Environment and Natural Resources Trust Fund

Research Addendum for Peer Review

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Project Title: **Identifying the Cause of Exceptionally High Mercury in Fish**

Project number: **045-B**

1. Abstract

We define "exceptionally high mercury in fish" as mercury concentrations in standardized-length top predator fish that exceed the 90th percentile mercury concentration of 0.57 mg/kg. As of the 2012 assessment of impaired waters in Minnesota, five rivers—all in northern Minnesota—had mercury levels in walleye above that threshold. The purpose of this mercury study is to test the hypothesis that high mercury concentrations in fish from these northern Minnesota rivers is caused by enhanced bioavailability of mercury in their watersheds, rather than because of more mercury input to the watersheds. This study will measure the key ecosystem processes that drive mercury bioavailability to identify the relative contribution of each process within and among rivers. Mobilization of mercury, methylmercury, and ancillary water quality parameters will be measured in the Roseau River and two other tributaries to the Red River of the North (in northwestern Minnesota). In the Roseau and four other rivers with high fish-mercury levels, this study will measure the rates of mercury converted to methylmercury, the removal of methylmercury, and the accumulation of methylmercury in the food webs. Stable isotope ratios of carbon and nitrogen will be measured at all levels of the food web to identify possible differences in carbon source and the rates of biomagnification. In addition to enhancing the understanding of mercury bioavailability, the results of this study are expected to reveal if there are ways to reduce mercury levels beyond reduction of mercury sources.

2. Background

Mercury is well known as a neurotoxin, and is especially damaging to the developing nervous system of fetuses and children. The health benefits of eating fish are compromised because fish consumption is the major route of mercury exposure to humans and wildlife. The Minnesota Pollution Control Agency has a Mercury Reduction Implementation Plan to achieve necessary reductions in the ultimate sources of mercury to the state's waters. Despite the benefit of these reductions to all lakes and rivers in Minnesota, some lakes and rivers are not expected to achieve the state's mercury water quality standard for mercury in edible fish tissue. To fully address the mercury problem in Minnesota, we need to understand what is driving the enhanced accumulation of mercury in this subset of waters.

Accumulation of mercury in fish is obviously a function of source contributions (e.g., combustion emissions and subsequent atmospheric deposition). Less obvious are the ecosystem processes that control mercury accumulation in the food web after the mercury is deposited in the watershed. These

processes drive the transport, transformation (speciation), and bioaccumulation of the mercury. As a consequence of these processes, watersheds receiving equivalent inputs of mercury can have mercury concentrations in the same fish species that differ by a factor of ten [1].

Most of the research to understand these processes has been in lakes and their watersheds. These studies have identified the drivers of mercury variation, such as dissolved organic carbon (DOC), wetlands density, and productivity [2]. These drivers are interconnected: methylmercury and DOC are produced in wetlands; mercury and methylmercury are transported to lakes and rivers bound to the DOC (and particulate matter).

Ward et al. (2010) [3] describes a hierarchy of interacting processes, from the individual factors that control mercury bioaccumulation (methylmercury in prey, consumption rate, and growth rate); through the trophic transfer in the food web; to the biogeochemical characteristics of the stream and its watershed. When all these factors interact to increase the efficiency of mercury accumulation in the food web, the authors postulated this is a "bioaccumulation syndrome" in which no one factor could predict the mercury levels in fish across a region.

Research on mercury bioaccumulation in stream food webs has increased in the last decade. The most comprehensive study of mercury in streams was recently reported by the United States Geological Survey [4]. They studied multiple levels of the mercury cycle in eight streams in Florida, Oregon, and Wisconsin over three years. In the first of three companion papers, they showed water column mercury (total and methyl) were most strongly correlated to DOC and suspended sediment concentrations; and the dissolved fractions of total and methyl mercury were correlated with wetland density in the river basin [5]. The second paper focused on methylmercury production and partitioning in sediments and water, they reported water column methylmercury concentrations were not related to methylmercury in the river sediments, indicating the methylmercury production must happen upstream instead of in the river channel [6]. The third paper focused on the aquatic food web and bioaccumulation. They concluded mercury levels in top predator fish were primarily controlled by the methylmercury availability at the base of the food web [7].

The critical and conflicting roles DOC seems to play in mercury bioavailability has made it the focus of intense research. In a comparison of 30 streams in a north-south gradient in eastern Minnesota, Tsui and Finlay (2011) [8] showed methylmercury concentrations at the base of the food web were higher in watersheds having higher wetland and forest coverage. They found that DOC concentration and DOC quality (measured as SUVA—specific UV absorbance per unit DOC) were positively correlated with dissolved total and methyl mercury, but negatively correlated with methylmercury (MeHg) bioconcentration factor (MeHg in consumers/MeHg in seston). The negative correlations demonstrated that increased DOC quantity and aromaticity reduce the efficiency of methylmercury partitioning into the primary consumers.

