

Factors Influencing Insecticide Efficacy against Armored and Soft Scales

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ADDITIONAL INDEX WORDS. pine needle scale, oleander scale, calico scale, striped pine scale, chemical control, crawler duration

SUMMARY. Scale insects (Hemiptera: Coccoidea) are among the most economically important pests of ornamental plants. Soft scales (Coccidae) are phloem-feeding insects that produce large amounts of honeydew. By contrast, armored scales (Diaspididae) feed on the contents of plant cells and produce a waxy test that covers their bodies. We studied two species of armored scales [pine needle scale (*Chionaspis pinifoliae*) and oleander scale (*Aspidiotus nerii*)] and two species of soft scales [calico scale (*Eulecanium cerasorum*) and striped pine scale (*Toumeyella pini*)] to compare efficacy of selected insecticides. In addition, we assessed how the duration of first instar emergence might influence insecticide efficacy. Several reduced-risk insecticides (chlorantraniliprole, pyriproxyfen, spiromesifen, and spirotetramat), horticulture oil, and two broad-spectrum insecticide standards (bifenthrin and dinotefuran) were evaluated. Efficacy of insecticides was consistent within each scale family. Bifenthrin and pyriproxyfen were the only insecticides that killed soft scale insects. By contrast, all insecticides killed armored scales when the crawler stage was the target of application. Armored and soft scales may differ in susceptibility to pesticides because of likely differences in the chemical composition of their integuments and covers. Finally, we found that the effectiveness of a single application of insecticide declined by >15% when the duration of the crawling juvenile period was increased from 1 to 4 weeks. Increases in duration of a scale crawling period decreased the efficacy of a pesticide application.

Scale insects are among the most destructive pests that feed on leaves, stems, and fruit of vegetative and woody plants (Fulcher et al., 2012). High densities of scale insects can slow the rate of carbohydrate assimilation (Speight, 1991; Washburn et al., 1985) and reduce plant vigor. This can result in leaf chlorosis, branch dieback, or even death of the plant due to direct injury or outbreaks of other insects and pathogens (Hanson and Miller, 1984; Hubbard and Potter, 2005; Rebek and Sadof, 2003). Armored scales, mealybugs (Pseudococcidae), and soft scales are the most economically important insects in this group (Miller et al., 2005).

Outbreaks of scale insects are more frequent in protected culture and disturbed habitats, such as in greenhouses and urban landscapes, than in natural forests (Hanks and Denno, 1993; Tooker and Hanks, 2000). Outbreaks have been attributed to a reduction of natural enemies caused by low plant and prey diversity, pesticide residues, dust, and high temperature (Hanks and Denno, 1993; Luck and Dahlstein, 1975; Meineke et al., 2013; Price et al., 2011; Raupp et al., 2001). In addition, urban warming might contribute to pest outbreaks due to the effect of temperature on the fecundity and temporal synchrony of scale insects, and their natural enemies (Dale and

Frank, 2014; Meineke et al., 2014). Individuals tasked with protecting trees from scale insects in urban environments need to select and time pesticide applications so they target the most susceptible stages of scales while minimizing impact on natural enemies (Cloyd, 2010; Frank, 2012; Rebek and Sadof, 2003; Robayo-Camacho and Chong, 2015). As such, pesticides that have been categorized by the U.S. Environmental Protection Agency (EPA) as reduced risk to human health and the environment are favorable candidates for use in a pest management program (EPA, 2010). Although applications of reduced-risk products to the crawling scales can be as effective as those products with higher risk chemistries (Frank, 2012; Xiao et al., 2016), the relative effectiveness of low risk materials can vary.

Scale insects have flightless adult females and winged adult males. Winged males have undeveloped mouthparts and live just long enough to find mates [hours to days (Kosztarab, 1996; Miller and Davidson, 2005; Rosen, 1990)]. The mobile first instar will settle and feed on plant substrates within 24–72 h or crawl to a leaf edge and disperse in the wind (Ben-Dov and Hodgson, 1997). During this time, crawlers are subject to high rates of mortality due to depletion of available host material, adverse environmental conditions such as heavy rains, or pesticide use (Rosen, 1990). Both armored and soft scale species may have univoltine and multivoltine life cycles depending on the geographic and climatic conditions (Ben-Dov and Hodgson, 1997; Miller and Davidson, 2005; Robayo-Camacho and Chong, 2015; Rosen, 1990).

Armored scales feed primarily on the contents of plant cells (Sadof and

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Units

To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
29.5735	fl oz	mL	0.0338
0.3048	ft	m	3.2808
3.7854	gal	L	0.2642
2.54	inch(es)	cm	0.3937
25.4	inch(es)	mm	0.0394
6.4516	inch ²	cm ²	0.1550
1.6093	mile(s)	km	0.6214
7.4892	oz/gal	g·L ⁻¹	0.1335
11.1612	oz/inch	g·cm ⁻¹	0.0896
6.8948	psi	kPa	0.1450
(°F–32)/1.8	°F	°C	(°C × 1.8) + 32

Neal, 1993) and are characterized by their waxy covering (Rosen, 1990). These wax covers, also called tests, start forming within 24–48 h after they settle to feed. The test is not bound to the body of an armored scale. It simply encloses the insect and eggs. Within hours after armored scales hatch, scale insects crawl under or out of the test find, a place to settle and feed for the rest of their lives (Miller and Davidson, 2005; Rosen, 1990).

