Final Abstract

Final Report Approved on December 1, 2025

M.L. 2021 Project Abstract

For the Period Ending June 30, 2025

Project Title: Microgeographic Impact of Antibiotics Released from Identified Hotspots

Project Manager: Randall Singer

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Funding Source:

Fiscal Year:

Legal Citation: M.L. 2021, First Special Session, Chp. 6, Art. 6, Sec. 2, Subd. 04d and M.L. 2024, Chp. Sec. 2, Subd. 18

Appropriation Amount: \$508,000

Amount Spent: \$507,048

Amount Remaining: \$952

Sound bite of Project Outcomes and Results

The project demonstrated that the effluent from wastewater treatment plants was a key source of antibiotics entering small streams/rivers in Minnesota. We developed a model that can be used by anyone to predict the fate and transport of different chemicals in streams of various sizes.

Overall Project Outcome and Results

Antimicrobial resistance (AMR) threatens public health, animal health, and ecosystem health. Antibiotic use in hospitals, long-term care facilities, and animal husbandry operations (point sources) play a major role in AMR emergence. Discharges and runoff from these point sources enter the natural environment, in some cases after going through a treatment system at the point source itself or at a wastewater treatment plant. Antibiotic residues are found in the surface waters nearby the point sources. In this project, we successfully measured antibiotic residues in small streams/rivers receiving wastewater effluent. By sampling upstream, at, and downstream of the discharge, it was shown that the treatment plants were the source of antibiotics to these reaches of the streams.

We combined the collected field data with information from the literature and from laboratory experiments that we

conducted to build a model to allow simulation of antibiotic fate and transport in the stream. The model is publicly available and can be used by anyone to predict the fate and transport of different chemicals in streams of various sizes. By combining different types of data, the user can predict the concentration profile of an antibiotic (or other pollutant) in a stream as a function of distance. The model allows comparison of the prediction to collected field data. The program and manual were developed to be user friendly.

Overall, this project expanded our understanding of the sources, dissemination routes, and persistence of different antibiotics in Minnesota's waterways. Our model will allow others to make similar predictions in other geographic locations.

Project Results Use and Dissemination

We have disseminated the results of this project in multiple ways. First, we have published multiple peer-reviewed scientific papers and have presented these results at scientific meetings. Second, we have developed a model to allow simulation of antibiotic (or other organic pollutant) transport and degradation downstream of point sources and have made this model publicly available. The availability of the model was advertised through social media channels. Finally, we have kept other similar efforts apprised of our progress, such as the Minnesota One Health Antibiotic Stewardship Collaborative. We have shared the collected data with the partner wastewater treatment plants.



Environment and Natural Resources Trust Fund

M.L. 2021 Approved Final Report

General Information

Date: December 2, 2025

ID Number: 2021-144

Staff Lead: Michael Varien

Project Title: Microgeographic Impact of Antibiotics Released from Identified Hotspots

Project Budget: \$508,000

Project Manager Information

Name: Randall Singer

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Project Reporting

Final Report Approved: December 1, 2025

Reporting Status: Project Completed & Additional Update Approved

Date of Last Action: December 1, 2025

Project Completion: June 30, 2025

Legal Information

Legal Citation: M.L. 2021, First Special Session, Chp. 6, Art. 6, Sec. 2, Subd. 04d and M.L. 2024, Chp. Sec. 2, Subd. 18

Appropriation Language: \$508,000 the first year is from the trust fund to the Board of Regents of the University of Minnesota to inform protection of environmental, animal, and human health from proliferation of antibiotic resistance by quantifying and mapping the extent of antibiotic spread in waters and soils from locations identified as release hot spots.and (a) The availability of the appropriations for the following projects is extended to June 30, 2025: (15) Laws 2021, First Special Session chapter 6, article 6, section 2, subdivision 4, paragraph (d), Microgeographic Impact of Antibiotics Released from Identified Hotspots;

Appropriation End Date: June 30, 2025

Narrative

Project Summary: We will quantify the release of antibiotics from hotspots identified in our previous project to surface waters in Minnesota using field, laboratory, and modeling approaches to ultimately inform interventions.

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

Antimicrobial resistance (AMR) threatens public health, animal health, and ecosystem health. Antibiotic use in hospitals, long-term care facilities, and animal husbandry operations (point sources) play a major role in AMR emergence. Discharges and runoff from these point sources enter the natural environment, in some cases after going through a treatment system at the point source itself or at a wastewater treatment plant. More than 50% of the antibiotic compounds used at the point source may be released unchanged or as an active metabolite into the environment. Antibiotic residues are found in the surface waters nearby the point sources. We still do not fully understand how far these residues can be transported, how long they remain in the water, and how they vary depending on the antibiotic use at the point sources. It is imperative to understand these issues to establish appropriate mitigation strategies at the point source to minimize the impact to human, animal, and ecosystem health. This project will quantify the antibiotics released from hotspots in space and time and how the dissemination and persistence of these antibiotics is affected by environmental characteristics as well as specific traits of the chemicals themselves.