Methylation of inorganic mercury is a critical step in the bioavailability of mercury to aquatic food webs, which is understood to be a microbial process, affected by many biogeochemical drivers [9] [10]. Demethylation of methylmercury is less well understood. Biotic-mediated demethylation is important in

contaminated systems [11], whereas abiotic demethylation, or more specifically photolytic demethylation, is considered the most important process that destroys methylmercury in marine and freshwater systems [12]. Like much of the research on the mercury cycle, more is known about photodemethylation in lakes than in rivers.

Differences in food web relationships can potentially explain some of the variation in fish-mercury levels. As the USGS study concluded, the mercury at the base of the food web is a strong determinant of mercury levels in the top predator fish. Feeding relationships between the base and the top of the food web determine the biomagnification rate. Biomagnification in food webs is measured by the linear regression slope between mercury concentrations and nitrogen stable isotope ratios [13]. The biomagnification rates can also be affected by biogeochemical processes[14]. In addition to the transfer of mercury from one trophic level to the next, mercury levels in the top predator fish are affected by growth rates of the fish; slower growth rates result in higher mercury levels in fish tissue [15] [16] [17].

The main objective of this study is a conceptual model of how these multiple drivers of mercury bioavailability interact to determine mercury levels in top predator fish.

3. Hypothesis

Contaminant monitoring data of mercury levels in walleye have shown five rivers in northern Minnesota have exceptionally high mercury levels compared to other rivers in the state. Based on the premise that mercury deposition is the source of mercury to northern Minnesota watersheds and it is equivalent among the watersheds, we propose to test the hypothesis that fish-mercury concentrations in these rivers are higher because one or more ecosystem drivers have increased the efficiency of methylmercury accumulation in the aquatic food webs of these rivers. We propose to test the hypothesis by measuring four key processes: mercury transport, transformation, degradation, and biological uptake in the food web. We will compare the loadings, rates, and biomagnification factors among the rivers and to values in published scientific studies.

4. Methodology

Transport of Mercury and Methylmercury

Fundamental to understanding mercury availability is quantifying how much mercury and methylmercury is transported through the river. Concentrations of thirteen analytes (Table 1) will be measured 32 times over two years at the gaging station in the Roseau River and two other tributaries of the Red River of the North. Water samples will be collected as grab samples below the water surface in the main flow the stream (i.e., thalweg). Mercury and methylmercury samples will be collected using the "clean hands – dirty hands" technique (EPA Method 1669). In addition, field measurements will include pH, temperature, specific conductivity, and dissolved oxygen. Water flows are continuously measured at these sites, which are in the cooperative flow gaging network supported by the DNR and USGS (http://www.dnr.state.mn.us/waters/csg/index.html). Flow and concentration data will be used in the FLUX32 model to calculate annual mass loads at each of the sites.

Final selection of the two tributaries for comparison to the Roseau will be based on prior examination of land cover and stream flow characteristics. The likely candidates are the Red Lake River, Buffalo River, and Marsh River.

All sample collection and laboratory analysis will be contractual. Sample collection will most likely be done by the USGS or DNR under contract with the MPCA. Laboratory analysis methods and costs are based on Minnesota Department of Health's Environmental Laboratory.

Table 1 Laboratory analytes for water collections

Analytes	Method	Reporting Limit	Units	Holding Time (days)
Total Organic Carbon	SM 5310C	1	mg/L	28
Dissolved Organic Carbon	SM 5310C	1	mg/L	28
Sulfate	EPA 300.1	1.00	mg/L	28
Phosphorus, Total	EPA 365.1	0.01	mg/L	28
Iron Dissolved, Low Level	EPA 200.7	14.0	ug/L	180
UV Absorbance @ 254 nm	SM 5910B	0.0050	Abs units	2
Solids, Suspended	SM 2540D	1.0	mg/L	7
Suspended Solids, Volatile	EPA 160.4	10	mg/L	7
Solids, Total Dissolved	SM 2540C	10	mg/L	7
Low Level Mercury, Total	EPA 1631	0.400	ng/L	90
Low Level Mercury, Dissolved	EPA 1631	0.400	ng/L	90
Methyl Mercury, Total	EPA 1630	0.050	ng/L	180
Methylmercury, Dissolved	EPA 1630	0.050	ng/L	180

