



Environment and Natural Resources Trust Fund

M.L. 2022 Approved Work Plan

General Information

ID Number: 2022-224

Staff Lead: Michael Varien

Date this document submitted to LCCMR: December 15, 2022

Project Title: Is the Tire Chemical 6PPDq Killing Minnesota's Fish?

Project Budget: \$437,000

Project Manager Information

Name: Nicholas Phelps

Organization: U of MN - College of Food, Agricultural and Natural Resource Sciences

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Project Reporting

Date Work Plan Approved by LCCMR: December 19, 2022

Reporting Schedule: March 1 / September 1 of each year.

Project Completion: June 30, 2025

Final Report Due Date: August 14, 2025

Legal Information

Legal Citation: M.L. 2022, Chp. 94, Sec. 2, Subd. 04i

Appropriation Language: \$437,000 the second year is from the trust fund to the Board of Regents of the University of Minnesota to optimize detection methods, determine environmental occurrence, and evaluate risk to Minnesota's fish populations of the toxic tire-derived chemical 6PPDq.

Appropriation End Date: June 30, 2025

Narrative

Project Summary: A newly discovered and highly toxic tire-derived chemical (6PPDq) may be impacting Minnesota's fish populations - we will optimize detection methods, determine occurrence in the environment, and evaluate risk statewide.

Describe the opportunity or problem your proposal seeks to address. Include any relevant background information.

Last year Washington state researchers discovered that the previously unreported chemical 6PPDq (derived from the “parent” chemical 6PPD used in rubber tires) is associated with major fish kills (40-90% coho salmon mortality!). Understanding the occurrence and consequence of this contaminant of emerging concern (CEC) in Minnesota waters is a priority given the very high toxicity. Most CECs have subtle, sublethal effects that emerge upon long-term exposures. In contrast, 6PPDq has rapid effects, and even a short-term episodic exposure released to surface waters by stormwaters and road runoffs appear to be sufficient to cause severe fish kills. Given the ubiquitous use of 6PPD in tires, this CEC is suspected to be widespread in aquatic systems including Minnesota. Currently, there are no data about the occurrence of 6PPDq in Minnesota (and most other states), and the biological mechanisms by which it is rapidly killing fish are not understood. Our team is uniquely positioned to determine whether 6PPDq poses a threat to fish in Minnesota. We have researched and documented fish kills and factors driving those statewide. We also have specialized analytical expertise and a documented capacity to survey CECs, including 6PPDq, and to characterize their toxicity to inform management decisions.

What is your proposed solution to the problem or opportunity discussed above? Introduce us to the work you are seeking funding to do. You will be asked to expand on this proposed solution in Activities & Milestones.

Given the significance of the 6PPDq discovery and potential threat to Minnesota fish populations, it is imperative that baseline data are collected to guide appropriate management responses and future research plans. To that end, we have brought together a highly interdisciplinary team to conduct the first-ever research on this emerging CEC in Minnesota. First, we will identify and spatially define hypothesized risk-factors associated with potential 6PPDq exposure, such as proximity to major roads, rainfall and runoff patterns, and historical fish kills. The resulting risk map will be used to prioritize locations for surveillance, with special consideration for high-risk locations on tribal lands. Second, concurrent with the above we will develop and transfer methods for 6PPDq extraction and measurement in water and fish tissues to the Minnesota Department of Health. Third, we will collect samples using the locations and methods defined in the above steps and measure 6PPDq concentrations. Lastly, we will use cost-effective and rapid fish cell and larval toxicity tests to conduct pilot studies that will determine the biological basis of toxicity. Based on the above and complimentary toxicity information, we will determine whether 6PPDq poses a risk to Minnesota fish and it warrants further biological studies and monitoring.

What are the specific project outcomes as they relate to the public purpose of protection, conservation, preservation, and enhancement of the state’s natural resources?

