# Environment and Natural Resources Trust Fund
# 2012-2013 Request for Proposals (RFP)

**Project Title:** **ENRTF ID:** **151-I**

Wastewater Treatment to Destroy Antibiotic Resistance: Minnesotas Story

**Topic Area:** I. Water Resources

**Total Project Budget: $** 148,855

**Proposed Project Time Period for the Funding Requested:** 2 yrs, July 2013 - June 2015

**Other Non-State Funds: $** 0

**Summary:**

This project will evaluate the ability of Minnesotas wastewater treatment facilities to eliminate antibiotic resistant bacteria, thus providing protection to Minnesotas water, soil, and public health.

**Name:** Timothy LaPara

**Sponsoring Organization:** U of MN

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**Location**

**Region:** Statewide

**County Name:** Statewide

**City / Township:**

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

**PROJECT TITLE: Wastewater Treatment to Destroy Antibiotic Resistance: Minnesota's Story**

## I. PROJECT STATEMENT

Antibiotics are undoubtedly one of the greatest scientific discoveries of the 20th century. Antibiotics are extraordinarily effective at treating many common bacterial infections, such as urinary tract infections, conjunctivitis (pink eye), and ear infections; antibiotics are also critically important for much more dangerous bacterial diseases, such as tuberculosis and systemic staph-infections. Paradoxically, our extensive use, over-use, and mis-use of antibiotics since World War II have led to an increased prevalence of antibiotic resistance (i.e., the existence of bacterial infections for which antibiotic therapy is ineffective). Indeed, resolving the problem of antibiotic resistance represents one of the critical scientific challenges of the 21st century. Current efforts to slow the spread of antibiotic resistance currently focus on preventing the development of additional resistant bacteria by reducing antibiotic use and mis-use. Unfortunately, this approach is inherently flawed because appropriate antibiotic use will continue, and thus antibiotic resistance will continue to exist (and become more common).

For about a decade, I have been recommending the use of more stringent wastewater treatment technology to help ameliorate the problem of antibiotic resistance. My rationale is that people ingesting antibiotics shed large quantities of resistant bacteria when they defecate; thus, our wastewater treatment infrastructure is well-positioned to actively kill/inactivate the overwhelming majority of antibiotic resistant bacteria.

My research continues to demonstrate that wastewater treatment facilities can be effectively used to meet this goal. For example, I co-authored a manuscript in 2010 that demonstrated that high temperature anaerobic digestion is significantly more effective than "standard" anaerobic digestion (this research was financially supported by the Minnesota Environment and Natural Resources Trust Fund). Conversely, my research shows that changes to our existing facilities are required to meet this new goal; specifically, I co-authored another manuscript in which one of Minnesota's BEST wastewater treatment facilities was shown to discharge wastewater that contained more than 100-times higher than the background concentration of antibiotic resistance genes. Both of these publications were extremely well-received by the scientific community (both were highlighted in *Chemical and Engineering News*, a trade journal promoting research published by the American Chemical Society) and the local/regional/national news media (I have appeared on Minnesota Public Radio and the evening news on Duluth television; my research has been profiled on the homepage of the National Science Foundation and in the University's Alumni Magazine).

In my most recent study (in collaboration with Kristine Wammer and Dwight Stoll; funding provided by the Minnesota Environment and Natural Resources Trust Fund), we have learned that the performance of wastewater treatment facilities varies substantially (as expected), presumably depending on their design. This leads to the question: How do the various wastewater treatment facilities in Minnesota compare with respect to their ability to destroy antibiotic resistant bacteria? To answer this question, I propose a study in which treated wastewater (discharged to Minnesota's rivers and lakes) and wastewater sludge (applied to Minnesota's agricultural land, among other places) is compared. This research will provide invaluable information to assess the ability of Minnesota's wastewater treatment infrastructure to help protect Minnesotans against the spread of antibiotic resistance. More importantly, the proposed research will help identify opportunities to improve Minnesota's wastewater treatment facilities to kill/inactivate/destroy antibiotic resistant bacteria.

## II. DESCRIPTION OF PROJECT ACTIVITIES

In this project, influent wastewater, treated wastewater, untreated wastewater sludge, and treated wastewater sludge samples will be collected from as many wastewater treatment facilities as possible. We will collect at least three different samples from each facility so that variation within a facility can be assessed. We will attempt to collect effluent samples during both the summer (while effluent disinfection is performed) and the winter (when effluent disinfection is not required) to discern the relative importance

of effluent disinfection. We will catalog wastewater treatment facilities according to their size (i.e., flow rate), wastewater type (i.e., the relative importance of residential, commercial, and/or industrial wastewater), process design, and other pertinent factors. In general, our approach will be to maintain the anonymity of wastewater treatment facilities to encourage their participation in the project. If we have difficulty obtaining effluent samples from different treatment facilities, then we can directly collect treated wastewater samples at their sewage outfalls, which are always located in publicly-accessible waterways.

