



EVALUATING GRASSLAND WILDLIFE EXPOSURE TO SOYBEAN APHID INSECTICIDES ON PUBLIC LANDS IN MINNESOTA

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SUMMARY OF FINDINGS

Increasing evidence suggests that pesticides may be an important factor explaining declines in grassland-dependent wildlife in agricultural landscapes. Minnesota Department of Natural Resource (MNDNR) wildlife managers and members of the public have reported concerns about foliar-application insecticides in particular. Such insecticides are used on a variety of crops but their use has been especially important for controlling soybean aphid outbreaks in Minnesota. Concerns have been raised about the impacts of chlorpyrifos, a broad-spectrum organophosphate, and other foliar-application insecticides on water quality and human health, prompting the Minnesota Department of Agriculture (MDA) to release guidelines for voluntary best management practices for their use. Although lab studies have shown chlorpyrifos and other insecticides used to target aphids are highly toxic to non-target organisms, including economically important game species and pollinators, few studies have investigated the environmentally-relevant exposure of free-ranging wildlife to these chemicals. Our objective was to assess the direct and indirect exposure of grassland wildlife to the 3 most common soybean aphid insecticides (i.e., chlorpyrifos, lambda-cyhalothrin, and bifenthrin) along a gradient from soybean field edge to grassland interior. During summer 2017 and 2018, we sampled 5 treatment and 4 control sites across western and southern Minnesota. We detected chlorpyrifos at all distances examined (0-400 m) at both treatment and control sites, suggesting that some background level of chlorpyrifos exposure is occurring in the environment regardless of landowner activities in the adjacent row crop field. Our preliminary analyses of filter paper samples (used to quantify direct exposure) showed that insecticide deposition tended to be greater at the field edge than the grassland interior at treatment sites. Furthermore, we detected chlorpyrifos deposition amounts above levels known to cause mortality or morbidity in lab tests for some bird and pollinator species. Our future analyses will use a model-selection approach to determine the effects of weather, vegetation, distance from field edge, and spray application method (i.e., airplane or ground boom) on direct and indirect exposure of wildlife and their invertebrate food resources to these insecticides. Our results will be used to help natural resource managers and private landowners better design habitats set aside for grassland wildlife in Minnesota's farmland region.

INTRODUCTION

Grassland loss and fragmentation is a major concern for grassland-dependent wildlife throughout the Midwestern United States (U.S.). In particular, habitat loss due to agricultural intensification has been implicated as a primary reason for the declines of many grassland nesting birds (Sampson and Knopf 1994, Vickery et al. 1999). However, concerns are

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increasingly being raised about the impacts of pesticides on birds and other wildlife in agriculturally-dominated landscapes (e.g., Hopwood et al. 2013, Hallmann et al. 2014, Main et al. 2014, Gibbons et al. 2015), and some evidence exists that acute toxicity to pesticides may be more important than agricultural intensity in explaining grassland bird declines in the U.S. (Mineau and Whiteside 2013).

Soybean aphids were first discovered in southeastern Minnesota during 2000 and subsequently spread throughout the farmland zone by 2001 (Venette and Ragsdale 2004). Although these aphids pose significant risks to agriculture, their presence does not automatically translate to reduced yield or income (Venette and Ragsdale 2004). In response to concerns over yield loss, the University of Minnesota Extension Office (hereafter, UM Extension) released guidelines on how to scout for aphids and when to consider treatment for infested fields (UM Extension 2014). Foliar applications of insecticides using ground sprayers or airplanes are common treatment methods when chemical control of aphids is necessary. The 2 most common insecticides used are chlorpyrifos and lambda-cyhalothrin (MDA 2005, MDA 2007, MDA 2009, MDA 2012, MDA 2014a) but bifenthrin is also frequently used (N. Davros, unpublished data; E. Runquist, unpublished data). Withholding times vary by chemical (chlorpyrifos: 28 d; lambda-cyhalothrin: 45 d; bifenthrin: up to 14 d); thus, the timing of product use within the growing season needs to be considered. If retreatment is necessary due to a continued infestation, landowners are encouraged to use an insecticide with a different mode of action to prevent resistance (UM Extension 2014) or reduce the impact of insecticide-resistant aphids (UM Extension 2017, UM Extension 2018). Therefore, multiple chemicals may be used on the same field at different times of the year in some situations. Alternatively, landowners may choose to use a product that combines 2 or more chemicals together (e.g., chlorpyrifos + lambda-cyhalothrin), and such products are readily available on the market.

Chlorpyrifos (common trade names include Dursban, Govern, Lorsban, Pilot, Warhawk, and Yuma) is a broad-spectrum organophosphate insecticide that disrupts the normal nervous system functioning of target- and non-target organisms through direct contact, ingestion, and inhalation (Christensen et al. 2009). Although first registered for use in the U.S. in 1965, its use as an ingredient in residential, pet, and indoor insecticides was removed in 1997 (except for containerized baits) due to human health concerns (Christensen et al. 2009, Alvarez et al. 2013 and references therein, MDA 2014b). Furthermore, MDA released guidelines for best management practices for the use of chlorpyrifos due to water quality concerns (MDA 2014b). Lab studies have shown chlorpyrifos to be toxic to a variety of aquatic and terrestrial organisms (reviewed in Barron and Woodburn 1995), and some bird and beneficial insect species are especially susceptible to acute toxicity from chlorpyrifos exposure (Christensen et al. 2009, MDA 2014a). Chlorpyrifos is very highly toxic to gallinaceous bird species such as the ring-necked pheasant (*Phasianus colchicus*) and domesticated chickens (*Gallus gallus domesticus*), with a lethal oral dose causing death in 50% of treated animals (LD₅₀) of 8.41 mg/kg and 32-102 mg/kg, respectively (Tucker and Haegele 1971, Christensen et al. 2009). Several other bird species are also particularly susceptible to chlorpyrifos, including American robins (*Turdus migratorius*), common grackles (*Quiscalus quiscula*), and mallards (*Anas platyrhynchos*; Tucker and Haegele 1971, Christensen et al. 2009). Yet few field studies have been able to document direct mortality of birds from chlorpyrifos exposure (e.g., Buck et al. 1996, Martin et al. 1996, Booth et al. 2005), and an ecotoxicological risk assessment conducted by Solomon et al. (2001) concluded that the available evidence did not support the presumption that chlorpyrifos use in agroecosystems will result in extensive mortality of wildlife. However, chlorpyrifos exposure leading to morbidity (e.g., altered brain cholinesterase activity, altered behaviors, reduced weight gain, impaired migratory orientation) has been documented in both lab and field studies of several avian species (McEwen et al. 1986, Richards et al. 2000, Al-Badrany and Mohammad 2007, Moyer 2008, Eng et al. 2017). Thus, sub-lethal effects leading to

indirect mortality (e.g., via increased predation rates) or lost breeding opportunities may be a concern for wildlife, especially birds, exposed to chlorpyrifos.

