Environment and Natural Resources Trust Fund
2018 Request for Proposals (RFP)

Project Title: **ENRTF ID: 054-B**

Emerging Pathogens in Lakes, Rivers, and Tap Water

Category: B. Water Resources

Total Project Budget: $ 355,244

Proposed Project Time Period for the Funding Requested: 3 years, July 2018 to June 2021

Summary:

This research project will provide critical information regarding how to treat surface water (used by 25% of Minnesota's population) to prevent outbreaks of Legionnaires' disease and infections by Mycobacterium avium.

Name: Timothy LaPara

Sponsoring Organization: U of MN

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Minneapolis MN 55455

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Web Address

Location

Region: Statewide

County Name: Statewide

City / Township:

Alternate Text for Visual:

We will quantify Legionella and Mycobacteria from surface water to tap water. We have detected Legionella in Norwegian water mains but not in water mains in Minnesota. We have detected benign Mycobacteria so far in Minnesota, but the potential exists for Mycobacterium avium to be present.

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

PROJECT TITLE: Emerging pathogens in lakes, rivers, and tap water

I. PROJECT STATEMENT

Minnesota's surface waters are a critically important natural resource that are used for numerous purposes ranging from recreation (e.g., boating and fishing) to transportation (e.g., shipping). Perhaps most importantly, however, surface waters serve as the source for drinking water for 25% of Minnesota's population. Water utilities in the State of Minnesota generally do an excellent job of treating this surface water so that it is safe for all Minnesotans to drink. From regulatory and microbiological perspectives, drinking water safety is usually evaluated based on the presence of "enteric organisms" (i.e., organisms found in the digestive tracts of warm-blooded animals), such as total coliforms or *E. coli*.

This first goal of this project is to quantify the presence and occurrence of two pathogens of emerging concern, *Mycobacteria* and *Legionella*, in Minnesota surface waters that serve as the source for drinking water supplies (e.g., Mississippi River, Red River, Lake Superior). These organisms are fairly unique because they naturally live in the environment, unlike enteric pathogens that are found in feces. The second goal of this project is to assess the ability of Minnesota's drinking water treatment facilities to remove these organisms, thus protecting the public from these pathogens of emerging concern. The third goal of this project is to quantify the presence and occurrence of these organisms in tap water. This goal is pertinent because, while the treated water leaving a public water utility is typically safe, water can spend days to weeks in water distribution systems and these organisms can potentially re-grow during this time. To combat this potential re-growth problem, water utilities attempt to maintain a low concentration of disinfectant (chlorine or chloramine) throughout their distribution system. This "residual disinfectant", however, can be very difficult and expensive to maintain.

Legionnaires' disease is a deadly form of pneumonia; there was a substantial outbreak of Legionnaires' disease in Hopkins, MN in the Fall of 2016, affecting more than 20 people including one fatality. The causative agents of Legionnaires' disease are bacteria of the genus *Legionella*. Similarly, *Mycobacterium avium* is an organism that causes a deadly pneumonia in immunocompromised people. Both organisms are relatively common in the environment, including surface waters, but little is known about how the abundance of these organisms varies with location (i.e., land use) and over time (i.e., season). Similarly, we know relatively little about the presence of *Legionella* and *Mycobacteria* in surface waters or in tap water because they are not enteric pathogens, which are historically the primary organisms of concern in terms of public health.

There is growing evidence and concern that public water supplies are pertinent carriers of *Legionella* and *Mycobacteria*. These bacteria in drinking water are literally distributed to every home, business, and industry connected to a water distribution system. We have observed *Legionella* in tap water, even in systems with a substantial disinfectant residual. Thus, we suspect that most public water supplies can serve as the "seed" for *Legionella* to grow in so-called premise plumbing inside of buildings. For example, *Legionella* are known to grow in hot water pipe loops in apartment buildings and hospitals and in hot water heaters operated at less than 125°F. Similarly, we have detected non-pathogenic strains of *Mycobacteria* in drinking water, perhaps suggesting that *Mycobacterium avium* could be present in some systems.

This project will provide critically important information for Minnesota's policy makers, water treatment engineers, and water treatment operators regarding how to best treat surface water and how to maintain water distribution systems to prevent outbreaks of Legionnaires' disease and infections by Mycobacterium avium. For example, this research could show that specific surface waters should be avoided as a source of drinking water because of excessively high levels of Legionella and/or Mycobacteria. Alternatively, this research could identify specific treatment technologies that are particularly effective at removing Legionella and/or Mycobacteria.

II. PROJECT ACTIVITIES AND OUTCOMES

Activity 1: *Measure the quantities of Legionella and Mycobacteria in Minnesota's lakes and rivers used as sources for public water supplies, in Minnesota's water utilities, in* **Budget: $ 347,744**

treated water leaving Minnesota's water utilities, and in tap water samples.

