

Variable concentration of soil-applied insecticides in potato over time: implications for management of *Leptinotarsa decemlineata*

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Abstract

BACKGROUND: Select populations of Colorado potato beetle, *Leptinotarsa decemlineata*, in Wisconsin have recently become resistant to soil-applied neonicotinoids in potato. Sublethal insecticide concentrations persisting in foliage through the growing season may select for resistance over successive years of use. Over the 2 years of this study, the aim was to document the in-plant insecticide concentrations over time that result from four different types of soil-applied insecticide delivery for thiamethoxam and imidacloprid in potato, and to measure the impact upon *L. decemlineata* populations following treatments. After plant emergence, insect life stages were counted and plant tissue was assayed weekly for nine consecutive weeks using ELISA.

RESULTS: Peak concentration of both imidacloprid and thiamethoxam occurred in the first sample week following plant emergence. The average concentration of both insecticides dissipated sharply over time as the plant canopy expanded 50 days after planting in all delivery treatments. Both insecticides were detected at low levels during the later weeks of the study. Among-plant concentrations of both neonicotinoids were highly variable throughout the season. Populations of *L. decemlineata* continued to develop and reproduce throughout the period of declining insecticide concentrations.

CONCLUSION: Sublethal, chronic exposure to soil-applied systemic insecticides resulting from these delivery methods may accelerate selection for resistant insects in potato.

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Keywords: insecticide resistance management; Colorado potato beetle; systemic insecticides; imidacloprid; thiamethoxam

1 INTRODUCTION

In-plant or at-plant delivery of insecticides, using both transgenic and conventional approaches, has become one of the most widely adopted arthropod management technologies in integrated pest management (IPM) programs.^{1,2} Flexibility in application type, diversity of active ingredients and focused broad-spectrum control of herbivorous arthropods has driven widespread adoption of these systemic insecticides in nearly every major commodity group worldwide.^{1,3} One of the most popular mode-of-action classes, the neonicotinoids, has occupied approximately 24% of the total global insecticide market share since 2008 (est. revenue \$US 1.17 billion).⁴ Neonicotinoid insecticides have registrations in an estimated 120 countries worldwide, partially owing to an exceptionally wide range of activity against piercing-sucking pests such as aphids, whiteflies, leafhoppers, planthoppers and thrips.⁴ As seed treatments, these compounds also have excellent activity on several other economically important pests, including the Colorado potato beetle (*Leptinotarsa decemlineata*, Say), corn rootworm (*Diabrotica* spp.), seed maggots (*Delia* spp.) and wireworms (*Agriotes* spp.).⁵ Other benefits of this insecticide mode-of-action group (Insecticide Resistance Action Committee mode-of-action group 4A) include versatile application methods (e.g. foliar, seed treatment, soil application, drip, drench), longer periods of residual activity when applied at planting and limited

non-target toxicity.^{6–10} Delivery of these insecticidal active ingredients, which result in systemic movement in plants, has often been classified as an EPA-designated reduced-risk alternative that limits impacts on non-target organisms, decreases additional pesticide use, limits acute and chronic exposure to humans and translates into positive economic benefits to growers.^{5,10–12} Although soil-applied neonicotinoid insecticides have been regarded as beneficial to the agricultural community, considerable concerns about non-target impacts of this mode-of-action class have recently emerged.¹³ To date, exposure of beneficial organisms to soil-applied neonicotinoid insecticides is not well documented in the potato agroecosystem. Undoubtedly a better

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understanding of non-target impacts of neonicotinoids and other systemic insecticides is needed for potato as well as for many other crops and cropping systems.

Since 1995, the majority of potato production acres in Wisconsin have received at-plant neonicotinoid insecticides to manage colonizing populations and early-summer generations of *L. decemlineata*. Repetitive use over a period of 18 years has resulted in resistance to this class in select populations of *L. decemlineata* throughout the United States and Europe.^{14–18} Where resistance occurs, growers continue to apply systemic neonicotinoids for *L. decemlineata* control with limited success, often applying supplemental foliar applications of organophosphate or carbamate insecticides to maintain populations below economic injury levels. Although the mechanisms of insecticide resistance have been extensively studied in this insect, very few studies have focused on examining whether specific insecticide delivery methods could contribute to the development of resistant *L. decemlineata*. Development of resistance to neonicotinoids is expected to vary, depending on the mode of insecticide application (e.g. foliar or systemic);¹⁹ however, few studies speculate on how spatio-temporal changes in the concentration of insecticides may be affected by different types of application.²⁰ Research in other annual crops has shown that in-plant concentrations of the soil-applied insecticides are both spatially and temporally variable throughout the growing season,^{20–23} and diminishing in-plant concentrations often closely coincide with increased arthropod herbivory and economic damage to the crop.^{24,25} In the potato production system, these variable distributions of the systemic insecticides may elicit physiological and behavioral responses that, in turn, could accelerate insecticide resistance development in populations of *L. decemlineata*.²⁶ The long-term impact of variable in-plant concentrations is unknown, but sublethal declining insecticide concentrations that result from soil application methods could compromise the longevity (e.g. selection for broad-spectrum detoxification mechanisms) of future systemic insecticide registrations for *L. decemlineata* and other pests in potato.¹⁹ In the present study, two common neonicotinoids were used as model insecticides to examine changes in the concentration of soil-applied systemic insecticides in potato following different types of insecticide delivery and the response of *L. decemlineata* herbivory resulting from these different delivery methods under commercial potato management conditions. Specifically, the temporal variation in imidacloprid and thiamethoxam concentrations among plants that resulted from four systemic insecticide delivery methods under field conditions was documented using enzyme-linked immunosorbent assay (ELISA) over 2 years, and the population density of a moderately resistant *L. decemlineata* population exposed to each treatment under field conditions over two consecutive seasons was measured concurrently.