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.

To fill in the important knowledge gaps that are needed to establish interventions to combat the impact of AMR on health-related outcomes, we propose to assess the influence of specific point sources on the dissemination of antibiotics in surface waters spatially and temporally. We will choose these point sources based on our previous large-scale footprint model of the entire state of Minnesota established with previous ENRTF support. By evaluating specific point sources (i.e. hospitals, wastewater treatment plants, animal husbandry operations) at a micro-geographical scale and over time, we will be able to correlate antibiotic prescription data throughout the year with environmental concentrations of antibiotic residues. We will be collecting field samples following our refined methodology from our previous project, as well as conducting laboratory experiments to fully understand factors such as degradation and association with particulates in the water column, which are critical variables to include in modeling efforts. Our models will ultimately indicate what type of interventions are needed at these point sources to minimize the impact of antibiotics on human, animal, and ecosystem health.

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 will generate data that correlates the impact of specific point sources with the dissemination of antibiotics into the environment both temporally and spatially. That information will allow improvement of waste management interventions at each one of the point sources studied, and better mitigation strategies to decrease the spread of antibiotics and AMR into the environment.

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: Point source data collection and modeling of antibiotic residues on surface waters

Activity Budget: \$273,750

Activity Description:

We will select point sources in specific watersheds in Minnesota that represent hospitals, wastewater treatment plants, and animal husbandry operations. This selection will be based on our previously developed footprint model for the entire state, which highlighted hotspots of antibiotic levels in surface waters in relation to point sources. We will collect data on antibiotic use and on waste management practices at these sources. This information will be provided leveraging the Minnesota One Health Antibiotic Stewardship Collaborative, as well as through collaborations that spearheaded from our previous ENTRF funded projects. Using these data and results from Activity 2, we will develop geospatial and hydrological models to predict the dissemination of antibiotic residues from the point sources over time and over space in the watersheds of interest. We will also conduct epidemiological analyses to correlate antibiotic use and waste management practices with the field measurements for the antibiotic residues (Activity 2). Finally, we will integrate all results into a user-friendly interface to be used by managers at the point sources to visualize their impact into the watershed. That interface will also allow us to model intervention strategies and assess how the antibiotic emission levels change based on different treatment scenarios.

Activity Milestones:

Description	Approximate Completion Date
Data collection on antibiotic use and waste management practices at the identified point sources	December 31, 2021
Identify specific point sources within a watershed based on previous state wide footprint model	June 30, 2022
Development of user-friendly interface	June 30, 2024
Geospatial and hydrological modeling using data from Activity 2	June 30, 2024
Epidemiological analyses	June 30, 2024

Activity 2: Field measurements downstream of point sources and laboratory transformation studies

Activity Budget: \$234,250

Activity Description:

Once point sources are identified, the levels emitted into the environment and any seasonal variations need to be quantified with field measurements. Based on information collected in Activity 1, up to 10 sites that cover different sources (e.g., hospitals, wastewater treatment plants, animal husbandry operations) will be selected. For three of these sites, monthly sampling from April to October will be conducted (upstream, at the discharge point, and five locations downstream). For the other seven sites, quarterly sampling will be performed. To fully understand the persistence of antibiotics in aquatic systems and fully parameterize models (Activity 1), information about degradation rates of different compounds is needed. Relevant removal processes include transformation by sunlight, degradation by bacteria, and sorption to settling particles. While degradation by bacteria seems counter-intuitive, at subtherapeutic levels, bacteria can degrade antibiotics. For the targeted compounds, a literature review will be conducted to compile relevant rate constants. Because biodegradation rates will be affected by season and the native microbial community and sorption will be affected by the type and load of particles present, experiments will be needed to obtain rate constants relevant to Minnesota. Experiments for sunlight degradation will be used to fill in literature gaps.

Activity Milestones:

Description	Approximate Completion Date
Literature review of transformation rates	July 31, 2022

Photolysis studies	December 31, 2022
Sorption studies	July 31, 2023
Field measurements of antibiotic levels over distance from point sources (monthly sites)	October 31, 2023
Biodegradation studies	May 31, 2024
Field measurements of antibiotic levels over distance from point sources (quarterly sites)	May 31, 2024

Project Partners and Collaborators

Name	Organization	Role	Receiving Funds
Amy Kinsley	University of Minnesota	Co-Investigator	Yes
Amanda Beaudoin	Minnesota Department of Public Health	Co-Investigator	No
Kristine Wammer	University of St. Thomas	Co-Project Manager	Yes
Irene Bueno	University of Minnesota	Co-Project Manager	Yes
William Arnold	University of Minnesota	Co-Project Manager	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.