Lambda-cyhalothrin (common trade names include Charge, Demand, Excaliber, Grenade, Hallmark, Icon, Karate, Kung-fu, Matador, Samurai, and Warrior) is a broad-spectrum pyrethroid insecticide that affects the nervous systems of target- and non-target organisms through direct contact, ingestion, and inhalation [National Pesticide Information Center (NPIC) 2001]. Although lambda-cyhalothrin is considered low in toxicity to birds, it is highly toxic to pollinators such as bees (NPIC 2001). Furthermore, field studies have shown lower insect diversity and abundance in fields exposed to lambda-cyhalothrin (Galvan et al. 2005, Langhof et al. 2005, Devotto et al. 2006). Because insects are an especially important source of protein for birds during the breeding season, fewer insects could mean reduced food availability for fast-growing chicks.

Bifenthrin (common trade names include Bifenture, Brigade, Discipline, Empower, Tundra, and Xpedient) is a broad-spectrum pyrethroid insecticide that affects the central and peripheral nervous systems of organisms by contact or ingestion (Johnson et al. 2010). Bifenthrin is low in toxicity to birds, including game species such as northern bobwhite (*Colinus virginianus*) and mallards (oral LD₅₀ values of 1800 mg/kg and <2150 mg/kg, respectively; Johnson et al. 2010). However, there are exposure risks for birds that feed on fish and aquatic insects because bifenthrin is very highly toxic to aquatic organisms (Siegfried 1993, Johnson et al. 2010). Some non-target terrestrial insects are also susceptible to bifenthrin (Siegfried 1993). For example, bifenthrin is very highly toxic to bumblebees, with one study showing 100% mortality by contact (Besard et al. 2010).

Minnesota DNR wildlife managers and members of the public have reported concerns about the effects of soybean aphid insecticides on non-target wildlife, including economically important game bird and pollinator species. Although perhaps unfounded, a frequent public concern is that indiscriminate spraying without first scouting for aphid outbreaks has become the norm and fewer birds and insects are observed after spraying has occurred. Yet little is known about the true exposure of birds and terrestrial invertebrates to these insecticides in Minnesota's grasslands. Distances reported for drift from application of foliar insecticides vary widely in the literature (5-75 m; Davis and Williams 1990, Holland et al. 1997, Vischetti et al. 2008, Harris and Thompson 2012), and a recent butterfly study in Minnesota found insecticide drift on plants located up to 1,600 m away from potential sources (E. Runquist, personal communication). The distance of travel for spray drift is dependent on several factors including droplet size, boom height or width, and weather conditions (e.g., humidity, wind speed, dew point) at the time of application. Guidelines for pesticide application are readily available to landowners and licensed applicators (MDA 2014b, MDA 2014c) so that the likelihood of spray drift can be minimized but there is likely large variation in typical application practices.

OBJECTIVES

Our goal was to assess the environmentally-relevant exposure of grassland wildlife to the 3 most commonly-used soybean aphid insecticides (i.e., chlorpyrifos, lambda-cyhalothrin, and bifenthrin; hereafter, target chemicals) in Minnesota's farmland region. Specific objectives included:

1. *Direct and Indirect Exposure:* Quantified the concentration of target chemicals along a gradient from soybean field edge to grassland interior to assess the potential for grassland wildlife (particularly nesting birds and their young, and beneficial insects) to be exposed to these chemicals: 1a) directly via contact with spray drift, and 1b) indirectly through consumption of insect prey items exposed to the insecticides.
2. *Indirect Effects:* Quantified and compared the relative abundance, richness, diversity, and

biomass of invertebrate prey items along a gradient from soybean field edge to grassland interior prior to and post-application to assess the indirect impact of the target chemicals on food availability for grassland nesting birds and other wildlife.

STUDY AREA

We conducted our study within the southwest (SW), west central (WC), and central (C) regions of Minnesota's farmland zone (Figure 1). Corn and soybeans combined account for approximately 90%, 67%, and 71% of the landscape across these three regions, respectively [U.S. Department of Agriculture (USDA) 2019a, USDA 2019b]. Area set aside as grassland cover on public and private land accounted for 6.9%, 10.0%, and 5.6% of the landscape in these regions, respectively (Messinger and Davros 2018). Since 2003, these regions have also experienced some of the highest estimated use of chlorpyrifos and lambda-cyhalothrin (MDA 2005, MDA 2007, MDA 2009, MDA 2012, MDA 2014a).

METHODS

Experimental Design

A treatment study site consisted of a MNDNR Wildlife Management Area (WMA) immediately adjacent to a soybean field that was sprayed to control for aphids. We worked closely with wildlife managers and private landowner cooperators to select treatment sites. We used sites dominated by a diverse mesic prairie mix containing warm-season grasses and forbs because this mix is commonly used by MNDNR managers and agency partners in the farmland zone to restore habitats for the benefit of grassland birds and beneficial insect species. We also selected control study sites with similar site characteristics except that control sites had corn as the adjacent crop and they were not sprayed with any chemicals to control aphids. We chose sites that were predicted to be downwind (typically east or north) from cooperators' agricultural fields based on typical wind direction patterns determined from archived daily summaries of National Weather Service data.

We sampled 5 treatment sites and 4 control sites across 2 field seasons (summer 2017 and summer 2018; Table 1). Within each treatment site prior to spraying, we established sampling stations at distances of <1 m, 5 m, 25 m, 50 m, 100 m, and 200 m along each of 3 transects. If the site was large enough, we also established a station at a distance of 400 m along each transect. This design gave us a total of 18-21 stations per site. We established transects and stations the same way within control sites. At all sites, transects ran perpendicular to the edge of the cooperator's field and were spaced 90-100 m apart to reduce the likelihood of duplicate insecticide exposure from the spraying event.