2 METHODS

2.1 Experimental site and design

Experiments were conducted at the University of Wisconsin Hancock Agricultural Research Station, Hancock, Wisconsin (44.11726° N, –89.539797° W). The potato cultivar ‘Russet Burbank’ was chosen as a long-season cultivar commonly grown in Wisconsin’s potato production system. Experiments were planted on 27 April 2010 and on 28 April 2011. A 0.2 ha field was planted at a seeding rate of 1 seed piece 0.3 m^{–1} with a 1 m row spacing. Individual plots were four rows wide (6.1 m × 3.7 m, length × width), with an additional untreated guard row adjacent to each plot. Soil

composition was loamy sand with <2% organic matter and pH 7. Selected fields did not have a neonicotinoid-treated crop grown for two prior growing seasons and were considered to be free of residual insecticide in the soil. Best management practices for weed, disease, irrigation and nutrient management for potato in Wisconsin were used.²⁷ Two neonicotinoid insecticides (imidacloprid and thiamethoxam) and three different application methods, plus an untreated control, were included in each year of this study. In 2010, application methods included a conventional in-furrow polyacrylamide impregnation and first-hilling neonicotinoid side dress + soil surfactant. In 2011, the side dress treatment was replaced with a conventional preplant seed treatment application. Plots were arranged in a randomized complete block design with four replications, and the study was terminated when all plots exceeded 90% defoliation or following senescence. Daily irrigation and rain inputs were recorded over the same time interval in each season.²⁸

To reduce chances of false positive signals as a result of misapplication in the field (e.g. insecticide carryover in the soil, maintenance spray tank contamination), a second set of untreated plants were grown under greenhouse conditions as an additional untreated control in the chemical assay component of this study. The potato cultivar ‘Russet Burbank’ was grown in sterile soil-less media (Metro-Mix 300; Sun Gro, Agawam, MA) in the greenhouse and sampled concurrently with the field experiments in each season.

2.2 Insecticides and application treatments

Insecticide treatments of imidacloprid (Admire® Pro and Gaucho® 600; Bayer CropScience, Research Triangle Park, NC) and thiamethoxam (Platinum® 75SG and Cruiser® 5FS; Syngenta, Greensboro, NC) were selected to represent both the majority of at-plant potato applications and two currently labeled insecticides with documented levels of *L. decemlineata* resistance. Insecticide products were commercially formulated and applied at maximum labeled rates (thiamethoxam 140 g AI ha^{–1} or imidacloprid 261 g AI ha^{–1}) for potato in Wisconsin.²⁷

A CO₂-pressurized backpack sprayer with a single nozzle boom was used to deliver an application volume of 94 L ha^{–1} at 207 kPa through a single, extended-range, flat-fan nozzle tip (TeeJet XR80015VS; Spraying Systems, Wheaton, IL) for in-furrow applications. Spray applications were directed onto seed pieces in the furrow at a speed of 1 m s^{–1}, and furrows were immediately closed following application. Polyacrylamide horticultural copolymer granules (JCD-024SM; JRM Chemical, Cleveland, OH) were impregnated with insecticides and subsequently delivered in-furrow at an application rate of 16 kg ha^{–1}. Imidacloprid (2.84 mL) and thiamethoxam (0.834 g) were each diluted (250 mL) in deionized water, and blue dye (Brilliant Blue; Sigma-Aldrich, St Louis, MO) (100 µL) was incorporated into the solution to assist in visualizing uniform mixing. Insecticide solutions were mixed with polyacrylamide (75 g) and then stirred until the liquid was absorbed and a uniform color was observed. Impregnated granules were vacuum dried in the absence of light for 24 h at 20 °C. Dry granules were divided into even quantities per row and evenly distributed into the two center rows for each treatment respectively. Flanking rows received an at-plant, in-furrow application of the same compound and rate. Insecticide applications for side dress treatments occurred simultaneously with fertilizer application (21-0-0-24S), and first hilling occurred on 17 May 2010. A Harriston Model 2010 potato hilling implement (Harriston Industries, Minto, ND) was modified with extended-range flat-fan nozzles (TeeJet TP-4001E;

Spraying Systems, Wheaton, IL) mounted above the hill (30 cm) to apply a banded spray of soil surfactant plus insecticide in a delivery volume of 0.766 L ha^{-1} . Soil surfactant (IrrigGold®; Aquatrols, Paulsboro, NJ) was applied at a rate of 0.5% in water, and a full rate of each insecticide was mixed and applied directly to potato hills. Surfactant and insecticide were covered with soil following a hilling application and immediately irrigated (1.89 cm). All other plots received soil surfactant only. In 2011, a CO_2 -pressurized backpack sprayer with a single nozzle boom delivering an application volume of 102.2 L ha^{-1} at 207 kPa through a single, extended-range, flat-fan nozzle (TeeJet XR8001SVS; Spraying Systems, Wheaton, IL) was used for delivery of seed treatments in water (130 mL) directly to suberized cut seed pieces (23 kg) 24 h prior to planting.

