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Evaluation of Systemic Chemicals for Avocado Thrips and Avocado Lace Bug Management

Year 2 of 3

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Benefit to the Industry

Insecticides are an important component of pest management in California avocado groves. In this study, we are evaluating the efficacy of systemic neonicotinoid insecticides against an established pest, the avocado thrips, and a newly introduced pest, the avocado lace bug. The potential benefits to the industry from the use of neonicotinoids are numerous. This class of insecticide has a novel mode of action, thereby minimizing the risk of cross-resistance between the neonicotinoids and those chemicals currently being used for avocado pest management. Neonicotinoids can be applied through the irrigation system, offering growers a very convenient method for treating their trees. One of the principal advantages of systemic applications of neonicotinoids may be the improved timing of treatments, which will allow growers to deal more effectively with pest outbreaks. The resulting decrease in the number of foliar applications will also reduce pressure for helicopter availability for those growers choosing to maintain foliar applications, and will also lessen the pressure for resistance development by adding another mode of action to the arsenal of chemicals available to growers.

Summary of Results to Date

Completion of Fallbrook Trial

During year one of this project (CAC Annual Report 2005), we evaluated the uptake of imidacloprid in two tree sizes located at a commercial avocado grove in Fallbrook. For up to one year following the insecticide applications, leaves were collected for bioassays against the avocado thrips and the avocado lace bug. In order to make a direct correlation between insecticide levels and insect mortality, imidacloprid concentrations were determined within the same leaves that were used for insect bioassays. Although imidacloprid was detected within both tree sizes, the results for large trees were not encouraging. The imidacloprid concentrations that were detected within the large trees did not reach sufficient levels to provide effective avocado thrips mortality in bioassays. The efficacy against the avocado lace bug was better,

owing to the greater sensitivity of this pest to the insecticide, and the higher concentrations of insecticide within the older leaves. The bioassay data for leaves sampled from the smaller trees were also most encouraging for the avocado lace bug, while data for the avocado thrips were not satisfactory. At 5-weeks post-application, the residues of imidacloprid in mature leaves provided excellent control of avocado lace bug. On the same sampling day, the younger leaves, which are more preferred by the thrips, had lower levels of toxicant and there was no mortality observed in bioassays. The only time we observed any significant mortality against thrips was when the insects were exposed to mature leaves. These leaves are not likely to be the preferred feeding site for thrips if there are young leaves also present on the same trees.

Since submitting our 2005 report, we have conducted 3 additional bioassays against the avocado lace bug (Oct 11 and Nov 15, 2005; Mar 1, 2006). On Oct 11, we observed over 70% mortality against the avocado lace bug feeding on leaves from both tree sizes, including the small trees that were treated with the half rate of Admire Pro (Figure 1). There was reduced mortality in the Nov 15 bioassay with leaves from the large trees, while the leaves sampled from the small trees treated with either the half or full label rates still returned excellent mortality (>85%) (Figure 2). Perhaps the most surprising result was the mortality that was observed in bioassays conducted in March 2006, 9 months after the trees were treated (Figure 3). In trees treated at the full label rate of Admire Pro (14 fl oz/acre), we observed almost 70% avocado lace bug mortality in bioassays. We also observed mortality in insects exposed to leaves from small trees that were treated at half the label rate, and in the large trees that were treated at the full label rate. This is a clear demonstration of the amazing persistence of imidacloprid and its potential value for pest management within crops where there is a need to combat pests that may infest groves over a prolonged period of time.