Data Collection

To assess the potential for direct exposure of birds and other wildlife to our target chemicals (*Objective 1a*), we deployed passive sampling devices (PSDs) to absorb any chemical drift that occurred. We placed PSDs in treatment fields on the day of but prior to spraying of soybeans. The PSDs were 14 cm tall by 7 cm in diameter and consisted of Whatman™ Qualitative Filter Paper (grade 2; GE Healthcare U.K. Ltd, Little Chalfont, United Kingdom) attached to 0.5-in² hardware cloth formed to a cylinder shape to approximate the size and shape of a large songbird or a gamebird chick. We placed the PSDs at 2 heights (ground and 0.5 m high [hereafter, mid-canopy]) at each of the 18-21 sampling stations per site for a total of 36-42 PSDs/site. Ground-level sampling represented ground-nesting birds and other wildlife that spend the majority of their time on the ground (e.g., gamebirds, small mammals, many species of invertebrates). Mid-canopy sampling represented above-ground nesting birds, songbirds, and many species of spiders and insects. We retrieved the PSDs from the field ≤ 2.25 h after spraying and properly stored them for later chemical analysis. At control sites, we placed PSDs

at both ground and mid-canopy levels at each of the stations. We left the PSDs on site for the same amount of time as PSDs at treatment sites before we collected and stored them for later analysis.

During 2017 only, we used water-sensitive cards (Syngenta Global, Basel, Switzerland) to collect spray droplets from chemical drift. These cards changed from yellow to dark blue when they encountered liquid. We attached 4 cards next to each PSD (2 cards on the vertical plane and 2 cards on the horizontal plane) at each canopy layer (ground, mid) of each sampling station. We used these cards to determine if they could be used as a quicker and cheaper method for qualitatively detecting spray drift in grasslands.

During 2018 only, we deployed PSDs during the pre-spraying period (i.e., 1-3 d prior to spraying) at each <1 m sampling station at 3 treatment and 2 control sites. These samples provided us with a secondary field-based control to determine if our target chemicals could be detected within a site prior to known sampling events (treatment sites only).

To assess the potential for birds and other insectivorous wildlife to be exposed to the target chemicals indirectly via consumption of prey items (hereafter, indirect exposure; *Objective 1b*), we sampled invertebrates ≤ 4 h post-spraying at the <1 m, 5 m, and 25 m stations along each transect (total = 9 stations/site). We sampled ground-dwelling invertebrates using a hand-held suction vacuum and canopy-dwelling invertebrates using a sweepnet. We collected vacuum and sweepnet samples along a 30-m doubled transect (30 m x 2 = 60 m total length sampled) to the right side of the sampling stations and parallel to the soybean field. We combined vacuum and sweepnet samples taken from the same station into 1 sample and properly stored them for later chemical analysis. We sampled control sites using the same methods and timing, with the timing based on when we deployed the PSDs at these sites.

To quantify and compare the effects of target chemicals on the abundance, richness, diversity, and biomass of invertebrate prey items (hereafter, indirect effects; *Objective 2*), we collected vacuum and sweepnet samples from the <1 m, 25 m, and 100 m distances along the 3 transects at each site (total = 9 stations/site). We collected these samples 1-3 d prior to spraying and between 3-5 d and 19-21 d post-spraying at treatment sites. We collected samples along a 20-m doubled transect (20 m x 2 = 40 m total length sampled) but on the left side of the sampling stations and parallel to the soybean field. We combined vacuum and sweepnet samples into 1 sample per station per sampling period and stored them in ethanol for later sorting, identification, counting, and measuring. Each time we returned to the site, we started sampling from the endpoint of the previous 20-m sampling transect. During the 3-5 d and 19-21 d sampling efforts, we also collected invertebrate samples at the same 3 distances along 1 additional transect established >60 m away from but parallel to our 3 main transects. This additional transect provided us with post-spraying control samples to address any concerns about whether our repeat disturbance of the main transects impacted our estimates of indirect effects. We used the same methods and timing to collect our indirect effect samples at each of our control sites. During identification in the lab, we placed emphasis on 4 invertebrate orders important in the diets of grassland nesting birds: Araneae (spiders), Orthoptera (grasshoppers, crickets, and katydids), Coleoptera (beetles), and Hemiptera (true bugs). We sorted all individuals from these orders and identified them to at least the family level for analysis. Quantifying the spider community also allowed us to examine potential impacts on an additional trophic level because spiders are an important predator of insects.

We used portable weather meters (Kestrel 5500AG Agricultural Weather Meters) mounted on tripods and equipped with weather vanes to measure relevant weather data (e.g., temperature, wind speed, wind direction, humidity, dew point) along the center transect at the <1 m, 100 m,

and 200 m stations during the deployment of PSDs and at the <1 m, 25 m, and 100 m stations during pre- and post-spraying insect sampling at each site.

At each site, we collected vegetation data 1-3 d prior to spraying at all stations and again at 3-5 d and 19-21 d post-spraying at the reduced subset of stations (i.e., those that coincided with the indirect effects sampling efforts for invertebrates). We sampled multiple vegetation plots at each station: 1 plot at each PSD station and 1 plot at each end of the 20-m and 30-m insect sampling transects. Data collected at each plot included percent ground cover, percent canopy cover, maximum height of live and dead vegetation, litter depth, vertical density, and species richness. Using a modified point-intercept method, we categorized ground cover into bare ground, litter, or other [i.e., woody debris, rock, or gopher mound; Bureau of Land Management (BLM) 1996]. To determine canopy cover, we took a nadir digital photograph of a 30 cm x 55 cm quadrat at a height of 1.5 m above the ground and used the program SamplePoint to estimate percent canopy cover (Booth et al. 2006). Canopy cover categories included grass, forb, standing dead vegetation, woody vegetation, and other. We recorded the maximum height of live and dead vegetation within each plot to the nearest 0.5 dm. We measured litter depth to the nearest 0.1 cm at 1 point within the plot that represented the average condition of the plot. We measured vertical density by placing a Robel pole in the center of each plot and estimating the visual obstruction reading (VOR) from 4 m away and 1 m above the ground in each of the 4 cardinal directions (Robel et al. 1970). We counted the unique number of grass and forb species in each plot to estimate species richness. Finally, we recorded the dominant grass and forb species (up to 3 species in each category) at each PSD station to obtain a qualitative assessment of the vegetation present at each site.

