

2011-2012 MAIN PROPOSAL

PROJECT TITLE: Using Gene Expression Analysis to Assess Water Resource Health

I. PROJECT STATEMENT

This project will implement a pilot program to evaluate the use of gene expression analysis in fish (using a technology called cDNA microarrays) to assess water resource health throughout Minnesota. Results from this work will: 1) advance the development of standards and monitoring protocols for emerging contaminants throughout Minnesota and the upper-Midwest; 2) develop an analytical approach that will support the Intensively Monitored Watersheds (IMW), Watershed Assessment, and Total Maximum Daily Load (TMDL) Programs currently being implemented by Minnesota Pollution Control Agency (MPCA); and 3) provide technical data to support the development of a watershed-wide TMDL study and local-level management of the Elm Creek watershed in Hennepin County.

Management of chemical contaminants is often limited by three major questions: 1) how can exposure to chemical contaminants be effectively monitored when environmental concentrations are highly variable over time and/or sensitive analytical chemistry procedures are unavailable (as for “emerging” contaminants); 2) how can the effects of chemical contaminants on ecosystem health be predicted before they manifest into a large-scale ecological problem; and 3) since organisms are exposed to a continually variable mixture of contaminants and changing environmental conditions over time, how can ecosystem changes be attributed to specific chemicals or stressors (and ultimately remediated)? Gene expression analysis in fish provides an efficient, cost-effective means to simultaneously answer all of these questions.

Since fish are continuously resident in the water, changes in their gene expression patterns mirror changes in environmental condition and chemical contaminant exposure in lakes, streams, rivers...etc. Additionally, by characterizing changes in the expression of specific genes that correspond with exposure to known contaminants (often called biomarkers) and combinations of genes that regulate important physiological processes (often called pathways), its is possible to identify individual chemicals that fish have likely been exposed to and predict their effects on the ecosystem. This proposal will characterize changes in gene expression in fish throughout the Elm Creek watershed using cDNA microarrays, and evaluate the ability this tool predict changes in contaminant exposure and ecosystem health (both of which will be independently monitored through parallel efforts by MPCA, the Elm Creek Watershed Management Commission and Three Rivers Park District).

II. DESCRIPTION OF PROJECT ACTIVITIES

Activity 1: Fish Tissue Collection _____ **Budget: \$** __\$20,000__

Tissues from liver and reproductive organs will be collected from wild and caged fish at 13 sites throughout the Elm Creek watershed for gene expression analysis.

Outcome	Completion Date
1. Fathead minnows will be deployed in cages for two weeks at 6 sites	June-Aug 2011
2. Tissues will be collected caged and wild fathead minnows at 6 sites	June-Aug 2011
3. Tissues will be collected from wild largemouth bass from 7 lakes	June-Aug 2011

Activity 2: Environmental Chemistry Analysis _____ **Budget: \$** __\$60,000__

Chemical concentrations in water, fish tissue and sediment will be measured throughout the Elm Creek watershed to identify the suite of contaminants potentially affecting the fish collected in Activity 1. Chemical analyses will be leveraged against water quality assessment work currently being conducted as part of the Elm Creek Watershed-wide TMDL.

Outcome	Completion Date
1. Water chemistry will be measured on weekly intervals at 13 stream and lake sites throughout the Elm Creek watershed	July 2011
2. Contaminant concentrations will be measured in wild fish tissues and sediment from all sites	Sept 2011-Feb 2012
3. Contaminant concentrations will be summarized in a report to support the Stressor Identification process in the Elm Creek Watershed Wide TMDL	July 2012

Activity 3: Gene Expression Analysis in Fish Tissues _____ **Budget:** \$ _\$270,000_

Changes in gene expression will be characterized in exposed fish and used to relate contaminant exposure (Activity 2) with changes in fish and invertebrate community composition (measured simultaneously through the MPCA Intensive Watershed Monitoring program).

