruno Anacleto de Andrade[3]*; Tiago Augusto Lima Cardoso[4]; Elton Lucio de Araujo[5]; Brenda Carla Rosendo Martins[6]; Luiz Antonio Freire Alencar Silva[7]; Emanoely Karoliny Santos da Silva[6]

Highlights

Residues of Spinosyn insecticides cause deaths of *Apis mellifera.*
Residues of Anthranilic Diamides insecticides do not harm *Apis mellifera.*
Formulations with Abamectin insecticide cause mortality of honey bees.

Abstract

The bee *Apis mellifera* is crucial in pollinating melon (*Cucumis melo* L.) and producing its fruit. Proper use of insecticides and understanding their toxicity to pollinators is necessary to protect bees in the field. This study aimed to evaluate the residual toxicity of insecticides from the anthranilic diamide and spinosyn on *A. mellifera.* The experiment was conducted under laboratory conditions, evaluating two commercial doses of the anthranilic diamides Chlorantraniliprole, Cyantraniliprole, and Chlorantraniliprole + Abamectin, and the spinosyns Spinetoram and Spinosad when applied to melon leaves. After exposure to insecticide residues, survival for up to 72 hours and flight capacity of bee were assessed. Spinosad,

[1] Bachelor in Agronomy, Universidade Federal de Campina Grande, UFCG, Pombal, PB, Brazil. E-mail: henriquepeixotodb8@gmail.com
[2] Ph.D. in Agronomy, Crop Science, Associate Professor, UFCG, Pombal, PB, Brazil. E-mail: ewerton.marinho@professor.ufcg.edu.br
[3] Ph.D. in Plant Protection, Researcher in the Graduate Program in Tropical Horticulture, PPGHT, UFCG, Pombal, PB, Brazil. E-mail: bdeandrade3@gmail.com
[4] Ph.D. in Biological Sciences, Laboratory Technician, UnUFCG, Pombal, PB, Brazil. E-mail: tiagoipj@yahoo.com.br
[5] Ph.D. in Agricultural Entomology, Associate Professor, Universidade Federal Rural do Semi Árido, UFERSA, Mossoró, Brazil. E-mail: elton@ufersa.edu.br
[6] Master's Students in the Graduate Program in Tropical Horticulture, PPGHT, UFCG, Pombal, PB, Brazil. E-mail: bcrosendomartins@gmail.com; emanoelyka@gmail.com
[7] Student in Undergraduate Agronomy, UFCG, Pombal, PB, Brazil. E-mail: luiz-tn2@hotmail.com
* Author for correspondence

Received: Nov. 06, 2024 Approved: Apr. 28, 2025

Spinetoram, and Chlorantraniliprole + Abamectin, regardless of the dose, were toxic to bees, causing mortality rates above 85%. Chlorantraniliprole and Cyantraniliprole were less harmful, resulting in lower mortality rates. Anthranilic diamide insecticides did not affect the flight capacity of *A. mellifera.* Residues from insecticides tested in field doses on melon crops harm the survival of pollinating bees. However, the insecticides Chlorantraniliprole and Cyantraniliprole are viable for use in management strategies that prevent bees from being exposed to highly toxic products.
Key words: Mortality. Pollinators. Toxicity. Selective insecticide.

Resumo

A presença de *Apis mellifera* é fundamental para polinização do meloeiro (*Cucumis melo* L.) e produção de frutos. Para preservar as abelhas em campo é imprescindível o uso adequado de inseticidas, sendo necessário conhecer a toxicidade dos produtos sobre esses polinizadores. Diante disso, objetivou-se avaliar a toxicidade residual de inseticidas dos grupos químicos antranilamida e espinosina sobre *A. mellifera.* O experimento foi realizado em condições de laboratório, sendo avaliadas duas doses comerciais das antranilamidas Clorantraniliprole, Ciantraniliprole e Clorantraniliprole + Abamectina e das espinosinas Espinetoram e Espinosade aplicados em folhas de meloeiro. Após a exposição aos resíduos dos inseticidas foram avaliadas a sobrevivência até 72 horas e capacidade de voo das abelhas. Os inseticidas Espinosade, Espinetoram e Clorantraniliprole + Abamectina, independente da dose utilizada, foram tóxicos as abelhas, ocasionando mortalidade superior a 85%. Os inseticidas Clorantraniliprole e Ciantraniliprole foram pouco nocivos, ocasionando os menores percentuais de mortalidade. Os inseticidas do grupo antranilamida, não afetaram a capacidade de voo da *A. mellifera.* Os residuos nas folhas dos inseticidas testados com doses para uso em campo nos cultivos de melão pejudicam a sobrevivência das abelhas polinizadoras, porém os insetisidas Chlorantraniliprole e Cyantraniliprole são viáveis para uso em estratégias de manejo que evite a exposição das abelhas a produtos altamente tóxicos.
Palavras-chave: Mortalidade. Polinizadores. Toxicidade. Inseticidas seletivos.

Introduction

The presence of *A. mellifera* L. (Hymenoptera: Apidae) is essential for many economically important crops, making it one of the most valuable pollinators worldwide (Potts et al., 2010; Hung et al., 2018; Hünicken et al., 2022). In Brazil's main melon (*Cucumis melo* L.) production areas, placing beehives around fields is a common practice. This ensures effective pollination, good fruit development, and increased productivity (Sousa et al., 2013; Ribeiro et al., 2015; Tschoeke et al., 2019; Fernandes et al., 2023; Messellem et al., 2024).

