Evaluation of the impact of neonicotinoid insecticides on the glassy-winged sharpshooter *Homalodisca vitripennis* and its egg parasitoids

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ABSTRACT
A study was conducted to determine the toxic effects of systemic imidacloprid against the egg stage of the glassy-winged sharpshooter *Homalodisca vitripennis*, and its mymarid egg parasitoid *Gonatocerus ashmeadi*. During the development of the sharpshooter embryo within the egg, imidacloprid did not have a lethal effect, even at very high concentrations. However, upon emergence from the egg mass, the 1st instar nymph encountered residues of imidacloprid in the surrounding leaf tissues. There was an excellent dose-response between the imidacloprid concentration and emerging nymph mortality, giving an LC_{50} of 39 ng imidacloprid/cm^2 leaf. In the same way, *G. ashmeadi* did not succumb to toxic levels of imidacloprid during its development within the sharpshooter egg. However, parasitoid adults were sensitive to imidacloprid residues during emergence from the sharpshooter egg. The LC_{50} for parasitoid mortality was 66 ng imidacloprid/cm^2 leaf. In a survey of commercial citrus trees that were treated with imidacloprid, the mean residues of the insecticide within the leaves did not exceed the LC_{50} concentrations for either insect. Our work with imidacloprid has been completed. We will use the same techniques to evaluate the impact of thiamethoxam and dinotefuran on the emerging eggs and parasitoids.

INTRODUCTION
In a previous study, imidacloprid was shown to be toxic to sharpshooter nymphs, but only during emergence from the egg (Byrne and Toscano, 2005). During development, the imidacloprid did not penetrate the egg membrane at sufficient levels to cause toxicity. In this study, we are continuing to evaluate the effects of the neonicotinoids against the sharpshooter nymphs and its egg parasitoid, *Gonatocerus ashmeadi*. Neonicotinoid insecticides are the most widely used insecticides on citrus for the area-wide management of the glassy-winged sharpshooter. Dose-response data are being generated from bioassays to indicate threshold residue levels of insecticides that are lethal to nymphs and parasitoids as they emerge from the sharpshooter egg.

OBJECTIVES
1. Determine the relative toxicities of neonicotinoids (imidacloprid, thiamethoxam and dinotefuran) to the adult and egg stages of the glassy-winged sharpshooter
2. Determine the impact of neonicotinoids (imidacloprid, thiamethoxam and dinotefuran) on egg parasitoids of the glassy-winged sharpshooter

RESULTS
The uptake of imidalcoprid into excised cotton leaves was variable and ensured a broad range of concentrations to determine its impact against the developing embryo and emerging first instar *H. vitripennis*. Even at the highest concentrations of imidacloprid (almost 1 µg imidacloprid per cm^2 leaf), there was no mortality of the developing embryo. The only mortality that occurred was during the

emergence of the first instar from the egg mass. At the highest concentrations, the 1st instars died as soon as they broke through the egg sac. At lower doses, many of the nymphs emerged half way through the egg sac before succumbing to insecticide. There was an excellent dose-response, resulting in an LC_{50} of 39 ng imidacloprid per cm^2 leaf (Figure 1).

We observed a similar pattern of mortality in bioassays with the parasitoid. As with the sharpshooter nymphs, there was no toxic effect on the developing parasitoid within the confines of the sharpshooter egg. However, once the insect began to emerge, it encountered insecticide and there was a dose-response between the concentration of imidacloprid and mortality. The LC_{50} for the adult *G. ashmeadi* was 66 ng imidacloprid per cm^2 leaf, indicating its greater tolerance to the insecticide (Figure 1).

Imidacloprid concentrations were measured in the xylem and leaf tissues of citrus trees treated with 2.34 l ha^{-1} of 240 g imidacloprid $liter^{-1}$ SC (Figure 2). There was a steady increase in xylem fluid levels of imidacloprid up to 40 days after the application. At this time, the titers remained steady, and only declined after 110 days. Residues of imidacloprid within leaves were measured at 60 days after the initial treatment when the imidacloprid concentration within the xylem system had stabilized (Figure 2). The level of variation between the four trees was not significant ($F_{3,40} = 0.77$, $P = 0.52$). The apparent differences between the imidacloprid concentrations in leaves sampled from the north and south sides of the trees were also not significant ($F_{1,40} = 2.30$, $P = 0.14$). The combined leaf residue data for the four trees was used to generate a frequency distribution curve (Figure 3), which showed more clearly that the residues present within most of the leaves were lower than the LC_{50} values for the two insects.

