Environment and Natural Resources Trust Fund (ENRTF)
M.L. 2014 Work Plan

Date of Report: January 14, 2014

Date of Next Status Update Report: January 31, 2015

Date of Work Plan Approval:

Project Completion Date: June 30, 2017

Does this submission include an amendment request? _No_

PROJECT TITLE: Evaluation of Wastewater Nitrogen and Estrogen Treatment Options

Project Manager: Paige J. Novak

Organization: University of Minnesota

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Location: Statewide

Total ENRTF Project Budget:		
	ENRTF Appropriation:	**$500,000**
	Amount Spent:	**$0**
	Balance:	**$500,000**

Legal Citation: M.L. 2014, Chp. 226, Sec. 2, Subd. 03d

Appropriation Language:
$500,000 the second year is from the trust fund to the Board of Regents of the University of Minnesota to examine the performance of new wastewater contaminant treatment options under Minnesota weather conditions in order to understand how to improve wastewater treatment of nitrogen and estrogenic compounds, decrease costs and energy use, and safeguard aquatic species. This appropriation is available until June 30, 2017, by which time the project must be completed and final products delivered.

I. PROJECT TITLE: Wastewater estrogen: removal options, fish abundance, and cost

II. PROJECT STATEMENT:
Wastewater treatment plants discharge effluent that contains contaminants of emerging concern (CECs), including estrogens. These estrogens have caused dramatic ecological effects such as fish feminization and fish population collapses, with unknown long-term consequences. The most important estrogen exiting wastewater treatment plants, in terms of contributing to the feminization potential of effluent, is a chemical called estrone, which is an estrogen that is released naturally from women via the waste stream. Although this and other estrogens are present in Minnesota lakes and rivers and can be ecologically harmful, their treatment and discharge are not regulated.

Interestingly, the discharge of estrone and other estrogens is a function of how (and how well) a treatment plant removes nitrogen. Nitrogen discharge is regulated to some extent in Minnesota and will be more heavily regulated in the future, requiring additional wastewater treatment plant upgrades. With this research we will determine how different nitrogen removal processes perform over the range of temperatures experienced in Minnesota with respect to both CEC (and in particular, estrone) and nitrogen removal so that the very best processes for the protection of Minnesota's natural resources can be put into place. In addition, we will determine how fish vulnerability changes seasonally so that treatment to extremely low levels of CECs is only required during critical periods (e.g., during egg maturation or spawning) to save energy and costs from excessive (and unnecessarily rigorous) treatment. Finally, we will combine laboratory efforts with predictive mathematical models so that we can extrapolate to cost and whole population behavior.

III. PROJECT STATUS UPDATES:

Project Status as of January 31, 2015:

Project Status as of July 31, 2015:

Project Status as of January 31, 2016:

Project Status as of July 31, 2016:

Project Status as of January 31, 2017:

Overall Project Outcomes and Results:

IV. PROJECT ACTIVITIES AND OUTCOMES:

ACTIVITY 1: Determine the performance of different wastewater treatment processes with respect to nitrogen removal, CEC and estrone removal, energy use, and cost

Description: Five laboratory-scale reactor systems will be set-up to mimic different wastewater treatment systems, including conventional (NH_3 removal-only) treatment (*CONV*) and four treatment systems designed for total nitrogen removal. Reactors will be constructed from glass and will be designed to mimic the most basic total nitrogen removal process (the Modified Ludzack-Ettinger (MLE) process), a nitritation-denitritation system (*N-D*), an *ANAMMOX* system, and a low-oxygen granular sludge process (*GRAN*). All of the reactors, except for the *GRAN* reactor, will be fed continuously and operated with solids residence times of 10-25 days depending on the reactor. A membrane separation system will be used with each reactor set-up to retain the biomass for recirculation. Experiments will initially be performed at approximately 72°F followed by a second set of experiments performed at approximately 59°F. Two exceptions are the experiments performed with the *N-D* and *ANAMMOX* reactors, which require heating of the aerobic (both, 90°F) and second anaerobic (*ANAMMOX*