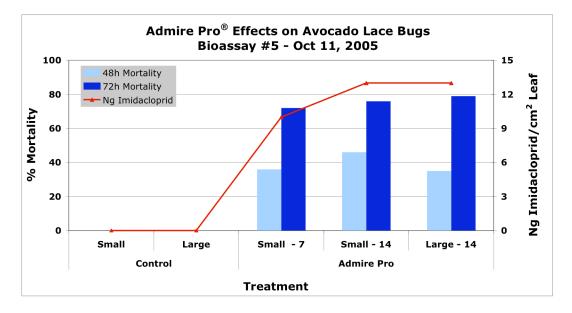


Figure 1. Bioassay of avocado lace bugs with leaves collected from trees treated with Admire Pro. Four leaves were sampled from each of 5 trees at each treatment. Small trees were treated with either 7 or 14 fl oz/acre, while large trees were only treated with the 14 fl oz rate. Imidacloprid levels were quantified in the same leaves used in Munger cell bioassays.

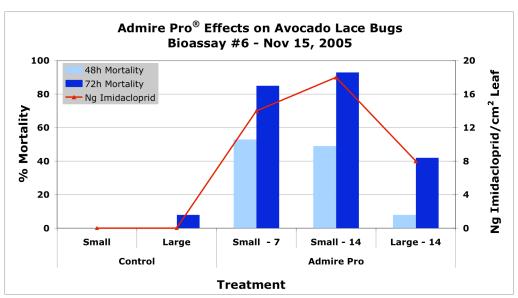


Figure 2. Bioassay of avocado lace bugs with leaves collected from trees treated with Admire Pro. Four leaves were sampled from each of 5 trees at each treatment. Small trees were treated with either 7 or 14 fl oz/acre, while large trees were only treated with the 14 fl oz rate. Imidacloprid levels were quantified in the same leaves used in Munger cell bioassays.

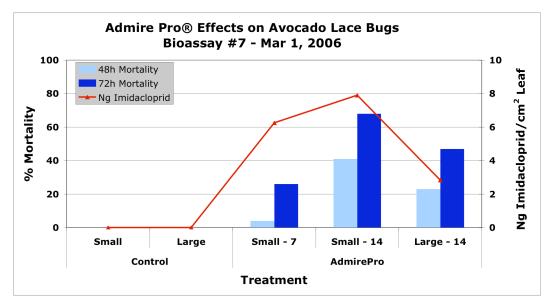


Figure 3. Bioassay of avocado lace bugs with leaves collected from trees treated with Admire Pro. Four leaves were sampled from each of 5 trees at each treatment. Small trees were treated with either 7 or 14 fl oz/acre, while large trees were only treated with the 14 fl oz rate. Imidacloprid levels were quantified in the same leaves used in Munger cell bioassays.

The results we obtained from the Fallbrook trial provided important information about the behavior of imidacloprid in avocado groves. The results also generated many interesting

questions such as why the rates of uptake were lower than expected (based upon our experiences with other systems – citrus and grapes).

Arising from our work in Fallbrook, we were able to establish the importance of imidacloprid for the management of the avocado lace bug. The insect prefers to feed on mature leaves where the highest imidacloprid residues are found. Although there may be problems in attaining sufficient concentrations of imidacloprid to protect very large trees from avocado thrips attack, the low levels of insecticide that move into trees of this size still seem to have an effect against the avocado lace bug.

Riverside County Trial

In a major collaborative effort between UC scientists, Bayer CropScience field and laboratory personnel, the CAC, and most importantly, the growers themselves, we have established three trials within commercial avocado groves in the Temecula region. The overall objectives of the trials are to compare the uptake of imidacloprid (applied as Admire Pro at 14 fl oz/acre via sprinkler irrigation systems) under different conditions of soil type and irrigation. The treatments were applied in late March/early April by Bayer CropScience personnel (Manuel Jimenez and Ed Ishida). The tree ages at all three sites were estimated to be 30 years old. Our focus at these sites has been on uptake into larger trees because these represent the greatest challenge to the effective deployment of neonicotinoids within avocado groves. The soil type at each location was determined using the Web Soil Survey facility provided by the USDA Natural Resources Conservation Service. In addition, during the process of soil sampling at each site, we examined the soil profile in the region of sample data trees. In Table 1, we have summarized the main features of the soils typical of the three sites that had an impact on imidacloprid availability for uptake through the tree roots.

