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## TOXICITY OF INSECTICIDES TO *FRANKLINIELLA* *INVASOR* (THYSANOPTERA: THIRIPIDAE) UNDER LABORATORY CONDITIONS

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### ABSTRACT

Mango cv. 'Ataulfo' is perhaps the most popular mango produced in Mexico. The presence of high densities of thrips in mango blossoms, mainly the species *Frankliniella invasor* Sakimura, has been linked to yield decline. Growers spray synthetic insecticides on a regular basis against thrips to reduce their numbers, but no studies on the effectiveness of these insecticides have been conducted. The present study was undertaken with the objective of assessing the toxicity of 4 insecticides commonly used by growers. Commercial formulations of spinosad, imidacloprid, malathion, and  $\alpha$ -cypermethrin were evaluated on adults of *F. invasor* under laboratory conditions. Six concentrations of each insecticide were assessed: 0.1, 1, 10, 100, 1000, and 10,000 ppm. Snap bean pods were submerged into the different concentrations of insecticides, dried on paper towels, and placed in a plastic container with the thrips adults. A completely randomized design with 10 replicates per treatment was performed. Probit analyses revealed that spinosad and  $\alpha$ -cypermethrin were the most toxic insecticides for *F. invasor* with estimated  $LC_{50}$  values of 0.413 and 0.636 ppm, respectively. No significant differences in toxicity were found between imidacloprid ( $LC_{50}$  = 23.013 ppm) and malathion ( $LC_{50}$  = 34.422 ppm). Mortality in control treatments (distilled water) was never higher than 14%. Our study suggests the use of spinosad and  $\alpha$ -cypermethrin as the best control for *F. invasor*. However, these results should be complemented with field evaluations before being recommended to mango growers.

Key Words: 'Ataulfo' mango, *Frankliniella*,  $\alpha$ -cypermethrin, spinosad, Mexico

### RESUMEN

El Mango cv. 'Ataulfo' es probablemente el mango más popular de México. La presencia de altas densidades de trips durante la floración, principalmente de la especie *Frankliniella invasor* Sakimura, ha sido vinculada a la baja productividad. Los productores asperjan insecticidas sintéticos de forma regular contra los trips para reducir sus poblaciones, pero no se han llevado a cabo estudios sobre la efectividad de esos productos. El presente estudio se llevó a cabo con el objetivo de evaluar la toxicidad de cuatro insecticidas comúnmente usados por los productores. Las formulaciones comerciales del espinosad, imidacloprid, malathion, and -cipermetrina fueron evaluados sobre adultos de *F. invasor* bajo condiciones de laboratorio. Fueron evaluadas seis concentraciones de cada insecticida: 0.1, 1, 10, 100, 1000, y 10,000 ppm. Para la evaluación fueron utilizados ejotes, los cuales fueron sumergidos en diferentes concentraciones de insecticidas, secados en papel toalla, y colocados en contenedores de plástico con los trips adultos. Se utilizó un diseño completamente al azar con 10 réplicas por tratamiento. El análisis Probit reveló que el espinosad y la  $\alpha$ -cipermetrina fueron los insecticidas más tóxicos a *F. invasor*, con  $CL_{50}$  estimadas de 0.413 y 0.636 ppm, respectivamente. No se detectaron diferencias significativas entre imidacloprid ( $CL_{50}$  = 23.013 ppm) y malathion ( $CL_{50}$  = 34.422 ppm). La mortalidad en el control (agua destilada) no fue más del 14% de los individuos. Nuestro estudio sugiere el uso del espinosad y la  $\alpha$ -cypermethrin para el control de *F. invasor*. Sin embargo, estos resultados deberán ser complementados con pruebas de campo antes de ser recomendados a los productores.

