Environment and Natural Resources Trust Fund (ENRTF)
M.L. 2019 ENRTF Work Plan (Main Document)

Today's Date:	August 3, 2018
Date of Next Status Update Report:	March 1, 2020
Date of Work Plan Approval:	
Project Completion Date:	June 30, 2022

Does this submission include an amendment request? __

PROJECT TITLE:	Stimulating bacteria to degrade chlorinated industrial contaminants
Project Manager:	Paige J. Novak
Organization:	University of Minnesota
College/Department/Division:	Department of Civil, Environmental, and Geo- Engineering
Mailing Address:	122 Civil Engineering Building, 500 Pillsbury Drive SE
City/State/Zip Code:	Minneapolis, MN 55455
Telephone Number:	(612) 626-9846
Email Address:	novak010@umn.edu
Web Address:	N/A

Location: Statewide

Total Project Budget:	$150,000
Amount Spent:	$0
Balance:	$150,000

Legal Citation: M.L. 2019, Chp. xx, Sec. xx, Subd. xx

Appropriation Language:

I. PROJECT STATEMENT:

Minnesota contains a large number of contaminated sites that require clean-up at a large cost. Indeed, according to the Minnesota Pollution Control Agency's most recent report, there are 92 contaminated sites on the Minnesota "Superfund" List. These sites are either abandoned or the contamination is uncontrolled, causing concern. At these sites alone, $13,500,000 was spent in FY 2015-16 on clean-up tasks. In addition to these Superfund sites, there are 621 additional contaminated sites in Minnesota that currently require clean-up. Over half of the contaminated sites in Minnesota contain chlorinated pollutants that are known or suspected to cause serious human health effects. Research is needed to develop ways to affordably clean up chlorinated pollutants, safeguarding current and future human and economic health.

Interestingly, bacteria exist that can "breathe" toxic chlorinated pollutants (so-called halorespiring bacteria). To survive, however, they require the presence of chlorinated pollutants. As a result, higher concentrations of chlorinated pollutants typically sustain these organisms more effectively. Nevertheless, during remediation, we want to remove or degrade chlorinated pollutants to very low concentrations, which can make it difficult to sustain these halorespiring bacteria. If these bacteria are being used to clean-up a site containing chlorinated pollutants, the result can be a "stalling" of the process at concentrations of pollutant that are too low to sustain the halorespiring bacteria, but too high to be protective of human and ecological health.

Natural chlorinated compounds (not pollutants) also exist in low concentrations in uncontaminated sites as a natural part of soil. In our research, we have found that these natural chlorinated compounds can stimulate pollutant dechlorination in both halorespiring bacteria and other bacteria that use the dechlorinated carbon for growth, called "non-respiratory dechlorinators." We suspect that these non-respiratory dechlorinators are able to dechlorinate pollutants to lower concentrations because, while they can use them and therefore degrade them, those bacteria do not rely solely on the chlorinated pollutants to survive.

We hypothesize that amendments with different amounts of soil-based carbon versus natural (non-pollutant) chlorinated compounds will stimulate halorespiring bacteria and non-respiratory dechlorinators differently. This can be used to verify that non-respiratory dechlorinators can dechlorinate pollutants to desired low concentrations and enable the addition of amendments to control the rate and extent of pollutant dechlorination based on the amount of pollutant present. In the proposed research we will test this hypothesis with the goal of determining the best way to stimulate both groups of bacteria with natural compounds for pollutant dechlorination to low concentrations, saving money and time, and reducing risk.

II. OVERALL PROJECT STATUS UPDATES:

First Update March 1, 2020

Second Update September 1, 2021

Third Update March 1, 2021

Fourth Update September 1, 2022

Fifth Update March 1, 2022

Final Report between project end (June 30) and August 15, 2022

III. PROJECT ACTIVITIES AND OUTCOMES:

ACTIVITY 1 Title: Determine how different amendments of natural compounds improve dechlorination

Description:
Experiments will be performed with the common pollutant trichloroethene (TCE) and sediment from contaminated and uncontaminated sites containing different initial amounts of TCE.

ACTIVITY 1 ENRTF BUDGET:

ENRTF Budget:	**$70,000**
Amount Spent:	**$0**
Balance:	**$70,000**

Outcome	Completion Date
*1. Measure the dechlorination of PCE and TCE in sediments with **high ratios** of soil-based carbon to natural chlorinated compounds when amended with stimulants of varying ratios of soil-based carbon to natural chlorinated compounds*	*6/30/21*
*2. Measure the dechlorination of PCE and TCE in sediments with **low ratios** of soil-based carbon to natural chlorinated compounds when amended with stimulants of varying ratios of soil-based carbon to natural chlorinated compounds*	*6/30/21*

First Update March 1, 2020

Second Update September 1, 2021

Third Update March 1, 2021

Fourth Update September 1, 2022

Fifth Update March 1, 2022

Final Report between project end (June 30) and August 15, 2022

ACTIVITY 2 Title: Determine how the different groups of dechlorinating bacteria ("halorespiring" and "non-respiratory dechlorinators") are affected by these amendments

Description:
Genes are the codes that "tell" organisms which functions to perform (such as breathing chlorinated compounds). By analyzing genes, we can understand which organisms dominate (and by what mechanism) in a given sample. Samples will be taken from the experiments described above and the genetic material will be extracted and analyzed over time. From this we will learn which genes are stimulated by the different amendments, which genes are responsible for different patterns of dechlorination, and which genes are initially present in different types of starting materials. By understanding how to "read" the genes used to dechlorinate pollutants, we will know the best amendment to add to stimulate dechlorination at a site without having to perform labor-intensive and expensive experiments.

