IMPACTS OF AGRICULTURAL PESTICIDES AND PERSISTENT ORGANIC POLLUTANTS ON NEOTROPICAL MIGRATORY BIRDS DURING MIGRATION

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PROJECT PLAN

Objectives

The overall objective of this study is to examine seasonal patterns of contaminant accumulation in Neotropical migratory songbirds and in addition, evaluate genetic damage and cholinesterase (ChE) activity as biomarkers of exposure to contaminants. The potential for exposure differs among feeding guilds due to specific ways in which contaminants move through food webs. This project also aims to evaluate the diet and trophic position of migratory birds. Specifically, the research objectives that will be addressed include:

I. Determine accumulation and seasonal variation of contaminants in Neotropical migratory songbirds.
II. Assess the sublethal impacts of contaminants on migratory birds by evaluating genetic damage and cholinesterase activity.
III. Evaluate the method of using dried blood spots as a biomonitoring tool to measure cholinesterase activity.
IV. Determine if there are significant changes in diet using stable isotopes of C and N during migration.

Location of Project

Sample sites will be near agricultural areas located in Texas, Yucatan, Mexico, and Costa Rica. Samples from Texas will be collected during the fall migration (September-October). Samples will also be collected while migrants are on their wintering grounds (December-January) at sites located in Mexico and Costa Rica.

Methods

All animal procedures in this study are approved by the Texas A&M University Institutional Animal Care and Use Committee. This study will use the yellow-rumped warbler (*Setophaga coronata*), a fairly common and widely distributed insectivorous bird, as an indicator species. If yellow-rumped warblers are not present in the area, alternative species will be collected including, the chestnut-sided warbler (*Setophaga pensylvanica*), yellow warbler (*Setophaga petechia*), Kentucky warbler (*Geothlypis formosa*), mourning warbler (*Geothlypis philadelphia*), Tennessee warbler (*Vermivora peregrina*), Wilson's warbler (*Cardellina pusilla*), American redstart (*Setophaga ruticilla*), prothonotary warbler (*Protonotaria citrea*), ovenbird (*Seiurus aurocapilla*), hooded warbler (*Setophaga citrina*), and black and white warbler (*Mniotilta varia*). Resident birds will be used as a reference species to the yellow-rumped warbler and include, the Carolina wren (*Campylorhynchus brunneicapillus*), resident flycatchers (*Empidonax species*), Yucatan flycatcher (*Myiarchus yucatanesis*), dusky

capped flycatcher (*Myiarchus tubericulifer*), and brown crested flycatcher (*Myiarchus tyrannulus*). All birds that will be captured and analyzed in this study will be listed in the category of least concern under the International Union for Conservation of Nature (IUCN) Red List of Threatened Species. A total of 30 (15 migrant and 15 resident) birds will be collected per sampling event.

Birds will be captured using 12 m mist nets and removed immediately after being detected in the net. Approximately 0.2-0.5 ml of blood will be collected from the jugular vein with a 1 ml syringe and a 26 ½ gauge needle for analysis of genetic damage and ChE activity. Blood will be transferred to hepranized capillary tubes to prevent coagulation. Two to three drops of blood will be placed in a second capillary tube and placed on dry ice until they are transferred to the lab for storage and analysis of genetic damage. One or two drops will be placed onto Whatman blood stain cards and then stored in a plastic bin containing a desiccant to thoroughly dry (~24 hr). The dried blood spots (DBS) will then be enclosed in glassine weighing paper and sealed in an envelope containing a tablespoon of drierite until further analysis. The remaining blood contained in the capillary tubes will be centrifuged in the field and the plasma frozen on dry ice until taken to the lab. The DBS on the blood stain cards and the plasma will be used to analyze ChE activity.

After blood collection, birds will be euthanized using thoracic compression. The first and second primary feathers from each wing will be removed, wrapped in aluminum foil, and placed in an individual envelope for stable isotope analysis. Carcasses will be weighed and stored on ice temporarily until they can be placed in a freezer at -20 or -80 °C. Bird carcasses will be analyzed for organochlorines (OCs) and brominated flame retardants (BFRs) at the Geochemical and Environmental Research Group, at Texas A&M University, using procedures described elsewhere [1, 2, 3]. The plasma and dried blood spots stored on filter paper will be analyzed for cholinesterase activity using methods described previously [4, 5]. In addition, reactivation analysis of OP-inhibited ChE will be used as an adjunct to baseline data for ChE activity using the 2-pyridinealdoxime methiodide (2-PAM) method. Flow cytometry will be used to measure genetic damage from blood samples stored in cryogenic vials, using methods described previously [6].

All statistical analysis of the data will be carried out using JMP 9.0 software. An ANCOVA will be used to determine differences in contaminant concentrations, cytogenetic damage, and ChE activity for migrant and resident birds during the different collection times (fall, winter and spring) with sex and species as covariates. A methods comparison for measuring ChE activity between plasma and DBS from filter paper will be conducted using Pearson correlations.

Anticipated Benefits

Understanding how pollutants impact migratory songbirds is critical for their protection and conservation. Information from this study can be used by other researchers, wildlife managers and conservation agencies in conducting risk assessments of avian wildlife and incorporating this knowledge further into conservation efforts. Additionally, a better comprehension of how contaminants impact migratory birds can facilitate policy makers and managers in making decisions regarding the use and regulation of pesticides and other hazardous chemicals.

In addition, current knowledge on the use and accumulation of BFRs in Latin America is limited. Determining the presence and levels of BFRs in migratory birds and resident birds sampled in Mexico and Costa Rica would provide the first baseline data for evaluation of BFR impacts on migratory songbirds in Latin America and future monitoring efforts.

This project will also provide the first field data on the use of DBS to measure ChE activity in songbirds. To our knowledge there are no publications that have evaluated this method as a biomonitoring tool in the field for migratory songbirds. A comparison of the accuracy for using DBS on filter paper relative to plasma will provide information required to assess its efficacy and potential usefulness in future studies.

This project also fosters collaborative work with universities and researchers in other countries, particularly in Mexico and Costa Rica. Since migratory birds cross geographical, political and cultural boundaries they are a shared resource and therefore a shared concern. Increased collaboration with other countries could strengthen cooperation on the development and improvement of environmental laws, regulations, policies and practices in regards to migratory bird conservation and toxic substance usage.

This project will facilitate in elucidating the impacts that pesticides and persistent organic pollutants may have on migratory songbirds. A better understanding of how contaminants impact migratory birds will also aid in their protection and conservation. Migratory bird conservation is important not only because they act as sentinels of environmental health for human populations, but also because they provide many ecosystem services. Ecosystem services are natural processes that benefit humans and generally fall into four types of services; provisioning, regulating, cultural, and supporting services [7]. As members of ecosystems they play important roles such as predators, pollinators, scavengers, seed dispersers, pest controllers, and ecosystem engineers [7]. Migratory birds are especially important since they link ecosystems processes and fluxes that are separated by space and time [7].

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