2.3 Beetle sampling and damage

During the 2010 and 2011 season, plots were visually assessed for *L. decemlineata* life stages at a weekly interval from 90% stand emergence until plant senescence. Ten randomly selected plants from each plot were visually assessed for the presence of all *L. decemlineata* life stages. Adult beetles and egg clusters were counted directly. Larval life stages were classified into two groups, small larvae (first and second instars) and large larvae (third and fourth instars), on each of ten plants. In Wisconsin, commercial application decisions are most often based on counts of small and large larvae.²⁷ As decision thresholds used by growers in Wisconsin are based upon numbers of immature stages, only counts of small and large larvae are reported.

2.4 Tissue collection and storage

To measure neonicotinoid concentrations in plants over time, leaf tissue collection began at 90% plant emergence on 2 June 2010 and 10 June 2011. Sampling occurred weekly until all plots exceeded 90% defoliation or until plant senescence for a total of nine consecutive weeks. Specifically, one terminal leaflet was selected from mid-canopy and from three randomly selected plants in each plot. Samples were immediately placed on ice until processing. Immediately following collection, a size 4 cork borer (0.52 cm^2) was used to remove four random cores from each individual leaf while avoiding primary venation. Leaf discs from each individual plant were placed into preweighed 1.5 mL microcentrifuge tubes (Eppendorf North America, New York, NY), weighed and frozen at -80°C until chemical analysis.

2.5 Chemical extraction and quantification

Thiamethoxam and imidacloprid residue were measured by ELISA (imidacloprid kit, cat. number 006, Envirologix Inc., Portland, ME; thiamethoxam plate, cat. number CPP-022, Beacon Analytical Systems Inc., Saco, ME) according to the manufacturer's specifications with the following modifications. Owing to antibody specificity of the imidacloprid ELISA kit, a small proportion of measured binding may be the result of secondary imidacloprid metabolites, not the parent molecule. However, other studies have previously documented arthropod efficacy with independent secondary imidacloprid metabolites,^{29,30} and thus the reported signal is considered to be insecticidal and will hereafter be reported as imidacloprid. Sensitivity ranges reported by the manufacturer were $0.2\text{--}6 \mu\text{g imidacloprid L}^{-1}$ and $0.05\text{--}2 \mu\text{g thiamethoxam L}^{-1}$. Prior to analysis of field-collected samples, the assays were calibrated to account for matrix effects of potato tissue.^{20–24} Untreated leaf extracts,

were used to determine the optimum dilution of homogenates in order to avoid matrix effects.^{22,25} Untreated leaf-disc samples were macerated with pellet pestles (K749520; Kontes, Vineland, NJ) in analytical-grade methanol (400 μL ; Sigma-Aldrich, St Louis, MO) to produce pure leaf extract. Two separate sets of five 400 μL serial dilutions of pure leaf extract were prepared in phosphate-buffered saline solution (PBS) containing 0.05% Triton X-100 (Sigma-Aldrich, St Louis, MO) and between 2 and 100% leaf extract. Each dilution series was spiked with standard calibrators supplied in each ELISA kit ($0.2 \mu\text{g imidacloprid L}^{-1}$ and $0.05 \mu\text{g thiamethoxam L}^{-1}$) to produce samples containing identical concentrations of each compound.²¹ Results of this initial experiment showed pure leaf extract had to be diluted to less than 5% to minimize matrix effects. For the 2 and 5% plant extract concentration groups, five separate dilution series of both imidacloprid and thiamethoxam were prepared in deionized water (100 μL) between 0.1 and 10 $\mu\text{g compound L}^{-1}$. To each dilution series, an equal volume of diluted plant extract was added to produce identical extract background.²² Results from this preliminary experiment indicated matrix effects of diluted plant extract would not interfere with the sensitivity range of the ELISA kits.

At the conclusion of the 9 week sample interval in each year, all field samples were removed from the freezer and prepared for assay using a methanol extraction procedure. Samples were homogenized in analytical methanol (400 μL) with pellet pestles. Homogenates were shaken vigorously on an orbital shaker table at 250 rpm overnight at room temperature and then centrifuged at $10\,000 \times g$ for 5 min to pelletize the particulate matter. Supernatants were diluted 80-fold in PBS containing 0.05% Triton X-100 and used directly for quantification by ELISA. Samples outside the sensitivity range of the assay were diluted further and retested. Greenhouse-grown untreated controls were included in all analyses as an internal standard. All results were quantitatively measured using a microtitre plate reader (VersaMax microplate reader; Molecular Devices, Sunnyvale, CA). Endpoint absorbance values were obtained for samples at an optical density of 450 nm to determine the insecticide concentration of each sample with respect to the standard curve. Calibrator standards provided by the manufacturer served as standard curves and were also used to determine whether non-specific binding had occurred.