We sent the PSD samples and invertebrate samples (i.e., the direct and indirect exposure samples, respectively) to the USDA Agricultural Marketing Service's National Science Lab (USDA/AMS-NSL) in Gastonia, NC for chemical residue analysis. Samples were analyzed using a solvent-based extraction method. Extracts were concentrated by evaporation and then analyzed using a gas chromatography/mass spectrometry-negative chemical ionization (GC/MS-NCI) technique or other appropriate method. The USDA/AMS-NSL equipment was capable of an extremely high degree of sensitivity in the limit of detection (LOD) and reported all results to us in parts per billion (ppb). Additionally, although our experimental design focused on soybean fields sprayed with foliar insecticides to control aphids, the chemical analyses allowed us to quantify residue of additional pesticides (e.g., neonicotinoids, fungicides) at minimal extra cost. Obtaining information about other pesticide residues provided us with valuable supplementary information that can be used to support other Section of Wildlife research and management goals.

As an additional control, we sent 5 filter paper samples to the USDA/AMS-NSL lab for chemical residue analysis. These samples were not deployed in the field but had been attached to PSD wire frames and held in a storage bin in the back of a field truck prior to shipment to the lab.

Data Analyses

Data analyses are ongoing at the time of this report. Preliminary analyses related to Objective 1a (direct exposure) are discussed below, and we report means and standard deviations unless otherwise noted. Analyses related to Objective 1b (indirect exposure) and Objective 2 (indirect effects) are too preliminary and are not included here. Results may be subject to change by our final reporting.

RESULTS AND DISCUSSION

During fall 2016, we surveyed 12 farmer cooperatives in 12 counties to gather more specific, localized information about chemical spraying (e.g., type of insecticide, spray application

method) in southern Minnesota. Congruent with MDA's pesticide usage reports (MDA 2007, MDA 2009, MDA 2012, MDA 2014a), the cooperatives reported that chlorpyrifos, lambda-cyhalothrin, and bifenthrin were the most commonly-used foliar soybean insecticides in recent years. Additionally, we learned that neonicotinoids are also present in the chemical mixes used as foliar treatment of crop pests. This information is contrary to the widespread belief that neonicotinoids are only used as a prophylactic seed treatment to protect plants systemically. Based on estimates provided by 8 of the 12 farmer cooperatives, an average of 63% of fields were sprayed by airplane (range: 40-85%) whereas 37% of fields (range: 15-60%) were sprayed from the ground in 2016. Fields are less accessible by tractor when conditions are wet or soybeans are too tall; thus, these factors can influence a landowner's decision on the type of application method (airplane or ground) to use.

In late winter and early spring 2017, we also mailed surveys to landowners adjacent to potential WMA study sites to learn about their soybean aphid spraying practices and to ask for their cooperation with our study (see Appendix 1 in Davros and Goebel 2016 for details). Although our mail surveys helped us identify willing cooperators, we ultimately found that soliciting landowner cooperation by visiting their residences or calling them was more effective. Therefore, we abandoned the mail survey in 2018. Once we secured landowner cooperation, we kept in contact with them throughout the growing season to determine if and when they would be spraying their soybeans for aphids. After they sprayed, we followed up with them to obtain additional relevant data (e.g., insecticide product used, application rate, tank pressure); however, some landowners declined to provide some of the information (Table 1).

We sampled sites between 28 July – 14 September 2017 and 18 July – 5 September 2018, coinciding with peak activity for aphid spraying in the farmland zone (Table 2). We collected a total of 368 direct exposure PSD samples (*Objective 1a*), 81 indirect exposure invertebrate samples (*Objective 1b*), and 297 indirect effects invertebrate samples (*Objective 2*) across both years. Additionally, we collected 30 pre-spraying PSD samples as our secondary field-based controls in 2018.

Our preliminary analyses of direct exposure to drift (*Objective 1a*) indicated that target chemicals were detected on PSDs at all distances examined (0-400 m) at both treatment and control sites (Table 3; Figure 2). These results suggest that some background level of deposition is occurring in the environment at the time of our sampling regardless of spraying status of the adjacent cooperator's field. Although our control sites did not have target chemicals sprayed during our sampling timeframe, our experimental design did not control for nearby fields, including other row crop fields that were adjacent to our WMA sites but not included in our landowner coordination efforts. If other landowners sprayed for aphids near the time of our sampling and drift occurred, then our PSDs would have detected any drift that traveled onto the WMA site. Although shorter distances of 5-75 m for drift from application of foliar insecticides are reported in the literature (e.g., Davis and Williams 1990, Holland et al. 1997, Vischetti et al. 2008, Harris and Thompson 2012), a recent butterfly study in western Minnesota found insecticide drift on plants located up to 1,600 m away from potential sources (E. Runquist, personal communication).

Our preliminary analyses of our secondary field-based controls (i.e., the PSDs deployed during the pre-spraying period in 2018) found that target chemicals were present at very low levels within control and treatment sites (6 ± 2 ppb and 7 ± 4 ppb, respectively) prior to spraying. These results further support our conclusion that chemical deposition is occurring from elsewhere in the environment besides our cooperating landowners. Similar to our other samples, chlorpyrifos was the main chemical detected in these field-based control samples.

Our preliminary analyses also found that target chemical deposition on PSDs tended to be greater and more variable at the field edge than the grassland interior at treatment sites, particularly for sites sprayed by airplane (Table 3; Figure 2). Although an edge effect of drift from adjacent treatment fields might be expected, further analyses are underway to examine this pattern and determine if spray method may be an important factor.

Our cooperating landowners used chlorpyrifos more often than lambda-cyhalothrin or bifenthrin. Thus, we also conducted a preliminary examination of chlorpyrifos deposition levels independent of these other chemicals. Similar to all target chemicals combined, chlorpyrifos deposition tended to be greater along the field edge and treatment fields sprayed by airplane also showed more variable deposition levels out to 50 m compared to treatment fields sprayed from the ground (Table 4; Figure 3). Additionally, the levels of chlorpyrifos we detected as drift onto WMAs (Table 4) were sometimes above the LD₅₀ values reported by Christensen et al. 2009 and Corbin et al. 2009 (Table 5). Chlorpyrifos is highly toxic to honey bees (*Apis* sp.) and can poison non-target insects for up to 24 h after spraying (Christensen et al. 2009). Chlorpyrifos is also very highly toxic to several common farmland bird species, including ring-necked pheasants, American robins, and common grackles. Our preliminary results suggest that birds, pollinators, and other grassland wildlife are being directly exposed to chlorpyrifos drift in Minnesota's farmland regions. However, we did not collect field data to determine if grassland wildlife species were experiencing lethal or sublethal (e.g., impaired movement, reduced foraging, lethargy, reduced body condition) effects from this exposure, and further research would be needed to address these potential impacts.