Palabras Clave: Mango 'Ataulfo', *Frankliniella*, spinosad, Mexico,  $\alpha$ -cypermethrin

Mango (*Mangifera indica* L.) is a highly popular tropical fruit and important agricultural commodity in the global market. Because of its exquisite taste, pleasant fragrance, and nutritional

value, it is termed the 'King of Fruits' (Tharathan et al. 2006). There are more than a thousand cultivars (= varieties) of mango (Mukherjee 1953) of which only around 30 are commercially impor-

tant and dominate mango plantations worldwide (Galán-Saúco 2009). 'Ataulfo' is a yellow-fruited cultivar, perhaps the most popular mango from Mexico among consumers. It was discovered serendipitously, growing freely at Tapachula, Chiapas, at the end of the 1950's (Magallanes-Cedeño 2004; Infante et al. 2011). Due to its wide acceptance in local and international markets, the popularity of this cultivar has increased rapidly over the last few years.

'Ataulfo' mango is considered the best agricultural option for farmers along the Pacific Coast; namely, the Soconusco region (Gehrke 2008). At present, there are about 18,000 ha planted with this cultivar (Magallanes-Cedeño 2004). In recent years, a complex of thrips has drawn the attention of growers because they appear in large numbers in mango blossoms. The complex consists of 7 species in the genus *Frankliniella*, with *Frankliniella invasor* Sakimura being the dominant species. Over 600 adults plus immature stages of *F. invasor* were found in a single mango inflorescence (Rocha et al. 2012). Although the pest status of *F. invasor* has not been established, these large numbers of thrips are associated with a gradual decline of mango yields from 15 to 4 tons per ha (Gehrke 2008).

According to Sakimura (1972), *F. invasor* is native to the Caribbean-Central American region, and it was first reported feeding on mango in Hawaii. Adults feed and reproduce on mango inflorescences, and insecticides are sprayed on a regular schedule during flowering to reduce thrips numbers (Rocha et al. 2012). However, there are no studies to document the toxicity of various insecticides on this species and the benefits of chemical insecticides are unknown. The objective of these experiments was to determine the toxicity of 4 insecticides (spinosad,  $\alpha$ -cypermethrin, imidacloprid and malathion) that are currently recommended and used in mango orchards against the flower thrips *F. invasor*. We determined and compared the median lethal concentrations ( $LC_{50}$ ) of these insecticides on the adult stage of *F. invasor* under laboratory conditions.

## Materials and Methods

During Dec 2012, when there were many mango flowers in the field, we collected numerous inflorescences infested with thrips from the mango orchard 'El Vergel' (N 14° 42' 06" -W 92° 18' 57"; 25 m asl). The samples were collected early in the morning (usually before 7:00 am), and each inflorescence was placed in a labeled plastic bag. Inflorescences were immediately taken to the laboratory where *F. invasor* adults were separated in plastic vials using an aspirator. Adult thrips were separated in batches of 10 individuals to be subsequently used in the bioassays. *Frankliniella invasor* was distinguished from other species

by the abdominal shadings (paired lateral spots) that are characteristic of this species (Mound & Marullo 1996).

Commercial insecticides were purchased at agrochemical local stores. In the bioassays we used the following products: spinosad (Spintor® 12 CS; Dow AgroSciences),  $\alpha$ -cypermethrin (Arrivo® 200 EC; FMC Agroquímica de México), imidacloprid (Confidor® 350 CS; Bayer CropScience), and malathion (Malathion® 1000 EC; Agroquímica Tri-dente, México). Before running the bioassays, 6 concentrations of each insecticide were prepared using distilled water: 0.1, 1, 10, 100, 1,000, and 10,000 ppm. The toxicity of insecticides on *F. invasor* adults was evaluated in bioassays according to the following protocol.

Fresh snap bean pods (*Phaseolus vulgaris* L.) were obtained from the supermarket. In the laboratory they were rinsed with tap water, dried on paper towels, and chopped in pieces of approximately 5 cm long.

Insecticides were prepared according to the established concentrations mentioned above and the solutions were placed in glass containers.

