Environment and Natural Resources Trust Fund (ENRTF)
M.L. 2018 ENRTF Work Plan (Main Document)

Today's Date: December 15, 2017

Date of Next Status Update Report: January 31, 2019

Date of Work Plan Approval:

Project Completion Date: June 30, 2021

Does this submission include an amendment request? No

PROJECT TITLE: Evaluate Emerging Pathogens in Lakes, Rivers, and Tap Water to Keep Drinking Water Safe

Project Manager: Timothy M. LaPara

Organization: University of Minnesota

College/Department/Division: Department of Civil, Environmental, and Geo- Engineering

Mailing Address: 500 Pillsbury Drive SE

City/State/Zip Code: Minneapolis, MN 55455

Telephone Number: (612) 624-6028

Email Address: lapar001@umn.edu

Web Address:

Location: Statewide

Total Project Budget: $325,000

Amount Spent: $0

Balance: $325,000

Legal Citation: M.L. 2018, Chp. xx, Sec. xx, Subd. xx

Appropriation Language:

I. PROJECT STATEMENT:

Minnesota's 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 meets all regulatory requirements, suggesting 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*. Recent research has suggested, however, that drinking water may also serve as the source for two opportunistic pathogens (i.e., non-enteric organisms), *Legionella pneumophila* and *Mycobacterium avium*. Unlike enteric pathogens, where exposure is via the so-called fecal-oral route, these opportunists are environmental bacteria that tend to occur naturally in water and the exposure route is through inhalation of droplets (aerosols) containing the organisms. This exposure can occur through showering, use of hot tubs, and other means.

Legionella pneumophila is the causative agent of Legionnaires' disease, a deadly form of pneumonia. There was a recent outbreak of Legionnaires' disease in Hopkins, MN in the Fall of 2016, affecting more than 20 people, including one fatality. The elderly and heavy smokers are typically the most likely to develop Legionnaires' disease. 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 and hence the ones that are targeted by monitoring programs (i.e., fecal indicators).

There is growing evidence and concern that public water supplies are pertinent carriers of *Legionella* and *Mycobacteria*. 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 in our previous research, even in systems with a substantial disinfectant residual. Thus, we suspect that most public water supplies can serve as the inoculum 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. Although we have not specifically detected *M. avium* in public water supplies, other researchers have detected *M. avium*, suggesting that this opportunistic pathogen could be present in some public water supplies in Minnesota. Fortunately, there is also evidence that a residual of monochloramine (resulting from the reaction of free chlorine and ammonia) in the water can be effective at controlling *Legionella* spp. in building hot water systems.

This project will characterize and quantify both *L. pneumophila* and *M. avium* from source water to tap in four or five water systems in the State of Minnesota. We hypothesize that the majority of surface water samples will have low but measurable levels of both *L. pneumophila* and *M. avium.* We hypothesize that drinking water treatment technologies will substantially reduce the quantity of all microorganisms in the water, including (but not specifically) *L. pneumophila* and *M. avium.* We hypothesize that the maintenance of a chlorine or chloramine disinfectant will further suppress *L. pneumophila* but select for *Mycobacteria* species (these organisms are known to be resistant to disinfectants used in drinking water). We finally hypothesize that the maintenance of a strong residual will select against *M. avium* compared to other species within the genus of *Mycobacteria*.

II. OVERALL PROJECT STATUS UPDATES:

First Update January 31, 2019

Second Update June 30, 2019

Third Update January 31, 2020

Fourth Update June 30, 2020

Fifth Update January 31, 2021

Final Update June 30, 2021

III. PROJECT ACTIVITIES AND OUTCOMES:

ACTIVITY 1: Sample collection, DNA extraction and preservation
Description: Water samples will be collected from the major surface water supplies in the State (the Mississippi River, the Red River, and Lake Superior) and from four or five public water utility systems that use these water sources. Participating water utilities will remain anonymous and will be identified only by their source water type (lake or river) and the data that we generate. We will collect the "raw" or source water (i.e., lake or river water) from near the intake point for each utility. We will also collect treated water prior to disinfection and following disinfection (i.e., finished water). Finally, we will collect tap water from a publically accessible location (city hall, fire department, etc) within the distribution system of each participating City. We will collect five replicate samples from each utility and from each sample location within the first 16 months of the project period (i.e., 5 utilities $\times$ 4 sample locations $\times$ 5 replicates = 100 samples).