However, billions of bees have disappeared from agricultural areas worldwide, and the use of insecticides is a major cause of this population decline (Leonhardt et al., 2013; Godfray et al., 2014; Fent & Christen, 2017). In the field, bees can be exposed to insecticides through contact with airborne particles, ingestion of contaminated nectar and pollen, and contact with product residues on plants, such as leaves after spraying (Costa et al., 2014; Silva et al., 2015; Heard et al., 2017). Contact with insecticides can be lethal or cause sublethal effects, including changes in behavior, physiology, biochemistry, histology, and gene expression. These effects impact the survival of pollinators (Pham-Delègue et al., 2002; Ibrahim et al., 2023; Shi et al., 2023; Wu et al., 2023).

As explained above, residual exposure after spraying is a major way bees get contaminated in the field. In Brazil, some studies have evaluated the residual toxicity of insecticides on melon leaves to *A. mellifera.* They found that residues of Thiamethoxam, Abamectin, Chlorfenapyr, Spiromesifen, Imidacloprid and Sulfoxaflor on melon on melon leaves are extremely toxic to *A. mellifera,* while Chlorantraniliprole is less toxic, causing low mortality but reducing the bees' flight capacity (Costa et al., 2014; Gomes et al., 2020; Costa et al., 2024; Souza et al., 2024).

In this way, to sustainably use pollinators in agriculture, we need to understand the effects of pesticides on them. Continuous studies on these insecticides lethal and sublethal effects are essential (Pinheiro & Freitas, 2010; Rosa et al., 2019). In melon cultivation, some products with recommended doses haven't been evaluated yet or were recently registered, like insecticides from the anthranilic diamide and spinosyn groups. Using product combinations also matters. For example, applying Chlorantraniliprole with an adjuvant in the field kills adult honey bees during almond bloom (Walker et al., 2022).

Therefore, studies on these products are necessary to ensure proper management of *A. mellifera* in production áreas. Therefore, this study aimed to evaluate the residual toxicity of insecticides from the anthranilic diamide and spinosyn groups on melon leaves to *A. mellifera,* to help in choosing products to compose strategies to preserve these pollinators in melon production areas.

Material and Methods

The experiment was conducted in the Entomology Laboratory of the Center for Agro-Food Science and Technology (CCTA) at the Federal University of Campina Grande (UFCG), in Pombal, Paraíba. Adult worker bees of *A. mellifera* were collected from honey frames of three colonies at the CCTA/UFCG apiary, located at 6° 47' 14.266" S; 37° 48' 7.765" W. The bees were collected between 5:30 AM and 7:00 AM and transported to the laboratory in plastic containers, which were partially covered with anti-aphid mesh and had lateral openings of approximately 2mm.

The insecticides tested belonged to two chemical groups: Anthranilic Diamides, which included Chlorantraniliprole, Cyantraniliprole, and Chlorantraniliprole + Abamectin; and Spinosyns, which included Spinetoram and Spinosad. We also used Thiamethoxam as a positive control. All insecticides were tested at the minimum and maximum doses recommended by the manufacturers for pest control in melon crops, except for Spinosad, which is only registered for watermelon crops (Table 1). The average application volume was considered to be 500 liters per hectare, and the solution was prepared in the laboratory to maintain this ratio, with dilutions made for an application volume of 1.0 liter.

Table 1
Insecticides and their respective dosages (minimum and maximum) evaluated for residual toxicity on *Apis mellifera*

Chemical group	Active ingredient	Doses	
		Product label	Active ingredient
Anthranilic Diamides*	Chlorantraniliprole	7.5 mL/100 L	0.015 g a.i. L^{-1}
Anthranilic Diamides	Cyantraniliprole	250 and 500 mL ha^{-1}	0.05 and 0.1 g a.i. L^{-1}
Anthranilic Diamides + Avermectin	Chlorantraniliprole +Abamectin	300 and 500 mL ha^{-1}	0.027+0.0108 and 0.045+0.018 g a.i. L^{-1}
Spinosyn	Spinetoram	120 and 160 g ha^{-1}	0.06 and 0.08 g a.i. L^{-1}
Spinosyn	Spinosad	150 and 200 mL ha^{-1}	0.144 and 0.192 g a.i. L^{-1}
Neonicotinoid**	Thiamethoxam	60 g ha^{-1}	0.03 g a.i. L^{-1}

*Registered single dosage for the crop, **Positive control.

The residual toxicity bioassay followed the methodology proposed by Costa et al. (2014). The experimental design was completely randomized with 9 treatments plus an absolute control (distilled water) and a positive control (Thiamethoxam - 0.03 g a.i./L), with 10 repetitions. Each experimental unit comprised 10 adult bees.

To evaluate the residual toxicity of the insecticides, we grew Iracema cultivar yellow melon plants (one of the main cultivars in northeastern Brazil) in the CCTA/UFCG greenhouse. The plants were kept in 1-kg pots with a substrate of soil and organic matter (2:1 ratio) and were watered three times a day. When the plants reached at least six true leaves, 10 plants were selected for each treatment.

The selected plants were sprayed with the respective treatments using a manual sprayer to simulate field application. The plants were then moved to a shaded, ventilated area to dry for 1 hour. After drying, the leaves were cut at the petiole and placed in arenas (plastic containers 15 cm in diameter x 15 cm in height, with the top partially covered with anti-aphid mesh to allow proper air circulation). Cotton soaked in water and artificial diet (Candi paste) were also placed in the arenas. Adult *A. mellifera* worker bees were then released into the arenas to contact the residues on the leaves and were maintained under controlled laboratory environmental conditions.

After the bees came into contact with the insecticide residues on the leaves, we assessed mortality and behavior (prostration, tremors, paralysis) at 1, 2, 3, 4, 5, 6, 9, 12, 15, 18, 21, 24, 30, 36, 42, 48, 60, and 72 hours after exposure. Bees that did not respond to mechanical stimuli (touches on their bodies with a fine brush during each evaluation period) were recorded as dead.