Different concentrations of insecticides were applied to different groups of bean pods by immersion for 5 min, and subsequently, dried on paper towels for about 15 min. The same procedure was applied to the control, except that bean pods were immersed in distilled water only.

A thin layer of liquid paraffin was applied to both ends of the bean pods to seal them.

Bean pods treated with insecticide were individually placed inside 30 mL transparent plastic cups (3 cm diam).

Ten thrips adults of *F. invasor* were placed inside each plastic cup with the appropriate green bean pod coated with insecticide. There were 10 replicates per concentration plus the control. Cups were left in the laboratory at a constant temperature of 28.3 °C and 60% RH.

Mortality was evaluated at 6 hours. Cups were opened, and thrips were considered dead when they were unable to stand upright or move after being probed for several seconds.

Living and dead thrips were separated and preserved in vials with 70% ethanol. They were mounted on slides using Hoyer's medium, and dried in an oven at 45 °C for 2 weeks. Then, the taxonomic identification of each specimen was corroborated by the taxonomic keys of Mound & Marullo (1996).

The concentration-mortality responses were determined using a regression log-Probit model (Finney 1971). A goodness of fit test using the Hosmer-Lemeshow  $\chi^2$  statistic was used to corroborate the approximation of the data to the Probit model (at the  $P < 0.05$  level) (Agresti 2002). The median lethal concentration ( $LC_{50}$ ) estimated to kill 50% of the tested population and 95% confidence intervals (CI) were calculated based on methods by Robert-

son & Priesler (1992). The failure of 95% confidence limits to overlap was used as a criterion to separate LC values between insecticides. When control mortalities reached more than 5%, the insecticide mortality rates were adjusted with control mortalities using the Abbott's procedure (Abbott 1925). The statistical package R (R Development Core Team 2013) was used for data analysis.

RESULTS

The response of *F. invasor* adults exposed to imidacloprid and malathion closely approximated the Probit model (last column of Table 1). In contrast, the dosage mortality equation obtained for spinosad and  $\alpha$ -cypermethrin, did not conform adequately to this model due to the over-dispersion of data. In models with binary response, i.e., Probit analysis, over-dispersion is a frequent problem that appears when variability in data is higher than expected. For this reason, data were adjusted through the quasi-likelihood approach (Agresti 2002), prior to the calculations of lethal concentrations for the different insecticides.

The dose response curves are presented in Fig. 1. A marked difference in thrips mortality among tested insecticides was observed. The dose response was nearly quadratic for spinosad ( $R^2 = 0.47$ ;  $P < 0.001$ ) and  $\alpha$ -cypermethrin ( $R^2 = 0.83$  ;  $P < 0.001$ ), while a linear response was obtained for imidacloprid ( $R^2 = 0.81$ ;  $p < 0.001$ ) and malathion ( $R^2 = 0.89$ ;  $P < 0.001$ ). The  $LC_{50}$  values and confidence limits for adults of *F. invasor* are presented in Table 1. Significant differences in  $LC_{50}$  values were observed in most comparisons of insecticides. Spinosad and  $\alpha$ -cypermethrin were the most toxic insecticides against *F. invasor*, with  $LC_{50}$  values of 0.413 and 0.635 ppm, respectively. No significant differences in toxicity were found between imidacloprid ( $LC_{50} = 23.013$  ppm) and malathion ( $LC_{50} = 34.422$  ppm). Spinosad and  $\alpha$ -cypermethrin at a concentration of 10 ppm killed 99% of the thrips adults. At 100 ppm and higher concentrations, it killed 100% of individuals. Imidacloprid and malathion did not kill 100% of individuals at any of the concentrations tested. Mortality in control treatments (distilled water) was never higher than 14%.