Identification of the risk factors and occurrence assessment will allow managers to understand sources and systemwide distribution of 6PPDq. The chemical extraction and analysis methods developed herein will be transferred to the Minnesota Department of Health, allowing Minnesota to monitor this highly toxic CEC as early as 2023. Determination of whether 6PPDq’s association with fish kills extends to habitats/fish species outside Washington will inform and potentially incentivize the tire industry's efforts to design less toxic alternatives. Lastly, this research will fill knowledge gaps related to fish kills in Minnesota and inform science-based mitigation strategies.

Project Location

What is the best scale for describing where your work will take place?

Statewide

What is the best scale to describe the area impacted by your work?

Statewide

When will the work impact occur?

During the Project and In the Future

Activities and Milestones

Activity 1: Develop methods for the detection of 6PPDq in water and fish tissue

Activity Budget: \$103,800

Activity Description:

Evaluating the risks of 6PPDq and other CECs to aquatic organisms requires a well-developed and validated analytical method to measure their concentrations. While a basic analytical method exists at the University of Washington (and two private labs), it needs optimization to be sufficiently sensitive, accurate, and available for use in Minnesota. Working with the experts in 6PPDq detection, we will optimize existing analytical methods for Minnesota samples (e.g., snowmelt and roadway runoff) and develop protocols to transfer the technology to the Minnesota Department of Health. Briefly, we will optimize sample cleanup and C-18 or HLB solid-phase extraction procedures to measure concentrations of 6PPDq down to 10 ng/L, while minimizing interference from background organic matter and salts by selective elution and extract cleanup steps.

Working from the basic tissue method described by Du et al. 2017, we will also develop our analytical capabilities in roadway-runoff exposed fish tissue deemed most suitable (e.g. plasma, liver, gill) to better measure bioavailable concentrations of 6PPDq and other roadway chemicals. Once this method is optimized, we can use it together with runoff water sampling to better understand how quickly and how much roadway chemicals, like 6PPDq, accumulate inside runoff exposed aquatic organisms.

Activity Milestones:

Description	Approximate Completion Date
Finalize standard operating procedures to determine 6PPDq concentrations in water and roadway runoff	March 31, 2023
Transfer water analysis methods to the Minnesota Department of Health via in-person or virtual meetings	April 30, 2023
Finalize standard operating procedures to determine 6PPDq concentrations in fish tissue	June 30, 2024
Transfer fish tissue methods to the Minnesota Department of Health via in-person or virtual meetings	June 30, 2024

Activity 2: Conduct a risk-based survey of water and fish for 6PPDq in Minnesota

Activity Budget: \$250,163

Activity Description:

We know the potential exists for 6PPDq to be widespread in Minnesota lakes and rivers; however, the distribution and concentration of the chemical is unknown – critical information for risk assessment and effective management. As a first step, we will identify and spatially define risk-factors associated with potential 6PPDq exposure, such as proximity to major roads, rainfall and runoff patterns, and historical fish kill records. The resulting risk map will prioritize locations for surveillance, with special consideration for high-risk locations on tribal lands. We will strategically survey water and fish from high-risk locations and reference sites across Minnesota during rainfall and snowmelt events to maximize the detection of 6PPDq. In addition, we will actively collect samples from fish kill events to understand the frequency of 6PPDq-associated mortality. To ensure reliable and rapid response to potential 6PPDq exposure, we will create standardized sampling protocols to be used during our study and make them available for future management use. We will leverage existing fish kill response protocols and online reporting by the public and managers (<http://z.umn.edu/fishkill>). The results of this activity will be critical to informing an appropriate response by state and local managers to this emerging chemical contaminant.