Soft scales feed on phloem sap and produce honeydew (Ben-Dov and Hodgson, 1997; Kosztarab, 1996). Honeydew is composed mostly of sugars and amino acids (Ben-Dov and Hodgson, 1997; Leroy et al., 2011; Völkl et al., 1999; Woodring et al., 2004) and can lead to accumulation of black sooty mold on bark and leaves, which can reduce photosynthesis and plant growth (Ben-Dov and Hodgson, 1997; Hubbard and Potter, 2005). In addition, honeydew supports symbiotic mutualisms with ants that provide protection from natural enemies (Hanks and Sadof, 1990; Vanek and Potter, 2010). Soft scale insects are covered mostly with honeydew (Ben-Dov and Hodgson, 1997; Kosztarab, 1996). Once soft scales settle, some species, such as striped pine scale, remain in the same location (Clarke et al., 1989). Other species retain functioning legs so they can move to different parts of their host plant. For example, second instar calico scales feed on leaves during the summer and move to the bark during the fall where they complete their life cycle (Hubbard and Potter, 2005).

In previous studies, we found that differences in the morphology

and biology of armored and soft scales could explain why these two groups of insects responded differently to applications of insecticidal soap and horticultural oil (Quesada and Sadof, 2017). For this reason, we hypothesize that armored and soft scales could respond differently to other pesticides with that share a mode of action. In addition, we hypothesize that temporal differences in the duration of the dispersing crawler stage can affect the capacity of the insecticide applications to reduce scale populations. Armored and soft scales vary widely in the duration of crawler periods because of environmental conditions and biology (Frank et al., 2013). Thus, because the crawler stage of scales is most vulnerable to insecticides, application timing and frequency may need to be adjusted to account for differences in the biology of scale insects. Scale species, such as calico scale and pine needle scale, that lay all eggs at one time are more likely to be controlled with single application of insecticide if it is timed to coincide with their brief crawler period. By contrast, other species, such as oleander and striped pine scale who lay a few eggs at a time, have a more extended crawler period and are more likely to avoid mortality from a single application of a short residual insecticide. Our laboratory colony of oleander scale produced a continuous supply of crawlers that allowed us to manipulate the crawler period to directly test this hypothesis.

The objectives of this research were to determine how the survival of armored and soft scales was affected by 1) the application of selected reduced-risk and broader spectrum

insecticides and 2) the duration of the scale crawler period.

Material and methods

SCALE INSECTS AND INSECTICIDES. Reduced-risk insecticides (chlorantraniliprole, pyriproxyfen, spiromesifen, and spirotetramat) were chosen from a list provided by EPA (2016) because of their potential to be part of an integrated pest management program that conserves beneficial insects (Cloyd et al., 2006; Frank, 2012; Planes et al., 2013; Xiao et al., 2016). Even though horticultural oil can kill natural enemies on contact (Oetting and Latimer, 1995), it was included in our list of insecticides because of its ability to kill scale insects with a minimal impact on natural enemies due to the short-term residual toxicity (Raupp et al., 2001; Rebek and Sadof, 2003). Dinotefuran and bifenthrin were chosen to represent the two most commonly used broad-spectrum insecticide classes, neonicotinoids, and pyrethroids (Sparks and Nauen, 2015). All insecticides were applied using the highest labeled rate except for spirotetramat, which was applied to striped pine scale at two different rates. A total of seven insecticides with six different modes of action were studied on either soft or armored scale (Table 1).

We conducted experiments with four species of economically important scale insects to determine how these different insecticides controlled soft and armored scales. Calico and striped pine scales represented soft scale, whereas pine needle and oleander scale represented armored scales. Five experiments were conducted during 4 years in Indiana. Three of

Table 1. Insecticides and formulation rates tested for efficacy against armored and soft scale.

Active ingredient	Trade name ^z	Rate (a.i.) ^y	Application method	IRAC mode of action no. ^x
Water				
Horticultural oil	Ultrafine Oil	20.00 g·L ⁻¹	Foliar	NA
Pyriproxyfen	Fulcrum	0.099 g·L ⁻¹	Foliar	3A
Spiromesifen	Judo	0.067 g·L ⁻¹	Foliar	23
Spirotetramat (low rate)	Kontos	0.032 g·L ⁻¹	Foliar	23
Spirotetramat (high rate)	Kontos	0.063 g·L ⁻¹	Foliar	23
Bifenthrin	Talstar S	0.118 g·L ⁻¹	Foliar	3A
Chlorantraniliprole	Acelepryn	0.058 g·L ⁻¹	Foliar	28
Dinotefuran	Transtect 70WSP	0.567 g·cm ⁻¹ DBH	Soil	4A

^zUltrafine Oil (Whitmire Micro-Gen Research Laboratories, St. Louis, MO), Fulcrum, Judo, and Kontos (OHP, Mainland, PA), Talstar S (FMC Corp., Philadelphia, PA), Acelepryn (E.I. du Pont de Nemours and Co., Wilmington, DE), and Transtect 70WSP (Rainbow Treecare Scientific Advancements, Minnetonka, MN).

^y1 g·L⁻¹ = 0.1335 oz/gal, g·cm⁻¹ = 0.0896 oz/inch, DBH = diameter at breast height.

^xInsecticide Resistance Action Committee (IRAC): NA (smothering agents not included in IRAC classification), 3A (juvenile hormone mimics), 23 (inhibitors of acetyl-CoA carboxylase), 3A (sodium channel modulators), 28 (ryanodine receptor modulators), and 4A [nicotinic acetylcholine receptor (nAChR) competitive modulators].

the experiments were conducted under field conditions on trees that were naturally infested with scale insects. The other two experiments were conducted in a shade house or laboratory using plants artificially infested with scale insects.