Methylmercury Production and Destruction

Mercury methylation and demethylation will be measured by the stable isotope technique [18 19]. In situ rates of methylation activity is measured by adding inorganic mercury isotope (e.g., $^{201}Hg^{2+}$) to each sample and monitoring formation of the labelled methylmercury (e.g., $Me^{201}Hg^{+}$). Similarly, for demethylation, the same sample is spiked with another stable-isotope labelled methylmercury (e.g., $Me^{199}Hg^{+}$) and it is monitored for a decrease in concentration. The ratio of methylation rate/demethylation rate gives the net methylation rate potential. The methylation-demethylation study will be done at nine sites, with at least three samples per site visit, and each site will be visited three times to capture seasonal differences of the rates. This study will begin in the second year of this project because sites will be selected based on the first year of water quality monitoring.

Photodemethylation is considered the most important removal process of methylmercury in lakes, but it has been studied much in rivers. To measure photodemethylation of mercury, clear ("light") and opaque ("dark") bottles will be suspended in situ with sufficient replicates to measure methylmercury concentrations before and after incubation. Ultraviolet and photosynthetically active radiation will be measured continuously during the incubations. The sites for the photodemethylation study will be downstream of the nine methylation-demethylation sites. The methylation-demethylation sites are

expected to be wetlands hydrologically connected to the rivers and the photodemethylation sites will be downstream sites unobstructed by tree canopy.

The quantity and quality of dissolved organic matter (DOM) strongly influences mercury methylation and demethylation. Therefore, the DOM from the selected river sites will be quantified as total organic carbon (TOC) and dissolved organic carbon (DOC) as part of Activity 1 and the quality of the DOM will be assessed using several state-of-the-art techniques[20]. The characterization of DOM will show if the DOM sources differ among the rivers, which could determine how they affect methylation and demethylation.

Food Web and Fish Life Histories

Aquatic food webs in five rivers will provide a comparison of biomagnification rates and possible differences in growth rates of top predator fish. Food web collections will include seston, benthic macro invertebrates, forage fish, and predator fish. Isotopic ratios of naturally-occurring stable isotopes for nitrogen (N) are carbon (C) will be measured in the biota samples to identify feeding relationships in the food webs and carbon sources. Food web collection and sample analysis will follow the procedures described in Scudder et al. (2008) [21]. This USGS publication describes the procedures for collection and processing of biota samples for mercury, methylmercury, and stable isotope analyses. Biomagnification of mercury in the food webs will be measured as the linear regression slope of log-transformed methylmercury concentration versus the N-isotope ratios (delta-15 N) in the biota samples. Growth rates in top predator fish will be calculated based on the aging and length data. USGS procedures include collection of otoliths from top predator fish for age determination.

Because of the natural variability in mercury and isotope levels in biota, meaningful comparisons require multiple sites and adequate samples of biota at each site. A total of 63 biota samples at each of 17 sites (1071 samples) was used to estimate the budget for the food web comparison.

GIS Database and Conceptual Model Development

All data will be compiled in a geo-referenced database, along with land cover summaries for each watershed. The compiled data will be statistically analyzed with as yet unspecified tests. Likely statistical tools are spatial statistics/plotting, principal components analysis, and mixed effects modeling. These statistical tools can provide a measure of the relative importance of the multiple variables and processes measured in this study. From the statistical analysis, an empirically-based conceptual model will be developed for the important drivers of mercury accumulation in the riverine food webs.

5. Results and Deliverables

Expected Outcomes

Activity 1. Transport of Mercury: mass loads of mercury, methylmercury, and other ancillary analytes, based on two years of flow and water quality monitoring in Roseau and two other rivers.

Activity 2. Mercury Production/Destruction: (1) methylation potentials (net methylation rates) at nine sites. (2) Photodemethylation rates at nine sites.

Activity 3. Food Web: (1) methylmercury and mercury concentrations in the food webs (insects to fish) from five rivers. (2) Carbon and nitrogen stable isotope ratios in all biota samples. (3) Trophic magnification factors for all measured food webs. (4) Growth rates of top predator fish in five rivers based on age and length data.

Activity 4. GIS Database & Modeling: (1) All data compiled into a geographic information system database. (2) Statistical modeling results showing relative importance of drivers of methylmercury accumulation in riverine food webs.

Deliverables

Written products will be six-month progress reports and the final report to LCCMR.

