Environment and Natural Resources Trust Fund
2019 Request for Proposals (RFP)

Project Title: **ENRTF ID: 093-B**

Rapid Detection of Algal Toxins in Minnesota Lakes

Category: B. Water Resources

Sub-Category:

Total Project Budget: $ 599,051

Proposed Project Time Period for the Funding Requested: June 30, 2022 (3 yrs)

Summary:

In this project we will use novel environmental genomics techniques, coupled with citizen science sampling in order to develop a statewide rapid-alert system for harmful algae blooms.

Name: Andrew Bramburger

Sponsoring Organization: U of MN - Duluth

Title:

Department: Natural Resources Research Institute

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Web Address

Location

Region: Statewide

County Name: Statewide

City / Township:

Alternate Text for Visual:

Samples will be analyzed for algal toxins and predictor genes in 200 lakes. Citizen science samples will be analyzed for calibrated predictor genes to produce a statewide cyanotoxin risk map.

_____ Funding Priorities	_____ Multiple Benefits	_____ Outcomes	_____ Knowledge Base
_____ Extent of Impact	_____ Innovation	_____ Scientific/Tech Basis	_____ Urgency
_____ Capacity Readiness	_____ Leverage	_____ TOTAL	_____%
____ If under $200,000, waive presentation?			

PROJECT TITLE: Rapid Detection of Algal Toxins in Minnesota Lakes

I. PROJECT STATEMENT

Recently, harmful algae blooms (HABs) and associated threats to human and ecosystem health have increased both globally and within Minnesota. However, projects previously funded by LCCMR have focused largely on changing distribution patterns of single HAB species or facilitating collaborations among HAB researchers in the state and have done little to increase our understanding of HAB biology or to protect the health of Minnesotans and the integrity of Minnesota's freshwater resources. As such, the proposed project will quantify the distribution and abundance of multiple HAB-forming species in MN inland lakes and evaluate algal toxin exposure risk at a statewide scale in order to achieve the following principal objective:

To develop a robust, cost-effective, rapid system for monitoring statewide algal toxin exposure risk in Minnesota.

In order to achieve our project objectives, we will:

1. **Use cutting edge genetics and toxin characterization techniques in 200 focal lakes statewide in order to identify genomic indicators of high algal toxin concentrations and produce a gene-based model of toxin exposure risk.**
2. **Train a network of citizen science volunteers to collect genetic samples from algae occurrences in a large number (~2,000) of Minnesota lakes in order to apply risk model at a statewide scale.**

Recent advances in environmental genomics technology, coupled with the abundance and diversity of inland lakes in Minnesota, provide a unique opportunity to produce a cost-effective and widely applicable long-term monitoring tool that will be effective for protecting Minnesotans from risks associated with HABs at a statewide scale.

II. PROJECT ACTIVITIES AND OUTCOMES

Activity 1: *Citizen scientist recruitment and outreach*

Citizen scientist volunteers will be recruited through social media, public outreach events, and through partner organizations. Volunteers will be equipped with sampling kits and trained in sampling technique. We will work with project partners to produce HAB-related outreach materials to be distributed to stakeholders. Targeted avenues for outreach include publication of an article in the MN Sea Grant publication, The Seiche; a podcast episode on The Sea Grant Files; and participation in MN Sea Grant and UMN Extension outreach activities including the state fair booth, as well as specific products and outcomes described below.

ENRTF BUDGET: $140,443

Outcome	Completion Date
1. Prepare 2,000 Citizen Scientist sampling kits for deployment in lakes statewide.	July 15, 2019
2. Develop 2 online awareness/sampling training videos (YouTube).	July 15, 2019
3. Host 3 regional training events (proposed in southern MN, Brainerd, and Ely) to discuss HABs, distribute sampling kits (500 for year 1), and train volunteers.	July 31, 2019
4. Host 3 regional follow up/outreach and training events (proposed in southern MN, Brainerd, Ely), distribute additional sampling kits (1,500 for year 2), and to train volunteers on proper sampling techniques.	May 31, 2020
5. Produce outreach materials including visual guide to potentially harmful algae (1), instructions for reporting suspicious blooms (1), initial report providing feedback and results to citizen scientists and snapshot of algal toxin risk in Minnesota lakes (1).	June 30, 2022

