Environment and Natural Resources Trust Fund
2016 Request for Proposals (RFP)

Project Title: **ENRTF ID: 035-B**

Neonicotinoid Insecticide Effects on Aquatic and Soil Communities

Category: B. Water Resources

Total Project Budget: $ 412,000

Proposed Project Time Period for the Funding Requested: 3 years, July 2016 to June 2019

Summary:

Neonicotinoid insecticide breakdown products produced in water and plant leaves will be identified and their toxicity to soil and aquatic species tested to allow informed use and management.

Name: William Arnold

Sponsoring Organization: U of MN

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Location

Region: Statewide

County Name: Statewide

City / Township:

Alternate Text for Visual:

Neonicotinoid insecticides and their breakdown products produced in water and plants may be toxic to aquatic and soil species

_____ Funding Priorities _____ Multiple Benefits _____ Outcomes _____ Knowledge Base

_____ Extent of Impact _____ Innovation _____ Scientific/Tech Basis _____ Urgency

_____ Capacity Readiness _____ Leverage _______ TOTAL ______%

PROJECT TITLE: Neonicotinoid Insecticide Effects on Aquatic and Soil Communities

I. PROJECT STATEMENT

Neonicotinoid insecticides were introduced in the 1990s and now represent 25% of global insecticide use. Current estimates for the U.S. are that neonicotinoids are used on 95% of corn and half of sugar beets and soybeans, all important Minnesota crops. Neonicotinoids are also used on food crops, including seeds purchased by home gardeners. These insecticides are applied as seed dressings, so a portion of the insecticide is taken up by the plant, and the remainder enters the soil and water. Thus, neonicotinoid compounds have been detected in soil, surface water, and groundwater, but their persistence in the environment and potential toxic effects are not fully understood. Reactions of neonicotinoids in water or in sunlight will give rise to new chemicals of similar chemical structure and unknown toxicity. Because neonicotinoids are applied as seed dressings and taken up by plants, water/solar driven reactions within the plant itself must also be explored. While the potential toxic effects of neonicotinoids on honey bees and birds are known, toxic effects on aquatic or soil species have received less attention. *Consequently, new studies regarding the environmental movement, fate, and toxicity of neonicotinoids are urgently needed to determine any potential effects in Minnesota waters and to develop guidelines for safe use of neonicotinoids*. The hypothesis to be tested by this project is that the neonicotinoid breakdown products formed in water and plants will have residual toxicity. The goals of the project are to:

- Identify reaction products from neonicotinoids in water in the presence of natural trace metals and minerals,
- Identify reaction products in water and simulated plant leaves upon neonictonioid exposure to sunlight,
- Assess toxicity of neonictinoids to soil and aquatic species before and after reaction in water and plants, and
- Disseminate the findings to stakeholders, regulators, and the public.

Data on neonicotinoid use and environmental detection in Minnesota are limited. Recent surveys by the Minnesota Department of Agriculture, however, have shown that imidacloprid, the most widely used neonicotinoid, is commonly applied to Minnesota soybean crops and thus neonicotinoids are likely present in Minnesota waters. Hydrolysis (water driven transformation) and photolysis (solar driven transformation) are two important pathways for pollutant degradation. The transformation of neonicotinoids in water shows that rates are slow at the pH conditions of natural waters. Other system components, however,such as natural trace metals and minerals (which are key plant nutrients), may increase transformation rates via hydrolysis and photolysis and lead to previously unidentified reaction products. For neonicotinoids, which are taken up by plants, reaction *within the plant leaves* is also potentially important. This will be tested using artificial plant leaves (wax films). *The results of this work will have direct impacts on management of neonicotinoid use and the environmental health of Minnesota's waters.* More specifically, these studies will provide evidence for which neonicotinoids are transformed most quickly in surface waters, if transformation in plant leaves occurs, and whether the breakdown products have residual toxic activity for soil and aquatic species. This information will enable management and regulatory decisions for these compounds.

II. PROJECT ACTIVITIES AND OUTCOMES

Activity 1: Neonicotinoid reaction in water: role of trace metals and minerals **Budget: $118,000**

This activity will quantify reaction rates and characterize transformation products of reactions of neonicotinoids in the presence of natural trace metals present in soil that are critical for plant growth (copper, iron, calcium, etc.) and soil minerals (e.g., clays, iron oxides). Three neonicotinoids will be tested with variables including pH, temperature, trace metals, and minerals. This will require 200 reactors (approximately 2000 samples) to be run. Product identification, which is time consuming, will be performed for each reactor.