only, 97°F) reactors to function optimally. In these cases the feed to the reactors will be heated and the energy used for heating will be incorporated into the cost calculations. When appropriate, reactors will be aerated. The flow rate of oxygen required to meet the dissolved oxygen set-point will be monitored daily and will be incorporated into energy utilization and cost calculations. Reactors will be fed synthetic wastewater amended with estrone. Reactor effluents will be monitored for soluble COD, estrone, NH_3, NO_3^-, NO_2^-, and dissolved oxygen. Two of the experiments will be repeated in triplicate to verify reproducibility. Finally, two to three experiments will be repeated with influent secondary wastewater from the Metropolitan Plant in St. Paul, MN to which estrone has been amended. This will allow verification of the trends observed with synthetic wastewater amended with estrone with a more complex feed. The effluent will also be collected, and the CECs present will be extracted and amended to aquaria in fish exposure experiments (described under Activity 2). We have experience operating similar systems.

Summary Budget Information for Activity 1:

ENRTF Budget: $ 139,250
Amount Spent: $ 0
Balance: $ 139,250

Activity Completion Date: February 28, 2017

Outcome	Completion Date	Budget
1. Nitrogen removal efficiency in five different wastewater treatment plant configurations	10/31/2016	$62,125
2. Estrone/CEC removal efficiency in five different wastewater treatment plant configurations	10/31/2016	$62,125
3. An estimate of the energy use for the various treatment options	02/28/2017	$5,000
4. An estimate for the cost of the various treatment options	02/28/2017	$10,000

Activity Status as of January 31, 2015:

Activity Status as of July 31, 2015:

Activity Status as of January 31, 2016:

Activity Status as of July 31, 2016:

Activity Status as of January 31, 2017:

Final Report Summary:

ACTIVITY 2: Determine how temperature and life stage alter the reproduction and survival of fathead minnows and smallmouth bass after exposure to treated synthetic or real wastewater

Description: We will employ a staggered blocked design using two species, two life stages, two temperatures and five exposure treatments to determine windows of vulnerability for fathead minnows and smallmouth bass to effluent exposure. The life history of non-migrating North American fishes usually contains two life stages during which the fish are assumed to be particularly vulnerable to the effects of environmental estrogens: (i) the embryonic/early larval stage during which organogenesis occurs and (ii) the period during which adult fish produce gametes and reproduce. We will expose two species of native freshwater fish (fathead minnow, smallmouth bass) to estrone- or treated effluent-amended water during both stages. These species were chosen because they are native to North America, widespread and abundant in many aquatic environments, readily available from controlled culture facilities, represent two levels of the aquatic food chain (fathead minnow – primary consumer; smallmouth bass – apex predator whose prey include juvenile fathead minnows), and have been used as model species for laboratory and field studies of CECs in the past. Both life stages and species will

be exposed at two temperatures (59°F = 15°C; 72°F = 23°C) to mimic conditions across the life history stages of these fishes and to match the treatment conditions in Activity 1. Following exposure, larvae will be assessed in their ability to perform innate predator avoidance behaviors. Adult fishes (males and females) will be assessed for changes in their reproductive behavior. Fish will also be analyzed for vitellogenin concentrations (a precursor protein involved in egg production and a sign of feminization of male fish) and their livers and reproductive organs will be evaluated for changes. The resultant data will be used to develop the fish biomass model described in Activity 3.

Summary Budget Information for Activity 2:

ENRTF Budget: $ 187,000
Amount Spent: $ 0
Balance: $ 187,000

Activity Completion Date: March 31, 2017

Outcome	Completion Date	Budget
1. Reproduction and survival data for larval and adult fathead minnows exposed to differentially treated wastewater at seasonally-appropriate temperatures	03/31/2016	$93,500
2. Reproduction and survival data for larval and adult smallmouth bass exposed to differentially treated wastewater at seasonally-appropriate temperatures	03/31/2017	$93,500

Activity Status as of January 31, 2015:

Activity Status as of July 31, 2015:

Activity Status as of January 31, 2016:

Activity Status as of July 31, 2016:

Activity Status as of January 31, 2017:

Final Report Summary:

ACTIVITY 3: Conduct a cost-benefit analysis that links the cost of different wastewater treatment options to mathematical predictions of fathead minnow and smallmouth bass abundance

Description: During Activity 3 we will conduct an empirical analysis of alternative water quality trading systems, incorporating information about costs of upgrading wastewater treatment facilities and ongoing operating costs from Activity 1 and from the literature, and solve for the cost of attaining a set of water quality levels. Mathematical modeling will link these water quality levels to the equilibrium biomass of fathead minnows and smallmouth bass in a river that receives treated wastewater. To link treatment options to fish biomass, we will develop a mathematical simulation model that uses environmental cues (e.g., seasonal wastewater effluent temperature) and fish biology to predict minnow and bass biomass under various scenarios of exposure to treated effluent. This information will allow us to express the cost of treating effluent in terms of benefits related to the biomass of different fish species

Summary Budget Information for Activity 3:

ENRTF Budget: $ 173,750
Amount Spent: $ 0
Balance: $ 173,750

Activity Completion Date: May 31, 2017

Outcome	Completion Date	Budget
1. A predictive mathematical model that simulates minnow and bass abundance in a pristine, Minnesota river during different seasons	12/30/2015	$52,325
2. A predictive mathematical model that simulates minnow and bass abundance under exposure to treated wastewater effluent during different seasons	12/30/2016	$52,325
3. A cost-benefit analysis of treatment options and fish abundance	05/31/2017	$69,100

Activity Status as of January 31, 2015:

Activity Status as of July 31, 2015:

Activity Status as of January 31, 2016:

Activity Status as of July 31, 2016:

Activity Status as of January 31, 2017:

Final Report Summary:

V. DISSEMINATION:

Description: The target audience for results from this research will be professionals in the areas of wastewater treatment and natural resource management. Specific targets will be environmental engineers and scientists in academia, industry, state agencies such as the DNR and 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 and if possible, at targeted seminars at the DNR and MPCA. Results will be used to determine which wastewater treatment upgrades offer the most ecological protection while incorporating the value of fisheries and energy use.

Status as of January 31, 2015:

Status as of July 31, 2015:

Status as of January 31, 2016:

Status as of July 31, 2016:

Status as of January 31, 2017:

Final Report Summary:

VI. PROJECT BUDGET SUMMARY:

A. ENRTF Budget Overview:

Budget Category	$ Amount	Explanation
Personnel:	$ 282,800	Over the course of the 3-year project, two years of support for two graduate students (Activity 1 and the economic aspects of Activity 3), two years of support for a postdoctoral researcher (the population modeling aspects of Activity 3), and two years of support for a research associate (Activity 2), are budgeted. Funds for the research associate will be covered under a subcontract to St. Cloud State University (see below). The PI (Novak) will each receive 2 weeks of salary a year for the first 2 years of the project. The Co-PIs Venturelli and Homans will each receive 1 week of salary a year for the first 2 years of the project. No salary is requested for Schoenfuss who will be granted one semester 100% re-assign time by St. Cloud State University to focus on the analysis of biological data. The PIs will be responsible for project oversight, guidance of the graduate students and postdoctoral researchers, data interpretation and analysis, and report preparation and submission. Two graduate student research assistants will each devote 100% of their research time to the project over a 2-year period. Fringe benefits for graduate students include tuition, health insurance, and summer FICA. All fringe benefit rates are set by the University of Minnesota and St. Cloud State University.
Equipment/Tools/Supplies:	$30,000	Funds ($30,000) are requested for materials, supplies, consumables, analytical costs and upkeep associated with the LC-MS, computers (to be used only on this project), and software. Required materials include, but are not limited to: pipette tips, glassware, solid phase extraction cartridges for extractions, chemicals for standards and experiments, pumps, analytical consumables, analytical fees, solvents, reagents, gloves, digital data storage media, and laboratory notebooks. A portion of the Materials & supplies are budgeted for support of the fish exposure experiments (fish, chemicals, pumps, aquaria maintenance, etc., $54,000/3 years) and will be part of the subcontract to St. Cloud State University (see below).