Soft scale studies

CALICO SCALE. We selected 25 honey locust trees (*Gleditsia triacanthos*), located in a restaurant parking lot in Fishers, IN (lat. 39.928119°N, long. 86.0336224°W), that were naturally infested with calico scale. Selected trees ranged in size from 6 to 16 inches in diameter at breast height and had a buffer tree between them. The experiment was designed to target first instar calico scales that had settled on leaves in 2012 followed by applications that targeted overwintering females in Spring 2013. Five treatments (chlorantraniliprole, bifenthrin, dinotefuran, pyriproxyfen, and water) were allocated to each of five trees in a randomized complete block design. Trees were blocked by location in the parking lot. Foliar and soil applications were applied on 6 June 2012 and 2–3 May 2013. In both years, foliar insecticides were applied to each tree canopy until runoff, using a high-pressure sprayer (FMC John Bean, Philadelphia, PA) at 100 psi, whereas the systemic insecticide (dinotefuran) was applied at 20 psi with a subsoil injector whose probe penetrated 8 inches below the soil surface at four points within 1 ft of the trunk (Eliaison and Potter, 2000; Hubbard and Potter, 2006).

Live scale densities were estimated from 30 leaflets per tree. Leaflets were obtained from five leaves collected from different locations on trees from each treatment near sites where there was evidence of egg-laying females. Six leaflets from the middle of each leaf were used. Each scale was visually examined to determine if it was dead or alive based on color (Hubbard and Potter, 2006). Scales that were yellow were counted as live, whereas those brown to orange were counted as dead. The leaf area of each sample was measured using a leaf area meter (LI-COR Biosciences, Lincoln, NE) and the density of live scales was defined as the total number per square centimeter of leaf. In 2012, leaves were collected on

6 and 20 June and 16 and 30 July [0, 14, 40, and 58 d after treatment (DAT), respectively]. On 2 May 2013, density of overwintering females was assessed to determine the effect of insecticides from the previous year. Four branches were chosen randomly for each of the 25 trees and the number of live female scale insects was counted on the terminal 30 cm of the branches. On 16 and 23 May 2013 (14 and 21 DAT, respectively), survival of overwintering females was assessed to determine the effect of the second application by counting the number of dead and live females from four branches per each treatment. If the scale was shriveled and the color was faded brown, it was counted as dead. By contrast, if the scale was smooth and with white tufts, it was counted as live. Also, leaf samples were collected to assess the effect of insecticides on the first and second instars on 17 June and 17 and 29 July (46, 76, and 88 DAT, respectively). Densities of live scales were calculated as during the previous year.

In addition to field tests, a laboratory experiment was conducted in 2012 on calico scale crawlers to determine effects of fresh and aged insecticide residues on calico scale survival. In this study, we used scale-free leaves collected on 21 June from honey locust trees located in a nursery in Westfield, IN (lat. 40.020767°N, long. 86.188962°W). These trees had been treated with a foliar insecticide (chlorantraniliprole, bifenthrin, or pyriproxyfen) on 8 June, 2 weeks before the laboratory assay. Foliar applications were made with a 3-gal sprayer (Solo Inc., Newport News, VA) to runoff. Leaves that were treated on the day of the assay were collected from untreated trees and sprayed to runoff with one of three foliar insecticides using a 32-fl oz bottle sprayer (The Bottle Crew, Farmington Hills, MI) on 22 June and allowed to dry.

Leaflets from each treatment were placed in petri dishes atop of cotton wool dampened with water at a constant temperature of 25 °C and 16/8 h (light/dark). There were six replications for each of the eight treatments arranged in a completely randomized design. On the day of the assay, 22 June, ovipositing female calico scales (previously collected from

trees in downtown Indianapolis on 15 May and refrigerated at 4 °C) were placed on each leaflet. After 12 h, females were removed, leaving only crawlers on the leaflets. Survival of calico scale crawlers was estimated 7 d later on 29 June. Crawlers that died before and after settling on the leaflet were used to calculate survival (those that crawled off the leaf into the cotton substrate were not counted).

STRIPED PINE SCALE. The third experiment was conducted on striped pine scales feeding on scots pine trees (*Pinus sylvestris*). In 2014, a total of 42 naturally infested trees were selected in a working Christmas tree farm in Fort Wayne, IN. They were ≈6 years old and 6 ft tall. The trees were planted on 6-ft centers in rows 10 ft apart. On 26 June, trees were blocked by location into groups of six. Treatments were randomly assigned to each tree in the block (water, bifenthrin, pyriproxyfen, oil, spirotetramat low rate, and spirotetramat high rate) and applied at first observation on the crawler stage on the selected trees. Applications were made to the canopy with a 3-gal backpack sprayer until runoff. Efficacy of insecticides was assessed by collecting three infested twigs from each tree. Twigs were collected from sites covered with sooty mold on 10 and 19 July, 24 Aug. 2014, and 6 May 2015 (14, 23, 59, and 314 DAT, respectively). We chose branches with remnants of sooty mold because it indicated that scales were present at the start of the study. We assessed effects on male and female striped pine scales separately because females develop on stems, whereas males develop on the needles (Clarke et al., 1989). In the laboratory, three 5-cm twig sections were used to estimate densities of striped pine scale females. Also, six needles were randomly selected from twigs to evaluate densities of males, using the number of live scales and length of needles.

