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Introduction

The monarch butterfly population has experienced an 80% decline in North America over the past two decades (Brower et al., 2012; Pleasants and Oberhauser, 2013; Jepsen et al., 2015). The three to four hectares of occupied overwintering forest in 2016 and 2015 was well below a target of six hectares needed to support a resilient population and reduce the risk of quasi-extinction (loss of the North American migration) in the next 10 to 20 years (Semmens et al., 2016). In response to a petition to evaluate the status of the species, the U.S. Fish and Wildlife Service (USFWS) is evaluating listing the monarch as a threatened species under the Endangered Species Act (USFWS, 2014 a, b). Under a court-supervised schedule, the USFWS must propose a listing decision in June 2019, which underscores the urgency of establishing viable, voluntary, state-based monarch conservation programs to provide USFWS a credible rationale to not list the species. If the monarch butterfly is listed, it could lead to significant regulatory and management burdens for farmers and livestock producers.

Increasing summer reproductive success in the North Central United States has been identified as a high priority for monarch conservation (Oberhauser et al. 2017; Flockhart et al. 2015). Up to 58% of the monarchs overwintering in Mexico originate in Iowa and neighboring states in the North Central U.S. (Flockhart et al. 2017). Monarch butterflies oviposit only on milkweed species (mainly *Asclepias* spp.) and primarily on common milkweed (*Asclepias syriaca*) in the North Central states (Malcolm et al. 1993). To increase monarch populations to levels that would reduce the probability of quasi-extinction by 50% over 20 years, Thogmartin et al. (2017) estimated that 1.3 to 1.8 billion additional milkweed stems need to be added to the North Central U.S. landscape.

The current amount of common milkweed stems in the North Central U.S. is estimated to be approximately 1.3 billion, the majority of which is in publically owned grasslands, land enrolled in conservation programs, such as the United States Department of Agriculture's Conservation Reserve Program (CRP), crop field borders, road rights-of-way, urban and suburban land cover, and miscellaneous non-agricultural habitat (Pleasants and Oberhauser 2013; Thogmartin et al. 2017). Analyses by Thogmartin et al. (2017) indicate that adding 1.3 to 1.8 billion new milkweed stems in the North Central U.S. will require an 'all hands on deck' approach across all land cover/land use categories. Habitat conservation goals can only be achieved through significant adoption of habitat restoration in privately owned land in agricultural landscapes. Establishment of milkweed and forage plants in rural roadsides; marginal crop land; portions of existing CRP land, pastures and grassland; and grassy areas bordering crop fields, will create a patchwork of habitat within agricultural landscapes dominated by corn and soybean production.

Overview of monarch conservation efforts in Iowa

In 2015, in response to the decline of the monarch butterfly, Iowa commodity and livestock organizations, pesticide technology providers, utility companies, city and county conservation organizations, Iowa colleges and universities, the Iowa Department of Agriculture and Land Stewardship, Iowa Department of Natural Resources and the College of Agriculture and Life Sciences at Iowa State University formed the Iowa Monarch Conservation Consortium (https://monarch.ent.iastate.edu). The consortium currently includes 42 participating organizations, with USFWS and USDA serving as *ex officio* members. The mission of the Iowa Monarch Conservation Consortium is to enhance monarch butterfly reproduction and survival in Iowa through collaborative and coordinated efforts of farmers, private citizens and their organizations through research, education and direct action.

The consortium's outreach and extension efforts draw upon all the member organizations to ensure the broad delivery of practical, science-based information on monarch butterfly conservation practices for Iowa's landscapes. Habitat improvements in rural landscapes are being designed to target underutilized areas that do not conflict with agricultural production, are sufficient in scale to support improved monarch breeding success, and strive to complement other conservation programs. In this regard, monarch habitat will be beneficial for pollinators and other wildlife species and can be 'stacked' with other conservation practices designed to reduce soil erosion and minimize nutrient runoff. The consortium published Version 1.0 of Iowa Monarch Conservation Strategy in February 2017 (https://monarch.ent.iastate.edu/files/file/iowa-monarch-conservation-strategy.pdf). Version 2.0 of the strategy is under development and will include Iowa's habitat goals, which will be integrated with neighboring North Central States' habitat targets (see: http://www.mafwa.org/?page_id=2347).

The research arm of the Iowa Monarch Conservation Consortium is based on the campus of Iowa State University and includes University and USDA scientists, technicians, graduate and undergraduate students. Below is a synopsis of ongoing demonstration and research activities.

Habitat establishment practices

A seed mix has been developed that includes three milkweed species and several nectar-producing forbs that benefit butterflies and bees (Appelgate et al. 2017). Grass species included in the mix are mostly short to medium height in order to reduce competition with forbs. All species are native to Iowa and should thrive in well-drained and moderately-drained soils. This seed mix has been used to establish monarch habitat at 46 demonstration sites. Thirteen demonstration sites are associated with establishment of approved edge-of-field practices in the Iowa Nutrient Reduction Strategy, including six saturated buffers sites and seven bioreactors sites. The remaining sites are in underutilized grass areas (21 sites) and land near hog confinements (12 sites). Supporting experiments are ongoing to develop methods to establish milkweed into land dominated by smooth brome grass, *Bromus inermis*.