ACTIVITY 2 ENRTF BUDGET:

ENRTF Budget:	**$80,000**
Amount Spent:	**$0**
Balance:	**$80,000**

Outcome	Completion Date
1. Analyze the genes in initial starting material for the experiments	*1/31/20*

2. Analyze the types and quantities of genes in the experiments described in Activity 1 over time	*5/1/22*

First Update March 1, 2020

Second Update September 1, 2021

Third Update March 1, 2021

Fourth Update September 1, 2022

Fifth Update March 1, 2022

Final Report between project end (June 30) and August 15, 2022

IV. DISSEMINATION:

Description:
The target audience for results from this research will be professionals in the area of hazardous waste treatment. Specific targets will be environmental engineers and scientists in academia, industry, state agencies such as the MPCA, and environmental consultants. Results will be disseminated through scholarly publications in peer-reviewed journals such as *Environmental Science and Technology*. Results from the research project will also be presented at regional conferences such as the *Minnesota Water* conference.

The Minnesota Environment and Natural Resources Trust Fund (ENRTF) will be acknowledged through use of the trust fund logo or attribution language on project print and electronic media, publications, signage, and other communications per the ENRTF Acknowledgement Guidelines.

First Update March 1, 2020

Second Update September 1, 2021

Third Update March 1, 2021

Fourth Update September 1, 2022

Fifth Update March 1, 2022

Final Report between project end (June 30) and August 15, 2022

V. ADDITIONAL BUDGET INFORMATION:
A. Personnel and Capital Expenditures

Explanation of Capital Expenditures Greater Than $5,000: N/A

Explanation of Use of Classified Staff: N/A

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

Enter Total Estimated Personnel Hours for entire duration of project: 2,160	Divide total personnel hours by 2,080 hours in 1 yr = TOTAL FTE: 0.35 FTE/yr

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

Enter Total Estimated Contract Personnel Hours for entire duration of project: N/A	Divide total contract hours by 2,080 hours in 1 yr = TOTAL FTE: N/A

VI. PROJECT PARTNERS:

A. Partners outside of project manager's organization receiving ENRTF funding
None

B. Partners outside of project manager's organization NOT receiving ENRTF funding
None

VII. LONG-TERM- IMPLEMENTATION AND FUNDING:
Minnesota has impressive environmental resources but also a large number of sites that need to be remediated at a large cost. Novak has worked on the halorespiration of chlorinated pollutants for about 20 years. She is the first to perform research on the existence of halorespiring bacteria in uncontaminated environments and the first to show that pollutant degradation can be stimulated through the addition of uncontaminated soil extracts to the bacteria present. The goal of this project is to identify how the organisms that naturally cycle chlorine in uncontaminated Minnesota environments can best be deployed to detoxify chlorinated pollutants. This research should enable the development of new remediation technologies that are more effective and less expensive than those currently used, cleaning more sites and improving Minnesota's environment.

VIII. REPORTING REQUIREMENTS:

- Project status update reports will be submitted March 1 and September 1 each year of the project
- A final report and associated products will be submitted between June 30 and August 15, 2022

IX. SEE ADDITIONAL WORK PLAN COMPONENTS:

A. Budget Spreadsheet
B. Visual Component or Map
C. Parcel List Spreadsheet: N/A
D. Acquisition, Easements, and Restoration Requirements: N/A
E. Research Addendum

Attachment A:
Environment and Natural Resources Trust Fund
M.L. 2019 Budget Spreadsheet
Legal Citation:
Project Manager: Paige J. Novak
Project Title: Stimulating bacteria to degrade chlorinated industrial contaminants
Organization: University of Minnesota
Project Budget: $150,000
Project Length and Completion Date: 3 years, June 30, 2022
Today's Date: 8/3/18

