



Environment and Natural Resources Trust Fund

2021 Request for Proposal

General Information

Proposal ID: 2021-160

Proposal Title: Establishment of Safe Water Reuse

Project Manager Information

Name: Satoshi Ishii

Organization: U of MN, College of Food, Agricultural and Natural Resource Sciences

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Project Basic Information

Project Summary: The goal of this project is to contribute to the establishment of safe water reuse in Minnesota by clarifying the potential health risks associated with water reuse.

Funds Requested: \$362,000

Proposed Project Completion: 2024-06-30

LCCMR Funding Category: Water Resources (B)

Project Location

What is the best scale for describing where your work will take place?

Region(s): Metro

What is the best scale to describe the area impacted by your work?

Statewide

When will the work impact occur?

During the Project and In the Future

Narrative

Describe the opportunity or problem your proposal seeks to address. Include any relevant background information.

Stormwater has potential to be a useful resource in the State of Minnesota. Water reuse can provide many benefits, such as reduced use of highly treated drinking water for watering the lawn or toilet flushing, reduced pressure on groundwater aquifers and surface waters, better control of pollutants in stormwater, and long-term savings for irrigation. But the risks of water reuse to public health are not fully understood, limiting its implementation. Even if the water is not consumed, water reuse applications likely pose some risk to public health, which needs to be understood so that water reuse applications can proliferate without compromising public health. Our previous studies have detected genes (DNA) for potential pathogens (i.e., microbes capable of making people sick) as well as antibiotic resistance genes (ARGs) in water reuse systems. But dead pathogens can be also detected by DNA-based assays, so detection of genes does not necessarily reflect a genuine risk. In addition, non-pathogenic bacteria may also carry ARGs. Additional research is therefore needed to determine the risk posed by live pathogenic and antibiotic resistant microorganisms in the water being reused.

What is your proposed solution to the problem or opportunity discussed above? i.e. What are you seeking funding to do? You will be asked to expand on this in Activities and Milestones.

The proposed project will (1) quantify various live infectious pathogens in Minnesota's water reuse systems, (2) analyze antibiotic resistance of the isolated bacteria, and (3) assess potential human health risks associated with the water reuse systems.

We will use culture (growth)-based analysis to detect infectious pathogens and clarify whether pathogenic bacteria are carrying the ARGs. By quantifying the levels of infectious pathogens with culture-based analysis and identifying the host of ARGs, it becomes possible to assess potential health risks associated with water reuse for various applications such as toilet flushing and irrigation.

What are the specific project outcomes as they relate to the public purpose of protection, conservation, preservation, and enhancement of the state's natural resources?

The overall goal of this project is to contribute to the establishment of safe water reuse by clarifying the potential health risks associated with water reuse. The final outcome of this project, information needed to develop water quality recommendations for safe water reuse, can be used by agencies working to develop regulatory and non-regulatory guidance for safe and sustainable reuse in Minnesota and help to increase water reuse in Minnesota. In turn, increased water reuse will conserve our drinking water supplies in groundwater aquifers and improve surface water quality.

Activities and Milestones

Activity 1: Quantify various infectious pathogens in Minnesota's water reuse systems

Activity Budget: \$232,000

Activity Description:

We will collect samples from water reuse facilities in Minnesota every two to three weeks during the reuse seasons (May-November) for two years (2021-2022). If water is treated (filtration, disinfection, etc.) or stored (stormwater pond) before reuse, both source water and treated water samples will be collected. Environmental parameters such as temperature and precipitation will be recorded for each site. General water quality parameters such as the nutrient concentrations and the levels of fecal indicator bacteria will be also measured. Pathogens previously frequently detected by DNA-based analysis as well as antibiotic-resistant bacteria will be isolated from the water samples using culture-based methods. In addition to culture-based analysis, DNA-based pathogen and ARG quantification will be also performed. By comparing the results obtained by culture-based and DNA-based methods, the proportion of infectious pathogens out of total pathogens will be calculated.

Activity Milestones:

Description	Completion Date
Water sample collection and characterization (from 3-4 facilities over 2 years)	2022-12-31
Isolation and quantification of pathogens and antibiotic resistant bacteria in water samples	2023-06-30

Activity 2: Analyze antibiotic resistance of the isolated bacteria

Activity Budget: \$67,000

Activity Description:

Bacteria isolated in Activity 1 will be analyzed for their levels of antibiotic resistance and the possession of ARGs. Bacterial species will be identified by gene sequencing analysis. Genomes of the representative strains will be also sequenced to analyze the possibility of ARGs being horizontally transferred between different bacteria (e.g., from non-pathogens to pathogens).