Findings will be disseminated and archived via reports to LCCMR, peer-reviewed publications (note funds are requested to allow publication in open access journals to enhance dissemination), and presentations at conferences. We will also, when appropriate, disseminate results via the media. The audience is not only the scientific community, but also the public, policymakers, and practitioners. The work will also be of interest to the medical community, and we will seek avenues to share the results with this community. Acknowledgment of funding from the Environment and Natural Resources Trust Fund will be made following the guidelines described at

https://www.lccmr.leg.mn/pm_info/acknowledgement_guidelines.pdf. For example, all presentations will include the ENTRF logo on the acknowledgment slide. All published manuscripts will acknowledge the specific grant that was awarded.

Water, soil, sludge, and manure samples will be freeze-dried for potential future analyses. Extracts will also be labeled and archived (frozen) for potential future analyses.

In July 2016, Minnesota Departments of Health and Agriculture, Minnesota Pollution Control Agency, and Minnesota Board of Animal Health pledged multi-agency support to address the problem of AMR. Since that time, the Minnesota One Health Antibiotic Stewardship Collaborative, consisting of professionals from state government agencies, academia, clinical human and animal health practice, industry, and professional groups, have worked together to meet the goals of the Minnesota One Health Antibiotic Stewardship strategic plan. The Collaborative will provide ongoing support and access to subject matter expertise throughout this project. In addition, this group of professionals, organizations, and supporting policy makers, will be kept up to date on progress and provided with practical documents developed as a result of this work (e.g., fact sheets, reports, publications). The broad Collaborative structure will also facilitate dissemination of outcomes from this work to the public, medical and agricultural industries, and human, animal, and environmental health professionals throughout Minnesota.

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?

Our project will define relationships between essential activities (e.g., healthcare, wastewater treatment, animal

agriculture) and the maintenance and proliferation of AMR in Minnesota's natural environment. The long-term goal is to develop scientific and risk-based guidance in human, animal, and environmental health for the mitigation of AMR in the natural environment. Our approach will also be useful to explore other biologically active chemicals in Minnesota's environment, such as hormones and endocrine disruptors. Results will be shared at local conferences, in open-access scientific publications, by publicly available final report, and through the Minnesota One Health Antibiotic Stewardship Collaborative.

Other ENRTF Appropriations Awarded in the Last Six Years

Name	Appropriation	Amount Awarded
Solar Drivan Doctruction of Docticidos	M.L. 2014 Chr. 226 Cos. 2 Subd. 025	
Solar Driven Destruction of Pesticides,	M.L. 2014, Chp. 226, Sec. 2, Subd. 03a	\$291,000
Pharmaceuticals, Contaminants in Water		
Antibiotics and Antibiotic Resistance Genes in	M.L. 2014, Chp. 226, Sec. 2, Subd. 03e	\$300,000
Minnesota Lakes		
Assessing Techniques for Eliminating Contaminants to	M.L. 2016, Chp. 186, Sec. 2, Subd. 04d	\$287,000
Protect Native Fish and Mussels		
Assessing Neonicotinoid Insecticide Effects on Aquatic	M.L. 2016, Chp. 186, Sec. 2, Subd. 04e	\$400,000
and Soil Communities		
Assessment of Household Chemicals and Herbicides in	M.L. 2017, Chp. 96, Sec. 2, Subd. 04a	\$236,000
Rivers and Lakes		
Mapping Antibiotic Resistance in Minnesota to Help	M.L. 2018, Chp. 214, Art. 4, Sec. 2, Subd. 04h	\$750,000
Protect Environmental, Animal, and Human Health		
Determining Influence of Insecticides on Algal Blooms	M.L. 2019, First Special Session, Chp. 4, Art. 2, Sec. 2,	\$350,000
	Subd. 04a	
Benign Design: Environmental Studies Leading to	M.L. 2019, First Special Session, Chp. 4, Art. 2, Sec. 2,	\$415,000
Sustainable Pharmaceuticals	Subd. 04b	

Budget Summary

Category / Name	Subcategory or Type	Description	Purpose	Gen. Ineli gible	% Bene fits	# FTE	Class ified Staff?	\$ Amount	\$ Amount Spent	\$ Amount Remaining
Personnel										
Undergraduate		Assist with field sampling/laboratory experiments and literature review			0%	0.5		\$56,000	-	1
Post-Doctoral Associate		Perform field measurements/laboratory experiments			25%	1.5		\$146,000	-	-
Amy Kinsley		Co-Investigator			27%	0.06		\$1,500	-	-
Irene Bueno		Co-Project Manager and Geospatial Modeler			20%	2		\$16,100	-	-
William Arnold		Co-Project Manager			27%	0.12		\$60,000	-	-
Randall Singer		Project Manager			27%	0.12		\$66,000	-	-
Graduate Student		Assist with field sampling and sample processing			20%	0.5		\$53,400	-	-
							Sub Total	\$399,000	\$398,827	\$173
Contracts and Services										
University of St. Thomas	Subaward	Dr. Wammer will oversee some of the field work and assist with the laboratory transformation experiments. She will include hire two undergraduate students to assist with field work and sample processing and to help with laboratory transformation experiments, in-state travel to collect samples, and supplies for the transformation experiments.				3		\$36,700	\$36,614	\$86
							Sub Total	\$36,700	\$36,614	\$86
Equipment, Tools, and Supplies										
	Tools and Supplies	Maintenance of mass spectrometry service contracts for use on the project	Instrument fees for analysis of ~500 samples					\$30,000	\$29,795	\$205
	Tools and Supplies	Laboratory consumables, including solvents, standards, gloves, vials, columns	For processing of field samples and conducting laboratory experiments					\$30,700	\$30,397	\$303
	Tools and Supplies	Laboratory services for mass spectrometry time	Instrument fees for analysis of ~500 samples					\$5,000	\$5,000	-

