

# **Entomology**

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Evaluation of Insecticide Treated Seeds to Control Green Peach Aphids

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**Index Words:** Green Peach Aphids, Kale, Seed Treatments

**Significance to Industry:** Film coating technology has been adapted to apply insecticides for seed coating. This technology has been used in other cropping systems to provide control of insect pests early in the crop production cycle. We evaluated the impact of film coating ornamental kale seeds with spinosad, fipronil, flonicamid, and chlorantraniliprole to suppress green peach aphid, *Myzus persicae*. These data indicate that treating seeds with flonicamid provides the resulting plants with protection against aphids. This technology greatly reduces the amount of pesticide need to control pest with no labor costs.

**Nature of Work:** Film coating is a technique originally developed for the pharmaceutical and confectionery industries to produce a colored, exterior finish. This method was adapted for seed coating, whereby a liquid formulation that includes a film-forming polymer is sprayed at a controlled rate onto a tumbling mass of seeds over time to achieve a uniform deposition of materials (2). Formulations may also contain plasticizers, colorants and other ingredients that are commercially available in aqueous suspensions. Seed treatments include application of certain compounds or processes, but in this paper refer to the application of insecticides to seeds. New chemistry insecticides, including fipronil, clothianidin and spinosad, were effective in controlling onion maggot (1). We evaluated the impact of film coating ornamental kale seeds with spinosad, fipronil, flonicamid, and chlorantraniliprole to suppress green peach aphid, *Myzus persicae*.

A seed lot of 'Osaka Formula Mix' ornamental kale was provided by Takii Seed Co for this project. Seed treatments were conducted at the New York State Agricultural Experiment Station, Geneva, NY. Spinosad (Entrust, Dow AgroSciences), fipronil (Regent 750, BASF), flonicamid (Aria, ISK) and chlorantraniliprole (E2Y45, Dupont) were applied at 0.25 mg ai/seed. All treatments were applied with a rotary pan seed treater (model R-6, GTG, Gilroy, CA) (2), and the commercial liquid binder, DISCO L-159 (Incotec Inc., Salinas, CA). Seed treatments were dispersed in the liquid film coating formulation and applied onto a 25 gram sample of seeds. Seeds were air dried overnight under ambient conditions, followed by a laboratory germination test as seed treatments may cause phytotoxicity (3).

Two trials were conducted to evaluate the efficacy of the seed treatments against green peach aphid, *Myzus persicae* (Sulzer). In both studies the kale seeds were sown in

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plug trays (200 cells per tray) and grown in a research glasshouse at the Texas A&M Agriculture Research and Extension Center at Overton. Nineteen days after seeding the seedlings were transplanted in 15.2 cm azalea pots. In the first trial, each plant was inoculated with five aphids when they were transplanted and again each sample period if no aphids were found on the plants. In the second trial the plants had a naturally occurring aphid infestation so aphids were not inoculated. The number of green peach aphids on each plant was counted approximately every two days.

For each trial, the mean number of aphids was subjected to analysis of variance (Randomized Complete Block AOV, Statistix 8). Means separation was accomplished by using Tukey's HSD test at the  $P < 0.05$  level.

**Results and Discussion:** In the first trial, none of the insecticide treated seeds resulted in a significantly lower aphid populations compared to non-treated seeds (Table 1). However, the flonicamid treated seeds had the lowest number of aphids on all but one sample period. In the second trial, seedlings had a natural infestation of aphids at the start of the trial. The flonicamid treated plants had significantly fewer aphids compared to the non-treated seeds on every sample period (Table 2). These data indicate that treating seeds with flonicamid provides the resulting plants with protection against aphids.

Additional studies are underway comparing the treatments against caterpillar pests. We are further evaluating the flonicamid treatment by evaluating both an increased rate (2x) and different seed coating materials to enhance efficacy.

The flonicamid seed treatment will also aid in greenhouse resistance management programs. Currently, many ornamental cabbage and kale producers make a preventative neonicotinoid insecticide application shortly after potting with the assumption that aphids will infest the plants. We are concerned about the overuse of the neonicotinoid insecticides resulting in the development of resistant pests. Flonicamid has a novel mode of action and there is less concern about cross-resistance.

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Table 1. Mean number of *Myzus persicae* per kale plant in the first trial.

Treatment	Days After Seeding							
	21	23	28	30	33	35	37	40
Non-treated	2.8 ab	4.8 ab	6.0 a	8.2 a	21.2 ab	24.4 ab	30.4 a	33.7 ab
Spinosad	5.9 b	11.5 b	18.0 b	26.8 b	35.5 b	36.6 b	33.4 a	33.7 ab
Fipronil	3.1 ab	7.7 ab	11.1 ab	10.8 a	25.4 ab	38.3 b	35.5 a	36.3 ab
Fonicamid	0.6 a	1.5 a	5.7 a	8.6 a	15.3 a	18.3 a	20.3 a	18.7 a
Chlorantraniliprole	4.1ab	6.4 ab	8.0 ab	9.9 a	20.5 ab	31.2 ab	37.8 a	49.9 b

Means within columns followed by the different letters are significantly different (Tukey's HSD,  $P < 0.05$ ).

Table 2. Mean number of *Myzus persicae* per kale plant in the second trial.

Treatment	Days After Seeding							
	21	24	27	30	34	38	41	44
Non-treated	36.8 c	43.5 c	47.5 c	38.1 b	39.9 b	30.3 b	34.9 bc	35.8 ab
Spinosad	24.7 bc	28.1 b	33.0 b	38.2 b	37.3 b	32.9 b	35.3 bc	35.9 ab
Fipronil	20.0 b	24.0 b	31.1 b	33.9 b	37.2 b	26.6 b	30.3 b	32.6 b
Fonicamid	2.3 a	1.7 a	3.6 a	4.6 a	8.9 a	5.8 a	8.5 a	9.4 c
Chlorantraniliprole	27.7 bc	28.8 b	37.7 bc	44.0 b	44.8 b	37.9 b	42.4 c	43.3 a

Means within columns followed by the different letters are significantly different (Tukey's HSD,  $P < 0.05$ ).

**Evaluation of Neonicotinoid Insecticides for Control of the Strawberry Rootworm, Part II**

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**Significance to Industry:** The strawberry rootworm, *Paria fragariae* Wilcox, continues to be the major insect pest of container azalea production in the southeastern United States. This small (1/8 inch diam) chrysomelid beetle damages plants by feeding on their foliage. The larvae develop in potting substrate but their feeding is not believed to cause economic damage. [For more information on strawberry rootworm biology in the southeastern U.S. see Boyd and Hesselein (1) and Hesselein and Boyd (2)]. In previously reported trials, several insecticides have killed beetles and/or prevented beetle damage. Stephenson (5) first reported an outbreak of strawberry rootworm in container grown azaleas in the Semmes, AL area in 1983. In a subsequent trial, Stephenson reported that sprays of Dursban, Ficam, Sevin and Orthene all reduced SRW populations by more than 85 percent. In a series of experiments, Hesselein and Boyd (2, 3, 4) found that Sevin 80 WSP, DuraGuard ME, DeltaGard, Scimitar, Discus, Tame, Decathlon, Talstar, Pylon, Orthene TT&O Spray97, Marathon II, Celero, and Safari all either killed and/or prevented damage by the strawberry rootworm as spray and/or drench treatments.

**Nature of Work:** The experiment was conducted at the Ornamental Horticulture Research Center; Mobile, AL. Insecticides used in the trial included Flagship (thiamethoxam), Scimitar (Lambda-cyhalothrin), BAS320 I (metaflumazone) and DuraGuard ME (chlorpyrifos). All insecticides were applied as a foliar spray; Flagship was also applied as a drench. Drenched plants were treated with 150 ml of formulated solution. Plants were treated on July 16, 2007. The experimental design was a completely randomized design with four replications of each treatment. Experimental units consisted of single one-gallon containerized azalea. Treatment efficacy was evaluated by removing a small similarly-sized branch from each experimental unit and immersing basal end of the stem in a capped, water-filled, #53 Aquapick tube inserted in a hole in the bottom of an eight-fl oz capped sample cup (arena). Three strawberry rootworm beetles were placed in each arena. Arenas were suspended on a wire covered laboratory bench for the duration of the evaluation. Stem removal took place according to the schedule outlined in table 1. Data collected consisted of a foliar damage rating based on the 1-12 rating system developed by Horsfall and Barratt where 1= 0 percent, 2= 0-3 percent, 3= 3-6 percent, 4= 6-12 percent, 5= 12-25 percent, 6= 25-50 percent, 7= 50-75 percent, 8= 75-88 percent, 9= 88-94 percent, 10= 94-97 percent, 11= 97-100 percent and 12= 100 percent of the foliage within the arena was damaged by strawberry rootworm feeding. Treated stems were evaluated for beetle damage prior to exposing stems to strawberry rootworm beetles to ensure there were

no differences among treatments. In addition to rating beetle feeding damage, numbers of dead and live beetles were counted. Data were collected one and three days after exposing beetles to treated foliage unless otherwise stated. Percent dead beetles ( $\text{dead}/(\text{dead} + \text{alive}) * 100$ ) and damage rating were analyzed and are presented in tables 2 and 3. Data were analyzed using ANOVA ( $P \leq 0.05$ ) and means were separated using Fisher's Protected LSD ( $\alpha = 0.05$ ).