Activity Milestones:

Description	Approximate Completion Date
Create publicly available online risk map of high-risk sites in Minnesota for 6PPDq exposure	April 30, 2023
Collect, analyze, and archive ~300 water and fish samples for the presence of 6PPDq	March 31, 2025
Actively sample fish kills following runoff events to determine 6PPDq exposure	June 30, 2025
Disseminate results to diverse audiences via media interviews, website, publications, and presentations	June 30, 2025

Activity 3: Evaluate the hazard 6PPDq poses to Minnesota fish

Activity Budget: \$83,037

Activity Description:

Fish cell and larval tests will be used for a rapid and cost-effective evaluation of toxicity. This portion of work is intentionally of limited scope because the chemical is not currently available/affordable. Cell and larval tests, unlike adult ones, require a small amount of chemical. Economically/ecologically important species, such as rainbow trout, bluegill and coho salmon, and chemical regulation-relevant fathead minnow will be used for cell tests. Effects of 6PPDq on cell death and mitochondria (cell “power plants” that generate energy essential for survival) will be measured because highly toxic chemicals often exert their toxicity by affecting mitochondrial function. In addition, live fish studies will be conducted with rainbow trout and fathead minnow larvae to determine whether cell/mitochondrial toxicity can lead to organismal effects (death and impairment of behaviors/physiological parameters associated with survival). If available, larvae of the additional native species (walleye and brook trout) may be tested in collaboration with the Grand Portage Band of Lake Superior Chippewa. Rapid assessment proposed here will allow us to determine if 6PPDq is likely to be toxic to a variety of Minnesota fish species and to hone in on the biological mechanisms underlying 6PPDq-associated fish kills in Minnesota.

Activity Milestones:

Description	Approximate Completion Date
Evaluate effects of 6PPDq on fish cell survival and mitochondrial function	August 31, 2023
Evaluate effects of 6PPDq on fish survival and behavior	August 31, 2024
Integrate chemical occurrence and effects data to estimate risk to fish	May 31, 2025
Disseminate results to diverse audiences via media interviews, website, publications, and presentations	June 30, 2025

Project Partners and Collaborators

Name	Organization	Role	Receiving Funds
Dalma Martinovic	University of St. Thomas	Environmental Toxicologist – Environmental sample extraction, toxicity method development and toxicity assessment. Assistance with sampling strategy design, analytical method development and data analyses associated with all the above. Student supervision, coordination of sample transfer and research activities across groups. Outreach and preparation of reports and manuscripts.	Yes
Seth Moore	Grand Portage	Director of Biology and Environment - will provide biological and ecological expertise, environmental sampling, logistics, and staff time for field sampling.	No
Mark Ferrey	MN Pollution Control Agency	Environmental scientist – will work in an advisory role to the project, contributing to the development of the project sampling plan, quality assurance, and the analysis of chemistry results.	No
Ed Kolodziej	University of Washington	Environmental Chemist - No lab in MN has a basic 6PPDq method developed. To assess the presence of the 6PPDq rapidly Dr. Kolodziej was identified as an essential out-of-state collaborator. Roles: analytical method development, sample analysis, data analyses, technology transfer, contribute to manuscript and report writing.	Yes
Mark Jankowski	US Environmental Protection Agency	Environmental toxicologist - will serve as a technical consultant for the project, providing his toxicological perspective and expertise to aid in the interpretation of the project chemical concentration data. He will serve in this role as an in-kind contribution to the project.	No

Dissemination

Describe your plans for dissemination, presentation, documentation, or sharing of data, results, samples, physical collections, and other products and how they will follow ENRTF Acknowledgement Requirements and Guidelines.

We anticipate significant interest in the results of this work and long-term impacts. With baseline data and reliable methods (Activity 1), we expect 6PPDq will be added to the regular monitoring list by the Minnesota Pollution Control Agency and may also become a candidate for Minnesota Department of Health (MDH) screenings of toxicity and exposure potential as a part of the MDH CEC - Protecting Minnesota's Water Resources Initiative. Such review may result in the development of Health Based Values and aquatic life screening values that are important for human and ecosystem health. Furthermore, we will engage with the State of Minnesota's inter-departmental contaminants working group (currently chaired by MN Department of Natural Resources) to get advice on prioritizing high-risk locations, share data and results, and inform risk assessments for future management (Activity 2). In addition, we will ensure the results of this study are made widely available to the scientific community with conference presentations, the publication of at least two open-access manuscripts, and all associated data made publicly available in the University of Minnesota's Data Repository (Activity 1-3).

