M.L. 2016 Project Abstract
For the Period Ending June 30, 2019

PROJECT TITLE: Evaluating insecticide exposure risk for grassland wildlife on public lands
PROJECT MANAGER: Nicole M. Davros
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FUNDING SOURCE: Environment and Natural Resources Trust Fund
LEGAL CITATION: M.L. 2016, Chp. 186, Sec. 2, Subd. 03n

APPROPRIATION AMOUNT: $250,000
AMOUNT SPENT: $240,096
AMOUNT REMAINING: $9,904

Sound bite of Project Outcomes and Results
Insecticide drift from soybean aphid spraying occurred in grasslands and was greatest along field edges, but wind direction, air temperature, and grassland vegetation structure also played a role. We will work with natural resource professionals and agricultural groups to develop recommendations for reducing impacts of spray drift on grasslands to protect and conserve declining wildlife in Minnesota.

Overall Project Outcome and Results
Concerns about the impact of insecticides on birds, pollinators, and other wildlife are gaining increasing attention. Chlorpyrifos, lambda-cyhalothrin, and bifenthrin (hereafter, target chemicals) are three insecticides commonly used to control soybean aphids in Minnesota's farmland region. Lab studies have shown these chemicals to be highly toxic to non-target organisms including several bird and beneficial insect species, but few studies have investigated exposure of free-ranging wildlife to these chemicals. During 2017 and 2018, we collected samples from public grasslands across southwest, west central, and central Minnesota to determine direct and indirect exposure of wildlife to target chemicals, and indirect effects of the chemicals on insect prey important in the diets of grassland birds. We detected target chemicals at all distances examined (0-400 m from grassland edge) at both treatment and control sites, suggesting that some baseline amount of spray drift occurred in the environment regardless of landowner activities in the adjacent crop field. We also examined the importance of weather, vegetation, and other factors in explaining direct and indirect exposure. Notably, we found insecticide deposition directly onto passive sampling devices (used to measure direct exposure) was greater at the field edge than grassland interior, and deposition was also greater at mid-canopy than ground level. We also detected chemical residues on invertebrates (used to measure the potential for indirect exposure of insectivorous wildlife to these insecticides) but we did not find a strong relationship with distance from edge, possibly because we only evaluated indirect exposure ≤25 m from the field edge. We are currently evaluating the indirect effects of spray drift on invertebrate richness, diversity, and biomass. This fall, we will further interpret our findings to understand potential impacts (e.g., sublethal, lethal) of spray drift on various species of grassland wildlife. We will also begin more broadly sharing our findings with multiple constituent groups, including cooperating landowners, agricultural groups, and natural resource professionals. Ultimately, our research on the factors influencing soybean aphid insecticide deposition in grasslands in the agricultural matrix of Minnesota will help improve management of these set-aside habitats for wildlife.

Project Results Use and Dissemination
To date, we have presented our preliminary results at wildlife professional society conferences, DNR regional wildlife meetings, LCCMR/University of Minnesota (UM) pollinator and partner project meetings, graduate student symposia, and a webinar focused on prairie habitat conservation issues. We have also prepared annual

progress reports for DNR and the USGS/Minnesota Cooperative Fish and Wildlife Research Unit. Finally, we have mentioned the study during several media interviews when appropriate. The final results of this research will form the main chapters of a Master's thesis for a graduate student at UM, and the thesis is expected to be completed during fall 2019 as part of her graduation requirements. These thesis chapters will be used to create peer-reviewed publications that will be shared with other scientists and natural resource professionals. We will continue to disseminate our results with DNR wildlife managers and other staff so they can incorporate our findings into their habitat acquisition, restoration, and management activities. We will also share our findings with our private landowner cooperators and the larger agricultural community to bring awareness to the issue of and factors influencing soybean aphid insecticide drift onto grasslands and other set-aside habitats.

Environment and Natural Resources Trust Fund (ENRTF) M.L. 2016 Work Plan – Final Report

Date of Report: August 15, 2019

Final Report: August 15, 2019

Date of Work Plan Approval: June 7, 2016

Project Completion Date: June 30, 2019

PROJECT TITLE: Evaluating Insecticide Exposure Risk for Grassland Wildlife on Public Lands

Project Manager: Nicole M. Davros

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Location:

Regions - Study sites were located across the Southwest, West Central, and Central regions of Minnesota (Fig. 1). However, the results from our study also have implications for the South Central, Southeast, East Central, and Northwest regions where these insecticides are commonly used in agricultural applications.

Counties - Specific study sites were located in Jackson, Kandiyohi, Lac qui Parle, Lyon, Murray, Stearns, and Yellow Medicine Counties.

Total ENRTF Project Budget:

ENRTF Appropriation:	**$250,000**
Amount Spent:	**$240,096**
Balance:	**$9,904**

Legal Citation: M.L. 2016, Chp. 186, Sec. 2, Subd. 03n

Appropriation Language:

$250,000 the second year is from the trust fund to the commissioner of natural resources to evaluate exposure risks of grassland wildlife to soybean aphid insecticides, to guide grassland management in farmland regions of Minnesota for the protection of birds, beneficial insects, and other grassland wildlife. This appropriation is available until June 30, 2019, by which time the project must be completed and final products delivered.

I. PROJECT TITLE: Evaluating insecticide exposure risk for grassland wildlife on public lands

II. PROJECT STATEMENT:

Grassland habitat loss due to agricultural intensification has been implicated as a primary reason for the decline of many grassland-dependent wildlife species, but concerns are increasing about the impacts of pesticides on birds and other wildlife in agricultural landscapes. Indeed, some evidence exists that acute toxicity to pesticides may be more important than agricultural intensity in explaining grassland bird declines in the United States. Although neonicotinoids (a systemic insecticide routinely used on corn and soybeans) are currently under scrutiny for their effects on birds and pollinators, other insecticides are commonly used in Minnesota's farmland regions that may also have negative effects on non-target organisms. Minnesota Department of Natural Resource (MN DNR) wildlife managers and members of the public have reported concerns about foliar-application insecticides in particular, especially chlorpyrifos. These insecticides are used on a variety of crops but their use has been especially important for controlling soybean aphid outbreaks in Minnesota's farmland regions. A common public perception is that indiscriminate aerial spraying without first scouting for aphid outbreaks has become the norm and many people have reported that they observe fewer birds and insects after aphid spraying has occurred. Many grasslands in Minnesota are highly fragmented and surrounded by row crops, including record-high soybean acres (>7 million acres planted) in recent years. Thus, the potential is high for grassland wildlife to be exposed to these common soybean aphid insecticides.

The public's concerns about the impact of these chemicals on wildlife may be well warranted. Lab studies have shown that chlorpyrifos and lambda-cyhalothrin, the two most common insecticides used to treat soybean aphids in Minnesota, are highly toxic to non-target organisms, including several grassland bird and pollinator species. Further, the Minnesota Department of Agriculture (MDA) released guidelines in July 2014 on voluntary best management practices (BMPs) for the use of pesticides in general and chlorpyrifos in particular due to water quality concerns. However, very little is known about the actual exposure risk of upland wildlife to these insecticides in Minnesota's agricultural landscape under typical application conditions. Distance of travel for spray drift is dependent on weather conditions (e.g., humidity, wind speed) at the time of application and the drift distances reported vary widely (e.g., 16 ft to 1 mi). Renewed interest in riparian buffers to help protect water quality and provide wildlife habitat was a key outcome of the 2014 Minnesota Pheasant Summit. In 2015, a new buffer law was established that will require perennial vegetation buffers up to 50 ft wide along public waters and ditches, but buffer practices may be less effective for wildlife conservation if grassland birds, their insect prey, and beneficial insects such as pollinators using these buffers are exposed to spray drift from adjacent field operations. Further, undisturbed grassland habitat acres in the form of Conservation Reserve Program (CRP) fields are declining. The Minnesota Prairie Conservation Plan aims to partially offset these habitat losses by establishing grassland/wetland habitat complexes within the agricultural matrix. However, we need better information on the environmentally-relevant exposure risk of wildlife under typical field application conditions to help land managers and private landowners alike better design grassland habitats set aside for Minnesota's wildlife.

The goal of our research project is to assess the environmentally-relevant exposure risk of grassland wildlife to common soybean aphid insecticides, especially chlorpyrifos, in Minnesota's farmland region. In particular, we will: 1) quantify the concentration of insecticides along a gradient from soybean field edge to grassland interior to assess the potential for grassland wildlife (e.g., nesting birds and their young, beneficial insects) to be exposed to chemicals directly via contact with spray drift and indirectly through insect prey items exposed to the insecticides, and 2) quantify and compare 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 insecticides on food availability for grassland nesting birds and other wildlife.

III. OVERALL PROJECT STATUS UPDATES:

Project Status as of January 1, 2017: We recruited a Masters student, Katelin Goebel, through the U.S. Geological Survey's Cooperative Fish and Wildlife Research Unit at the University of Minnesota (UM) to work on the project. The graduate student is further refining the methods for the field sampling portion of the study through her graduate research proposal. We contacted farmer cooperatives to gather more information about spraying patterns and chemicals used to control soybean aphids in our study area. We also began to identify potential study sites. We drafted an introductory letter and survey that will be sent to neighboring landowners to learn more about their soybean aphid spraying patterns and to ask them to be cooperators with the project. We attended a butterfly & soybean aphid insecticide symposium held on the University of Minnesota campus. The event allowed us to meet and exchange ideas with other researchers interested in the topics of wildlife and soybean aphid insecticides. Finally, we introduced our project at a symposium attended by other researchers with LCCMR/ENRTF funding for projects relating to pollinators.

Project Status as of June 30, 2017: Landowner cooperation is vital to helping us time our field sampling efforts. To enlist the cooperation of landowners, we mailed surveys to landowners and identified several potential cooperators; however, not all of the cooperators are certain that they will spray for aphids this growing season. Therefore, we have continued to identify additional potential WMAs that meet our site criteria and have begun contacting additional landowners via phone and in-person to ask for their cooperation with the project. We have also coordinated with DNR wildlife managers regarding our selection of study sites and we have purchased equipment and supplies for the project. Katelin had her first UM graduate committee meeting in early May and solicited further feedback from her committee to help refine details of the project. Finally, we have identified potential labs that have the expertise necessary to complete the chemical analyses and we are beginning the process to set up a contract with a lab following DNR purchasing policies.

Project Status as of January 1, 2018: We secured cooperation from landowners and completed our first field season of data collection between July-September 2017. The process to secure a contract with a lab to process our samples for chemical analysis (Activities 1a & 1b) is still ongoing. The process of sorting insect samples (Activity 1c) from our first season is underway and we have recruited student volunteers to help us sort the samples. We have started identifying potential study sites for our 2018 field season, and we will be contacting landowners this spring to ask for their cooperation. Finally, we have continued to disseminate information about our project through multiple avenues, including conference talks/posters, regional DNR Wildlife meetings, a multi-agency webinar, and through the media when appropriate.

Project Status as of June 30, 2018: We finalized a contract with a USDA lab and sent our 2017 samples to them for chemical analysis. We recruited three unpaid student volunteers (see previous project status update) and one UM work study student to sort the insect samples from the 2017 season. We also hired a person with insect identification expertise via a temporary appointment through the UM/Coop Unit to identify our insects. We presented project updates at five different meetings (two scientific conferences, two DNR Regional Wildlife meetings, one UM/LCCMR Pollinator Project meeting). After identifying potential WMA study sites for the 2018 growing season, we asked DNR wildlife managers to review the list and provide us information on recent management activities (e.g., planned/completed prescribed burns, grazing) to refine our site list. Currently, we are further refining the list by visiting the sites in-person to see what crops have been planted in the adjacent fields and to make in-person contact with adjacent landowners to solicit their cooperation for this year's sampling efforts.

Amendment Request (06/30/2018)
We are requesting two amendments. First, we want to shift funds within and between categories in Activity 1 of the budget to partially offset a third year of stipend support for the graduate student which was not budgeted for in the original proposal but which is necessary for her to finish the project and complete her degree. Specifically, we want to move $15,240 from the chemical analysis lab contract line (under

Professional/Technical/Service Contracts category) to the University of Minnesota (UM) contract line (also under Professional/Technical/Service Contracts category). This amount reflects the significant savings we have from our chemical analyses lab contract which was cheaper than expected (i.e., expected costs were ≥$350/sample; final costs are $220/sample). We also want to move $14.15 from the miscellaneous sampling equipment and supplies line (under Equipment/Tools/Supplies category) to the UM contract line (Professional/Technical/Service Contracts category) since we have finished purchasing all of our expected equipment and supplies. Our second amendment request is to change the dates under Outcomes 1 and 3 for Activity 1 below. These newly proposed dates better reflect the time that is needed to process samples from summer 2018 and begin data analyses.
Amendment Approved by LCCMR 7/24/2018

Project Status as of January 1, 2019: We received all 2017 chemical residue sample results from the USDA lab by the end of July 2018. We completed our second season of field sampling during July-September but we sampled fewer sites than anticipated this summer due to events out of our control (see "Amendment Request" below for more details). We sent the 2018 samples to the USDA lab for residue analysis in mid-September, and the lab returned all 2018 results to us in mid-December. During the fall 2018 academic semester, we recruited four undergraduate students (three part-time work study students; one part-time non-work study student) through the UM/Coop unit to sort the insect samples from the 2018 season and begin biomass estimation for the 2017 insect samples. We have continued with data entry and proofing, and we have begun preliminary data analyses. We presented project updates at two different meetings (one scientific conference, one DNR Regional Wildlife meeting). Finally, we recruited three unpaid student volunteers for the 2019 J-term to continue processing the 2017 and 2018 insect samples for biomass estimation.

Amendment Request (01/01/2019)
We are requesting a budget amendment to shift Activity 1 funds from the USDA lab contract budget line to the UM contract budget line as well as a new personnel budget line. During summer 2018, multiple events prevented us from sampling our goal number of sites. These events included: 1) a wet spring and early summer which resulted in the aphid outbreak period being temporally compacted (i.e., aphid spraying happened in a shorter window of time and we couldn't get to multiple sites at the same time to complete our sampling as outlined by our experimental design); 2) a cooperating landowner who had a farming accident which resulted in his hospitalization and inability to coordinate with us to time our sampling; 3) landowners not providing enough notice for us to complete our pre-spray sampling; 4) aphid populations not reaching threshold levels for spraying in the cooperator's field, and 5) lower soybean prices which resulted in some landowners deciding not to spray because the economics (i.e., cost of spraying aphids vs. price of soybeans) weren't in their favor. With fewer sites sampled and thus fewer samples sent to the lab, we have a cost savings of $38,280 on the USDA lab contract line. We want to shift this money to our UM contract and to a new budget line for personnel. The additional funds ($21,424) towards the UM contract will: 1) support our graduate student through August, which is a few months longer than previously anticipated but which is needed for her to finish all of her degree requirements, including final data analyses and defending/revising/depositing her thesis, and 2) help us recruit additional work study students to process the insect samples. The funds towards a new personnel budget line ($16,856) will be used to retain our full-time technician, currently on DNR funding that is nearly expended, for three additional months. This technician is also helping process insect samples and enter/proof data. The technician is a temporary employee currently on soft funds, and her work is directly related to the project and necessary for meeting the project outcomes. The insect processing and the data entry/proofing tasks have both been a larger, more time-consuming effort than anticipated, and we need to retain our full-time technician and recruit more work-study students during the spring 2019 semester to keep the project on track with deadlines.
Amendment Approved by LCCMR 2/1/2019

Overall Project Outcomes and Results: Concerns about the impact of insecticides on birds, pollinators, and other wildlife are gaining increasing attention. Chlorpyrifos, lambda-cyhalothrin, and bifenthrin (hereafter, target chemicals) are three insecticides commonly used to control soybean aphids in Minnesota's farmland region. Lab studies have shown these chemicals to be highly toxic to non-target organisms including several bird and beneficial insect species, but few studies have investigated exposure of free-ranging wildlife to these chemicals. During 2017 and 2018, we collected samples from public grasslands across southwest, west central, and central Minnesota to determine direct and indirect exposure of wildlife to target chemicals, and indirect effects of the chemicals on insect prey important in the diets of grassland birds. We detected target chemicals at all distances examined (0-400 m from grassland edge) at both treatment and control sites, suggesting that some baseline amount of spray drift occurred in the environment regardless of landowner activities in the adjacent crop field. We also examined the importance of weather, vegetation, and other factors in explaining direct and indirect exposure. Notably, we found insecticide deposition directly onto passive sampling devices (used to measure direct exposure) was greater at the field edge than grassland interior, and deposition was also greater at mid-canopy than ground level. We also detected chemical residues on invertebrates (used to measure the potential for indirect exposure of insectivorous wildlife to these insecticides) but we did not find a strong relationship with distance from edge, possibly because we only evaluated indirect exposure ≤25 m from the field edge. We are currently evaluating the indirect effects of spray drift on invertebrate richness, diversity, and biomass. This fall, we will further interpret our findings to understand potential impacts (e.g., sublethal, lethal) of spray drift on various species of grassland wildlife. We will also begin more broadly sharing our findings with multiple constituent groups, including cooperating landowners, agricultural groups, and natural resource professionals. Ultimately, our research on the factors influencing soybean aphid insecticide deposition in grasslands in the agricultural matrix of Minnesota will help improve management of these set-aside habitats for wildlife.

IV. PROJECT ACTIVITIES AND OUTCOMES:

ACTIVITY 1: Data Gathering and Analysis – *Assess the potential for grassland wildlife to be exposed directly and indirectly to spray drift from common soybean aphid insecticides, especially chlorpyrifos.*

Description: We will choose Wildlife Management Areas (WMAs) and other MN DNR properties adjacent to soybean fields in Southwest and South Central Minnesota as study sites in consultation with DNR staff, private landowners and operators, and partner agency personnel. Within each study site, we will conduct sampling at stations placed at multiple distances (<5 m to ≥100 m) along each of three transects extending from a treated soybean field edge to an adjacent grassland interior (Fig. 2). Our sampling will be conducted to assess both direct and indirect exposure risk of grassland wildlife, especially birds and insects, immediately after spraying and at additional time periods post-application. Invertebrates in grasslands adjacent to untreated soybean fields will also be sampled as a control.

a) *Direct exposure risk* will be assessed by placing sampling devices at mid-canopy and ground level at each station prior to insecticide spraying. We will collect sampling devices ≤3 days post-spraying for chemical analysis. The sampling devices will be made of a silicone material that will passively absorb organic chemicals, representing the potential for a grassland-dwelling animal to come into direct contact with spray drift during insecticide application.

b) *Indirect exposure risk* will be assessed by collecting invertebrates via sweep-net and pitfall trap sampling at each station prior to insecticide spraying and at ≤3 days, 10 days, and 20 days post-spraying. We will combine sweep-net and pitfall trap samples into one sample per station for chemical analysis. This sampling approach will assess the potential for grassland birds, predatory insects, and other insectivores to be exposed to insecticides indirectly through consumption of insects that were directly exposed to spray drift.

c) *Indirect effects of exposure* will be assessed by collecting invertebrates and sorting them to estimate their relative abundance, richness, diversity, and biomass prior to insecticide spraying and ≤3 days, 10 days, and 20 days post-spraying. We will focus our sampling on two insect orders [Orthoptera (including grasshoppers, crickets, katydids) and Coleoptera (beetles)] due to their importance in grassland nesting bird diets. This sampling approach will help assess potential reductions in prey items due to insecticide spray drift.