Travel Expenses in MN:	$400	Travel funds are extremely minimal ($400) are included for travel to meetings at either St. Cloud State University or the University of Minnesota for project coordination.
Other: Subcontract to St. Cloud State University	$186,800	The subcontract amount ($186,800) will include salary for a research associate ($48,825 salary, $17,575 fringe (36% fringe rate) per year for 2 years) and supplies for experiments (fish, chemicals, pumps, aquaria maintenance, etc., $54,000 for 3 years) to complete Activity 2.
TOTAL ENRTF BUDGET:	**$500,000**	The total proposed project amount is $500,000. No indirect costs for the University of Minnesota or St. Cloud State University are included in the budget.

Explanation of Use of Classified Staff: N/A

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

Number of Full-time Equivalents (FTE) Directly Funded with this ENRTF Appropriation: 4.15

Number of Full-time Equivalents (FTE) Estimated to Be Funded through Contracts with this ENRTF Appropriation: 2

B. Other Funds: N/A

VII. PROJECT STRATEGY:

A. Project Partners: The project team consists of the Principal Investigator (PI) Paige Novak (University of Minnesota) and co-PIs Dr. Heiko Schoenfuss (St. Cloud State University), Paul Venturelli (UMN), and Frances Homans (UMN). Novak will direct Activity 1; Schoenfuss will direct Activity 2; Venturelli will direct Activity 3; Homans will direct the cost analysis and economic modeling efforts. MCES has agreed to provide access to wastewater.

B. Project Impact and Long-term Strategy: The proposed work fits into a larger research agenda centered at UMN and St. Cloud State focused on environmental estrogens and improved wastewater treatment. The proposed research complements current and prior research in this area. This project builds on what we have learned and takes it further, factoring in cost, how CEC removal is impacted by changes in treatment (focused on nitrogen removal), and how temperature impacts both removal efficiency and fish vulnerability. It also expands the impact of the research by incorporating fish population modeling to scale the findings to a whole-state level. When taken together, this research will provide a more complete picture of how to improve treatment, decrease costs and energy use, and safeguard our fish populations.

C. Spending History: N/A

VIII. ACQUISITION/RESTORATION LIST: N/A

IX. VISUAL ELEMENT or MAP(S): See attached graphic.

X. ACQUISITION/RESTORATION REQUIREMENTS WORKSHEET: N/A

XI. RESEARCH ADDENDUM: See attached Research Addendum

XII. REPORTING REQUIREMENTS:
Periodic work plan status update reports will be submitted no later than January 31, 2015, July 31, 2015, January 31, 2016, July 31, 2016, and January 31, 2017. A final report and associated products will be submitted between June 30 and August 15, 2017.

Environment and Natural Resources Trust Fund

M.L. 2014 Project Budget

Project Title: Evaluation of Wastewater Nitrogen and Estrogen Treatment Options

Legal Citation: M.L. 2014, Chp. 226, Sec. 2, Subd. 03d

Project Manager: Paige J. Novak

Organization: University of Minnesota

M.L. 2014 ENRTF Appropriation: $ 500,000

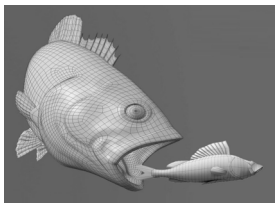
Project Length and Completion Date: 3 Years, June 30, 2017