**Results and Discussion:** Flagship sprays (4 and 6 oz), Scimitar (5 fl oz) and DuraGuard (50 fl oz) all effectively killed and/or prevented damage of the strawberry rootworm on one or more evaluation (table 1). In terms of killing beetles, none of these treatments could be distinguished from the untreated control from 14 DAT through the end of the trial. The Flagship drench treatments (4 and 6 oz) did not kill any more beetles than the control at any evaluation date. Scimitar had significantly greater percentage dead beetles than the control at 1 DAT and was indistinguishable from both rates of sprayed Flagship and DuraGuard. Both rates of sprayed Flagship had significantly greater percentage dead beetles than the control at 1, 4 and 7 DAT and were indistinguishable from each other, Scimitar at 1 and 4 DAT and DuraGuard at 1 and 7 DAT. DuraGuard had significantly more percentage dead beetles than the control at 1 and 7 DAT and was indistinguishable from Scimitar and both rates of sprayed Flagship at 1 DAT and both rates of sprayed Flagship at 7 DAT.

For damage ratings, Scimitar (5 fl oz) had lower damage ratings than the control 1, 4, 7, 14 and 21 DAT (table 2). Scimitar damage ratings were indistinguishable from both rates of Flagship sprays (4 and 6 oz) 1, 7 and 21 DAT and both rates of Flagship drenches (4 and 6 oz) at 7 and 21 DAT. Scimitar had higher damage ratings than the 4 oz rate of sprayed Flagship at 4 DAT and both rates of sprayed Flagship at 14 DAT. Flagship spray treatments were indistinguishable and had lower damage ratings than the control at 1, 7, 14, 21 and 42 DAT. At 64 DAT the 6 oz spray Flagship treatment had a lower damage rating than the control but more damage than would be commercially acceptable. Flagship drench treatments (4 and 6 oz) were indistinguishable and had lower ratings than the control 7, 21, 42, and 64 DAT. DuraGuard (50 fl oz) had lower damage ratings than the control at 1, 7 and 21 DAT. At 1 DAT DuraGuard was indistinguishable from Scimitar and both rates of sprayed Flagship. At 7 DAT DuraGuard was indistinguishable from Scimitar and all Flagship treatments. There were no differences among treatments at 28 and 84 DAT.

Scimitar proved effective at killing beetles 1 DAT but also prevented economically damaging feeding for at least a week following treatment. Flagship sprays but not drenches also proved effective at killing beetles for at least seven days in the case of the 6 oz rate. Flagship also prevented commercially unacceptable damage for at least 42 days in the case of both rates of sprayed Flagship and 64 days in the case of the 6 oz rate of Flagship as a drench. Commercially acceptable damage would encompass ratings under 4 ( $\leq 6\%$  damage). DuraGuard proved effective at killing beetles for at least 7 DAT.

Given the expense involved in drenching plants both in terms of insecticide used and labor, I would recommend that growers use Flagship as a spray rather than a drench unless there is some evidence that the drench killed rootworm larvae in potting substrate as well as on the foliage. Even with the added incentive of killing larvae most growers would probably opt for spray treatments, except where infestations involved only a small number of plants. Results from this trial indicate that Scimitar and Flagship would be two excellent insecticides to use in a rotation for controlling strawberry rootworm.

In the course of the trial, beetles that weren't killed were placed in a colony. Some of these beetles were reused in the trial. We have evidence that beetles exposed to Flagship treatments were able to recover and feed on untreated foliage. We first tested this hypothesis on beetles used in the 64 DAT evaluation (data not shown). In addition, we suspect that beetles were recovering and surviving in our colony after being exposed to foliage treated by Flagship prior to this date. Based on this evidence, beetles that are not exposed to a fatal dose of Flagship may be able to revive, fly to untreated plants and recover. On the other hand, beetles exposed to a sublethal insecticide dose are at increased risk of other mortality factors such as predation, starvation and desiccation, which may lead to their death before they are able to recover from insecticidal toxicity. These are questions that could be addressed in future studies.

In 2006, a preliminary report of a study evaluating neonicotinoid insecticides applied as drenches was reported in the SNA Proceedings (4). Unreported in that paper were the 56, 106 and 173 DAT results. Briefly, at 56 DAT, Marathon II (21.8 fl oz/ 100 gal  $\approx$  label rate of 244, 1-gal pots treated with 1.7 fl oz Marathon II) and Discus (120.3 and 167.7 fl oz/ 100 gal  $\approx$  label rates of 340 and 244, 1-gal pots treated with 13 fl oz Discus respectively) all kept beetle damage ratings below four. At 106 and 173 DAT, only the Marathon II treatment kept beetle damage below four (data not shown).

Marathon II and Discus rates reported in the 2006 paper were based on mathematical extrapolations, including conversions, of actual application volumes. As a result the rates presented are roughly 3% greater than the actual label rates. For Marathon II, the label rate for treating 244 1-gallon pots is 21.2 fl oz/ 100 gal; for Discus, the label rate for treating 340 and 244 1-gallon pots is 162.3 fl oz and 116.5 fl oz respectively. These rates are based on an application volume of 4.2 fl oz per 1-gallon container.

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Table 1. Mean percentage dead strawberry rootworm beetles per arena

Product	Oz or fl oz/ 100 gal	Method	1 DAT <sup>z</sup>	4 DAT	7 DAT	14 DAT	21 DAT	28 DAT	42 DAT	64 DAT <sup>v</sup>	84 DAT <sup>u</sup>
Untreated			8 <sup>y</sup> b <sup>x</sup>	17 b	8 b	17 NS	8 NS	8 NS	0 NS	8 NS	67 NS
Flagship	4	D <sup>w</sup>			17 b		50 NS		8 NS	33 NS	22 NS
Flagship	6	D			0 b		33 NS		17 NS	33 NS	17 NS
Flagship	4	S	100 a	83 a	67 a	42 NS	8 NS	8 NS	0 NS	17 NS	25 NS
Flagship	6	S	88 a	100 a	92 a	42 NS	50 NS	33 NS	8 NS	25 NS	27 NS
Scimitar	5	S	100 a	58 ab	8 b	17 NS	17 NS	0 NS			
BAS320 I*	16	S	0 b		8 b						
BAS320 I*	11.4	S	25 b		8 b						
BAS320 I*	8	S	0 b		0 b						
BAS320 I	8	S	0 b		0 b						
DuraGuard	50	S	100 a		92 a	42 NS	50 NS	25 NS			

\* Treatments included adjuvant BAS9084 at 32 fl oz per 100 gal

<sup>z</sup> DAT is Days after Treatment

<sup>y</sup> Mean percentage dead beetles per arena, dead/(dead+alive)\*100

<sup>x</sup> Means separated using Fisher's Protected LSD,  $P \leq 0.05$ ,  $\alpha = 0.05$ . Means followed by the same letter are not significantly different

<sup>w</sup> D = insecticide applied as a drench. S= insecticide applied as a spray

<sup>v</sup> Data collected six days after exposing beetles to treated foliage

<sup>u</sup> Data collected nine days after exposing beetles to treated foliage

Table 2. Mean damage rating caused by strawberry rootworm beetle damage per arena.

Product	Oz or fl oz/ 100 gal	Method	1 DAT <sup>z</sup>	4 DAT	7 DAT	14 DAT	21 DAT	28 DAT	42 DAT	64 DAT <sup>v</sup>	84 DAT <sup>u</sup>
Untreated			5 <sup>y</sup> a <sup>x</sup>	4.5 a	5.5 a	6.5 a	5.5 a	4.2 NS	5.2 a	7.5 a	4.8 NS
Flagship	4	D <sup>w</sup>			2.8 cd		2.5 c		3 b	4.5 bc	3.7 NS
Flagship	6	D			3.3 bc		2.8 c		2.8 b	3.8 c	5.5 NS
Flagship	4	S	2.2 b	1 c	2.3 cd	2.2 c	3 bc	3.5 NS	3.8 b	6.5 ab	5.8 NS
Flagship	6	S	2 b	1.5 bc	1.5 d	2.2 c	2.8 c	2 NS	3.5 b	5 bc	5.5 NS
Scimitar	5	S	2.2 b	2 b	2.8 cd	4.2 b	3.5 bc	3.8 NS			
BAS320 I*	16	S	4.5 a		4.8 ab						
BAS320 I*	11.4	S	4 a		4.8 ab						
BAS320 I*	8	S	4 a		5 a						
BAS320 I	8	S	4.8 a		5.5 a						
DuraGuard	50	S	2.2 b		2.5 cd	5 ab	4 b	4.2 NS			

\* Treatments included adjuvant BAS9084 at 32 fl oz per 100 gal

<sup>z</sup> DAT is Days after Treatment

<sup>y</sup> Mean damage rating using 12 point Horsfall-Barratt rating system

<sup>x</sup> Means separated using Fisher's Protected LSD,  $P \leq 0.05$ ,  $\alpha = 0.05$ . Means followed by the same letter are not significantly different

<sup>w</sup> D = insecticide applied as a drench. S= insecticide applied as a spray

<sup>v</sup> Data collected six days after exposing beetles to treated foliage

<sup>u</sup> Data collected nine days after exposing beetles to treated foliage

**Species Variation within the *Chrysobothris femorata* “complex” (Flatheaded Appletree Borer): Evidence and Implications of DNA Sequencing**

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**Index Words:** Buprestidae, Coleoptera, genetics, molecular systematics, speciation, wood boring beetle

**Significance to Industry:** Flatheaded apple tree borer (FHAB) beetle (*Chrysobothris femorata*) is a destructive pest of woody ornamentals in the southeastern United States, which is part of a species complex. Morphologically, species in this group are difficult to separate due to presence of intermediate forms. If currently described species can be confirmed and separated within the complex using DNA analysis, new more efficient methods to control FHAB can be explored and more accurate diagnostic tools for growers can be created.