Our objective with using the water-sensitive cards was to obtain an immediate, qualitative visual assessment of insecticide deposition. However, even moderately high humidity levels produced a color change in the absence of insecticide deposition (Figure 4a and 4b). The cards also picked up dew droplets from the surrounding vegetation that caused discoloration. Thus, we were unable to reliably detect insecticide deposition and quantify drift using these cards. We discontinued their use in 2018.

We will be finalizing our analyses over the next 2-3 months. This fall, we will begin sharing our findings with multiple constituent groups. Our first step will be to share individual, field-level results with each cooperating landowner to engage them, make them aware of how their participation benefited our research efforts, and show them how the aggregated data will be shared with other groups. Subsequently, we will invite these landowners, other agricultural groups (e.g., University of Minnesota's Southwest Agricultural Experiment Station personnel; Soybean Growers Association), and various natural resource professionals to a seminar where we will present our overall findings and public land management recommendations. Our proximate goal with these agricultural community outreach events is multifold: 1) bring awareness to the issue of soybean aphid insecticide drift onto grasslands, 2) engage agricultural partners in coming up with solutions to reduce the potential for drift to occur on these grasslands, and 3) promote good will and communication that could be beneficial if MNDNR conducts further pesticide research in the future. However, our ultimate goal is to provide natural resource managers with information on patterns of soybean aphid insecticide drift onto grassland cover in the agricultural matrix of Minnesota. Understanding these patterns will help us improve management of public lands and better design private lands conservation programs to aid grassland wildlife conservation.

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Table 1. Spray method and application data for soybean aphid spraying events by cooperating landowners adjacent to Wildlife Management Areas (WMA) that were sampled for insecticide spray drift between 28 Jul - 14 Sep 2017 and 18 Jul - 5 Sep 2018 in Minnesota's farmland zone.

Site ID ^a	Spray method	Insecticide trade name	Insecticide active ingredients	Insecticide application rate (L/ha)	Sprayer application rate (L/ha)	Application speed (m/s)	Beheading height (m)
tA	Ground	Endigo	lambda-cyhalothrin + thiamethoxam	0.26	140.3	4.0	0.2
tB	Airplane	Bolton	chlorpyrifos + gamma-cyhalothrin	0.88	18.7	67.9	
tC	Ground	Lorsban 4E	chlorpyrifos	NA ^b	93.5	NA	
tD	Airplane	Lorsban Advanced	chlorpyrifos	1.17	18.7	55.9	2.7
tE ^c	Airplane	Lorsban Advanced; Warrior II	chlorpyrifos; lambda-cyhalothrin	0.44; 0.22	NA	NA	

^aWMA names are not provided to protect private landowner cooperators.

^bData is not available because cooperator declined to provide this information.

^cThis cooperating landowner combined two different trade name insecticides during the spraying event.

Table 2. Location, site type, year sampled, and timing of sampling for Wildlife Management Areas (WMA) sampled for insecticide drift from adjacent row crop fields sprayed for soybean aphids during summers 2017 and 2018 in Minnesota's farmland zone.

Site ID ^a	Region ^b	County	Site type ^c	Year sampled	Range of dates when field sampled
tA	SW	Jackson	Treatment	2017	28 July - 18 Aug
tB	SW	Murray	Treatment	2017	9 Aug - 30 Aug
cA	SW	Jackson	Control	2017	21 Aug - 14 Sept
cB	SW	Lyon	Control	2017	7 Aug - 31 Aug
tC	WC	Lac qui Parle	Treatment	2018	10 Aug - 29 Aug
tD	C	Stearns	Treatment	2018	28 July - 16 Aug
tE	WC	Yellow Medicine	Treatment	2018	7 Aug - 28 Aug
cC	C	Kandiyohi	Control	2018	17 Aug - 5 Sept
cD	WC	Lac qui Parle	Control	2018	18 Jul - 8 Aug

^aWMA names are not provided to protect private landowner cooperators.

^bRegions sampled in this study include the southwest (SW), west central (WC), and central (C) regions. The boundaries for these regions follow the same boundaries as outlined in the Minnesota Department of Natural Resources' annual August Roadside Survey.

^cTreatment sites had adjacent soybean fields that were sprayed for aphids; control sites had adjacent corn fields that were not sprayed for aphids.

^dIncludes first day of pre-spray sampling through last day of post-spray sampling for data collection activities.

Table 3. Mean (\pm SD) values of target chemicals detected on passive sampling devices (PSDs) by distance from soybean field edge to grassland interior on Wildlife Management Areas (WMAs) between 28 Jul - 14 Sep 2017 and 18 Jul - 5 Sep 2018 in Minnesota's farmland zone. Target chemicals included chlorpyrifos, lambda-cyhalothrin, and bifenthrin. Values are reported in parts per billion (ppb).

Site type ^a	Distance from soybean field edge (m)					
	0 m	5 m	25 m	50 m	100 m	200 m
Treatment ^b	35,322 (\pm 145,015)	16,260 (\pm 64,298)	26,712 (\pm 92,827)	385 (\pm 906)	40 (\pm 68)	14 (\pm 10)
Airplane	57,198 (\pm 185,976)	27,080 (\pm 82,113)	44,504 (\pm 117,734)	629 (\pm 1,115)	50 (\pm 84)	7 (\pm 10)
Ground	2,510 (\pm 5,538)	30 (\pm 30)	25 (\pm 27)	19 (\pm 21)	24 (\pm 30)	23 (\pm 30)
Control	41 (\pm 76)	21 (\pm 20)	21 (\pm 19)	21 (\pm 20)	22 (\pm 23)	19 (\pm 23)

^aTreatment sites had adjacent soybean fields that were sprayed for aphids; control sites had adjacent corn fields that were not sprayed for aphids.

^bCooperating landowners at treatment sites sprayed for aphids using either airplane or ground booms.

Table 4. Mean (\pm SD) values of chlorpyrifos detected on passive sampling devices (PSDs) by distance from soybean field edge to grassland interior on Wildlife Management Areas (WMAs) between 28 Jul - 14 Sep 2017 and 18 Jul - 5 Sep 2018 in Minnesota's farmland zone. Values are reported in parts per billion (ppb).