						Sub Total	\$65,700	\$65,192	\$508
Capital Expenditures									
						Sub Total	-	-	-
Acquisitions and Stewardship									
						Sub Total	-	-	-
Travel In Minnesota									
	Miles/ Meals/ Lodging	Mileage for sample collection	To get water samples from selected sites				\$2,000	\$2,000	-
	Conference Registration Miles/ Meals/ Lodging	Conference fees	To present results at local conferences				\$1,000	\$1,000	-
	3.0					Sub Total	\$3,000	\$3,000	-
Travel Outside Minnesota									
	Conference Registration Miles/ Meals/ Lodging	Conference fees	To present results at conferences	Х			\$100	\$79	\$21
	Loughig					Sub Total	\$100	\$79	\$21
Printing and Publication									
	Publication	Open access fees for peer-reviewed journals	To disseminate results to the scientific community and the general public				\$3,500	\$3,336	\$164
						Sub Total	\$3,500	\$3,336	\$164
Other Expenses									

			Sub Total	-	-	-
			Grand Total	\$508,000	\$507,048	\$952

Classified Staff or Generally Ineligible Expenses

Category/Name	Subcategory or Type	Description	Justification Ineligible Expense or Classified Staff Request
Travel Outside	Conference	Conference fees	This project is developing methodology that is of interest to the broader scientific
Minnesota	Registration		community. It is important to present the results of this project at scientific conferences
	Miles/Meals/Lodging		outside of Minnesota. This budget item reflects some of the costs associated with
			presenting the results at high profile scientific conferences.

Non ENRTF Funds

Category	Specific Source	Use	Status	\$ Amount	\$ Amount Spent	\$ Amount Remaining
State						J
In-Kind	Waived facilities and administrative costs	The University of Minnesota is waiving the income normally generated from extramural research grants that contribute Facilities and Administrative (F&A). The current full rate is 55% of direct costs.	Pending	\$322,025	-	\$322,025
			State Sub Total	\$322,025	-	\$322,025
Non-						
State						
			Non State Sub Total	-	-	-
			Funds Total	\$322,025	-	\$322,025

Attachments

Required Attachments

Visual Component

File: <u>f57b8e5a-5d4.pdf</u>

Alternate Text for Visual Component

Antibiotic residues are found in surface waters near hotspots like hospitals or animal farms. We still need to understand how far these residues can be transported, how long they remain in water, and how they vary depending on the antibiotic use at the source. As shown in the image, this project will 1) select hotspots identified from the antibiotic footprint model of Minnesota developed in the previous ENRTF funded project, 2) collect field measurements over time upstream and downstream from...

Supplemental Attachments

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

Title	File
Letter of support for Dr. Amanda Beaudoin	<u>1a49a93c-da8.pdf</u>
UMN SPA Cover Letter	<u>2ecdd62e-310.pdf</u>
Research Addendum	<u>4ca83355-84a.docx</u>
Background Check Form	<u>6a3ca846-65d.pdf</u>
Figure 1- June 2022 progress report	<u>9f7f4c99-cf5.pdf</u>
GRC 2024 Poster	<u>8ade31c0-64c.pdf</u>
Bueno Science Total Environment 2023	f9fd815d-bcb.pdf
Chesley ACS 2023	<u>b4af4f39-e5a.pdf</u>
Chesley ACS 2024	dcba2732-291.pdf
Collyard ACS 2025	<u>ac647a08-d95.pdf</u>
Clausen ACS 2023	<u>1c12afc3-732.pdf</u>

Media Links

Title	Link
An Adaptable, Interactive Computer Program to Predict the	https://conservancy.umn.edu/items/232091e0-f51f-4970-aaf4-
Fate and Transport of Antibiotics in Streams Impacted by	801630dc4058
Wastewater Effluent	

Difference between Proposal and Work Plan

Describe changes from Proposal to Work Plan Stage

The budget for this project was adjusted to the recommended funding amount. To do this, salaries for some of the project participants were reduced, estimates for the costs of instrument time and certain supplies were reduced, and funding for publication in open access journals was reduced. No changes were made to the actual project, as the reductions in requested funding do not impact the scope of the project.

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 UMN Policy on travel 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? $\ensuremath{\text{N/A}}$

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?

Does the organization have a fiscal agent for this project?

