

**Environment and Natural Resources Trust Fund
2010 Request for Proposals (RFP)**

LCCMR ID: 227-G

Project Title:

Characterization of Environmentally Sensitive Aquatic Insect Communities

LCCMR 2010 Funding Priority:

G. Creative Ideas

Total Project Budget: \$ \$82,118

Proposed Project Time Period for the Funding Requested: 2 years, 2010 - 2012

Other Non-State Funds: \$ \$0

Summary:

Faculty and students from Augsburg College propose to conduct a 2-year project identifying insect species in the Rice Creek Watershed through DNA analysis. Results are useful in water management decisions.

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Sponsoring Organization: Augsburg College

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Location:

Region: Metro

County Name: Anoka

City / Township: Shoreview, Fridley, Centerville

_____ Knowledge Base	_____ Broad App.	_____ Innovation
_____ Leverage	_____ Outcomes	
_____ Partnerships	_____ Urgency	_____ TOTAL

MAIN PROPOSAL

I. PROJECT STATEMENT

Faculty and students from Augsburg College propose to conduct a 2-year project identifying insect species in the Rice Creek Watershed through DNA analysis. Currently, there is no project such as this in Minnesota. Aquatic insects are surveyed to assess stream water quality and environmental stress. Insect surveys are useful to assess watersheds because there are many kinds of insects in streams, and they vary in tolerance to environmental stress. The variety of insects can be used as a measurement of watershed "health". However, there are limitations to the methods in current use. This project will address these limitations. First, many insects have multiple juvenile life stages in streams and it can be extremely difficult to identify the species. Therefore, the insects are grouped into larger categories such as mayflies or caddisflies. However there are over 50 species of mayflies and over 250 species of caddisflies in Minnesota. Species of insects vary in their sensitivity to environmental stressors, therefore making the determination of water health difficult. Second, the current identification methods require expertise and are time consuming, expensive, and generally impractical. This project will overcome these limitations on stream insect identification.

We will use approaches that were recently implemented in an international effort to identify species based on their DNA. The Barcode of Life Initiative (BOLI) combines DNA analysis, international databases and the Internet to assign "DNA barcodes" to each species. DNA barcodes are determined by examining a particular feature of single organisms. The DNA organization (DNA sequence) is unique for every species. The term, DNA barcode, is used by BOLI for this feature of DNA because it is reminiscent of product barcodes in stores. Augsburg College supported pilot studies, and we have demonstrated feasibility for the work. We now wish to expand the effort to study insect communities in Rice Creek.

The work is broadly applicable to the 81 major watersheds in Minnesota. The method is cost effective, and does not require highly skilled technicians. Hundreds of organisms can be analyzed in relatively short time, and exact species identification is achieved. DNA barcode results will allow for more refined analysis of specific environmental stresses and for better and faster responses to stress. Results are added to the database, which will be accessible to Minnesotans, and will allow for better and faster decisions about stream health. Other colleges and volunteer groups in Minnesota can replicate the project. An added benefit is that students interested in conservation and the environment become directly involved in the field and laboratory studies.

II. DESCRIPTION OF PROJECT RESULTS

Result 1: Species identification of aquatic insects in Rice Creek by DNA

Budget: \$

62,251

A. Insect Collection: Insects will be collected each year from Rice Creek and tributary sites at three-week intervals from May through October using EPA protocol. Insects will be evaluated in the laboratory to characterize site biodiversity, preserved and recorded for DNA studies. Identification keys will be used to assign the Order or Family taxonomic level, while DNA analysis will identify the species.

B. Preparation and analysis of DNA: We will start with environmentally sensitive groups of insects including mayflies, stoneflies and caddisflies. As this first objective progresses, we will analyze the less sensitive groups of insects. Steps for DNA characterization are 1) extraction of DNA from the specimen 2) prepare multiple copies of a small part of the extracted DNA using the polymerase chain reaction (PCR) 3) clean up of the DNA and determine the order of it's A-T-G-C subunits using equipment at the University of Minnesota.

C. Barcode analysis: DNA barcodes are sent to BOLI over the Internet, and species identification is immediately fed back. Each DNA barcode is added to the growing database, which itself is available to the public. DNA barcodes for which no species is assigned are used to build new species files. We anticipate that we will encounter some DNA barcodes not in the database since we are working in a niche area. In this situation, it will be necessary to use detailed taxonomic keys to identify the organism at the species level, and the adult forms of the insects (rather than juvenile forms) may be required.

Deliverable**Completion Date** June 30, 2012**1. Specific, measurable deliverable (outcome) #1**

We will document 50 to 75 species by DNA barcodes. Results will contribute new results to the database useful for other researchers. Six students will participate, and become knowledgeable researchers.

Result 2: Analysis of Rice Creek habitat at insect collection sites**Budget: \$ 19,867**

Environmental conditions and water quality are important to any assessment and will be included in this project. Water chemistry will be determined each year according to the wadable stream chemistry manual. Some physical properties of the stream water, such as dissolved oxygen; pH, conductivity, temperature and flow rate will be monitored on-site with hand-held sensors. Stream samples will be collected for lab analysis, including biological oxygen demand (BOD), and iron, sulfate, calcium, chlorine, ammonia, copper, chromium, manganese, phosphate, nitrogen and nitrates.

Deliverable**Completion Date** June 30, 2012**1. Specific, measurable deliverable (outcome) #1**

Conditions of the collection sites, and water chemistry will be noted and summarized each year. Results give species level information relative to environmental conditions, and will help future studies at other sites. Each year students prepare research reports and present results at appropriate conferences.