2.6 Statistical analysis

To determine the impact of different neonicotinoid treatments on insecticide residue detected in the plant over time, the mean concentration over a 9 week sampling sequence was reported. Independent sampling dates for both insect counts and residue analyses were standardized as days after planting to provide a uniform, continuous graphical representation between years. All analyses considered days after planting as a discrete, ordinal factor level response. All data manipulation and statistical analyses of in-plant concentrations and pest counts were performed in R,³¹ using the base distribution package. Functions used in the analysis are available in the base package of R unless otherwise noted. To avoid pseudoreplication, individual leaf residue values and plot subsamples were pooled into an experimental unit level mean for each observation week prior to statistical analysis.³² Preliminary statistical analyses of untreated controls grown in the field and greenhouse were not significantly different, which shows that plants grown under field conditions were not exposed to carryover from either neonicotinoid in the soil or through misapplication. Plot means for each week in each year of the analyses were subjected to a repeated-measures analysis of variance (ANOVA)

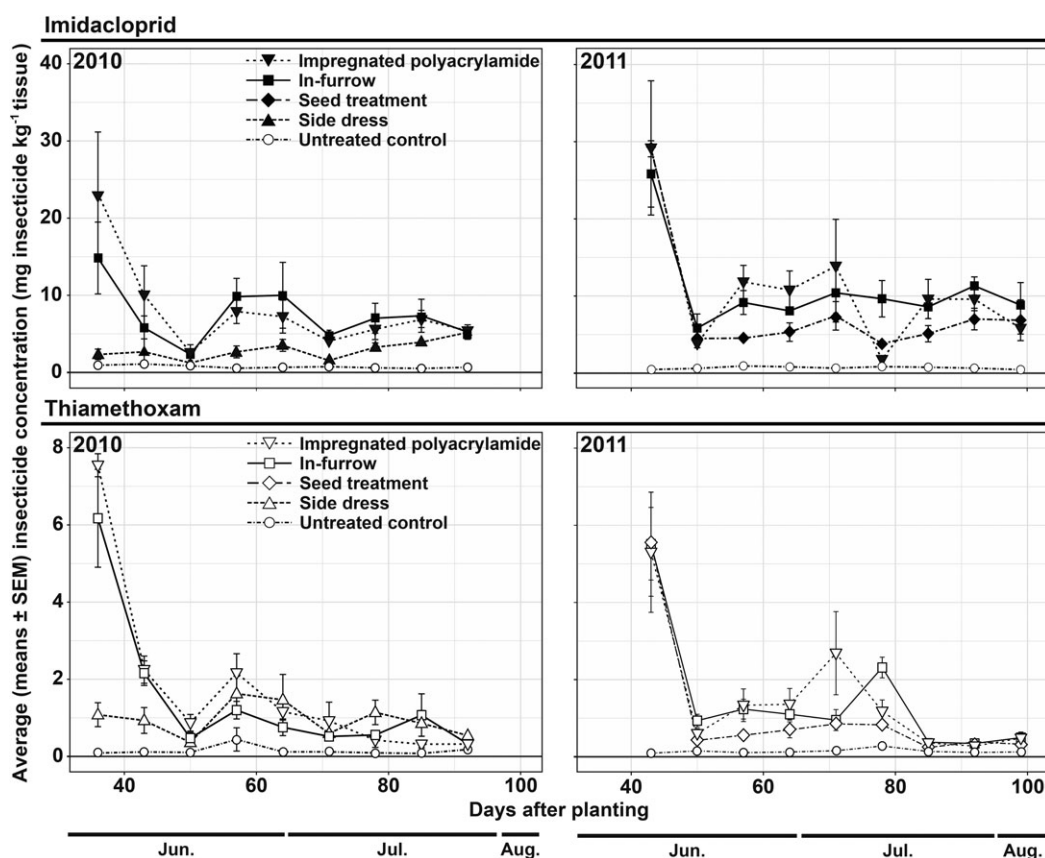


Figure 1. Average neonicotinoid concentration for four different soil application methods over nine consecutive weeks, estimated by enzyme-linked immunosorbent assays. Note that the side dress application in 2010 was replaced with seed treatment application in 2011.

using a linear mixed-effects model to determine significant delivery (i.e. treatment), date and delivery by date interaction effects ($P < 0.05$). Because the magnitudes of residues were markedly different between imidacloprid and thiamethoxam, and given that the comparison of interest was at the insecticide delivery treatment level, insecticide concentrations were analyzed separately for each year and active ingredient. Repeated-measures structured models were fitted using mixed-effects models employing the *lme* function.³³ Empirical autocorrelation plots from unstructured correlation model residuals were examined using the *ACF* function. Correlation among within-group error terms were structured and examined in three ways: (i) unstructured correlation, (ii) with compound symmetry using the function *corCompSymm* and (iii) with first-order autoregressive covariance using the function *corAR1*. As models were not nested, fits of unstructured compound symmetry and first-order autoregressive covariance were compared using Akaike's information criterion statistic with the function *anova* (test = *F*). Insect count and residue data were transformed with natural logarithms before analysis to satisfy assumptions of normality; however, non-transformed means are graphically presented.