Summary Budget Information for Activity 1: **ENRTF Budget: $250,000**
Amount Spent: $240,096
Balance: $9,904

Outcome	Completion Date
1. ***Assess risk of direct exposure to insecticide spray drift:*** Quantify the concentration of soybean aphid insecticides through passive absorption sampling within 3 days post-application at multiple distances from soybean field edge to grassland interior.	1/1/2019
2. ***Assess risk of indirect exposure to insecticide spray drift:*** Quantify the concentration of soybean aphid insecticides in invertebrates at multiple distances and multiple time periods post-application; compare with control fields.	1/1/2019
3. ***Assess indirect effects of insecticide exposure on prey food resources:*** Quantify and compare the relative abundance, richness, diversity, and biomass of insect prey items important to grassland nesting birds at multiple distances and multiple time periods post-application; compare with control fields.	4/15/2019
4. Report findings and make recommendations	6/30/2019

Activity Status as of January 1, 2017: The Masters student, Katelin Goebel, is furthering our research literature review and contacting subject matter experts in preparation for writing her graduate research proposal, which will further refine our field sampling methods. We contacted representatives at 12 farmer cooperatives across 5 counties to gather more information about spraying patterns and chemicals most frequently used to spray for aphids in these counties within our study area. We identified 25 potential study sites via a Geographic Information System (GIS) and further refined our site criteria. We conducted several site visits to further identify site characteristics and determine if the sites meet our criteria. We drafted an introductory letter and survey that will be sent in mid-January to landowners adjacent to potential study sites to learn more about their soybean aphid spraying patterns and to ask them to be cooperators with the project; their involvement will help us precisely time our field sampling during summer 2017.

Activity Status as of June 30, 2017: All of our activity to date has been project planning. No field samples have been collected yet as aphid spraying typically does not occur until late July into September. Spreading our sampling efforts over a large spatial and temporal (i.e., across years) scale is key to having robust, widely applicable results. Thus, our current goal is to sample 2-3 treatment sites and 1-2 control sites during summer 2017 with additional sampling at new sites during the 2018 growing season. We need to time our field sampling efforts closely with the timing of aphid spraying. To aid this effort, we solicited landowner cooperation by mailing two rounds of letters and surveys to landowners in late winter (late February – early April 2017) to identify potential cooperators. Our mailing list included all landowners directly adjacent to WMAs we had identified as potential study sites. We had a 28.1% overall survey return rate but not all landowners filled out the survey completely. Many landowners did not complete the survey because they rent their land and did not have information on aphid spraying practices. In some of these cases, the landowners provided the renter's contact information so that we could contact them. Approximately 13.6% of landowners completed the survey in its entirety and 7 landowners indicated that they will be planting soybeans adjacent to a WMA this season; however, not all of them were certain if they would spray for aphids. These 7 landowners were willing to be contacted again during the growing season so that we could monitor their spraying activities. In case several of them do not spray for aphids, we have continued to identify additional WMA sites and potential landowner cooperators. We have coordinated with DNR area managers to ensure that they do not control weeds with herbicide at our sites this summer as herbicide spraying could confound our results. We have further refined the field sampling methods, including protocols that will be used to collect vegetation data that will be needed as covariates in our data analyses. We have also completed purchasing for equipment and supplies using DNR funding. Finally, we have identified labs that have the expertise to analyze our pesticide residue samples and we are in the process of getting a contract in place, following DNR purchasing policies, to have one of the labs analyze our samples.

Activity Status as of January 1, 2018: We completed our first field season of data collection between July-September 2017. We had identified 16 potential study sites via GIS prior to the start of the field season but in-person site visits reduced our potential list to 7 treatment sites for various reasons (e.g., adjacent row crop was corn instead of soybeans) and 4 control sites. Four out of 7 landowners for our potential treatment sites agreed to cooperate with our study so that we could precisely time our sampling efforts. However, 1 landowner did not spray for aphids in 2017 and 1 landowner failed to give us advanced notice of his spraying efforts. Thus, we only sampled 2 treatment sites in 2017. We also sampled 2 control sites. Overall, we collected 166 direct exposure samples (Activity 1a), 36 indirect exposure samples (Activity 1b), and 132 indirect effect samples (Activity 1c) during our first field season. We are currently working with DNR contract/purchasing staff and the Department of Administration on a contract for a lab that can complete the chemical analyses. The lab is a USDA lab that has been used by other State of Minnesota researchers for similar pesticide analyses. We will want to send samples to this lab in >1 fiscal year; thus, we are working with the Department of Administration to determine whether a multi-year master contract would be most appropriate for our collective purposes and, if so, to establish the contract. This process has taken longer than expected but will be much easier in the future once we get the contract established. Further, it is important to note that the lab typically processes samples in 10 business days once they receive them. We expect that samples will be sent to the lab within the next 3 months and we will have preliminary results for our 2017 samples by late spring 2018. We have begun sorting our insect samples for Activity 1c. Given the volume of samples to be processed, we have recruited 3 J-term volunteers from two different undergraduate colleges (Gustavus-Adolphus College, St. Peter, MN; Luther College, Decorah, IA) to help with these efforts in January. Additional undergraduate students will be recruited to help during the spring semester at the University of Minnesota. Katelin continues to use GIS as a first step in identifying potential study sites, and we will be contacting landowners this spring to ask for their cooperation with our project. Based on our experience from last summer, we have learned that speaking to potential cooperators either in person or via phone is a better option compared to sending them a letter via postal mail. We will also remain in close contact with our DNR wildlife managers to coordinate our field activities with them. In particular, we would prefer that they not conduct any management activities (e.g., spring prescribed burns, herbicide weed control) that could affect our ability to use the sites that we select. Finally, we have continued to disseminate information about our project when/where appropriate. Most notably, Nicole has introduced the project and provided brief updates on it to several audiences (e.g., DNR wildlife managers at regional meetings, a multi-agency webinar, and an article in the Star Tribune) and Katelin submitted abstracts for presentations (one lightning talk; one poster) at two upcoming professional wildlife society conferences. Both abstracts have been accepted.

Activity Status as of June 30, 2018: With significant help and guidance from DNR and MN Department of Administration contract and purchasing specialists, we finalized our contract with the USDA lab in April and sent the 2017 samples to them at the end of May. Given the lab's processing timeline goals, we expect to have all 2017 raw data for Activities 1a and 1b by mid-July; partial raw data has been sent to us already. By recruiting 3 unpaid college J-term volunteers (see previous activity status update for details) and 1 undergraduate work study student from UM-Twin Cities, we were able to sort the 2017 invertebrate samples for Activity 1c by the end of May. Additionally, we hired a person trained in invertebrate taxonomy through the UM/Coop Unit to identify our samples for us. Invertebrates in the Orders Araneae, Coleoptera, Hemiptera, and Orthoptera were identified to Family whereas all other insects were identified to Order (e.g., Diptera, Hymenoptera). Our decision to identify these particular Orders to Family was based on several reasons: 1) Activity 1c places emphasis on Araneae, Coleoptera, and Orthoptera, 2) Hemiptera are a very diverse Order and additional information can be gained from sorting them to the level of Family, and 3) it is very time-consuming and cost-prohibitive to identify other Orders to a further level of resolution. Finally, our goal is to sample 6 treatment and 2 control sites during the 2018 field season. In preparation, Katelin identified >75 WMAs as potential study sites based on our study design criteria and we sent the preliminary site list to managers for review. After incorporating their feedback (e.g., site is too wet, site is planned for spring 2018 burning, etc) which reduced our list of potential sites, Katelin began conducting site visits in mid-June to: 1) further determine if the sites fit our criteria (e.g., appropriate plant diversity, correct row crop planted this growing season along the desired adjacent edge), and 2) to make first contact with landowners by visiting them in-person. Wet field conditions during planting and the early growing season has impacted soybean plant germination and growth; the impact of the rain on aphid populations (and thus the timing of our field sampling) this year is yet to be determined.

Activity Status as of January 1, 2019: During summer 2018, we visited 48 WMAs that had been identified as potential study sites in GIS. Of those sites, we identified 16 sites that met the criteria of our experimental design. We visited with landowners both over the phone and in-person, and 10 landowners agreed to cooperate with our study so that we could precisely time our sampling efforts. Despite having landowner cooperation, several of these sites were not sampled because: aphids did not reach threshold levels, landowners failed to provide enough lead time for us to conduct pre-spray sampling, multiple sites were sprayed during the same time period and we could not sample both sites at the same time, or landowners did not spray due to cost of spraying vs. economic value of soybeans. Thus, we sampled 3 treatment and 2 control sites during summer 2018 which brought our total to 5 treatment and 4 control sites over the course of the entire study. Total sample sizes across the 2017 and 2018 field seasons are: 398 filter paper samples for direct exposure analysis (Activity 1a), 81 invertebrate samples for indirect exposure analysis (Activity 1b), and 297 invertebrate samples for indirect effects analysis (Activity 1c). The USDA lab returned all 2017 results to us by the end of July. After completing field sampling in September, we shipped the 2018 samples for residue analysis (Activities 1a & 1b) to the USDA lab, and we received all results by mid-December. We plan to send an additional 5 samples to the lab as “true controls” or “blanks.” These additional samples are filter paper samples that have never been in the field and will serve to validate the lab’s quality control measures. During the fall academic semester, we recruited 3 part-time undergraduate work study students and 1 part-time non-work study student through the UM/Coop unit to process the insect samples for Activity 1c. We also retained our full-time DNR technician to help with all data entry/proofing duties and to help with processing the insect samples for Activity 1b. Currently, all 2017 invertebrate samples for Activity 1c have been sorted and identified but they have not been counted or measured. The 2018 invertebrate samples are 95% sorted and the sorting will be completed by mid-January. In December, we hired a person trained in invertebrate taxonomy through the UM/Coop Unit to begin identifying our 2018 samples after which they will be counted and measured. Three new undergraduate J-term volunteers (unpaid) have been recruited from Gustavus-Adolphus College and Luther College to help with the Activity 1c duties during January. We conducted preliminary analyses of the 2017 raw data related to Activities 1a & 1b. Nicole presented those preliminary results at a DNR Region 1 Wildlife Meeting in September and Katelin presented them at the national conference of The Wildlife Society in October. Katelin has also submitted a poster abstract to share these preliminary findings at the Minnesota Chapter meeting of The Wildlife Society in February 2019. The preliminary findings indicate that insecticide drift is occurring on our study sites, and wildlife have the potential to be exposed to these chemicals either directly (Activity 1a) or indirectly (Activity 1b) at multiple distances from the soybean field edge. Our future analyses will incorporate the 2018 data and covariates such as distance from edge, application method (i.e., plane or ground boom), weather data (e.g., wind speed, direction, humidity), and vegetation data (e.g., canopy cover, vertical density) to better elucidate the relationships between aphid spraying in soybean fields and the direct and indirect effects on wildlife in adjacent grasslands.

Final Report Summary: Through close cooperation from landowners adjacent to WMAs, we sampled a total of 5 treatment and 4 control sites during fieldwork between 28 July – 14 September 2017 and 18 July – 5 September 2018 (Table 1). Three treatment sites had adjacent soybean fields that were sprayed for aphids by airplane whereas 2 treatment sites had adjacent soybean fields that were sprayed by ground (Table 2). Our cooperators primarily used chlorpyrifos or lambda-cyhalothrin, or a combination of those two chemicals although other chemicals (gamma-cyhalothrin, thiamethoxam) were also used during the spraying event (Table 2). In total, we collected 398 filter paper samples for direct exposure analysis (Activity 1a), 81 invertebrate samples for indirect exposure analysis (Activity 1b), and 297 invertebrate samples for indirect effects analysis (Activity 1c). Precisely timing our field sampling with spraying operations was difficult and time-consuming because it required contacting many more landowners than sites were needed in order to secure enough cooperators; however, we felt this approach was the most appropriate for gathering the data needed to evaluate our objectives properly, given our experimental design. We also experienced hesitancy to cooperate on the part of many landowners because any drift is illegal and they feared they would be penalized if the data were made public. To garner their support, we assured them that we would not provide specific site names or locations to protect their identities; therefore, we plan to mask exact study site locations in all of our reports and manuscripts.

Our analyses of direct exposure to drift (Activity 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 3). Given that we detected target chemicals at control sites, our findings suggest that some baseline amount of deposition occurred in the environment on or before the day of our sampling, regardless of whether spraying occurred on the cooperator's adjacent field. Although our control sites were not sprayed with target chemicals, our experimental design did not control for other nearby fields, including additional row crop fields adjacent to other boundaries of our WMA sites. If other landowners in the same landscape 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. Using a hierarchical model selection approach, we also examined factors important in explaining direct exposure via target chemical deposition at treatment sites only. Our results indicate that mean air temperature and direction of the wind relative to the WMA during soybean spraying events, percent canopy cover of live vegetation (primarily grasses and forbs), distance from grassland field edge, and position in the canopy layer were all important factors explaining deposition and drift of target chemicals onto WMAs (Table 4). Notably, we found insecticide deposition onto PSDs was greater at the field edge than the grassland interior (Figure 3), and deposition was also greater at mid-canopy than ground level. Spray application method (i.e., ground or airplane) was not important in explaining patterns of target chemical deposition on our WMA sites.

We also detected target chemical residues on invertebrates (Activity 1b) at all distances examined (0-25 m) at both treatment and control sites (Figure 4). Using a hierarchical model selection approach with data from treatment sites only, we found that mean air temperature and the maximum height of live vegetation best explained patterns of deposition on invertebrates, although a model incorporating distance to field edge was competitive (Table 5). The relationship between chemical deposition on invertebrates and distance from field edge was not strong, however, and is likely due to the shorter range of distances that we evaluated for this activity. Similar to direct exposure (Activity 1a), spray application method was not important in explaining patterns of indirect exposure.

We are still evaluating indirect effects of spray drift on richness, diversity, and biomass of invertebrate prey (Activity 1c). Given the large volume of samples collected and the time limitations involved with using undergraduate students as lab assistants (e.g., working around their course schedules, semester breaks), we did not finish processing these invertebrate samples until early May which has delayed our analyses for this activity. Despite this delay in meeting project deadlines, we would readily use undergraduate students again for this type of work. They not only provide a cost-effective approach but we were also able to provide valuable experience and training opportunities to a wide diversity of undergraduate students, including international students, students from both a large university and smaller, private teaching colleges, and students who are undecided about their future career paths. We have begun exploratory data analyses to examine differences in biomass estimates between treatment and control sites during each sampling period, but these results were not ready at the time of this final report. Our future objectives include: a) making formal statistical comparisons of richness, diversity, and biomass estimates between treatment and control sites, and b) building models relating measures of chemical deposition at treatment sites across sampling periods to our richness, diversity, and biomass estimates to determine if spray drift impacted the availability of food for grassland birds and other insectivores.

Final analyses and final interpretation of results will be incorporated in a Master's thesis which will be completed during the fall 2019 academic semester. See "Final Report Summary" under part V (Dissemination) below for details on our planned approach for discussing management implications with agricultural and natural resource professionals.

V. DISSEMINATION:

Description: The results of this study will be reported in the annual MN DNR Summaries of Wildlife Research Findings publication, in a Master's thesis, in peer-reviewed scientific journal(s), and in presentations at professional conferences. The results will also be shared with MN DNR personnel (especially area wildlife managers and prairie habitat team members), University of Minnesota (UM) Cooperative Fish & Wildlife Research Unit, other government agencies [e.g., U.S. Geological Survey (USGS), MDA, U.S. Fish and Wildlife Service (USFWS), U.S. Department of Agriculture/Natural Resources Conservation Service (USDA/NRCS), U.S. Environmental Protection Agency (EPA)], and other partner groups [e.g., Minnesota Zoo, The Xerces Society, Pheasants Forever (PF), The Nature Conservancy (TNC)] via summary reports and direct consultation. We will work with MN DNR's Office of Communications and Outreach to publicize the progress and findings of the research. Finally, we will also work with partners to help inform the public about additional best management practices (BMPs; e.g., biocontrol) that can be used to help control crop pests.

Status as of January 1, 2017: We have presented information about the study in several internal MN DNR meetings and at a research symposium with partner groups who also have ongoing LCCMR projects. We have also submitted a poster abstract to introduce our study at an upcoming professional society meeting. See details below.

Status as of June 30, 2017: We have continued to share information about our project with other biologists who are interested in the topic of pesticide drift and/or are currently conducting related research. We have prepared a research summary for inclusion in the next publication of the MN DNR Summaries of Wildlife Research Findings and an annual report for the U.S. Geological Survey's (USGS) Minnesota Cooperative Fish & Wildlife Research Unit. Finally, Katelin presented two posters at professional society meetings. No formal media attention has been given to the project yet but we have mentioned the project to several reporters during other media interviews related to upland bird populations and habitat concerns. See details below.

Status as of January 1, 2018: Our project was mentioned in an article in the Star Tribune in October 2017. Additionally, we have provided further updates and/or overviews of the project during presentations to DNR wildlife staff, a USGS Minnesota Cooperative Fish & Wildlife Unit Cooperators Meeting, and a Minnesota Prairie Plan webinar. Katelin submitted two poster abstracts for presentation at professional society meetings. One abstract has been accepted for a lightning talk at the 2018 Midwest Fish & Wildlife Conference which will be held in late January 2018. The second abstract was accepted for a poster presentation at the 2018 Annual Meeting of the Minnesota Chapter of The Wildlife Society in February 2018. See details below.

Status as of June 30, 2018: Katelin presented a lightning talk at the 2018 Midwest Fish & Wildlife Conference in Milwaukee, WI and a poster presentation at the 2018 Minnesota Chapter of The Wildlife Society Meeting in St. Cloud, MN. Katelin also presented a project update at the 2018 UM/LCCMR Pollinator Project Meeting. Nicole provided a brief project update to wildlife managers at two DNR Regional Wildlife meetings (Region 3 and Region 4). Two agency project reports have been updated and submitted. Katelin submitted a research-in-progress poster abstract for the upcoming national meeting of The Wildlife Society which has recently been accepted. See details below.

Status as of January 1, 2019: Nicole presented a brief project update, including preliminary analyses of the 2017 data for direct and indirect exposure risk, at the DNR Region 1 Wildlife Meeting in September. Katelin presented a poster at the 2018 The Wildlife Society meeting in Cleveland, OH in October which also included preliminary analyses of the 2017 data for direct and indirect exposure risk. See details below.

Final Report Summary: To date, we have presented our preliminary research results via one oral presentation and five poster presentations at professional conferences (one annual meeting of The Wildlife Society, three annual meetings of The Minnesota Chapter of The Wildlife Society, and two annual Midwest Fish and Wildlife Conferences). We have also given oral presentations at six DNR regional wildlife meetings (covering Regions 1, 3, and 4), two LCCMR/UM Pollinator and Partner Projects meetings, two UM Natural Resources Association of Graduate Students research symposia, and one DNR/partner webinar focused on topics related to Minnesota's Prairie Plan. We have provided annual progress reports in two different agency publications (i.e., DNR Summaries of Wildlife Research Findings, USGS/Minnesota Cooperative Fish and Wildlife Research Unit annual reports). When appropriate, we have also mentioned the study during DNR media interviews. See below for details.

During fall 2019, Katelin will prepare her thesis and submit it as part of her Master's degree requirements for graduation. Concomitantly during fall 2019, we will begin sharing our findings more broadly 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., UM 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 and factors influencing 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 between the agricultural and natural resource sectors. 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 and the factors that influence them will help us improve management of public lands and better design private lands conservation programs to aid grassland wildlife conservation.

Over the next 4-8 months, we will also prepare at least two manuscripts for submission to peer-reviewed, scientific journals. By summer 2020, we will also submit final reports to DNR and the USGS describing our findings.