Table 1. Soil types at the three trial sites located west of Temecula in Riverside County. Data were obtained using the Web Soil Survey facility provided by the USDA Natural Resources Conservation Service.

Site	Soil Name	Soil Texture	% Organic Matter	% Clay Content
P68	Lodo rocky loam	Gravelly loam	1 - 6%	18-35%
GGG	Lodo rocky loam	Gravelly loam	1-6%	18-35%
P37	Fallbrook rocky	Sandy loam	0.5 - 1%	10-20%
	sandy loam			

Summary of Trial Objectives

- 1. At all three sites, the Admire Pro was injected within 6 hours of the irrigation water being switched on (the "early" injection schedule). The injection period was at least 1 hour, and the irrigation was continued for an additional 14 hours as per the grower's normal irrigation practice.
- 2. At one of the three sites (P68), an additional injection was conducted on a separate group of trees at 7 hours <u>before</u> the end of the irrigation period (the "late" injection schedule). The injection period was at least 1 hour to ensure a minimum of 6 hours watering before

the end of the irrigation cycle. This allowed us to compare imidacloprid uptake using early versus late injections.

- 3. At two sites (P37 and P68), we assessed the impact of leaf litter on the uptake of imidacloprid into trees. The leaf litter was removed from a number of trees <u>before</u> the insecticide was injected into the irrigation lines, and the uptake compared with neighboring trees in the same grove where the leaf litter was left undisturbed.
- 4. The movement/persistence of imidacloprid was determined in the soil at 1 and 3-months post-treatment (Bayer to analyze soil cores data not provided in this report).
- 5. The distribution of imidacloprid in the flower, nectar and pollen of treated trees will be determined (UCR and Bayer to analyze data not provided in this report).

Insecticide Applications

Admire Pro was applied at each location by Manuel Jimenez and Ed Ishida of Bayer CropScience. The application strategy is summarized in Figure 4. Trees at P68 were treated on March 30 (late injection) and April 4 (early injection). Trees at P37 and GGG were treated on March 31.

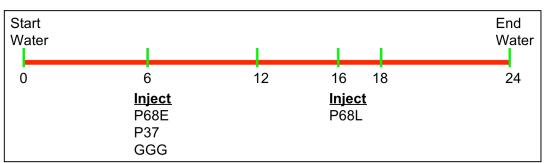


Figure 4. Injection protocol for the application of Admire Pro at the three trial sites located west of Temecula in Riverside County. The full label rate (14 fl oz/acre) was applied either early (at least 1 hour injection at 6 hours after the irrigation was commenced) or late (to ensure at least 6 hours of water before the end of the24 hour irrigation cycle).

<u>P68</u>

At this site, we tested two objectives – the timing of the Admire Pro injection during the chemigation cycle, and the impact of leaf litter on the uptake of imidacloprid. A flower study was also conducted at this location, the results of which will be presented at a later date.

Apart from two trees sampled at 6-weeks post-treatment, the overall level of imidacloprid uptake was very poor at the P68 site. The timing of the Admire Pro injection during the 24 h chemigation cycle had no major impact on uptake (Figure 5). Raking the heavy leaf litter from the sprinkler treatment area under the trees did not improve the uptake at this location (Figures 6 & 7). At week 11 in the P68 "early inject" trees, the levels of imidacloprid measured in leaves of three raked trees were higher than in the unraked trees (Figure 7). However, threshold concentrations required to generate even minimal thrips mortality in Munger cell bioassays (20

ng imidacloprid/cm² leaf) were not reached in either condition. We can conclude that there was no dramatic impact of leaf litter removal on imidacloprid uptake.