Outcome	Completion Date
1. All fish tissues will be analyzed to determine relative gene expression using cDNA microarrays	Sept 2011-Feb 2012
2. Microarray data will be analyzed to identify changes in contaminant-specific biomarkers and alteration of important physiological pathways	Sept 2011-Feb 2012
3. Biomarker and pathway data will be evaluated as a tool to identify contaminant exposure and predict ecological effects	Sept 2011-Feb 2012
4. All data will be summarized in a final project report and presented to MPCA	July 2012

III. PROJECT STRATEGY

A. Project Team/Partners

This project will be implemented through a partnership between Three Rivers Park District, Elm Creek Watershed Management Commission and University of Florida. Dr. Randy Lehr from Three Rivers Park District will serve as the overall project lead, coordinating the collection and analysis of water chemistry data, collection fish tissue samples and project reporting. Dr. Nancy Denslow from University of Florida will coordinate all gene expression analysis. Funds requested in this proposal will be leveraged against funding from two ongoing grants from MPCA, the state-wide IMW program and in-kind contributions from Three Rivers Park District.

B. Timeline Requirements

This project will be implemented from 2011-2012. Field data collection will occur in the summer of 2011 to coincide with likely maximum contaminant exposure and spawning seasons. Laboratory chemical and cDNA microarray analyses will be conducted throughout 2011-2012 and the final project report and presentation will be completed in the summer of 2012.

C. Long-Term Strategy and Future Funding Needs

This project is self-contained. Results from this work will provide a potential model to conduct multi-stressor analysis as part of the IMW and TMDL programs throughout the state.