Outcome	Completion Date
1. Rates of neonicotinoid reaction in water	12/31/16
2. Rates of neonicotinoid reaction in water with natural trace metals	6/30/17
3. Rates of neonicotinoid reaction in water with natural minerals	12/31/17
4. Identification of reaction products	12/31/18

Activity 2: Solar effects on neonicotinoids in water and plants **Budget: $113,000**

Photolysis experiments will be performed in pure water solutions using artificial sunlight (which provides control and reproducibility) as an energy source. Validation of results will use natural sunlight and natural waters. Following these experiments, photolysis rates in "artificial leaves" (cuticular wax films) will be investigated. This method has been used in recent pesticide transformation studies to mimic the chemical environment of a plant leaf. The waxy leaf environment may lead to different transformation rates and products. Again, three neonicotinoids will be used for experiments, with a total of at least 100 experiments (1000 samples) required. Finally, transformation products will be identified for reactions in water and "artificial leaves" to find any structural or comparative differences in product compositions.

Outcome	Completion Date
1. Rates of solar-driven neonicotinoid reaction in water	6/30/17
2. Rates of solar-driven neonicotinoid reaction in "artificial leaves"	6/30/18
3. Identification of products of aqueous and "artificial leaf" photolysis	12/31/18
4. Dissemination of Activity 1 & 2 findings via open access journal publication(s)	12/31/18

Activity 3: Toxicity of transformation products to soil and aquatic species **Budget: $181,000**

Toxicity tests will be performed with the neonicotinoid insecticides, the reaction mixtures from Activity 1 and 2, and, when possible, with individual identified/isolated transformation products. The potential impacts on soil and aquatic organisms need to be explored to fully evaluate impacts of neonicotinoids. The tests will use springtails (a soil arthropod commonly used in assessment of environmental contaminants), mosquito larvae, and tadpoles from three native frog species that breed in vernal pools, often impacted by agricultural runoff. This will provide a representation of a range of species native to Minnesota. For each reaction condition, a minimum of seven doses are needed for each species tested (up to 2500 total experiments). Efforts will focus on reactions where the reaction product is likely to have residual activity based on its structure.

Outcome	Completion Date
1. Quantify levels of neonicotinoids and breakdown products toxic to springtails	6/30/18
2. Quantify levels of neonicotinoids and breakdown products toxic to mosquito larvae	12/31/18
3. Quantify levels of neonicotinoids and breakdown products toxic to tadpoles (3 species)	6/30/19
4. Dissemination of findings via open access journal publication(s)	6/30/19

III. PROJECT STRATEGY

A. Project Team/Partners: The project will be led by William Arnold (U of MN, Department of Civil, Environmental, and Geo- Engineering), who will be responsible for Activities 1 and 2, and Ann Fallon (U of MN, Department of Entomology) who will be responsible for Activity 3. The team will consist of two graduate and two undergraduate student researchers. Arnold is an expert in chemical reactions of pollutants in water, and Fallon is an expert in insecticide toxicology, insecticide resistance, insect physiology and molecular biology.

B. Project Impact and Long-Term Strategy: This project will provide an understanding of neonicotinoid interactions with the natural environment and their potential transformation pathways. Results of the proposed work will provide a strong basis for evaluating the persistence and toxicity of neonicotinoids thus allowing for informed use, management, and, if needed, regulatory actions. Additionally, these studies will provide the first evidence of neonicotinoid hydrolysis and photolysis beyond simple baseline experiments in pure water solutions. The results will be disseminated via open-access scientific literature and publically available reports.

C. Timeline Requirements: The project will be completed in a three year period. The experiments to gather sufficient data to discern neonicotinoid hydrolysis and photolysis trends, the method development for product identification, and the toxicity testing are all time consuming and require attention to detail and replication.

