

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

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**Project Title:**

**ENRTF ID: 051-AH**

Modern eDNA Technology for Better Game Fish Census

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**Category:** H. Proposals seeking \$200,000 or less in funding

**Sub-Category:** A. Foundational Natural Resource Data and Information

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**Total Project Budget:** \$ 200,000

**Proposed Project Time Period for the Funding Requested:** June 30, 2022 (2 yrs)

**Summary:**

We will develop an inexpensive and rapid eDNA measurement tool to determine the abundance and distribution of Walleye in MN lakes, which greatly enhances current counting methods to manage Walleye.

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**Name:** Michael Sadowsky

**Sponsoring Organization:** U of MN

**Job Title:** Dr.

**Department:** BioTechnology Institute

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St. Paul MN 55113

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**Email:** sadowsky@umn.edu

**Web Address:** \_\_\_\_\_

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**Location:**

**Region:** Statewide

**County Name:** Statewide

**City / Township:**

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**Alternate Text for Visual:**

Lakes and streams in Minnesota providing fish habitats

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



**PROJECT TITLE: Modern eDNA Technology for Better Game Fish Census**

**I. PROJECT STATEMENT**

Fishing in Minnesota is a big deal, with nearly 32% of its resident population participating in some form of fishing activity. It has been estimated that the state boasts more than 1 million licensed anglers and opening day brings nearly 500,000 people to the states more than 5,400 fishing lakes and 18,000 miles of fishable rivers and streams. The most prized fish among anglers are walleyes (the State Fish), northern pike, panfish, and muskies, in order of preference. Together fishing supports about 35,000 Minnesota jobs, pumping nearly \$2.4 billion to the state’s economy. **Despite its importance to the state, establishing fish abundance, distribution, and catch limits in Minnesota’s lakes is difficult, costly, and often imprecise because these estimates presently rely on a limited array of traditional methods, including gill netting, trap netting, and electrofishing.** The MN DNR is the main agency responsible for fisheries management, mainly using some 650, 3-5 year, lake surveys to guide stocking and management actions. The DNR reports that \$5 million is used annually for lake and steam surveys. While Minnesota’s largest lakes are surveyed annually, smaller and more remote lakes may only be surveyed once in 10-20 years. Despite this large and costly effort, the current sampling technology cannot alone provide accurate fish census data for all locations, let alone in the large number of lakes and streams in MN.

**In this project, we develop and test an inexpensive, rapid, and sensitive detection tool to better quantify the numbers and distribution of Walleye in Minnesota’s lakes using the DNA these fish release (environmental or “eDNA”).**

The new method to detect Walleye relies on measuring the eDNA released by the fish, similar to that used in crime forensics. **It will greatly improve upon traditional techniques by enhancing the sensitivity and accuracy of measuring Walleye. Notably, the technique will also allow biologists and managers to sample waters that are otherwise inaccessible to DNR netting (e.g. are too deep, ice covered, heavily vegetated, bad weather, too isolated).** Quantification of eDNA has gained favor by federal agencies scientists for the detection of fish, including aquatic invasive species – so why not game fish. Nevertheless, although taking lake water samples for eDNA is faster than netting, standard techniques require use of inefficient filtration methods to capture DNA, and requiring relatively large volume of water. This has meant that eDNA technology is not yet widely available for routine quantification of fish, such as walleye. **The proposed research will solve this problem by developing and testing a rapid and quantitative DNA detection system to quantify Walleye eDNA in up to 88 samples at a time.** This technology will augment current net surveys to more accurately and easily determine fish population data that can be used to better manage Walleye fisheries.

**II. PROJECT ACTIVITIES AND OUTCOMES**

The project will proceed in several steps to detect and quantify numbers and distribution of Walleye in a Minnesota lake. We will also expand detection ability and sensitivity by developing new filtration methods to capture eDNA and to utilize inexpensive qPCR technology that simultaneously measures eDNA in up to 88 samples at a time (using the MFQPCR technique). We will disseminate results and approaches to government biologists including those in MN DNR, USGS, and USFWS. We will establish the technique as a service for the state to use for some time, until adopted by them for the long term. Key steps in our project are described below:

**Activity 1:** eDNA Marker gene development and validation.

**Budget:** \$96,000

While a potential marker gene for Walleye has been published, we will modify this DNA sequence to improve sensitivity and specificity.