Publications

Publications	Title	Authors
2016 Summary of Wildlife Research Findings, Division of Fish & Wildlife Minnesota Department of Natural Resources, St. Paul, Minnesota	Evaluating insecticide exposure risk for grassland wildlife on public lands	Nicole Davros, Katelin Goebel, & David Andersen
2016 Annual Report, Minnesota Cooperative Fish & Wildlife Research Unit, U.S. Geological Survey	Insecticide exposure risk for grassland wildlife on public lands	Nicole Davros, Katelin Goebel, & David Andersen
2017 Annual Report, Minnesota Cooperative Fish & Wildlife Research Unit, U.S. Geological Survey	Insecticide exposure risk for grassland wildlife on public lands	Nicole Davros, Katelin Goebel, & David Andersen
2017 Summary of Wildlife Research Findings, Division of Fish & Wildlife Minnesota Department of Natural Resources, St. Paul, Minnesota	Evaluating insecticide exposure risk for grassland wildlife on public lands	Nicole Davros, Katelin Goebel, & David Andersen

<u>Publications</u>	<u>Title</u>	<u>Authors</u>
2018 Summary of Wildlife Research Findings, Division of Fish & Wildlife Minnesota Department of Natural Resources, St. Paul, Minnesota * *In review*	Evaluating grassland wildlife exposure to soybean aphid insecticides on public lands in Minnesota	Nicole Davros & Katelin Goebel

Presentations

Presentations (Event, Location, & Date)	***Topic** (Oral talk unless otherwise noted)*	***Lead Presenter***
MN DNR Region 4 Wildlife Meeting, New Ulm, MN – July 2016	Overview of grassland wildlife/ insecticide exposure study	Nicole Davros
LCCMR Pollinator & Partner Projects Meeting, UM – St. Paul Campus – Dec. 2016	Introduction of grassland wildlife insecticide exposure study	Nicole Davros
MN DNR Region 4 Wildlife Meeting, Lamberton, MN – Dec. 2016	Update on grassland wildlife/ insecticide exposure study	Nicole Davros
Annual Meeting of the MN Chapter of The Wildlife Society – Feb. 2017	Poster: Insecticide exposure risk for grassland wildlife on public lands	Katelin Goebel
Midwest Fish & Wildlife Conference Lincoln, NE – Feb. 2017	Poster: Insecticide exposure risk for grassland wildlife on public lands	Katelin Goebel
Little Lunch on the Prairie Webinar WebEx Meeting – Dec. 2017**	Does diversity matter? Ring-necked pheasant nest site selection & nest survival in grassland reconstructions	Nicole Davros
MN DNR Region 4 Wildlife Meeting, Lamberton, MN – Jan. 2018	Update on grassland wildlife/ insecticide exposure study	Nicole Davros
Midwest Fish & Wildlife Conference, Milwaukee, WI – Jan. 2018 **Lightning Talk Session*	Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota	Katelin Goebel
Annual Meeting of the MN Chapter of The Wildlife Society – Feb. 2018	Poster: Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota	Katelin Goebel
LCCMR Pollinator & Partner Projects Meeting, UM – St. Paul Campus – March 2018	Introduction of grassland wildlife/ insecticide exposure study	Katelin Goebel
MN DNR Region 3 Wildlife Meeting, Zimmerman, MN – April 2018	Update on grassland wildlife/ insecticide exposure study	Nicole Davros
UM Natural Resources Association of Graduate Students Research Symposium – April 2018 **Won an Oral Presentation Award*	Insecticide exposure risk for grassland wildlife on public land in southwest Minnesota	Katelin Goebel
MN DNR Region 1 Wildlife Meeting, Thief River Falls, MN – September 2018	Update on grassland wildlife/ insecticide exposure study	Nicole Davros

Presentations (Event, Location, & Date)	***Topic** (Oral talk unless otherwise noted)*	***Lead Presenter***
The Wildlife Society Conference Cleveland, OH – Oct. 2018	Poster: Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota	Katelin Goebel
Annual Meeting of the MN Chapter of The Wildlife Society – Feb. 2019	Poster: Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota	Katelin Goebel
UM Natural Resources Association of Graduate Students Research Symposium – April 2019	Grassland wildlife exposure to insecticides on public land in	Katelin Goebel
MN DNR Region 3 Wildlife Meeting, Zimmerman, MN – April 2019	Poster: Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota	Katelin Goebel
MN DNR Region 1 Wildlife Meeting, Thief River Falls, MN – September 2019	Poster: Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota	Katelin Goebel

**pending as of August 2019*
***Webinar can be viewed online at:*
https://www.youtube.com/watch?v=kidTWvK0a30&index=9&list=PLeh-ajY3F3JK8MgVek1eeWwtKibPLgzdc&t=2647s

Media Interviews

- Star Tribune, reporter Tony Kennedy interviewed Nicole Davros, published on Oct. 11, 2017 (http://www.startribune.com/dnr-wildlife-researcher-nicole-davros-working-to-help-upland-birds-thrive/450349283/)
- Outdoor News, reporter Rob Drieslein, interviewed Nicole Davros on June 22, 2018

VI. PROJECT BUDGET SUMMARY:
A. ENRTF Budget Overview:

Budget Category	$ Amount	Overview Explanation
Personnel:	$9,681	1 DNR technician for 3 months to support data data entry/proofing duties and processing of invertebrate samples in the lab
Professional/Technical/Service Contracts:	$220,490	1 graduate student ($114,010) recruited through University of Minnesota – Twin Cities (Dr. David Andersen, MN Cooperative Fish & Wildlife Research Unit) on a 50% research assistantship for 3.5 years to lead fieldwork, lab work, and analysis of data; funds will also cover part-time work study or other undergraduate student research assistants and a temporary casual appointment/trained taxonomist to identify invertebrate samples Lab analysis ($106,480) – U.S. Department of Agriculture/Agricultural Marketing Services – National Science Lab in Gastonia, NC (hereafter, USDA/AMS – NSL)

Budget Category	$ Amount	Overview Explanation
		to complete chemical analysis of samples
Equipment/Tools/Supplies:	$639	Miscellaneous sampling equipment & supplies
Travel Expenses in MN:	$6,939	Fleet & mileage, lodging, and meals
Other:	$2,347	Direct & Necessary Costs[1] ($2,347) – services to support this appropriation
TOTAL ENRTF BUDGET:	**$240,096**	

[1]Department Support Services. MN DNR's Direct & Necessary costs pay for activities that are directly related to and necessary for accomplishing appropriated programs/projects. In addition to itemized costs captured in our proposal budget, direct and necessary costs cover Financial Support ($138) that is necessary to accomplishing our funded project. Department Support Services are described in the agency Service level Agreement, and billed internally to divisions based on rates that have been developed for each area of service. These services are directly related to and necessary for the appropriation. Department leadership services (Commissioner's Office and Regional Directors) are not assessed. Those elements of individual projects that put little or no demand on support services (e.g., large single-source contracts, large land acquisitions, and funds that are passed through to other entities) are not assessed Direct & Necessary costs for those activities.

Explanation of Use of Classified Staff:
Funds will not be used to pay for classified staff.

Explanation of Capital Expenditures Greater Than $5,000:
N/A

Number of Full-time Equivalents (FTE) Directly Funded with this ENRTF Appropriation:
N/A

Number of Full-time Equivalents (FTE) Estimated to Be Funded through Contracts with this ENRTF Appropriation:
1.0 FTE

B. Other Funds:
The MN DNR Section of Wildlife provided funding from the State Game and Fish (G&F) Fund and the Pheasant Habitat Improvement Program (PHIP) Fund to directly support this research project for additional expenses (graduate student stipend, UM work study students, UM temporary casual appointment for insect identifications, travel, project supplies, and additional field technicians) that were incurred from spring 2016 through FY19. Additionally, multiple employees from the MN DNR Section of Wildlife, Farmland Wildlife Populations and Research Group (FaWPRG) devoted effort to the project throughout its 36-month duration: Nicole Davros at approximately 20% effort, FaWPRG clericals, and multiple FaWPRG seasonal field technicians.

Source of Funds	$ Amount Proposed	$ Amount Spent	Use of Other Funds
State			
MN DNR Section of Wildlife (G&F Fund & PHIP Fund)	$5,180	$3,521	Travel to project-related meetings, travel to select and sample study sites, meals for project staff and graduate student while traveling
MN DNR Section of Wildlife (G&F Fund & PHIP Fund)	$11,500	$13,116	Supplies (field and lab sampling equipment, GPS units, safety & first aid equipment, etc.)

Source of Funds	$ Amount Proposed	$ Amount Spent	Use of Other Funds
MN DNR Section of Wildlife, Farmland Populations and Research Group	$79,190	$81,207	Multiple employees (36 months, 1 FTE @ 20% effort, 3 FTE @ 5% effort, 3 full-time, temporary technicians @ 100% effort) – project management, field work, data management & analyses, reporting
MN DNR Section of Wildlife (PHIP Fund)	$33,720	$15,990	Contract with UM to support unmet costs associated with the graduate student stipend, undergraduate work study students, and a temporary casual appointment for the insect identifications
TOTAL OTHER FUNDS:	**$129,590**	**$113,834**	

VII. PROJECT STRATEGY:

A. Project Partners:

Dr. Nicole Davros, MN DNR, project manager
Dr. David Andersen, UM Cooperative Fish & Wildlife Research Unit, co-investigator & graduate student advisor
Dr. Pamela Rice, USDA Agricultural Research Service and UM Department of Soil, Water, & Climate, co-investigator and graduate student committee member
Dr. Theresa Kissane Johnston, Loyola University Chicago, Institute of Environmental Sustainability, co-investigator

Additional project partners (e.g., MDA, USDA/NRCS) will be included as we begin implementing this research project.

No project partners other than the University of Minnesota (through which the graduate student is being recruited) will be receiving funds. The university will receive $151,424 to support the graduate student and hire additional staff (including undergraduate work study students, a temporary casual appointment, and a seasonal field technician) as follows: $116,678 via the LCCMR/ENRTF grant and $34,745.85 via MN DNR Section of Wildlife funding.

B. Project Impact and Long-term Strategy:

Concerns have previously been raised about the impacts of chlorpyrifos and other agricultural insecticides on water quality and human health, prompting the MDA to release guidelines for voluntary BMPs for their use. Our research will address additional mounting concerns about the impacts of these insecticides on wildlife in Minnesota's farmland regions by determining exposure risk of grassland wildlife to commonly-used soybean aphid insecticides under typical field application conditions. Our research will allow us to make recommendations to land managers and private landowners alike on how to better design grassland habitats surrounded by an agricultural matrix to reduce the impacts of spray drift on upland wildlife, including birds and beneficial insects. Additionally, results from our study will assist in improving riparian buffer designs to better protect waterways, their associated wildlife, and humans who may recreate in or consume water from these water bodies. We will also work with partners to help inform the public about additional BMPs that can be used to control crop pests, thereby potentially reducing our reliance on pesticides.

C. Funding History:

No portions of this project have been previously funding by the Environment and Natural Resources Trust Fund (ENRTF).

VIII. FEE TITLE ACQUISITION/CONSERVATION EASEMENT/RESTORATION REQUIREMENTS:

A. Parcel List:
N/A

B. Acquisition/Restoration Information:
N/A

IX. VISUAL COMPONENT or MAP(S):
Please see attached map (Fig. 1) and graphic (Fig. 2).

X. RESEARCH ADDENDUM:
Please see attached Research Addendum.

XI. REPORTING REQUIREMENTS:
Periodic work plan status update reports will be submitted no later than January 1, 2017; June 30, 2017; January 1, 2018; June 30, 2018, and January 1, 2019. A final report and associated products will be submitted between June 30 and August 15, 2019.

Table 1. 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 summer 2017 and summer 2018 in Minnesota's farmland regions.

Site ID[a]	Region[b]	County	Site type[c]	Year sampled	Range of dates when field sampling occurred
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

[a] WMA names are not provided to protect private landowner cooperators.
[b] Regions 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 reports.
[c] Treatment sites had adjacent soybean fields that were sprayed for aphids; control sites had adjacent corn fields that were not sprayed for aphids.
[d] Includes first day of pre-spray sampling through last day of post-spray sampling for data collection activities.

Table 2. 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 July - 14 September 2017 and 18 July - 5 September 2018 in Minnesota's farmland regions.

Site ID	Spray method	Insecticide trade name	Insecticide active ingredients	Insecticide application rate (L/ha)	Sprayer application rate (L/ha)	Application speed (m/s)	Boom height (m)	Tank pressure (kPa)
tA	Ground	Endigo	lambda-cyhalothrin + thiamethoxam	0.26	140.3	4	0.2-0.3	275.8
tB	Airplane	Bolton	chlorpyrifos + gamma-cyhalothrin	0.88	18.7	67.9	1.5	275.8
tC	Ground	Lorsban 4E	chlorpyrifos	NA[b]	93.5	NA	NA	137.9-206.8
tD	Airplane	Lorsban Advanced	chlorpyrifos	1.17	18.7	55.9	2.7-4.0	275.8
tE[c]	Airplane	Lorsban Advanced; Warrior II	chlorpyrifos; lambda-cyhalothrin	0.44; 0.22	NA	NA	NA	NA

[a] WMA names are not provided to protect private landowner cooperators.

[b] Data is not available because cooperator declined to provide this information.

[c] This cooperating landowner combined two different trade name insecticides during the spraying event.

Table 3. Mean (± 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 regions. Target chemicals were chlorpyrifos, lambda-cyhalothrin, and bifenthrin. Values reported in parts per billion (ppb).

	Distance from soybean field edge (m)						
Site type[a]	0 m	5 m	25 m	50 m	100 m	200 m	400 m
Treatment[b]	35,322 ± 145,015	16,260 ± 64,298	26,712 ± 92,827	385 ± 906	40 ± 68	14 ± 20	699 ± 3,508
Airplane	57,198 ± 185,976	27,080 ± 82,113	44,504 ± 117,734	629 ± 1,115	50 ± 84	7 ± 9	8 ± 8
Ground	2,510 ± 5,538	30 ± 30	25 ± 27	19 ± 21	24 ± 30	23 ± 26	2,254 ± 6,322
Control	41 ± 76	21 ± 20	21 ± 19	21 ± 20	22 ± 23	19 ± 18	30 ± 30

[a] Treatment sites had adjacent soybean fields that were sprayed for aphids; control sites had adjacent corn fields that were not sprayed for aphids.
[b] Cooperating landowners at treatment sites sprayed for aphids using either airplane or ground booms.

Table 4. Number of parameters (*K*), difference from Akaike's Information Criterion (calculated for small sample sizes) of the best-supported model (ΔAIC_c), conditional R^2 value (variation explained by the entire model, including random effects), and deviance (*d*) for models explaining chemical deposition onto passive sampling devices (PSD) in Wildlife Management Areas (WMAs) in the farmland region of Minnesota during July-September, 2017 and 2018. The PSDs were used to assess direct exposure of wildlife to drift from target chemicals (i.e., chlorpyrifos, lambda-cyhalothrin, and bifenthrin) sprayed to control soybean aphids. We used a hierachical model selection approach in which our first set of models assessed weather conditions during the spraying event. Our best supported weather model was used as a base model to assess WMA vegetation covariates. The best weather + vegetation model was then used to assess our key factors of interest which included distance from grassland/soybean edge to the WMA interior (edge distance), position in the canopy layer (ground level or mid-canopy) height), and spray application method (airplane or ground boom). The column ΔAICc compares models within each step of model development; the ΔAIC^i compares models to the best-supported model of the previous step; negative values indicate a decrease in AIC_c. All models included site as a random effect.

Model[a]	*K*	ΔAIC_c	ΔAIC^i	R^2	*d*
Weather					
wind direction + temperature	5	0.00		0.10	5161.96
temperature	4	0.85		0.08	5164.91
wind direction + wind speed + temperature	6	2.12		0.10	5161.95
wind speed + temperature	5	2.86		0.08	5164.82
wind direction	4	7.67		0.07	5171.73
wind direction + wind speed	5	9.77		0.07	5171.73
wind speed	4	10.08		0.07	5174.14
Weather and Vegetation					
Weather[b] + % cc live	6	0.00	-1.90	0.11	5157.94
Weather + % cc live + mhl	7	1.31	-0.59	0.12	5157.10
Weather + mhl	6	1.73	-0.17	0.11	5159.67
Weather + % cc live + density	7	2.13	0.23	0.12	5157.92
Weather + % cc live + mhl + density	8	3.02	1.12	0.12	5156.65
Weather + density	6	3.69	1.79	0.10	5161.63
Weather + mhl + density	7	3.78	1.88	0.11	5159.57
Weather, Vegetation, and Key Factors of Interest					
Veg[c] + edge distance + canopy layer	8	0.00	-1.68	0.14	5151.96
Veg + edge distance	7	0.62	-1.06	0.13	5154.74
Veg + canopy layer	7	1.07	-0.61	0.13	5155.19
Veg + edge distance + canopy layer + spray method	9	1.89	0.21	0.14	5151.66
Veg + edge distance + spray method	8	2.50	0.82	0.13	5154.45
Veg + canopy layer + spray method	8	2.99	1.31	0.13	5154.94
Veg + spray method	7	3.57	1.89	0.12	5157.69

[a] Weather covariates were estimated within each WMA during the spraying event using a portable weather meter and included: temperature = mean temperature; wind direction = WMA was either upwind or down wind of the predominant wind direction; wind speed = mean wind speed. Vegetation metrics estimated within each WMA included: % cc live = percent canopy cover of live vegetation (grasses, forbs, woody stems); mhl = maximum height of live vegetation; density = vertical density of the vegetation as estimated by a visual obstruction reading from 4 m away at a height of 1 m.

[b] Weather = covariates in the top-ranked Weather model (wind direction + temperature).

[c] Veg = covariates in the top-ranked Weather and Vegetation model (wind direction + temperature + % cc live).

Table 5. Number of parameters (*K*), difference from Akaike's Information Criterion (calculated for small sample sizes) of the best-supported model (ΔAIC_c), conditional R^2 value (variation explained by the entire model, including random effects), and deviance (*d*) for models explaining chemical deposition on invertebrate samples collected from Wildlife Management Areas (WMAs) in the farmland region of Minnesota during July-September, 2017 and 2018. The invertebrates were used to assess potential for indirect exposure of wildlife to drift from target chemicals (i.e., chlorpyrifos, lambda-cyhalothrin, and bifenthrin) sprayed to control soybean aphids. We used a hierarchical model selection approach in which our first set of models assessed weather conditions during the spraying event. Our best supported weather model was used as a base model to assess WMA vegetation covariates. The best weather + vegetation model was

then used to assess our key factors of interest which included distance from grassland/soybean edge to the WMA interior (edge distance) and spray application method (airplane or ground boom). The column ΔAICc compares models within each step of model development; the ΔAIC^i compares models to the best-supported model of the previous step; negative values indicate a decrease in AICc. All models included site as a random effect.

Model[a]	*K*	ΔAIC_c	ΔAIC^i	R^2	*d*
Weather					
temperature	4	0.00		0.25	877.60
wind direction + temperature	5	0.25		0.28	875.31
wind speed + temperature	5	2.47		0.25	877.53
wind direction + wind speed + temperature	6	2.91		0.28	875.30
wind direction	4	6.22		0.19	883.82
wind speed	4	8.57		0.19	886.17
wind direction + wind speed	5	8.76		0.19	883.82
Weather and Vegetation					
Weather[b] + mhl	5	0.00	-1.38	0.31	873.68
Weather + mhl + density	6	0.18	-1.20	0.35	871.19
Weather + % cc live + mhl	6	2.33	0.95	0.32	873.33
Weather + % cc live + mhl + density	7	2.97	1.59	0.35	871.16
Weather + % cc live	5	3.88	2.50	0.25	877.56
Weather + density	5	3.90	2.52	0.25	877.59
Weather + % cc live + density	6	6.52	5.14	0.25	877.53
Weather, Vegetation, and Key Factors of Interest					
Veg[c]	5	0.00		0.31	873.68
Veg + edge distance	6	1.25	1.25	0.33	872.26
Veg + spray method	6	2.32	2.32	0.32	873.33
Veg + edge distance + spray method	7	3.58	3.58	0.34	871.77

[a] Weather covariates were estimated within each WMA during the spraying event using a portable weather meter and included: temperature = mean temperature; wind direction = WMA was either upwind or down wind of the predominant wind direction; wind speed = mean wind speed. Vegetation metrics estimated within each WMA included: % cc live = percent canopy cover of live vegetation (grasses, forbs, woody stems); mhl = maximum height of live vegetation; density = vertical density of the vegetation as measured by a visual obstruction reading from 4 m away at a height of 1 m. [b] Weather = covariates in the top-ranked Weather model (temperature). [c] Veg = covariates in the top-ranked Weather and Vegetation model (temperature + mhl).