Activity 2: *Determination of genomic indicators of cyanotoxin exposure risk and construction of statewide risk map*

We will characterize water quality conditions and sample water and environmental DNA annually in 200 Minnesota lakes. Metagenomics and toxin signatures of focal lake samples will be fully characterized using cutting-edge techniques. Citizen scientist samples will be scanned for presence of indicator genes. Gene-based risk models will be constructed and calibrated using focal lake data and applied to citizen science data.

ENRTF Budget: $458,608

Outcome	Completion Date
1. Focal lake genetic and toxin sampling and analysis (Round 1; 100 samples)	Nov 30, 2019
2. Analysis of citizen scientist sample kits (Indicator gene scan; Round 1; 500 samples)	Jan. 31, 2020
3. Focal lake genetic and toxin sampling and analysis (Round 2; 100 samples)	Nov. 30, 2020
4. Analysis of citizen scientist sample kits (Indicator gene scan; Round 2; 1,500 samples)	Jan 31, 2021
5. Quality control of genetic, toxin, and water quality data, data analysis construction/calibration of toxin risk model for application to citizen science data.	June 30, 2022

III. PROJECT PARTNERS:

A. Partners receiving ENRTF funding N/A

B. Partners NOT receiving ENRTF funding

Name, Title, Affiliation	Role
Minnesota Coalition of Lake Associations	Participant in / facilitator of citizen science activities
White Iron Chain of Lakes Association	Participant in / facilitator of citizen science activities
Riley Purgatory Bluff Creek Watershed	Participant in / facilitator of citizen science activities
Minnesota Pollution Control Agency	Participant in / facilitator of citizen science activities

IV. LONG-TERM- IMPLEMENTATION AND FUNDING:

Upon successful demonstration of a genomics/citizen science-based monitoring network, we will construct a near real-time cyanotoxin exposure risk map that may be accessed via mobile app. Potential funders and users for this initiative include the Minnesota Pollution Control Agency. Further, several lake associations and watershed districts have expressed interest in participating in ongoing citizen-science monitoring and employing this technique as a fee-for service HAB monitoring program in their jurisdictions. UMN Extension Citizen Science have expressed interest in collaborating with the project team to develop and implement best management practices for citizen science activities of this monitoring program after the LCCMR project end date.

V. TIME LINE REQUIREMENTS:

Three years, from July 2019 through June 2022.

VI. SEE ADDITIONAL PROPOSAL COMPONENTS:

A. Proposal Budget Spreadsheet
B. Visual Component or Map
C. Parcel List Spreadsheet
D. Acquisition, Easements, and Restoration Requirements
E. Research Addendum (not required at proposal stage)
F. Project Manager Qualifications and Organization Description
G. Letter or Resolution
H. Certified Audit or 990 Tax Information