We assessed the flight capability of the surviving bees in each treatment using a flight tower, following the methodology by Gomes et al. (2020). The flight tower was made of wood (35 × 35 × 115 cm), open inside, and had a fluorescent lamp at the top. All sides of the tower were covered with transparent plastic to allow perfect visualization of the bees' flight. The evaluations took place in a dark room with an average temperature of 25 ± 2 °C and a relative humidity of 60 ± 10%, where the only light source was the lamp at the top of the tower, designed to attract the bees through positive phototropism.

Each surviving bee was individually placed at the base of the tower (0 cm height) and given 60 seconds to complete its flight. We then recorded the final height each bee reached. The flight tower had five height levels: 1 (base of the tower), 2 (1 cm to 30 cm), 3 (31 cm to 60 cm), 4 (61 cm to 90 cm), and 5 (91 cm to 115 cm, where the lamp was located).

The mortality averages were corrected using Abbott's formula (Abbott, 1925) and then subjected to the non-parametric test of Kruskal-Wallis at a 5% significance level, followed by the Wilcoxon test. Survival data were analyzed using the "Survival" package (Therneau & Lumley, 2010) in R software. Treatments with similar effects (toxicity and mortality rate) were grouped using contrasts. The median lethal time (TL_{50}) was also calculated for each group. The survival percentage was calculated using the formula $f(x) = e^{((-\mu^{-\alpha})(t^{\alpha}))}$, where μ = lethal time, α = 1.41844 (for Anthranilic Diamide Group), α = 1.392758 (for Spinosyn Group), t = time (hours), and $f(x)$ = survival (%). Survival data were fitted to the Weibull distribution. Flight capability data were evaluated using Permutation Anova (Permanova) and Scott-Knott tests. The analyses were conducted in R 4.2.2 (R Core Team [R], 2022).

Results

In assessing *A. mellifera* mortality after exposure to insecticide residues on melon leaves, we found a significant difference among treatments (χ^2 = 183, df = 10, p-value < 2.2^{e-16}). The insecticides spinosad, spinetoram, and chlorantraniliprole + abamectin, regardless of dosage, proved toxic to the bees, causing more than 85% mortality. At the highest doses of spinetoram and chlorantraniliprole + abamectin, we observed 100% bee mortality, comparable to the positive control (Thiamethoxam) (Figure 1).

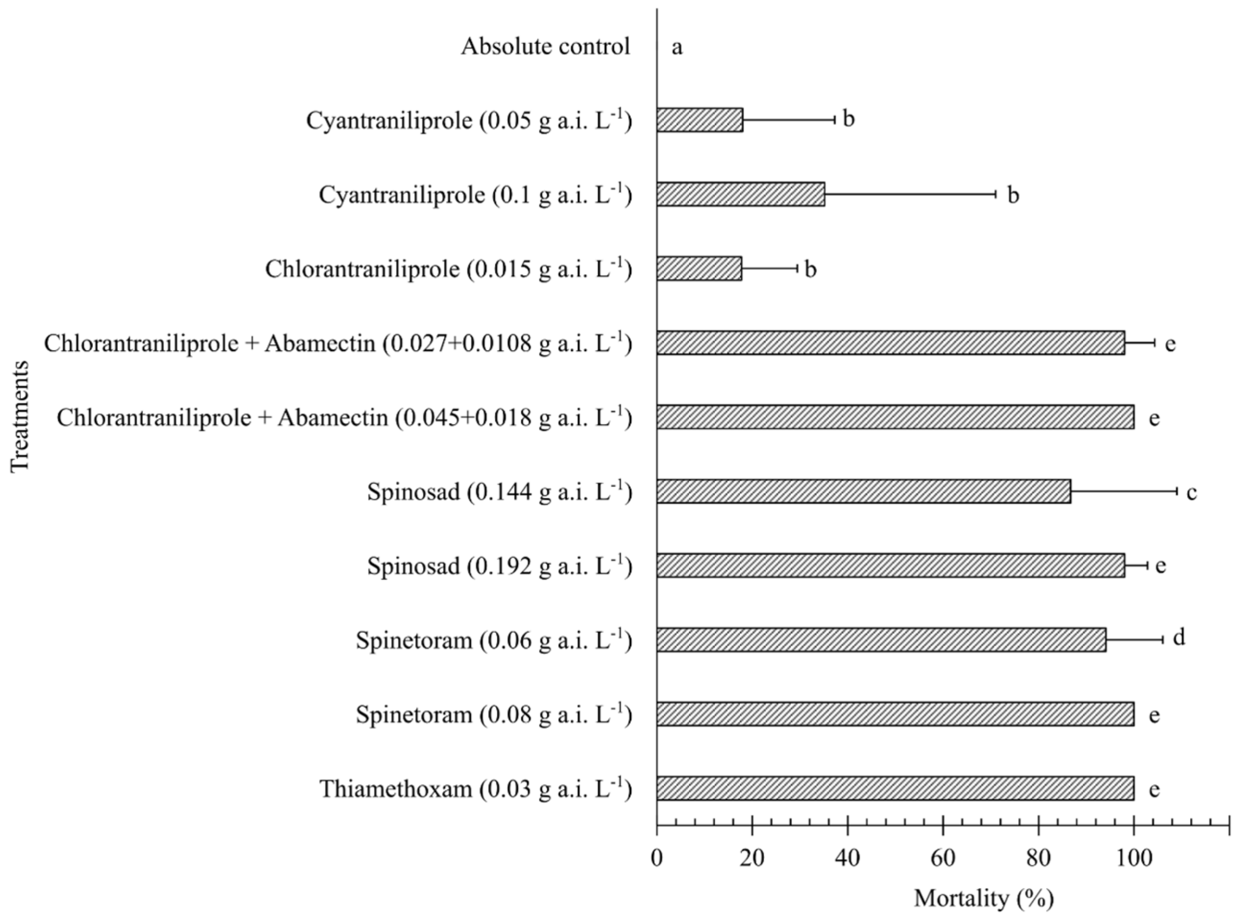

Figure 1. Mortality (%) of *A. mellifera* after exposure to insecticide residues on melon leaves. Different letters indicate significant differences according to the Wilcoxon multiple comparison test.