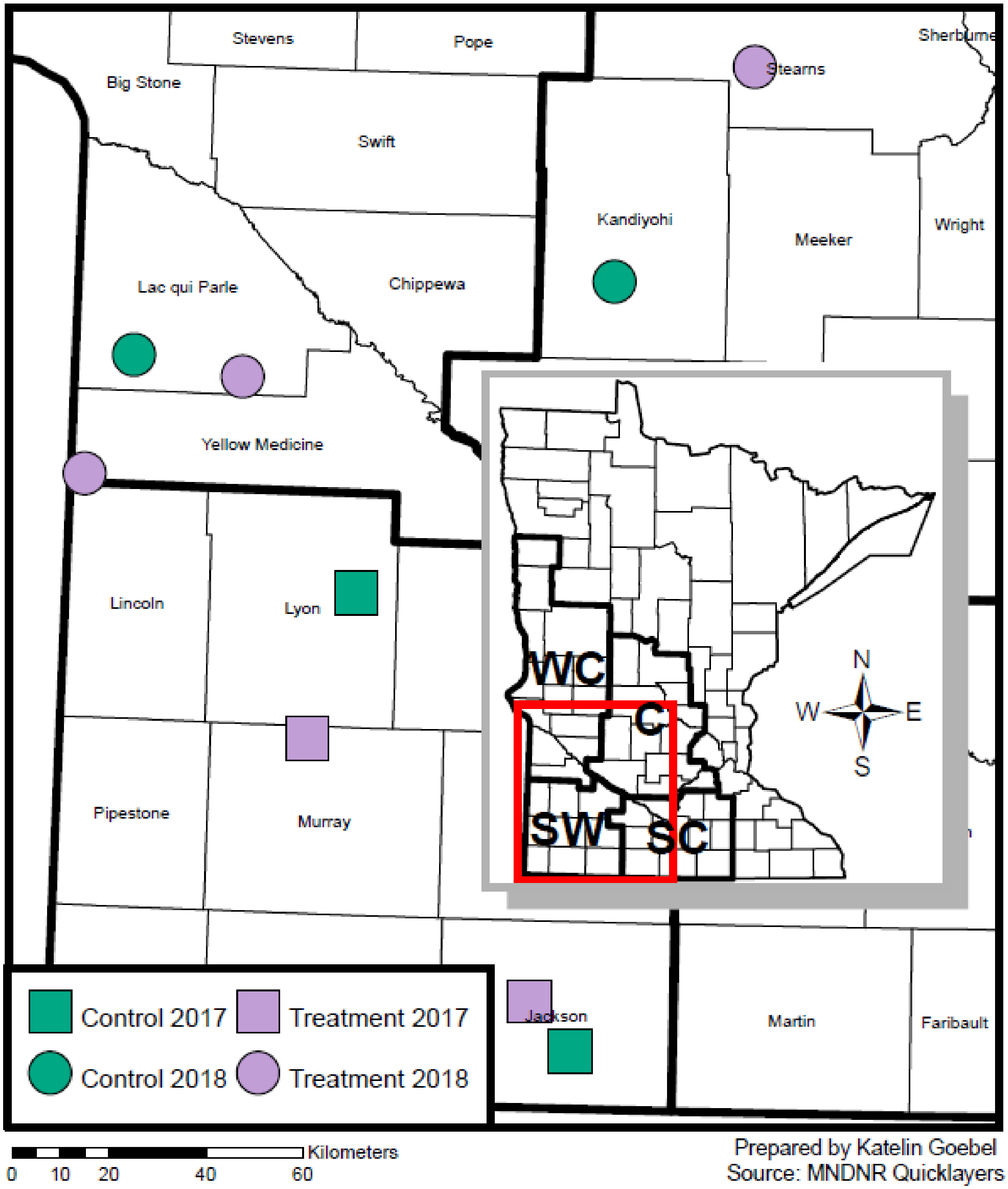

Figure 1. Location of treatment (purple symbols) and control (green symbols) sites during 2017 (square symbols) and 2018 (round symbols) field sampling efforts, July-September each year. Treatment sites were Wildlife Management Areas (WMA) adjacent to soybean fields sprayed for aphids; control sites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

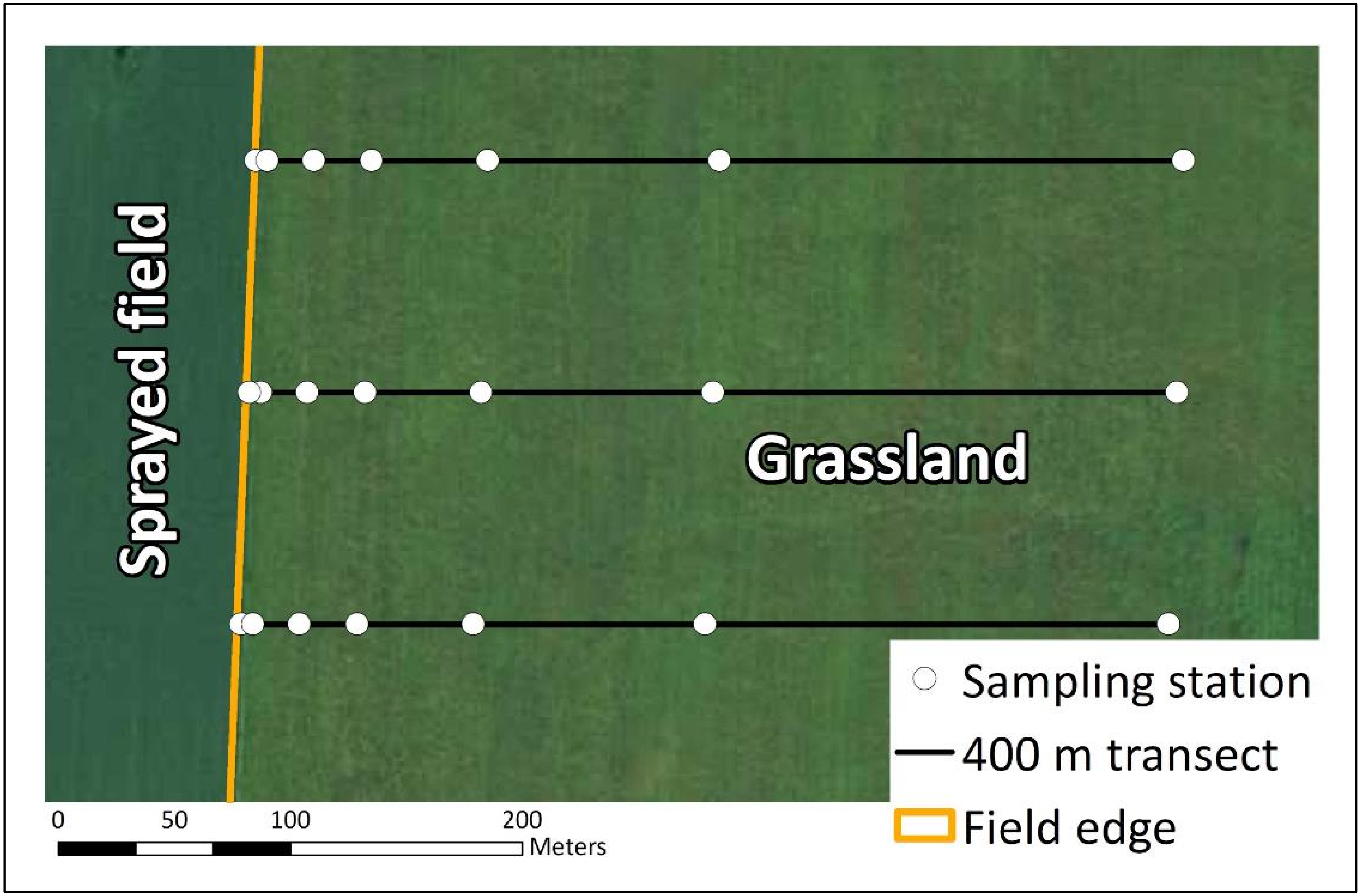

Figure 2. Field sampling design used to assess the exposure of grassland wildlife to soybean aphid insecticides, especially chlorpyrifos, commonly used in Minnesota's farmland regions. Sampling occurred on MN DNR-owned grasslands adjacent to privately-owned soybean fields sprayed for aphid infestations. Black lines indicate sampling transects established perpendicular to the soybean field edge and extending 400 m into the grassland; white circles represent distances (0 m, 5 m, 25 m, 50 m, 100 m, 200 m, and 400 m) at which sampling occurred along each transect.

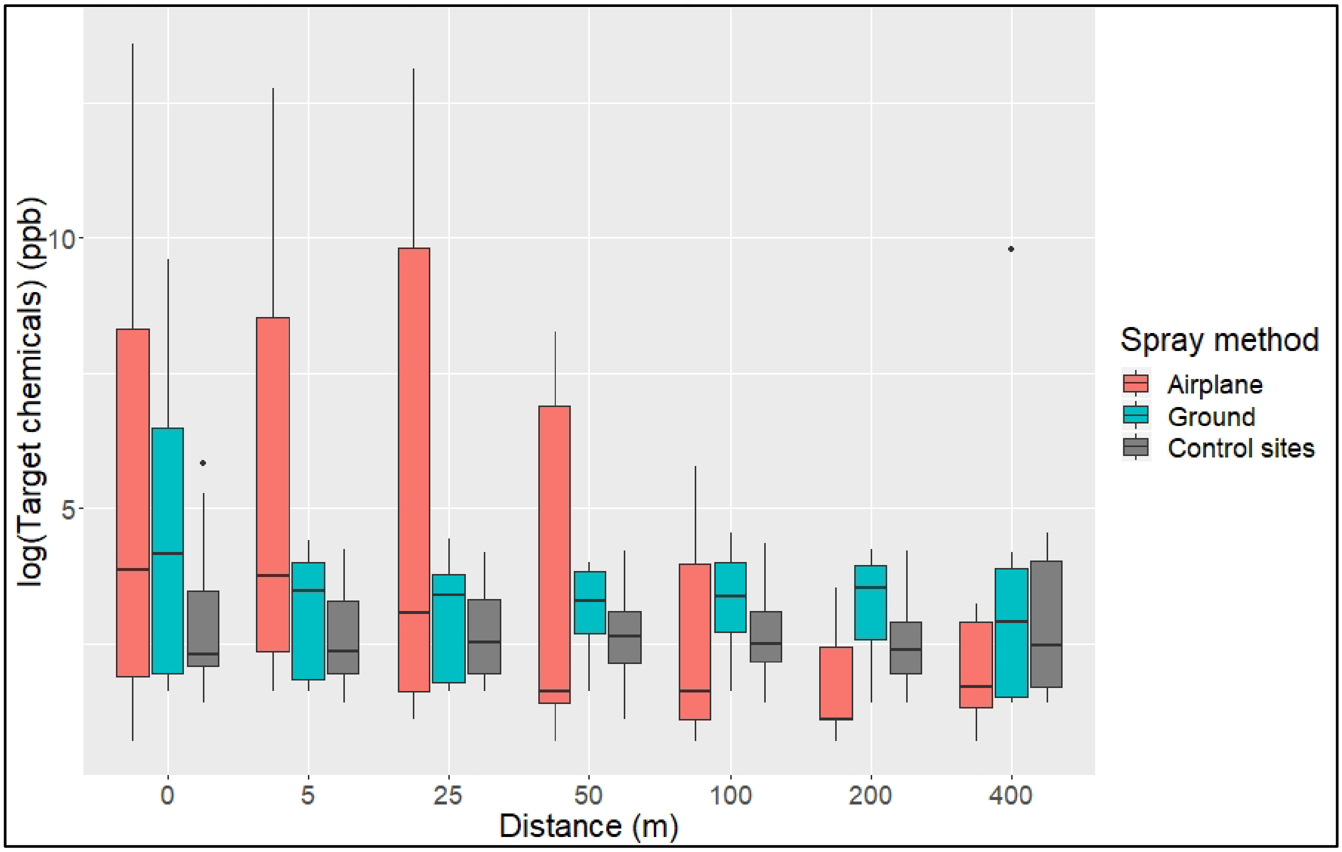

Figure 3. Box plot summaries of target chemical deposition on passive sampling devices (PSDs; *n* = 398) by distance from field edge to grassland interior for treatment sites sprayed by airplane (orange) or ground boom (blue) and control sites (gray), July-September 2017 and 2018 in Minnesota's farmland regions. The PSDs were used to quantify the potential for grassland wildlife to be exposed to chlorpyrifos, lambda-cyhalothrin, and bifenthrin directly through spray drift (Activity 1a). 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.

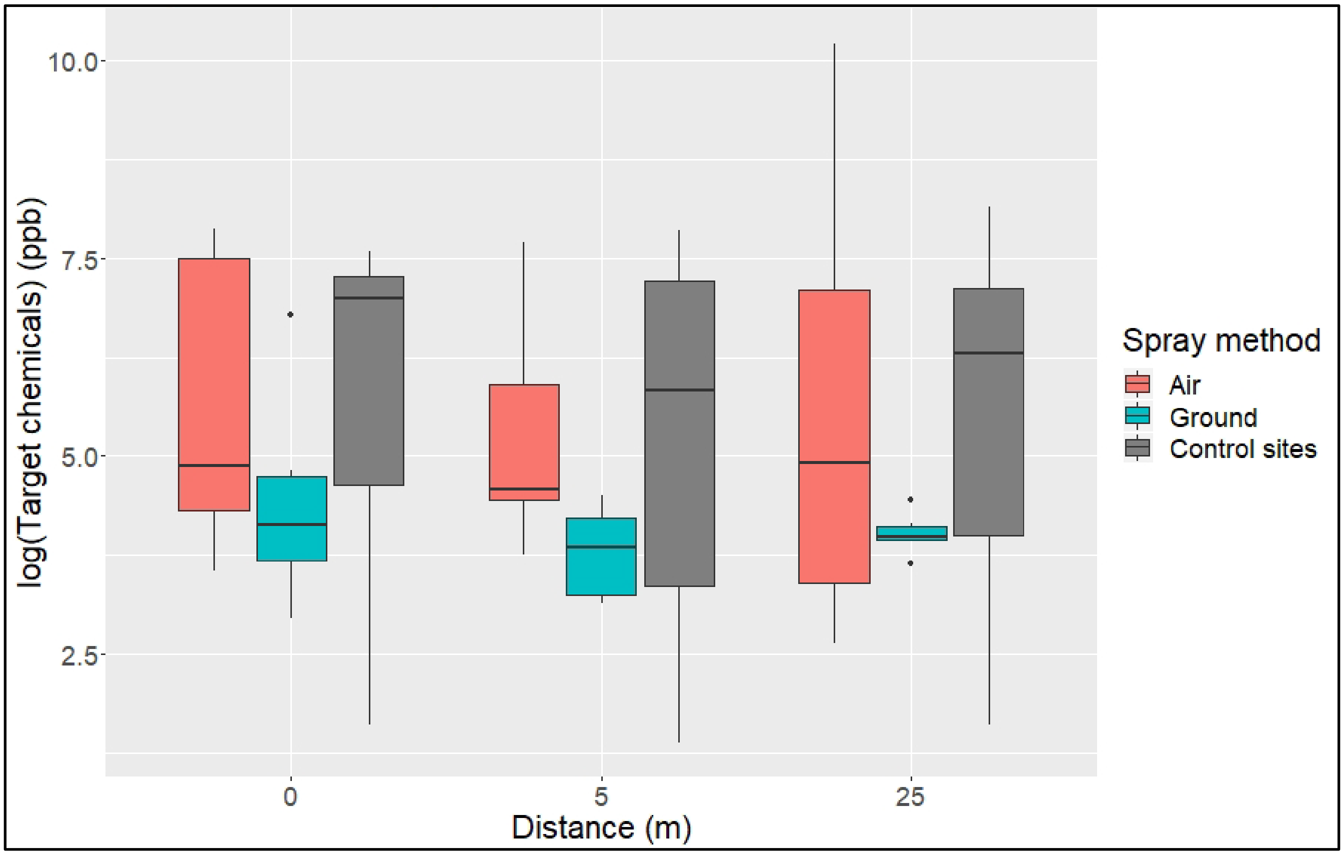

Figure 4. Box plot summaries of target chemical deposition on invertebrates (*n* = 81) by distance from field edge for treatment sites sprayed by airplane (orange) or ground boom (blue) and control sites (gray), July-September 2017 and 2018 in Minnesota's farmland regions. The invertebrates were used to quantify the potential for grassland wildlife to be exposed to chlorpyrifos, lambda-cyhalothrin, and bifenthrin indirectly through consumption of invertebrate prey items (Activity 1b). 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.

Environment and Natural Resources Trust Fund M.L. 2016 Project Budget

Project Title: Evaluating Insecticide Exposure Risk for Grassland Wildlife on Public Lands
Legal Citation: M.L. 2016, Chp. 186, Sec. 2, Subd. 03n
Project Manager: Nicole M. Davros
Organization: Minnesota Department of Natural Resources, Division of Fish & Wildlife, Section of Wildlife
M.L. 2016 ENRTF Appropriation: $250,000
Project Length and Completion Date: 3 years; June 30, 2019
Date of Report: August 15, 2019 (FINAL)

ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Activity 1 Budget 2/1/2019	Amount Spent	Activity 1 Balance	TOTAL BUDGET	TOTAL BALANCE
BUDGET ITEM					
Personnel (1/1/2019)					
1 DNR technician for 3 months (688 hours at $24.50/hour including fringe) to support processing of invertebrate samples in the lab (sorting, measuring, counting) and multiple data entry/proofing duties related to the entire project.	$16,856	$9,681	$7,175	$16,856	$7,175
Professional/Technical/Service Contracts					
University of Minnesota - Twin Cities (single-source contract): 1 graduate student; research assistantship @ 0.5 FTE for 3.5 years ($40,000/yr); 75% salary, 25% benefits; recruited in collaboration with Dr. David Andersen, Minnesota Cooperative Fish & Wildlife Research Unit; Note: remaining stipend support needs not covered by the ENRTF funds shown here will be covered by MN DNR funds.	$116,678	$114,010	$2,668	$116,678	$2,668
U.S. Department of Agriculture/Agricultural Marketing Services - National Sciences Lab (Gastonia, NC): Lab processing of samples using solvent extraction/evaporation process and GC/MS-NCI analysis method; 658 samples @ $220/sample	$106,480	$106,480	$0	$106,480	$0
Equipment/Tools/Supplies					
Miscellaneous sampling equipment & supplies (e.g., insect sample collection jars, tweezers, sorting trays, chemical protection body suits, gloves, etc.)	$639	$639	$0	$639	$0
Travel expenses in Minnesota					

Travel to and between study sites in south-central and southwest Minnesota by graduate student and MN DNR research staff. Fleet & mileage: $5,500; lodging: $1,000; meals: $500	$7,000	$6,939	$61	$7,000	$61
Other					
Direct and Necessary Costs: These expenses include Department Support Services (specifically, Financial Support @ $2,347) necessary to accomplish the funded project.	$2,347	$2,347	$0	$2,347	$0
COLUMN TOTAL	**$250,000**	**$240,096**	**$9,904**	**$250,000**	**$9,904**

EVALUATING INSECTICIDE EXPOSURE RISK FOR GRASSLAND WILDLIFE ON PUBLIC LANDS

Nicole M. Davros and Katelin Goebel[1]

SUMMARY OF FINDINGS

Increasing evidence suggests that acute toxicity to pesticides may be more important than agricultural intensity in explaining declines in grassland-dependent wildlife. Although neonicotinoids (systemic insecticides routinely used on corn and soybeans) are currently under scrutiny for their effects on birds and pollinators, other insecticides are commonly used in Minnesota's farmland regions that may also have negative effects on non-target organisms. 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's farmland regions. Concerns have previously 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, fewer studies have investigated the environmentally-relevant exposure risk of free-ranging wildlife to these chemicals. Our research project will assess the direct and indirect exposure risk of grassland wildlife to common soybean aphid insecticides along a gradient from soybean field edge to grassland interior. The data we obtain on the environmentally-relevant exposure risk of wildlife to these insecticides 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 habitat 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 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).