2019 Proposal Budget Spreadsheet

Project Title: Rapid Detection of Algal Toxins in Minnesota Lakes

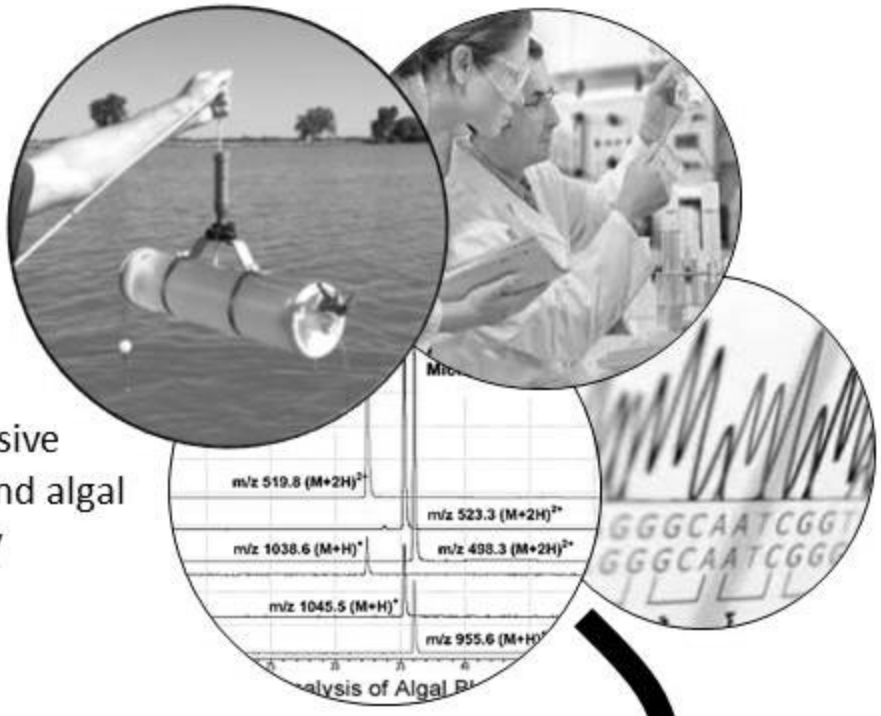
IV. TOTAL ENRTF REQUEST BUDGET 3 years

BUDGET ITEM	AMOUNT
Personnel:	**$ 402,608**
Andy Bramburger, Principal Investigator: $47,613 (fringe rate 33.5%); 15% FTE each year for 3 years. Dr. Bramburger will be responsible for leading algal taxonomy and ecology aspects of this program, and coordinating overall program activities.	
Cody Sheik, Co-Investigator: $33,499 (fringe rate 33.5%); 100% FTE 1 SUM months each year for 3 years. Dr. Sheik will lead molecular analysis for this project. His is uniquely qualified to perform these duties.	
Chris Filstrup, Co-Investigator: $33,432 (fringe rate 33.5%); 15% FTE each year for 3 years. Dr. Filstrup will coordinate field activities and site selection. His expertise in HAB monitoring is vital to this project.	
Kathryn Schreiner, Co-Investigator: $34,709 (fringe rate 33.5%); 100% FTE 1 SUM months each year for 3 years. Dr. Schreiner will lead the toxin quantification activities for this project. She is uniquely qualified to conduct these analyses.	
Julia Halbur, Lab Technician (Genomics Sample Prep): $87,588 (fringe rate 27.2%); 60% FTE each year for 3 years. Ms. Halbur will perform all genomics extractions for the program. The high sample throughput of this project warrants a dedicated technician.	
Marte Kitson, Minnesota Sea Grant, Outreach Coordinator: $17,161 (fringe rate 33.5%); 10% FTE each year for 3 years. Ms. Kitson will coordinate the outreach activities of this project. She will facilitate access to the outreach capabilities of Minnesota Sea Grant.	
Elizabeth Alexson, Lab Tech: $17,878 (fringe rate 27.2%); 10% FTE each year for 3 years	
Graduate Student: $130,728 (fringe rate 15%) and tuition reimbursement; 50% FTE each year for 3 years	
Professional/Technical/Service Contracts:	**$ -**
Equipment/Tools/Supplies:	**$ 86,005**
YSI EX02 Water Quality Sonde used to quantify water quality ($19,560). Quote from YSI. Will follow University policy for selection of vendor. State-of-the-art water quality sonde for measurement of multiple parameters. Equipment will be retained for use in future HAB research and monitoring, and shared with another LCCMR-funded HAB progam if both programs receive funding (see support letters).	