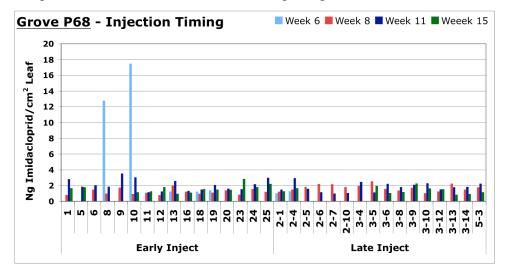


Figure 5. Impact of injection timing on imidacloprid uptake. During the 24 hour chemigation cycle, the Admire Pro (at 14 fl oz/acre) was either injected at 6 hours after irrigation was begun (Early Inject) or at 7 hours before the end of the irrigation cycle (Late Inject). For each tree, there are four data points, representing the mean imidacloprid concentrations in 10 young leaves/tree measured at four times during the trial (6, 8, 11 and 15 weeks after chemigation). The numbers on the x-axis represent the tree identification numbers.

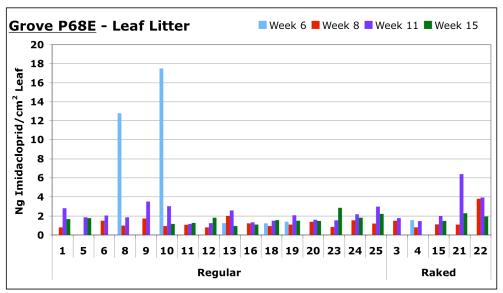


Figure 6. Impact of leaf litter on imidacloprid uptake into the P68 "Early Inject" trees. The leaf litter under five trees was removed from the sprinkler treatment area before the insecticide was injected with 14 fl oz/acre Admire Pro. For each tree, there are four data points, representing the mean imidacloprid concentrations in 10 young leaves/tree measured at four times during the trial (6, 8, 11 and 15 weeks after chemigation). The numbers on the x-axis represent the tree identification numbers.

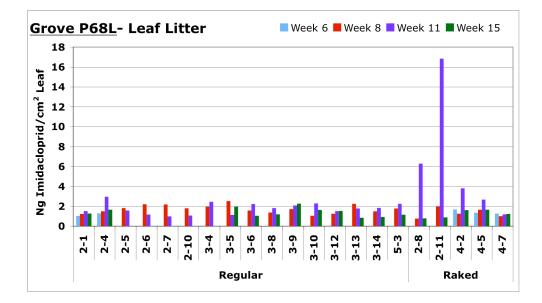


Figure 7. Impact of leaf litter on imidacloprid uptake into the P68 "Late Inject" trees. The leaf litter under five trees was removed from the sprinkler treatment area before the insecticide was injected with 14 fl oz/acre Admire Pro. For each tree, there are four data points, representing the mean imidacloprid concentrations in 10 young leaves/tree measured at four times during the trial (6, 8, 11 and 15 weeks after chemigation). The numbers on the x-axis represent the tree identification numbers.

<u>P37</u>

At this site, where trees were treated using the "early injection" strategy (Figure 4), we also investigated the impact of leaf litter on the uptake of imidacloprid (Figure 8). Again, the leaf litter was raked from the sprinkler treatment area under 5 trees, and the uptake compared with trees where the leaf litter was left undisturbed. Thresholds required to generate avocado thrips mortality in bioassays were not reached until late (15 weeks post-treatment) in the sampling program when leaves were fully expanded and had started to harden off.

<u>GGG</u>

At this site, the Admire Pro (14 fl oz/acre) was also injected early in the chemigation cycle, and the leaf litter was left undisturbed under all trees included in the sampling program. During the early weeks of monitoring, there were very low levels of imidacloprid being deposited in the young flush leaves (Figure 9). At week 15, the residue levels had improved considerably due to the increased maturity of the leaves at this time.