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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 (Vennette 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 boom sprayers or planes are common treatment methods when chemical control of aphids is considered necessary. The 2 most common insecticides used are chlorpyrifos and lambda-cyhalothrin (MDA 2005, MDA 2007, MDA 2009, MDA 2012, MDA 2014a) but bifenthrin use has also been reported (R. Riley, personal communication; E. Runquist, unpublished data). Withholding times vary by chemical (lambda-cyhalothrin: 45 d; chlorpyrifos: 28 d; bifenthrin: up to 14 d); thus, the timing of product use within the growing season should 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 the development of resistance (UM Extension 2014). 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.

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). Further, 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 bobwhite quail (*Colinus virginianus*) and mallards (*Anas platyrhynchos*)(LD_{50} 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 terrestrial insects are also susceptible to bifenthrin (Siegfried 1993). Bifenthrin is very highly toxic to bumblebees, with one study showing 100% mortality by contact (Besard et al. 2010).

Chlorpyrifos (common trade names include Dursban, Govern, Lorsban, Pilot, Warhawk, and Yuma) is a broad-spectrum organophosphate insecticide that also 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). Further, MDA recently 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 dose causing death in 50% of treated animals (LD_{50}) 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 (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 ecotoxological 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) has been documented in both lab and field studies (McEwen et al. 1986, Richards et al. 2000, Al-Badrany and Mohammad 2007, Moye 2008). Thus, sub-lethal effects leading to indirect mortality (e.g., via increased predation rates) may be a concern for wildlife exposed to chlorpyrifos.

Minnesota DNR wildlife managers and members of the public have reported concerns about the effects of these soybean aphid insecticides on non-target wildlife, including economically important bird and pollinator species. The common public perception 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 actual exposure risk 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 1600 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 is to assess the environmentally-relevant exposure risk of grassland wildlife to commonly-used soybean aphid insecticides, especially chlorpyrifos, in Minnesota's farmland region. In particular, we will:

1) Quantify the concentration of insecticides 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 chemicals directly via contact with spray drift and indirectly through insect prey items exposed to the insecticides.

2) Quantify and compare 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 insecticides on food availability for grassland nesting birds and other wildlife.

STUDY AREA

Our study is being conducted within the south-central and southwest regions of Minnesota's farmland zone (Figure 1). These regions are intensively farmed, and corn and soybeans combined account for approximately 75% of the landscape [U.S. Department of Agriculture

(USDA) 2013a, USDA 2013b]. Acres set aside as grassland habitat on public and private land account for 5.8% and 4.6% of the landscape, respectively, in these regions (Davros 2015). 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 will consist of a MNDNR Wildlife Management Area (WMA) immediately adjacent to and downwind from a soybean field that will be sprayed to control for aphids. We are working in close consultation with wildlife managers and private landowner cooperators to choose 6-8 treatment sites. We will choose 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 will also chose 2-4 control study sites with similar site characteristics except that control sites will not be sprayed with any chemical to control aphids.

Field sampling will occur during summer 2017 and 2018, and approximately half of the study sites will be sampled each year. Within each treatment site prior to spraying, we will establish 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 is large enough, we will also establish a station at a distance of 400 m along each transect. This design will give us a total of 18-21 stations per site. We will establish transects and stations the same way within control sites. At all sites, transects will run perpendicular to the edge of the soybean field and will be spaced 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 soybean aphid insecticides (hereafter, target chemicals), we will deploy passive sampling devices (PSDs) to absorb any chemical drift that occurs. The PSDs will be placed in treatment fields on the morning of but prior to spraying of soybeans. They will be made of Whatman™ Qualitative Filter Paper (grade 2) that is attached to 0.5 in^2 hardwire cloth formed to a cylinder shape to approximate the size and shape of a large songbird or a gamebird chick. We will place the PSDs at two heights (ground and mid-canopy) at each of the sampling stations. Ground-level sampling will help represent 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 will help represent above-ground nesting birds and many species of spiders and insects. We will retrieve the PSDs from the field ≤1 h after spraying and properly store them for later chemical analysis. All ground-level and mid-canopy samples will be analyzed independent of one another. At control sites, we will place PSDs at both ground and mid-canopy levels at each of the stations. We will leave the PSDs on site for the same amount of time as PSDs at treatment sites before we collect and store them for later analysis.

To assess the potential for birds and other insectivorous wildlife to be exposed to the target chemicals indirectly via consumption of prey items, we will sample invertebrates ≤2 h post-spraying at each of the sampling stations. We will sample ground-dwelling invertebrates using a vacuum trap and canopy dwelling invertebrates using a sweepnet. Vacuum trap and sweepnet samples will both be taken along 60 m transects to the left side of the sampling stations and parallel to the soybean field. We will combine vacuum trap and sweepnet samples taken from the same station during the same time period into one sample and properly store them for later chemical analysis. We will sample control sites using the same methods and timing, with the timing based on when we deploy the PSDs at these sites.

To quantify and compare the abundance, richness, diversity, and biomass of invertebrate prey items, we will collect vacuum trap and sweepnet samples from the <1-5 m, 25 m, and 100 m distances along the 3 transects at each site (total = 9 stations/site). The <1 m and 5 m distances will be combined into 1 transect parallel to the soybean field for this effort. We will collect these samples 1-3 d prior to spraying and between 3-5 d and 19-21 d post-spraying at treatment sites. Samples will be taken along 40 m transects but on the right side of the sampling stations and parallel to the soybean field. We will combine vacuum trap and sweepnet samples into 1 sample per station per sampling period and store them in ethanol for later sorting and identification. We will place emphasis on 3 invertebrate orders important in the diets of grassland nesting birds: Araneae (spiders), Orthoptera (grasshoppers, crickets, and katydids) and Coleoptera (beetles). All individuals from these orders will be sorted and identified to at least the family level for analysis. Quantifying the spider community will allow us to examine potential impacts on an additional trophic level since spiders are an important predator of insects.

We will use portable weather meters (Kestrel 5500AG Agricultural Weather Meters) 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 during insect sampling periods at each site.

At each site, we will also collect vegetation data 1-3 days prior to spraying at all sampling stations and again at 3-5 d and 19-21 d post-spraying at the reduced subset of sampling stations coinciding with invertebrate sampling efforts. Data collected will include percent canopy cover, maximum height of live and dead vegetation, litter depth, and vertical density. We are still developing our methods for vegetation data collection but we will likely use the program SamplePoint (Booth et al. 2006) to estimate percent canopy cover as it provides a more objective measure than visual estimation techniques.

We will send the PSD samples and invertebrate samples to an external lab to be analyzed using a solvent-based extraction method. Extracts will be concentrated by evaporation and then analyzed using a gas chromatography/mass spectrometry-negative chemical ionization (GC/MS-NCI) method. Although our experimental design will focus on soybean fields sprayed with foliar insecticides to control aphids, the chemical analyses will allow us to quantify additional pesticides (e.g., neonicotinoids, fungicides) at minimal extra cost. Obtaining information about other pesticide exposure will be valuable supplementary information in support of other Section of Wildlife research and management goals.

Data Analyses

We will use mixed regression models to examine factors related to risk of direct and indirect exposure of wildlife to target chemicals. Chemical concentration will be the dependent variable. We will specify distance from soybean field edge and canopy height (when relevant) as a fixed effect. We may also include other covariates such as site, ordinal date, vegetation data, and weather condition variables where appropriate. We will use similar models to examine differences in the abundance, richness, diversity, and biomass of Aranaeans, Orthopterans and Coleopterans. We will use the sampling period (i.e., 1-3 d prior to spraying, and 7-9 d or 18-20 d post-spraying) as a repeated measure in these analyses, specifying a covariance structure [e.g., autoregressive 1 (AR1)] when appropriate.

RESULTS AND DISCUSSION

To date, we have surveyed 12 farmer cooperatives in 12 counties to gather more specific information about chemical spraying (e.g., type of insecticide, application method) in southern

Minnesota. Congruent with MDA's pesticide usage reports (MDA 2007, MDA 2009, MDA 2012, MDA 2014), the coops reported that chlorpyrifos, lambda-cyhalothrin, and bifenthrin have been the most commonly-used foliar soybean insecticides in recent years. Additionally, we learned that neonicotinoids have also been used 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 treat plants systemically.

We also surveyed landowners adjacent to potential WMA study sites to learn more about their soybean aphid spraying practices and to ask for their cooperation with our study (see Appendix 1) since cooperation will be key to timing our field sampling. We mailed 221 letters during the first week of March 2017; 24 letters were returned as undeliverable. The overall response rate for the first mailing was 24.4%. In early April, we sent a second round of 164 letters and had a response rate of 6.1%. Some landowners opted to call us instead and provide their renter's contact information; however, not all landowners provided renter information when they returned the survey by mail. Overall, we were able to identify 11 landowners adjacent to and upwind from a WMA during 2017 who are willing to be cooperators with our study. We are currently contacting these landowners again to determine if they have planted soybeans this year and whether they will be spraying their soybeans for aphids this growing season. Several landowner cooperators have indicated that they do not plan on scouting for aphids. Rather, they plan to spray regardless of infestation levels. This approach to soybean management may be a primary reason why reports of aphid resistance to pyrethroid insecticides are increasing in Minnesota and parts of North Dakota this year (UM Extension 2017).

Further results are forthcoming as no field sampling has occurred yet. Our first year of field sampling will occur during late summer 2017 once soybean aphid spraying begins. A second season of field sampling is also planned for summer 2018.

ACKNOWLEDGMENTS

We would like to thank MNDNR wildlife managers, especially Cory Netland, Don Nelson, Randy Markl, Judy Markl, Wendy Krueger, Kent Schaap, and Bill Schuna, for discussions on the topic and for providing information on potential study sites. We are especially grateful to the landowners who have cooperated with us by returning our mail survey, calling us to provide information, and/or coordinated with us on the timing of spraying and field sampling. Tonya Klinkner contacted the cooperatives to ask about soybean spraying practices. Veronique St-Louis and John Giudice provided valuable feedback on initial study design, and David Andersen, Pamela Rice, Theresa Kissane Johnston, and Jessica Petersen have provided further feedback on study design and methodologies. This project has been funded by the Environment and Natural Resources Trust Fund (ENRTF) as recommended by the Legislative-Citizen Commission on Minnesota Resources (LCCMR). The MNDNR Section of Wildlife has also contributed funding through the Federal Aid in Wildlife Restoration Act. Finally, the Cooperative Fish and Wildlife Research Unit at the University of Minnesota has provided in-kind support.

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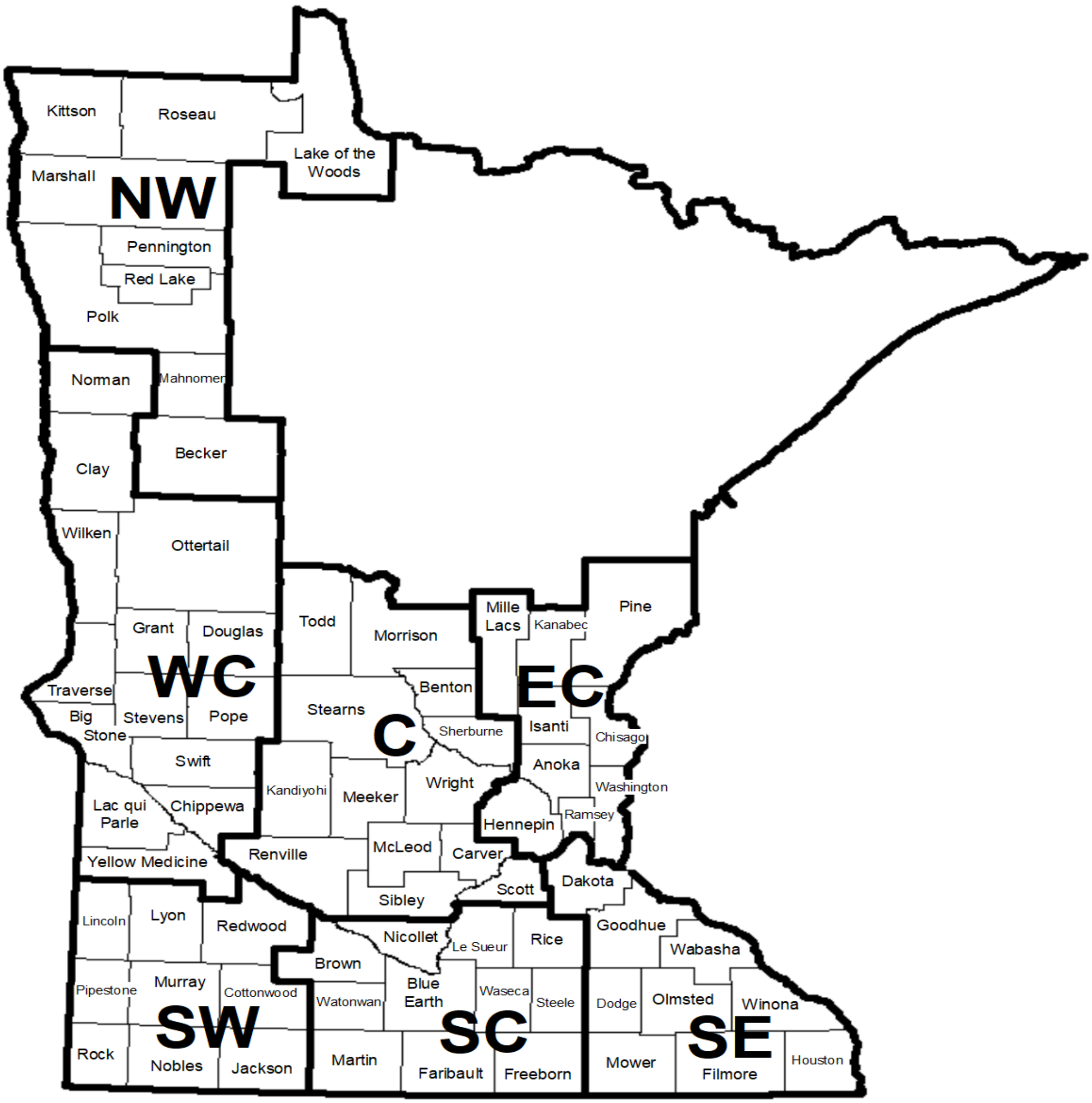

Figure 1. Minnesota's agricultural regions as outlined in MNDNR's annual August Roadside Surveys.

Appendix 1. Survey sent to neighboring landowners (i.e., private landowners with property immediately adjacent to potential Wildlife Management Area study sites) in March and April 2017 to assess soybean aphid spraying practices and to solicit cooperation for summer 2017 sampling efforts.

Print your name here ___________________

Spraying Practices Survey

PART I

1. **Have you planted soybeans on your land in the past 3-5 years?**
   ◦ Yes
   ◦ No → *please continue to Part II*

2. **Were your soybeans treated with foliar insecticides in the past 3-5 years?**
   ◦ Yes
   ◦ No → *please continue to Part II*

3. **On what date(s) were foliar insecticides applied on your soybeans?**

4. **How was the majority of foliar insecticides sprayed on your soybeans in the past 3-5 years?**
   ◦ Ground boom
   ◦ Aerial
   ◦ Other (please specify):

5. **Please list the foliar insecticide trade names and/or the application logistics used on your soybeans in the past 3-5 years to control aphids.**
   Example: "2016: Lorsban - 20 gpa through 8004 nozzles @ 50-60 psi from a 854 Rogator traveling at 6 mph to apply a 90' swath"

6. **Did you hire an applicator (e.g. agricultural consultant company) to treat your soybeans with foliar insecticides in the past 3-5 years?**
   ◦ Yes (please specify company or individual):
   ◦ No, I applied insecticides myself

PART II

1. **Will you be planting soybeans on your land that borders a Wildlife Management Area (WMA) or other protected grassland in 2017?**
   ◦ Yes
   ◦ No → *end of survey - thank you*
   ◦ I'm not sure

2. **Will you be treating these soybeans with foliar insecticides in 2017 if significant numbers of aphids occur?**
   ◦ Yes
   ◦ No → *end of survey - thank you*
   ◦ I'm not sure

3. **How will foliar insecticides likely be sprayed on these soybeans in 2017?**
   ◦ Ground boom
   ◦ Aerial
   ◦ Other (please specify):
   ◦ I'm not sure

4. **Please list the foliar insecticide trade names and/or the application logistics that will likely be used on these soybeans in 2017 to control aphids.**
   Example: "Lorsban - 20 gpa through 8004 nozzles @ 50-60 psi from a 854 Rogator traveling at 6 mph to apply a 90' swath"

5. **Will you hire an applicator (e.g. agricultural consultant company) to treat these soybeans with foliar insecticides in 2017 if chemical treatment is needed?**
   ◦ Yes (please specify company or individual):
   ◦ No, I will apply insecticides myself
   ◦ I'm not sure

Please return to Katelin Goebel in the envelope provided. Thank you.

Print your name here ____________________

Contact Information Form

1. **May we contact you to identify foliar insecticide spraying date(s) in the summer of 2017?**
   ○ Yes
   ○ No

2. **What is the best way to reach you?**
   ○ Home phone
   | | | |
   ○ Cell phone
   | | | |
   ○ Both home & cell phones

3. **In order to identify the exact date(s) of spraying, how often are you comfortable with us contacting you during the late summer of 2017?**
   ○ Weekly
   ○ Semi-weekly
   ○ As often as necessary as the spraying date approaches (no more than once daily)

4. **Would you like to receive a paper copy of the LCCMR work plan for our project?**
   This can also be found at: http://www.lccmr.leg.mn/projects/2016/work_plans_may/_2016_03n.pdf
   ○ Yes
   ○ No

5. **Would you like to receive a paper copy of your responses to the Spraying Practices Survey and Contact Information Form?**
   ○ Yes
   ○ No

6. **If you rent your land, please provide the name and address of your renter so we may send them a letter and survey:**

Please return to Katelin Goebel in the envelope provided. Thank you.

Insecticide Exposure Risk for Grassland Wildlife on Public Lands[1]

2016 Annual Report

23 January 2017

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Insecticide Exposure Risk for Grassland Wildlife on Public Lands

2016 Annual Report

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Abstract: There is growing concern about the potential effects of insecticides on grassland wildlife that inhabit landscapes dominated by agriculture. In the agricultural region of southern and western Minnesota, there is particular concern about the risk of exposure of wildlife on public lands to insecticides used to control soybean aphids. Our objectives are to assess the direct and indirect exposure risks of grassland birds and their insect food resources to insecticides encountered via aerial drift. We will quantify chemical residues in public grasslands, measure chemical residues on invertebrates, and assess effects of insecticide exposure to invertebrate abundance near sprayed fields before and after routine applications of insecticides are used to control soybean aphids. We have secured funding for this project, recruited a graduate student, and identified the chemicals that we will focus our sampling efforts on. We are in the process of determining study sites, requesting landowner cooperation with our project, and refining the details of the study design. Our research will inform land managers and private landowners on how best to manage grasslands to reduce risks of wildlife to insecticide drift.