Furthermore, the insecticides Spinetoram and Chlorantraniliprole + Abamectin also induced tremors, paralysis, and prostration in the insects before death. In contrast, the insecticides chlorantraniliprole and cyantraniliprole resulted in the lowest mortality rates (Table 1), showing minimal harm to *A. mellifera* and no adverse effects on the bees' motor activities.

In analyzing bee survival after exposure to insecticides from the Anthranilic Diamide group, we found that doses combined with the Avermectin group resulted in the lowest median lethal times (TL_{50}), rapidly decreasing bee survival compared to the absolute control (Figure 2A). The Spinosyn insecticide doses had TL_{50} values below 33 hours, closely matching the positive control (Thiamethoxam). The highest dose of the Spinosyn insecticide (0.08 g a.i. L^{-1}) grouped with the positive control, with a TL_{50} of 9.6 hours (Figure 2B).

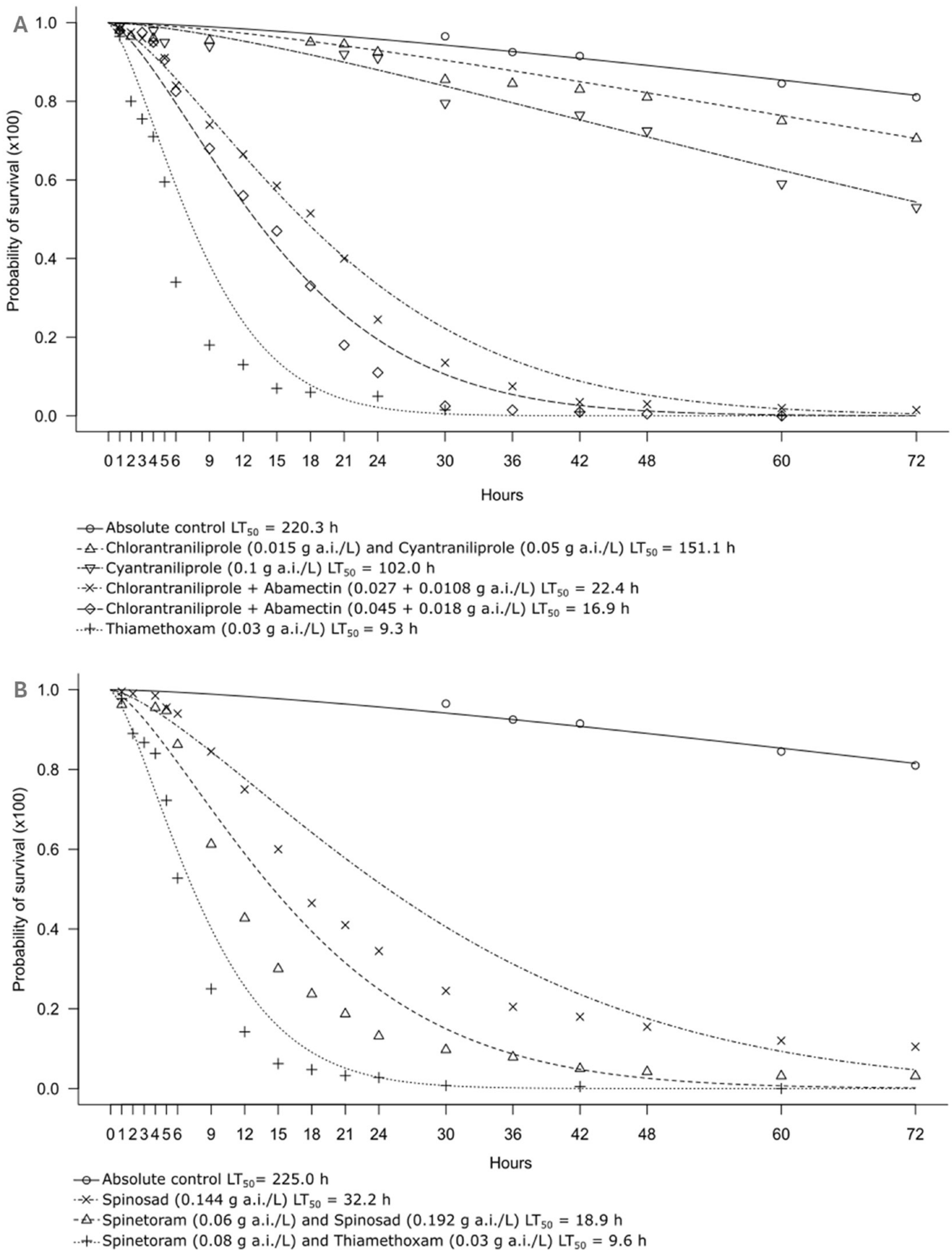

Figure 2. Survival (%) of *A. mellifera* and median lethal time (TL_{50}) after contact with insecticide residues on melon leaves.
(A) Anthranilic Diamide Group: Absolute control, Chlorantraniliprole (0.015 g a.i. L^{-1}) + Cyantraniliprole (0.05 g a.i. L^{-1}), Cyantraniliprole (0.1 g a.i. L^{-1}), Chlorantraniliprole + Abamectin (0.027+0.0108 g a.i. L^{-1}), Chlorantraniliprole + Abamectin (0.045+0.018 g a.i. L^{-1}), and positive control (Thiamethoxam (0.03 g a.i. L^{-1})). (B) Spinosyn Group: Absolute control, Spinosad (0.144 g a.i. L^{-1}), Spinetoram (0.06 g a.i. L^{-1}) + Spinosad (0.192 g a.i. L^{-1}), Spinetoram (0.08 g a.i. L^{-1}), and positive control (Thiamethoxam (0.03 g a.i. L^{-1})).