Site type ^a	Distance from soybean field edge (m)					
	0 m	5 m	25 m	50 m	100 m	200 m
Treatment ^b	34,875 (\pm 144,686)	16,049 (\pm 63,954)	26,489 (\pm 92,626)	373 (\pm 879)	38 (\pm 65)	14 (\pm 10)
Airplane	56,451 (\pm 185,631)	26,729 (\pm 81,703)	44,132 (\pm 117,524)	608 (\pm 1,082)	48 (\pm 80)	7 (\pm 10)
Ground	2,509 (\pm 5,538)	30 (\pm 30)	25 (\pm 27)	19 (\pm 21)	24 (\pm 30)	23 (\pm 10)
Control	38 (\pm 72)	20 (\pm 20)	19 (\pm 20)	21 (\pm 20)	21 (\pm 23)	18 (\pm 10)

^aTreatment sites had adjacent soybean fields that were sprayed for aphids; control sites had adjacent corn fields that were not sprayed for aphids.

^bCooperating landowners at treatment sites sprayed for aphids using either airplane or ground booms.

Table 5. Acute toxicity (lethal dose [LD₅₀] values^a) of chlorpyrifos for various species as reported in Christensen et al. 2009^b and Corbin et al. 2009^b.

Species	Scientific name	Oral LD ₅₀	Overall toxicity
Pollinator species			
Honey bees	<i>Apis</i> sp.	59-360 ng/bee	Toxic
Avian species			
Ring-necked pheasants	<i>Phasianus colchicus</i>	8.41 mg/kg	Very highly toxic
Mallards - adults	<i>Anas platyrhynchos</i>	76-490 mg/kg	Moderately toxic
Mallards - ducklings	<i>Anas platyrhynchos</i>	112 mg/kg	Moderately toxic
Northern bobwhite	<i>Colinus virginianus</i>	32 mg/kg	Highly toxic
Canada geese	<i>Branta canadensis</i>	40-80 mg/kg	Highly toxic
Sandhill cranes	<i>Grus canadensis</i>	25-50 mg/kg	Highly toxic
Common grackles	<i>Quiscalus quiscula</i>	5.62 mg/kg	Very highly toxic
Red-winged blackbirds	<i>Agelaius phoeniceus</i>	13.1 mg/kg	Highly toxic
American robins ^c	<i>Turdus migratorius</i>	NA	
Rock doves	<i>Columba livia</i>	10-26.9 mg/kg	Highly toxic
House sparrows	<i>Passer domesticus</i>	10 mg/kg	Highly toxic
European starlings	<i>Sturnus vulgaris</i>	75 mg/kg	Moderately toxic
Lab species			
Domestic chickens		32-102 mg/kg	Highly toxic
Mice		60 mg/kg	Moderately toxic
Rats		95-270 mg/kg	Moderately toxic
Rabbits		1,000-2,000 mg/kg	Slightly toxic

^aThe LD₅₀ value is one common measure of acute toxicity and represents the lethal dose that causes death in 50% of treated animals from a single or limited exposure. The LD₅₀ does not reflect any effects from chronic exposure that may occur at levels below those that cause death.

^bSee Literature Cited at end of report for full citations.

^cAmerican robins are the most frequently reported avian species killed in field incidents; however, the LD₅₀ values are unknown.

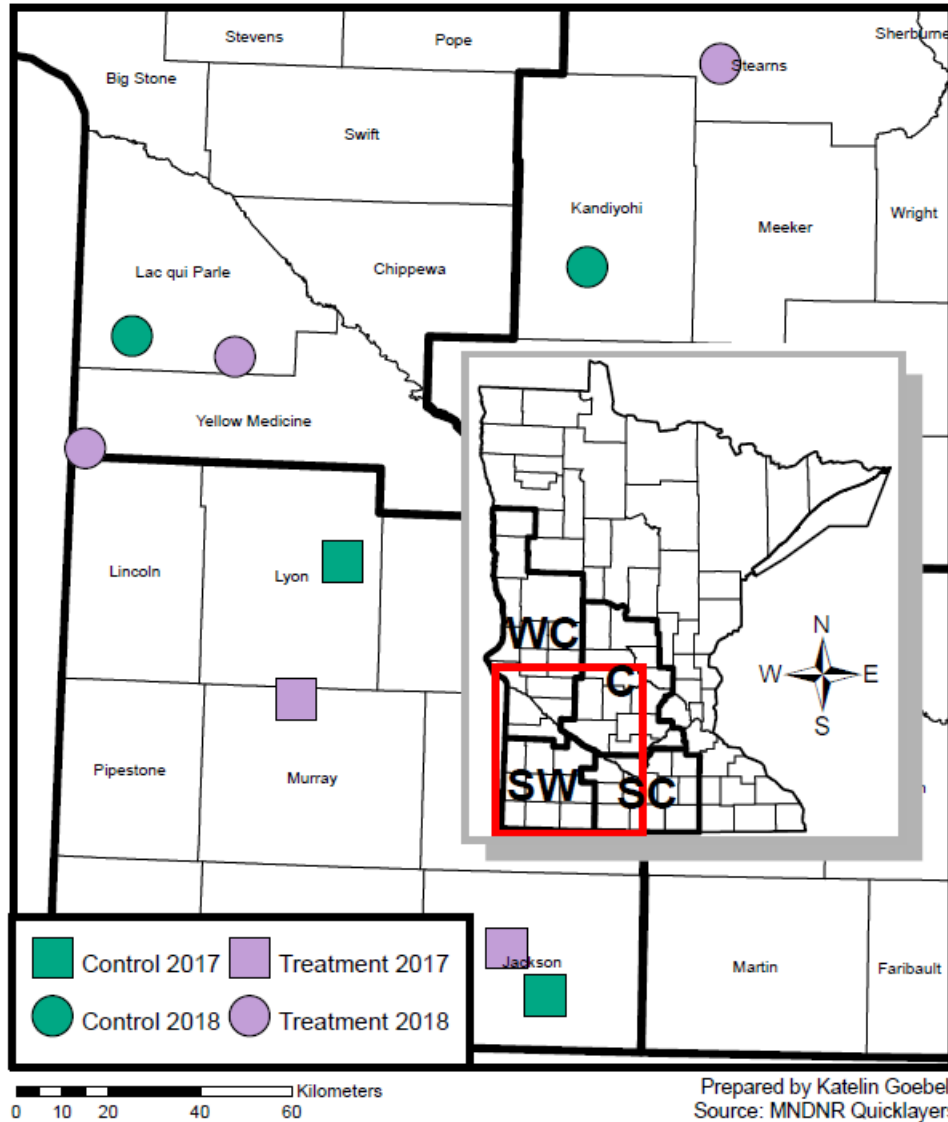


Figure 1. Location of treatment (purple symbols) and control (green symbols) sites during 2017 (square symbols) and 2018 (round symbols) field sampling efforts. Treatment sites were Wildlife Management Areas (WMA) adjacent to soybean fields sprayed for aphids; control sites were WMAs adjacent to corn fields that were not sprayed with insecticides to control soybean aphids. Regions shown are the same as those outlined in Minnesota Department of Natural Resource’s annual August Roadside Survey reports and include: SW = southwest, SC = south central, WC = west central, and C = central.