6. Timetable

	2014		2015				2016				2017	
Activity	Q3	Q4	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4	Q1	Q2
1. Transport of Mercury												
Water quality sampling												
Laboratory analysis												
Flux (mass load) calculations												
Product delivery										X		X
2. Methylmercury Production & Destruction												
Methylation potential												
Photodemethylation												
DOM characterization												
Product delivery										X		X
3. Food Web Relationships												
Biota collection												
Fish age determination												
Laboratory Analysis												
Product delivery										X		X
4. Database and Statistical Analysis												
Database development												
Statistical analysis												
Product delivery										X		X
Reporting		X		X		X		X		X		X

7. Budget

The recommended funding from LCCMR for this project is $743,000. The justification is as follows:

Personnel: Over the three-year study, Dr. Bruce Monson will manage the project at an estimated 50% of his time. His salary is being provided as in-kind support;. Other MPCA staff will likely provide support to Dr. Monson for database development, contracts, and miscellaneous administrative tasks. None of the ENTRF funding will contribute to MPCA staff salary or benefits. All personnel costs will be included in the contracts with other government or private contractees.

Contracts: All field sampling, experiments, and laboratory analysis will be completed under contracts with the MPCA. Therefore, a total of $739,000 is budgeted for contracts. The original proposal was developed with the United States Geological Services (USGS) and Minnesota Department of Natural Resources (MDNR) as partners and both agencies committed to matching funds. Because the LCCMR recommended less than the requested funding in the original proposal, partnerships with these two agencies have not been renegotiated at this time; however, the project manager expects USGS and MDNR will play major roles in this project, possibly contracting to complete all the activities.

Supplies: No supplies outside of the contracts are requested.

Travel Expenses: A total of $4,000 is requested for in-state travel for MPCA staff to oversee or assist with field sampling and to attend in-state meetings.

Budget Category	$ Amount	Explanation
Personnel:	$ 0	Project manager salary is in-kind support and all other personnel costs included in contracts
Professional/Technical/Service Contracts: Professional/Technical (Activities 1-4) Laboratory (Activities 1 and 3)	 $ 454,000 $ 285,000	All field sampling, experiments, and laboratory analysis will be completed under contracts with the MPCA; Where possible, MPCA will use existing master contracts. Requests for proposals will be issued by the MPCA in the third quarter of 2014 with the intent of signed contracts by January 1, 2015.
Travel Expenses in MN:	$ 4,000	
Other:	$ 0	
TOTAL ENRTF BUDGET:	**$ 743,000**	

8. Credentials

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Education

Ph.D.	1997	University of Minnesota, Minneapolis (Civil Engineering; minor: Water Resources Science). Dissertation: *Mercury Cycling in Low Alkalinity Lakes and Factors Influencing Bioavailability.*
M.S.	1982	University of Illinois, Champaign-Urbana (Biology)
B.S.	1979	University of Minnesota, Minneapolis (Zoology)

Positions

2002-Present	Research Scientist 3, Minnesota Pollution Control Agency
1993-2002	Limnologist, Environmental Engineer, Barr Engineering, Minneapolis, Minnesota.
1991-1994	Graduate Research Assistant, Civil Engineering, University of Minnesota
1988-1991	Environmental Scientist, Limno-Tech, Inc., Ann Arbor, Michigan
1985-1988	Executive Director, Rouge River Watershed Council, Friends of the Rouge, Huron River Watershed Council, Detroit and Ann Arbor, Michigan

Professional Affiliation

Society of Environmental Toxicology and Chemistry

Recent Publications

Coleman-Wasik, JK, Mitchell CPJ, Engstrom DR, Swain EB, Monson BA, Balogh SJ, Jeremiason JD, Branfireun BA, Eggert SL, Kolka RK, Almendinger JE. 2012 Methylmercury Declines in a Boreal Peatland When Experiment Sulfate Deposition Decreases. Environ. Sci. Technol. 46, 6663-6671

Wiener JG, Sandheinrich MB, Bhavsar SP, Bohr JR, Evers DC, Monson BA, Shrank CS. 2012. Toxicological significance of mercury in yellow perch in the Laurentian Great Lakes region. Environmental Pollution 161, 350-357.

Evers DC, Wiener JG, Driscoll CT, Gay DA, Basu N, Monson BA, Lambert KF, Morrison HA, Morgan JT, Williams KA, and Soehl AG. 2011. Great Lakes Mercury Connections: The Extent and Effects of Mercury Pollution in the Great Lakes Region. Biodiversity Research Institute Report BRI 2011-18, Gorham, Maine. 44 pp. http://www.briloon.org/mercuryconnections/greatlakes

Monson BA, Staples DF, Bhavsar SP, Holsen TM, Schrank CS, Moses SK, McGoldrick DJ, Backus SM, Williams KA. 2011. Spatiotemporal trends of mercury in walleye and largemouth bass from Laurentian Great Lakes Region. Ecotoxicology 20 (7): 1555-1567.