3 RESULTS

3.1 Neonicotinoid concentrations in plant tissue

Imidacloprid and thiamethoxam concentrations associated with all application methods declined sharply during the first 50 days after planting, with the exception of the side dress treatment, which did not have a pronounced peak in 2010 (Fig. 1).

Concentrations varied differentially among treatment methods through time for thiamethoxam in 2010 (treatment \times day interaction, $F = 2.5$; $df = 24, 105$; $P < 0.0001$) and 2011 ($F = 6.5$; $df = 24, 105$; $P < 0.0001$). Results for imidacloprid concentrations were also significantly different among treatments over time in 2010 (treatment \times day interaction, $F = 6.5$; $df = 24, 105$; $P < 0.0001$) and 2011 ($F = 4.3$; $df = 24, 105$; $P < 0.0001$). The significant treatment by day interactions presented here indicate that the effect of time is not the same for all soil application methods for each of the insecticides. Residues of both insecticides declined sharply between 36 and 50 days after planting in 2010 and between 43 and 50 days in 2011 (Fig. 1). Water inputs did not reflect a major weather event that contributed to leaching loss of insecticides in either 2010 or 2011 (supporting information Fig. S1).

Insecticide concentration was variable between individual plants both within and between treatments throughout the season in both years (Fig. 2). Concentrations of imidacloprid averaged 6.2 mg kg^{-1} (± 8.9 ; minimum 0.5; maximum 106.6) in 2010 and 9.1 mg kg^{-1} (± 11.6 ; minimum 0.4; maximum 95.3) in 2011, averaging over the nine sample dates and delivery methods. Average annual thiamethoxam concentrations were estimated as 1.4 mg kg^{-1} (± 2.1 ; minimum 0.1; maximum 13.3) in 2010 and 1.4 mg kg^{-1} (± 2.3 ; minimum 0.1; maximum 21.3) in 2011, again over the nine sample dates and delivery types.

3.2 Insect control

High densities of *L. decemlineata* small and large larvae were observed in untreated plots (Figs 3 and 4) in both years of the study, indicating consistent infestation pressure. *L. decemlineata*

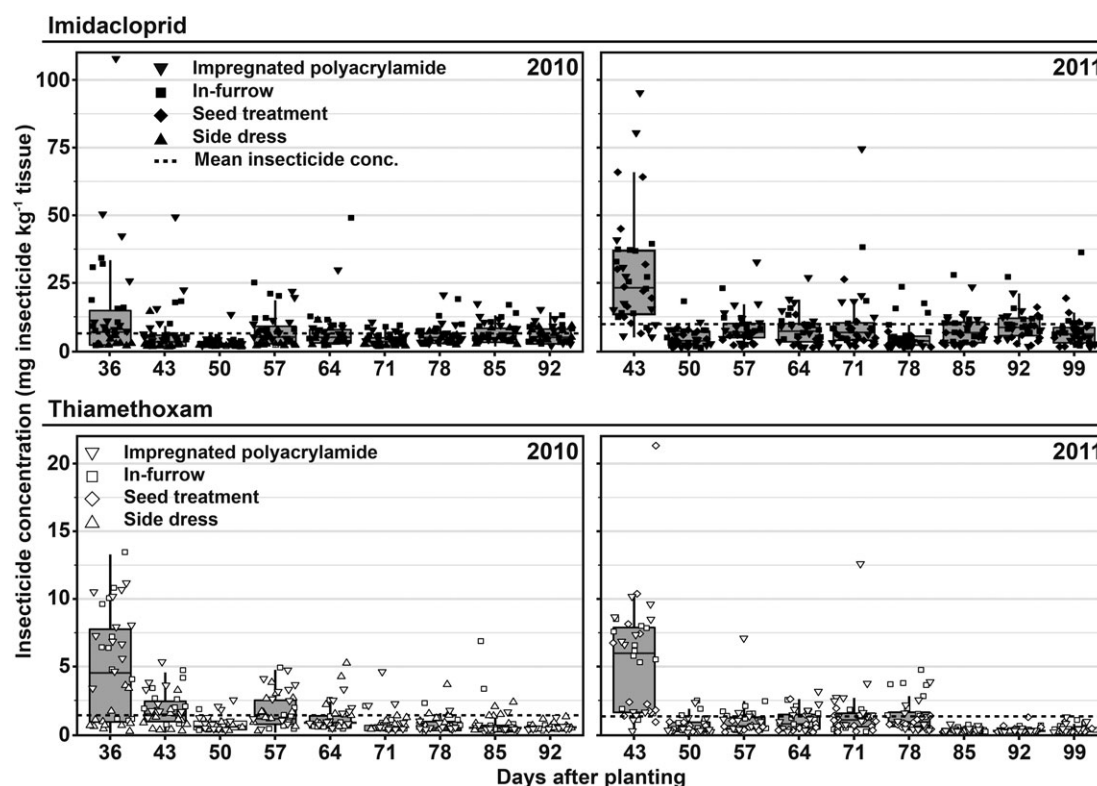


Figure 2. Neonicotinoid concentration estimates in individual potato leaves for four different soil application methods over nine consecutive weeks using enzyme-linked immunosorbent assays. Box plots show pooled variation for all treatments at each sample date. Dotted lines designate the mean insecticide concentration averaging over all sample dates and insecticide delivery treatments for each compound.