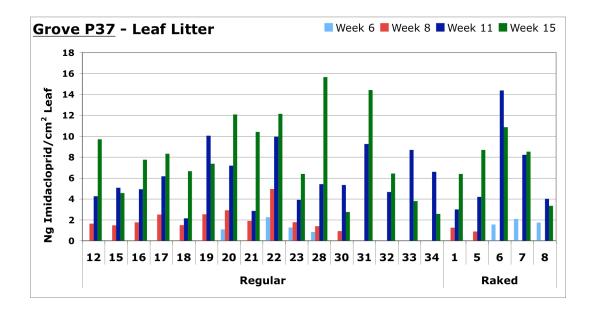


Figure 8. Impact of leaf litter on imidacloprid uptake into the P37 trees. The leaf litter under five trees was removed from the sprinkler treatment area before the insecticide was injected with 14 fl oz/acre Admire Pro. For each tree, there are four data points, representing the mean imidacloprid concentrations in 10 leaves/tree measured at four times during the trial (6, 8, 11 and 15 weeks after chemigation). The numbers on the x-axis represent the tree identification numbers.

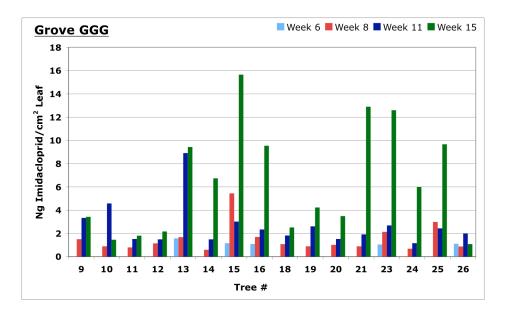


Figure 9. Impact of leaf litter on imidacloprid uptake into the GGG trees. The leaf litter under five trees was removed from the sprinkler treatment area before the insecticide was injected with 14 fl oz/acre Admire Pro. For each tree, there are four data points, representing the mean imidacloprid concentrations in 10 leaves/tree measured at four times during the trial (6, 8, 11 and 15 weeks after chemigation). The numbers on the x-axis represent the tree identification numbers.

Leaf Age

On Aug 18 (19 weeks post-treatment), leaves of different ages (young flush versus fully developed, mature leaves) were sampled from trees at each site to compare imidacloprid residues. The attractiveness of leaves to avocado thrips and avocado lace bugs varies with leaf age, and as a consequence, differences in the concentrations of imidacloprid in leaves of different ages could have an important impact on the management of these pests. Despite the poor imidacloprid uptake into the trees at the P68 site (in both Early and Late Injection treatments), we were still able to show that the residues of imidacloprid were higher in the mature leaves (Figure 10). At GGG and P37, the differences in residue levels between the young and mature leaves were even more pronounced.

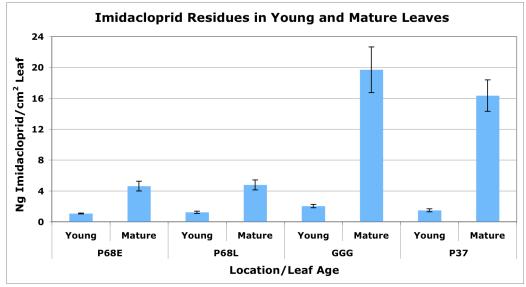


Figure 10. Impact of leaf age on imidacloprid uptake. The timing of the Admire Pro injection during the chemigation cycle (P68E = Early, P68L = Late) had no effect on uptake into either the young or mature leaves. Similarly, the grove location had no impact on uptake into young leaves. There was, however, a significant difference between the residues of imidacloprid in the mature leaves at both the P37 and GGG sites and the P68 site. Each bar represents the mean residue levels for 16 trees measured 18 weeks after the chemigation. For each tree, the imidacloprid residues were determined in 10 leaves.

<u>Bioassays</u>

The residues of imidacloprid in the mature leaves at the P37 site were varied and resulted in expected differences in mortality when avocado lace bugs were exposed to them (Figure 11). The threshold of activity for imidacloprid in this series of bioassays was close to 10 ng/cm² leaf. These data are consistent with the data we obtained from the bioassays conducted during the Fallbrook trial (Figures 1-3 and 2005 CAC Annual Report).