<b>Outcome</b>	<b>Completion</b>
1. Modify current DNA marker genes for walleye.	September 2020
2. Test new markers for specificity and sensitivity using DNA detection methods.	December 2020



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**2020 Main Proposal**

**Project Title: Better Game Fish Technology**

4. Optimize protocols to enhance detection sensitivity in water from several lakes.	January 2021
5. Develop new filtration-based technologies to increase fish detection.	June 2021
6. Optimize filtration technologies to specifically capture eDNA from fish.	December 2021

**Activity 2:** Assay validation

**Budget:** \$96,000

<b>Outcome</b>	<b>Completion</b>
7. Validate correlation between lab and field data obtained using DNA standards created from known fish census data to that obtained from tank studies (if not walleye then carp ).	May 2022
8. Test protocols using lake water containing known numbers of walleye based on net census data obtained by DNR. Determine decay rate of walleye eDNA in lake water.	May 2022
9. Validate test assays using real-time lake net data.	June 2022

**Activity 3:** Dissemination of results

**Budget:** \$8,000

To be meaningful, the assay we develop will need to be used by others.

<b>Outcome</b>	<b>Completion</b>
1. Publish results in peer-reviewed journals.	June, 2022
2. Sponsor a workshop to train DNR and other biologists.	June, 2022
3. Present results at scientific meetings and symposia.	June, 2022
4. Work with UMN extension service and BTI outreach personnel to publicize our technology, including development of a web site resource.	June, 2022

**III. PROJECT STRATEGY**

**A. Project Team/Partners**

Our team consists of Michael Sadowsky, Peter Sorensen (FWCB), and Ping Wang (BTI). Dr. Sadowsky will coordinate all laboratory activities, Dr. Sorensen will coordinate fish studies including working with ongoing DNR surveys (collecting water samples concurrently and then comparing data). Dr. Wang will work with a postdoctoral fellow to develop markers and implement qPCR assays, as well as to help with water filtration technology development. The research team assembled has all the necessary qualifications to perform the proposed studies. Their areas of expertise are complementary. Michael Sadowsky is a microbial ecologist with expertise in genomics and quantitative PCR. Peter Sorensen is an expert in fish biology, and Dr. Wang has expertise in qPCR and the development of primers and probes to detect fish in aquatic systems. The MN DNR has expressed interest in this technology in conversations with Dr. Sorensen, as has the USGS in discussion with Dr. Sadowsky.

**B. Project Impact and Long-Term Strategy**

Dr. Sorensen has already developed eDNA techniques to measure the presence of invasive fish in inland lakes and rivers. This study extends this work by focusing on game fish, initially walleye. This project will create a new practical technique that could be used to aid in monitoring and managing game fish in Minnesota’s lakes and waterways. This technology will not replace current net surveys, but will augment this data to provide better predictions of fish census that can be used to better manage this critical fisheries resource. We will conduct initial studies to demonstrate its use in MN waterways. The next phase would be to work directly with DNR biologists to implement this technology more broadly. We propose to train DNR and other biologists is how to do these assays in workshops at the University of Minnesota. Our ultimate goal is to obtain competitive national grants to continue this research and extend its application.

**C. Timeline Requirements:** We request two years for this project.

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



Legal Citation:  
 Project Manager: Michael J. Sadowsky  
 Project Title: Modern eDNA Technology for Improved Game Fish Census  
 Organization: University of Minnesota  
 Project Budget: \$200,000  
 Project Length and Completion Date: 2 years, August 31, 2022  
 Today's Date: 04/03/2019

ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET		Budget	Amount Spent	Balance
<b>BUDGET ITEM</b>				
<b>Personnel (Wages and Benefits)</b>		\$ 152,000	\$ -	\$ 152,000
Peter Sorensen, Professor 2 weeks summer support (benefits = 36.0%) for two years; supervision of research technician, data analysis, project reporting. (salary 74%, fringe 26% of total) (5% FTE)				
Postdoctoral research associate (24.3% benefits); 100% FTE for two years; perform experiments, analyze data, write manuscripts (80% salary, 20% fringe of total)				
Undergraduate student support for two years; maintain fish tanks, and help with field studies. All salary, no fringe benefits				
<b>Professional/Technical/Service Contracts; University of Minnesota Genomics Center: MiSeq DNA sequencing (\$3,000/run x 2 runs = \$6,000), Sangar sequencing (\$5/reads x 2 reads/sample x 200 samples = \$2,000)</b>		\$ 8,000		\$ 8,000
<b>Equipment/Tools/Supplies: Lab and/ or Field: Field sampling (\$2,250), water filtration (\$20 x 500 samples = \$10,000), DNA extraction (\$5/sample x 500 samples = \$2,500), PCR/qPCR primers and reagents (\$5,600), qPCR standards (\$95/assay x 20 assays = \$1,900), fish feed (\$2,000), chemicals (\$2,000), disposable plasticware (\$1,750), aquarium supplies (\$2,000), Fish (\$2,000)</b>		\$ 32,000		\$ 32,000
<b>Printing /Publication: Publication costs for dissemination. \$500-\$2000 per manuscript. Total: 3-4</b>		\$ 4,000	\$ -	\$ 4,000
<b>Travel expenses in Minnesota: In-state sampling – Transportation: 5 field trips per year (~862 miles) X \$0.58/mile x 2 years = \$1,000. Lodging and meals during field trips: \$150/person/trip x 2 person/trip x 5 trips x 2 years = \$3,000</b>		\$ 4,000		\$ 4,000
		\$ -	\$ -	\$ -
<b>Other</b>				
		\$ -	\$ -	\$ -
<b>COLUMN TOTAL</b>		\$ 200,000	\$ -	\$ 200,000
<b>SOURCE AND USE OF OTHER FUNDS CONTRIBUTED TO THE PROJECT</b>				
	<b>Status (secured or pending)</b>	<b>Budget</b>	<b>Spent</b>	<b>Balance</b>
<b>Non-State:</b>		\$ -	\$ -	\$ -
<b>State:</b>		\$ -	\$ -	\$ -
<b>In kind:</b> The University of Minnesota does not charge the State of Minnesota its typical overhead rate of 54% of the total modified direct costs.		\$ 108,000	\$ -	\$ 108,000
<b>Other ENRTF APPROPRIATIONS AWARDED IN THE LAST SIX YEARS</b>				
	<b>Amount legally obligated but not yet spent</b>	<b>Budget</b>	<b>Spent</b>	<b>Balance</b>
		\$ -	\$ -	\$ -

## Every lake & river in Minnesota

*Pioneer Press graphic by David H. Montgomery*



Lakes and rivers in Minnesota (blue outlines), some of which heavily support Minnesota's walleye and game fish industry.

**PROJECT TITLE: Modern eDNA Technology for Better Game Fish Census**

**Project Manager Qualifications and Organization Descriptions**

**Michael Sadowsky**

Michael Sadowsky is McKnight University Professor in the Dept. of soil, Water and Climate. He is also director of the BioTechnology Institute. He has been studying microbes and their DNA in the environment for the last 38 years and has recently been working on microbiota associated with invasive species (fish, mussels, and plants). His laboratory has expertise in quantitative PCR and DNA sequencing technologies to track organisms in the environment, including game and invasive fish. He has published over 325 peer-reviewed papers in scientific journals. The Sadowsky Lab is well equipped with all the necessary items for the proposed research. In addition, his group has a full access to qPCR and all needed instruments and access to the University of Minnesota supercomputing institute.

**Peter Sorensen**

Peter Sorensen is a professor in the Dept. Fisheries, Wildlife and Conservation Biology. He has been studying invasive fish for 29 years since he arrived at the University of Minnesota and has over 140 peer-reviewed articles in addition to nearly 50 book chapters and a patent on sea lamprey control. Dr. Sorensen has considerable experience measuring fish DNA, which was a prelude to this work. His laboratory is well equipped for this study and he also has access to two wet lab facilities to hold and handle fish. He also has all necessary permits for this work and government colleagues who could acquire them.

**Organization Descriptions**

The University of Minnesota is the main research and graduate teaching institution in the state of Minnesota. The BioTechnology Institute provides advanced research, training, and university-industry interaction in biological process technology. In the Department of Soil, Water, and Climate, we seek to improve and protect the quality of soil, air, and water resources in natural and managed ecosystems, through research, reaching, and extension.