Activity Milestones:

Description	Completion Date
Species name of the antibiotic resistant bacteria isolated from water samples	2023-06-30
Genome of the antibiotic resistant bacteria strains	2023-12-31

Activity 3: Assess potential health risks associated with the reuse water systems

Activity Budget: \$63,000

Activity Description:

Quantity of potentially infectious pathogens obtained in Activity 1 will be used to assess potential human health risks associated with the water samples. Results of these analyses could then be used to develop water quality recommendations for best public health practices in Minnesota. In addition to written reports, we will discuss outcomes with managers of local water reuse facilities in various venues, such as conferences, training sessions, workshops or stakeholder meetings. Through these activities, we will contribute to the establishment of safe water reuse in Minnesota.

Activity Milestones:

Description	Completion Date
Events to disseminate the results	2024-06-30
Assessment of potential health risks associated with the reuse water systems	2024-06-30

Project Partners and Collaborators

Name	Organization	Role	Receiving Funds
Nancy Rice	Minnesota Department of Health	Data analysis	No
Anita Anderson	Minnesota Department of Health	Technical support	No
Timothy LaPara	University of Minnesota	Co-supervision of a post-doctoral researcher.	Yes

Long-Term Implementation and Funding

Describe how the results will be implemented and how any ongoing effort will be funded. If not already addressed as part of the project, how will findings, results, and products developed be implemented after project completion? If additional work is needed, how will this be funded?

This project will maximize the potential of water reuse to conserve Minnesota's groundwater and improve surface water quality by providing the pathogen/ARB data needed to eliminate barriers to water reuse. This research will help provide assurances to the public, regulators and system owners that water reuse can become common practice without negative effects on public health and safety.

Other ENRTF Appropriations Awarded in the Last Six Years

Name	Appropriation	Amount Awarded
Assessment of Water Quality for Reuse	M.L. 2017, Chp. 96, Sec. 2, Subd. 04f	\$148,000

Project Manager and Organization Qualifications

Project Manager Name: Satoshi Ishii

Job Title: Associate Professor

Provide description of the project manager's qualifications to manage the proposed project.

Satoshi Ishii is Associate Professor in the BioTechnology Institute (BTI) and the Department of Soil, Water, and Climate (SWC) at the University of Minnesota. Dr. Ishii's research focuses on environmental microbiology and biotechnology, including water quality and public health microbiology. He has led several projects related to water quality and reuse, funded by LCCMR and Minnesota Stormwater Research Council (MSRC).

Organization: U of MN - Twin Cities

Organization Description:

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.

Budget Summary

Category / Name	Subcategory or Type	Description	Purpose	Gen. Ineligible	% Benefits	# FTE	Classified Staff?	\$ Amount
Personnel								
2 Professors		Project supervision, supervision of a post-doctoral researcher, project reporting.			27%	0.48		\$76,000
Postdoctoral research associate		Collect samples, perform lab experiments, and analyze data for Activities 1, 2, and 3; write reports			20%	3		\$186,000
2 Undergraduate research technicians		Sample collection and processing, water quality analysis for Activity 1			0%	0.54		\$13,000
							Sub Total	\$275,000
Contracts and Services								
University of Minnesota Genomics Center	Internal services or fees (uncommon)	Microfluidic qPCR user fee (Approx \$300/run x 12 runs); Complete genome sequencing (Approx. \$5,000 for 8 genomes)				0		\$9,000
							Sub Total	\$9,000
Equipment, Tools, and Supplies								
	Equipment	CO2 incubator	Necessary to grow enteric viruses					\$6,000
	Tools and Supplies	Field supplies (a high-performance vacuum pump: \$1,800; tubings: \$1,000; other supplies: \$400)	Necessary to collect water samples from water reuse facilities					\$3,000
	Tools and Supplies	Lab supplies (Membrane filters: 400 samples at \$20/sample; DNA/RNA extraction kits: 400 samples at \$10/sample; Reagents for microfluidic qPCR pathogen and ARG detections: 400 samples at \$60/sample; general water quality analysis: 400 samples at \$20/sample; Culture media for pathogen isolation \$4,000; Quanti-Tray kits for fecal indicator bacteria testing: \$4,800; Glassware and plastic consumables: \$4,000)	Necessary to grow and detect pathogens and antibiotic resistant bacteria in water samples					\$57,000

							Sub Total	\$66,000
Capital Expenditures								
							Sub Total	-
Acquisitions and Stewardship								
							Sub Total	-
Travel In Minnesota								
	Miles/ Meals/ Lodging	In-state travel to collect samples (Approximately 8,700 miles at \$0.575/mile per U of M travel policy)	Necessary to collect water samples from several water reuse facilities in Minnesota					\$5,000
							Sub Total	\$5,000
Travel Outside Minnesota								
							Sub Total	-
Printing and Publication								
	Publication	Open access publication fee (\$2,500/publication x 2 publications)	Necessary to make the results publicly available					\$5,000
							Sub Total	\$5,000
Other Expenses								
		Equipment repair and maintenance	Necessary for maintaining existing incubators, biosafety cabinet, etc.					\$2,000
							Sub Total	\$2,000
							Grand Total	\$362,000