DISCUSSION

The median lethal concentration of insecticides is the most widely accepted basis for toxicity tests at a particular length of exposure (Nwani et al. 2010). Pesticides play an important role in modern agriculture. Therefore, it is very important to know the  $LC_{50}$  of pesticides before using them in the field (Raj et al. 2013). Considering that *F. invasor* is the most abundant species of thrips in mango orchards (Rocha et al. 2012), and apparently one of the factors contributing to mango productivity decline in Chiapas (Gehrke 2008), the lack of studies on this particular topic is unfortunate. As far as we know, this is the first study dealing with the toxicity of insecticides on *F. invasor* in the laboratory, and it could be an important starting point for further evaluations of other insecticides. The knowledge generated in this study of the toxicities of these insecticides to *F. invasor* will facilitate resistance monitoring for this pest (Eger et al. 1998).

Spinosad was one of the most toxic insecticides to the adults of *F. invasor*. This toxicity was evident even at the lowest doses used. At 0.1 ppm it caused 38% mortality and approximately 100% mortality at the dose of 10 ppm. Higher doses of spinosad produced 100% mortality in this species. Our results concur with those reported for other species of thrips in the same genus. For instance, in toxicity bioassays of spinosad against several species of flower thrips in Florida, namely, *Frankliniella occidentalis* (Pergrande), *Frankliniella tritici* (Fitch) and *Frankliniella bispinosa* (Morgan),  $LC_{50}$  values of 0.594, 0.31 and 0.72, were reported, respectively (Eger et al. 1998). Similarly, spinosad has been reported as a very effective insecticide for controlling *F. occidentalis* in horticultural vegetables in Australia (Kay & Herron 2010). A mortality as high as 98-100% was recorded for spinosad in laboratory bioassays when tested on *F. occidentalis* adults, whereas in field experiments, the population of larvae and adults of this species significantly decreased in comparison to the control (Kay & Herron 2010).

*Frankliniella invasor* was sensitive to  $\alpha$ -cypermethrin. This insecticide was effective in avocado and mango orchards of Florida when

TABLE 1. TOXICITY IN PPM OF INSECTICIDES AGAINST ADULTS OF *FRANKLINIELLA INVASOR* UNDER LABORATORY CONDITIONS. LETHAL CONCENTRATIONS ARE IN PPM.

Insecticide	N	Slope $\pm$ SE	$LC_{50}$ (95% CL)	$\chi^2$	$P > \chi^2$
Spinosad	100	0.862 $\pm$ 0.1045	0.413 a (0.061 - 0.881)	13.171	0.010
$\alpha$ -Cypermethrin	100	1.388 $\pm$ 0.1168	0.636 a (0.316 - 1.281)	12.813	0.012
Imidacloprid	100	0.856 $\pm$ 0.0564	23.013 b (16.768 - 31.452)	1.992	0.737
Malathion	100	1.018 $\pm$ 0.0613	34.422 b (26.447- 44.714)	0.729	0.947

Lethal concentrations ( $LC_{50}$ ) followed by the same lowercase letter are not significantly different if the confidence limits (CL = 95%) overlap.