CONCLUSIONS

In this study, we have provided further evidence that imidacloprid residues within leaves do not penetrate through the chorion of the sharpshooter egg. There was, however, a toxic effect on the emerging 1st instar nymph, which was most dramatic at higher insecticide concentrations within the leaves. Imidacloprid residues within the leaves had a similar effect against parasitoids. All parasitoids developed fully to the adult stage, and only upon emergence did they encounter insecticide residues. As with the emerging sharpshooter nymphs, there was a dose response between the levels of parasitoid mortality and imidacloprid concentrations present within the leaves.

In the treated citrus trees (Figures 2 and 3), measurements of imidacloprid were taken from leaves when the insecticide levels within the xylem system had stabilized. The distribution of imidacloprid levels within the leaves (Figure 3) clearly showed that the residues present would not have a dramatic effect on the emergence of either the sharpshooter nymph or the parasitoid. However, nymph populations have been shown to decline on treated citrus when concentrations of imidacloprid were as low as 3 ng/ml xylem fluid (Castle et al., 2005). Therefore, the greater impact of the insecticide treatment would occur on the sharpshooter nymphs when they began to feed on the xylem fluid since the imidacloprid concentrations were above threshold levels necessary to induce mortality (Figure 2).

G. ashmeadi is an important parasitoid of the glassy-winged sharpshooter. The success of this parasitoid against the sharpshooter has highlighted the importance of biological control in the management of the sharpshooter (Triapitsyn et al., 1998). There is much interest in enhancing biological control through the search and release of additional egg parasitoids, both *Gonatocerus* species and others (Hoddle and Triapitsyn, 2003, 2004; Morgan et al., 2000; Morse and Stouthamer, 2005). The success of these efforts could depend in part upon the impacts that insecticide treatments have on their survival. One of the primary objectives of the release program is to enhance the impact of biological control during the early part of the year when the first generation is developing. However, it is at this time that the first systemic treatments of imidacloprid are applied. It is important, therefore, that the interaction between the two management systems is fully understood, so that the benefits of both systems can be maximized. Our data show that there is great potential for the integration of the two systems – imidacloprid has no toxic effect on the sharpshooter embryo during its development within the egg, ensuring that the parasitoid will be able to develop to the adult stage. The greater tolerance of the parasitoid to the insecticide will further ensure that the leaf residues of insecticide will have a lesser impact on its survival during emergence. We will continue with this work by evaluating potential non-target effects of thiamethoxam and dinotefuran on the egg parasitoids.

REFERENCES

Byrne, F.J., Toscano, N.C., 2005. Characterization of neonicotinoids and their plant metabolites in citrus trees and grapevines, and evaluation of their efficacy against the glassy-winged sharpshooter and the egg parasitoid *Gonatocerus ashmeadi*. In: Tariq, M.A., Blincoe, P., Mochel, M., Oswalt, S., Esser, T. (Eds.), Proceedings of the Pierce's Disease Research Symposium, 5-7 December 2005, San Diego, California, pp. 287-289.

Castle, S.J., Byrne, F.J., Bi, J.L., Toscano, N.C., 2005. Spatial and temporal distribution of imidacloprid and thiamethoxam in citrus and impact on *Homalodisca coagulata* Wells populations. Pest Manag. Sci. 61, 75-84.

Hoddle, M., Triapitsyn, S., 2003. Investigating the behavior and biology of exotic mymarid parasitoids released for glassy-winged sharpshooter control, and evaluating techniques for enhancement in the field. In: Tariq, M.A., Oswalt, S., Blincoe, P., Spencer, R., Houser, L., Ba, A., Esser, T. (Eds.), Proceedings of the Pierce's Disease Research Symposium, 8-11 December 2003, Coronado, San Diego, California, pp. 292-294.

Hoddle, M., Triapitsyn, S., 2004. Searching for and collecting egg parasitoids of glassy-winged sharpshooter in the central and eastern USA. In: Tariq, M.A., Oswalt, S., Blincoe, P., Ba, A., Lorick, T., Esser, T. (Eds.), Proceedings of the Pierce's Disease Research Symposium, 7-10 December 2004, Coronado, San Diego, California, pp. 339-341.