On 16 June 2015, a second application of insecticide was made when crawlers were observed for the first time on the trees. Effects of insecticides were examined as in the previous year by collecting twigs on 1 and 15 July and 5 Aug. (15, 30, and 50 DAT, respectively). In addition, percentage of dead branches was assessed by visually estimating the percentage of brown needles on 26

May and 15 July after the normal period of spring needle drop (Sadof, 1997).

Armored scale studies

Scots pine trees infested with were used in our fourth experiment located in a Christmas tree farm in Thorntown, IN. Naturally infested trees were ≈ 6 ft tall. In 2015, a total of 48 trees were selected and grouped into eight blocks of six trees. The six different treatments (water, bifenthrin, pyriproxyfen, spiromesifen, horticultural oil, and spirotetramat) were randomly allocated to trees in the eight replicate blocks. On 12 May, applications were made with a 3-gal backpack sprayer to the canopy until runoff, targeting young crawlers of the first generation of pine needle scale insects. Three old needles (previous year's growth) and three young needles (current year's growth) were randomly selected from each plant and taken to the laboratory to assess the number of live scale insects per cm length on 0, 15, 30, and 77 DAT. Under stereoscopic inspection, an ultrafine needle (12.7 mm) was used to remove the shell of the insect and perforate its body. Scale insects were considered dead if the body was dry and dark colored. By contrast, scales were counted as alive if it was yellowish to orange and excreted hemolymph when their bodies were perforated with an ultrafine needle.

OLEANDER SCALE. Oleander scales were used to compare how the duration of the crawler emergence period affected effectiveness of a single insecticide application. Oleander scales produce crawlers continuously year round (Uygun and Elekçioğlu, 1998). The life cycle is completed in ≈ 26 d and the seasonal oviposition period of a population can last up to 126 d (Uygun and Elekçioğlu, 1998). We used a laboratory colony of oleander scale (Quesada and Sadof, 2017) maintained on the fruit of butternut squash (*Curcubita maxima*) to artificially infest english ivy (*Hedera helix*) plants to mimic either 1 or 4 weeks of crawler emergence.

Plants were purchased as plugs obtained from Midwest Groundcovers LLC (Chicago, IL) because they had no history of pesticide use. These were transplanted into 1-gal pots during Summer 2015. The plants were maintained in a shade

house located at the Throckmorton Purdue Agricultural farm, 15 km south of the West Lafayette campus of Purdue University. The plants were fertilized by mixing 15 mL of 19N–2.6P–10K slow-release fertilizer (Start-N-Grow 19–6–12; Ferti-lome, Bonham, TX) into the top 3 cm of soil at the time of planting at the labeled rate and watered as needed.

On 14 Sept., 42 plants were selected to create a population of scales that mimicked a week-long crawler emergence period during which scales could colonize plants. The plants were taken to the laboratory for 1 week to infest them with the crawler stage of oleander scales by brushing live crawlers from a colony of scale-infested butternut squash each day for 5 d. Enough scales were transferred to assure a visually even dusting of crawlers across treatments. The actual number of scales transferred was not counted because of logistical difficulties in counting moving scale crawlers. After infestation, plants were returned to the shade house and randomly assigned to one of the six treatments (water, bifenthrin, pyriproxyfen, spiromesifen, horticulture oil, or spirotetramat) on 21 Sept. 2015 in a completely randomized design. Applications were made with a 3-gal sprayer until runoff targeting first instars between 2 and 7 d old. At 15 and 21 DAT, five leaves infested with oleander scale were selected from each plant and taken to the laboratory to assess scale survival. Under a stereoscope, an ultrafine needle (12.7 mm) was used to puncture the first 100 scale insects encountered to determine if it was alive. Insects that expelled hemolymph when punctured were considered alive.

On 16 Oct., a second set of 42 plants were taken to the laboratory to establish an infestation of scales that mimicked a 4-week crawler emergence period. This was achieved by continually transferring scales from the colony to the plants every other day for 4 weeks, the approximate duration of oviposition by an oleander scale (Miller and Davidson, 2005). By Nov. 16, when the six pesticides were applied, each infested plant contained first instar, second instar, and adult stages. Treatments were applied and allocated in a completely randomized design as described previously. Effect of insecticide on

insect survival was assessed as described previously at 14 and 21 DAT.

Data analysis

The effects of insecticides on densities, mortality, and survival of scale insects were examined using a repeated measures analysis of variance (ANOVA) with PROC MIXED (SAS version 9.3; SAS Institute, Cary, NC) when response variables were quantified more than once. One-way ANOVA was used to analyze the laboratory assay and the effect of insecticides on females the following year after application. Also, a two-way factorial ANOVA was used to determine the effect of 1- vs. 4-week crawler period on the efficacy of the six treatments. Before data analysis, Shapiro–Wilk and Levene's tests were conducted to determine normality and homogeneity of variance, respectively. If data did not meet these assumptions, square-root or arcsine square-root transformations were applied to densities or proportion (mortality or survival) of scale insects, respectively. Live scale densities, mortality, and survival were used as a response variable and insecticide treatment as the independent variable. Means were compared with a Fisher's protected least significant difference. In addition, linear contrasts were used to compare means of armored scales that survived when the host plant was colonized for 1 or 4 weeks after insecticide application.