INTRODUCTION

Grassland habitat loss and fragmentation is a major concern for grassland-dependent wildlife throughout the Midwestern United States. Increasing evidence suggests that acute toxicity to pesticides may be a greater threat to grassland bird populations than habitat loss due to agricultural intensification (Mineau and Whiteside 2013). In Minnesota, many remaining grasslands are highly fragmented and surrounded by row crops, including over 3 million hectares of soybeans (USDA 2016b). The insecticides used to combat soybean aphids, namely chlorpyrifos, lambda-cyhalothrin, and bifenthrin, have been shown to be highly toxic to non-target organisms such as birds and pollinators (Christensen et al. 2009, NPIC 2001, Johnson et al. 2010). Members of the public and Minnesota Department of Natural Resources (MNDNR) wildlife managers have observed fewer birds and insects after these insecticides are applied in late summer, raising concerns regarding the impacts of these chemicals on populations of grassland wildlife. However, little is known about the deposition of these pesticides in grasslands and the exposure risk to wildlife in an agricultural matrix under typical application conditions.

Restoring grasslands within the agricultural matrix is a priority conservation concern in western Minnesota. Information about risk of exposure of grassland wildlife to insecticides in this landscape is lacking, but this knowledge would help managers with grassland conservation efforts. Agricultural practices and policies that influence cover-type composition (e.g., a 2016 Minnesota law that requires perennial vegetation buffers up to approximately 15 m (50 ft) wide along public waters and ditches) may result in addition of grasslands to the landscape. However, how and to what extent grassland birds, their insect prey, and beneficial insects such as pollinators using these buffers are exposed to spray drift from adjacent field operations is unknown. Similarly, Minnesota's Pheasant Summit Action Plan and Prairie Conservation Plan both aim to offset grassland cover losses due to declining Conservation Reserve Program (CRP) enrollments by establishing grassland/wetland habitat complexes within the agricultural matrix.

One important avenue of exposure of grassland wildlife to agricultural insecticides is through aerial drift. Drift occurs when insecticides are sprayed on crops but environmental factors result in their transport to areas beyond the targeted application area. Distance of travel for insecticide drift is highly dependent on factors such as humidity, wind speed, and application method. Furthermore, the reported drift distances vary widely, ranging from 5 m to 1,600 m (Davis and Williams 1990, E. Runquist, personal communication). For many standard insecticide application regimes in agricultural landscapes, there is little or no information about drift and exposure risk to wildlife in grasslands - information necessary to effectively design grasslands set aside and managed for wildlife.

The objectives of our research are to assess the direct and indirect exposure risks of grassland birds and their insect food resources to soybean aphid insecticides in Minnesota's farmland region. First, we will quantify the concentration of insecticides along a gradient from soybean field edge to grassland interior to assess the potential for grassland wildlife to be directly exposed to chemicals via contact with insecticides resulting from spray drift. Second, we will quantify the chemical residue on invertebrates that serve as prey items of grassland birds, predatory insects, and other insectivores. This will allow us to assess the indirect exposure risk of birds and other wildlife to these chemicals through consumption of invertebrates. Finally, we will quantify and compare the relative abundance, richness, diversity, and biomass of invertebrates along a gradient from soybean field edge to grassland interior prior to and post-application to assess the indirect impact of insecticides on food availability for grassland nesting birds and other wildlife. Our research will allow us to inform decision-making by land managers and private landowners so they can better design grasslands and buffers, thus reducing the impacts of spray drift on wildlife in these habitats.

STUDY AREA

We are currently evaluating potential study sites on Wildlife Management Areas (WMAs) in the west-central (WC), central (C), southwest (SW), and south-central (SC) agricultural regions of Minnesota (Fig. 1). These WMAs are owned by the MNDNR and are managed with the intent of providing high quality habitat for wildlife. Corn and soybean fields account for approximately 50% of the landscape in these four regions. The SW and SC regions are the most intensively farmed; corn and soybeans are planted on 75% of those landscapes (USDA 2016a, USDA 2016b).

Each treatment study site will consist of a WMA including upland grassland directly adjacent to and downwind of a soybean field. We have been consulting area wildlife managers and will be contacting private landowner cooperators to choose 5-7 treatment study sites. We will prioritize 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. The treatment study sites will be adjacent to fields sprayed by the same application method (i.e., either ground boom or plane). We will also choose 2 control study sites with similar site characteristics except that control sites will not be sprayed with any chemicals to control aphids.

METHODS

Within each treatment study site, we will conduct sampling at stations placed at multiple distances (<5 m to ≥100 m) along each of 3 transects extending from a treated soybean field edge to an adjacent grassland interior (Fig. 2). We will align transects perpendicular to the soybean field edge and space them ≥200 m apart to reduce the likelihood of duplicate insecticide exposure during the spraying event. We will conduct sampling to assess both direct and indirect exposure risks to grassland wildlife, especially birds and insects, immediately after spraying and at additional periods post-application. As a control, we will also sample invertebrates in grasslands adjacent to untreated soybean fields. We will use portable weather stations or pocket weather meters to estimate relevant weather data (e.g., temperature, wind speed, wind direction, humidity, dew point) near the time of spraying and at several periods post-spraying, including insect sampling periods.

Direct Exposure Risk

To assess the potential for direct exposure of birds and other wildlife to soybean aphid insecticides, we will measure the amounts of chemicals deposited in grasslands during and after soybean fields are sprayed. We are evaluating the most appropriate method by which this will be accomplished. One option is to measure the amount of organic chemicals passively adsorbed to a hydrophobic silicone surface (Wennrich et al. 2002, T. Johnston, personal communication). We may use feather-covered samplers instead to simulate chemical accumulation on the body of birds, as pesticide concentrations have been shown to be detectable in bird feathers (Abbasi et al. 2016). Alternatively, we may analyze insecticide residues on grassland vegetation. We will take samples within 24 hours of spraying and properly store them for later chemical analysis. At control study sites, we will sample at 4 stations. We will collect these samples within the same timeframe as at treatment study sites and store them for later analysis.

Indirect Exposure Risk

To assess the potential for birds and other insectivorous wildlife to be exposed to insecticides indirectly, we will examine the chemical residues on invertebrates collected once prior to spraying and several days post-spraying at each treatment study site. We will sample ground-dwelling invertebrates using pitfall traps and canopy dwelling invertebrates via sweep netting (Brown and Matthews 2016, Doxon et al. 2011). We will combine pitfall trap and sweep net samples taken from the same station during the same period into 1 sample and properly store them for later chemical analysis.

Indirect Effects of Exposure
To quantify and compare the abundance, richness, diversity, and biomass of invertebrate prey items before and after spraying, we will collect additional pitfall trap and sweep net samples at the same stations. We will store them for later sorting and identification to at least the family level. We will place emphasis on invertebrate orders important in the diets of grassland nesting birds, including: Araneae (spiders), Orthoptera (grasshoppers, crickets, and katydids), and Coleoptera (beetles).

We will send samples that require chemical analysis to a lab that is to be determined. Samples will undergo a thermal desorption gas chromatography-mass spectrometry (GC-MS) process in which concentrations of each insecticide are determined by comparing peaks and retention times of standards to samples. An alternative analytical method may be used for the invertebrate samples. This alternative method will chemically extract the target chemicals from invertebrate samples prior to the GC-MS analysis. This will require dilution of the samples, however, and will result in less sensitive measurements. Although our experimental design will focus on soybean fields sprayed with chlorpyrifos, lambda-cyhalothrin, and/or bifenthrin, the chemical analyses are designed to allow us to quantify additional pesticides present in the samples.

The specifics of the experimental design and statistical analysis are being developed by the graduate student in cooperation with the project's principal investigators and collaborators.

RESULTS

To date, we have (1) established the intra-agency agreements that support this project at the Minnesota Cooperative Fish and Wildlife Research Unit, (2) secured funding for this project through the Minnesota Environment and Natural Resources Trust Fund (ENRTF) as recommended by the Legislative-Citizen Commission on Minnesota Resources (LCCMR), (3) recruited a graduate student currently working on this project at the University of Minnesota, (4) contacted representatives at 12 farmer cooperatives across 5 counties to gather information about current spraying methods used in our study area, (5) identified the insecticides that will constitute the focus of our sampling efforts, (6) identified 25 WMAs that fit our criteria as potential treatment study sites, (7) compiled a list of 180 landowners who own property adjacent to these WMAs, (8) drafted a research summary letter and survey to be sent to these landowners to request their cooperation with our project, and (9) introduced our research project to MNDNR Wildlife Managers and other researchers with LCCMR/ENRTF funding for projects relating to pollinators. During spring 2017, we will introduce this project at the annual meeting of the Minnesota Chapter of The Wildlife Society and Midwest Fish and Wildlife Conference, identify additional potential study sites, send research intent letters and surveys to landowners, complete the project design, organize logistics, and purchase equipment. We will initiate data collection in summer 2017.

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Figure 1. Minnesota's agricultural regions as outlined in Minnesota Department of Natural Resources (MNDNR) annual August Roadside Surveys. The study sites for this project will include Wildlife Management Areas owned and managed by the MNDNR in the west-central (WC), central (C), southwest (SW), and south-central (SC) regions.

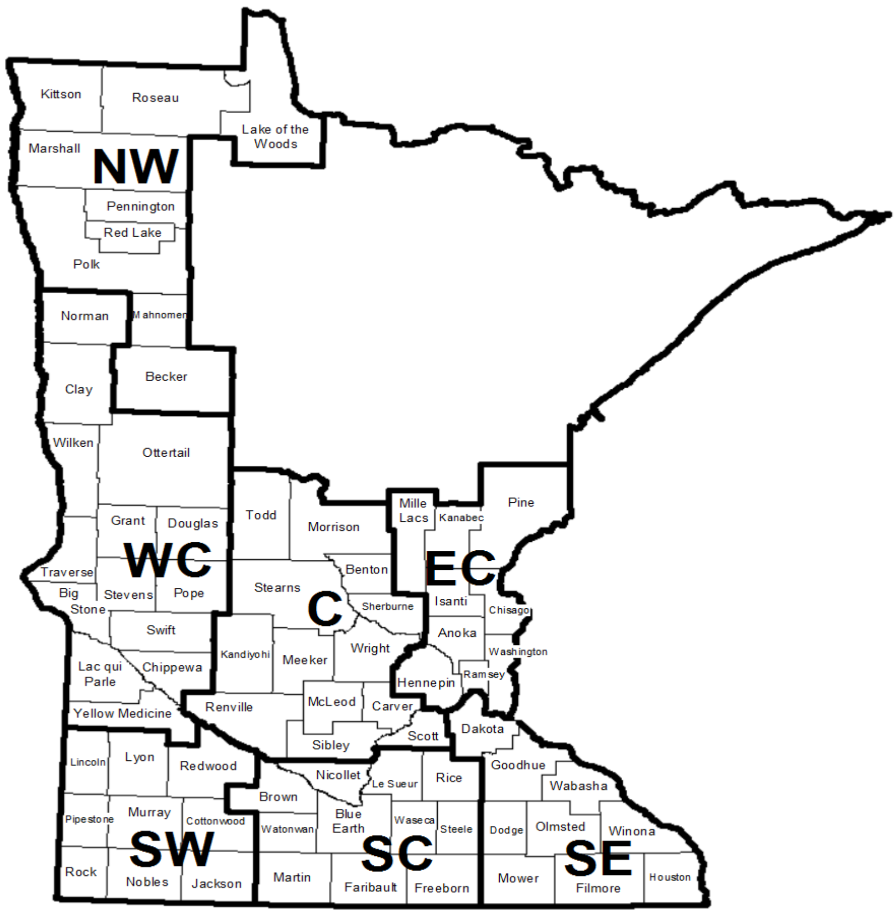

Figure 2. Example field sampling design that will be used to assess the exposure risk of grassland wildlife to soybean aphid insecticides. Sampling will be conducted on Wildlife Management Areas (outlined in black) adjacent to privately-owned soybean fields sprayed for aphid infestations. White lines indicate sampling transects established perpendicular to the soybean field edge and extending ≥100 m into the grassland.

Insecticide Exposure Risk for Grassland Wildlife on Public Lands[1]

2017 Annual Report

16 March 2018

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[1] Minnesota Department of Natural Resources Agreement no. 113497

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Abstract: There is growing concern about the potential effects of insecticides on grassland wildlife that inhabit landscapes dominated by agriculture. In the agricultural region of southern and western Minnesota, there is particular concern about the risk of exposure of wildlife on public lands to insecticides used to control soybean aphids. Our objectives are to assess the direct and indirect exposure risks of grassland birds and their insect food resources to insecticides encountered via aerial drift. We are quantifying chemical residues in public grasslands, measuring chemical residues on invertebrates, and assessing effects of insecticide exposure to invertebrate abundance near sprayed fields before and after routine applications of insecticides are used to control soybean aphids. We collected data from 2 treatment and 2 control sites during summer 2017 and are preparing for our final field season in summer 2018. Our results will inform land managers and private landowners on how best to manage grasslands to reduce risks of wildlife to insecticide drift.

INTRODUCTION

Loss and fragmentation of grassland cover is a major concern for grassland-dependent wildlife throughout the Midwestern United States. Increasing evidence suggests that acute toxicity to pesticides may be a greater threat to grassland bird populations than habitat loss due to agricultural intensification (Mineau and Whiteside 2013). In Minnesota, many remaining grasslands are highly fragmented and surrounded by row crops, including over 3 million hectares of soybeans (USDA 2016*a*). The insecticides used to combat soybean aphids, namely chlorpyrifos, lambda-cyhalothrin, and bifenthrin, disrupt nervous system functioning of organisms and are highly effective against target insect pests; however, they are highly toxic to non-target organisms such as birds and pollinators (NPIC 2001, Christensen et al. 2009, Johnson et al. 2010). Chlorpyrifos is an organophosphate insecticide, whereas lambda-cyhalothrin and bifenthrin are pyrethroids. Members of the public and Minnesota Department of Natural

Resources (MNDNR) wildlife managers have observed fewer birds and insects after these insecticides are applied in late summer, raising concerns regarding the impacts of these chemicals on populations of grassland wildlife. However, little is known about the deposition of these pesticides in grasslands and the exposure risk to wildlife in an agricultural matrix under typical application conditions.

One important avenue of exposure of grassland wildlife to agricultural insecticides is through aerial drift associated with routine application to prevent and control pest outbreaks. Drift occurs when insecticides are sprayed on crops but environmental factors result in their transport to areas beyond the targeted application area. Distance of travel for insecticide drift is highly dependent on factors such as humidity, wind speed, and application method. Furthermore, the reported drift distances vary widely, ranging from 5 m to 1,600 m (Davis and Williams 1990, E. Runquist, MN Zoo, personal communication). For many standard insecticide application regimes in agricultural landscapes, there is little or no information about drift and exposure risk to wildlife in grassland cover types - information necessary to effectively design grasslands set aside and managed for wildlife.

Restoring grasslands within the agricultural matrix is a priority conservation concern in western Minnesota. Information about risk of exposure of grassland wildlife to insecticides in this landscape is lacking, but this knowledge would help managers with grassland conservation efforts. Agricultural practices and policies that influence cover-type composition [e.g., a 2016 Minnesota law that requires perennial vegetation buffers of an average of 15 m (50 ft) width and 9 m (30 ft) minimum width along public waters and 5 m (16.5 ft) width along public drainage systems] may result in addition of grassland cover to the landscape. However, how and to what extent grassland birds, their insect prey, and beneficial insects such as pollinators using these buffers are exposed to spray drift from adjacent field operations is unknown. Similarly, Minnesota's Pheasant Summit Action Plan (MNDNR 2015), Prairie Conservation Plan (MN Prairie Plan Working Group 2011), and Wildlife Action Plan (MNDNR 2016) aim to offset grassland cover losses by establishing grassland/wetland complexes within the agricultural matrix.

Chlorpyrifos, lambda-cyhalothrin, and bifenthrin have all been shown to have detrimental effects on non-target organisms. Lab studies have shown chlorpyrifos to be very highly toxic to several bird species including ring-necked pheasants (*Phasianus colchicus*), American robins (*Turdus migratorius*), common grackles (*Quiscalus quiscula*), and mallards (*Anas platyrhynchos*; Tucker and Haegele 1971). Furthermore, sub-lethal effects in birds resulting from chlorpyrifos exposure (e.g., altered brain cholinesterase activity, altered behaviors, reduced weight gain) have been documented in both lab and field studies (McEwen et al. 1986, Richards et al. 2000, Al-Badrany and Mohammad 2007, Moye 2008, Eng et al. 2017). Thus, exposure to sub-lethal doses of chlorpyrifos has the potential to cause indirect mortality of wildlife through factors such as increased predation risk or exposure to harsh weather conditions. Lambda-cyhalothrin is highly toxic to pollinators including bees and mildly toxic to birds (NPIC 2001). Insect abundance and diversity has decreased in fields exposed to this insecticide during field studies (Galvan et al. 2005, Langhof et al. 2005, Devotto et al. 2007). Birds relying on insects as a source of protein therefore may face reduced food availability when lambda-cyhalothrin is applied in agricultural landscapes. Bifenthrin is low in toxicity to upland birds; however, it is very highly toxic to aquatic organisms and its use may decrease food availability for birds that feed on fish and aquatic insects (Siegfried 1993, Johnson et al. 2010). Bifenthrin is also very highly toxic to bumblebees (*Bombus* spp.), with one study showing 100% mortality by contact (Besard et al. 2010). Consequently, these insecticides have the potential to detrimentally affect both birds and their insect food resources.

Reduced insect abundance and diversity resulting from insecticide application may pose a threat to grassland wildlife that use insects as a food source. Protein-rich insects are especially important for breeding grassland birds during egg-laying and the nestling and fledgling periods. The majority of breeding grassland birds' diets consist of insects, and insects are the primary food item fed to nestlings (Wiens and Rotenberry 1979, Kaspari and Joern 1993). Furthermore, there is correlative evidence between reduced insect food supplies and reduced nesting success for birds in fragmented habitat surrounded by cultivated fields (Zanette et al. 2000). Thus, the reduction of food availability via mortality of non-target insects from insecticides has the potential to negatively impact grassland bird reproduction and survival.

The objectives of our research are to assess the direct and indirect exposure risks of grassland birds and their insect food resources to soybean aphid insecticides in Minnesota's farmland region. First, we are quantifying the concentration of insecticides along a gradient from soybean field edge to grassland interior to assess the potential for grassland wildlife to be directly exposed to chemicals via contact with insecticides resulting from spray drift. Second, we are quantifying the chemical residue on invertebrates that serve as prey items of grassland birds, predatory insects, and other insectivores. This will allow us to assess the indirect exposure risk of birds and other wildlife to these chemicals through consumption of invertebrates. Finally, we are quantifying and comparing the relative abundance, richness, diversity, and biomass of invertebrates along a gradient from soybean field edge to grassland interior prior to and post-application to assess the indirect impact of insecticides on food availability for grassland-nesting birds and other wildlife. Our research will allow us to inform decision-making by land managers and private landowners so they can better incorporate areas of grassland cover within agricultural landscapes, thus reducing the impacts of spray drift on wildlife in these systems.

STUDY AREA

Our potential study sites are in the west-central (WC), central (C), southwest (SW), and south-central (SC) agricultural regions of Minnesota (Fig. 1). Corn and soybean fields account for approximately 50% of the landscape in these four regions. The SW and SC regions are the most intensively farmed; corn and soybeans are planted on 75% of those landscapes (USDA 2016*a*, *b*). Our 2017 study sites consisted of Wildlife Management Areas (WMAs) owned by the MNDNR and managed with the intent of providing high quality habitat for wildlife. We may consider other public lands (e.g., Scientific and Natural Areas) in addition to WMAs for our 2018 study sites.

We identified 16 potential study sites via GIS prior to the start of the field season but in-person site visits reduced our potential list to 7 treatment sites for various reasons (e.g., adjacent row crop was corn instead of soybeans) and 4 control sites. Four out of 7 landowners for our potential treatment sites agreed to cooperate with our study so that we could precisely time our sampling efforts. However, 1 landowner did not spray for aphids in 2017 and 1 landowner failed to give us advanced notice of his spraying efforts. Ultimately, we sampled 2 treatment and 2 control sites in summer 2017 (Table 1).