Yes, Sponsored Projects Administration

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	 Budget - Personnel Budget - Professional / Technical Contracts Budget - Capital, Equipment, Tools, and Supplies Budget - Travel and Conferences 	Need to create an official subaward for the University of St. Thomas. This agreement was described in the original budget but not officially written as a subaward. This Amendment will officially create the subaward with University of St. Thomas.	August 9, 2022	Yes	August 9, 2022
2	Amendment Request	Budget - Personnel	We are requesting that funding be shifted from the Co-project manager and geospatial modeler to the post doctoral associate and graduate student research assistant support. Due to personnel changes, the responsibilities of tasks among project staff need to be shifted, and additional funding for the postdoctoral associate is required to run the 2023 field campaign and to assist with the geospatial modeling. The graduate student researchers will help with sampling and processing of collected samples.	March 1, 2023	Yes	March 6, 2023
3	Amendment Request	 Budget Budget - Personnel Budget - Professional / Technical Contracts Budget - Capital, Equipment, Tools, and Supplies Budget - Travel and Conferences Budget - Printing and Publication 	The postdoctoral researcher is taking on responsibilities previously assigned to the co-project manager/geospatial modeler. We request funds be moved from the co-project manager/geospatial modeler to the postdoctoral researcher. We need additional laboratory supply funds to continue our field work. We request to move funds from the co-project manager/geospatial modeler line to laboratory supplies and \$3000 to the publication line to allow payment of future open access fees.	October 9, 2023	Yes	November 6, 2023

4	Completion Date	Previous Completion Date: 06/30/2024 New Completion Date: 06/30/2025; Governor Approved on 04/15/2024	As described in the Oct. supplemental update, an additional sampling season is needed to fully validate the model we are building. The first two seasons, while providing interesting data, did not provide the resolution of data needed for the microgeographic model. Personnel turnover (unexpected) has also impeded progress.	November 10, 2023	Yes	May 21, 2024
5	Amendment Request	 Budget Other Budget - Personnel Budget - Professional / Technical Contracts Budget - Capital, Equipment, Tools, and Supplies Budget - Travel and Conferences Budget - Printing and Publication 	Our project is completed and we have recently finalized the funding spent. The final budget was underspent, but we had to reallocate some of the funding across categories. This amendment reflects those changes.	September 15, 2025	Yes	October 9, 2025

Additional Status Update Reporting

Additional Status Update July 30, 2025

Date Submitted: September 15, 2025

Date Approved: October 9, 2025

Overall Update

this additional status update not needed

Activity 1

this additional status update not needed

Activity 2

this additional status update not needed

Dissemination

this additional status update not needed

Additional Status Update Reporting

Additional Status Update July 28, 2025

Date Submitted: September 15, 2025

Date Approved: October 9, 2025

Overall Update

this additional status update not needed

Activity 1

this additional status update not needed

Activity 2

this additional status update not needed

Dissemination

this additional status update not needed

Final Status Update August 14, 2025

Date Submitted: September 15, 2025

Date Approved: October 9, 2025

Overall Update

The project successfully measured the profiles of antibiotics in small streams/rivers receiving wastewater effluent. By sampling upstream, at, and downstream of the discharge, it was shown that the treatment plants were the source of antibiotics to these reaches of the streams. A literature review was conducted to compile the degradation rate constants for antibiotics by photolysis and biodegradation processes and identify knowledge gaps. Laboratory experiments were conducted to measure additional rate constants. A model was built in the python programming language that takes input from spreadsheets about the stream (depth, flow, suspended sediment concentration/composition), local conditions (wind, sunlight intensity), and chemical parameters (partitioning and rate constants) to allow simulation of antibiotic profiles in the stream and optimizing of the stream and chemical parameters to field data. The information from this project will aid in predicting the persistence of antibiotics in impacted streams so that sites where mitigation may be needed to limit spread of antibiotics or antibiotic resistance proliferation can be identified.

Activity 1

A final version of the fate and transport model was developed, tested, and made publicly available. While designed for the specific sites sampled and chemicals analyzed in Activity 2, the interface allows the user to provide information and/or modify the code so that it can be used 1) in streams of various sizes and 2) for any desired chemical. By using stream geometry measurements, measured or publicly available flow data, and partitioning and reaction rate constants, the user can predict the concentration profile of an antibiotics (or other pollutant) in a stream as a function of distance. The model allows comparison of the prediction to collected field data, and an optimization step can be used to adjust the reaction or transport parameters to fit the data. This fitting allows the introduction of a reaction parameter (e.g., biodegradation) that may vary from site to site. The program and manual were developed to be user friendly (no programing knowledge needed), use readily available software (Microsoft Excel for data input/output, python to run the code) and run on both Mac and PC platforms.

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

Activity 2

Analysis of all field data is complete. In general, the antibiotics detected at the wastewater outfalls were largely attenuated from the water column within 5 km of the discharge points. We are preparing a manuscript to present these results. We completed the indirect photolysis experiments to obtain additional rate constants with hydroxyl radical and singlet oxygen. Unfortunately, the low concentrations needed to avoid altering the microbial community led to experimental and analytical challenges that we could not overcome that prevented measurement of biodegradation rate constants of the antibiotics.