Total genomic DNA will be extracted and purified from these samples and then used as template to quantify specific genes known to encode antibiotic resistance by quantitative real-time PCR. As of April 2012, my laboratory has substantial experience in quantifying more than 5 genes that encode for resistance to tetracycline, one gene that encodes for resistance to erthyromycin, one gene that encodes for sulfa drugs, and one gene involved in Class 1 integrons (Class 1 integrons are associated with multiple antibiotic resistance). We will also quantify total *Bacteroides* spp. (a measure of total fecal material) and human-specific *Bacteroides* spp. (a measure of human fecal material). This research will allow us to identify which of Minnesota's wastewater treatment facilities perform the best with respect to eliminating antibiotic resistant bacteria and antibiotic resistance genes, allowing us to make recommendations to the other wastewater treatment facilities regarding various alternatives to improve their performance with respect to this newly-emerging category of contaminant.

**Activity 1:** *Sample collection and quantification of specific microbes* **Budget:** $148,855

*Include detailed description of the activity you are proposing to do here.*

| **Outcome** | **Completion Date** |
|---|---|
| *1. Sample Collection* | July 2014 |
| *2. Genomic DNA extraction and purification* | August 2014 |
| *3. Quantification of specific microorganisms by quantitative real-time PCR* | December 2014 |

## III. PROJECT STRATEGY

### A. Project Team/Partners

Dr. Timothy M. LaPara (Department of Civil Engineering, University of Minnesota) will be responsible for coordinating the entire project and co-mentoring of a graduate student (yet to be hired). Dr. LaPara has considerable expertise in municipal wastewater treatment and environmental microbiology, having published more than 40 manuscripts that have been published in the peer-reviewed literature.

### B. Timeline Requirements

The first year of this project will be devoted to collecting (and preserving) wastewater and wastewater solids samples and training the graduate student on the techniques required to quantify the specified organisms. The second year will be devoted to quantifying the organisms of concern (this should take 3-6 months), collating the data and interpreting its meaning (this should take < 3 months), and sharing the results with interested end-users. We anticipate that, in addition to the unnamed participating wastewater treatment facilities, the Minnesota Department of Health, and the Central States Water Environment Association will all be interested parties.

### C. Long-Term Strategy and Future Funding Needs

The proposed project will be completed within the two-year project period. Additional work could be carried out (at additional wastewater treatment facilities) in the future, with the cost borne by the individual wastewater treatment facilities (or other sources).

# 2012-2013 Detailed Project Budget

## IV. TOTAL ENRTF REQUEST BUDGET: 2 years

| BUDGET ITEM | AMOUNT |
|---|---|
| **Personnel:** Timothy M. LaPara, Project Manager. 6 weeks of salary per year plus associated fringe benefits. Duties: Project management, graduate student supervision, results dissemination | $ 47,010 |
| **Personnel:** Graduate Student, University of Minnesota. Funding for a graduate student for two years plus associated fringe benefits (including tuition). Duties: sample collection, quantification of genes, assistance with report preparation | $ 83,344 |
| **Equipment/Tools/Supplies:** Supplies to collect samples (vaccuum pump, glassware, filters, etc), extract genomic DNA & RNA (commercially available kits), and perform quantitative polymerase chain reaction (reagents) | $ 12,500 |
| **Travel:** *Travel to Central Minnesota to collect wastewater/sludge samples* | $ 6,000 |
| **TOTAL ENVIRONMENT AND NATURAL RESOURCES TRUST FUND $ REQUEST =** | $ 148,855 |

## V. OTHER FUNDS

| SOURCE OF FUNDS | AMOUNT | Status |
|---|---|---|
| **Other Non-State $ Being Applied to Project During Project Period:** | $ - | |
| **Other State $ Being Applied to Project During Project Period:** | $ - | |
| **In-kind Services During Project Period:** | $ - | |
| **Remaining $ from Current ENRTF** | $ - | |
| **Funding History:** | $ - | |

## Project Manager Qualifications

***Timothy M. LaPara,* Associate Professor, Department of Civil Engineering, University of Minnesota**

Education
B. S. C. E., 1995, Civil Engineering, University of Notre Dame
Ph.D., 1999, Environmental Engineering, Purdue University

Research and Teaching
Dr. LaPara's research is focused on the role of drinking water treatment, municipal wastewater treatment, and industrial wastewater treatment in preserving environmental quality and in protecting public health. Dr. LaPara teaches courses in the design of municipal water and wastewater treatment facilities and in environmental microbiology.

**Organization Description**
The University of Minnesota-Twin Cities is the state of Minnesota's largest institution of higher education