**Nature of Work:** Pest status of FHAB beetles is well acknowledged by ornamental, fruit and nut bearing tree growers (2, 8). Larval feeding injury slows tree growth, causes trunk scarring and can lead to premature tree death, costing growers thousands of dollars in lost revenue (6). Currently, industry growers rely on chlorpyrifos (e.g., Dursban) to control FHAB. However, increased Federal regulation of organophosphate insecticides may result in discontinued use, prompting interest in other effective and sustainable control methods (1). As scrutiny of pesticides rises, the need for a more detailed knowledge of pest biology and behavior becomes critical to effective and economical pest management. Recognizing new species is the first step in this process.

Originally, the *C. femorata* “complex” was comprised of only four species: *C. femorata* (*sensu stricto*), *C. rugosiceps*, *C. adelpha* and *C. viridiceps* (3). Today, the complex is believed to be composed of at least twelve different morphospecies (5). Unfortunately, physical variation of these characters within this group is so extensive that intermediate forms are common and outward appearance alone may be insufficient to delimit species. Molecular data can be used to test for the existence of species divisions within this borer complex.

If currently described species within the *C. femorata* group can be validated using molecular techniques, each could be found to have different host plant preferences, seasonal biology and behaviors, and chemical pesticide susceptibilities. More efficient management and control strategies could be devised to reduce grower reliance on chemical pesticide applications. This would be expected to limit costs associated with FHAB management.

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Specimens for this study were collected in 2006 and 2007 from several states (i.e. IA, OH, KY, TN, GA, and TX) using purple panel traps coated with Pestick® (Phytotronics, Earth City, MO). Beetles were removed once each week and preserved in 95% ethanol. Pestick® was removed from the beetles with Histoclear™ (National Diagnostics, Somerville, NJ) before DNA extraction. Purified DNA was used as a template for polymerase chain reactions (PCR), in which the COX-I gene and other nuclear genes were amplified. PCR products were sequenced in both directions for each specimen, then aligned and compared to observe any differences. A phylogeny of the *C. femorata* complex was generated using PAUP (Sinauer Associates, Sunderland, MA) (Fig. 1). The objective of this research is to investigate nuclear and mitochondrial markers within the *C. femorata* complex to ascertain whether species can be reliably separated within this difficult-to-characterize group.

**Results and Discussion:** Variation in the COX-I gene sequences was significant, with up to eight separate maternal lineages, or groups, being evident in the reconstructed phylogeny. Two of the monophyletic groups correspond to *C. adelpha* and *C. viridiceps* as distinct morphospecies. Individuals of *C. femorata sensu stricto* were dispersed among the remaining six monophyletic lineages, with one also containing individuals of *C. rugosiceps*.

While both morphological and molecular data support placement of *C. adelpha* and *C. viridiceps* as distinct species, *C. rugosiceps* and *C. femorata* appear to be more closely related than previously reported. Surprisingly, *C. rugosiceps* is clustered within one of the inferred maternal lines with *C. femorata*. Both species originate from a common maternal line and in addition, both appear so similar that intermediate forms can make identification difficult.

Analysis of COX-I sequences supports the existence of several well-supported maternal lineages within the *C. femorata* complex, at least some of which could represent distinct species. Interestingly, *C. femorata* and *C. rugosiceps* share a maternal lineage and overlapping host preference (oak trees) (3, 5). Despite previous research dividing the *C. femorata* complex into twelve species (5), our data provides evidence for no more than eight distinct groups. Due to sampling limitations, specimens of western and northeastern species were unavailable for testing (4, 5). However, sampling in several habitats in multiple eastern states over a two-year period has increased the probability that any major southeastern species component of the complex is included in this study. If representatives of the other reported morphospecies can be collected and included, these could reveal additional lineages.

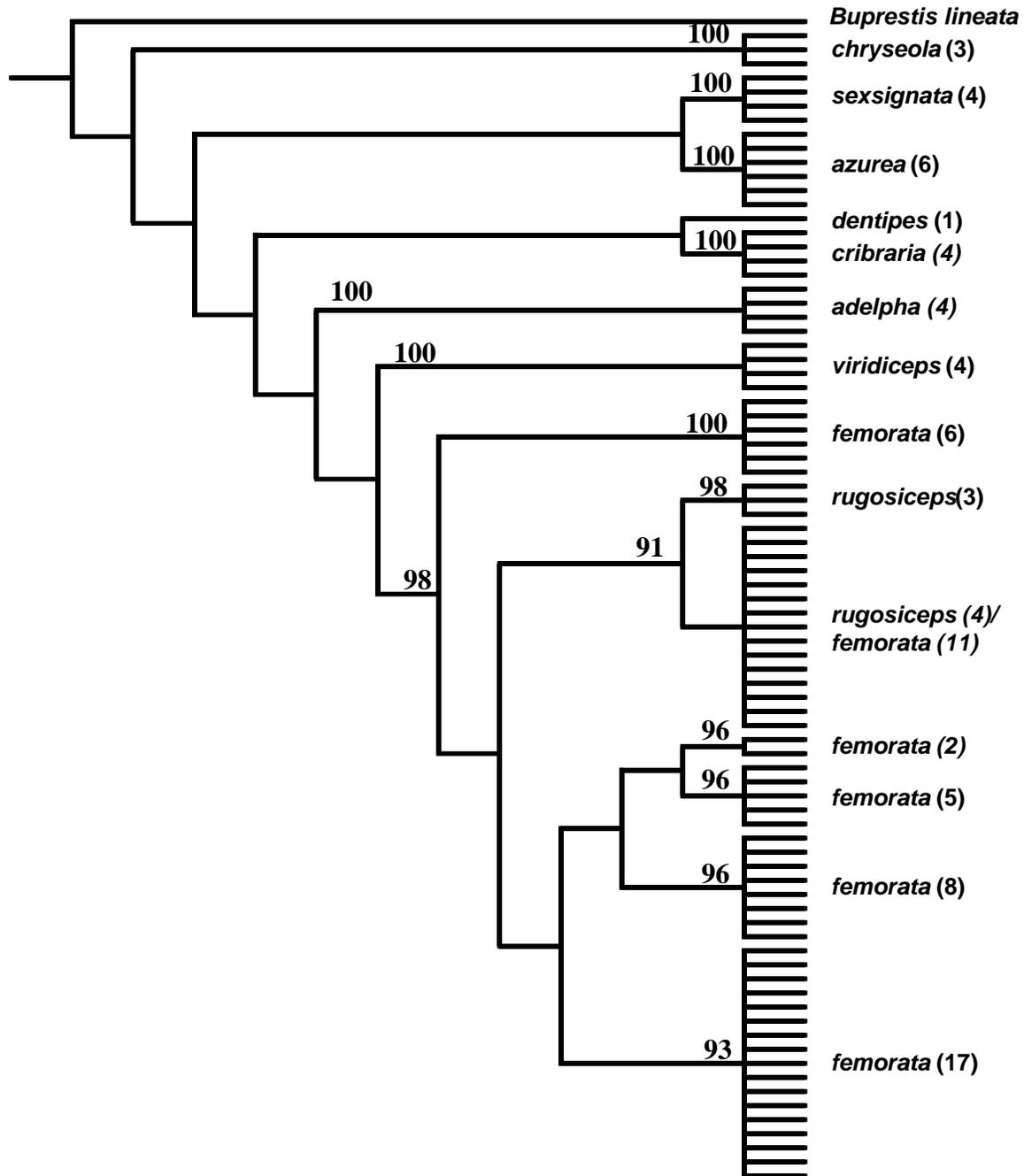
Several nuclear genes were also sequenced and found to be largely uninformative. While the OTT-MAL nuclear gene does possess sufficient variation, it has been difficult to amplify across all available specimens. To date, OTT-MAL sequences have been obtained from representatives of only five of the eight observed maternal groupings. At present, the phylogeny generated from OTT-MAL is not congruent with the mitochondrial-based tree. Further interpretation will be difficult without additional taxon sampling. In order to more fully assess species status within this complex, we intend to obtain and amplify the OTT-MAL gene from the remaining three maternal lineages and include more individuals from the observed maternal lines.

