

# Final Abstract

Final Report Approved on November 27, 2024

## M.L. 2020 Project Abstract

For the Period Ending June 30, 2024

**Project Title:** Invasive Didymosphenia Threatens North Shore Streams

**Project Manager:** Mark Edlund

**Affiliation:** Science Museum of Minnesota - St. Croix Watershed Research Station

**Mailing Address:** 120 Kellogg Blvd W c/o Dr. Laurie Fink

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**Website:** <https://www.smm.org/scwrs>

**Funding Source:**

**Fiscal Year:**

**Legal Citation:** M.L. 2021, First Special Session, Chp. 6, Art. 5, Sec. 2, Subd. 06g

**Appropriation Amount:** \$197,000

**Amount Spent:** \$197,000

**Amount Remaining:** -

### Sound bite of Project Outcomes and Results

Didymo or rocksnot has invaded our North Shore. Two years of sampling showed eight streams whose future recreational opportunities, stream ecologies, and local economies may be threatened. We've informed resource managers, the public, stakeholders, colleagues, even kids about the threat of this aquatic invasive species and how we will respond.

### Overall Project Outcome and Results

The invasive alga, *Didymosphenia geminata* (aka didymo or rocksnot), has invaded clean, cold streams worldwide impacting recreational opportunities, stream ecology, and local economies. Invasive didymo appeared in the Poplar River in 2018 raising alarms. Didymo is known to grow "well-behaved" in nearshore Lake Superior, but it had never been noted in any North Shore streams. Two years of sampling North Shore didymo showed the problem is not isolated and requires a multi pronged response. We showed that didymo is now confirmed in eight North Shore streams—Caribou, Devil Track, Two Island, Poplar, Kimball, Carlson, Flute Reed, and Kadunce—all north of the Caribou River and representing the primary sites with documented colonizations. The Devil Track had didymo densities greater than 100000 cells/cm<sup>2</sup>, a bloom comparable to other worldwide invasions. The ecology of didymo in North Shore streams

followed a pattern of higher abundances in late summer and fall following spring stream scouring. Stream sites with didymo were characterized by having minimal tree canopy, cobble/boulder substrate, low or base flow, and low nutrient levels. Bacterial communities in the algal mats associated with didymo invasion are less diverse than non-didymo mats. In contrast, didymo in nearshore Lake Superior showed lower density, was limited to very nearshore, and had highly variable populations due to regular wind and wave scouring. The source of didymo in North Shore streams remains enigmatic. Molecular efforts were challenging but did show connections between North Shore didymo and Montana populations; this continues to be a research focus. Our findings have been communicated broadly to state and tribal resource managers, stakeholders, and the public through press, interviews, citizen science, presentations, fliers, a children's book, even coloring sheets. Raising awareness on the threat of didymo remains paramount as we protect our state and North Shore resources.

### **Project Results Use and Dissemination**

We identified three important findings in this project: 1) the didymo problem is larger than ever expected and requires continued study; 2) something has changed on our North Shore to allow the didymo invasion; and 3) the time to respond is now. Our results have been used to inform resource management and response (proposed signage, handouts, and felt-soled wader bans). Dissemination has been extensive and included radio, newspaper, TV, blogposts, presentations to resource managers, colleagues, and stakeholder groups, and tabled events at partner institutions. Outreach and education resources have been developed to target all audiences from policy makers to kids.



## Environment and Natural Resources Trust Fund

M.L. 2020 Approved Final Report

### General Information

**Date:** December 11, 2024

**ID Number:** 2020-035

**Staff Lead:** Noah Fribley

**Project Title:** Invasive Didymosphenia Threatens North Shore Streams

**Project Budget:** \$197,000

### Project Manager Information

**Name:** Mark Edlund

**Organization:** Science Museum of Minnesota - St. Croix Watershed Research Station

**Office Telephone:** (701) 317-4245

**Email:** medlund@smm.org

**Web Address:** <https://www.smm.org/scwrs>

### Project Reporting

**Final Report Approved:** November 27, 2024

**Reporting Status:** Project Completed

**Date of Last Action:** November 27, 2024

**Project Completion:** July 31, 2023

### Legal Information

**Legal Citation:** M.L. 2021, First Special Session, Chp. 6, Art. 5, Sec. 2, Subd. 06g

**Appropriation Language:** \$197,000 the second year is from the trust fund to the Science Museum of Minnesota to evaluate the recent spread, origin, cause, and economic and ecological threat of didymo formation in North Shore streams and Lake Superior to inform management and outreach.