ENRTF BUDGET: $ 94,450

Outcome	Completion Date
1. Sample Collection	October 31, 2019
2. DNA extraction and purification	January 31, 2020

First Update January 31, 2019

Second Update June 30, 2019

Third Update January 31, 2020

Fourth Update June 30, 2020

Fifth Update January 31, 2021

Final Update June 30, 2021

ACTIVITY 2: Quantification and characterization of microorganisms (especially *Legionella* and *Mycobacteria*)
Description: We will use quantitative polymerase chain reaction (qPCR) to quantify total bacteria (as 16S rRNA genes), total *Legionella* spp. (*ssrA* genes), *Legionella pneumophila* (*mip* genes), and *Legionella pneumophila* serogroup 1 (*wzm* genes), total *Mycobacteria* spp. (*atpE*), and *M. avium* (ITS region) in the samples collected from Activity 1. The composition of the bacterial communities in the water samples from Activity 1 also will be determined by using the polymerase chain reaction (PCR) to amplify the V3-region of the 16S rRNA gene, which will then be sequenced using an Illumina MiSeq instrument. The *Mycobacteria* in the samples will be

characterized to the strain level using PCR to amplify the gene encoding a 65-kDa heat-shock protein (*hsp65*) and sequencing the PCR products by Illumina MiSeq.

ENRTF BUDGET: $ 230,550

Outcome	Completion Date
1. qPCR targeting *Legionella* and *Mycobacteria*	May 31, 2020
2. PCR and Illumina MiSeq analysis of 16S rRNA genes	May 31, 2020
3. PCR and Illumina MiSeq analysis of *hsp65* genes	August 31, 2020
4. Data Analysis	December 31, 2020

First Update January 31, 2019

Second Update June 30, 2019

Third Update January 31, 2020

Fourth Update June 30, 2020

Fifth Update January 31, 2021

Final Update June 30, 2021

IV. DISSEMINATION:

Description:

Findings will be disseminated directly to each of the sampled utilities as a written report and an in-person presentation (if desired by the utility). Findings will also be disseminated and archived via reports to LCCMR, peer-reviewed publications (published open-access), and presentations at conferences (particularly the annual meeting of the State's American Water Works contingency in Duluth). We will also, when appropriate, disseminate results via press releases to the media and via the MDH website. The audience is not only the scientific community, but also the public, policymakers, and practitioners. The work will also be of interest to the medical community and we will seek avenues to share the results with this community. We will preserve the anonymity of participating utilities in our press releases and publications upon their request and work with them and the MDH to determine how best to communicate the results from their individual systems to their customers.

First Update January 31, 2019

Second Update June 30, 2019

Third Update January 31, 2020

Fourth Update June 30, 2020

Fifth Update January 31, 2021

Final Update June 30, 2021

V. PROJECT BUDGET SUMMARY:

A. Preliminary ENRTF Budget Overview: See attached budget spreadsheet

Explanation of Capital Expenditures Greater Than $5,000: NA

Explanation of Use of Classified Staff: NA

Total Number of Full-time Equivalents (FTE) Directly Funded with this ENRTF Appropriation:

Enter Total Estimated Personnel Hours: 8090	Divide by 2,080 = TOTAL FTE: 3.9

Total Number of Full-time Equivalents (FTE) Estimated to Be Funded through Contracts with this ENRTF Appropriation:

Enter Total Estimated Personnel Hours:	Divide by 2,080 = TOTAL FTE:

B. Other Funds:

SOURCE OF AND USE OF OTHER FUNDS	Amount Proposed	Amount Spent	Status and Timeframe
Other Non-State $ To Be Applied To Project During Project Period:			
	$	$	
Other State $ To Be Applied To Project During Project Period:			
	$	$	
Past and Current ENRTF Appropriation:			
M. L. 2016, Chp 186, Sec. 2, Subd. 04f	$299,000	$178,259	Project is currently ahead of schedule and should be completed by June 30, 2019
Other Funding History:			
	$	$	

VI. PROJECT PARTNERS:

A. Partners receiving ENRTF funding

Name	Title	Affiliation	Role

B. Partners NOT receiving ENRTF funding

Name	Title	Affiliation	Role

VII. LONG-TERM- IMPLEMENTATION AND FUNDING:

The implications of this project could be substantial with respect to the operation and management of public water supplies in the State of Minnesota. We will provide specific technical support to the participating utilities

regarding strategies to minimize the risk posed by *Legionella* and *Mycobacteria* to public health; we will provide analogous general technical support to non-participating utilities within the State of Minnesota also (i.e., the quality of our technical support will depend, to a certain extent, on our knowledge of a public water supplier). It is also possible that this project will uncover new questions that require further research; in that case, we will apply to the Minnesota Environment and Natural Resources Trust Fund or other sources (e.g., National Science Foundation) for financial support.