Regardless of these challenges, the current study clearly demonstrates considerable variation in this species complex, especially among 'species' collectively referred to as *C. femorata*. Further research may elucidate differences in host plant preference, which can be exploited by growers to manage *C. femorata* populations by scouting only host plants of the pest. If differences in pesticide sensitivity or seasonal biology can also be shown between these maternal lines, such knowledge will be critical to developing successful pest management strategies and would aid in eventual development of improved on-site diagnostic tools.

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Figure 1. Phylogeny of the *Chrysobothris femorata* complex based on COX-I gene sequence. Numbers in parenthesis next to specific names indicate the number of individuals sequenced for that clade. Bootstrap values of 70 or higher at nodes indicate good support for the inferred clade.



## Imported Fire Ant Mortality Following Exposure to a Biopesticide (Armorex)

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**Index Words:** Armorex, Hybrid, Mound drench, *Solenopsis*, Talstar

**Significance to Industry:** Imported fire ants (IFA) are a serious pest in nursery industry and are readily transported long distances when nursery stock and other farm items are shipped outside the infested areas. In Tennessee, approximately 84% of nursery stock from IFA infested areas is shipped to locations outside the quarantine (Brooker et al. 2000). Nursery plants must be treated with insecticides before shipment in compliance with the Federal Imported Fire Ant Quarantine enacted in 1958 by the US Congress to prevent the spread of IFA from areas already infested (USDA-APHIS 2007). However, most insecticides registered for IFA control by the nursery industry are expensive and potentially damaging to the environment. A new IFA management alternative to organophosphate insecticides is therefore a research priority in the nursery industry. One solution to the problems posed by conventional pesticides is to increase the utilization of EPA-approved biopesticides in nursery production.

**Nature of Work:** The IFA was accidentally introduced in the United States from South America in the early 1900s. Currently, over 325 million acres in North America are infested by IFA, causing billions of dollars in damage each year. The IFA is readily transported long distances when articles such as soil, nursery stock, and other items are shipped outside the infested area. These ants prosper in human modified sites that receive full sunlight such as cropland, pastures and urban lawns (Williams et al. 2001). A major issue for the U.S. nursery industry is the Federal Imported Fire Ant Quarantine (7CFR 301.81) enacted to prevent the spread of IFA from infested areas (USDA-APHIS 2007). Based on nursery trade estimates (Brooker et al. 2000), approximately 84% of nursery stock from IFA infested areas in Tennessee is shipped to locations outside the quarantine. Nursery stock destined for non-infested areas must be treated with insecticides before being shipped. However, most insecticides registered for IFA control by the nursery industry are expensive and potentially damaging to the environment. One solution to the problems posed by conventional pesticides is to increase the utilization of biopesticides in nursery production. Efficacious biopesticides can potentially be incorporated into existing nursery-pest-management-programs. One such biopesticide, Armorex (Soil Technologies Corp.), is exempt from EPA residue tolerance requirement and is labeled for ornamentals and ants in general, but its effects against IFA are unproven. Armorex contains sesame oil (84.5%), rosemary oil (1%), garlic (2%), clove (2%), white pepper (0.5%), as well as paprika, lecithin and citric acid (10%). In this study we tested the effectiveness of Armorex at different concentrations in combination with low rates of Talstar to manage individual colonies of IFA both in the lab and field situation. In lab bioassays, we evaluated toxic effects of Armorex at

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different rates (7.5, 10 and 15 ml / gallon respectively) and Talstar (0.1 ppm), plus combinations of Armorex and Talstar over 72 h periods. Laboratory conditions were at  $26 \pm 2^{\circ}\text{C}$  and 65 - 70 % RH. Only IFA workers were used in laboratory bioassays.

Field evaluations were performed along a highway right-of-way in Sequatchie County with the permission of the Tennessee Department of Transportation (TDOT). These sites have suitable infestations of hybrid IFA. Samples of IFA were taken from randomly selected mounds and identified to species using cuticular hydrocarbon analysis. Four plots, each containing 8 active IFA colonies were established. Distance between adjacent plots was set at 80 -100 ft. Location of each IFA mound was recorded by a hand-held GPS (sub-meter accuracy) and marked with a numbered surveyor's flag a day before treatments were applied. Mound activity was determined by probing the mound with a wire flag and only those mounds where more than 10 worker ants emerged following probing were used. Treatments were mixed in water buckets and applied as a 1-gallon drench. Treatments in each plot were assigned randomly, and included: 1) water-drench control, 2) Armorex at three rates (5, 10 and 15 ml / gal), 3) Talstar (1 ppm), and 4) Armorex concentrations combined with Talstar (1 ppm) resulting in 8 treatments per plot. Ant colonies were checked at 1, 3, 7, 14, 21 and 28 days after treatment. Data were analyzed using analysis of variance (ANOVA) procedures with means separated using LSD test ( $P < 0.05$ ).

**Results and Discussion:** In laboratory bioassays, we evaluated toxic effects of Armorex at different rates (7.5, 10 and 15 ml / gallon respectively) and Talstar (0.1 ppm), plus combinations of Armorex and Talstar to worker IFA over 72 h period. The recommended quarantine treatment dose when drench treating container media for IFA control is 25 ppm of Talstar. There was no significant difference between Armorex, Talstar and control treatments at 24, 48 and 72 h post treatment (Table 1). Significant ant mortality was observed when all rates of Armorex were combined with Talstar (0.1 ppm), suggesting a synergistic effect between the ingredients. In the field evaluations, however, no synergistic effect was observed between different rates of Armorex combined with a lower than labeled rate of Talstar (Table 2). Mortality in all IFA colonies treated with either Armorex alone or in combination with Talstar (1 ppm) were significantly different from Talstar and control treatments after 14 d post-treatment. No significant differences were detected ( $P < 0.05$ ) between Talstar and water-treated controls. We chose to use relatively low amounts of Talstar that might be more compatible with other IPM strategies and safer for the environment. Individual mound treatments are usually more environmentally and ecologically acceptable because less insecticide is used and treated areas are limited, resulting in less impact to non-target insects. Furthermore, a low dose of Talstar combined with a biopesticide may also be amendable for broadcast acreage treatments, especially since the lower Talstar rate will have less environmental impact. Only colonies from two Armorex (5 ml/gal and 10 ml/gal + Talstar) treatments and a single control relocated three days after treatment.

**Acknowledgements:** We thank Jane King and Bedie Bland for their assistance with experiments. Soil Technologies Corporation donated the Armorex used in this study.

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Table 1. Mean percentage mortality of hybrid imported fire ants compared to water treated control, different rates of Armorex, and 0.1 ppm Talstar in laboratory bioassays.

Treatments	24 hrs	48 hrs	72 hrs
Control	10a	25a	27.5a
Armorex (7.5 ml/gal)	3.8a	15.0a	17.5a
Armorex (10 ml/gal)	5.0a	27.5a	42.5a
Armorex (15 ml/gal)	12.5a	32.5a	47.5a
Talstar (0.1 ppm)	11.25a	27.5a	38.75a
Armorex (7.5 ml) +Talstar	63.75b	85.0b	93.75b
Armorex (10 ml) +Talstar	75.0b	91.25b	95.0b
Armorex (15 ml) +Talstar	68.75b	90.0b	93.75b

All Talstar treatments were applied at 0.1 ppm rate. Means followed by the same letter within the same column were not significantly different using ANOVA and means separated using Tukey's studentized range (HSD) test ( $P < 0.05$ ). Ten ants were used per each of 8 replicates per treatment.

Table 2. Cumulative percentage mortality of imported fire ant colonies following mound drenches with water (control), Armorex (different rates), Talstar (1 ppm) and Armorex plus Talstar in the field.

Treatments	Days After Treatment					
	1	3	7	14	21	28
Control	0.0±0.0a	12.5±12.5a	25.0±25.0a	25.0±25.0a	25.0±25.0a	25.0±25.0a
Armorex (5 ml/gal)	12.5±12.5ab	47.5±27.5ab	62.5±23.9ab	92.5±4.8b	100.0±0.0b	100.0±0.0b
Armorex (10 ml/gal)	50.0±28.9bc	50.0±28.9ab	80.0±20.0b	97.5±2.5b	100.0±0.0b	100.0±0.0b
Armorex (15 ml/gal)	45.0±26.0c	95.0±2.9b	100.0±0.0b	100.0±0.0b	100.0±0.0b	100.0±0.0b
Talstar (1 ppm)	37.5±21.7abc	47.5±27.5ab	50.0±28.9ab	50.0±28.9a	50.0±28.9a	50.0±28.9a
Armorex (5 ml/gal) + Talstar	17.5±14.4ab	85.0±11.9b	100.0±0.0b	100.0±0.0b	100.0±0.0b	100.0±0.0b
Armorex (10 ml/gal) + Talstar	45.0±26.0abc	60.0±22.7ab	92.5±4.8b	100.0±0.0b	100.0±0.0b	100.0±0.0b
Armorex (15 ml/gal) + Talstar	75.0±2.9c	95.0±5.0b	97.5±2.5b	100.0±0.0b	100.0±0.0b	100.0±0.0b

All Talstar treatments were applied at 1 ppm rate. Means followed by the same letter within the same column were not significantly different using ANOVA and means separated using LSD test ( $P < 0.05$ ). Each treatment was replicated four times.