Date of Report: January 14, 2014

ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Activity 1 Budget	Amount Spent	Activity 1 Balance	Activity 2 Budget	Amount Spent	Activity 2 Balance	Activity 3 Budget	Amount Spent	Activity 3 Balance	TOTAL BUDGET	TOTAL BALANCE
BUDGET ITEM	***Determine the performance of different wastewater treatment processes with respect to nitrogen removal, CEC and estrone removal, energy use, and cost***			***Determine how temperature and life stage alter the reproduction and survival of fathead minnows and smallmouth bass after exposure to treated synthetic or real***			***Conduct a cost-benefit analysis that links the cost of different wastewater treatment options to mathematical predictions of fathead minnow and smallmouth bass***				
Personnel (Wages and Benefits)	$109,050	$0	$109,050	$0	$0	$0	$173,750	$0	$173,750	$282,800	$282,800
Paige Novak, PI ($12,700 salary, $4,300 fringe, 33.6% fringe rate; total for 2 years; 3.8% effort),											
Paul Venturelli, Co-PI ($4,300 salary, $850 fringe, 19.8% fringe rate; total for 2 years; 1.9% effort)											
Frances Homans, Co-PI ($5,650 salary, $1,400 fringe, 24.7% fringe rate; total for 2 years; 1.9% effort)											
One Postdoctoral Researcher ($82,400 salary, $17,100 fringe (includes healthcare); total for 2 years; performing the mathematical modeling of fish populations)											
Two Graduate Research Assistants ($84,700 salary, $69,400 fringe (includes healthcare and tuition); total for 2 years for each student; one student will perform the research on the removal of nitrogen and CECs during wastewater treatment and the other will perform research on the cost and value of wastewater treatment upgrades with respect to the preservation of fish populations))											
Equipment/Tools/Supplies Laboratory supplies and analytical costs (includes, but is not limited to, chemicals for all analyses, supplies to maintain analytical equipment, supplies for reactor construction, and pumps ($30,000/3 years))	$30,000	$0	$30,000	$0	$0	$0	$0	$0	$0	$30,000	$30,000
Travel expenses in Minnesota Travel between St. Cloud and Minneapolis for research progress meetings (in state)	$200	$0	$200	$200	$0	$200	$0	$0	$0	$400	$400
Other: Subcontract Some of the work will be conducted at St. Cloud State University (Activity 2). The subcontract amount will include salary for a research technician ($55,000 salary, $11,400 fringe (20.75% fringe rate) per year for 2 years) and supplies for experiments (fish, chemicals, pumps, aquaria maintenance, etc., $54,000/3 years)	$0	$0	$0	$186,800	$0	$186,800	$0	$0	$0	$186,800	$186,800
COLUMN TOTAL	**$139,250**	**$0**	**$139,250**	**$187,000**	**$0**	**$187,000**	**$173,750**	**$0**	**$173,750**	**$500,000**	**$500,000**

What we have learned from previous research (ENRTF-funded and funded by other sources)

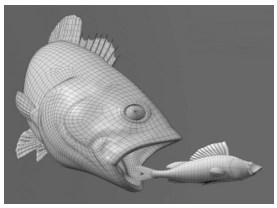




	Wastewater treatment	Contaminant effects	Fish abundance
 Operation	• MN effluent contains CECs and nitrogen (N) • N treatment removes CECs	• Estrogen harms vulnerable minnow stages (sexual differentiation, spawning, early development)	
 Temperature	• N removal declines in cold weather		
 Cost	• N treatment is expensive		

Questions to relate CEC removal costs to fish abundance (proposed ENRTF project)



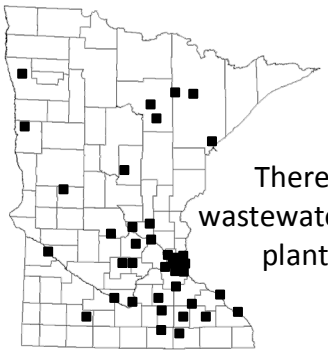


	Activity 1	Activity 2	Activity 3
 Operation	• How well do more efficient N-removal systems remove CECs?	• Do CECs affect minnows and bass differently?	• How do changes in minnows affect bass and vice versa?
 Temperature	• How does seasonality affect CEC removal?	• How does CEC seasonality correspond to vulnerable life stages?	• How does CEC seasonality affect abundance?
 Cost	• What does it cost to operate more efficient N-removal systems?		• How can more efficient N-removal systems benefit fish abundance?

Why?

- Improve N-removal systems to save money, decrease energy use, and **provide the added benefit of CEC removal**
- **Safeguard** Minnesota's **fish populations for the long-term**

Where?



There are ~35 wastewater treatment plants in MN