Results

SOFT SCALE STUDIES. Compared with the water control, densities of live immature calico scales were significantly reduced ($F = 3.35$; $df = 4, 20$; $P = 0.030$) by bifenthrin (45%) when applications targeted settled crawlers in 2012 (Fig. 1A). During Spring 2013, density of live overwintering females on branches was also significantly reduced ($F = 4.49$; $df = 4, 24$; $P = 0.009$) on trees treated with bifenthrin (43%) and pyriproxyfen (58%) compared with the water control (Fig. 1B).

In 2013, after a second application targeted egg-laying adult females calico scale, bifenthrin was the only insecticide that significantly reduced scale survival compared with the water control [$F = 16.9$; $df = 4, 20$; $P > 0.001$ (Fig. 2A)]. However, all the

treatments of adult scales except for chlorantraniliprole, ultimately reduced the density of crawlers on leaves ($F = 5.13$; $df = 4, 20$; $P = 0.005$) compared with the control. The greatest reduction was produced by pyriproxyfen (57%) and bifenthrin (48%) followed by dinotefuran (36%) (Fig. 2B).

Laboratory assays of calico scale survival showed significant differences among treatments [$F = 11.83$; $df = 8, 53$; $P < 0.001$ (Fig. 3)]. As a group, survivorship on 7- and 21-d-old residues were not statistically different ($F = 0.14$; $df = 1, 40$; $P = 0.712$). However, both 7- and 21-d-old residues significantly reduced survivorship compared with the water control [$(F = 11.69$; $df = 1, 40$; $P < 0.001$) and ($F = 21.19$; $df = 1, 40$; $P < 0.001$), respectively]. Survival of crawlers exposed to 7 and 21 DAT residues was significantly reduced by bifenthrin (to 15% and 16%) and chlorantraniliprole (to 28% and 19%, respectively) when compared with water control (70% surviving).

In 2014, application of insecticides that targeted the peak of striped pine scale crawler emergence significantly reduced the number of females that settled on the branches [$F = 7.02$; $df = 5, 36$; $P > 0.001$ (Fig. 4A)] and males that settled on the needles [$F = 4.69$; $df = 5, 35$; $P = 0.002$ (Fig. 4B)]. Bifenthrin reduced immature female populations by 78% and immature male populations by 57% compared with the controls. Pyriproxyfen reduced the same groups by 48% and 25%, respectively.

None of the insecticides applied to striped pine scale crawlers in 2014 reduced the density of overwintering adult females when compared with the control in Spring 2015 [$F = 3.51$; $df = 5, 37$; $P = 0.012$ (Fig. 4C)]. However, pyriproxyfen provided the same level of control as bifenthrin at this time. Densities of adult scales on untreated control trees were low in 2015 because striped pine scales were already causing tree mortality. Comparison of tree canopy mortality showed statistically significant differences ($F = 3.41$; $df = 5, 36$; $P = 0.013$) among treatments. All treatments produced significantly less branch mortality than the water control (27%). No branch mortality was detected for bifenthrin, pyriproxyfen, and spiromesifen showed the greatest reduction of 52%, 45%, 39%, and 38%,

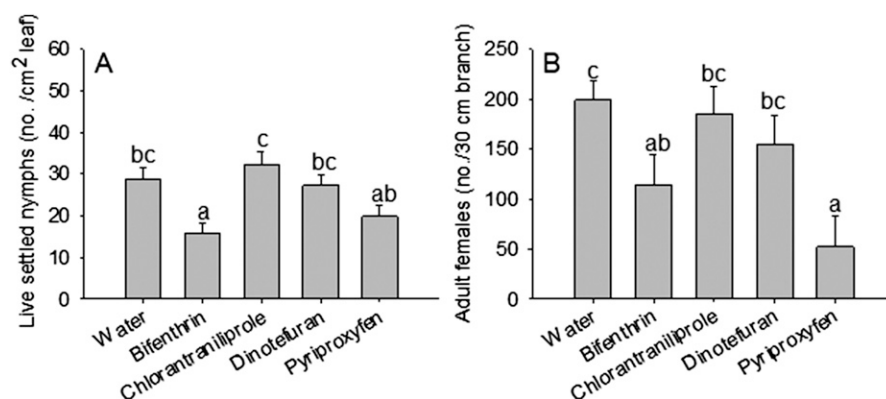


Fig. 1. Effects of insecticide applications on mean \pm SE of settled (A) crawlers on leaves and (B) adult females on branches, when applied to calico scale crawlers on honey locust trees on 6 June 2012. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test; 1 nymph/cm² = 6.4516 nymphs/inch², 1 adult female/30 cm = 1.0160 adult female/ft.

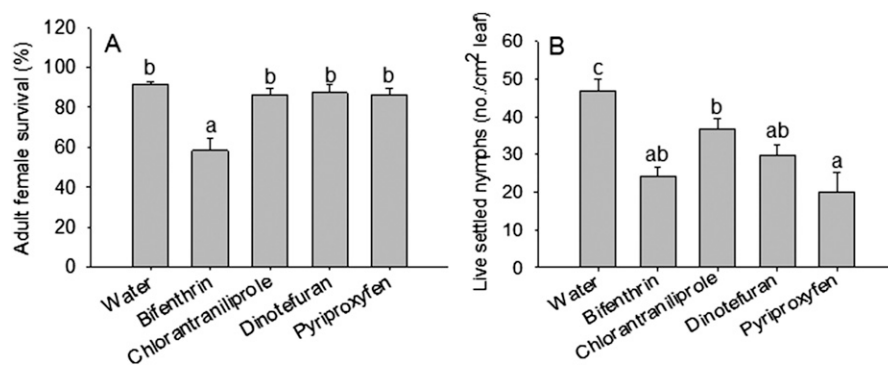


Fig. 2. Effects of insecticide applications on mean \pm SE of (A) egg-laying females on branches and (B) settled crawlers on leaves, when applied to overwintering stages of female calico scales on honey locust trees on 2–3 May 2013. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test; 1 nymph/cm² = 6.4516 nymphs/inch².