We will collect large volume samples of surface water (~500 liters), treated drinking water (i.e., leaving the water utility), and tap water from 5 to 10 different cities. We will attempt to collect samples throughout the year (as feasible) to assess seasonal variability. We will also collect filter media samples from water utilities (if possible) because these samples often have fairly high levels of microbes. We will primarily measure *Legionella* and *Mycobacteria* using highly-specific genetic assays (i.e., quantitative polymerase chain reaction), but we will also use a newly available commercial kit that can culture *Legionella* (Legiolert).

Outcome	Completion Date
1. Collect surface water samples (river or lake) used as the source of 5 to 10 different water supplies at least 4 times a year to capture seasonal variability	August 31, 2019
2. Collect filter media and finished water samples from 5 to 10 different water utilities at least every 6 weeks to capture seasonal variability.	August 31, 2019
3. Collect tap water samples from at least 3 different locations from at least 5 different cities on at least two occasions (summer and winter).	December 31, 2019
4. Enumerate viable indicator bacteria (coliforms) and viable Legionella	December 31, 2019
5. Extraction and purify DNA from water and filter media samples	June 30, 2020
6. Quantify Legionella and Mycobacteria by quantitative polymerase chain reaction	December 31, 2020

Activity 2: *Disseminate our results to Minnesota's public water utilities* **Budget: $ 7,500**

Outcome	Completion Date
1. Presentations at in-sate scientific conferences (on-going/continuous)	June 30, 2021
2. Meetings with stakeholders to disseminate results (on-going/continuous)	June 30, 2021

III. PROJECT STRATEGY

A. Project Team/Partners

The project team will be led by Timothy LaPara (University of Minnesota; Dept. of Civil, Environmental, and Geo- Engineering) and Raymond Hozalski (UMN; CEGE). Drs. LaPara and Hozalski are experts in environmental microbiology and the analysis of bacteria in drinking water. Drs. LaPara and Hozalski will oversee all aspects of the project, including meetings with stakeholders. A graduate student and two undergraduate students (to be hired) will collect samples, extract/purify DNA, perform quantitative polymerase chain reaction, analyze data, and present the results at scientific conferences. We will partner with water utilities on a voluntary and confidential basis.

B. Project Impact and Long-Term Strategy

The long-term goal of the proposed research is to gain a better understanding of the potential sources of problematic microorganisms in Minnesota water supplies and the overall microbiological safety of drinking water in Minnesota. Specifically, our knowledge of emerging pathogens in Minnesota water supplies and drinking water systems is relatively limited. *Legionella* bacteria, for example, were discovered fairly recently (i.e., about 40 years ago), the organism is difficult to cultivate in the laboratory, and its concentration in drinking water is sometimes very low. This research is especially important because about 25% of Minnesota's population drinks water obtained from a surface water source (e.g., Minneapolis, St. Paul, St. Cloud, Duluth). The proposed study, therefore, will generate novel and critically important knowledge on the microbiological quality of Minnesota's public water supplies.

C. Timeline Requirements

The proposed project will be completed in a three-year period. The sample collection, chemical analysis, and microbiological analysis are time-consuming and will require detailed quality assurance/quality control.

2018 Detailed Project Budget

Project Title: *Emerging pathogens in lakes, rivers, and tap water*

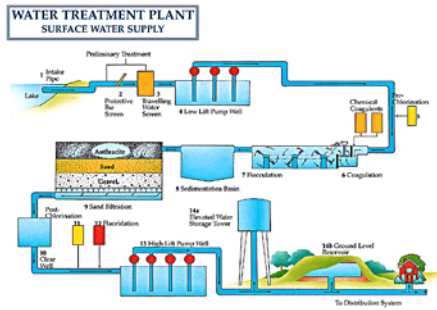
IV. TOTAL ENRTF REQUEST BUDGET *3* **years**

BUDGET ITEM	AMOUNT
Personnel:	
Timothy LaPara, Project Manager (75% salary, 25% benefits); 8% FTE; Project supervision, supervision of post-doctoral and graduate student, project reporting, dissemination and outreach.	$ 53,878
Raymond M. Hozalski, co-Project Manager (75% salary, 25% benefits); 8% FTE; Project supervision, supervision of post-doctoral and graduate student, project reporting, dissemination and outreach.	$ 59,762
Graduate researcher to be hired (100% salary, 41% benefits); 50% FTE; Collect samples, extract/purify DNA, quantify Legionella and Mycobacteria	$ 164,744
Undergraduate researchers, to be hired (100% salary, 0% benefits); 25% FTE during academic year, sample collection and processing	$ 24,360
Professional/Technical/Service Contracts: University of Minnesota Genomics Center: next generation DNA sequencing, quantitative polymerase chain reaction, DNA quantification, etc.	$ 15,000
Equipment/Tools/Supplies	
Membrane Filters ($20 per sample; 500 samples)	$ 10,000
DNA extraction kits ($3 per sample; 500 samples)	$ 1,500
Legiolert supplies ($10 per sample; 500 samples)	$ 5,000
qPCR supplies ($2 per assay; 6 assays per sample; 500 samples)	$ 6,000
Miscellanous supplies (expendable glassware, reagents, etc)	$ 5,000
Travel: *In-state travel to collect samples and to present results to stakeholders and at in-state water conferences*	$ 5,000
Additional Budget Items: *Publication charges. We will publish our manuscripts 'open access' to maximize data availability to Minnesotans*	$ 5,000
TOTAL ENVIRONMENT AND NATURAL RESOURCES TRUST FUND $ REQUEST =	$ 355,244