Morgan, D.J.W., Triapitsyn, S.V., Redak, R.A., Bezark, L.G., Hoddle, M.S., 2000. Biological control of the glassy-winged sharpshooter: current status and future potential. In: Hoddle, M.S. (Ed.), [Proceedings] California Conference on Biological Control, July 11-12, 2000, Mission Inn, Riverside, California, 205 pp.

Morse, J.G., Stouthamer, R., 2005. The *Anagrus epos* complex: a likely source of effective classical biological control agents for glassy-winged sharpshooter control. In: Tariq, M.A., Blincoe, P., Mochel, M., Oswalt, S., Esser, T. (Eds.), Proceedings of the Pierce's Disease Research Symposium, 5-7 December 2005, San Diego, California, pp. 373-375.

Triapitsyn, S.V., Mizell III, R.F., Bossart, J.L., Carlton, C.E., 1998. Egg parasitoids of *Homalodisca coagulata*. Florida Entomol. 81, 241-243.

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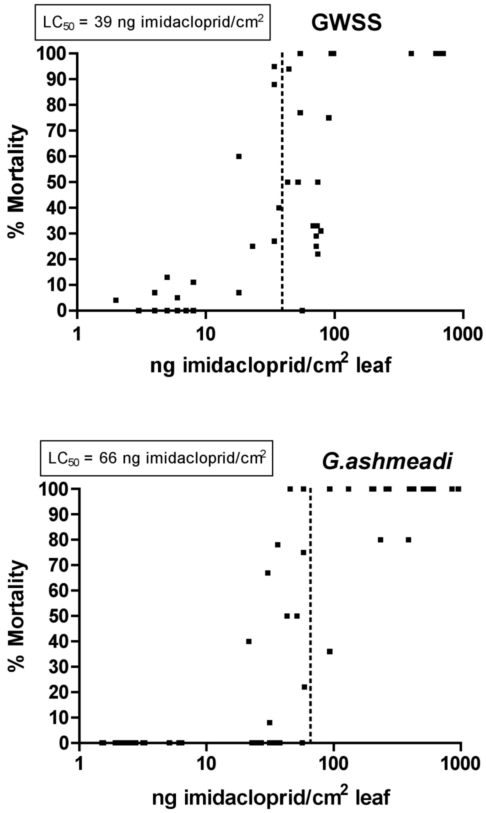

Figure 1. Dose response for GWSS nymphs and *G. ashmeadi* adults as they emerge from sharpshooter eggs that developed on imidacloprid-treated leaves. Data for the GWSS is copied from Byrne and Toscano (2005), and is included to show comparative toxicity of imidacloprid to emerging nymphs and parasitoid adults.

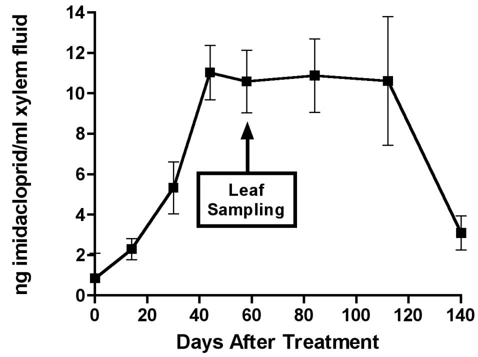

Figure 2. Temporal profiles of imidacloprid concentrations within the xylem fluid of citrus trees treated with 240 g l^{-1} imidacloprid. Each point represents the mean (± SEM) imidacloprid concentration in extracts from 12 trees. The arrow indicates the day on which leaves were sampled from four of the trees to determine the leaf concentrations of imidacloprid.

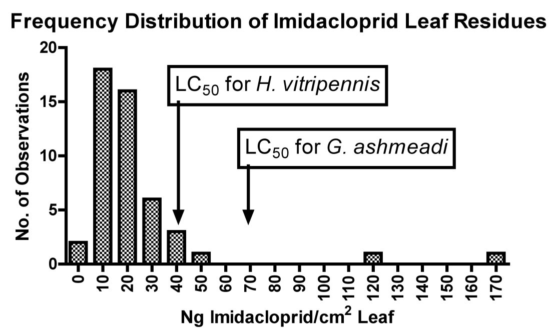

Figure 3. Frequency distribution of imidacloprid concentrations in citrus leaves sampled from trees treated with 240 g^{-1} l imidacloprid. The arrows indicate the LC_{50} values for *H. vitripennis* and *G. ashmeadi*.