**Optimizing Fertilization and Predator Combinations  
for Biological Control of Western Flower Thrips on Cut Roses**

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**Index Words:** *Amblyseius swirskii*, Biological Control, Cut flower, Fertilizer, *Frankliniella occidentalis*, *Rosa hybrida*

**Significance to Industry:** Commercial growers in the US are presently faced with challenges associated with the economical production of quality plant material and management of persistent pest problems. To meet these challenges, growers need management strategies for floricultural and ornamental crops that not only improve resource management but also provide more effective insect pest management. Despite both widespread and intensive use of insecticides, western flower thrips, *Frankliniella occidentalis* (Pergande), is a difficult pest to control on cut rose crops. Cultural manipulation of host plant quality could be used to enhance the effectiveness of biological control for *F. occidentalis*, if fertilization rate and host plant quality could be manipulated to reduce thrips population growth without compromising crop growth or productivity. We compared cut roses grown under fertilizer regimes representing 30% or 100% of the commercially recommended rate (150 ppm N) and measured: crop yield, abundance of *F. occidentalis* in the harvested crop, and control of *F. occidentalis* with releases of a predatory mite [*Amblyseius swirskii* (Anthias-Henriot) (Acari: Phytoseiidae)]. Rose plants fertilized with either 30% or 100% of the recommended rate produced similar numbers of harvestable flower stalks, but plants fertilized with the low rate had, on average, 30% fewer thrips than plants fertilized with the recommended rate. Lowering fertilization enhanced control of *F. occidentalis* with *A. swirskii* because plants protected by predatory mites and fertilized with the recommended rate had twice as many thrips, on average, as protected plants fertilized with the low rate. Our findings show that manipulation of fertilization for cut roses may be easily accomplished and a desirable pest management outcome generated; both of which are important for practical implementation of this strategy in commercial production of potted or cut roses.

**Nature of Work:** Roses are by far the most important ornamental crop worldwide and can be grown as cut flowers, potted flowering plants, and nursery crops. Among the major cut flowers still grown in the U.S., roses remain the highest earner with an average of \$696,933 in sales per large grower (1). Western flower thrips, *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae), is a serious pest of roses and other ornamental crops worldwide (2). On roses, conventional chemicals are the primary means to control *F. occidentalis* despite growing concerns about insecticide resistance in field populations of this pest. *Frankliniella occidentalis* shows resistance to as many as 22 compounds from at least 5 different classes of insecticides, including spinosad,

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an insecticide commonly used to manage *F. occidentalis* in greenhouses (3, 4). Many conventional insecticides are also detrimental to worker health and disrupt production or harvest routines. Novel or alternative management approaches that address resistance and human health concerns associated with chemical control of *F. occidentalis* on roses are essential to the continued success of this industry.

Cultural manipulation of host plant quality could be used to enhance the effectiveness of biological or chemical control for *F. occidentalis*, if fertilization rate and host plant quality could be manipulated to reduce thrips population growth without compromising crop growth or productivity. On potted chrysanthemum, we demonstrated that fertilization could be lowered to a level that reduces population growth of *F. occidentalis* while still maintaining plant productivity (5). In another study, we treated roses (*Rosa hybrida* cv. 'Tropicana') with four fertilization levels (50, 75, 100, 125, or 150% of the recommended level of 150 ppm N) and measured the effect of fertilization level on rose yield, quality, leaf tissue nutrient content, and post-harvest longevity (vase life) (6). We found that lowering fertilization to 50% of the recommended level did not adversely affect crop productivity, quality, or vase life. Our findings show that fertilizer inputs can be significantly reduced with no adverse effects on yield or quality of this cut flower crop; thereby, providing an ideal system to test whether rose plant quality could be manipulated to reduce thrips populations.

For this study, we compared cut roses grown under fertilizer regimes representing 30% or 100% of the commercially recommended rate (150 ppm N); and we measured in each regime: crop yield, abundance of *F. occidentalis* in the harvested crop, and the ability to control *F. occidentalis* with releases of a predatory mite [*Amblyseius swirskii* (Anthias-Henriot) (Acari: Phytoseiidae)]. *Amblyseius swirskii* is commercially available and has been found to be an effective predator of *F. occidentalis* on greenhouse roses (7, 8). In two trials conducted in research greenhouses on the Texas A&M University, College Station campus, we examined the feasibility of manipulating rose plant quality to reduce *F. occidentalis* populations and enhance biological control of this pest with *A. swirskii*.

The first trial was conducted from November 2006 to December 2006 and we compared infestations of *F. occidentalis* on roses treated with either 30% or 100% of the recommended rate (150 ppm N). Rose plants were grown from bare-root roses (*Rosa hybrida* L. cv. 'Tropicana' grafted onto 'Dr. Huey' rootstock) individually planted in 14-L, plastic nursery-containers with soilless mix (Sunshine Mix no.1, Sun Gro Horticulture Canada Ltd., Bellevue, WA, USA), pine bark, and sand (3:1:1 ratio). These plants were cultivated as a cut flower crop following conventional guidelines for greenhouse production (9). For 60 days prior to our experiment, we fertilized one set of plants with the low fertilization rate (30 %) and the second set with the 'High' recommended rate (100 %). The number of replications was six per treatment and each replicate consisted of twelve potted plants arranged in a 6-by-2 grid and confined within a thrips-proof, screened cage (120-inch long x 50-inch wide x 48-inch high). We produced a synchronous crop of cut roses that was harvested during the final week of the 8-week production period. At the start of the production period, we released 32 adult females and 8 adult males of *F. occidentalis* twice each week over five consecutive weeks and then once each week over three consecutive weeks into each cage. During the eighth week of the crop, we cut all the harvestable flower shoots and counted all stages of *F. occidentalis* (except eggs) extracted from flower shoots harvested from each cage.

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The second trial was conducted from March 2007 to April 2007 and we used the same experimental design but compared four treatments: low fertilization and thrips, high fertilization and thrips, low fertilization with both thrips and predatory mites, high fertilization with both thrips and predatory mites. During the same week of the first thrips release (week 1), we hung a single sachet (Swirski-Mite Plus™) of *A. swirskii* near the center of each potted plant in all replicates assigned predatory mites. Four weeks later, we replaced all the old sachets with new ones (week 5). All predatory mites were obtained from Koppert Biological Systems Inc. (Romulus, Michigan, USA) and three replications were completed for each treatment in the second study. We analyzed counts of harvested flower shoots and *F. occidentalis* with one-way ANOVA.

**Results and Discussion:** In the first trial, rose plants fertilized with either 30% or 100% of the recommended rate produced similar numbers of harvestable flower stalks ( $19.9 \pm 0.9$  flowers per replicate,  $n = 12$ ) (one-way ANOVA:  $F_{1,10} = 0.80$ ;  $P = 0.784$ ). However, plants fertilized with the low rate had, on average, 30% fewer thrips (one-way ANOVA:  $F_{1,10} = 21.08$ ;  $P = 0.001$ ) than plants fertilized with the recommended rate (Figure 1). In the second trial, we found no significant effects of fertilization or predatory mites releases on the numbers of flower stalks harvested ( $22.2 \pm 0.1$  flowers per replicate,  $n = 12$ ) (one-way ANOVA:  $F_{1,10} = 0.62$ ;  $P = 0.621$ ). However, lowering fertilization enhanced control of *F. occidentalis* with *A. swirskii* because plants protected by predatory mites and fertilized with the recommended rate had twice as many thrips, on average, as protected plants fertilized with the low rate (Figure 2). Numbers of thrips recovered from unprotected plants fertilized with the low fertilization rate were, on average, not statistically different from the numbers recovered from protected plants fertilized with the recommended rate (Figure 2). Unprotected plants fertilized with the recommended rate had, on average, twice as many thrips as plants fertilized at the same rate but protected by predatory mites (Figure 2).

Our findings clearly show that manipulation of fertilization for cut roses may be easily accomplished and a desirable pest management outcome generated; both of which are important for practical implementation of this strategy in commercial production of potted or cut roses. Use of biological control for thrips and reduction of both fertilizer and insecticide use should benefit growers by reducing production costs, chemical run-off and risks of insecticide resistance or plant phytotoxicity. Given the promising results of our study, we recommend further investigations to determine whether this approach could be extended into pest management systems for other floricultural crops.