Research completed at Iowa State University (Pocius et al. 2017 a, b) indicate a diversity of milkweed species including: *Asclepias exaltata* (Poke Milkweed), *A. hirtella* (Tall Green Milkweed), *A. incarnat*a (Swamp Milkweed), *A. speciosa* (Showy Milkweed), *A. sullivantii* (Prairie Milkweed), A. syriaca (Common Milkweed), *A. tuberosa* (Butterfly Milkweed), *A. verticillata* (Whorled Milkweed), and *Cynanchum laeve* (Honeyvine Milkweed) could benefit monarchs in their summer range. Ultimately, monarch seed mixes and planting guidelines will provide farmers and landowners practical and reliable methods to expand the benefits of existing groundcover to promote monarch caterpillar development and provide nectar sources for monarch butterfly adults. Pollinators and other wildlife species also will benefit from the increased diversity of plant species.

Monarch population modeling

Research is needed to better understand how habitat patch size, composition, and spatial arrangements (Oberhauser et al. 2017; Pleasants and Oberhauser, 2013; Zalucki et al. 2001, 2002; Zalucki and Lammers 2010; Zalucki et al. 2016) influence monarch survival and breeding success. This understanding provides a foundation for evaluating how different habitat patch distributions influence monarch population growth rates, which in turn supports conservation planning. Modeling is a useful tool for investigating how monarch butterfly reproductive performance, larval development, and population growth respond to habitat plant composition and quality, including milkweed stem density and spatial arrangement; habitat patch size; and habitat fragmentation patterns within a spatially explicit landscape. Previous modeling efforts in artificial landscapes have identified monarch perceptual range and habitat attractiveness, which influences the probability a butterfly will choose a given habitat patch, as important data inputs (Zalucki et al. 2016). Modeling efforts to date (Zalucki et al. 2016, Zalucki and Lammers, 2010) have also identified the important role individual, lone milkweed plants and small low-density patches of milkweed play in increasing the number of monarch eggs laid on a landscape. To further evaluate and advance understanding of how female monarchs utilize breeding habitat in Iowa, we have developed a model that uses spatially explicit landscapes that reflect current habitat conditions and spatial patterns (Grant et al., 2017a). The model can incorporate results from on-going studies that are refining estimates of monarch perceptual range and flight patterns (Fisher et al., 2017a, b).

Estimating risk of herbicides and insecticides

Monarch conservation benefits and risks associated with restoring habitat near conventional crop fields, due to potential pesticide exposure, are uncertain. The National Resources Conservation Service (NRCS) *Monarch Butterfly Wildlife Habitat Evaluation Guide* (2016) discourages placement of monarch breeding habitat within 38m (125ft) of crop fields that are treated with herbicides or insecticides; a risk-based rationale for the buffer size is not available. Employing a buffer of this size would result in a significant area of land unavailable for the establishment of breeding habitat. In Story County, Iowa, a 38m buffer around corn and soybean fields would eliminate approximately 84% of rural roadside habitat and 38% of grassland, Conservation Reserve Program (CRP) land, pastures, railroad rights-of-way, riparian corridors and wetlands. Based on possible pesticide use patterns, do the potential risks from pesticide exposure reduce the benefits of establishing habitat in close proximity to crop fields?

Herbicides

The effect of nonlethal herbicide injury to milkweed on monarch oviposition and larval development has not been investigated. Utilization of forbs by pollinators was reduced by exposure to low concentrations of herbicides (Bohnenblust et al. 2016), largely due to fewer floral resources. The mechanism of toxicity for growth-regulator herbicides may be due to the stimulation of abscisic acid and ethylene production in affected plants (Grossman 2009). At sublethal doses, the growth-regulator herbicides cause responses such as epinasty and malformed leaves. Since a high percentage of milkweed occurs adjacent to crop fields, there is a need for an off-target risk assessment of herbicides on the value of milkweed to monarchs. Such an assessment is particularly necessary for evaluating the value of new habitat patches.

Common milkweed persists in crop fields due to perennial rootstocks that are resilient to most herbicides; post-emergence herbicides can cause significant damage to shoots present at the time of application. Hartzler and Lizotte-Hall (Department of Agronomy, ISU) are undertaking dose response studies to determine the effect of fomesafen, a herbicide commonly used on soybeans, on biomass production of common milkweed. Field experiments are determining how herbicide injury influences utilization of common milkweed by adult monarchs. Fomesafen is being used to evaluate effects of in-field herbicide use, whereas dicamba is being used to study effects of off-field exposure.