Each treatment site consisted of a WMA including upland grassland directly adjacent to and east of a soybean field. We chose this configuration based on the prevailing wind direction in the region to increase the likelihood that our treatment study sites would be downwind of sprayed soybean fields. We prioritized 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 and create grassland bird and beneficial insect habitat. Control sites had similar characteristics except that control sites were east of cornfields. We plan to sample an additional 6-8 treatment sites and 2 control sites during summer 2018.

METHODS

We conducted sampling to assess both direct and indirect exposure risks to grassland wildlife, especially birds and insects, immediately after spraying and at additional periods post-application. Within each treatment site, we conducted sampling at stations placed at 7 distances (0, 5, 25, 50, 100, 200, and 400 m) along each of 3 transects extending from a treated soybean field edge to an adjacent grassland interior (Fig. 2). Thus, there were 21 drift sampling stations at each treatment (i.e., pesticide application) or control (i.e., non-application) site. We aligned transects perpendicular to the soybean field edge and spaced them 100 m apart to reduce the likelihood of duplicate insecticide exposure during the spraying event. As a control, we also conducted sampling in grasslands adjacent to cornfields. We used Kestrel 5500AG agricultural weather meters (Nielsen-Kellerman Co., Boothwyn, PA, U.S.A.) mounted on tripods and equipped with weather vanes to measure relevant weather data including temperature, wind speed, wind direction, humidity, and dew point at the time of spraying and during insect sampling periods (use of trade names does not imply endorsement by the U.S. Government, the University of Minnesota, or the MNDNR). We also measured vegetation characteristics at each station. Weather conditions and vegetation characteristics may influence deposition rates of insecticides and we will include covariates related to these factors in mixed linear models of insecticide deposition. In addition, weather influences the availability of insects to be collected (Southwood and Henderson 2000).

Direct Exposure Risk

To assess the potential for direct exposure of birds and other wildlife to soybean aphid insecticides, we measured the amount of chemicals deposited in grasslands within hours of soybean fields being sprayed. We placed passive sampling devices (PSDs) and water-sensitive cards at ground level and mid-canopy level at each station described in the 2 hrs prior to spraying. Ground level sampling measures potential insecticide drift exposure of ground-nesting birds and other ground-dwelling wildlife. Mid-canopy sampling measures potential exposure of above-ground-nesting birds and many species of spiders and insects to insecticide drift. Ground-level and mid-canopy samples will be analyzed independent of one another. We collected 42 filter paper and 42 water-sensitive card samples at each site. We retrieved these samples within 2.5 hrs of treatment sites being sprayed, wrapped them in aluminum foil, enclosed them in airtight plastic storage bags, and placed them in a cooler with dry ice. At control sites, we sampled at the same 21 stations within the same timeframe as at treatment sites. We are storing filter paper from the PSDs in a -80° C freezer until we send them to the laboratory for chemical analysis.

PSDs were composed of Whatman grade 2 filter paper (GE Healthcare UK Ltd., Little Chalfont, UK) covering 1.27 cm (0.5 in) mesh steel hardware cloth in the shape of a cylinder. Organic molecules adhere to the surface of filter paper, and a 3-dimensional PSD mimics an animal being exposed to insecticides. We attached PSDs to upright plastic fence posts with zip ties. Similarly, we attached 4 water-sensitive cards (Syngenta, Basel, CH) to steel mesh hardware cloth (2 on the vertical plane and 2 on the horizontal plane) to collect spray droplets. These cards change color from yellow to purple when they encounter liquid.

Indirect Exposure Risk

To assess the potential for birds and other insectivorous wildlife to be exposed to insecticides indirectly, we will examine the chemical residues on invertebrates collected on the day of spraying at each treatment site. We sampled ground-dwelling invertebrates using vacuum sampling and canopy dwelling invertebrates via sweep netting (Southwood and Henderson 2000). These sampling methods collected invertebrates of differing size classes and taxa (Doxon et al. 2011). We collected these samples on paired 30-m transects extending perpendicular to the field edge from the 0-, 5-, and 25-m stations for a total of 9 stations (Fig. 3). We placed vacuum sampling and sweep netting transects 1-2 m apart at each station to minimize disturbance of sampling and maximize the likelihood that the invertebrate communities being sampled were similar (Doxon et al. 2011). We combined vacuum samples and sweep net samples taken from the same station into 1 sample in sterilized plastic bags and placed them on dry ice immediately after collection in the field. We are storing them in a -80◦ C freezer until later chemical analysis.

We will send samples that require chemical analysis to the U. S. Department of Agriculture - Agricultural Marketing Service (USDA-AMS) National Science Laboratory (Gastonia, NC, U.S.A.) to test for chemical levels of our three primary chemicals of interest (chlorpyrifos, lambda-cyhalothrin, and bifenthrin) and several additional pesticides (particularly those classified as neonicotinoids and fungicides) commonly used in Minnesota's agricultural region. Chemical analyses will use a solvent extraction method followed by concentration of the extracts by evaporation. Concentrated extracts will then be subjected to Gas Chromatography/Mass Spectrometry (MS)-Negative Chemical Ionization to test for organophosphates and pyrethroids, and Liquid Chromatography/MS/MS for neonicotinoids and fungicides. Chemical residues will be reported in parts per billion.

Indirect Effects of Exposure

To quantify and compare the abundance, richness, diversity, and biomass of invertebrate prey items before and after spraying, we collected additional vacuum and sweep net samples 1-3 d prior to spraying and 3-5 d and 19-21 d post-spraying. We collected these samples between the 0- and 5-m stations and at the 25- and 100-m stations on paired 20-m transects. Additionally, we collected insect samples at the same 3 distances along an added transect 3-5 d and 19-21 d post-spraying. This transect was not adjacent to our 3 original transects to ensure that these samples were not affected by our previous disturbance of the area due to sampling activities. We combined vacuum and sweep net samples from each station into 1 Whirl-Pak plastic bag and preserved insects in ethanol. We are sorting and identifying these insects to the family level. We are placing emphasis on invertebrate orders important in the diets of grassland nesting birds, including: Araneae (spiders), Orthoptera (grasshoppers, crickets, and katydids), and Coleoptera (beetles). After identification, we will dry and weigh invertebrates to measure biomass and measure them to sort into size classes preferred by grassland birds and nestlings.

Vegetation Measurements

We measured ground cover, canopy cover, litter depth, maximum height of live and dead vegetation, vertical vegetation density, and species richness at 3 locations parallel to the field edge at each station and at both ends of insect sampling transects (Fig. 3). We recorded these vegetation characteristics at the 21 drift sampling stations 1-3 d prior to spraying and at each of the 9 insect sampling stations at 1-3 d prior to spraying and 3-5 d and 19-21 d post-spraying. Using a modified point-intercept method, we categorized ground cover into bare ground, litter, and other (i.e., woody debris, rock, or gopher mound; BLM 1996). We determined canopy cover from nadir digital photographs taken of each plot from 1.5 m above the ground using the program SamplePoint (Booth et al. 2006). Canopy cover categories included grass, forb, standing dead, woody vegetation, and other. 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 recorded the maximum height of live and dead vegetation within each plot to the nearest 0.5 dm. We measured vertical vegetation density by placing a Robel pole in the center of each plot and estimating the visual obstruction reading (VOR) in each of the 4 cardinal directions (Robel et al. 1970). We recorded the lowest 0.5-dm mark visible on the pole from 4 m away and 1 m above the ground. Finally, we listed the dominant grass and forb species in each plot along the center transect only. This list was composed of up to 3 species of grasses and 3 species of forbs that constituted significant portions of the canopy cover within the sampling frame and provided a qualitative assessment of the vegetation present at each site. We will include covariates derived from these measurements in mixed linear models of chemical deposition and abundance, richness, and diversity of invertebrates.

Researcher Safety

Long-term exposure to organophosphate and pyrethroid insecticides have been linked to increased human health risks in pesticide applicators. These chronic health risks include adverse respiratory effects (e.g., asthma and wheezing) and lung cancer (Lee et al. 2007, Hoppin et al. 2017). Bifenthrin is listed by the EPA as a possible human carcinogen (Johnson et al. 2010). The specimen labels of insecticide mixes including chlorpyrifos, lambda-cyhalothrin, and bifenthrin contain warnings of short-term side effects of exposure including eye, skin, nose, and throat irritation; headaches; nausea; and dizziness (Dow AgroSciences LLC 2014, Syngenta Crop Protection LLC 2014).

To reduce our exposure to these chemicals, we followed the Personal Protective Equipment (PPE) recommendations listed on the specimen labels of mixes containing chlorpyrifos. These mixes had more PPE recommendations than those containing lambda-cyhalothrin or bifenthrin, because chlorpyrifos has more severe health risks. We were equipped with more PPE than necessary, because the PPE recommendations on specimen labels are aimed at pesticide applicators who spend several days per year working in close proximity to these chemicals (D.

Herzfeld, University of MN, personal communication). Our overall exposure levels were very low, as we spent 4-5 h in grasslands adjacent to sprayed fields on only 1 d per each treatment site. We wore Tychem QC 127 series hooded Tyvek coveralls (DuPont, Wilmington, DE, U.S.A.), StanSolv 15 mil nitrile gloves (MAPA Professional, Colombes, FR), and rubber boots while collecting samples in treatment sites on the day of spraying immediately after chemical application. We had chemical-resistant goggles and half-mask air-purifying respirators on-hand should we have experienced eye, skin, nose, or throat irritation while in the field, but did not need to use them during our fieldwork in summer 2017.

Contacting Farmer Cooperatives

We contacted 12 farmer cooperatives (with the assistance of T. Klinkner, MNDNR) during fall 2016 to request the trade names of the soybean insecticides they most commonly applied during summer 2016 to decide the active ingredients upon which to focus our sampling efforts. We also requested information regarding the application method of these chemicals (i.e., ground boom or aerial). These cooperatives were located in Cottonwood, Kandiyohi, Redwood, Stearns, Swift, Meeker, and Watonwan counties in Minnesota. Several representatives reported using multiple active ingredients to combat soybean aphids (e.g., chlorpyrifos + lambda-cyhalothrin). This is a common practice, as active ingredients have differing withholding times and modes of action, and such products are readily available commercially (Koch et al. 2016).

Landowner Contact

Landowner cooperation is vital to timing our field sampling efforts. To request the cooperation of landowners and learn about their soybean-aphid-spraying practices, we mailed surveys to 206 landowners who owned land bordering 29 potential study sites in March and April 2016. We ultimately solicited landowner cooperation for our treatment sites by directly calling landowners and visiting their residences. This approach was more effective than mailing surveys and we will contact landowners in this manner to request their cooperation in early summer 2018.

RESULTS

Insecticides Used in Our Study Area

Lambda-cyhalothrin was the most common active ingredient reported to us by farmer cooperative representatives in our study area, followed by chlorpyrifos and bifenthrin. This reflects statewide insecticide usage trends from 2013: the Minnesota Department of Agriculture found that lambda-cyhalothrin was the most widely used chemical on 16% of surveyed soybean acres, followed by 13% being treated with chlorpyrifos and 5% with bifenthrin (MDA 2016). Overall, cooperative representatives estimated 56% of insecticide applications to control soybean aphids were by air and 44% were via ground boom in recent years. Our 2 treatment sites in summer 2017 were sprayed with insecticides with the trade names Bolton (chlorpyrifos + gamma-cyhalothrin; Cheminova, Inc., Research Triangle Park, NC, U.S.A.) and Endigo (lambda-cyhalothrin + thiamethoxam; Syngenta Canada Inc., Guelph, ON, CA). One treatment site was sprayed by air and the other was sprayed via ground boom.

Landowner Contact
Of the 206 surveys we sent to landowners who owned land adjacent to potential study areas, 28.1% were returned. However, not all landowners filled out the survey completely. Many landowners did not complete the survey because they rent their land and did not have information on aphid-spraying practices; this was the case for the landowners with soybean fields adjacent to our 2 treatment sites in 2017. We then called these landowners to request their renters' information. Approximately 13.6% of landowners completed the survey in its entirety and 7 landowners indicated that they would be planting soybeans adjacent to a WMA in 2017 and were willing to be contacted during the growing season. However, we did not select these WMAs at treatment sites for summer 2017.

Field Sampling
During our first field season in summer 2017, we collected 166 direct-exposure samples, 36 indirect-exposure invertebrate samples, and 132 indirect-effect invertebrate samples in 2 treatment and 2 control sites (Table 2). We will send samples requiring chemical analysis to the USDA-AMS National Science Laboratory upon approval of a multi-year master contract. This process has taken longer than expected, but the lab typically processes samples in 10 business days once they receive them; thus, analyses will commence quickly upon contract approval.

The aim of using water-sensitive cards was to provide an immediate visual assessment of whether drift occurred at our treatment sites in low humidity. However, at high humidity levels these cards demonstrated a color change in the absence of chemical drift. We were unable to attain quantifiable measures of chemical drift from these cards and thus, we will be discontinuing their use in 2018.

ACCOMPLISHMENTS

Through January 2017
- Contacted representatives at 12 farmer cooperatives across 7 counties to gather information about current spraying methods used in our study area (T. Klinkner, MNDNR)
- Identified the insecticides that will constitute the focus of our sampling efforts
- Drafted a research summary letter and survey to be sent to potential cooperating landowners

January 2017-present
- Sent a research summary letter and survey to 206 landowners who own property adjacent to 29 potential study site WMAs
- Contacted landowners via phone and in-person to request their cooperation with the project
- Refined the sampling design
- Purchased project supplies and equipment
- Hired 2 technicians through the MN DNR to assist with field sampling efforts
- Identified 16 potential study sites using GIS and in-person site visits

- Collected 166 direct exposure samples, 36 indirect exposure invertebrate samples, and 132 indirect effect invertebrate samples in 2 treatment and 2 control sites during our first field season
- Recruited 3 undergraduate students to process indirect effect invertebrate samples

Work in Progress

- Setting up a multi-year master contract with the USDA-AMS National Science Laboratory in Gastonia, NC, U.S.A. following MNDNR purchasing policies
- Sorting insect samples collected in summer 2017
- Analyzing preliminary vegetation data
- Identifying additional study sites for 2018 field season
- Advertising a job posting to hire 1 technician for our 2018 field season

PUBLICATIONS

Davros, N. M., and Goebel, K. M. *In press*. Evaluating insecticide exposure risk for grassland wildlife on public lands. 2016 Summary of Wildlife Research Findings, Division of Fish & Wildlife: Minnesota Department of Natural Resources. St. Paul, Minnesota, U.S.A.

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PRESENTATIONS

Davros, N. M. 2016. Overview of grassland wildlife/insecticide exposure study. MN DNR Region 4 Wildlife Meeting, New Ulm, Minnesota, U.S.A. (July 2016)

Davros, N. M. 2016. Introduction of grassland wildlife/insecticide exposure study. LCCMR Pollinator & Partner Projects Meeting, St. Paul, Minnesota, U.S.A. (December 2016)

Davros, N. M. 2016. Update of grassland wildlife/insecticide exposure study. MN DNR Region 4 Wildlife Meeting, Lamberton, Minnesota, U.S.A. (December 2016)

Goebel, K. M., Davros, N. M., and D. E. Andersen. 2017. Insecticide exposure risk for grassland wildlife on public lands. Poster. Midwest Fish and Wildlife Conference, Lincoln, Nebraska, U.S.A. (February 2017)

Goebel, K. M., Davros, N. M., and D. E. Andersen. 2017. Insecticide exposure risk for grassland wildlife on public lands. Poster. Annual Meeting of the MN Chapter of The Wildlife Society, Callaway, Minnesota, U.S.A. (February 2017)

Davros, N. M. 2017. Does diversity matter? Ring-necked pheasant nest site selection and nest survival in grassland reconstructions. Little Lunch on the Prairie Webinar, WebEx Meeting. <https://www.youtube.com/watch?v=kidTWvK0a30&index=9&list=PLeh-ajY3F3JK8MgVek1eeWwtKibPLgzdc&t=2647s> (December 2017)

Davros, N. M. 2018. Update on grassland wildlife/insecticide exposure study. MN DNR Region 4 Wildlife Meeting, New Ulm, Minnesota, U.S.A. (January 2018)

Goebel, K. M., Davros, N. M., and D. E. Andersen. 2018. Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota. Lightning Talk. Midwest Fish and Wildlife Conference, Milwaukee, Wisconsin, U.S.A. (January 2018)

Goebel, K. M., Davros, N. M., and D. E. Andersen. 2018. (Poster) Insecticide exposure risk for grassland wildlife on public land in southwestern Minnesota. Annual Meeting of the MN Chapter of The Wildlife Society, St. Cloud, Minnesota, U.S.A. (February 2018)

Goebel, K. M., Davros, N. M., Andersen, D. E., and P. J. Rice. 2018. Evaluating insecticide exposure risk for grassland wildlife on public lands. LCCMR Pollinator and Partner Projects Meeting, St. Paul, Minnesota, U.S.A. (March 2018)

MEDIA OUTLET

Kennedy, T. October 2017. DNR wildlife researcher Nicole Davros working to help upland birds thrive. Star Tribune, Minneapolis, Minnesota, U.S.A. <http://www.startribune.com/dnr-wildlife-researcher-nicole-davros-working-to-help-upland-birds-thrive/450349283/>

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Figure 1. Minnesota's agricultural regions as outlined in the Minnesota Department of Natural Resources (MNDNR) annual August Roadside Surveys. The study sites for this project include Wildlife Management Areas owned and managed by the MNDNR and potentially other publically-owned grasslands in the west-central (WC), central (C), southwest (SW), and south-central (SC) regions of the state.

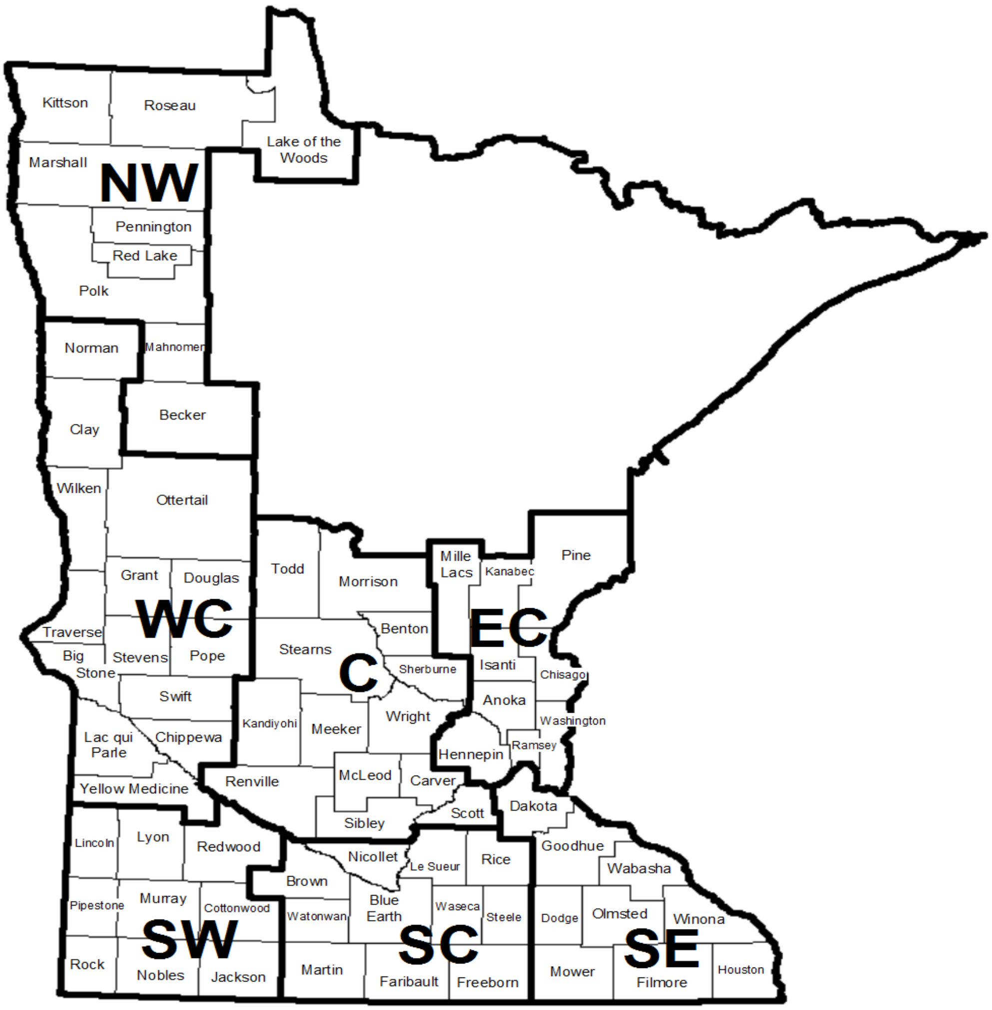


Figure 2. Field sampling design used to assess the exposure risk of grassland wildlife to soybean aphid insecticides. We conducted sampling on 2 WMAs east of privately owned soybean fields treated with insecticides to combat aphids in 2017. Our control sites are WMAs adjacent to cornfields. White lines indicate sampling transects established 100 m apart, perpendicular to the soybean field edge, and extending 400 m into the grassland. Yellow circles represent sampling stations at 0, 5, 25, 50, 100, 200, and 400 m from the field edge. This transect and station layout is used in both our treatment and control sites.