To evaluate flight capability, we used bees only from the absolute control group (distilled water) and those exposed to residues of the insecticides Chlorantraniliprole and Cyantraniliprole, both from the Anthranilic Diamide group, as there were no survivors in the other treatments.

The bees exhibited similar behavior across all treatments. Both insecticides showed effects that were not significantly different from the absolute control (Figure 3). However, at its highest dose (0.1 g a.i. L^{-1}), Cyantraniliprole had a higher number of bees unable to fly, remaining at the base of the flight tunnel and walking instead. Nonetheless, there was no significant difference compared to the other treatments (Figure 3).

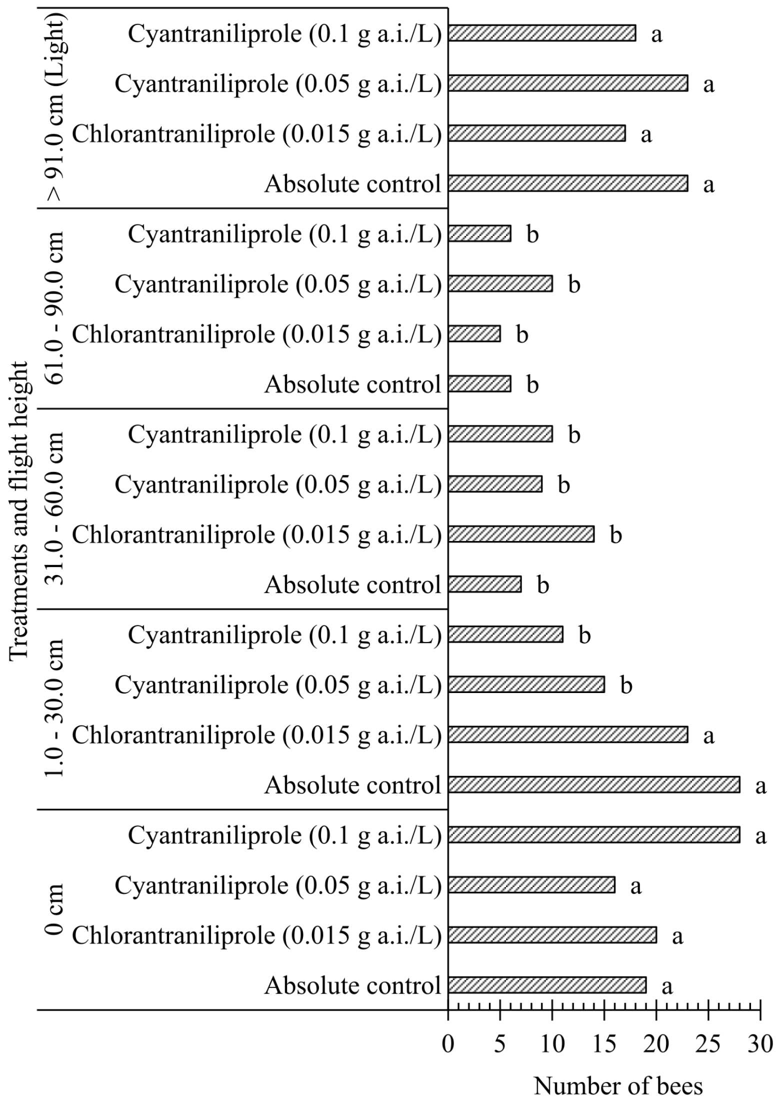

Figure 3. Flight capability of Apis mellifera after exposure to residues of the insecticides Chlorantraniliprole (0.015 g a.i. L^{-1}) and Cyantraniliprole (0.05 g a.i. L^{-1} and 0.1 g a.i. L^{-1}) and distilled water (absolute control).

Discussion

The high mortality rate caused by Spinosyn insecticides was also observed in other exposure methods for honey bees in Central America and Pakistan. In these studies, it was observed that the toxicity of these insecticides increased with higher concentrations. Among the tested insecticides, Spinosad was the most toxic and proved harmful to bees at the maximum recommended field doses (Edwards et al., 2003; Farooqi & Arshad, 2016). Additionally, high concentrations of the insecticide Spinosad in honey and waxes are linked to colony losses due to the significant presence of dead and dying adult bees (Naccari et al., 2024). In hazelnut cultivation, it was observed that insecticides Spinosad caused high bee mortality after 5 days of contact (Akça & Saruhan, 2022). Furthermore, exposure to lower doses of these insecticides can reduce bees' flight capacity, walking ability, respiratory rate, and detoxification enzyme activity. This exposure also leads to oxidative stress and cell death, demonstrating sublethal yet complex effects that impair individual bee homeostasis (Lopes et al., 2018; Araújo et al., 2023). In the present study, the recommended doses for melon cultivation resulted in high bee mortality, making it impossible to observe these sublethal effects.