Insight was gained by use of the developed model. Simulations and fitting to our collected field data and revealed that processes for which rate constants were well-known, specifically, partitioning to sediment particles and photolysis, could not account for the attenuation rate of the antibiotics. This indicates that other processes, such as biodegradation are important. Even though we could not measure these rate constants in the lab, the model provides ranges for what these values are likely to be in the streams.

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

Dissemination

The model developed to allow simulation of antibiotic (or other organic pollutant) transport and degradation

downstream of point sources is available in the Data Repository for UMN at https://hdl.handle.net/11299/273641). A "user guide" and "how the program works" document are included to provide user instructions for use of the model and instructions on how that the code can be modified if desired. The availability of the model was advertised through social media channels.

We are preparing a manuscript that presents the profiles of antibiotics in the sampled streams. The experimental results and the model were presented at meetings of the American Chemical Society and the Association of Environmental Engineering and Science Professors.

We have kept the Minnesota One Health Antibiotic Stewardship Collaborative apprised of our efforts. We also shared the data collected with the partner wastewater treatment plants and will place it in the Data Repository for UMN when it is finalized.

Status Update June 1, 2025

Date Submitted: September 15, 2025

Date Approved: October 9, 2025

Overall Update

LCCMR staff waived the June 1, 2025, reporting status update because the appropriation ends June 30, 2025, and the reporting final status update is due August 14, 2025

Activity 1

LCCMR staff waived the June 1, 2025, reporting status update because the appropriation ends June 30, 2025, and the reporting final status update is due August 14, 2025

Activity 2

LCCMR staff waived the June 1, 2025, reporting status update because the appropriation ends June 30, 2025, and the reporting final status update is due August 14, 2025

Dissemination

LCCMR staff waived the June 1, 2025, reporting status update because the appropriation ends June 30, 2025, and the reporting final status update is due August 14, 2025

Status Update December 1, 2024

Date Submitted: December 20, 2024

Date Approved: January 13, 2025

Overall Update

Key components of the work to collect data and develop a model for the microgeographical distribution and fate of antibiotics is continuing. We have complete data collect of antibiotics in two streams and one small river that receive wastewater effluent. At all three sites, we observe an increase in antibiotics at the outfall, followed by dissipation with distance. Using the rate constants compiled from our literature review and measured in the laboratory, we are currently building a model into which hydrological site information, relevant water chemistry and weather information, and photolysis/sorption/biodegradation rate parameters are used to simulate antibiotic levels in stream/rivers that receive wastewater effluent. We are using our collected field data to calibrate the model, to assess to which parameters the model output is most sensitive, and to identify which processes are likely responsible for antibiotic dissipation in the field.

Activity 1

A model that accounts for hydrology and chemical and biological loss processes of the antibiotics is progressing. The model is being build in python and allows the user to input various hydrological, physical, and seasonal site parameters as well as information about the specific chemical of interest. Data collected from the literature review and laboratory experiments in Activity is being used for rate constants in the model. Field data from Activity 2 is being used to assess the utility of the model and which processes have the greatest effect on predicted antibiotics levels downstream of wastewater inputs. The goal is to have a user interface where once the initial parameters are set, the user can vary rate constants and hydrological/systems parameters to see which processes influence the results. Initial testing shows that flow rate is a key variable, and direct photolysis is likely the most important loss process.

Activity 2

In the spring and summer, we completed our last rounds of field sampling in Litchfield, New Prague, and Rochester. All of the antibiotic concentrations have been measured and we are working on analyzing and interpreting this data for publication and dissemination. Work on finding indirect photolysis rate constants for compounds for which the literature review did not find values is continuing. These experiments are focusing on hydroxyl radical and singlet oxygen. Similarly, we are continuing work on biodegradation rate constants in river water. The biodegradation experiments have proved challenging, and we are processing the data from the trials to evaluate if changes in the experimental design are needed. We will continue these experiments in the winter/spring, and add the relevant rate constants into the model being developed in Activity 1.

Dissemination

We are currently working on publications that describe the field data and model.

Status Update June 1, 2024

Date Submitted: June 3, 2024

Date Approved: October 14, 2024

Overall Update

We have compiled the data collected of the past two field season about antibiotic concentrations in the targeted streams. Over a span of a few kilometers, we observe that the detected antibiotics are largely removed from the water column. In parallel, we have develop the model that includes the governing equations for the potential processes that lead to attenuation of antibiotics in the streams. By using a combination of literature data and our own measurements, we are defining the parameters needed in the model. By comparing the field data with the model results, we can see we are on the right track, but that the model needs further refinement. We are continuing efforts on the model development, assessment of the model sensitive to various terms, preparation of the model for use by others, measurement of necessary rate constants, and collection and analysis of sediment samples in the three streams.