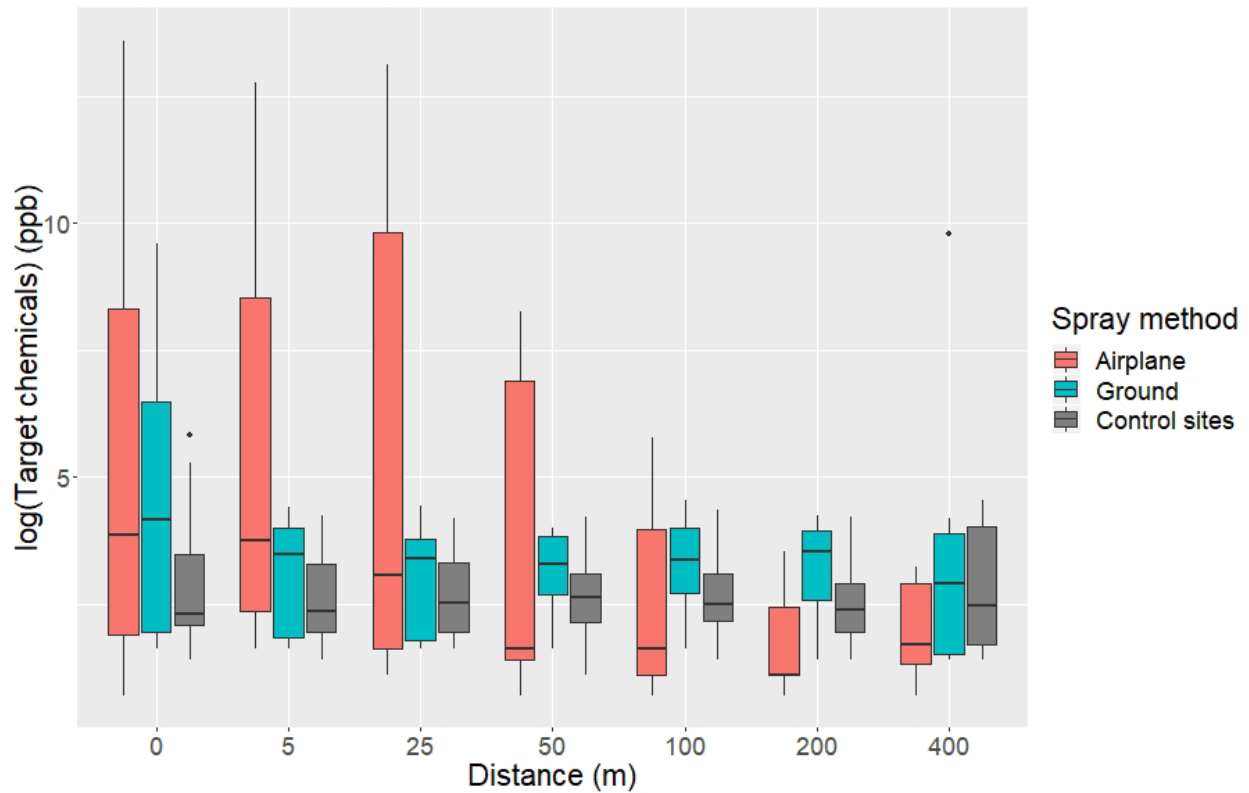


Figure 2. Box plot summaries of target chemical deposition on passive sampling devices (PSDs; $n = 368$) by distance from field edge to grassland interior for treatment sites sprayed by airplane (orange) or ground boom (blue) and control sites (gray), July-September 2017 and 2018 in Minnesota's farmland zone. The PSDs were used to quantify the potential for grassland wildlife to be exposed to chlorpyrifos, lambda-cyhalothrin, and bifenthrin directly through spray drift. Spraying at treatment sites occurred on soybean fields adjacent to grasslands; control sites were grasslands adjacent to unsprayed corn fields. The 0 m distance represents the grassland/row crop edge. Note that distances shown on the x-axis are not graphed to scale.