**Appropriation End Date:** June 30, 2024

## Narrative

**Project Summary:** We examine the recent spread, origin, cause, and economic and ecological threat of nuisance rock snot formation in North Shore streams and Lake Superior to inform management and outreach.

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

A North Shore stream was invaded by rock snot and the time to stop it is now! In 2018, the first nuisance growth of rock snot (*Didymosphenia geminata* or “didymo”) developed in the North Shore’s Poplar River and we don’t know why. Didymo is a freshwater diatom or algae that can form nuisance goo in coldwater streams worldwide, in its native range and where it is invasive. Formation of didymo mats has aesthetic, economic, and recreational impacts. Economic impacts to tourism have exceeded \$20M annually following invasions elsewhere, a serious threat to the North Shore \$250M summer economy. Didymo disrupts normal community structure and ecosystem function in streams, alter habitat and food webs, impact fish and invertebrate abundance and diversity, and result in major shifts in natural bacterial composition.

Two hypotheses may explain rock snot: 1) The aggressive colonization hypothesis maintains an aggressive strain of didymo is being introduced and invading coldwater streams. 2) The changing environmental conditions hypothesis states environmental conditions (e.g., nitrogen to phosphorus ratios or timing of nutrient delivery) have become favorable to the formation of didymo mats. Understanding which of these models is supported by data is vital to management response.

**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.**

With LCCMR support we will understand:

- 1) Distribution, dynamics, and effect of didymo in North Shore streams
- 2) Why rock snot formed in the Poplar River and what other streams are at risk?
- 3) The source of didymo in North Shore streams—which strain is it? A new invasive? A native gone bad?
- 4) Share information and solutions with resource managers, citizen groups, and users to stop the rock snot invasion

While didymo has been documented in the near shore algal community of Lake Superior with increasing frequency since the 1960s, the Poplar River, near Lutsen, is the first stream ever colonized. Didymo is unique because it only blooms in low nutrient waters and recently, mats have been observed more frequently in streams similar to those on the North Shore around the world, including New Zealand, South America, Canada, and the US. Research shows thicker didymo mats have formed along the Superior shoreline annually for over a decade; however, it was only in 2018 that didymo was first observed colonizing North Shore streams as single cells or mat form. It is unclear why the mat formed in the Poplar River and whether didymo is already invading other North Shore streams.

**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?**

We can solve rock snot! If populations in North Shore streams and Lake Superior are not each others closest relatives, efforts for prevention of didymo mat formation will be focused on preventing movement of alien didymo among streams, paralleling practices that prevent the spread of microbes in freshwater systems. Alternatively, if the stream didymo originated from Lake Superior populations, management practices will focus on understanding the specifics that promote mat formation. Regardless, we will fully understand the source and cause of mat formation and broadly communicate the threat, implications, and management response to didymo invasion of North Shore streams.

## Project Location

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

Region(s): NE

**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: Understand didymo mat formation and distribution in North Shore streams and Lake Superior

**Activity Budget:** \$131,700

**Activity Description:**

We will monitor the Poplar River and Lake Superior shoreline near the mouth of the Poplar to determine if a didymo mat reforms in the next two years and monitor the timing and environmental conditions associated with mat formation in the stream and lake. We will similarly sample 3-4 other stream-lakeshore pairs along the North Shore to document changes in the algal community and associated environmental conditions. Sampling will be monthly from July-November 2021 and April-November 2022 and will include sampling of the algal community and chemical (e.g., nutrients, dissolved organic carbon) and physical characteristics of the stream. Temperature, water depth, and flow will be measured continuously throughout the project. During peak didymo growth (late Aug-Sept) a survey will target 20 major North Shore stream-lakeshore pairs to fully assess current didymo presence and susceptibility of North Shore resources. All sampling will adhere to MNDNR protocols for preventing spread of aquatic invasive species.