24c Admire Pro Survey

In 2006, Admire Pro use within avocado groves was permitted under a special 24c label. This afforded us the opportunity to assess the efficacy of Admire Pro treatments in a broader range of sites than was originally possible with our trial in Riverside County. While the latter trial sites permitted us to examine the impact of several variables (leaf litter, injection timing, etc) on the

uptake of imidacloprid, the 24c sites were more representative of how things might occur under prevailing agronomic conditions and across the variability of commercial avocado growing sites in California. For our monitoring study, we chose sites in Riverside, Ventura and Santa Barbara Counties. Although our choice of location within each county was limited to those growers who made the decision to apply Admire Pro (we had no control over their final decision to use the product), we did get a good cross-section of tree sizes and growing conditions for our study. The sampling program was difficult to establish owing to the quite variable application dates across locations. However, as a standard rule, we tried to take the first leaf samples within the 4-6 week window following application. The monitoring effort is still ongoing and we present here preliminary data for one location in Riverside County. These early data provide important insight into the dynamics of imidacloprid uptake in avocado groves and support many of our findings from the 2006 trial in Temecula. We anticipate that analysis of all our 2006 survey data will be even more informative.

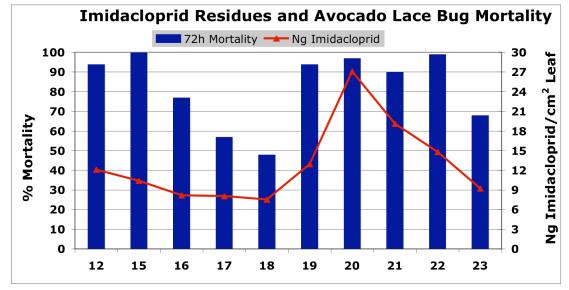


Figure 11. Bioassay of avocado lace bugs with leaves collected from trees treated with Admire Pro. Five mature leaves were sampled from each of 10 trees. Imidacloprid levels were quantified in the same leaves used in Munger cell bioassays. The numbers on the x-axis represent the tree identification numbers at the P37 site.

Riverside County – 24c Site 1

Description of Site: 3-year old Hass on Escondido fine sandy loam with 0.5-1% organic matter in the top 5" layer of soil. Sample trees (n=20) were on flat ground near the injection site. The trees were treated with 14 fl oz Admire Pro on June 6, 2006. The leaves that were sampled were not fully developed, as we believe these leaves to be most attractive to avocado thrips feeding.

At three weeks post-treatment, we detected imidacloprid in young leaves sampled from all trees (Figure 12). Although we observed variation in the titers of insecticide measured, there was good consistency in the uptake levels between trees. Ten trees were chosen from the original 20 trees for the specific investigation of two key objectives. First, was the uptake of imidacloprid greater into mature fully-developed leaves versus younger flushing leaves? And second, would

the residues present in the leaves have a significant impact on avocado thrips and avocado lace bugs feeding on them?

To address the first objective concerning age-related differences in uptake, we conducted a second set of samples on July 13 (5 weeks post-treatment). Five young and five mature leaves were sampled from each of the 10 trees for residue measurements. There was a dramatic difference (3- to 13-fold) in the concentrations of imidacloprid present in leaves of different ages, with the highest concentrations present in the mature leaves (Figure 13). At the time of this sample, a bioassay was conducted to determine the effects of imidacloprid residues in mature leaves against avocado lace bugs (Figure 14). Mortality reached 100% in all bioassays after 72 h exposure to the leaves in Munger cells, confirming the suitability of Admire Pro for the protection of young trees against avocado lace bug infestations.

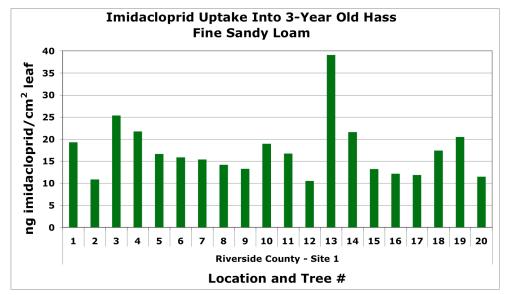


Figure 12. Imidacloprid residues in young leaves collected from avocado trees 3 weeks after they were treated with 14 fl oz Admire Pro. Five leaves were sampled from each tree and imidacloprid residues determined by ELISA. The numbers on the x-axis represent the tree identification number.