Such mortality can be explained by the chemical composition of the insecticides and their mode of action. The insecticides spinosad and spinetoram are composed of a macrocyclic lactone isolated from the soil bacterium *Saccharopolyspora spinosa.* They act on the central nervous system of insects by directly causing prolonged activation of acetylcholine receptor proteins. This activation impacts genes involved in lysosomal function and reproduction, resulting in continuous and uncontrolled transmission of nerve impulses. This leads to tremors, intense excitement, muscle fatigue, paralysis, and eventually the insect's death (Salgado, 1998; Martelli et al., 2023), consistent with the bee behaviors observed in this study.

On the other hand, the insecticides from the Anthranilic Diamide group, Chlorantraniliprole and Cyantraniliprole, showed low toxicity and consequently low mortality. This finding aligns with studies using other exposure methods, where Chlorantraniliprole was deemed selective for *A. mellifera* (Carmo et al., 2017; Ratnakar et al., 2017). While contact and ingestion of this insecticide show low toxicity, Chlorantraniliprole has been observed to reduce flight capability (Gomes et al., 2020). Direct exposure to Chlorantraniliprole has been observed to cause lethargy and reduced locomotor activity in bees (Williams et al., 2020). Furthermore, Cyantraniliprole increases larval mortality within the colony but does not cause deformities in adult insects (Kim et al., 2022). This study observed low mortality, corroborating the literature. However, it did not alter the flight behavior of surviving individuals, which may be related to the kind of exposure the bees were subjected.

Pollinating insects can be exposed to insecticides through three different ways: contact with spray droplets, residues on plants, and ingestion of contaminated food. The exposure method can change the toxicity intensity of insecticides to bees (Costa et al., 2014). The sublethal effects caused by

Chlorantraniliprole in *A. mellifera* are long-lasting when applied to the dorsal abdomen, suggesting direct cardiotoxicity due to alterations in calcium channels in the brain and muscles (Kadala et al., 2019; Kaabeche et al., 2024). Finally, the observed low survival for the insecticide chlorantraniliprole + abamectin could be attributed to the active ingredient abamectin, as chlorantraniliprole alone caused low mortality. Abamectin has been reported to be highly toxic to *A. mellifera,* even in residual exposure (Carvalho et al., 2009; Costa et al., 2014). Combining active ingredients or adjuvants can harm bees in the field, especially during flowering periods. For example, applying chlorantraniliprole with adjuvants caused acute honey bee mortality at recommended doses (Walker et al., 2022; Shannon et al., 2023). These combinations can also disrupt bees' natural behaviors, such as pheromone signaling, leading to reproductive issues in colonies (Wu et al., 2023). These findings, along with the high mortality rates from chlorantraniliprole + abamectin combinations used in melon cultivation observed in this study highlight the need for more research on the impacts of these formulations on bees.

Conclusions

Residues on melon leaves from insecticides such as Spinetoram, Spinosad, and Chlorantraniliprole + Abamectin cause high mortality in *A. mellifera,* harming the survival of these crucial pollinators for melon productivity. The recommended doses of the insecticides Chlorantraniliprole and Cyantraniliprole decrease bee survival but do not affect their flight capacity. This suggests that these insecticides can be viable for use in management strategies that avoid exposing bees to highly toxic products.

References

Abbott, W. S. (1925). A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology, 18*(1), 265-267. doi: 10.1093/jee/18.2.265a

Akça, R., & Saruhan, I. (2022). The effects of some insecticides on honeybees (*Apis mellifera*). *Israel Journal of Ecology and Evolution, 69*(1-2), 37-43. doi: 10.1163/22244662-bja10043

Araújo, R. D. S., Lopes, M. P., Viana, T. A., Bastos, D. S. S., Machado-Neves, M., Botina, L. L., & Martins, G. F. (2023). Bioinsecticide spinosad poses multiple harmful effects on foragers of *Apis mellifera. Environmental Science and Pollution Research, 30*(25), 66923-66935. doi: 10.1007/s11356-023-27143-6

Carmo, D. G., Marsaro, A. L., Jr., Costa, T. L., Farias, E. de S., Ribeiro, A. V., & Picanço, M. C. (2017). Toxicidade de inseticidas comerciais, por ação de contato, para *Apis mellifera. Insetos e Entomologia,* 145-148.

Carvalho, S. M., Carvalho, G. A., Carvalho, C. F., Carvalho, J. S. S., & Baptista, A. P. M. (2009). Toxicidade de acaricidas/inseticidas empregados na citricultura para a abelha africanizada *Apis mellifera* L., 1758 (Hymenoptera: Apidae). *Arquivos do Instituto Biológico, 76*(4), 597-606. doi: 10.1590/1808-1657v76p5972009

Costa, E. M., Araujo, E. L., Maia, A. V. P., Silva, F. E. L., Bezerra, C. E. S., & Silva, J. G. (2014). Toxicity of insecticides used in the Brazilian melon crop to the honey bee *Apis mellifera* under laboratory conditions. *Apidologie, 45*(1), 34-44. doi: 10.1007/s13592-013-0226-5

Costa, E. M., Augusto, L. P., Silva, E. K. S. da, Rocha, V. H. M., Cardoso, T. A. L., Araujo, E. L., & Almeida, F. A. (2024). Honey bee survival and flight capacity after exposure to sulfoxaflor residues. *Sociobiology, 71*(4), e10729-e10729. doi: 10.13102/sociobiology.v71i4.10729

Edwards, C. R., Gerber, C. K., & Hunt, G. J. (2003). A laboratory study to evaluate the toxicity of the Mediterranean fruit fly, Ceratitis capitata, bait, Success 0.02 CB, to the honey bee, *Apis mellifera. Apidologie, 34*(2), 171-180. doi: 10.1051/apido:2003005

Farooqi, M. A., & Arshad, M. (2016). Toxicity of three commonly used nicotinoids and spinosad to *Apis mellifera* L. (Hymenoptera: Apidae) using surface residual bioassays. *Pakistan Journal of Zoology, 48*(6), 1983-1987.