Classified Staff or Generally Ineligible Expenses

Category/Name	Subcategory or Type	Description	Justification Ineligible Expense or Classified Staff Request
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Non ENRTF Funds

Category	Specific Source	Use	Status	Amount
State				
Cash	Minnesota Stormwater Research Council (5/1/2020-11/31/2021)	Stormwater samples will be collected from several reuse facilities in the Twin Cities Metro area, for the quantification of pathogens and antibiotic resistance genes. This MSRC-funded research does not grow pathogens and antibiotic resistant bacteria by culture-based methods, and therefore, there is a minimum overlap between the MSRC-funded research and the proposed LCCMR research.	Secured	\$59,756
In-Kind	Minnesota Department of Health	Minnesota Department of Health provides salary + benefits to its members for this project.	Secured	\$10,000
In-Kind	University of Minnesota	The University of Minnesota is not allowed to charge the State of Minnesota its typical overhead rate of 55% of the total modified direct costs. We are listing our unrecoverable indirect cost as in-kind contribution.	Secured	\$195,800
			State Sub Total	\$265,556
Non-State				
			Non State Sub Total	-
			Funds Total	\$265,556

Attachments

Required Attachments

Visual Component

File: [8d611653-a03.pdf](#)

Alternate Text for Visual Component

This graphic shows how this project contributes to maximizing the benefit of water reuse in improving surface water quality and reducing demand on groundwater aquifers in Minnesota.

Optional Attachments

Support Letter or Other

Title	File
Letter from MDH	026f1f48-ee7.pdf

Administrative Use

Does your project include restoration or acquisition of land rights?

No

Does your project have patent, royalties, or revenue potential?

No

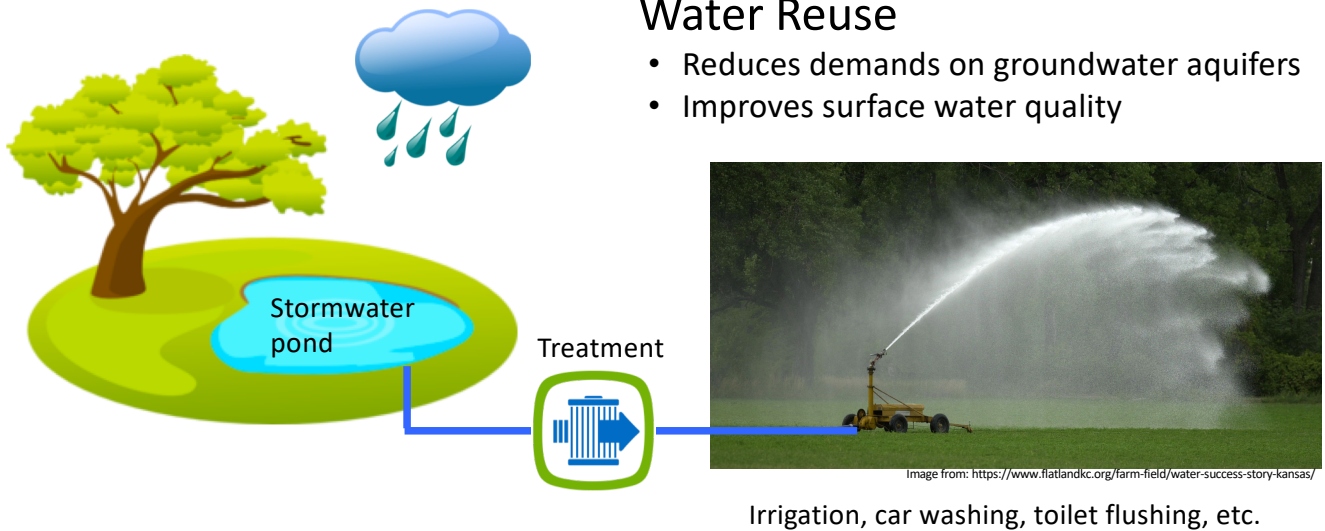
Does your project include research?

Yes

Does the organization have a fiscal agent for this project?

Yes, Sponsored Projects Administration

Establishment of Safe Water Reuse



Barrier for to the expansion of water reuse

- Public health concerns
 - Pathogens
 - Antibiotic resistant bacteria (ARB)

Currently, no water quality standard or recommendation available for reuse

Project Goal

To contribute to the establishment of safe water reuse in Minnesota by clarifying the potential health risks associated with water reuse.

Activities

1. Quantify various infectious pathogens in Minnesota's water reuse systems
2. Analyze antibiotic resistance of the isolated bacteria
3. Assess potential health risks associated with the reuse water systems

Project Team

- Satoshi Ishii (U of M)
- Tim LaPara (U of M)
- Anita Anderson (MN Dept. Health)
- Nancy Rice (MN Dept. Health)



