

**Environment and Natural Resources Trust Fund
2020 Request for Proposals (RFP)**

Project Title:

ENRTF ID: 099-B

Rapid Detection of Algal Toxins in Minnesota Lakes

Category: B. Water Resources

Sub-Category:

Total Project Budget: \$ 830,326

Proposed Project Time Period for the Funding Requested: June 30, 2023 (3 yrs)

Summary:

We will use satellite data to identify potentially HAB-impacted lakes, then apply genomics based models developed in this project to quantify cyanotoxin exposure risk in target lakes.

Name: Andrew Bramburger

Sponsoring Organization: U of MN - Duluth NRRI

Job Title: Dr

Department: Natural Resources Research Institute (NRRI)

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Duluth MN 55811

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Web Address:

Location:

Region: Statewide

County Name: Statewide

City / Township:

Alternate Text for Visual:

Visual shows a satellite swath over a stylized landscape depicting satellite identification of target lakes. Below is a schematic of genomic output for inferring cyanotoxin risk, and a map of Minnesota showing lakes where high cyanotoxin exposure risk has been detected.

_____ Funding Priorities	_____ Multiple Benefits	_____ Outcomes	_____ Knowledge Base
_____ Extent of Impact	_____ Innovation	_____ Scientific/Tech Basis	_____ Urgency
_____ Capacity	_____ Readiness	_____ Leverage	_____ TOTAL _____%



**Environment and Natural Resources Trust Fund (ENRTF)
2020 Main Proposal**

PROJECT TITLE: Rapid Detection of Algal Toxins in Minnesota Lakes

I. PROJECT STATEMENT

This project aims to develop a robust, cost-effective, rapid system for monitoring statewide algal toxin exposure risk in Minnesota lakes.

In order to achieve this objective, we will:

- 1. Use cutting edge genetics and toxin characterization techniques in 100 focal lakes statewide in order to identify genomic indicators of high algal toxin concentrations and produce a gene-based HAB Nowcasting model of toxin exposure risk.**
- 2. Use a near real-time water quality monitoring system (WQMS) of > 10,000 lakes that is being built with a current LCCMR funded project to identify lakes with potential for HABs using a chlorophyll threshold developed in Activity 1. Develop cutting edge methods using daily Sentinel 3 satellite imagery to identify HAB pigments that indicate HABs in large lakes (>400 acres).**

Recently, harmful algae blooms (HABs) and associated threats to human and ecosystem health have increased in Minnesota. Despite the relatively low frequency of HAB occurrences in more forested areas of the state, these events are becoming more widely reported, even in “pristine” wilderness areas such as the Boundary Waters Canoe Wilderness Area. The mechanisms that regulate HAB formation and toxicity across multiple HAB species in various lakes are poorly understood, and there exists no statewide means of quantifying risk associated with HABs in order to protect lake users. As such, the proposed project will use cutting edge techniques to 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 protect public health and tourism revenues associated with Minnesota lake use.

Recent advances in environmental genomics technology, coupled with the abundance and diversity of inland lakes in Minnesota and newly available satellite imagery, 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. This proposal represents a collaborative effort among the Natural Resources Research Institute (UMD), Remote Sensing and Geospatial Analysis Laboratory and Water Resources Center (UMTC), and the Science Museum of Minnesota.

II. PROJECT ACTIVITIES AND OUTCOMES

Activity 1: Comprehensive Genomics and Toxin Characterization.

ENRTF Budget: \$432,098

In order to constrain and calibrate remote-sensing based HAB predictions, as well as provide a means of differentiating between toxic and non-toxic blooms, we will characterize water quality conditions and sample water and environmental DNA three times annually in 100 Minnesota lakes. Metagenomics (based on eDNA) and toxin signatures of focal lake samples will be fully characterized using cutting-edge techniques. Gene-based risk models will be constructed and calibrated using focal lake data, and used in Act. 2 to develop remote sensing methods. The resulting early detection system will consist of identification of potentially impacted lakes using remote sensing (Act. 2), and verification of cyanotoxin production hazard using genomic markers.