Rolfhus KR, Hall BD, Monson BA, Paterson MJ, Jeremiason JD. 2011. Assessment of mercury bioaccumulation within the pelagic food web of lakes in the western Great Lakes region. Ecotoxicology 20 (7): 1520-1529.

Monson BA. 2009. Trend Reversal of Mercury Concentrations in Piscivorous Fish from Minnesota

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Jeremiason JD, Engstrom DR, Swain EB, Nater EA, Johnson BM, Almendinger JE, Monson BA, Kolka RK. Sulfate addition increases methylmercury production in an experimental wetland. Environ. Sci. Technol. 40, 3800-3806.

Monson, B.A. and P.L. Brezonik. 1999. Influence of food, aquatic humus, and alkalinity on methylmercury uptake by Daphnia magna. Environmental Chemistry and Toxicology 18(3): 560-566.

Monson, B.A. and P.L. Brezonik. 1998. Seasonal patterns of mercury species in water and plankton from softwater lakes in Northeastern Minnesota. Biogeochemistry 40: 147-162

Conference Presentations

Monson BA, Engstrom DE, Swain EB, Balogh SJ, Parson K. 2013. Resolving the Cause of the Recent Rise of Fish-mercury Levels in the Western Great Lakes Region (Poster). 11th International Conference on Mercury as a Global Pollutant, 28 July – 02 August 2013, Edinburgh, Scotland.

Monson BA, Bhavsar SP, Sandheinrich MB, Wiener JG. 2011. Mercury in Fish from the Great Lakes Region: Spatiotemporal Trends and Ecological Risk (Oral). 10th International Conference on Mercury as a Global Pollutant, 24 July – 29 July 2011, Halifax, Nova Scotia, Canada.

Monson BA, Hoff P, McCann P, Solem L, Streets S. 2009. Distribution of PFCs in Fish from Minnesota Lakes and Rivers (Poster). Society of Environmental Toxicology and Chemistry North America 30th Annual Meeting, 19-23 November 2009, New Orleans, Louisiana

Monson BA. 2009. Assessing Temporal Trends of Mercury in Fish (Oral Plenary). Society of Environmental Toxicology and Chemistry Midwest Chapter Annual Meeting, 30 March – 1 April 2009, La Crosse, Wisconsin.

Monson BA. 2007. Total Mercury and Methylmercury in Constructed Stormwater Wetlands (Poster). Society of Environmental Toxicology and Chemistry North America 28th Annual Meeting, 11-15 November 2007,

Monson BA, Markus H, Swain EB, Jackson A, Brooks N. 2006 Minnesota's Statewide Mercury TMDL (Oral). 8th International Conference on Mercury as a Global Pollutant, 6 - 11 August 2006.

Monson B, Markus H. 2006 Use of Multi-media Monitoring to Develop a Statewide Mercury TMDL (Oral). 5th National Monitoring Conference 7-11 May 2006, San Jose, California.

Minnesota Pollution Control Agency Reports (Lead Author)

Perfluorochemicals in Mississippi River Pool 2: 2012 Update. 2013. http://www.pca.state.mn.us/index.php/view-document.html?gid=19516

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Total Mercury and Methylmercury Flux in a Constructed Wetland for Stormwater Treatment. 2008. http://www.pca.state.mn.us/index.php/view-document.html?gid=11441

Effectiveness of Stormwater Ponds/Constructed Wetlands in the Collection of Total Mercury and Production of Methylmercury. 2007. http://www.pca.state.mn.us/index.php/view-document.html?gid=289

Minnesota Statewide Mercury Total Maximum Daily Load. 2007. http://www.pca.state.mn.us/index.php/view-document.html?gid=8507

9. Dissemination and Use

The research findings will be initially disseminated as reports to LCCMR. From these reports, the project manager and other primary researchers in this project will prepare manuscripts for scientific peer-reviewed publications and present the results at conferences. Major results will undoubtedly be of interest to the news media, because of the public's interest in mercury and consuming fish. The results of this research should be useful for developing total maximum daily load studies for mercury in fish in these waters that do not qualify for the Statewide Mercury TMDL because of exceptionally high mercury levels. The mercury results for fish will be added to the fish contaminant database maintained by Minnesota's interagency Fish Contaminant Monitoring Program.

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