2016 Detailed Project Budget

Project Title: Neonicotinoid Insecticide Effects on Aquatic and Soil Communities

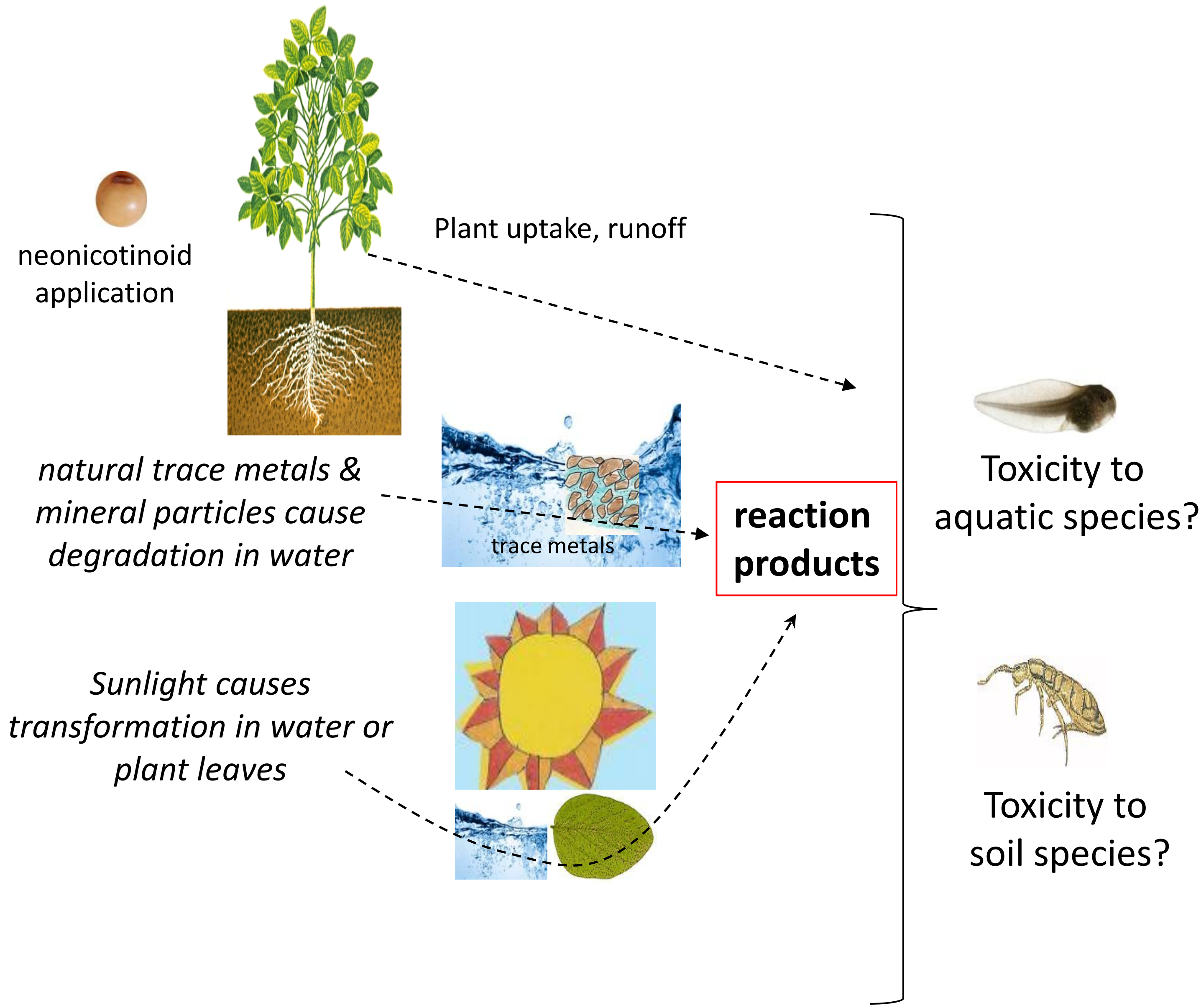
IV. TOTAL ENRTF REQUEST BUDGET 3 years

BUDGET ITEM	AMOUNT
Personnel: Prof. William Arnold (PI, 8% time per year over three years, salary 74.8% of cost, fringe benefits 25.2% of cost). Project supervision, design of experiments and data analysis of Activities 1 &2, supervision of graduate and undergraduate students and project reporting.	$ 58,550
Personnel: Prof. Ann Fallon (PI, 4% time per year over three years, salary 74.8% of cost, fringe benefits 25.2% of cost). Project supervision, design of experiments and data analysis of Activity 3, supervision of graduate and undergraduate students and project reporting.	$ 28,450
Personnel: Graduate student #1 (50% time during academic year, 50% time in summer in Y1 and Y2; 25% time in Y3; 56% salary, 33% tuition, 11% fringe benefits). Hydrolysis and photolysis experiments, development of analytical methods, identification of reaction products, data analysis and interpretation.	$ 119,050
Personnel: Graduate student #1 (50% time during academic year, 50% time in summer in Y1 and Y2; 25% time in academic Y3, 45% time in summer); 56% salary, 33% tuition, 11% fringe benefits). Rearing organisms for toxicity studies, toxicity studies, data analysis and interpretation.	$ 119,050
Personnel: Undergraduate students (100% time. In Y1 and Y2, two students for 40 hr/wk in the summer (10 weeks) and 10 hours per week for one semester (15 weeks). In Y3, summer only. 100% salary). Assist graduate students with all laboratory activities.	$ 45,000
Supplies: Supplies (chemical standards, chemical reagents for fate experiments and toxicity assays, necessary glassware, instrument/analytical time for product identification, solvents, consumable supplies, laboratory notebooks, software licenses; $22,900 total). Analytical time for product identification is a major portion of supply costs ($4,000 per year). Operating costs for laboratory instruments required for analyses and experiments; costs portioned based on usage by project ($9,000 total)	$ 31,900
Travel: charges and univeristy vehicle rental charges for trips to water samples. Hotel/meal charges if overnight stay required. Attendence for students at local conferneces to disseminate project findings to agriculture and environmental interests.	$ 4,000
Additional Budget Items: Publication charges to make published journal articles (four) immediately available via open access to maximize data availability and dissemination.	$ 6,000
TOTAL ENVIRONMENT AND NATURAL RESOURCES TRUST FUND $ REQUEST =	$ 412,000