**Activity Milestones:**

Description	Approximate Completion Date
Survey all major North Shore streams for presence of didymo and invasion susceptibility during peak	December 31, 2022
Describe algae and environmental conditions in 4-5 paired Lake Superior shoreline and North Shore tributaries	March 31, 2023
Communicate findings and solutions to resource managers and citizens through presentations, signage, social media, publications	June 30, 2023

### Activity 2: Genetic variability in Minnesota didymo populations and the associated bacterial community

**Activity Budget:** \$65,300

**Activity Description:**

We will collect genetic information on North Shore tributary and Lake Superior coastal didymo populations to determine if the populations in the Poplar River and other North Shore streams are most closely related to didymo populations in Lake Superior or to other didymo populations in North America. We will use whole-genome sequencing of pools of individuals to genotype multiple didymo populations from the Poplar River and Lake Superior, didymo populations encountered in any North Shore streams, and populations from the US and Canada. We will characterize the bacterial communities using 16S rRNA gene sequencing from total DNA extracted from the periphyton mat samples to predict broader ecological consequences of didymo and learn how nuisance blooms are triggered in ultra-clean waters.

**Activity Milestones:**

Description	Approximate Completion Date
Sequence genetics of North Shore didymo populations to determine source of rock snot	March 31, 2023
Determine genetic structure of the microbial mat community among lake and stream pairs	March 31, 2023
Communicate results to inform management through meetings, signage, and presentations to resource managers and stakeholders	June 30, 2023

## Project Partners and Collaborators

Name	Organization	Role	Receiving Funds
Dr Heidi Rantala	MN DNR	Dr. Heidi Rantala (MNDNR) will provide expertise related to stream ecology and coordinate communications with local stakeholders. All partners will contribute to data analyses and writing manuscripts.	No
Dr Robert Pillsbury	UW-Oshkosh	Dr. Robert Pillsbury (UW-Oshkosh) will provide specialized molecular analyses to the project. Funding to this institution is specifically for analytical molecular analyses and does not represent direct salary. All partners will contribute to data analyses and writing manuscripts.	Yes

## 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 will share the findings of this study with resource managers (MNDNR, including state parks; MPCA; watershed groups) throughout the study period through meetings and personal communication. Edlund and Rantala are periodically invited to give presentations within their organizations and to outside groups, and they will present this work upon invitation. We will communicate the findings of this study with the public through signage, factsheets, and social media (Twitter and Facebook) accounts associated with their agencies. We plan on publishing the results of this work as two peer-reviewed publications in relevant scientific journals.

The Minnesota Environment and Natural Resources Trust Fund (ENRTF) will be acknowledged through use of the trust fund logo or attribution language on project print and electronic media, publications, signage, and other communications per the ENRTF Acknowledgement Guidelines.

## 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 will share the findings of this study with resource managers (MNDNR, including state parks; MPCA; watershed groups) and stakeholders throughout the study period through meetings and personal communication. Edlund and Rantala are periodically invited to give presentations within their organizations and to outside groups, and they will present this work upon invitation. We will communicate the findings of this study with the public through signage, factsheets, and social media (Twitter and Facebook) accounts associated with their agencies. We plan on publishing the results of this work as two peer-reviewed publications in relevant scientific journals.

## Other ENRTF Appropriations Awarded in the Last Six Years

Name	Appropriation	Amount Awarded
Tracking and Preventing Harmful Algal Blooms	M.L. 2016, Chp. 186, Sec. 2, Subd. 04a	\$500,000
Determining Risk of a Toxic Alga in Minnesota Lakes	M.L. 2018, Chp. 214, Art. 4, Sec. 2, Subd. 06f	\$200,000