Trees treated with a low rate of spirotetramat and horticultural oil had 5% and 10% branch mortality, respectively. Later in the summer on 15 July, water control (36%), horticultural oil (32%), spirotetramat low rate (13%), and spirotetramat high rate (8%) showed increase in branch mortality. By contrast, bifenthrin and pyriproxyfen had no dead branches (Fig. 5).

ARMORED SCALE STUDIES. Under field conditions, densities of live pine needle scales were significantly reduced ($F = 4.68$; $df = 5, 36$; $P = 0.002$) by all the insecticides that targeted peak crawler activity compared with the water control. Horticultural oil, pyriproxyfen, bifenthrin, and spiromesifen showed the greatest reduction of 52%, 45%, 39%, and 38%,

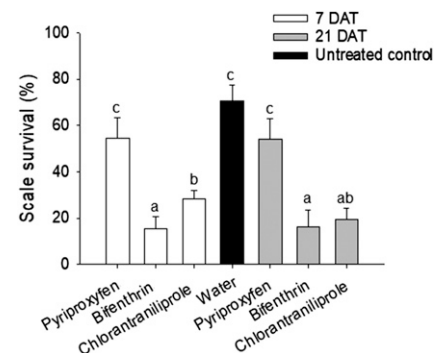


Fig. 3. Mean \pm SE survivorship of calico scale crawlers on leaves with 7- and 21-d-old insecticide residues. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test; DAT = days after treatment.

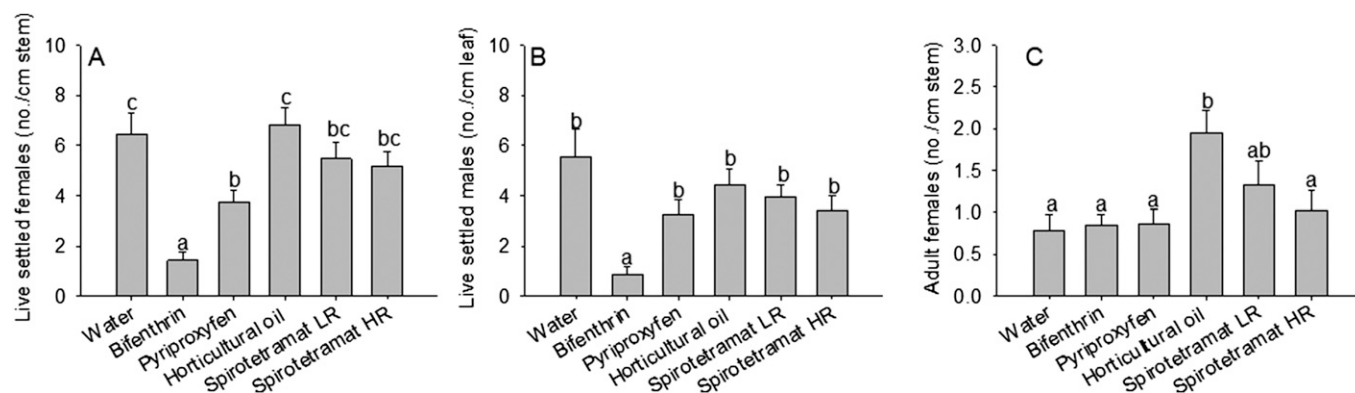


Fig. 4. Effects of insecticide applications on mean \pm SE of striped pine scale crawlers and females on scots pine trees applied on 26 June 2014. A and B represent the effect for each insecticides on immature and C represents the effect on adults. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test; LR = low rate, HR = high rate, 1 live settled female or male/cm = 2.54 live settled females or males/inch.

respectively, followed by spirotetramat with a 34% reduction (Fig. 6A).

Survival of oleander scale on english ivy was significantly lowered by all insecticidal treatments [$F = 108.90$; $df = 5, 65$; $P < 0.001$ (Fig. 7A and B)]. Also, when insecticide was applied to plants inoculated with crawlers for 1-week crawler survivorship was 15% lower than when crawlers were inoculated for a 4-week period [$F = 42.80$; $df = 1, 65$; $P < 0.001$ (Fig. 8)]. There was no significant interaction between insecticide treatments and crawler period ($F = 108.90$; $df = 5, 65$; $P < 0.001$). Plants treated with bifenthrin or horticultural oil were the treatments with lowest survivorship of oleander scale followed by spiromesifen, spirotetramat, pyriproxyfen, and the water control (Fig. 7A and B).

Discussion

We found the efficacy of insecticides with different modes of action differed between soft scales and armored scales. Yet, it is common to find U.S. labels of insecticides with generalized instructions for managing all scale insects. These instructions can result in ineffective control because they underestimate biological variability among species of scale can affect susceptibility to a particular insecticide. Scale insects differ greatly in their external morphology and mode of feeding on plant tissues. Soft scales lack the waxy cover of armored scales, and are phloem feeders, whereas armored scales feed on other plant tissues (Ben-Dov and Hodgson, 1997; Kosztarab, 1996; Miller and

Davidson, 2005; Rosen, 1990; Sadof and Neal, 1993). In addition, we found that prolonged periods of crawler activity decreased insecticide efficacy.