Sample Processing and Analysis Supplies ($32,615): Phytoplankton sample processing ($7,050): beakers (case of 100 = $400), centrifuge bottles (case of 1,000 X 2 = $1,500), slides (case of 1000 X 2 = $800), coverslips (case of 1,000 X 2 =$450), reagents and preservatives (HNO3 X 8L = $1,200, H2O2 X 16L = $700, K2Cr2O7 X 8L = $700, mounting media X 100 mL = $300, formalin X 3L = $300, acetic acid X 2L = $200, iodine X 500 mL = $300, KI crystals = $200). Purchase half Year 1 and Year 2. Molecular analysis supplies ($14,765): primers ($400), PCR MasterMix solution ($10,200 Agarose ($1150, TBE Bugger ($200), PCR plates ($895), PCR film ($420), Multi-channel pipettes ($1,500) Purchase half year 1 and half year 2 Toxin characterization supplies ($10,800): Liquid Chromatograph columns (2 sets/year $2,600), cyanotoxin standards (1 set / 6 months $8,200).	
Lab and field supplies ($4,360): Sample bottles and vials, preservatives, Rite in the Rain notebooks, sunscreen, insect repellent, gloves, test vials, labortory glassware, bags, general storage supplies $2,130 each in Y1 and Y2; $100 in Y3	
Data storage device ($450): 3x 3TB hard drives for genomics data storage @ $150	
Citizen science sampling kits ($26,000): $13/kit x 2,000 kits. Purchased in Y1.	
Li-Cor LI192 submersible light quantum sensor, logger, and cable for measuring underwater light levels in lakes ($3,020). Equipment will be retained for use in future HAB research and monitoring	
Acquisition (Fee Title or Permanent Easements):	**$ -**
Travel:	**$ 19,047**
Field Travel ($10,974): 4 trips per year to sample 100 inland lakes in MN / year (4 trips x 4 nights @ $120 + per diem for 22 travelers @ $64 + mileage + 32 days truck and boat use = $5487/year).	
Outreach ($8,073): distribution of citizen science kits (4 trips per year to attend public events and stakeholder meetings x 1 night @ $120 + partial perdiem for 2 ppl @ $48 + 24 days truck use =$2691/ yr)	
Additional Budget Items:	**$ 91,391**
Scientific services ($86,891): Water quality analysis ($9,000): 100 samples / year @ $30 per sample for total nitrogen and phosphorus + 100 samples / year x $15/sample for chlorophyll. = $4500/ year in Y1 and Y2 only. Genomics analysis ($49,500): Molecular-DNA extraction (2,200 samples x $6.25/sample =$13,750), genome sequencing ($16.25/sample x 2,200 samples = $35,750). Toxin characterization ($28,391): Extraction, prep, sample cleanup and solvents ($8,391) + LC-MS analysis time (200 samples * $80/ sample = $16,000) + LCMS calibration and standardization time (50 hours x $80/hr = $4,000)	
Mailing/shipping to cover cost for citizen scientists to mail back sampling kits ($4,500): $2.25/kit x 2,000 kits.	
TOTAL ENVIRONMENT AND NATURAL RESOURCES TRUST FUND $ REQUEST =	**$ 599,051**

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:	$ -	N/A
Unrecovered indirect: 54% modified total direct costs ($518,621 base; excludes equipment and grad tuition reimbursement)	$ 280,055	Secured
Past and Current ENRTF Appropriation:	$ -	N/A
Other Funding History:	$ -	N/A

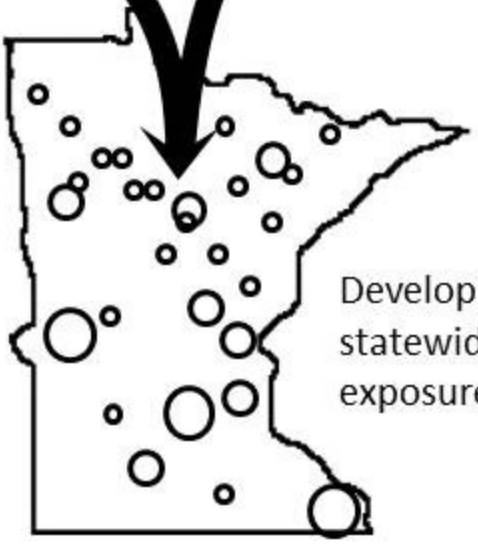
Rapid Detection of Algal Toxins in Minnesota Lakes