## Budget Summary

Category / Name	Subcategory or Type	Description	Purpose	Gen. Ineligible	% Benefits	# FTE	Classified Staff?	\$ Amount	\$ Amount Spent	\$ Amount Remaining
<b>Personnel</b>										
Dr. Mark Edlund, Project Director and Senior Scientist at Science Museum of Minnesota		Project direction, periphyton analysis, diatom identification			45.5%	0.6		\$57,000	-	-
2 Interns, Science Museum of Minnesota		Field work, lab tech			12%	0.4		\$13,642	-	-
Communication Specialist, Science Museum of Minnesota		Communication, outreach, social media			45.5%	0.05		\$2,000	-	-
Dr David Burge		Assistant Sci, molecular analysis			12%	0.5		\$21,147	-	-
							<b>Sub Total</b>	<b>\$93,789</b>	<b>\$93,789</b>	-
<b>Contracts and Services</b>										
University of Wisconsin-Oshkosh	Subaward	Bacterial Genetic Analysis: 200 samples @ \$130.80/sample (Total \$26,160; University of Wisconsin-Oshkosh, Dr. Robert Pillsbury, or competitive bid)				0		\$26,160	\$26,160	-
University of MN Genomics Center	Professional or Technical Service Contract	Didymo Genetic Analyses: Long-read molecular analyses, \$3500 Univ MN Genomics Ctr, or competitive bid				0		\$3,500	\$3,500	-
ZymoResearch Corp, Irvine, CA	Professional or Technical Service Contract	Molecular shotgun analyses of didymo collections, \$5872				0		\$5,872	\$5,872	-
							<b>Sub Total</b>	<b>\$35,532</b>	<b>\$35,532</b>	-



<b>Equipment, Tools, and Supplies</b>										
	Equipment	Stream Gauging Equipment: five 13-foot water levels @ \$495 each; software \$75; communication cable \$249	Equipment for stream gauging					\$2,799	\$2,799	-
	Tools and Supplies	Consumable Supplies: \$4,000 field supplies; \$1,320 microbial DNA extraction kits	Consumable supplies for field work and microbial DNA extraction					\$5,320	\$5,320	-
							<b>Sub Total</b>	<b>\$8,119</b>	<b>\$8,119</b>	-
<b>Capital Expenditures</b>										
							<b>Sub Total</b>	-	-	-
<b>Acquisitions and Stewardship</b>										
							<b>Sub Total</b>	-	-	-
<b>Travel In Minnesota</b>										
	Miles/ Meals/ Lodging	Travel for Field Work: Round-trip drive from St. Croix Watershed Research Station to Grand Marais, MN for 2 employees: 18 days each FY20 and FY21 (\$16,000 over two yrs)	Travel for Field Work					\$16,000	\$16,000	-
							<b>Sub Total</b>	<b>\$16,000</b>	<b>\$16,000</b>	-
<b>Travel Outside Minnesota</b>										
							<b>Sub Total</b>	-	-	-
<b>Printing and Publication</b>										
	Publication	Cost of open access publication in PLOS ONE peer-reviewed open access scientific journal (\$0)	This line has been removed to provide salary for project synthesis to project personnel					-	-	-
							<b>Sub Total</b>	-	-	-

Other Expenses									
		Water Chemistry Analyses	Water chemistry analyses, St. Croix Watershed Research Station, \$198/sample (for suite of 10 analyses), 110 samples in FY21 and 110 samples in FY22 (Total \$43,560)				\$43,560	\$43,560	-
						<b>Sub Total</b>	<b>\$43,560</b>	<b>\$43,560</b>	-
						<b>Grand Total</b>	<b>\$197,000</b>	<b>\$197,000</b>	-

Classified Staff or Generally Ineligible Expenses

Category/Name	Subcategory or Type	Description	Justification Ineligible Expense or Classified Staff Request
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Non ENRTF Funds

Category	Specific Source	Use	Status	\$ Amount	\$ Amount Spent	\$ Amount Remaining
<b>State</b>						
			<b>State Sub Total</b>	-	-	-
<b>Non-State</b>						
In-Kind	Science Museum of Minnesota	Contribution of portion of Rantala Salary: FY21 10% of time (208 hr), salary \$36.32/hr, fringe \$8.99/hr; FY22 10% of time (208 hr), salary \$37.63/hr, fringe \$9.39/hr (Total \$19,205 over 2 years)	Secured	\$19,205	\$19,205	-
In-Kind	Science Museum of Minnesota	Indirect Costs for the project are provided in-kind by the Science Museum of Minnesota (federally negotiated indirect rate 40.09% on all direct costs = \$78,977)	Secured	\$78,977	\$78,977	-
In-Kind	Pillsbury	Genetic Laboratory Fees are provided in-kind by Pillsbury (\$1,744 total over 2 years)	Secured	\$1,744	\$1,744	-
			<b>Non State Sub Total</b>	<b>\$99,926</b>	<b>\$99,926</b>	-
			<b>Funds Total</b>	<b>\$99,926</b>	<b>\$99,926</b>	-

## Attachments

### Required Attachments

#### *Visual Component*

File: [442e2249-e62.pdf](#)

#### *Alternate Text for Visual Component*

We examine the recent spread, origin, cause, and economic and ecological threat of nuisance rock snot formation in North Shore streams and Lake Superior to inform management and outreach....