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Figure 1. Numbers of *Frankliniella occidentalis* (+ SE) recovered from cut flowers harvested from roses treated with either 50 ppm N (low fertilization) or 150 ppm N (high fertilization), n = 6 per treatment. Different letter(s) above the bars indicate significant differences among fertilization treatments at  $P \leq 0.05$ .

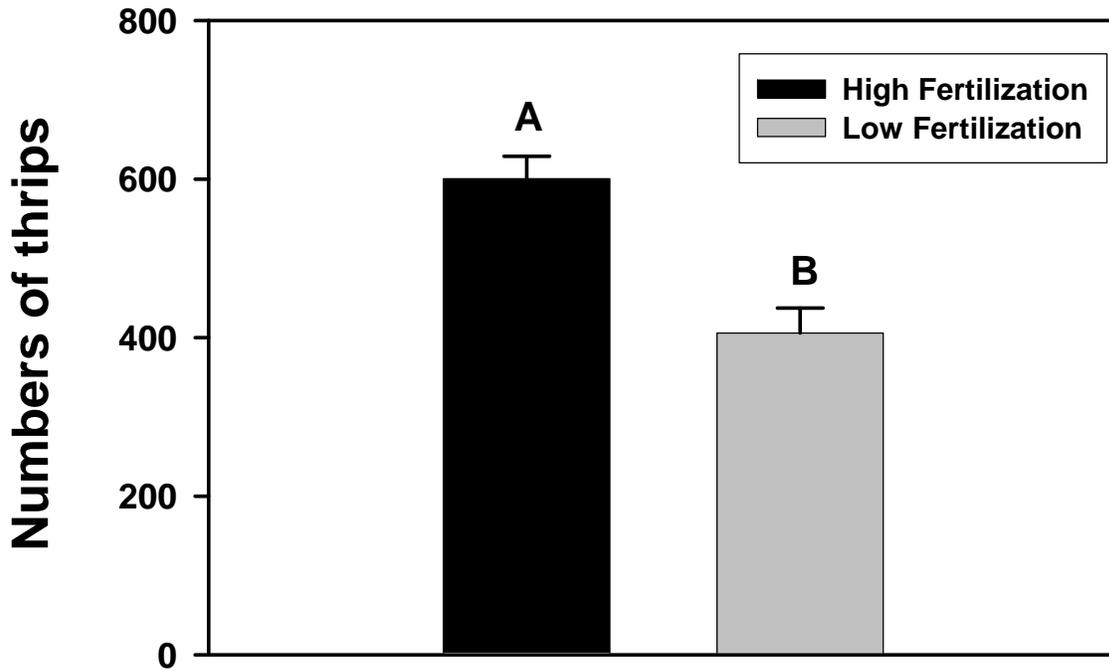
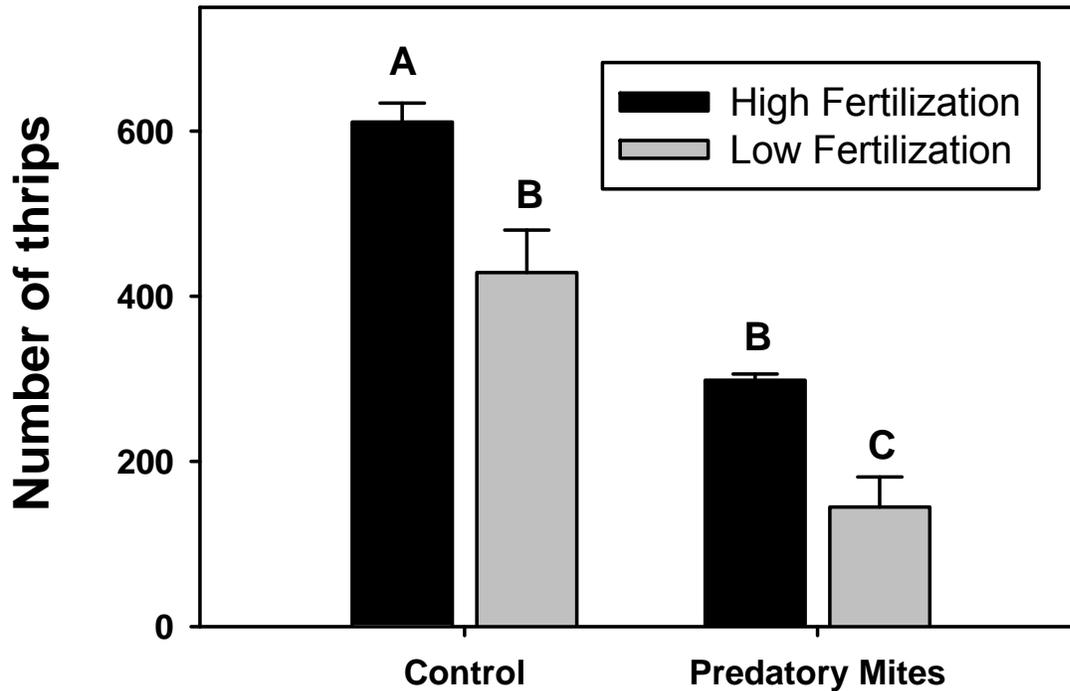


Figure 2. Numbers of *Frankliniella occidentalis* (+ SE) recovered from cut flowers harvested from roses with or without predatory mites and treated with either 50 ppm N (low fertilization) or 150 ppm N (high fertilization), N = 3 per treatment. Different letter(s) above the bars indicate significant differences among treatments at  $P \leq 0.05$  by one-way ANOVA and Tukey's HSD (one-way ANOVA:  $F = 34.29$ ,  $P < 0.001$ ).



**Reducing fertilization for potted *Mandevilla*: impacts on crop quality and spider mite control**

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**Index Words:** *Mandevilla splendens*, *Neoseiulus fallacis*, *Tetranychus urticae*, Fertilizer, Integrated Pest Management, Miticide, Predatory mite

**Significance to Industry:** Over fertilization of ornamental crops can contribute to higher pest control costs because the two-spotted spider mite (TSSM), *Tetranychus urticae* Koch, responds positively to high nutrient levels in the crop. Reduction of fertilization can be a useful pest management tactic if, when used alone or with other control practices, spider mite populations are reduced without loss of crop quality or yield. In this study, we show that halving the standard fertilization rate did not compromise growth of *Mandevilla splendens* 'Alice Dupont' during the first 10 weeks of the crop. Potted plants treated with 100% or 50% of the standard fertilization rate produced canopies with similar stem length and leaf number. However, weekly counts of spider mites and spider mite eggs on leaf samples did not differ statistically between plants treated with the grower miticide regime, 'Experimental product BYI 08330', predatory mite releases or the no application treatment at the two fertilization rates. We concluded that current fertilization rates for potted *Mandevilla* can be reduced by 50% without compromising crop quality but lowering fertilization did not enhance spider mite management.

**Nature of Work:** The two-spotted spider mite (TSSM), *Tetranychus urticae* Koch, is a worldwide pest of ornamental crops (1). Chemical control of TSSM is also often difficult in fast-growing plants, such as *Mandevilla*, that produce thick canopies in which good spray coverage is impossible to achieve. This leads to frequent and intensive applications that increase pesticide resistance selection, worker exposure, and environmental contamination risks. As an alternative to chemical control, predatory mites of the family Phytoseiidae are commercially available for biological control of spider mites; however, this tactic is most feasible when minimum crop injury may be tolerated during a part of the production cycle (2).

Manipulation of fertilization may be a useful tactic in an integrated pest management program, when altered fertilization regimes reduce pest populations with little loss in crop yield and quality. In a previous study, we compared cut roses grown under fertilizer regimes representing 10% or 100% of the commercially recommended rate (150 ppm N) and compared control of TSSM with either releases of a predatory mite (*Phytoseiulus persimilis* Athias-Henriot) or applications of Floramite® (3). Roses fertilized with 10% of the recommended rate and treated with predatory mites or the

miticide had, on average, 60 – 70% fewer spider mites and 70 – 80% fewer spider mite eggs than plants fertilized with 100% of the recommended rate and treated with similar control methods. Unprotected plants fertilized with the lower rate had, on average, around 40% fewer spider mites and spider mite eggs than those fertilized with the recommended rate. Lowering spider mite numbers should reduce the need for frequent miticide applications and lower production costs and selection for miticide resistance. Smaller spider mite populations are also easier to control with alternatives such as biological control. In this study, we compared crop quality and TSSM control on potted *Mandevilla* grown under fertilizer regimes representing 50% or 100% of the commercially recommended rate. We measured crop performance, TSSM abundance, and control of TSSM with miticide applications or releases of a predatory mite, *Neoseiulus fallacis*. In a preliminary test on potted *Mandevilla* (Carlos Bogran, unpublished data) *N. fallacis* was as effective in controlling TSSM as the more commonly used predatory mite *P. persimilis*, but at a lower release rate and a more competitive application-cost relative to that of weekly miticide applications.