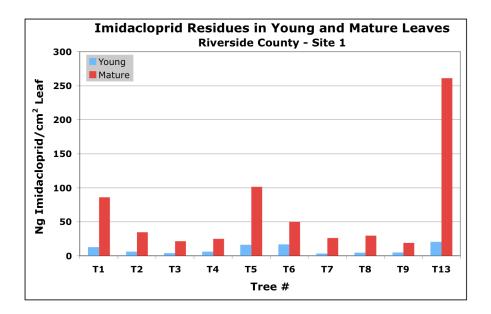


Figure 13. Impact of leaf age on imidacloprid uptake. There was a significant difference between the residues of imidacloprid in the young and mature leaves, with higher concentrations in the older leaves. Each bar represents the mean residue levels for 10 trees measured 5 weeks after the chemigation. For each tree, the imidacloprid residues were determined in either 5 young or 5 mature leaves. The numbers on the x-axis represent the tree identification number.

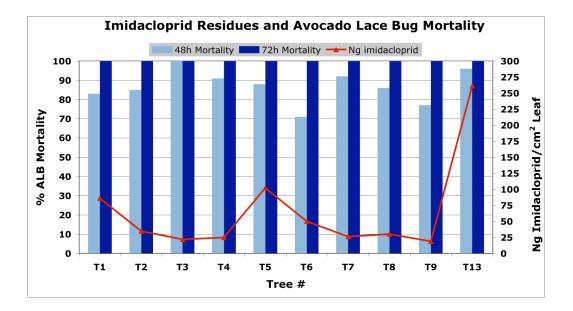


Figure 14. Bioassay of avocado lace bugs with leaves collected from trees treated with Admire Pro. Five mature leaves were sampled from each of 10 trees. Imidacloprid levels were quantified in the same leaves used in Munger cell bioassays. The numbers on the x-axis represent the tree identification numbers.

An avocado thrips bioassay was conducted on July 27. At this time, the imidacloprid residue levels were higher than those measured two weeks earlier, indicating that there was still movement of imidacloprid into the trees. Mortality was assessed in these bioassays at 48 h and 72 h (Figure 15).

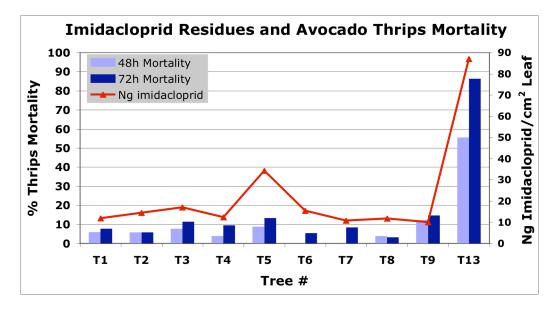


Figure 15. Bioassay of avocado thrips with leaves collected from trees treated with Admire Pro. Five young leaves were sampled from each of 10 trees. Imidacloprid levels were quantified in the same leaves used in Munger cell bioassays. The numbers on the x-axis represent the tree identification numbers.

The longer exposure time returned higher mortalities; however, apart from one tree (Tree #13) where the mortality was 86%, mortality did not generally exceed 15%. Our conclusion from this series of experiments is that the rate of delivery of imidacloprid to developing leaves does not keep pace with the rate of leaf growth, even in young trees. Although these leaves will eventually receive high levels of imidacloprid as they mature, they remain vulnerable to avocado thrips attack during leaf development.