Comprehensive genomics and algal toxin survey (200 lakes)

Citizen science sampling program (~2000 lakes)

Toxin Exposure Risk Model



Development of statewide algal toxin exposure risk map

Risk of Algal Toxin Exposure

Low Moderate High

PROJECT TITLE: Rapid Detection of Algal Toxins in Minnesota Lakes

Dr. Andrew Bramburger is a Research Associate at the Natural Resources Research Institute (NRRI) at the University of Minnesota Duluth (UMD). His research interests and expertise lie within the field of phycology (the study of algae), and he has been conducting research on freshwater algae for over 15 years. Bramburger has published 15 peer-reviewed articles on algal communities and presented over 50 conference presentations. Since 2010, Bramburger has served as PI or Co-PI on research programs totaling over $3.5 million in total funding, including ongoing EPA Great Lakes phytoplankton monitoring programs and several projects related to harmful algae blooms and the use of citizen science in both Canada and the U.S.

The NRRI is a U.S.-based research institute established by the Minnesota state legislature within UMD. NRRI is a non-profit applied research organization that works to develop and deliver the understanding and tools needed to utilize our mineral, forest, energy and water resources in a balanced and environmentally responsible manner. The NRRI facility in Duluth MN is a 110,000-square-foot facility dedicated to providing research-based solutions for empowering sustainable development. NRRI is equipped with the facilities for GIS, water quality, and algal analyses, including a wide variety of sampling equipment, boats and field vehicles, as well as sample processing, inverted microscopy and image analysis capabilities. NRRI is a well-established laboratory and research facility and can provide ~$500,000 in analytical equipment, computers, and microscope facilities at no cost to the project. NRRI works in close collaboration with other departments at UMD including the Large Lakes Observatory (LLO) and Minnesota Sea Grant.

The Phycology / Paleolimnology Lab (Bramburger) at NRRI is fully equipped for microscopic analysis of phytoplankton. The laboratory has several microscopes, including Olympus BH-2 and BX-60 compound microscopes equipped with DIC, RIC, and phase contrast optics, as well as Olympus CX-40 inverted microscopes equipped with phase contrast optics and epifluorescence accessories. Auxiliary equipment includes a freeze-dryer, centrifuges, hot-plates, and slide warmers, as well as a dedicated radioisotope preparation facility featuring a Hitachi Aloka Accu-Flex 8000 liquid scintillation counter. Shared facilities within NRRI also consist of a LaChat multi-channel flow-injection nutrient autoanalyzer and a Hitachi TM3030 Plus scanning electron microscope.

The Sheik Geomicrobiology lab housed at the Large Lakes Observatory (LLO) and associated with the Biology Department is equipped as a modern microbiology laboratory with emphasis on culturing and processing samples from the environment. The lab is outfitted with common area bench space with power and gas outlets, a laminar flow hood, fume hood, centrifuges, PCR machine, Qubit DNA quantification platform, agarose gel electrophoresis systems, transilluminator with gel capture camera system, diH2O, Milli-Q water system, incubator, lighted and refrigerated growth chambers, an autoclave, and -20 and -80 °C storage.

The Organic Geochemistry Laboratory (Schreiner) has two Agilent 6890 GCs, one interfaced to an Agilent 5973 quadropole MS and one interfaced to a flame ionization detector. Additionally, this laboratory contains various extraction and other equipment, including Soxhlet extractors, an Accelerated Solvent Extractor, and glassware, hoods, and other equipment necessary for organic geochemical analyses.

The aquatic chemistry laboratory houses an HPLC, Total Organic Carbon analyzer, and FTIR spectrometer, in addition to multiple ovens and furnaces, hoods, microscopes, and chemical glassware and other equipment. The LLO also houses a dedicated LC-MS laboratory, which contains an Agilent LC triple quadrupole MS, along with a variety of peripherals including fraction collectors.