During October to December of 2007, we conducted a field trial at a commercial nursery operation (Hines Horticulture Inc., Fulshear, TX). The grower established a crop of *Mandevilla splendens* from rooted plugs (cv. 'Alice du Pont') individually planted in 1-gallon, plastic pots. Plants were grown on 4.5 ft width x 40.0 ft length benches in a plastic-covered shade house following the grower's standard practices. Fertilization was made using a controlled-release, complete, 12-month fertilizer (Scotts Osmocote Pro 23-4-8 with Scottkote®, Scotts-Sierra Horticultural Products Company, Marysville, OH, USA) at the rate of 11 lb per cubic yard of potting media. To evaluate the impact of fertilization regime on crop performance and TSSM abundance, we tested two fertilization regimes (50% and 100% of the standard rate) and four mite control methods for a total of eight experimental treatments. The control methods were: no application (control treatment), releases of *N. fallacis*, a rotation of miticides (Floramite® and Avid®) favored by the grower, and a new insecticide/miticide product under development (Experimental product BYI 08330, OHP Inc.).

Sixteen benches of 720 potted *Mandevilla* were included in the test. Eight of the benches were located on the north side of the shade house and the other eight on the south side. Each bench was divided into two sections of equal length (20 feet) and we randomly assigned plants potted with media incorporating the standard fertilization rate (11 lb per cubic yard) to one section (360 plants per section) and plants potted with media incorporating half the standard fertilization rate (5.5 lb per cubic yard) to the other section. On each side of the greenhouse, we randomly assigned two benches to each TSSM control method (total benches per control method = 4). Thus, our experiment was a nested factorial design with fertilization rate nested within TSSM control method.

During each of the first nine weeks of the crop, we randomly selected four plants from each bench section and recorded the length of the main stem and the total number of leaves per plant. We also randomly selected a single leaf per plant from the center of the canopy and used a dissecting microscope to count the total numbers of all TSSM stages. To standardize counts for spider mites and spider mite eggs, we measured the area of all sampled leaves with a CI-203 Laser Area Meter (CID Inc., Camas, WA, USA) and converted raw counts to counts per cm<sup>2</sup>. Most of the rooted plugs were badly infested with TSSM upon delivery and all plants were treated by the grower with a

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rotation of Floramite® and Avid® during the first, second, and fourth weeks to prevent crop damage.

During the fifth week, we initiated the control treatments for our study and sprayed all plants on benches assigned BYI 08330 at the recommended rate (3.4 fluid oz per 100 gallons of water) until runoff was visible on all the foliage. *Neoseiulus fallacis* was obtained from IPM Laboratories, Inc. (Locke, NY, USA) and released during the fifth and sixth weeks. We uniformly sprinkled 1000 predatory mites with vermiculite onto all the plants on each bench assigned the predatory mites treatment to attain the recommended rate of one predatory mite per square foot. Each week, Hines Horticulture personnel were provided with TSSM counts for the leaf samples from the previous week and instructed to continue applications of their preferred miticides as needed on the grower-treatment assigned benches.

All plants were treated with Attrimec®, a plant growth regulator used to chemically pinch the crop, in the second week and then pruned in the ninth week. The trial was terminated when the entire crop was repotted into 2-gallon pots in the tenth week. We analyzed weekly counts of spider mites per cm<sup>2</sup> or spider mite eggs per cm<sup>2</sup> with repeated measures analysis using a nested design with fertilization rate and TSSM control method as main effects. Counts for spider mites and spider mite eggs from weeks 1-5 and weeks 6-9 were separately analyzed because the control methods of our trial were initiated during the fifth week. We also compared measurements of stem length or leaf counts with ANOVA using a randomized complete block design with bench as a block effect and both fertilization rate and week as main effects.

**Results and Discussion:** During the first five weeks of the crop, spider mite populations were similar on plants treated with the standard or lower fertilization rates. We found no significant interactions between fertilization and week for counts of spider mites ( $F_{8, 240} = 0.830$ ;  $P = 0.577$ ) or counts of spider mite eggs ( $F_{8, 240} = 0.355$ ;  $P = 0.943$ ). Differences between counts of spider mites ( $F_{1, 30} = 0.038$ ;  $P = 0.847$ ) or spider mite eggs ( $F_{1, 30} = 0.022$ ;  $P = 0.882$ ) on leaves sampled from plants treated with the standard or lower fertilization rate (Figures 1, 2) were not statistically significant. Mean counts of spider mites and spider mite eggs from both sets of plants sharply declined in weeks 3 and 5 after 'clean up' applications of miticides by the grower during weeks 2 and 4. However, we were surprised to find that all of the control treatments were equally ineffective in suppressing TSSM populations on the crop after week 5. During weeks 6 to 9, we found no significant interactions between fertilization and control method for counts of spider mites ( $F_{3, 24} = 0.298$ ;  $P = 0.827$ ) or spider mite eggs ( $F_{3, 24} = 0.166$ ;  $P = 0.918$ ). After week 5, counts of spider mites ( $F_{3, 24} = 2.601$ ;  $P = 0.075$ ) and spider mite eggs ( $F_{3, 24} = 2.544$ ;  $P = 0.080$ ) increased each week and did not statistically differ among control methods. Although TSSM populations seemed to be consistently larger each week on plants treated with the standard fertilization rate (Figures 1, 2), the differences were not statistically significant for spider mites ( $F_{1, 24} = 2.628$ ;  $P = 0.118$ ) or spider mite eggs ( $F_{1, 24} = 1.129$ ;  $P = 0.298$ ).

Reducing the fertilization rate by 50% did not appear to negatively affect the growth of *Mandevilla*. Since spider mite populations were statistically similar among plants treated with different control methods, we assumed that spider mite damage to the crop was uniform and tested for only bench and fertilization effects on stem length

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or canopy size (leaf count). Stem length of the *Mandevilla* crop differed among benches ( $F_{15, 1119} = 1.681$ ;  $P = 0.049$ ) but there was no significant interaction between fertilization and week ( $F_{8, 1119} = 0.477$ ;  $P = 0.873$ ). Mean stem length did not differ statistically with fertilization rate ( $F_{1, 1119} = 0.071$ ;  $P = 0.790$ ) and started at  $9.03 \pm 0.50$  cm ( $n = 128$ ) in the first week and increased to  $20.31 \pm 1.92$  cm ( $n = 128$ ) by the eighth week. Canopy size also differed among benches ( $F_{15, 1119} = 2.700$ ;  $P < 0.001$ ) but again there was no significant interaction between fertilization and week ( $F_{8, 1119} = 0.238$ ;  $P = 0.984$ ). Fertilization rate did not affect canopy size ( $F_{1, 1119} = 0.047$ ;  $P = 0.828$ ), which started at  $6.82 \pm 0.23$  leaves ( $n = 128$ ) in the first week and increased to  $10.45 \pm 0.40$  leaves ( $n = 128$ ) by the eighth week. During the ninth week, all plants were uniformly pruned which reduced the mean stem length to  $10.36 \pm 0.23$  cm ( $n = 128$ ) and mean canopy size to  $7.23 \pm 0.25$  leaves ( $n = 128$ ).

For ornamental production, reduction in both fertilizer and miticide use should benefit growers by reducing production costs and risks of plant phytotoxicity and chemical run-off. We have demonstrated that lowering fertilization level to 50% of the standard rate had no adverse effects on the growth of *M. splendens*; but unlike our findings with cut roses, it neither reduced the severity of TSSM infestations nor improved chemical or biological control of TSSM. Future studies are needed to evaluate the feasibility of significantly reducing spider mite populations and producing marketable crops of *Mandevilla* with fertilization below 50% of the standard rate.

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Figure 1. Mean numbers of two-spotted spider mites (adults and nymphs) (+ SE) per cm<sup>2</sup> counted on leaves from plants fertilized at 50% (n = 16) or 100% (n = 16) of the recommended fertilization rate.