Flower Study

In addition to monitoring the uptake of imidacloprid over time, at the request of Bayer CropScience we also initiated a flower study (they were unwilling to proceed with the 24c in 2006 unless we agreed to help obtain such flower data). In the flower study, we will determine the residues of imidacloprid in nectar, pollen and whole flower extracts. This study will complement our residue work on the leaves, and will provide important information on the distribution and concentration of imidacloprid within the various tissues of the trees. We are undertaking this study in the hope that our data will show a minimum likelihood of non-target impacts of the pesticide applications. This information will be used to guide the label recommendations for imidacloprid so that growers will be able to optimize the timing of their applications in order to maximize treatment efficacy.

Discussion

The results from our 2005 Fallbrook trial, and our ongoing Temecula trial, indicate several important points regarding the use of neonicotinoids within avocado groves. First, tree size is important. We observed better uptake into smaller trees, and levels of toxicant were reached that were effective against the avocado lace bug. While the insecticide was detected within the larger trees, it will be necessary to conduct further evaluations on the larger tree size to determine the feasibility of using imidacloprid as a pest management tool for avocado thrips control. While imidacloprid is taken up into the trees and did afford some thrips control (based on bioassay data for similar residue levels), satisfactory threshold levels were not reached on a consistent basis. We are especially concerned with the extremely slow uptake, even in younger trees. The latter suggests that many of the prevailing soil types may not be conducive to good uptake under current agronomic conditions. The soils at our three 2006 trial sites are rich in organic matter and clay content. These factors act as a strong sink for imidacloprid, reducing the ability of irrigation water to solubilize the chemical at sufficient concentrations necessary for rapid uptake. Second, quantitative differences exist between mature leaves (i.e. those that are present on the tree at the time of insecticide application) and young flush leaves. The leaves that receive the best protection from the insecticide treatment are those that are fully developed on the tree at the time of application. This favors the use of imidacloprid for avocado lace bug control. In addition to its exposure to feeding sites that are receiving the highest doses of imidacloprid, this pest is also more susceptible to the insecticide than the avocado thrips. Given its higher tolerance to imidacloprid, combined with its preference for feeding on leaves receiving only low doses of toxicant (i.e. young leaves), the avocado thrips will be difficult to manage with imidacloprid applied via chemigation.

Future Research Plans

During the second year of this project, we have established that there can be substantial differences between the residue levels of imidacloprid in young versus older leaves. While it seems that all leaves on the tree will receive imidacloprid, the rate of uptake does not keep pace with the rate of growth of leaves during flush. This is especially relevant in older trees, and must be considered when deciding on the suitability of imidacloprid for pest management in avocado groves. To overcome this problem of slow uptake, we would like to proceed with studies on alternative application strategies. Although not yet registered as a means of applying imidacloprid, it would be prudent to test the efficacy of trunk injections of this product. The main reason for testing this procedure is that it will bypass the soil factor in the uptake process. If we continue to see poor deposition of imidacloprid within the tree itself is governing the efficacy of the chemical. Our bioassay program established that imidacloprid is very effective against avocado thrips in bioassays. The use of trunk injections may be the way to attain threshold levels of toxicant necessary for control of avocado thrips,

It is also important to evaluate the uptake of other neonicotinoids. We are most interested in thiamethoxam (Platinum) and dinotefuran (Venom). These two neonicotinoids have greater water solubility (thiamethoxam is 8x, dinotefuran is 40x more soluble) than imidacloprid and

may be more readily available for uptake within soils that are high in organic matter. A comparison of soil treatments versus trunk injections for each of these three chemicals would provide valuable insight into the likely success of systemic treatments for pest management within the challenging conditions of avocado groves.

We will continue with our current trial in Temecula, and determine the longevity of residues within the trees. These data will complement the soil persistence study that Bayer CropScience is currently evaluating on our behalf. In 2007, we envisage a more comprehensive study on the distribution of neonicotinoids within the flowers, nectar and pollen. The latter study will provide important data necessary for formulating label recommendations that will allow growers to use these products when they are most needed.

Relevant Recent Publications

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