Fent, K., & Christen, V. (2017). Exposure of honeybees (*Apis mellifera*) to different classes of insecticides exhibit distinct molecular effect patterns at concentrations that mimic environmental contamination. *Environmental Pollution, 226,* 48-59. doi: 10.1016/j.envpol.2017.04.003

Fernandes, N. S., Luz, L. R., Alves, E. G., Fº., Aragão, F. A. S. D., Zocolo, G. J., & Freitas, B. M. (2023). Differences in the chemical composition of melon (*Cucumis melo* L.) nectar explain flower gender preference by its pollinator, *Apis mellifera. Journal of the Brazilian Chemical Society, 34*(7), 976-986. doi: 10.21577/0103-5053.20230010

Godfray, H. C. J., Blacquière, T., Field, F. M., Hails, R. S., Petrokofsky, G., Potts, S. G., Raine, N. E., Vandergen, A. J., & Mclean, A. R. (2014). A restatement of the natural science evidence base concerning neonicotinoid insecticides and insect pollinators. *Published by the Royal Society B, 281*(1786), 20140558. doi: 10.1098/rspb.2014.0558

Gomes, I. N., Vieira, K. I. C., Gontijo, L. M., & Resende, H. C. (2020). Honeybee survival and flight capacity are compromised by insecticides used for controlling melon pests in Brazil. *Ecotoxicology, 29,* 97-107. doi: 10.1007/s10646-019-02145-8

Heard, M. S., Baas, J., Dorne, J. L., Lahive, E., Robinson, A. G., Rortais, A., Spurgeon, D. J., Svendsen, C., & Hesketh, H. (2017). Comparative toxicity of pesticides and environmental contaminantsin bees: are honey bees a useful proxy for wild bee species? *Science of the Total Environment, 578*(1), 357-365. doi: 10.1016/j.scitotenv.2016.10.180

Hung, K. L. J., Kingston, J. M., Albrecht, M., Holway, D. A., & Kohn, J. R. (2018). The worldwide importance of honey bees as pollinators in natural habitats. *Proceedings of the Royal Society B: Biological Sciences, 285*(1870), 20172140. doi: 10.1098/rspb.2017.2140

Hünicken, P. L., Morales, C. L., Villalobos, A. E. de, & Garibaldi, L. A. (2022). Evaluation of interactions between honeybees and alternative managed pollinators: a meta-analysis of their effect on crop productivity. *Agriculture, Ecosystems & Environment, 340,* 108156. doi: 10.1016/j.agee.2022.108156

Ibrahim, E. D. S., Abd Alla, A. E., El-Masarawy, M. S., Salem, R. A., Hassan, N. N., & Moustafa, M. A. (2023). Sulfoxaflor influences the biochemical and histological changes on honeybees (*Apis mellifera* L.). *Ecotoxicology, 32*(5), 674-681. doi: 10.1007/s10646-023-02677-0

Kaabeche, M., Charreton, M., Kadala, A., Mutterer, J., Charnet, P., & Collet, C. (2024). Cardiotoxicity of the diamide insecticide chlorantraniliprole in the intact heart and in isolated cardiomyocytes from the honey bee. *Scientific Reports, 14*(1), 14938. doi: 10.1038/s41598-024-65007-2

Kadala, A., Charreton, M., Charnet, P., & Collet, C. (2019). Honey bees long-lasting locomotor deficits after exposure to the diamide chlorantraniliprole are accompanied by brain and muscular calcium channels alterations. *Scientific Reports, 9*(1), 2153. doi: 10.1038/s41598-019-39193-3

Kim, J., Chon, K., Kim, B. S., Oh, J. A., Yoon, C. Y., & Park, H. H. (2022). Assessment of acute and chronic toxicity of cyantraniliprole and sulfoxaflor on honey bee (*Apis mellifera*) larvae. *Pest Management Science, 78*(12), 5402-5412. doi: 10.1002/ps.7162

Leonhardt, S. D., Gallai, N., Garibaldi, L. A., Kuhlmann, M., & Klein, A. M. (2013). Economicgain, stability of pollination and bee diversity decrease from southern to northern Europe. *Basic and Applied Ecology, 14*(6), 461-471. doi: 10.1016/j.baae.2013.06.003

Lopes, M. P., Fernandes, K. M., Tomé, H. V. V., Gonçalves, W. G., Miranda, F. R., Serrão, J. E., & Martins, G. F. (2018). Spinosad-mediated effects on the walking ability, midgut, and Malpighian tubules of Africanized honey bee workers. *Pest Management Science, 74*(6), 1311-1318. doi: 10.1002/ps.4815

Martelli, F., Ravenscroft, T. A., Hutchison, W., & Batterham, P. (2023). Tissue-specific transcriptome analyses in Drosophila provide novel insights into the mode of action of the insecticide spinosad and the function of its target, nAChRα6. *Pest Management Science, 79*(10), 3913-3925. doi: 10.1002/ps.7585

Messellem, I., Aguib, S., Abed, R., & Abderrezak, S. (2024). Comparative study of pollination efficiency and yield components of melon crops by three bee species in algeria. *Journal of the Kansas Entomological Society, 97*(1), 1-12. doi: 10.2317/0022-8567-97.1.1

Naccari, V., Trevisi, G., Naccari, C., Ferrara, G., Bava, R., & Palma, E. (2024). Poisoning due to Spinosad in honey bees: toxicological report. *Journal of Apicultural Research, 63*, 1-8. doi: 10.1080/00218839.2024.2425913

Pham-Delègue, M. H., Decourtye, A., Kaiser, L., & Devillers, J. (2002). Behavioural methods to assess the effects of pesticides on honey bees. *Apidologie, 33*(5), 425-432. doi: 10.1051/apido:2002033

Pinheiro, J. N., & Freitas, B. M. (2010). Efeitos letais dos pesticidas agrícolas sobre polinizadores eperspectivas de manejo para os agroecossistemas brasileiros. *Oecologia Australis, 14*(1), 266-281.