III. PROJECT STRATEGY

A. Project Team/Partners The project team consists of Dr. Ralph Butkowski in the Biology Department, who will participate in and manage the project full time during summer sessions, and 4 hours per week during school terms. Dr. Vivian Feng in the Chemistry Department will work half time during summer sessions. Two students will work full time for 10 weeks during summer sessions, and one student will work each semester during the school year. Students and Dr. Butkowski will conduct the fieldwork. Students will participate in all aspects of the project under guidance from Dr. Butkowski and Dr. Feng. Students will present results in poster sessions at Augsburg College and possibly at other local meetings.

B. Timeline Requirements Because we have established feasibility for the project, lag time should be minimal. About 2 months are required to obtain initial results, after which the results will come in continuously. We are proposing a two-year project to obtain representative and as complete analysis as possible. Frequent field samplings are required to obtain specimens during peak activity, which varies with species.

C. Long-Term Strategy The project has long term potential to map species in Minnesota waterways, and to study unique species' response to change. Funding of this project does not require a long-term commitment.

Project Budget

IV. TOTAL PROJECT REQUEST BUDGET (2 years)

BUDGET ITEM	AMOUNT
Personnel:	\$ -
Dr. Ralph Butkowski 2/9 time, or 2 months per year for 2 years Fringe benefits figured at 7.6%. Dr. Vivian Feng 1/9 time (one month) per year for 2 years. Benefits	\$ 39,201
2 summer students full time for 10 weeks for each of 2 years. \$4,000 stipend and 800 housing allowance eah for 2 years. No fringe benefits.	\$ 19,200
School term students: One student per semester for 10 hours per week over two years at \$1000/student	\$ 4,000
Contracts:	\$ -
University of Minnesota for DNA sequencing services. \$20 per sample x 50 samples x two years. (The number includes some anticipated repeats.)	\$ 2,000
Equipment/Tools/Supplies:	\$ -
Microcentrifuge to spin samples \$3600 Micro-spectrophotometer to measure DNA in small volumes \$3500 DNA extraction kits, 20 at \$160 = \$3200 PCR kits, 20 at \$145 = \$2900 Water analysis chemicals and supplies, \$2000 Plastic test tubes and reagent tubes, \$2000 Stream chest waders, 1 pair, \$145 Insect guides \$30 and books \$150 = \$180	\$ 17,525
Acquisition (Fee Title or Permanent Easements): N/A	\$ -
Travel: 12 collection trips over 2 years x 40 miles x 40c per mile	\$ 192
Additional Budget Items:	\$ -
TOTAL PROJECT BUDGET REQUEST TO LCCMR	\$ 82,118

V. OTHER FUNDS

SOURCE OF FUNDS	AMOUNT	Status
Other Non-State \$ Being Applied to Project During Project Period: None	\$ -	
Other State \$ Being Applied to Project During Project Period: N/		
In-kind Services During Project Period: N/A	\$ -	
Remaining \$ from Current Trust Fund Appropriation (if applicable): N/A		
Funding History: Augsburg College supported this project as a start-up in 2008 and 2009. Four students received stipends and housing allowance (\$19,200), the manager received \$4,000. Supplies budget was \$2,000 over the period.	\$ 25,200	

Characterization of environmentally sensitive aquatic insect communities

Project Manager and Organization

The project will be managed by Dr. Ralph Butkowski. He is assistant professor in the biology department at Augsburg College, where he teaches biology courses to non-science students, and upper division courses in biochemistry and immunology. In also he teaches the "Introduction to Research" course.

Dr. Butkowski has been teaching at Augsburg College since 2001. He is interested in environmental education, and in engaging students in research in environmental issues. He is a Minnesota Master Naturalist, and is involved in various Master Naturalist activities in the Twin Cities. For the past 3 years, he participated as a volunteer in the Stream Health Evaluation Project (SHEP), where he initially became familiar with the Rice Creek watershed and its features, and the opportunities to enhance water quality assessment. The proposed project does not overlap with SHEP, but may in the future, aid in refining its approaches.

Prior to coming to Augsburg, he was a Research and Development Manager at DiaSorin in Stillwater, Minnesota. At DiaSorin he was in charge of research and development of products to detect autoimmune disease. Previously he was a scientist at the University of Minnesota, and the University of Kansas. His field of academic research was autoimmune disease and protein chemistry. He has published numerous manuscripts and chapters in his field.

Augsburg College, founded in 1869, is a private college in Minneapolis affiliated with the Evangelical Lutheran Church in America. Together with a strong academic curriculum, the College offers experiential programs to enhance classroom learning. Augsburg offers an education that unites the liberal arts with the professional to prepare students for vocations in a global, multicultural society. The college is legally incorporated as a Minnesota non-profit corporation and has been continuously accredited by the North Central Association of Colleges and Schools since 1954.

On its compact urban campus, the Augsburg community includes approximately 3,800 students from 42 states, 40 foreign countries and 24 tribal nations/reservations. The primary focus of Augsburg is its undergraduate program offered in the more traditional day program and a large Weekend College Program for working adults. Students may choose from over 50 major areas of study, culminating in the Bachelor of Arts, Bachelor of Music and Bachelor of Science undergraduate degrees. Augsburg College also offers the Master of Arts in Leadership, the Master of Social Work, the Master of Arts in Nursing, the Master of Arts in Education, the Master of Business Administration, and the Master of Science in Physician Assistant Studies (the only Physician Assistant training program in Minnesota).

Many students majoring in the sciences participate in research with a faculty member serving as a guide and mentor. Undergraduate participation in research is an important aspect to their education as scientists.