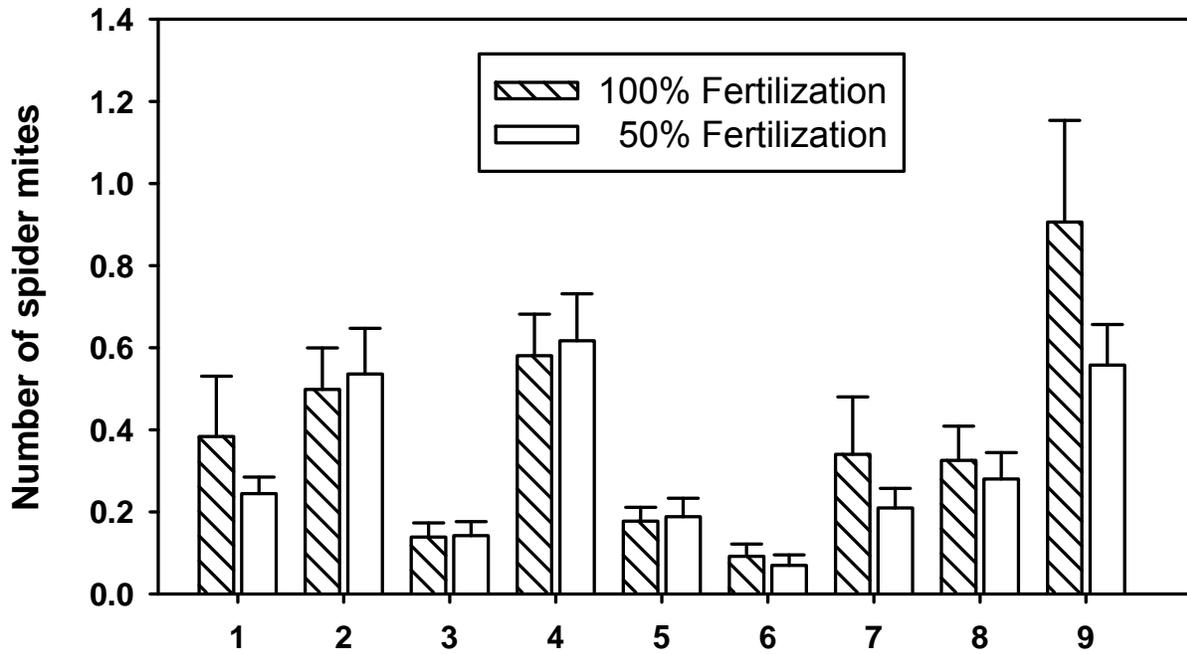
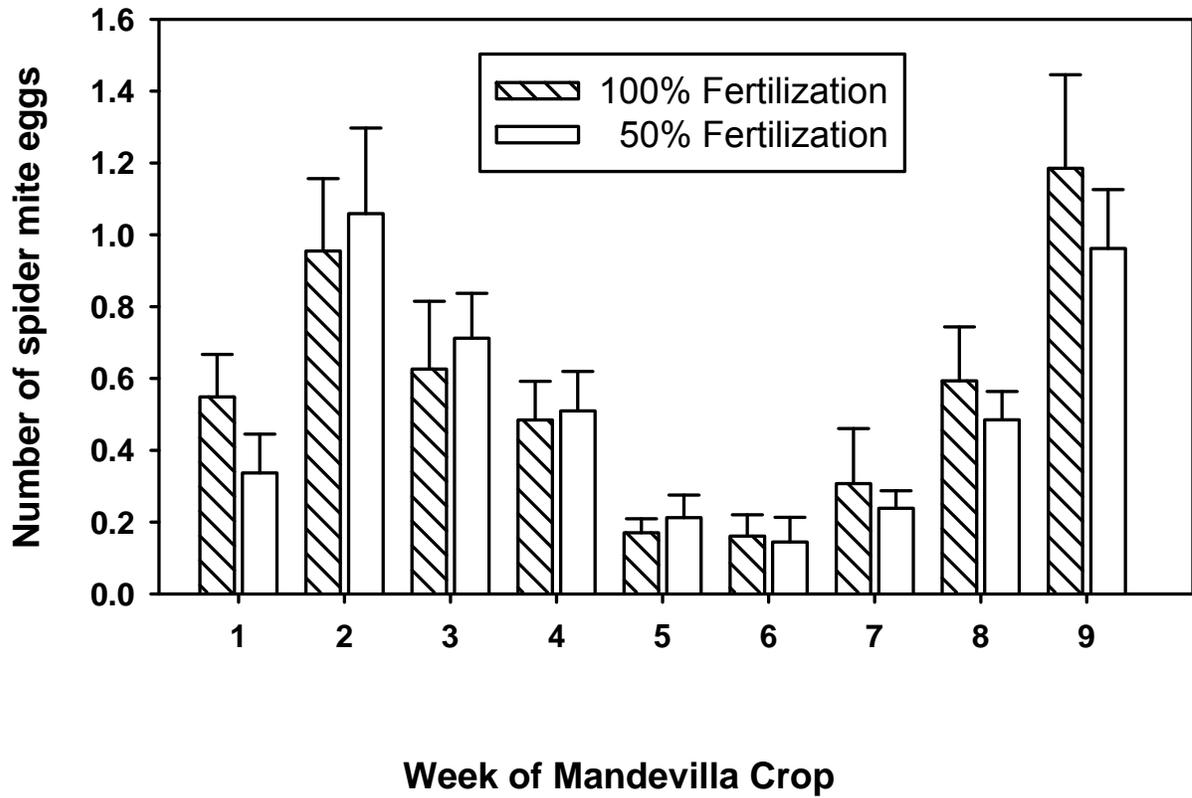


Figure 2. Mean numbers of two-spotted spider mite eggs (+ SE) per cm<sup>2</sup> counted on leaves from plants fertilized at 50% (n = 16) or 100% (n = 16) of the recommended fertilization rate.



**1 Species, 2 Species (Hybrid species...“True” Species?): A hip-cat in a cool-hat uses genetic analyses to investigate species complexity within both dogwood and lilac/ash clearwing moth borer populations (Lepidoptera: Sesiidae)**

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**Significance to Industry:** Economically important clearwing moths are sometimes difficult to identify. Lilac borers (*Podosesia* spp.) can cost growers thousands of dollars per acre in lost revenue (4). Male *Podosesia* species resemble each other to such an extent that outward appearance makes precise identification difficult. Similarly, dogwood borers (*Synanthedon scitula*) have been suspected to comprise a species complex. If true, elucidation of this complex could help to develop safer and more efficient control measures. In addition, with future use of organophosphate pesticides in question, better understanding of dogwood borer life history could expose new opportunities for control.

**Nature of Work:** Lilac borers (*Podosesia syringae syringae*, *P. syringae fraxini* and *P. aureocincta*) are almost identical morphologically, with only slight genitalic differences between males (6). Mating between *P. syringae* and *P. aureocincta* has been successful in the lab, in spite of the obvious differences in host preference and phenology (6). However, the two are commonly confused with each other due to their similar outward appearance and because males of both species are attracted to the same commercial lures used for monitoring. Intermediate forms between the two also hamper correct identification.

Like the *Podosesia* spp., the dogwood borer (*Synanthedon scitula*) has been suspected to comprise a species complex. Its host range is the largest of any known clearwing and its seasonal flight phenology poses enigmatic challenges. Most claim the dogwood borer is univoltine (9, 10). Even so, some point to evidence of semivoltinism and others suggest support for multivoltinism (7, 8, 9). As the dogwood borer becomes an increasing economic threat to apple growers, it is important to understand if the “species” is indeed a complex so populations can be managed effectively (1). Until recently, research investigating a dogwood borer species complex has been hampered by lack of an effective lure (5). Fortunately, a non-commercial lure is available that allows accurate sampling of populations and investigation of the putative species complex (11). The objectives of this research are: 1. To determine if the bimodal flight

peaks of the dogwood borer are composed of two sibling species and 2. investigate genetic variation within the lilac/ash borer complex.

**Results and Discussion:** Material was collected from several eastern and mid-western states (NY, MD, VA, WV, NC, GA, TN, KS, IA) in 2006 and 2007. Multipher 1 traps were modified to trap the moths in 80-95% ethanol and checked on a weekly basis. Moths were chosen for DNA extraction by selecting early and late season specimens. Moths from several locations were selected as well. Total DNA extract was used for amplification of the COX I mitochondrial gene. PCR products were sequenced in both directions for each specimen, were then aligned and compared looking for differences between nucleotide sequences. A phylogeny of *S. scitula* was generated using PAUP (Sinauer Associates, Sunderland, MA) (Fig. 1).

*Podosesia* spp. COX I sequences showed no differences between *P. syringae syringae* and *P. syringae fraxini*. Likewise, sequence variation between *P. syringae* and *P. aureocincta* also yielded little evidence of speciation. Early and late season dogwood borers varied little in their COX I sequences with two exceptions, one from Kansas and one from Mississippi differed significantly. The sequences of these two moths suggest a sister group relationship with other moths caught. These specimens were from separate seasonal flight peaks. Their divergence from other dogwood borers is interesting considering that another moth sequenced from Mississippi, caught the same day, was not significantly different from moths caught elsewhere. Further sampling is needed to investigate this cluster of mid-western dogwood borers.

The remaining dogwood borers included in the analysis clustered together, in a well-supported clade. This suggests that despite the idea that early and late season dogwood borer compose a species complex, they are actually one species. It has been suggested that differences in host plant material could account for the bimodal flight peak of the dogwood borer (3). The red-belted clearwing moth (*Synanthedon myopaeformis*), which fills a similar ecological niche, has been shown to have a shorter development time when feeding on apple burr knot tissue (2).

While this study does not support the idea of the observed phenology being composed of two separate species, it does show that some other factor is most likely responsible for observed bimodal flight peaks. Additionally, more sampling of the sister grouping in this study is needed to confirm its validity. The potential sister-grouping may result from insufficient sampling because only two specimens out of 18 sequenced fell into this group. Research into the rate of dogwood borer development on different plant material should also be investigated.

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Figure 1. Phylogeny of *Synanthedon scitula* based on *CoxI* gene sequence. Bootstrap values at nodes of 70 or higher indicate good support for the inferred clade. Bold indicate late-season trap captures.