Activity 1

For the fate and transport model, we established the governing equations to explain the attenuation of antibiotics in streams. We assumed that all processes are explained by a first-order rates. The processes that affect both the total mass and concentration of antibiotics in the stream can be classified into (i) chemical and biological reactions such as photolysis and biodegradation, and (ii) the removal of antibiotics to the sediments by particle settling. We identified that the dilution or flushing process is the main factor that affects the concentration of antibiotics but leaves the mass in the stream unchanged. To get all of the data for these physical, chemical, and biological processes, we surveyed the streams using the float surface method and collected water quality parameters to accurately input these variables into the fate and transport model. We have developed an intuitive Excel sheet to import all field and laboratory data. The predicted values have been reported between 24 and 38% of the real values. We are working on a more accurate model using volumetric discharge data obtained from a gauging station located in one of the studied sites in Rochester, MN.

Activity 2

To understand the area of study and the potential point sources of antibiotics, we investigated the location, stream order, sub-catchment area, and land cover percentage of each identified water body. The streams in Litchfield, MN (Jewett Creek) and New Prague, MN (Phillips Creek) are considered low-order streams (i.e., 2nd and 1st order, respectively) with catchment areas of 65 and 24 km2, respectively, and similar land cover (~20% of developed areas and ~66% of agriculture). However, the study site in Rochester, MN, (South Fork Zumbro River) is a 4th order with a bigger catchment area of 807 km2, developed area, agriculture, and forested area of 16, 72, and 9%, respectively. The most frequently detected antibiotics in 2023 were the class of sulfonamides (i.e., sulfamethoxazole, sulphapyridine, sulfadiazine, and sulfamethazine), fluoroquinolones (i.e., ciprofloxacin, ofloxacin, and norfloxacin), and the antibiotic trimethoprim. The highest concentrations (up to 650 ng/L) were quantified in the wastewater effluent discharge. We found that these levels attenuated within a few hundred meters downstream of the discharge point. We are continuing our sampling efforts in 2024 at these sites. We are also continuing our efforts to measure photolysis and biodegradation rate constants needed for the fate and

Dissemination

Batista-Andrade, J.A.; Wammer K.H.; Singer, R., Arnold, W.A.; A microgeographic approach to the occurrence, fate, and transport of antibiotic residues in wastewater effluent-impacted streams across rural areas. Environmental Sciences: Water. Gordon Research Conference. (Holderness, NH). Poster presentation, June 23 – 28, 2024

Chelsey, A.J.; Batista-Andrade, J.A.; Arnold, W.A.; Wammer K.H. A microgeographic approach to the determination of

antibiotic concentrations, fate, and transformations in Minnesota surface waters. Spring 2024 ACS National Meeting (New Orleans, LA). Poster presentation, March 17-20, 2024.

Knellwolf, C.D.; Chelsey, A.J.; Batista-Andrade, J.A.; Arnold, W.A.; Remucal, C. K.; Wammer K.H. Photodegradation of antibiotics and parameters for environmental relevance. Spring 2024 ACS National Meeting (New Orleans, LA). Poster presentation, March 17-20, 2024.

Status Update December 1, 2023

Date Submitted: December 20, 2023

Date Approved: January 30, 2024

Overall Update

The literature review on transformation and removal processes of antibiotics in natural waters has been published. We are analyzing samples collected in 2023 and have generated maps that show antibiotic levels in the waterways at sampled locations. We have begun building a global model of antibiotic fate and transport in Minnesota's waterways. As reported before, we lost the entire field season of 2021 (due to COVID) and the site selection used in 2022 was not ideal for the stated objectives. We will need a no cost extension so that we can have a full year of sampling in 2024 and use the data collected in that year to validate the model.

Activity 1

For the fate and transport model, we were able to measure the depth, cross-sectional area, and flow velocity at each sampling site. Moving forward, we will be developing rating curves to estimate the volumetric discharge of these small streams. Additionally, the Zumbro River has a gauge monitoring station from the United States Geological Survey (USGS MN-05372995) which will provide the necessary volumetric discharge data for each sampling time of this site. These data will be important to start the effort to make a fate and transport model. In terms of physical-chemical and biological processes, we were able to estimate the partition coefficient of organic carbon to water for the antibiotics that were measured in the water samples that were collected. This information is vital to estimate the rate constant of sorption processes that potentially occur in water-sediment interface in streams. More efforts related to the fate and transport model will be made in the following months where rate constants of direct and indirect photolysis and biodegration will be incorporated in the global model.

Activity 2

We sampled three sites impacted by wastewater effluent discharge from June to October 2023 monthly. The sites were located in the Zumbro River (a repeat of 2022), one stream in Litchfield, MN, and another stream in New Prague, MN. For each sampling site, we collected one sample upstream of the wastewater effluent discharge, at the wastewater discharge site, and three samples downstream separated by 200-800 meters. In these three sites, the highest antibiotic concentrations were detected at the wastewater effluent discharge sites (i.e., up to 650 ng/L). Of the measured antibiotics, sulfonamides were the most frequently detected, followed by fluoroquinolones, macrolides, and trimethoprim. The antibiotic levels were then attenuated with distance at the downstream sites. These data will serve as the input for the fate and transport model of antibiotics in small streams, then this model can be translated to other sites that are impacted by wastewater effluent discharge.