V. OTHER FUNDS *(This entire section must be filled out. Do not delete rows. Indicate "N/A" if row is not applicable.)*

SOURCE OF FUNDS	AMOUNT	Status
Other Non-State $ To Be Applied To Project During Project Period: *Indicate any additional non-state cash dollars secured or applied for to be spent on the project during the funding period. For each individual sum, list out the source of the funds, the amount, and indicate whether the funds are secured or pending approval.*	n/a	n/a
Other State $ To Be Applied To Project During Project Period: *Indicate any additional state cash dollars (e.g., bonding, other grants) secured or applied for to be spent on the project during the funding period. For each individual sum, list out the source of the funds, the amount, and indicate whether the funds are secured or pending approval.*	n/a	n/a
In-kind Services To Be Applied To Project During Project Period:*The University of Minnesota does not charge the State of Minnesota its typical overhead rate of 54% of the total modified direct costs (graduate tuition is excluded).*	$ 167,419	*secured*
Past and Current ENRTF Appropriation: *This project leverages knowledge obtained from ENTRF project Bacterial Assessment of Groundwater Supplies used for Drinking Water (2017-2019)*	$ 299,000	Obligated
Other Funding History: *Indicate funding secured but to be expended prior to July 1, 2018, for activities directly relevant to this specific funding request. State specific source(s) of funds and dollar amount.*	n/a	n/a

Emerging pathogens in lakes, rivers, and tap water



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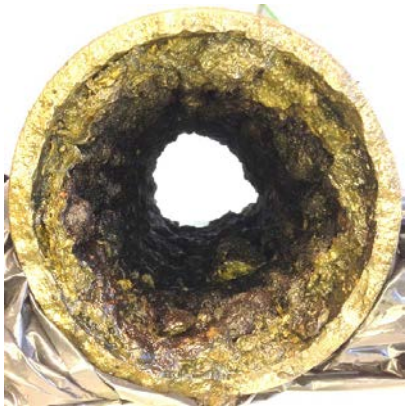

We have detected *Legionella* in Norwegian water mains (left) but not in water mains in Minnesota (right). We have detected benign *Mycobacteria* so far in Minnesota, but the potential exists for *Mycobacterium avium* to be present.

Project Manager Qualifications and Organization Description

Timothy M. LaPara
Professor, Department of Civil, Environmental, and Geo- Engineering, University of Minnesota

B.S.C.E., Civil Engineering, 1995, University of Notre Dame, Notre Dame, IN
Ph.D., Civil Engineering, 1999, Purdue University, West Lafayette, IN

Dr. LaPara's research focuses on the microbiology of municipal wastewater treatment and the treatment of public water supplies; the goal of his research is to preserve environmental quality and to protect public health. His research has a strong interdisciplinary nature, stemming from his unique background in both environmental engineering and microbiology.

Raymond M. Hozalski
Professor, Department of Civil, Environmental, and Geo- Engineering, University of Minnesota

B.ChE., Chemical Engineering, 1990, Villanova University, Villanova, PA
M.S., 1992, Environmental Engineering, Johns Hopkins University, Baltimore, MD
Ph.D., 1996, Environmental Engineering, Johns Hopkins University, Baltimore, MD

Dr. Hozalski's research focuses on water treatment systems including filtration, biofiltration, sorption, and chemical oxidation as well as water distribution system issues. His research has recently produced some of first detailed characterizations of bacterial communities in biofilms on the inside of water distribution system pipes and in tap water. His investigations span the full spectrum from basic research to applied research done in collaboration with water utilities in Minnesota and beyond. He is well connected in the drinking water community in Minnesota through his involvement with the Minnesota Section of the American Water Works Association.

Organization Description
The University of Minnesota is one of the largest, most comprehensive, and most prestigious public universities in the United States (http://www1.umn.edu/twincities/about/index.html). The laboratories and offices of the PIs contain the necessary fixed and moveable equipment and facilities needed for the proposed studies.