Figure 3. Field sampling design showing the layout of transects, chemical drift sampling, insect sampling, and vegetation sampling in each sample site. Features are color-coded to represent the timing of sampling in relation to the day of spraying. Numbers indicate distances in meters (not to scale).

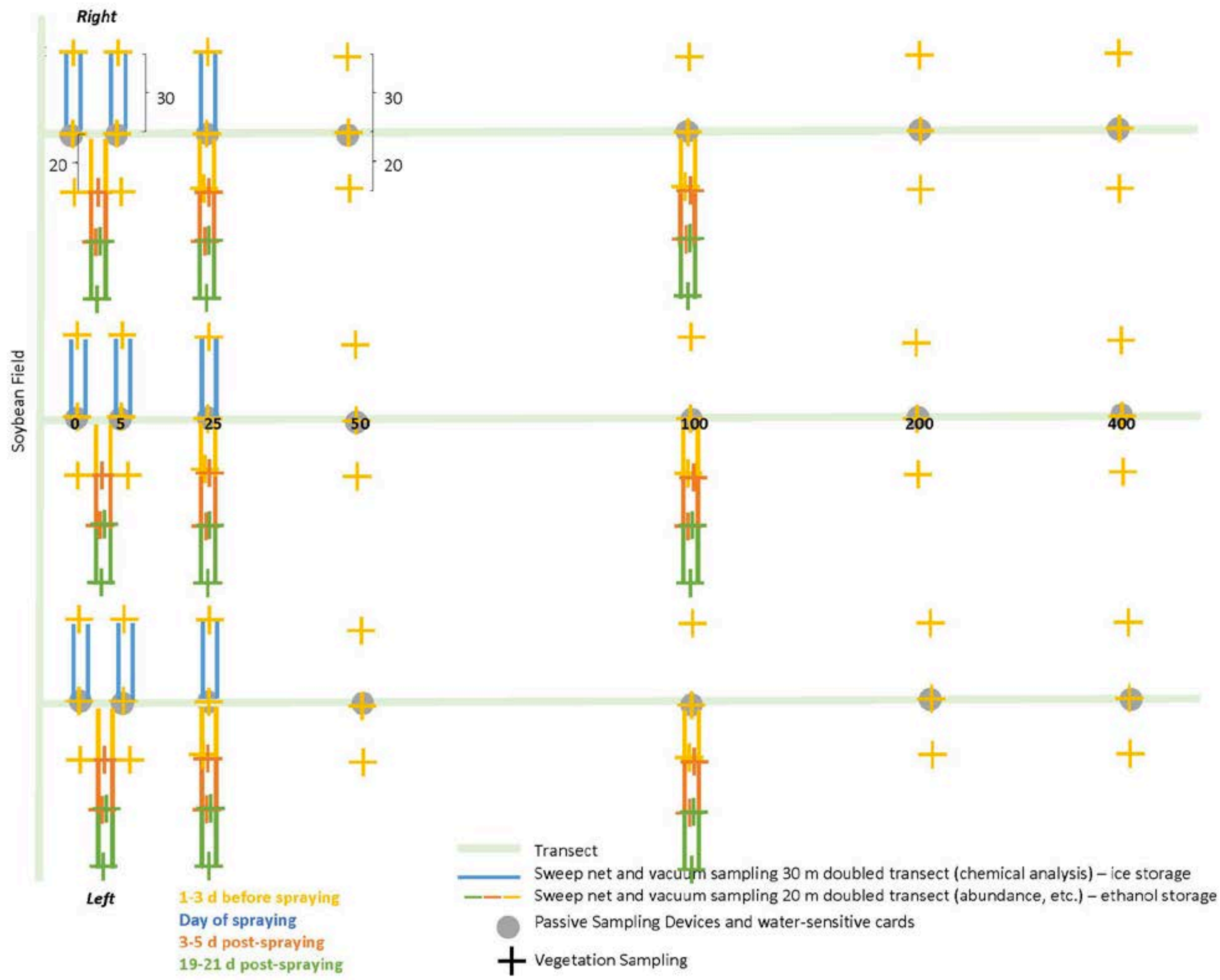


Table 1. Wildlife Management Areas (WMAs) selected as study sites for our summer 2017 field season. Agricultural region refers to the regions outlined in the Minnesota Department of Natural Resources annual August Roadside Surveys.

Site name	County	Site type	Agricultural region
Dead Horse WMA	Jackson	Control	Southwest
Heron Lake WMA: South Heron Unit	Jackson	Treatment	Southwest
Lake Maria WMA	Murray	Treatment	Southwest
Rolling Hills WMA	Lyon	Control	Southwest

Table 2. Timing and number of samples collected during our summer 2017 field season at 2 treatment and 2 control sites. (EF) denotes indirect effect invertebrate samples stored in ethanol and (EX) denotes indirect exposure invertebrate samples to be submitted for analysis of chemical residues.

Timeframe	Sample type	Number of samples/site	Total number of samples collected during July-Sept 2017
1-3 d before spraying	Insects (EF)	9	36
Day of spraying	PSDs & water-sensitive cards	*42	166
Day of spraying	Insects (EX)	9	36
3-5 d after spraying	Insects (EF)	12	48
19-21 d after spraying	Insects (EF)	12	48

*We omitted 1 400-m station at 1 treatment site (Heron Lake WMA: South Heron Unit) due to transect length constraints. We collected 40 PSD and water-sensitive card samples at that site.

Evaluating exposure of grassland wildlife to soybean aphid insecticides in Minnesota's farmland region

This project was funded by Funding provided by the Minnesota Environment and Natural Resources Trust Fund as recommended by the Legislative-Citizen Commission on Minnesota Resources (*M.L. 2016, Chp. 186, Sec. 2, Subd. 03n).*

Background

Concerns about the impact of insecticides on birds, pollinators, and other wildlife are gaining increasing attention. Chlorpyrifos, lambda-cyhalothrin, and bifenthrin (hereafter, target chemicals) are three insecticides commonly used to control soybean aphids in Minnesota's farmland region. Lab studies have shown these chemicals to be highly toxic to non-target organisms including several bird and beneficial insect species, but few studies have investigated the exposure of free-ranging wildlife to these chemicals. Chemical drift has been reported in other studies but very little Minnesota-specific data exists to understand this issue.

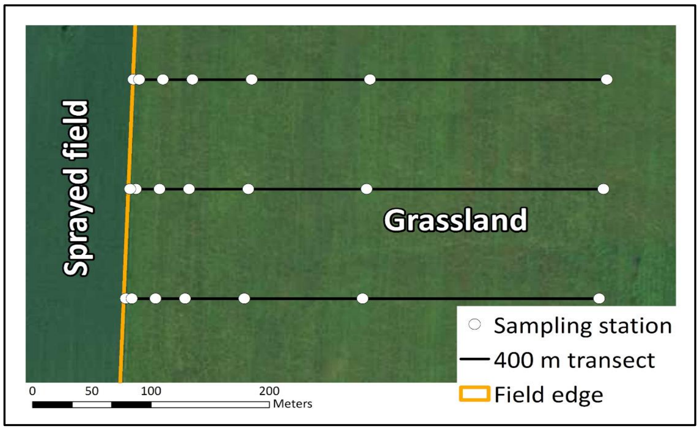
Objectives

Determine the environmentally-relevant exposure of Minnesota's grassland wildlife to insecticides sprayed to control soybean aphids. In particular:

1) Quantify the concentration of target chemicals along a gradient from soybean field edge to grassland interior to see if wildlife are (a) directly exposed via contact with spray drift and (b) indirectly exposed through their food (insect prey).

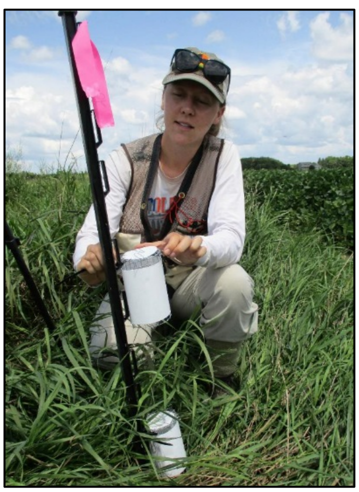
2) Compare relative abundance, richness, diversity, and biomass of insects along a gradient from soybean field edge to grassland interior prior to and post-spraying to assess the indirect impact of target chemicals on food availability for grassland nesting birds and other insectivorous wildlife.

Methods



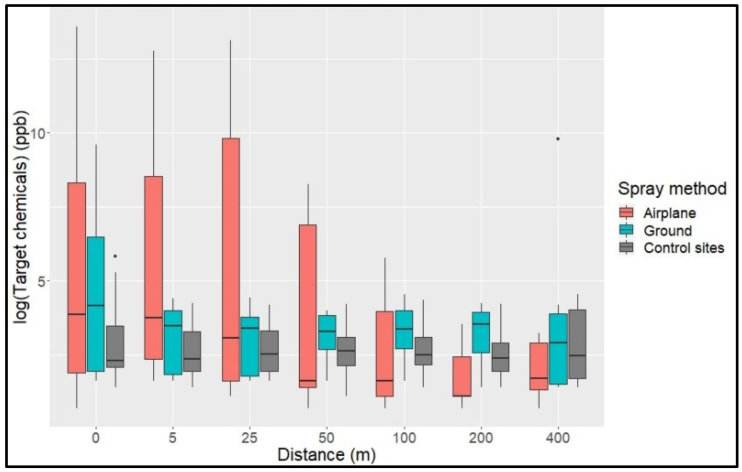
Our treatment sites were Wildlife Management Areas (WMAs) that had soybean fields immediately adjacent to the WMA. Our control sites were WMAs with a corn field immediately adjacent to the WMA. During summers 2017 and 2018, we worked closely with private landowner cooperators to precisely time our field data collection. At each Wildlife Management Area (WMA) site, we established three transects perpendicular to the soybean field edge. We then established sampling stations at various distances (0-400 m) along each transect.

The day before the spraying event, we collected data vegetation data at each sampling station. On the day of spraying, we deployed passive sampling devices made of filter paper immediately prior to the landowner spraying the adjacent soybean field. During the spraying event, we used a weather meter to collect data on weather conditions, including temperature, wind speed, wind direction, and relative humidity. Immediately after the spraying event, we collected our filter paper and insect samples and then properly stored these samples until later processing in the lab.



Results To Date

We sampled a total of five treatment and four control sites between July-September across our two field seasons. Our cooperators primarily sprayed chlorpyrifos but other insecticides were also used. We detected target chemicals at all distances examined (0-400 m from the grassland edge to the interior) at both treatment and control sites, suggesting that some baseline amount of spray drift occurred in the environment regardless of landowner spraying activities in the adjacent crop field.



Direct Exposure to Spray Drift

We also examined the importance of weather, vegetation, and other factors in explaining direct exposure. Our results indicate that mean air temperature and direction of the wind relative to the WMA during soybean spraying events, percent canopy cover of live vegetation (primarily grasses and flowering plants), distance from grassland field edge, and position in the grassland canopy layer were all important factors explaining deposition and drift of target chemicals onto WMAs. In particular, we found insecticide deposition was greater at the field edge than the grassland interior, and deposition was also greater at mid-canopy than ground level. Spray application method (i.e., ground or airplane) was not important in explaining patterns of target chemical deposition on our WMA sites.

Indirect Exposure to Spray Drift

We also detected target chemical residues on invertebrates at all distances examined (0-25 m) at both treatment and control sites. Further, our results showed mean air temperature and the maximum height of live vegetation best explained patterns of deposition on invertebrates. Distance to field edge had a weak relationship with chemical deposition on invertebrates, however, and is likely due to the shorter range of distances that we evaluated for this objective. Similar to direct exposure, spray application method was not important in explaining patterns of indirect exposure.

Future Work

We are still evaluating the indirect effects of spray drift on relative abundance, richness, diversity, and biomass of invertebrate prey.

Our final analyses and interpretation of all results will be completed this fall (2019) and incorporated into a thesis as part of our graduate student's Master's degree requirements for graduation. The chapters from her thesis will be turned into publications in peer-reviewed scientific journals and shared with other scientists and natural resource professionals.

We will continue to disseminate our results with DNR wildlife managers and other staff so they can incorporate our findings into their habitat acquisition, restoration, and management activities. Finally, we will also share our findings with our private landowner cooperators and the larger agricultural community to bring awareness to the issue of and factors influencing soybean aphid insecticide drift onto public grasslands. Ultimately, our research will help improve design and management of both public and private set-aside habitats for wildlife in Minnesota.

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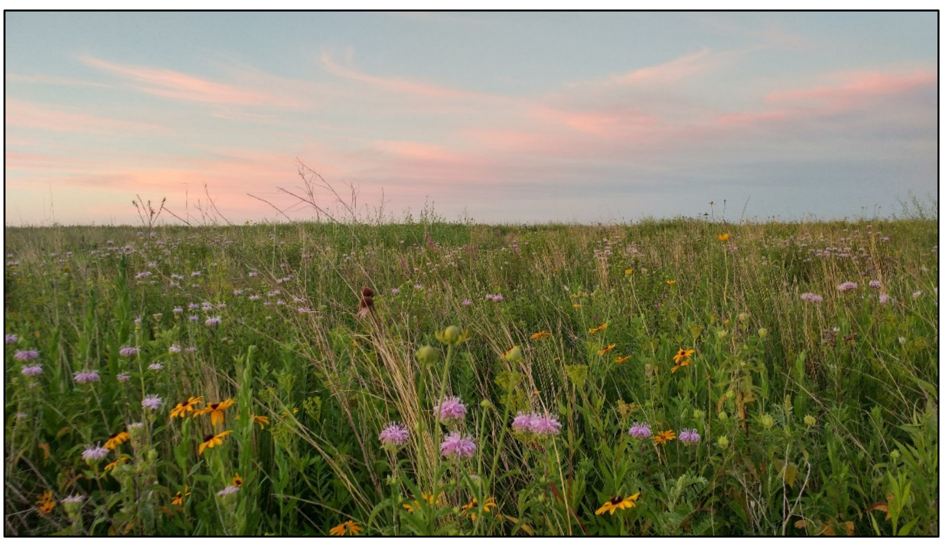

Updated: August 15, 2019

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DEPARTMENT OF NATURAL RESOURCES

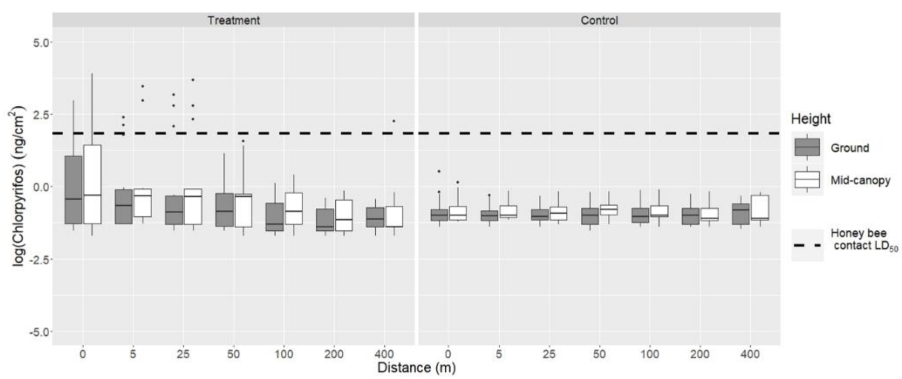
ENVIRONMENT AND NATURAL RESOURCES TRUST FUND

Wildlife Research Information Brief

Grassland Wildlife Exposure to Soybean Aphid Insecticides on Public Lands

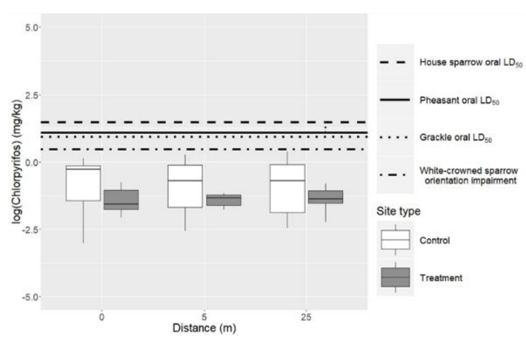
Research Summary

Foliar-application insecticides (chlorpyrifos, lambda-cyhalothrin, bifenthrin) are commonly used to treat soybean aphids in Minnesota but we have little information on exposure of free-ranging wildlife to chemical drift from spraying events. During summer 2017 and 2018, we sampled 5 treatment and 4 control sites in the farmland region to assess direct and indirect exposure of grassland wildlife to drift and the indirect impacts on their invertebrate food resources. We detected chlorpyrifos deposition in grasslands at all distances examined (0-400 m from soybean field edge) at both treatment and control sites, suggesting drift was ubiquitous on the landscape. Drift was greater near field edges than the grassland interior at treatment sites. We also found higher residue amounts on mid-canopy samples than ground-level samples. We detected chlorpyrifos residue amounts on arthropods that were below the acute oral LD_{50} values for common farmland bird species; however, residue amounts were above the contact LD_{50} for honey bees up to 25 m from the field edge. We found short-term reductions in overall arthropod abundance, bird prey abundance (specifically, individuals in the orders Araneae, Coleoptera, Lepidoptera larvae, and Orthoptera), and Coleopteran family richness in treatment sites but our other arthropod measures (i.e., biomass, richness of several other families) did not differ between treatment and control sites post-spraying.



Management Recommendations

Our results suggest grassland wildlife can be exposed directly and indirectly to drift from foliar-application insecticides. Furthermore, reductions in arthropod food abundance may impact breeding grassland birds, their young, or other insectivorous wildlife up to 21 days after spraying operations in the area. To reduce impacts of drift, natural resource managers should consider:



- Minimizing the perimeter-to-area ratio of grasslands. Acquiring and maintaining larger grassland tracts will help reduce edge effects of insecticide drift and provide refugia for arthropods to be able to recolonize affected areas.
- Using seed mixes and management techniques that create a thicker, more diverse canopy cover to reduce the amount of drift reaching mid-canopy and ground-dwelling wildlife.

Additional Resources

This research was completed by Katelin Goebel, University of Minnesota (Twin Cities), as part of her Master's degree. For more information, contact Dr. Nicole Davros, Farmland Wildlife Research Supervisor, at Nicole.Davros@state.mn.us, or view past Research Summaries under the "Habitats" tab here.