### Supplemental Attachments

#### *Capital Project Questionnaire, Budget Supplements, Support Letter, Photos, Media, Other*

Title	File
Institutional Letter of Support, Science Museum of Minnesota	<a href="#">7d296146-447.pdf</a>
SF-990, Institutional Tax Exempt Form, Science Museum of Minnesota	<a href="#">8204cf17-0af.pdf</a>
Background Check Certification Form	<a href="#">80769076-b99.pdf</a>
didymo outreach.pdf	<a href="#">269ce474-af0.pdf</a>
2022DidymoFactsheet_20230210.pdf	<a href="#">0ea973ee-7e4.pdf</a>
Didymo stream coloring sheet_A11Ys_FAW_0022_2023..pdf	<a href="#">95f9f721-38e.pdf</a>
Healthy stream coloring sheet_A11Ys_FAW_0021_2023.pdf	<a href="#">af0d1b9e-ff0.pdf</a>
2021NshoreDidymosummary.pdf	<a href="#">314b3ef3-2cf.pdf</a>

### Difference between Proposal and Work Plan

#### *Describe changes from Proposal to Work Plan Stage*

Sampling timetable and reporting timetable updated to reflect receipt of funding award in 2021.

## 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 understand that travel expenses are only approved if they 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 understand the Commissioner's Plan applies.

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

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?**

No

## Work Plan Amendments

Amendment ID	Request Type	Changes made on the following pages	Explanation & justification for Amendment Request (word limit 75)	Date Submitted	Approved	Date of LCCMR Action
1	Amendment Request	<ul style="list-style-type: none"> <li>• Project Collaborators - Project Partner Info</li> <li>• Budget - Personnel</li> <li>• Budget - Professional / Technical Contracts</li> <li>• Budget - Printing and Publication</li> </ul>	<p>We request budget amendments to 1) move \$20448 from Tech Services to Personnel (hired Dr David Burge at SMM, so contract not needed), 2) move \$699 from Publications to Personnel for project synthesis (Dr Burge). This changes the Project Personnel line from \$72642 to \$93789; the Tech Services line to Univ Arkansas from \$29820 to \$9372 for UMin Genomics (\$3500), ZymoResearch ((\$5872) molecular analyses (revised Tech Services line \$35532); and the Publications line to \$0.</p>	June 28, 2023	Yes	June 28, 2023

# Status Update Reporting

## Final Status Update September 14, 2023

**Date Submitted:** October 25, 2024

**Date Approved:** November 27, 2024

### Overall Update

We intensively sampled five stream and lakeshore sites in late summer and fall of 2021 as well as surveyed 20 additional sites along the North Shore in late August 2021 for didymo. We returned in 2022, and with the same sampling strategy. To date, eight streams that never had recorded didymo populations before 2021 were found, indicating the invasion of North Shore streams is worse than originally thought. A few 2022 samples are still undergoing microscopical analysis, water quality analyses are completed, and molecular analyses are underway to meet our project Outcomes. Outreach and communication for didymo has been extensive and has included newspaper pieces (Duluth Tribune, Strib), a radio interview (WTIP), blog posts (Quetico-Superior Foundation), TV (PrairieSportsman) and social media. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities.

### Activity 1

We sampled North Shore stream and lakeshore sites for didymo and water quality in 2021 including trips to our five premier monitoring sites and one trip that included blitz sampling of 20 additional stream/lakeshore. We returned from May-Oct 2022, and again sampled our premier and blitz sites. Results have been surprising. We found didymo populations in eight streams where didymo had never been reported! Water quality and physical stream measures suggest that streams with didymo have cobble or boulder/bedrock substrate, are at long-term baseflow, and have low nutrient concentrations of nitrogen and phosphorus. Analysis of didymo collections showed several rivers had greater abundance of didymo than the lakeshore collections with up to 100,000 cells per cm<sup>2</sup> in the Devil Track River—comparable to other worldwide invasion—higher densities in the late summer/fall, variable abundances among years, and some streams with didymo in 2021 did not have evident didymo in 2022. Lakeshore didymo ecology differed; there was high variability over the sampling years, likely associated with wind and wave action. Also of note is that the algae communities associated with didymo in the streams were significantly different than didymo communities in Lake Superior. See dissemination results below.