Long-Term Implementation and Funding

Describe how the results will be implemented and how any ongoing effort will be funded. If not already addressed as part of the project, how will findings, results, and products developed be implemented after project completion? If additional work is needed, how will this work be funded?

We anticipate significant interest in the results of this work and long-term impacts. With baseline data and reliable methods, we expect 6PPDq will be added to the regular monitoring list by Minnesota Pollution Control Agency and may also become a candidate for Minnesota Department of Health (MDH) screenings of toxicity and exposure potential as a part of the MDH CEC - Protecting Minnesota's Water Resources Initiative. Such review may result in the development of Health Based Values and aquatic life screening values that are important for human and ecosystem health.

Other ENRTF Appropriations Awarded in the Last Six Years

Name	Appropriation	Amount Awarded
Identification of Chemicals of Emerging Concern in Minnesota Fish	M.L. 2017, Chp. 96, Sec. 2, Subd. 04g	\$400,000

Budget Summary

Category / Name	Subcategory or Type	Description	Purpose	Gen. Ineligible	% Benefits	# FTE	Classified Staff?	\$ Amount
Personnel								
Nicholas Phelps		Project Manager; Oversee all aspects of the project, coordinate team meetings and external communication, prepare written reports and presentations, etc.			36.5%	0.15		\$25,806
Post-doctoral associate		The post-doctoral associate will be involved in all aspects of the project, with primary responsibilities for sample collection, data management, project team coordination, and scientific and stakeholder communication.			25.4%	2.3		\$157,500
							Sub Total	\$183,306
Contracts and Services								
University of St. Thomas	Sub award	Dalma Martinovic (University of St. Thomas, UST) will be involved with all aspects of the project, with primary responsibilities for Activities 1 and 3, including method development, sample collection and processing, student technician supervision, and coordination with the University of Washington for analytical testing and technology transfer.				0.6		\$233,224
							Sub Total	\$233,224
Equipment, Tools, and Supplies								
	Tools and Supplies	Field supplies for sample collection (bottles, bags, necropsy tools, etc.)	Supplies are requested to support the sample collection of water and fish tissues from high-risk sites in Minnesota.					\$8,000
	Tools and Supplies	Office supplies (e.g., lab notebooks, pens, etc).	Supplies are requested for project team members to record data, procedures, and share information.	X				\$150
	Equipment	One computer	One laptop computer is requested for the post-doctoral associate for use only on this project. The ability to track samples, record data, and prepare	X				\$2,000

			publications/reports (sometimes from field locations) is an essential component of this project.					
							Sub Total	\$10,150
Capital Expenditures								
							Sub Total	-
Acquisitions and Stewardship								
							Sub Total	-
Travel In Minnesota								
	Miles/ Meals/ Lodging	3250 total miles at a rate of \$0.56/mile to collect water and fish samples.	Project team members must travel across Minnesota to collect water and fish samples. The total number of trips is TBD, but we estimate a total of 3250 total miles.					\$1,820
	Miles/ Meals/ Lodging	Project team coordination meeting with travel for three collaborators. The meeting will be held over two days in St. Paul, MN. Funds requested for travel costs and facility fees.	It is important to hold face-to-face project team coordination meetings to share results, discuss analysis, and plan for stakeholder communication. The meeting will be held in St. Paul (local for most project team members) and include travel for three collaborators from Grand Portage (Moore), University of Washington (Kolodziej), and the US EPA in Seattle (Jankowski). A second team meeting will be hosted by Grand Portage with non-state funds.					\$3,000
							Sub Total	\$4,820
Travel Outside Minnesota								
	Conference Registration Miles/ Meals/ Lodging	Travel and registration for one scientific conference (Ecological Society of America), location within United States TBD.	We expect the results of this project to be of high interest to the scientific community. It is therefore prudent that one project team member (post	X				\$2,500

			doctoral associate) travel to attend a conference in Year 3 of the project to present.					
							Sub Total	\$2,500
Printing and Publication								
	Publication	Publication of two peer-reviewed manuscripts	We will publish two peer-reviewed manuscripts and make all methods and data associated with this project publicly available through open access journals and/or the UMN Data Repository.					\$3,000
							Sub Total	\$3,000
Other Expenses								
							Sub Total	-
							Grand Total	\$437,000