Insecticides

Insecticides for managing insect corn and soybean pests include organophosphate, pyrethroid, neonicotinoid, and anthranilic diamide (Hodgson and VanNostrand G 2016; University of Tennessee 2016; DuPont 2010). Monarch larvae in existing and newly established milkweed patches near crop fields could be exposed to insecticide spray drift following applications to manage soybean aphid (*Aphis glycines* Matsumura) between mid-July and mid-September (Iowa State University 2016), which coincides with estimated peak larval abundance of the 2nd and 3rd monarch generations in Iowa (Pleasants 2015; Prysby and Oberhauser 2014; Nail et al. 2015). With expanding use of cover crops in Iowa, potential economic injury to corn from true armyworm (*Mythimna unipuncta* Haworth) has been observed, with foliar insecticide applications documented between mid-May and late June (Dunbar et al 2015), which overlaps with the 1st and 2nd generation larvae in Iowa. Monitoring studies designed to systematically document insecticide levels on milkweeds following foliar application are not available; however, models used by the U.S. EPA, such as AgDRIFT (USEPA 2017), indicate that spray drift exposure to milkweed up to 38m or more downwind is likely.

Monarch larvae could also be exposed to insecticides through ingestion of milkweed. Corn and soybean are typically planted with neonicotinoid-treated seed (Douglas and Tooker 2015), including 70% of soybean acres in Iowa (Hodgson et al. 2016). Chlorantraniliprole also is entering the market as a corn seed treatment option (Corn and Soybean Digest 2015). Imidacloprid, clothianidin, and thiamethoxam have moved into Iowa streams (Hladik et al 2014), presumably due to subsurface flow (Hladik et al 2017), which raises concerns that plants downslope of cropped fields could absorb neonicotinoids systemically. Several studies (Botias et al. 2016, 2015; David et al. 2016; Krupke et al. 2012; Long and Krupke 2016; Paola and Kaplan 2015; Pecenka and Lundgren 2015) indicate a variety of non-crop plants, including milkweed, in the margins of fields previously sowed with neonicotinoid-treated seeds can have detectable levels of imidacloprid, clothianidin, and thiamethoxam in leaves, pollen and nectar; although the frequency of detections and concentrations are highly variable.

There is a paucity of monarch toxicity data to interpret the significance of insecticide exposures due to spray drift or systemic uptake by milkweed. Consequently we are examining the toxicity of six representative insecticides: beta-cyfluthrin (pyrethroid, foliar uses); chlorpyrifos (organophosphate, foliar); imidacloprid (neonicotinoid, seed treatment and foliar); clothianidin (neonicotinoid, seed treatment); thiamethoxam (neonicotinoid, seed treatment and foliar); and chlorantraniliprole (anthranilic diamide, seed treatment and foliar) on monarch larvae, eggs, pupae and adults (Krishnan et al., 2017 a, b, c). Larvae, eggs and chrysalides are being treated topically to mimic exposure to a spray drift plume. Larvae and adults are also being exposed orally to mimic insecticide exposure from milkweed leaves (larvae - systemic uptake and spray drift deposition) and nectar of flowering plants (adults - systemic uptake), respectively. Dose response curves for mortality and growth/development are being compared to exposure levels obtained from spray drift models, on-going residue surveys, and existing ecological risk assessments and open literature studies. These analyses provide the means to assess risks at the habitat patch scale.

Predicting landscape-scale risk of insecticide exposure to monarch populations

Using instar-specific, 96-hour mortality dose-response curves for beta-cyfluthrin, chlorpyrifos, imidacloprid, thiamethoxam and chlorantraniliprole and estimates of exposure using EPA's AgDRIFT model, we have quantified spray drift risks at distances up to 38m downwind from treated fields following simulated aerial and ground applications (Krishnan et al 2017 a, b, c). In general, predicted mortality rates downwind were highest for chlorantraniliprole and beta-cyfluthrin and lowest for thiamethoxam and imidacloprid. To assess monarch population responses at a landscape scale we created a GIS layer that delineates areas within 38m of crop fields in Story County, Iowa. This layer was incorporated into the model described previously. A projection model is used to estimate survival from eggs to adults. Estimates of adult recruitment over a 10-year period are derived under three scenarios: a) no new milkweed planted within 38m of crop fields; b) milkweed patches placed within 38m of crop fields, with no insecticide exposure; and c) milkweed patches placed within 38m of crop fields with spray drift exposure. Different frequencies of economically-significant pest-pressure and associated foliar applications using typical wind direction and speed are used in the simulations (Bradbury et al. 2017; Grant et al 2017b). In future studies, the projection model will incorporate larval mortality rates based on systemic insecticide uptake in milkweed downslope of fields planted with treated corn or soybean seeds. Estimates of landscape-scale adult recruitment under varying spatial-temporal pest management scenarios provide the means to evaluate monarch conservation costs and benefits of establishing habitat in areas potentially exposed to insecticides.

Summary

The USFWS is committed to make a decision regarding whether to list the monarch butterfly as a threatened species by June of 2019. On-going research is enhancing the scientific basis for supporting monarch conservation practices by elucidating relationships between landscape-level habitat patterns and monarch movement, survival, reproduction, and development in the context of pest management practices. Coordinated research, extension and outreach provides a foundation for advancing and implementing a proactive, science-based conservation program that will enhance North American recovery of the monarch while maintaining efficient agriculture and livestock production.

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