Outcome	Completion Date
1. Focal lake genetic and toxin sampling and analysis (Comprehensive analysis; Round 1-3; 100 samples/round)	Nov 30, 2020
2. Focal lake genetic and toxin sampling and analysis (Comprehensive analysis; Round 4-6; 100 samples/round, Paleolimnological analyses (10 lakes)	Nov. 30, 2021



Environment and Natural Resources Trust Fund (ENRTF)
2020 Main Proposal

3. Quality control of genetic, toxin, and water quality data, construction/calibration of toxin risk model for application to lakes statewide using Act 2.	Apr 30, 2023
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Activity 2: Remote sensing of HAB conditions in MN lakes.

ENRTF Budget: \$341,714

We will create and verify algorithms to track HABs from satellite images using data from lake sampling (Act. 1). We will use the WQMS that will be operational when this project starts to develop an inventory of HAB likelihood in lakes using a chlorophyll a proxy (Sentinel 2), and explore the applicability of Sentinel 3 satellite imagery to infer phycocyanin (cyanobacterial pigment) concentrations in lakes. To further validate and refine algorithms, we will process 30 years of historical satellite imagery for 10 lakes and compare it to HAB fossils (pigments, DNA, toxins) found in sediment cores of a subset of 10 of the focal lakes. These will be lakes that are part of the MN Sentinel Lake program, allowing us to leverage existing datasets. We will add the ability to flag lakes with potential HABs in the Enhanced Lake Browser created as part of the WQMS.

Outcome	Completion Date
1. Use cloud-based computing to measure historic water clarity in 10 lakes with sediment cores using Landsat imagery (30+ Years).	Sept. 30, 2021
2. Use sampling data (Act. 1) develop methods to predict likelihood of high cyanobacterial concentrations from Sentinel 3 satellite imagery.	Apr. 30, 2022
3. Add HAB flagging capability to Enhanced LakeBrowser	Jan. 30, 2023
4. Use paleolimnological techniques (fossil pigments and DNA) to validate remote sensing-inferred trends in cyanobacteria abundance and toxic potential.	June 30, 2023

Activity 3: Development and Dissemination of HAB Nowcasting Model ENRTF Budget: \$58,504

We will work with MN Sea Grant to disseminate materials to stakeholders. Targeted avenues for outreach include 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.

Outcome	Completion Date
1. Produce outreach materials including visual guide to potentially harmful algae, instructions for reporting suspicious blooms, initial feedback to citizen scientists and snapshot of algal toxin risk in Minnesota lakes.	Sept. 30, 2022
2. Produce statewide toxin risk map, disseminate algal toxin risk model to stakeholders, distribute results via conferences and research articles.	Apr. 30, 2023
3. Provide long-term monitoring outreach and guidance materials including user guide and training manual for sampling kit use, and recommendations for establishing a citizen science monitoring network for HAB toxins.	June 30, 2023

III. PROJECT PARTNERS: Minnesota Department of Natural Resources, Minnesota Department of Health

IV. LONG-TERM- IMPLEMENTATION AND FUNDING:

Potential future applications of this project include a continuously updated publicly-accessible statewide web-based toxin risk map that could be validated monthly based on citizen science sampling. This map could be accessed via a searchable, geo-linked cellular app.

V. TIME LINE REQUIREMENTS:

Three years, from July 2020 through June 2023.

VI. SEE ADDITIONAL PROPOSAL COMPONENTS

Attachment A: Project Budget Spreadsheet
 Environment and Natural Resources Trust Fund
 M.L. 2020 Budget Spreadsheet



Legal Citation:

Project Manager: A. Bramburger

Project Title: Rapid detection of algal toxins in Minnesota Lakes

Organization: University of Minnesota Duluth

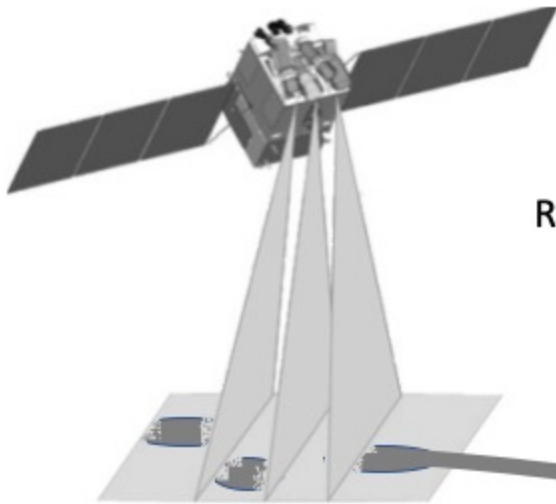
Project Budget: \$830,326

Project Length and Completion Date: 3 years; 06/30/2022

Today's Date: 04/08/2019

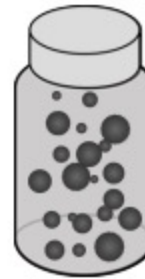
ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET		Budget	Amount Spent	Balance	
BUDGET ITEM					
Personnel (Wages and Benefits) *Salaries of all personnel except Schreiner and Sheik rely upon external funding)		\$ 497,688	\$ -	\$ 497,688	
Andrew Bramburger, UMD-NRRI (Lead PI) 15% FTE, 3 years based on salary of \$74,212 *NRRI staff salaries are largely supported by sponsored external funds.		\$ 48,195			
Christopher Filstrup, UMD-NRRI (Co-PI) 15% FTE, 3 years based on salary of \$69,999 *		\$ 45,459			
Kathryn Schreiner, UMD-LLO (Co-PI) 0.75 months per year summer appointment based on salary of		\$ 26,848			
Cody Sheik, UMD-LLO (Co-PI) 0.75 months per year summer appointment based on salary of \$70,249		\$ 25,342			
Leif Olmanson, UMN-Forest Resources (Co-PI) 15%FTE 3 years based on salary of \$64,440 *		\$ 41,848			
Marte Kitson, MN Sea Grant (Outreach Coordinator) 10%FTE in years 2,3 based on salary of \$58,251		\$ 12,793			
Benjamin Page, UMN Water Resources Center (Research Assoc. - Remote Sensing) 37.5% FTE based on salary of \$52,000 *		\$ 84,427			
Julia Halbur, UMD-LLO (Lab-Tehcnical - Genomics and Toxin Prep) 60% FTE in years 1,2, 40% FTE in year 3 based on salary of \$40,248 *		\$ 76,776			
Elizabeth Alexson, UMD-NRRI (Lab Technician - Algal Analysis) 10% FTE Based on salary of \$43,888 *		\$ 18,091			
M.Sc. Students (Water Resource Science Program, UMD) 2 students 50% appointment for 2 years each		\$ 117,909			
Professional/Technical/Service Contracts		\$ 96,933	\$ -	\$ 96,933	
Science Museum of Minnesota - Paleolimnological Analysis of target lakes		\$ 96,933			
Equipment/Tools/Supplies		\$ 46,613	\$ -	\$ 46,613	
Phytoplankton processing supplies (beakers (case of 100 = \$400), centrifuge bottles (case of 1000 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).		\$ 7,050			
Genomics analysis supplies (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)		\$ 14,765			
Toxin analysis supplies (Liquid Chromatograph columns (2 sets/year = \$2,600), cyanotoxin standards (1 set / 6 months = \$8,200), solvents, extraction materials (\$6,188)		\$ 16,988			
Water Quality Lab Supplies (Sampling bottles = 2 sets / site / year @ \$8.31/site = \$3,364, cuvettes,		\$ 4,060			
General field supplies (Gloves, insect repellent, sunscreen, notebooks = \$100/yr)		\$ 300			
Data Storage (3 - 3TB hard drives for storage of molecular data @ \$150)		\$ 450			
Outreach supplies (Printing of materials, production of videos)		\$ 3,000			
Capital Expenditures Over \$5,000		\$ 31,560	\$ -	\$ 31,560	
Water quality sonde (YSI EXO V2) Used for measuring multiple water quality parameters in situ. Will be dedicated to this project for duration of project and retained for algal monitoring activities after project.		\$ 19,600			
Hyperspectral Radiometer (SeaBird HyperOCR) Used for calibrating satellite data to in water light conditions. Will be dedicated to this project for duration of project and retained for algal monitoring activities after project.		\$ 12,000			
Fee Title Acquisition		\$ -	\$ -	\$ -	
Easement Acquisition		\$ -	\$ -	\$ -	
Professional Services for Acquisition		\$ -	\$ -	\$ -	
Printing		\$ -	\$ -	\$ -	
Travel expenses in Minnesota		\$ 44,445	\$ -	\$ 44,445	
Sampling trips to calibrate models in 100 lakes and verify models in 500 lakes (each trip = 16days truck use (\$240) + mileage (1040 miles = \$603) + hotels for 2 for 14 nights (\$3008) + meal per diem for 2 (\$1705) = \$5556 total. Yr 1 = 1 trip, yr 2 = 5 trips, yr 3 = 2 trips		\$ 44,445			
Other		\$ 113,087	\$ -	\$ 113,087	
Water Quality Analysis (100 samples/yr x \$62 (nutrients) + \$12 (chlorophyll) + \$25 (DOC))		\$ 19,800			
Genomics Analysis = 100 samples (yr 1/2) + 500 samples (yr 2/3) x (\$6.25 for qPCR + \$16.25 for genom seq.) + \$15000 for extraction and transcriptome sequencing to identify target genes = \$17,250		\$ 43,505			
Toxin characteruzation (100 samples year 1/2 + 500 samples year 2/3 @ \$50/sample + \$8391 per		\$ 46,782			
Remote sensing and geospatial analysis lab fees (\$1000/yr)		\$ 3,000			
COLUMN TOTAL		\$ 830,326	\$ -	\$ 830,326	
SOURCE AND USE OF OTHER FUNDS CONTRIBUTED TO THE PROJECT		Status (secured or pending)	Budget	Spent	Balance
Non-State:		\$ -	\$ -	\$ -	\$ -
State:		\$ -	\$ -	\$ -	\$ -
In kind:		\$ -	\$ -	\$ -	\$ -
Other ENRTF APPROPRIATIONS AWARDED IN THE LAST SIX YEARS		Amount legally obligated but not yet spent	Budget	Spent	Balance
			\$ -	\$ -	\$ -