*(This activity marked as complete as of this status update)*

### Activity 2

Samples for molecular analysis were taken at stream/lakeshore sites in 2021-2022. Samples were screened for presence/absence of didymo and degree of mat formation. A river and lake population were chosen for genomic screening using long-read sequencing to elucidate as much of the didymo genome as possible. DNA recovery was suboptimal and has limited use of these data. Other didymo positive samples were subjected to shotgun sequencing to detect population differences. We received samples from other USA populations for comparison with North Shore populations. We attempted barcode analysis of didymo populations from select rivers. In short we have been unable to fully ascertain the source of didymo in North Shore streams to date without strong long-read data. Barcode analysis shows strong connection to didymo populations from Montana (with a cautionary note we are currently without good comparison yet to Lake Superior populations), but we are continuing our efforts to ascertain the didymo source. We identified significant variation in nitrogen cycling genes among sites suggesting that didymo mats drive changes in nutrient dynamics in streams. Samples for bacterial genomics from were analyzed and show strong bacterial community differences between didymo and non-didymo mats and between lake and river sites.

*(This activity marked as complete as of this status update)*



## **Dissemination**

Outreach and communication for didymo has been extensive and has included three newspaper pieces (Duluth Tribune, Strib, Cook Co News Herald), web (Bring Me the News), two radio interviews (WTIP), blog posts (Quetico-Superior Foundation), two TV episodes (Prairie Sportsman), and social media. Our group has spoken within our own agencies and organizations (SMM, UWoshkosh, and MNDNR), presented seminars to stakeholders (Duluth Aquarium, Arrowhead FF), and hosted meetings with project partners to discuss findings, management response, and outreach activities. Other partners have been contacted during various meetings and conferences (NALMS, LoW Forum, DNR Fish Academy, 2022 North American Diatom Symposium, 2023 MN Waters Conference) where didymo information was presented. Peer-reviewed articles are being prepared. Outreach activities have primarily been on the floor of the Science Museum of Minnesota (Science Rocks, A Evening at the Museum), in the field (Field Encounters of the STEM Kind), in the classroom, through a partnership with Grand Marais Story Scouts to produce a rocksnot children's book, and have included distribution of project handouts and didymo coloring sheets outlining the problem, how to spot didymo, and invitations to help solve the didymo problem. Our dissemination activities are uploaded as attachments, and all dissemination included acknowledgement of ENTRF.

# Status Update Reporting

## Status Update April 1, 2023

**Date Submitted:** June 28, 2023

**Date Approved:** June 28, 2023

### Overall Update

We have intensively sampled five stream and lakeshore sites in late summer and fall of 2021 as well as surveyed 20 additional sites along the North Shore in late August 2021 for didymo. We returned in May-Oct 2022, with the same sampling strategy on five premier sites and 20 blitz sites. To date, eight streams that had never had recorded didymo populations before 2021 were found, indicating the invasion of North Shore streams is worse than originally thought. A few 2022 samples are still undergoing microscopical analysis, water quality analyses are completed, and molecular analyses are underway to meet our project Outcomes. Outreach and communication for didymo has been extensive and has included newspaper pieces (Duluth Tribune, Strib), a radio interview (WTIP), blog posts (Quetico-Superior Foundation), TV (Prairie Sportsman), and social media. Our group has also participated in outreach activities, spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities.

### Activity 1

We sampled North Shore stream and lakeshore sites for didymo and water quality in 2021 (August, September, October) including three trips to our five premier sites and one trip that included a blitz sampling of 20 additional streams. We returned from May-Oct 2022, and again sampled our premier and blitz sites. Results have been surprising. We found didymo populations in eight streams where didymo had never been reported. Water quality analyses are completed and are suggesting that streams with didymo were at long-term baseflow and with low nutrients. Microscopic analysis of didymo collections showed several rivers had greater abundance than lakeshore collections with up to 100,000 cells/cm<sup>2</sup> in (Devil Track River); lower abundances in general were seen in 2022. We note the algae communities associated with didymo in streams are significantly different than didymo communities in Lake Superior.