Both soft scale species, calico and striped pine scales, were consistently controlled by bifenthrin and pyriproxyfen. Pyrethroids, such as bifenthrin, affect the nervous system of the insect by preventing the closure of sodium channel voltage gates in membranes (Ishaaya, 2001; Yu, 2015). In this study, bifenthrin was the only insecticide that killed immature stages and ovipositing adult soft scale insects. It produced rapid and long-lasting effects in the field and laboratory. When bifenthrin was applied early in the season to target ovipositing females, its residues on leaves were toxic enough to kill crawlers for at least 3 weeks. Pyriproxyfen also provided consistent control of both species of soft scales. This insect growth regulator mimics juvenile hormone activity (Ishaaya, 2001; Yu, 2015), and it is considered to be a reduced-risk insecticide (EPA, 2016). By contrast to bifenthrin, pyriproxyfen is an insect growth regulator. As such, it took longer to kill the insects and affected molting immature stages only. However, our data showed that several months after application, both pyriproxyfen and bifenthrin had reduced population densities of soft scales to the same levels. These findings are similar to those reported by others working on soft scales (Hubbard and Potter, 2006). The effectiveness of pyriproxyfen may be attributed to more than its acute capacity to kill the scale

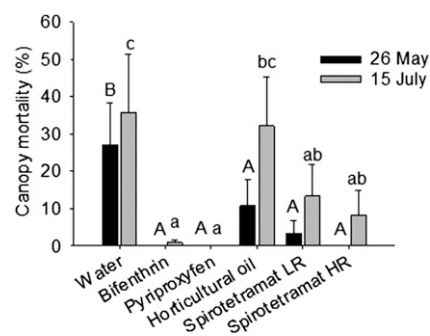


Fig. 5. Mean \pm SE of mortality of scots pine trees due to a striped pine scale infestation. Trees were sprayed on 26 June 2014 and 16 June 2015. Uppercase letters represent mortality on 26 May and lowercase letters represent mortality on 15 July 2015. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test; LR = low rate, HR = high rate.

insects. Its reported low residual toxicity to natural enemies can eventually spare the natural enemies that facilitate sustained rates of scale insect mortality (Ellsworth and Martinez-Carrillo, 2001; Frank, 2012; Naranjo, 2001; Naranjo and Ellsworth, 2009). Similar findings of what has been termed bioresidual activity have been reported in other studies of hemipterans treated with pyriproxyfen (Naranjo, 2001; Naranjo and Ellsworth, 2009).

Spirotetramat is a tetracyclic acid derivative that inhibits lipogenesis in treated insects (Lümmen et al., 2014). Its efficacy against striped pine scale was not different from water controls. However, in the following

year, it depressed populations of overwintering females and reduced dead canopy to the same extent as bifenthrin and pyriproxyfen. Like the long-term effects of pyriproxyfen, this bioresidual activity might also be explained by the purportedly low impact of spirotetramat on natural enemies (Frank, 2012; Planes et al., 2013). Chlorantraniliprole and horticultural oil failed to control either of the two soft scale insects.

Dinotefuran performed inconsistently against calico scale on honey locust. This highly mobile neonicotinoid affects insect nicotinic acetylcholine receptors and can be applied as a foliar, soil, or trunk applied systemic (Simon-Delso et al., 2015). Dinotefuran reduced calico scale densities when it was applied to the soil early in the spring against egg-laying

females before leaves fully expanded. Soil applications in early summer when crawlers were active failed to control scales. Although failure may have been due to a lack of uptake during the historic drought of 2012 that occurred in Indiana at the time of the study, others have also shown inconsistencies with the efficacy of soil-applied neonicotinoids against soft scales (Hubbard and Potter, 2006). Thus, successful use of this systemic material seems dependent on following manufacturer's recommendations for providing adequate moisture after application.

Like our studies of soft scales, both species of armored scale responded to each insecticide in a consistent manner. However, unlike the soft scales, all the insecticides we tested had a negative effect on pine needle scale and oleander scale populations. Bifenthrin and horticultural oil gave the best control against both armored scales. However, pyriproxyfen, spirotetramat, and spiromesifen also significantly reduced the abundance of scales when compared with the water control. Our findings are consistent with other studies of insecticides against armored scales (Frank, 2012; Raupp et al., 2001; Rebek and Sadof, 2003; Xiao et al., 2016).

Differences among the patterns of susceptibility to insecticides among armored and soft scales support our hypothesis that biological differences between these families of scale insects can alter insecticide efficacy. When soft scale insects feed on phloem sap, they produce a liquid excrement called honeydew which together with other substances form their bulky

cover (Ben-Dov and Hodgson, 1997; Kosztarab, 1996). Both cover and integument together can contain as much as 86% honeydew in wax scales (Coccidae), such as white wax scale (*Ceroplastes destructor*), noted for its high content of wax (13%) (Hackman, 1951; Tamaki and Kawai, 1969). This honeydew is 95% water, which can make the scale insect integument highly polar (Ben-Dov and Hodgson, 1997; Leroy et al., 2011; Völkl et al., 1999; Woodring et al., 2004). By contrast, armored scales feed primarily on the contents of plant cells (Sadof and Neal, 1993). These insects do not produce honeydew and are characterized by a waxy cover, which forms within 24–48 h after they settle and envelopes the top of their body. Covers are 50% wax and quite hydrophobic (Miller and Davidson, 2005; Rosen, 1990). We hypothesized that this reported variation in chemical composition of integuments may be partly responsible for observed differences in insecticide efficacy between soft and armored scales. The active ingredients of most insecticides are nonpolar and thus more likely to penetrate the hydrophobic integument of the armored scale (Yu, 2015).