small larva densities peaked in the insecticide-treated plots at 50 and 69 days after planting in 2010 and 2011 respectively (Fig. 3). *L. decemlineata* large larva densities peaked in the insecticide-treated plots at 57 and 75 days after planting in 2010 and 2011 respectively (Fig. 4). This increase in small larva density occurred concomitantly with the detection of low insecticide doses measured in the insecticide-treated plots (Figs 1 and 2). *L. decemlineata* density differences between treatment and untreated plots over time show clear differences in control over time (Figs 3 and 4). Furthermore, differences in control were apparent between the active ingredients and delivery methods, as numbers of small larvae varied significantly between treatments through time in 2010 (treatment \times day interaction, $F = 8.7$; $df = 42, 165$; $P < 0.0001$) and again in 2011 ($F = 2.4$; $df = 42, 165$; $P < 0.0001$). Numbers of large larvae varied significantly between treatments through time in 2010 (treatment \times day interaction, $F = 5.6$; $df = 42, 165$; $P < 0.0001$) and again in 2011 ($F = 3.6$; $df = 42, 165$; $P < 0.0001$). The significant treatment by days after planting interactions presented here indicate that the effect of time is inconsistent among application methods and insecticides.

4 DISCUSSION

In Wisconsin, high-intensity potato production has generated stable populations of *L. decemlineata* that are capable of annual economic damage to the crop. As a result, growers have adopted pest management plans largely based on the use of a systemic insecticide to control these recurrent early-season infestations of *L. decemlineata*. For the past 18 years, soil-applied neonicotinoid insecticides have been the backbone of *L. decemlineata* management, and this recurring use has resulted in resistance to these

compounds. Over a period of 5 years, the authors observed a considerable increase in the number of growers reporting loss of efficacy of neonicotinoids. Other studies conducted in intensive potato production areas of the East Coast and Midwest have reported similar losses in levels of control associated with the use of the systemic neonicotinoids.^{15,18,34}

In the present study, high concentrations of neonicotinoid insecticides were observed in potato foliage within 2 weeks of crop emergence, followed by a sharp decline as the season progressed (Fig. 1). Similar patterns in soil-applied insecticide concentrations have been documented for several different application methods (e.g. seed, in-furrow, drip and drench) in other annual herbaceous crops,^{35,36} as well as in perennial tree, shrub and vine crops.^{21–25} This rapid reduction in concentration is intriguing, as these declines closely correspond to expansion of the potato canopy in early June. Other studies in annual crops seldom indicate the potential of rapid plant growth as a factor affecting the dilution of concentrations of the insecticide. Reduction in the concentrations of these insecticides at the time of canopy expansion increases crop vulnerability to direct damage by insect herbivores and also increases the potential for pathogen transmission.^{26,36,37} Furthermore, the non-uniform distribution of systemic insecticides among plants and likely within plants creates the potential for refugia to be present within potato fields, which results in increased selection pressure for accelerated insecticide resistance development later in the season.^{26,38}

Two alternative delivery methods, the side dress and impregnated polyacrylamide treatments, were included in these investigations to determine whether the duration of high insecticide concentrations could be extended further into the growing