Outreach and communication for didymo has been extensive and included two newspaper pieces (Duluth Tribune, Strib), radio interview (WTIP), blog posts (Quetico-Superior Foundation), TV (PrairieSportsman), and social media. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted meetings with all project partners to discuss findings, management response, and outreach activities.

### Activity 2

Samples for molecular analysis were taken at all North Shore stream and lakeshore sites sampled in 2021 and 2022. Upon return to the laboratory, samples were screened under the microscope for presence/absence of didymo and degree of mat formation. For didymo analysis, a river and a lake population were chosen for initial genomic screening using long-read sequencing to elucidate as much of the entire didymo genome as possible. DNA recovery was suboptimal; however, the UMGC is currently running the analysis. The other didymo positive samples have been subjected to shotgun sequencing to detect population differences. We also received samples from other USA populations of didymo for comparison with North Shore populations. Samples for bacterial genomics from 2021 and 2022 have been sent to Dr. Pillsbury at UW-Oshkosh and are undergoing analysis.. Other didymo studies have shown strong bacterial differences between didymo and non-didymo mats, and we hope to use our results to help decipher how stream didymo on the North Shore have been able to get a foothold. Project partners have been updated on progress-to-date on our genomic analyses.

### Dissemination

Outreach and communication for didymo has been extensive and has included two newspaper pieces (Duluth Tribune, Strib), a radio interview (WTIP), blog posts (Quetico-Superior Foundation), TV (Prairie Sportsman) and social media. Our

group has also spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities. Other partners have been contacted during various meetings and conferences (NALMS, LoW Forum). Outreach activities have primarily been on the floor of the Science Museum of Minnesota (Science Rocks, A Evening at the Museum) and have included distribution of project handouts, preventing didymo, and didymo coloring sheets. Project results were reported at the 2022 North American Diatom Symposium.

# Status Update Reporting

## Status Update October 1, 2022

**Date Submitted:** October 17, 2022

**Date Approved:** October 17, 2022

### Overall Update

We have intensively sampled five stream and lakeshore sites in late summer and fall of 2021. In 2022 we repeated this sampling scheme once per month and have found didymo mats in different streams than 2021. We are sampling Kimball Creek intensively as it is blooming didymo this year. All samples are undergoing microscopical, water quality, and molecular analyses as planned to reach our project Outcomes. We have begun analysis of all data collected. Preliminary results indicate strong correlation between low dissolved carbon content and didymo mat formation in late summer. Outreach and communication for didymo has been extensive and has included newspaper pieces, a radio interview, TV interview with The Prairie Sportsman (PBS), blog posts and social media. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, conferences, webinars, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities.

### Activity 1

We sampled North Shore stream and lakeshore sites for didymo and water quality on 3 trips in 2021 (August, September, October) and 6 trips in 2022. Results were surprising. We found didymo populations in 12 streams to date whereas we reported 7 in 2021. Water quality analyses continue but are suggesting that streams were at baseflow and with low nutrient values. Specifically we have identified that lower dissolved organic carbon is associated with increased didymo abundance, and increased temperature (likely a seasonal signal) is associated with increased didymo abundance. Microscopic analysis of didymo collections showed several rivers had greater abundance of didymo than the lakeshore collections with up to 100,000 cells per cm<sup>2</sup> in the Devil Track River in 2021 and Kimball Creek in 2022. These streams exhibit extensive and thick coverage of didymo mucus-mats. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, presented our findings at the North American Diatom Symposium and the Diatom Web Academy where we met with other didymo researchers throughout the USA and world. We also hosted a meeting with all project partners to discuss findings, management response, and outreach activities.