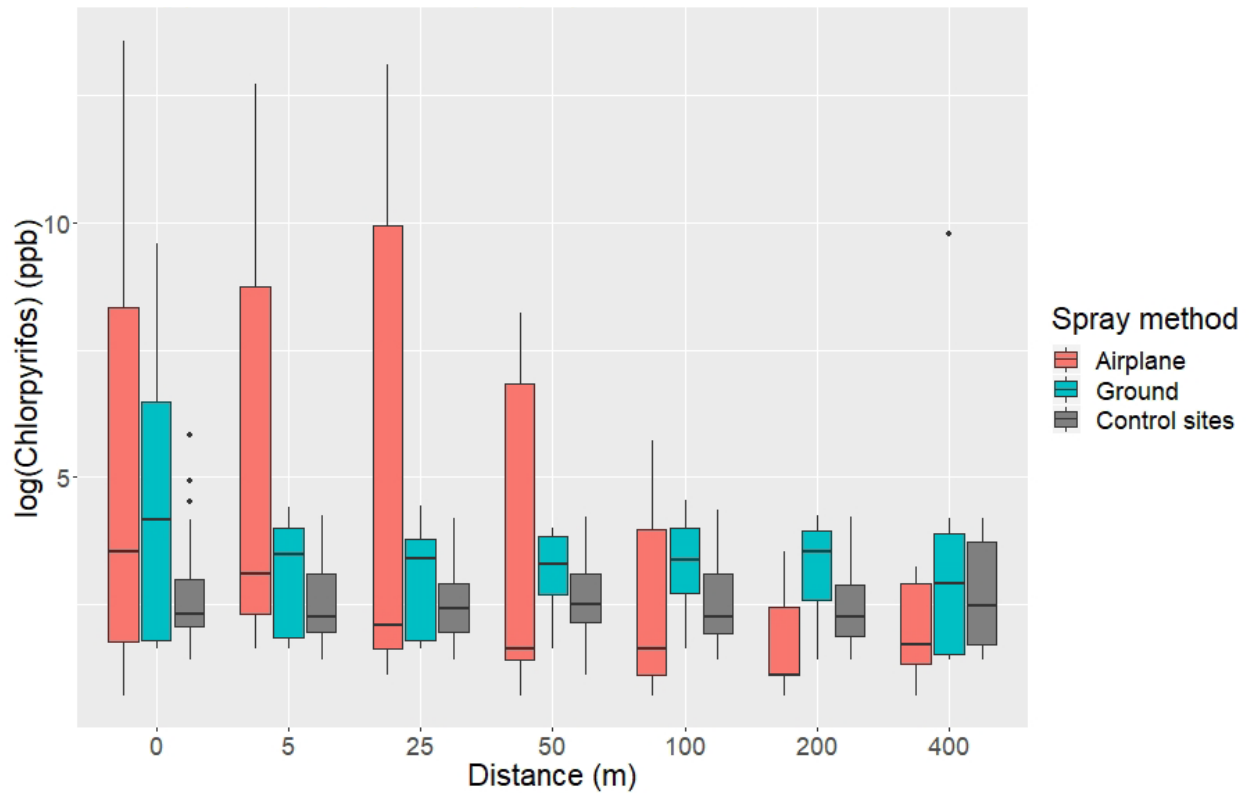


Figure 3. Box plot summaries of chlorpyrifos deposition on passive sampling devices (PSDs; $n = 368$) by distance from field edge to grassland interior for treatment sites sprayed by plane (orange) or ground boom (blue) and control sites (NA; gray), July-September 2017 and 2018 in Minnesota's farmland zone. The PSDs were used to quantify the potential for grassland wildlife to be exposed to chlorpyrifos directly through spray drift. Spraying at treatment sites occurred on soybean fields adjacent to grasslands; control sites were grasslands adjacent to unsprayed corn fields. The 0 m distance represents the grassland/row crop edge. Note that distances shown on the x-axis are not graphed to scale.

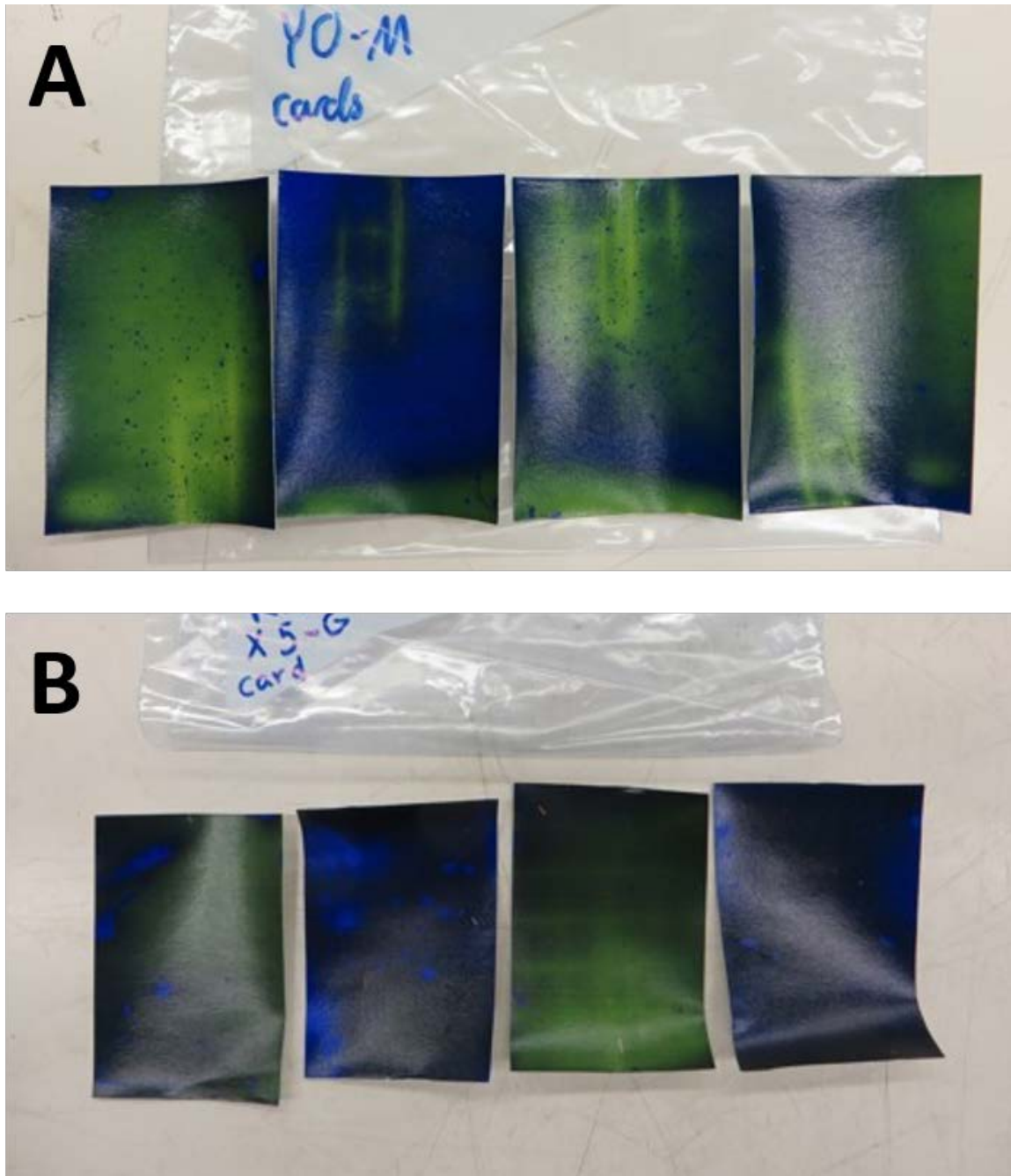


Figure 4. Water-sensitive cards were used during July-September 2017 in an attempt to qualitatively assess insecticide deposition along a gradient from soybean field edges to grassland interiors in Minnesota. Cards turned from yellow to blue when exposed to liquid but relative humidity (RH) levels above 60% also caused the cards to discolor significantly. A) Spray droplets are visible on cards placed at the mid-canopy height a distance of 0 m from the soybean edge at a treatment site; RH was 91% at the time of sampling and also caused major discoloration. B) No evidence of spray droplets is visible on cards placed on the ground at a distance of 5 m from the corn field edge at a control site; RH was 60% and caused the cards to be almost completely discolored; droplets from dew are also visible.