VIII. REPORTING REQUIREMENTS:

- **The project is for 3 years, will begin on July 1, 2018, and end on June 30, 2021.**
- **Periodic project status update reports will be submitted January 31 and June 30 of each year.**
- **A final report and associated products will be submitted between June 30 and August 15, 2021.**

IX. SEE ADDITIONAL WORK PLAN COMPONENTS:

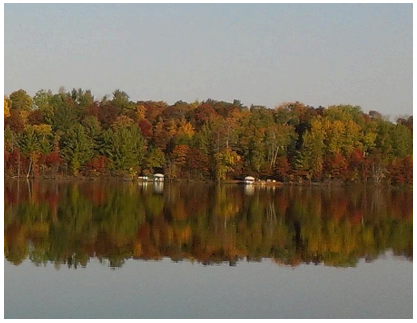
A. Budget Spreadsheet
B. Visual Component or Map
C. Parcel List Spreadsheet - NA
D. Acquisition, Easements, and Restoration Requirements - NA
E. Research Addendum – Separate document

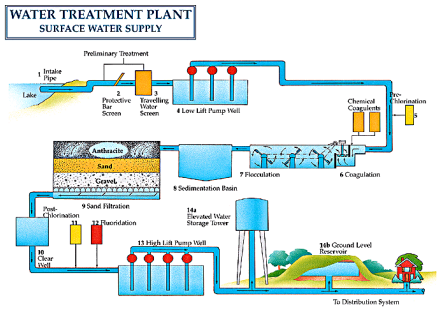
Attachment A:
Environment and Natural Resources Trust Fund
M.L. 2018 Budget Spreadsheet

Project Title: Evaluate Emerging Pathogens in Lakes, Rivers, and Tap Water to Keep Drinking Water Safe
Legal Citation:
Project Manager: Timothy M. LaPara
Organization: University of Minnesota
College/Department/Division: Department of Civil, Environmental, and Geo- Engineering
M.L. 2018 ENRTF Appropriation: $325,000
Project Length and Completion Date: 3 years; June 30, 2021
Date of Report: February 19, 2018

ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Budget	Amount Spent	Balance
BUDGET ITEM			
Personnel (Wages and Benefits) - Overall	$272,500	$0	$272,500
Project Management, Timothy LaPara ($53,877: $17,431 in year 1, $17,954 in year 2, $18492 in year 3;8.3% FTE 75% to salary, 25% to fringe benefits)			
Project Management, Raymond Hozalski ($59,761: $19334 in year 1, $19,914 in year 2, $20512 in year 3;8.3% FTE 75% to salary, 25% to fringe benefits)			
Post-Doc at U of M ($124,126; 1 FTE; 82% to salary, 18% to benefits)			
Undergraduate Researchers ($34,734; 1.4 FTE; 100% salary, 0% to benefits)			
Professional/Technical/Service Contracts	$15,000	$0	$15,000
UMGC for PCR and Illumina Sequencing			
Equipment/Tools/Supplies - Overall (estimated below)	$27,500	$0	$27,500
General lab supplies ($4500)			
IDEXX Supplies for measuring E coli and coliforms ($3000)			
Peristaltic pumps for sample collection (3; each = $2500)			
Membrane filters ($2500)			
Portable power generators (3; each = $1000)			
qPCR Supplies ($5000)			
DNA Extraction Kits ($1200)			
PCR purification kits ($800)			
Travel expenses in Minnesota			
Travel to water utilities in Minnesota to collect water samples; travel to MN water conferences to present research; travel to water utilities to disseminate results	$5,000	$0	$5,000
Other			
Open-access Publication Fees	$5,000	$0	$5,000
COLUMN TOTAL	**$325,000**	**$0**	$325,000

Evaluate Emerging Pathogens in Lakes, Rivers, and Tap Water to Keep Drinking Water Safe





We will quantify *Legionella* and *Mycobacteria* from surface water to tap water