Potts, S. G., Biesmeijer, J. C., Kremen, C., Neumann, P., Schweiger, O., & Kunin, W. E. (2010). Global pollinator declines:

trends, impacts and drivers. *Trends in Ecology and Evolution, 25*(6), 345-353. doi: 10.1016/j.tree.2010.01.007

R Core Team (2022). *R: uma linguagem e ambiente para computação estatística.* R Foundation for Statistical Computing.

Ratnakar, V., Koteswara, R. S. R., Sridevi, D., & Vidyasagar, B. (2017). Sublethal lethal exposure of certain newer insecticides molecules to honeybee, *Apis mellifera* Linnaeus. *Indian Journal of Pure & Applied Biosciences, 5*(4), 641-646. doi: 10.18782/2320-7051.5234

Ribeiro, M. D. F., Silva, E. M. S. D., Lima, I. D. O., & Kiill, L. H. P. (2015). Honey bees (*Apis mellifera*) visiting flowers of yellow melon (*Cucumis melo*) using different number of hives. *Ciência Rural, 45*(10), 1768-1773. doi: 10.1590/0103-8478cr20140974

Rosa, J. M., Arioli, C. J., Nunes-Silva, P., & Garcia, F. R. M. (2019). Disappearance of pollinating bees in natural and agricultural systems: is there an explanation?. *Revista de Ciências Agroveterinárias, 18*(1), 154-162. doi: 10.5965/223811711812019154

Salgado, V. L. (1998). Studies on the mode of action of spinosad: insect symptoms and physiological correlates. *Pesticide Biochemistry and Physiology, 60*(2), 91-102. doi: 10.1006/pest.1998.2332

Shannon, B., Walker, E., & Johnson, R. M. (2023). Toxicity of spray adjuvants and tank mix combinations used in almond orchards to adult honey bees (*Apis mellifera*). *Journal of Economic Entomology, 116*(5), 1467-1480. doi: 10.1093/jee/toad161

Shi, T., Jiang, X., Cao, H., & Yu, L. (2023). Exposure to sublethal concentrations of thiacloprid insecticide modulated the expression of microRNAs in honeybees (*Apis mellifera* L.). *Ecotoxicology and Environmental Safety, 264,* 115499. doi: 10.1016/j.ecoenv.2023.115499

Silva, I. P., Oliveira, F. A. S., Pedroza, H. P., Gadelha, I. C. N., Melo, M. M., & Soto-Blanco, B. (2015). Pesticide exposure of honeybees (*Apis mellifera*) pollinating melon crops. *Apidologie, 46*(6), 703-715. doi: 10.1007/s13592-015-0360-3

Sousa, R. M., Aguiar, O. S., Andrade, A. B. A., Medeiros, A. C., & Maracajá, P. B. (2013). Densidade de colméias com abelhas africanizadas (*Apis mellifera* L.) para polinização da cultura do melão (*Cucumis melo* L.) no estado do Ceará-Brasil. *ACTA Apicola Brasilica, 1*(1), 9-12. doi: 10.18378/aab.v1i1.3586

Souza, A. A., Silva, E. K. S., Costa, E. M., Cardoso, T. A. L., Costa, J. A. D. M. A., Silva, D. M. T., & Oliveira Gondim, A. R. (2024). Survival and flight capacity of *Apis mellifera* after contact with residues of spiromesifen on melon leaves. *Sociobiology, 71*(4), e10753-e10753. doi: 10.13102/sociobiology.v71i4.10753

Therneau, T., Lumley, T. (2020). *Survival: Survival analysis, including penalised likelihood. R package version.* http://CRAN.Rproject.org/package=survival

Tschoeke, P. H., Oliveira, E. E., Dalcin, M. S., Silveira-Tschoeke, M. C. A., Sarmento, R. A., & Santos, G. R. (2019). Botanical and synthetic pesticides alter the flower visitation rates of pollinator bees in Neotropical melon fields. *Environmental Pollution, 251,* 591-599. doi: 10.1016/j.envpol.2019.04.133

Walker, E. K., Brock, G. N., Arvidson, R. S., & Johnson, R. M. (2022). Acute toxicity of fungicide insecticide adjuvant combinations applied to almonds during bloom on adult honey bees. *Environmental Toxicology and Chemistry, 41*(4), 1042-1053. doi: 10.1002/etc.5297

Williams, J. R., Swale, D. R., & Anderson, T. D. (2020). Comparative effects of technical-grade and formulated chlorantraniliprole to the survivorship and locomotor activity of the honey bee, *Apis mellifera* (L.). *Pest Management Science, 76*(8), 2582-2588. doi: 10.1002/ps.5832

Wu, W. Y., Liao, L. H., Lin, C. H., Johnson, R. M., & Berenbaum, M. R. (2023). Effects of pesticide-adjuvant combinations used in almond orchards on olfactory responses to social signals in honey bees (*Apis mellifera*). *Scientific Reports, 13*(1), 15577. doi: 10.1038/s41598-023-41818-7