2011-2012 Detailed Project Budget

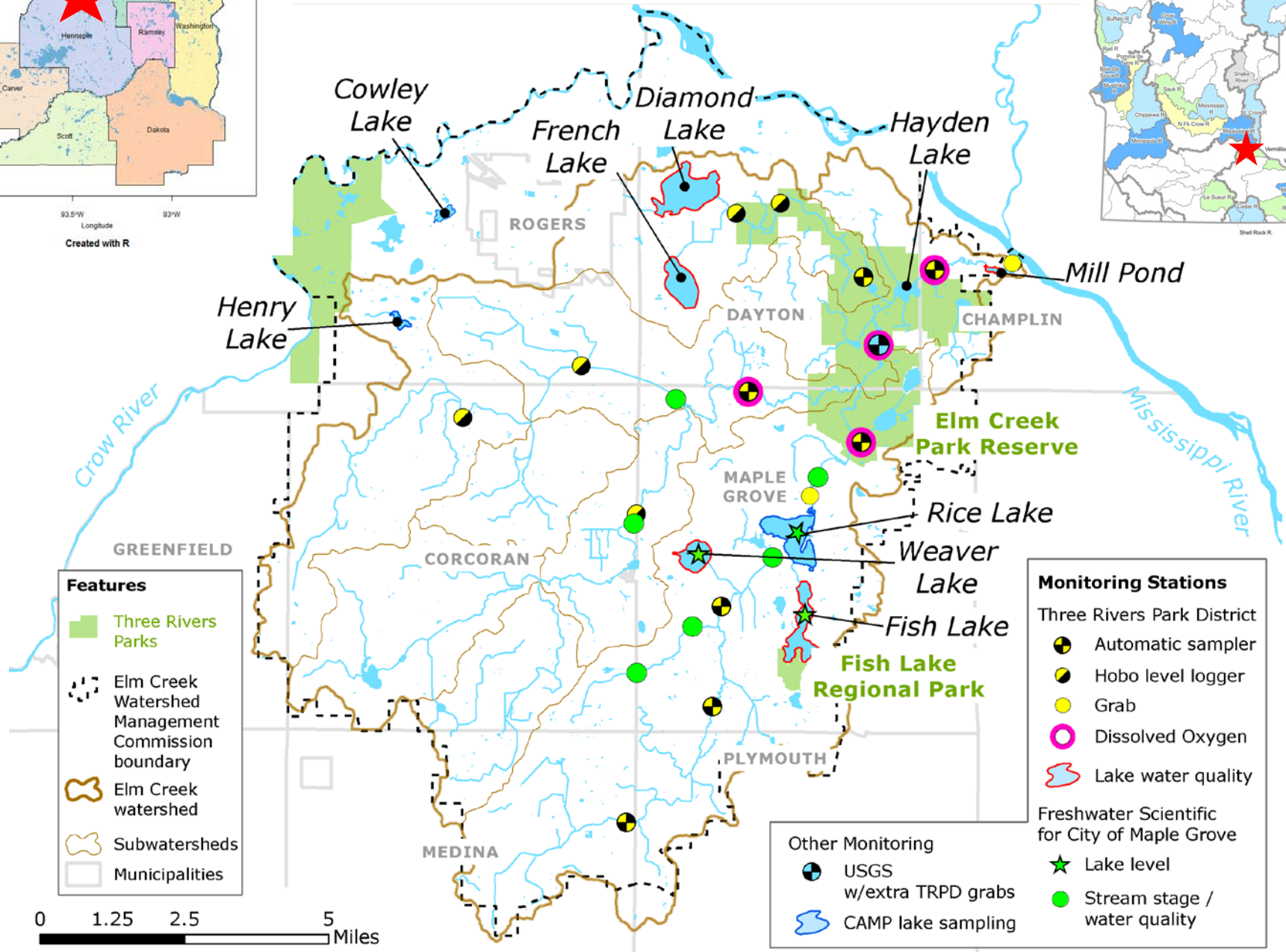
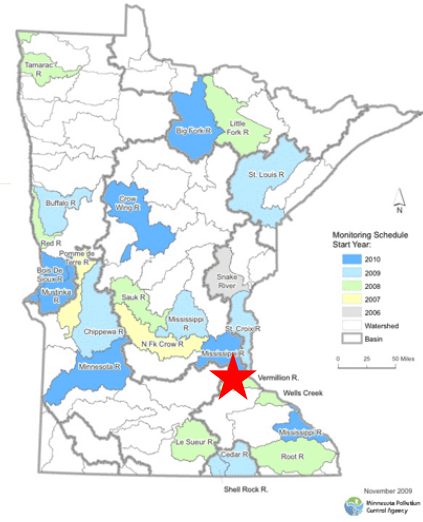
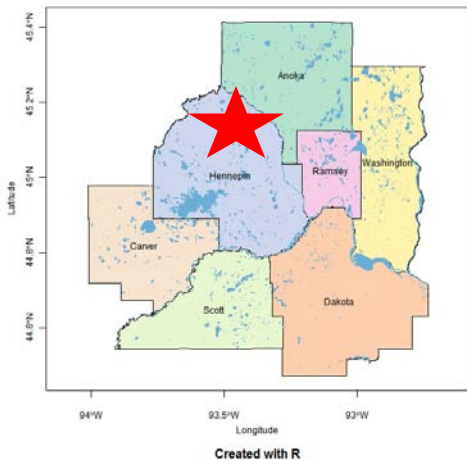
IV. TOTAL TRUST FUND REQUEST BUDGET - Project Length, 2-years

<u>BUDGET ITEM</u>	<u>AMOUNT</u>
Personnel: <i>Personnel expenditures will cover salary and benefits for: Project Manager (\$70/hr) and Field Technicians (\$35/hr). The Project Manager will serve as the overall project lead, coordinating the collection and analysis of water chemistry data, collection fish tissue samples and project reporting (\$20,000). Field Technician will be responsible for all field sample collection and site maintenance (\$25,000).</i>	\$ 45,000
Contracts: <i>Dr. Nancy Denslow, University of Florida (\$250,000). This contract will result in the microarray analysis of tissues from caged and wild fish from ~13 sites throughout the Elm Creek watershed. Analytical chemistry (\$50,000). This contract will be for analysis of chemical contaminants in water, sediment and fish tissue throughout the watershed.</i>	\$ 300,000
Equipment/Tools/Supplies: <i>Fish Exposure Equipment (\$2500), Chemical collection equipment (\$2500)</i>	\$ 5,000
Acquisition (Fee Title or Permanent Easements): <i>NA</i>	\$ -
Travel: <i>NA</i>	\$ -
Additional Budget Items: <i>NA</i>	\$ -
TOTAL ENVIRONMENT & NATURAL RESOURCES TRUST FUND \$ REQUEST	\$ 350,000