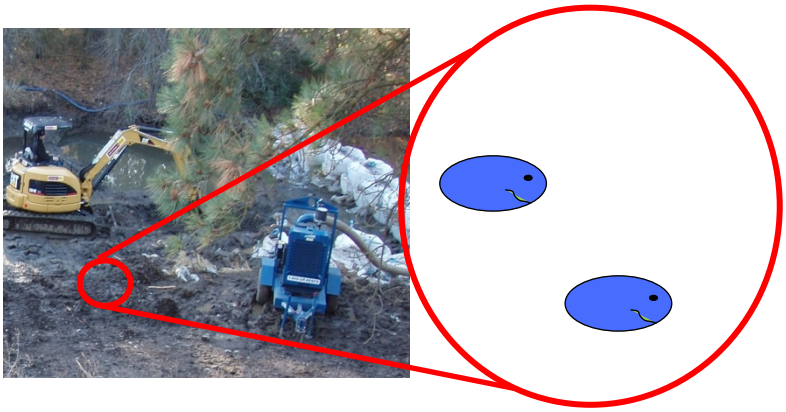
ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Budget	Amount Spent	Balance
BUDGET ITEM			
Personnel (Wages and Benefits)	$ 106,352	$ -	$ 106,352
Novak (PI, 1% time per year for three years, salary 75% of cost, fringe benefits 25% of cost). Project supervision, provide guidance experimental design and sample analysis. Total estimated cost is $11,136. Graduate student (33% time per year for three years, 57% salary, 32% tuition, 11% fringe benefits). Conducting laboratory experiments and analyzing samples using chemical and genetic techniques. Total estimated cost is $95,216.			
Professional/Technical/Service Contracts			
	$ -	$ -	$ -
Equipment/Tools/Supplies			
Funds for laboratory supplies are requested ($11,000/year). This includes, but is not limited to: DNA soil extraction kits, materials for quantifying genes present, primers for deep genetic sequencing, pipette tips, eppendorf tubes, glassware, chemicals for standards and experiments, analytical consumables, analytical fees, solvents, reagents, and gloves. Funds ($8,000 total) are also requested for sequencing via Illumina sequencing. Additional funds budgeted for equipment repair and maintenance ($2,648).	$ 43,648	$ -	$ 43,648
	$ -	$ -	$ -
COLUMN TOTAL	$ 150,000	$ -	$ 150,000

OTHER FUNDS CONTRIBUTED TO THE PROJECT	Status (secured or pending)	Budget	Spent	Balance
Non-State:		$ -	$ -	$ -
State:		$ -	$ -	$ -
In kind: Novak and LaPara will provide unpaid time to the project (including 2% cost-share). Because the project is overhead-free, laboratory space, electricity, and other overhead costs are provided in kind. The University of Minnesota overhead rate is 54%.		$ -	$ -	$ -

PAST AND CURRENT ENRTF APPROPRIATIONS	Amount legally obligated but not yet spent	Budget	Spent	Balance
Current appropriation:		$ -	$ -	$ -
Past appropriations:		$ -	$ -	$ -

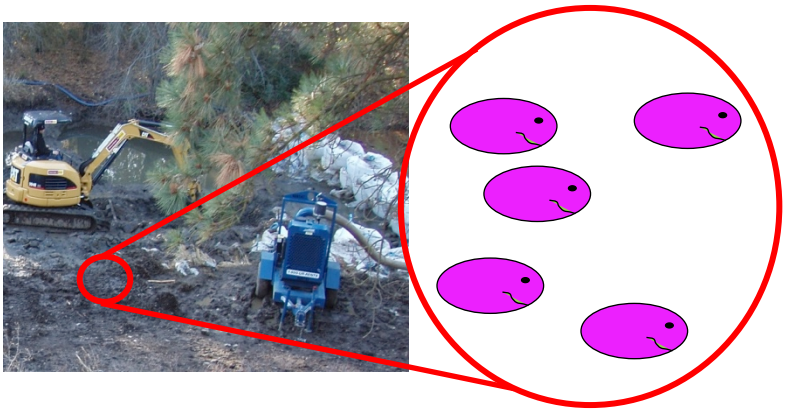
Stimulating bacteria to degrade chlorinated industrial contaminants

The problem



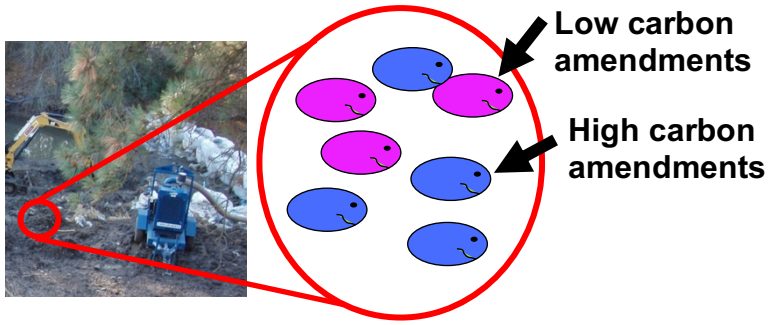
- Bacteria exist that "breathe" chlorinated contaminants (**blue bacteria**)
- Bacteria need higher concentrations of contaminants to thrive
- There is often too little contamination to support the bacteria, but too much to consider the site safe

A potential solution



- Other bacteria exist that dechlorinate compounds but don't "breathe" these compounds (**pink bacteria**)
- They work more slowly, but may degrade the contaminants to lower concentrations, cleaning the site to a greater extent

What we need to know



- How do we best stimulate both types of bacteria for fast and complete dechlorination?
- How can we monitor their progress without expensive and time-consuming experiments?