Classified Staff or Generally Ineligible Expenses

Category/Name	Subcategory or Type	Description	Justification Ineligible Expense or Classified Staff Request
Equipment, Tools, and Supplies		Office supplies (e.g., lab notebooks, pens, etc).	The requested supplies are essential to conducting this work and not provided by the University of Minnesota.
Equipment, Tools, and Supplies		One computer	One laptop computer is requested for the post-doctoral associate for use only on this project. The ability to track samples, record data, and prepare publications/reports (sometimes from field locations) is an essential component of this project.
Travel Outside Minnesota	Conference Registration Miles/Meals/Lodging	Travel and registration for one scientific conference (Ecological Society of America), location within United States TBD.	We expect the results of this project to be of high interest to the scientific community. It is therefore prudent that one project team member (post doctoral associate) travel to attend a conference in Year 3 of the project to present.

Non ENRTF Funds

Category	Specific Source	Use	Status	\$ Amount
State				
			State Sub Total	-
Non-State				
In-Kind	The University of St. Thomas	Waived indirect cost recovery (41.4%) for the professional service contract to the University of St. Thomas.	Secured	\$97,797
In-Kind	University of Minnesota	Waived indirect cost recovery (55% + 25% for UST subcontract) for the University of Minnesota.	Secured	\$141,226
Cash	Grand Portage Band of Lake Superior Chippewa	Sample collection, sample analysis, travel and personnel time.	Secured	\$75,000
In-Kind	Grand Portage Band of Lake Superior Chippewa	Sample collection, sample analysis, travel and personnel time.	Secured	\$75,000
			Non State Sub Total	\$389,023
			Funds Total	\$389,023

Attachments

Required Attachments

Visual Component

File: [87d1c729-ebc.pdf](#)

Alternate Text for Visual Component

The infographic provides a visual description of the project, including the problem, question and plan.

The problem: In late 2020, researchers discovered that an analog of a common tire chemical was responsible for significant salmon mortality events in the Pacific Northwest. This highly toxic chemical is now known as "6PPDq". The research was highlighted in the popular press and concerns have quickly grown across the country, including Minnesota.

The question: Is the tire chemical 6PPDq ki...

Optional Attachments

Support Letter or Other

Title	File
Research addendum	a4c7fb40-26f.pdf
Background check	8ecc6a9d-c05.pdf

Difference between Proposal and Work Plan

Describe changes from Proposal to Work Plan Stage

No significant changes were made to the scope of work. Budget reductions were made to align with approved funding by leveraging ongoing research activities and existing personnel, personnel reduction, and reduction in sample size.

Additional Acknowledgements and Conditions:

The following are acknowledgements and conditions beyond those already included in the above workplan:

Do you understand and acknowledge the ENRTF repayment requirements if the use of capital equipment changes?

N/A

Do you agree travel expenses must follow the "Commissioner's Plan" promulgated by the Commissioner of Management of Budget or, for University of Minnesota projects, the University of Minnesota plan?

Yes, I agree to the UMN Policy.

Does your project have potential for royalties, copyrights, patents, or sale of products and assets?

No

Do you understand and acknowledge IP and revenue-return and sharing requirements in 116P.10?

N/A

Do you wish to request reinvestment of any revenues into your project instead of returning revenue to the ENRTF?

N/A

Does your project include original, hypothesis-driven research?

Yes

Does the organization have a fiscal agent for this project?

Yes, Sponsored Projects Administration