V. OTHER FUNDS

<u>SOURCE OF FUNDS</u>	<u>AMOUNT</u>	<u>Status</u>
Other Non-State \$ Being Applied to Project During Project Period: <i>Elm Creek Watershed Management Commission contribution to the Elm Creek Watershed-wide TMDL (secured)</i>	\$ 40,000	Secured
Other State \$ Being Applied to Project During Project Period: <i>MPCA grant to complete the Elm Creek Watershed-wide TMDL (secured)</i>	\$ 155,000	Secured
In-kind Services During Project Period: <i>In-kind contributions for water quality monitoring and watershed modeling to support the Elm Creek Watershed-wide TMDL (secured)</i>	\$ 100,000	Secured
Remaining \$ from Current ENRTF Appropriation (if applicable): <i>NA</i>	\$ -	
Funding History: <i>MPCA, Elm Creek Watershed Commission and Three Rivers Park District contributions to the Elm Creek Watershed-wide TMDL (\$150,000). MPCA grant to Three Rivers Park District to utilize cDNA microarrays to monitor lake health in the Twin Cities metro area (\$40,000).</i>	\$ 223,000	Secured

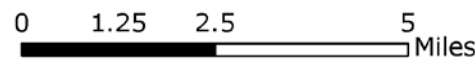
Elm Creek Watershed



- Features**
- Three Rivers Parks
 - Elm Creek Watershed Management Commission boundary
 - Elm Creek watershed
 - Subwatersheds
 - Municipalities

- Monitoring Stations**
- Three Rivers Park District
- Automatic sampler
 - Hobo level logger
 - Grab
 - Dissolved Oxygen
 - Lake water quality
- Freshwater Scientific for City of Maple Grove
- Stream stage / water quality
 - Lake level

- Other Monitoring**
- USGS w/extra TRPD grabs
 - CAMP lake sampling



Project Manager

Dr. Randy Lehr, Senior Manager of Water Resources for Three Rivers Park District (TRPD) will serve as the project manager. Dr. Lehr holds an M.S. and Ph.D. from University of Minnesota in Water Resources Science where his master's thesis and dissertation research focused on the development of chemical and molecular techniques to assess the potential impacts of a group of contaminants known as endocrine disrupting chemicals throughout the upper-Midwest. Specifically, this research focused on the use of gene and protein expression in fish to identify exposure to contaminants of concern. Dr. Lehr's current work and research focus on the assessment and restoration of water resource health throughout the Twin Cities Metropolitan area. Dr. Lehr has administered over \$1.8-million dollars in grant funded research and authored (or co-authored) a number of scientific and technical publications. Dr. Lehr also currently serves as the technical lead on two projects that will be directly leveraged against the proposed work. Current projects include:

Minnesota Pollution Control Agency, \$40,000 – "Using cDNA Microarrays to Assess Potential Ecological Impacts of Perfluorinated Chemical in Select Lakes Throughout the Twin Cities Metro Area", Randy Lehr, Three Rivers Park District, Nancy Denslow, University of Florida and Summer Streets, MPCA, May 2009

Minnesota Pollution Control Agency, \$460,500 (over 5 years) – "Elm Creek Watershed-Wide TMDL", Randy Lehr, Three Rivers Park District, February 2009

Three Rivers Park District

The mission of Three Rivers Park District is to "Promote natural resource stewardship through education and recreation in a natural resourced-based park setting". To this end, TRPD has been extensively involved in research, outreach and education related to natural resource management throughout the Twin Cities metropolitan area. The TRPD water resource program has a staff of five full-time scientists, four seasonal technicians and has served as the technical and outreach lead for a number of research and management efforts throughout the region (specifically in the Elm Creek watershed).