Rapid Detection of Algal Toxins in Minnesota Lakes



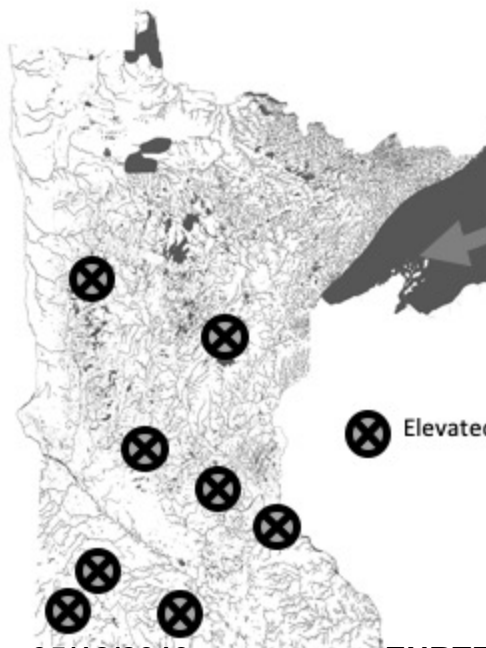
Remote identification of potentially HAB-impacted lakes.

Genomic screening and modeling of cyanotoxin exposure risk in potentially-impacted lakes



ACTGAGTTCCCT
TACTGAGTTCC
CCGTCTGGTAG
TTCCGAGTTCC
CTTCCGAGTTCC
TCCGAGTTCCCT
GGATAACCGTG
ACGCCATAGAG
TTCCGAGTTCC
CGGGACGCCAT
CGTCTGGTAGG

Statewide modeling and mapping of cyanotoxin exposure risk



⊗ Elevated cyanotoxin risk detected



Environment and Natural Resources Trust Fund (ENRTF) 2020 Project Manager Qualifications and Organization Description

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 20 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 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 Central Analytical Lab (Filstrup) 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.