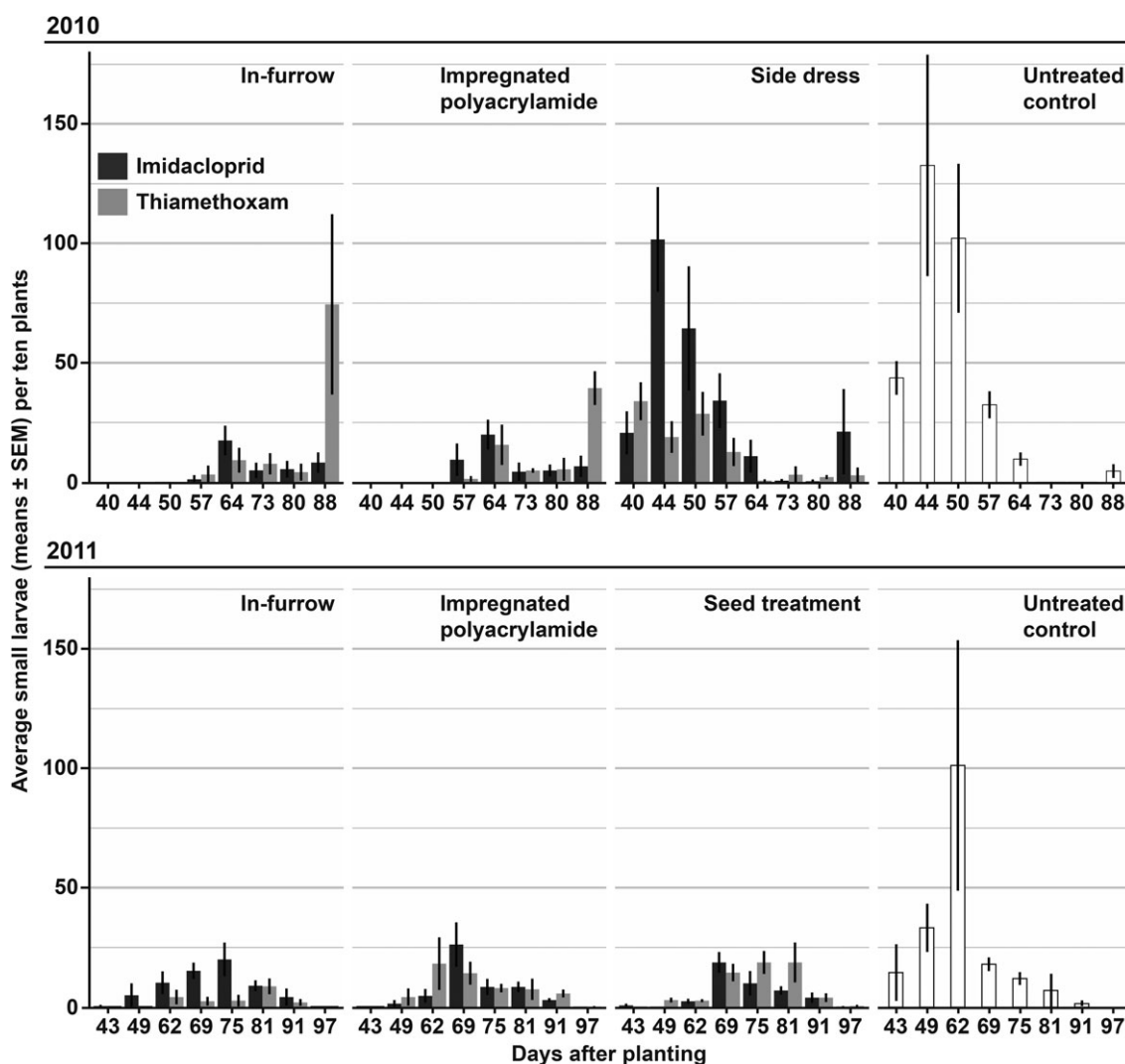


Figure 3. Mean *L. decemlineata* small larvae per ten plants among four different soil application methods for neonicotinoid insecticides.

season. Side dress applications were once common with older systemic compounds such as disulfoton (Di-Syston® 15G; Bayer Cropscience, Research Triangle Park, NC) and aldicarb (Temik® 15G; Bayer Cropscience), but have since been discontinued with registration of in-furrow neonicotinoids and voluntary cancellations. In the present study, full-rate side dress applications of neonicotinoids showed little benefit in extending the interval of insect control or increase in the residual concentrations of insecticides in the plant. An overall reduction in the residual concentration of both active ingredients associated with the side dress use pattern was observed. Interestingly, this method of neonicotinoid insecticide delivery is currently being utilized for the management of another season-long pest of potato, the potato psyllid, *Bactericera cockerelli* (Sulc).^{36,39} To achieve adequate control of this novel pest in potato, many producers are now experimenting with split applications of the neonicotinoid insecticides, incorporating an at-plant, in-furrow application combined with a supplementary side dress application. However, this study documents that side dress application methods did not result in an increased insecticide concentration during the growing season, and there was no statistical increase in control of *L. decemlineata* when compared with other common soil-application methods.

Polyacrylamide gels are commonly used in horticultural and nursery applications as a soil conditioning agent to improve water retention for plant growth.^{40–42} In the present study, polyacrylamide treatments had the highest observed residue levels for both insecticides and the greatest average concentration of active ingredients (Figs 1 and 2). Moreover, the polyacrylamide treatments controlled small larvae for both insecticides in both years (Fig. 3). Delivery of soluble agronomic products with polyacrylamide and other soil-conditioning agents has been researched for several years. In numerous studies these gels were used as a medium to carry plant nutrients in agricultural applications.^{43–45} Impregnation of polyacrylamide with ammonium has also positively increased the total nitrogen concentration in tomato (*Solanum lycopersicum*) and promoted water retention when compared with standard nutrient delivery methods when grown in the greenhouse.⁴⁶ Similarly, higher neonicotinoid concentrations and adequate pest control were documented when the insecticide was delivered using polyacrylamide as opposed to standard in-furrow treatments. Commercial application of impregnated gels could be accomplished, as many growers in Wisconsin are currently equipped to apply dry granular additives at planting. For large-scale applications, naturally derived starch-based