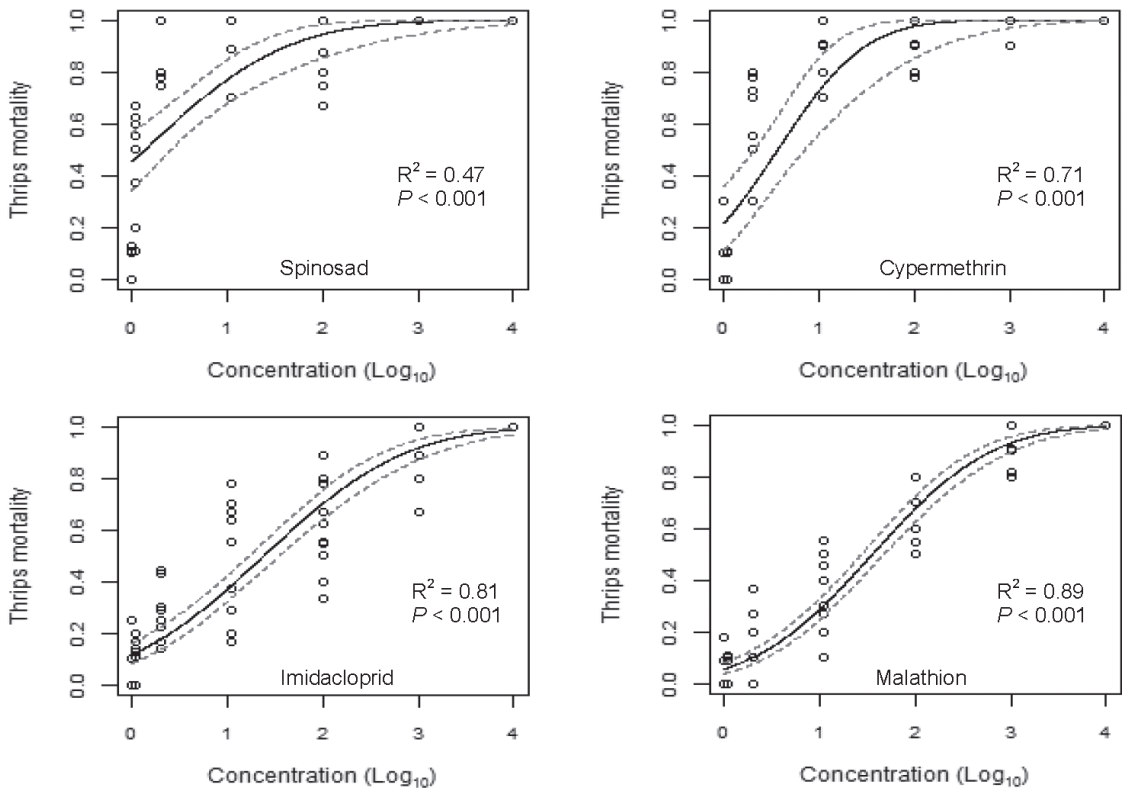


Fig. 1. Dose-Response Relationships in PPM of *Frankliniella invasor* Adults Exposed to Four Insecticides under Laboratory Conditions. The Two Dashed Lines Represent the Confidence Limits (95%) of the Slope (Solid Line).

sprayed on the flower thrips, *Frankliniella kel-liae* Sakimura and *F. bispinosa* (Peña et al. 2006). Regarding imidacloprid and malathion, bioassays revealed that they were similar in toxicity. Both insecticides were reported as very effective when applied to different species of thrips. Shibao et al. (2006) reported that imidacloprid was highly effective against *Scirtothrips dorsalis* (Hood) on grapes. Similarly, it has been successfully used for the control of the avocado thrips, *Scirtothrips perseae* Nakahara (Byrne et al. 2007). Malathion has been used to control *S. dorsalis* with good results and extensive persistence (Chandra et al. 2010).

Insecticides, including the ones under evaluation in this study, are sprayed by mango growers on a regular basis due to the presence of large numbers of thrips: i.e., *F. invasor*, during mango flowering (Rocha et al. 2012). Therefore, it is critical to have candidate insecticides for this insect. The present work could be the foundation for an eventual pest management program of thrips. However, further research is required to estimate the influence of biotic and abiotic factors on the performance of insecticides under field conditions. Therefore, large-scale experiments in mango orchards are needed to support the present findings. Effective and sustainable

management of *F. invasor* will only be achieved in the overall context of integrated pest management, with insecticide resistance management being a key component. Demirozer et al. (2008) recommended rotating the available insecticides in different chemical classes to avoid the development of resistant thrips. They emphasized that this integrated resistance management strategy should only be a component of a comprehensive integrated pest management program, and it is only recommended when multiple applications of insecticides are needed for thrips or other pests during the same cropping season.

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