V. OTHER FUNDS

SOURCE OF FUNDS	AMOUNT	Status
Other Non-State $ To Be Applied To Project During Project Period: N/A	$ -	
Other State $ To Be Applied To Project During Project Period: N/A	$ -	
In-kind Services To Be Applied To Project During Project Period: Because the project is overhead free, laboratory space, electricty, and other facilities/adminstrative costs (52% of direct costs excluding permanent equipment and graduate student academic year fringe benefits) are provided in-kind	$ 167,473	secured
Funding History: N/A	$ -	
Remaining $ From Current ENRTF Appropriation: no current project directly applicable	$ -	

Neonicotinoid Insecticides and Their Breakdown Products May Be Toxic to Non-Target Species



Outcomes

1. Understand neonicotinoid processing
2. Identify water/plant reaction products
3. Recognize toxic effects
4. Inform use, management, regulation

Project Manager Qualifications and Organization Description

William A. Arnold
Joseph T. and Rose S. Ling Professor and Associate Head
Department of Civil, Environmental, and Geo- Engineering, University of Minnesota

B.S., Chemical Engineering, 1994, Massachusetts Institute of Technology, Cambridge, MA.
M.S., Chemical Engineering, 1995, Yale University, New Haven, CT.
Ph.D., Environmental Engineering, 1999, The Johns Hopkins University, Baltimore, MD.

Dr. William Arnold will be responsible for overall project coordination and supervision and design of the hydrolysis and photolysis studies. He has been studying the fate of pharmaceutical and pesticide compounds in aquatic environments for sixteen years. As part of these studies, he has determined the transformation rates and identified reaction products of numerous compounds. Work has focused on the phototransformation of pesticides in prairie wetlands, pesticides losses in soils, and antibiotic fate in surface waters. He has published over twenty peer-reviewed papers on pesticide and pharmaceutical fate since 2003, and he is the co-author of a textbook on water chemistry published in 2011. Dr. Arnold is a Resident Fellow of the University of Minnesota Institute on the Environment, an Associate Fellow of the Minnesota Supercomputing Institute, and a member of the graduate faculty in Water Resources Science. He won the *Arcadis/Association of Environmental Engineering and Science Professors Frontier in Research Award* in 2012 and the University of Minnesota College of Science and Engineering *George W. Taylor Award for Distinguished Research* in 2011.

Ann Fallon
Distinguished McKnight University Professor
Department of Entomology, University of Minnesota

B.A., Biology, 1972, University of Connecticut, Storrs, CT.
M.S., Biology, 1973, Yale University, New Haven, CT.
Ph.D., Biology, 1976, Queen's University, Kingston, ON.

Dr. Ann Fallon will be responsible for supervision and design of the toxicology studies. She has extensive experience in insect physiology, biochemistry, and molecular biology, and she has investigated genetic processes involved in insecticide resistance. Using mosquitoes as a model system, she has explored physiological processes related to the insect repellent DEET, and most recently she has developed improved protocols for ecotoxicological studies with the springtail, *Folsomia candida*, a standard soil arthropod used for evaluation of environmental toxins. She has published over 120 papers in peer reviewed journals and has served on grant review panels for the National Institutes of Health, United States Department of Agriculture, and National Science Foundation. She is a Distinguished McKnight University Professor (1999). She received the University of Minnesota Distinguished Women Scholars Award in Science and Engineering (2009) and a FAME (Faculty Award for Mentorship in Entomology) Award (2014). Her research has been funded by the National Institutes of Health and US Department of Agriculture, and she has trained 12 MS and 15 PhD students, as well as numerous undergraduates.

Organization Description
The University of Minnesota is one of the largest, most comprehensive, and most prestigious public universities in the United States (http://twin-cities.umn.edu/about-us). The laboratories and offices of the PI contain the necessary fixed and moveable equipment and facilities needed for the proposed studies.