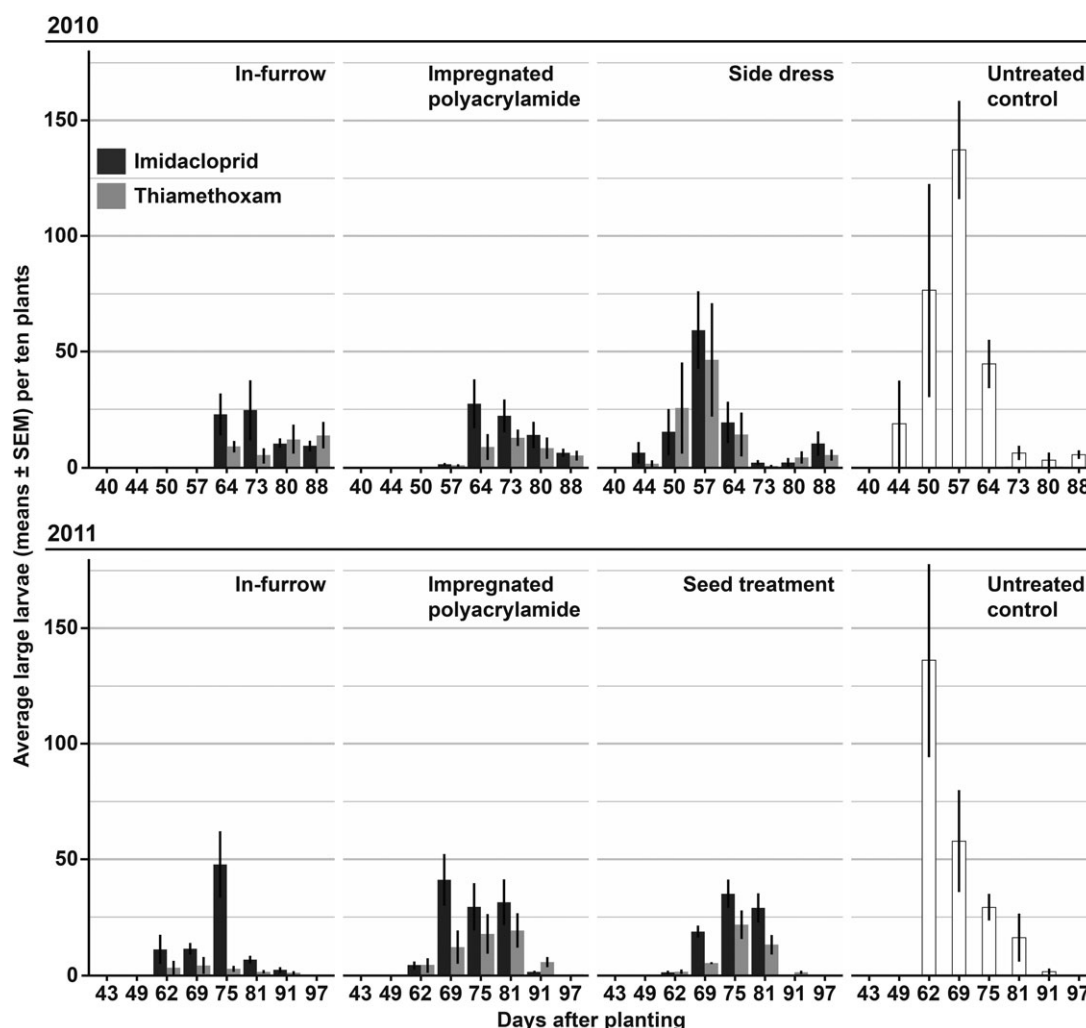


Figure 4. Mean *L. decemlineata* large larvae per ten plants among four different soil application methods for neonicotinoid insecticides.

polysaccharides made from corn or wheat (Zeba®; Absorbent Technologies Inc., Beaverton, OR) may be a suitable alternative to inorganic polyacrylamide-based products. Moreover, experimentation with different types of commercially available polymer could extend the duration of high insecticide concentration further into the season to provide better pest management and possibly reduce the incidence of resistance in a variety of pest species.

5 SUMMARY

In the state of Wisconsin, the majority of commercial potato acreage receives an in-furrow, at-plant neonicotinoid insecticide application to manage *L. decemlineata*, potato leafhopper *Empoasca fabae* (Harris), green peach aphid *Myzus persicae* (Sulzer) and potato aphid *Macrosiphium euphorbiae* (Thomas). Growers have relied upon the neonicotinoid insecticides to control these key insect pests since the initial registration of imidacloprid in 1995. Ease of the in-furrow application method, combined with a long period of residual activity, resulted in an ideal use pattern for large-scale conventional potato production. Unfortunately, widespread adoption and reliance on these at-plant use patterns to manage *L. decemlineata* has resulted in high selection pressure and now resistance to this mode-of-action group.^{16,47,48}

Management options for resistant beetle populations now often result in more frequent applications and tank mixing of several different insecticides, and these additional chemical inputs create considerable economic impacts to growers.⁴⁷

The present authors found that, regardless of the soil application method or neonicotinoid used, insecticides lost efficacy through time, but were still detectable in plant tissues at the conclusion of the study. Under commercial circumstances, growers would make additional foliar applications based on the insect population density and associated defoliation thresholds. For the insect population, multiple modes of action would be operating as a simultaneous selection factor for insecticide resistance within discrete *L. decemlineata* generations. Consideration of the manner in which insecticides are delivered, the residual time of each compound and exposure to the target pest are all critical components in sound resistance management plans. Sublethal chronic exposures from early-season soil-applied insecticides, irrespective of the mode-of-action group, should be carefully evaluated as a possible contributor to emergence of insecticide resistance. Results of this study improve current understanding about the effects of different insecticide delivery methods on among-plant concentration profiles through time and the effects of measured pesticide concentrations on *L. decemlineata*.

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SUPPORTING INFORMATION

Supporting information may be found in the online version of this article.

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