### Activity 2

Samples for molecular analysis were taken at all North Shore stream and lakeshore sites sampled in 2021. Upon return to the laboratory, samples were screened under the microscope for presence/absence of didymo and degree of mat formation. For didymo analysis, a river and a lake population were chosen for initial genomic screening using long-read sequencing to elucidate as much of the entire didymo genome as possible. DNA recovery was suboptimal so a revised extraction method will be used. The other didymo positive samples will be subjected to shotgun sequencing to detect population differences. We have also begun to receive samples from other USA populations of didymo for comparison with North Shore populations. Samples for bacterial genomics from 2021 have been sent to Dr. Pillsbury at UW-Oshkosh and are undergoing analysis. Sampling for 2022 will be complete in mid-October, and 2022 samples will be sent to Dr. Pillsbury shortly. Other didymo studies have shown strong bacterial differences between didymo and non-didymo mats, and we hope to use our results to help decipher how stream didymo on the North Shore have been able to get a foothold. Project partners have been updated on progress-to-date on our genomic analyses.

### Dissemination

Outreach and communication for didymo has been extensive and has included two newspaper pieces (Duluth Tribune, Strib), two radio interviews (WTIP), a TV spotlight (Prairie Sportsman/PBS; episode expected Spring 2023), blog posts (Quetico-Superior Foundation), and social media. Our group has also spoken within our own agencies and organizations,

presented seminars to stakeholders, presented at national and international venues (in-person and virtual), and hosted a meeting with all project partners to discuss findings, management response, and outreach activities. We also presented our initial findings at the North American Diatom Symposium (in-person/virtual) and the Diatom Web Academy (virtual) where we met with other diatom researchers throughout the USA and world. In all dissemination efforts, we acknowledge the ENTRF and LCCMR for their support.

# Status Update Reporting

## Status Update April 1, 2022

**Date Submitted:** May 17, 2022

**Date Approved:** May 18, 2022

### Overall Update

We have intensively sampled five stream and lakeshore sites in late summer and fall of 2021 as well as surveyed 20 additional sites along the North Shore in late August 2021 for didymo. Surprisingly, seven streams that had never had recorded didymo populations were found indicating the invasion of North Shore streams is worse than originally thought. All samples are undergoing microscopical, water quality, and molecular analyses as planned to reach our project Outcomes. Outreach and communication for didymo has been extensive and has included newspaper pieces (Duluth Tribune, Strib), a radio interview (WTIP), blog posts (Quetico-Superior Foundation), and social media. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities.

### Activity 1

We sampled North Shore stream and lakeshore sites for didymo and water quality on 3 trips in 2021 (August, September, October) including three trips to our five premier monitoring sites and one trip that included a blitz sampling of 20 additional streams. Results were surprising. We found didymo populations in seven streams where didymo had never been reported. Water quality analyses continue but are suggesting that streams were at baseflow and with low nutrient values. Microscopic analysis of didymo collections showed several rivers had greater abundance of didymo than the lakeshore collections with up to 100,000 cells per cm<sup>2</sup> in the Devil Track River. Also of note is that the algae communities associated with didymo in the streams were significantly different than didymo communities in Lake Superior.

Outreach and communication for didymo has been extensive and has included two newspaper pieces (Duluth Tribune, Strib), radio interview (WTIP), blog posts (Quetico-Superior Foundation), and social media. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities.

### Activity 2

Samples for molecular analysis were taken at all North Shore stream and lakeshore sites sampled in 2021. Upon return to the laboratory, samples were screened under the microscope for presence/absence of didymo and degree of mat formation. For didymo analysis, a river and a lake population were chosen for initial genomic screening using long-read sequencing to elucidate as much of the entire didymo genome as possible. DNA recovery was suboptimal so a revised extraction method will be used. The other didymo positive samples will be subjected to shotgun sequencing to detect population differences. Samples for bacterial genomics have been sent to Dr. Pillsbury at UW-Oshkosh and are undergoing analysis. Other didymo studies have shown strong bacterial differences between didymo and non-didymo mats, and we hope to use our results to help decipher how stream didymo on the North Shore have been able to get a foothold. Project partners have been updated on progress-to-date on our genomic analyses.

### Dissemination

Outreach and communication for didymo has been extensive and has included two newspaper pieces (Duluth Tribune, Strib), a radio interview (WTIP), blog posts (Quetico-Superior Foundation), and social media. Our group has also spoken within our own agencies and organizations, presented seminars to stakeholders, and hosted a meeting with all project partners to discuss findings, management response, and outreach activities.