Therefore, it is not surprising to find that more of the products we tested killed armored scales rather than soft scales. The reason that bifenthrin and pyriproxyfen killed both kinds of scales may be because their modes of action were unaffected by the differences in the properties of armored and soft scale integuments. Beyond differences in penetration due to integument composition (Benezet and Forgash, 1972; LeRoux and Morrison, 1954), other factors to consider may include differences in internal distribution, metabolism, target site interaction, and excretion of insecticides or their metabolites (Yu, 2015). For example, differences in the capacity of armored and soft scales to produce honeydew may be linked to fundamental differences in their excretion physiology that could affect insecticide metabolism, excretion, and ultimately, toxicity.

Soft and armored scales are among the most difficult insect pests to control (Fulcher et al., 2012). Timing of application is important for effective management of scales because they can become less susceptible to

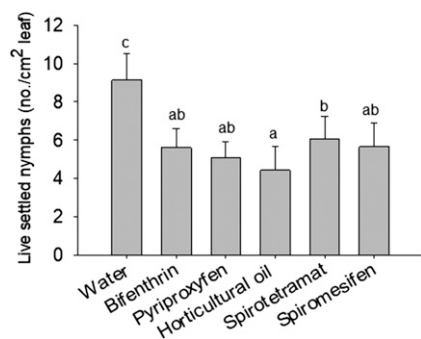


Fig. 6. Effects of insecticide applications on mean \pm SE of pine needle scale crawlers on scots pine trees treated on 12 May 2015. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test; 1 nymph/cm² = 6.4516 nymphs/inch².

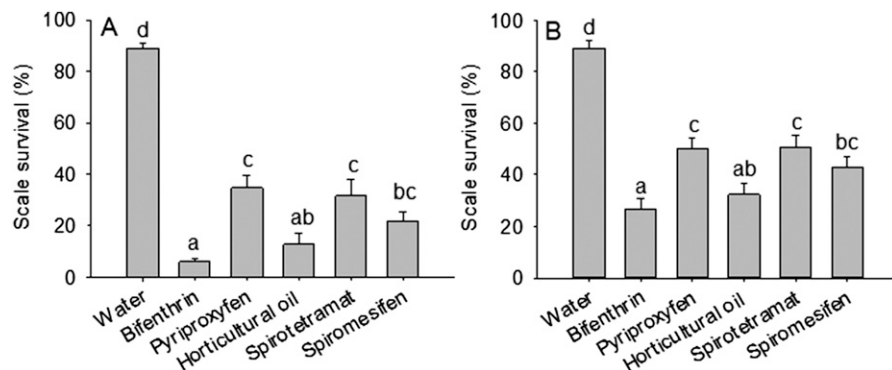


Fig. 7. Effects of insecticide applications on mean \pm SE survival of oleander scale mimicking (A) 1-week and (B) 4-week emergence periods on english ivy plants in Summer 2015. Means followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test.

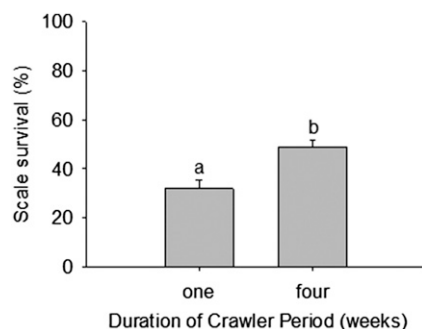


Fig. 8. Effect of 1- and 4-week emergence periods on insecticide efficacy against oleander scale feeding on english ivy plants in 2015. Means (\pm SE) followed by the same letter are not significantly different ($P < 0.05$) according to Fisher's protected least significant difference test.

insecticides as they age (Ebeling, 1936; Phillips and Smith, 1963; Quesada and Sadof, 2017). Our results are consistent with findings by others who indicate that targeting armored scale at the crawler stage is critical to the effectiveness of several insecticides with different active ingredients and classes (Fondren and McCullough, 2005; Frank, 2012; Sadof and Sclar, 2000; Salahuddin et al., 2015; Raupp et al., 2008; Xiao et al., 2016). By contrast, we found that soft scales were susceptible to a narrower range of the products we tested.

Our use of oleander scale to mimic species of scales with short (1 week) vs. long (4 weeks) periods of crawler activity showed that insects with shorter intervals are more susceptible to acute mortality caused by a single insecticide application. This is because a greater proportion of the population is in the susceptible stage at one time. This is consistent with hypotheses that suggest that the strategy of spreading the risk of neonatal mortality over time reduces the impact of acutely dangerous events (Fritz et al., 1982).

In conclusion, we suggest that differences in insecticide efficacy between soft and armored scales could be explained by reported differences in the chemical composition of their integument and covers (Quesada and Sadof, 2017). Most of the insecticides we tested were nonpolar and capable of penetrating the waxy cover of recently settled first instar-armored scales. Thus, the age of the armored scale was the most important factor in determining efficacy because younger scales had fewer layers

of wax to penetrate than older scales. By contrast, although crawlers were still the most susceptible stage of soft scale, the high polarity of soft scale integuments may have increased their tolerance to some insecticides. Thus, by contrast to armored scales, both timing and pesticide influenced the efficacy of the insecticides we tested for soft scales. For this reason, it is important for practitioners to consider that differences between the biology and life history of scale insects could alter the efficacy of products labeled for scale insects.

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