Dissemination

We have a new peer-reviewed publication that came out in November. The citation is: Bueno I, He H, Kinsley AC, Ziemann SJ, Degn LR, Nault AJ, Beaudoin AL, Singer RS, Wammer KH, Arnold WA. Biodegradation, photolysis, and sorption of antibiotics in aquatic environments: A scoping review. Sci Total Environ. 2023 Nov 1;897:165301. doi: 10.1016/j.scitotenv.2023.165301. Epub 2023 Jul 4. PMID: 37414169. The link to the publication is: doi: 10.1016/j.scitotenv.2023.165301.

Additional Status Update Reporting

Additional Status Update October 2, 2023

Date Submitted: October 9, 2023

Date Approved: November 8, 2023

Overall Update

While we have continued to collect samples and associated antibiotic data, we have recognized that the lost field season of 2021 (due to COVID) and the site selection used in 2022 have affected our ability to draw firm conclusions. The 2023 field sites are promising, but to build and validate the microgeographic model, an additional field season in 2024 will be needed.

Activity 1

Our modeling efforts are still in the early stages due to limited field data that allows building and testing of the model. Now that we have settled on the sites that should produce the desired spatial data, we can focus our efforts on identifying antibiotic usage in these areas. Personnel turnover (largely unplanned) has affected progress be we now have people with the necessary expertise.

Activity 2

Our literature review is complete and published, and this has guided the degradation studies. We are currently working to fill the gaps in photolysis and biodegradation data, and we have determined the previously established relationships can be used for needed sorption parameters. The field data collection is going. The 2021 (COVID) and 2022 (site selection) seasons have provided data that will be useful in evaluating spatial presence of antibiotics, but the 2022 sites are inadequate for the microgeographic model. We are now working at three sites (one a repeat of 2022), that we expect to provide the necessary data. While we will be able to build a model based on these data, and additional field season will be needed for model validation and verification. This is needed for the model and mapping to be translated to other sites under a variety of flow and seasonal conditions.

Dissemination

Our literature review has been published online (Bueno et al., Biodegradation, photolysis, and sorption of antibiotics in aquatic environments: A scoping review, Science of the Total Environment, 897, 165301; https://doi.org/10.1016/j.scitotenv.2023.165301)

We also are continuing to directly engage with wastewater treatment facilities and outreach centers to communicate our findings and gather data.

Status Update June 1, 2023

Date Submitted: June 25, 2023

Date Approved: June 26, 2023

Overall Update

The literature review on transformation and removal processes of antibiotics in natural waters has been completed. The manuscript is currently under review. We sampled four sites multiple times in 2022, and captured additional sites by leveraging other sampling efforts. At all of the sites sampled, antibiotics were detected. For three of the sites, the trends were variable, which limited their utility for geospatial modeling. This was likely due to a combination of weather and the volume of wastewater flow relative to that of the river. For the 2023 sampling season, we are continuing with one of the four sites from 2022. We have identified two additional sites where we expect to see a stronger signal and greater gradient of antibiotic concentrations. The streams at these sites do not have flow gauges, so we are developing the rating curves needed to determine flow rates.

Activity 1

We are continuing the gather the necessary data to build the modeling efforts. This includes gathering the parameters and measurements to determine flows at the two new sites for 2023. Modeling efforts this spring were disrupted by two team members receiving permanent positions outside of Minnesota and a third leaving the project. Development of the model and interface will resume later this summer when a new postdoctoral researcher arrives.

In our last report, we mentioned a hospital survey that was being developed. Unfortunately, this survey was not pursued because of a loss of some of the student help and one other researcher who were going to help with this task. However, we want to emphasize that this hospital survey was never part of our original proposal; we added this survey because we thought it would add useful information. There is a chance that we will be able to pursue it in the future, but for now, this survey is not being pursued.

Activity 2

With the literature review complete, we are continuing with the photolysis, sorption, and biodegradation experiments needed to provide the reaction/removal rate data necessary for the model. For the sampling efforts, we are still analyzing the data from 2022, but some key information has been obtained. For the Cannon River, Minnesota River, and St. Croix River, antibiotics were detected, but the signal from the wastewater treatment plant was muted in most cases. We believe this is likely because the flow of the wastewater made up only a small fraction of the total flow. For the Zumbro river, a more consistent influence of the input from wastewater and loss with distance was observed. Based on this result, we are again sampling the Zumbro River, and, in consultation with MPCA, we have identified and partnered two new sites/wastewater treatment plants where the effluent makes up a substantial portion of small streams. We are also narrowing the distance over which we sample downstream, which should give us a better data on reactive versus persistent antibiotics.

Dissemination

A manuscript containing the scoping review results was submitted for publication.

Two presentations were given at the Spring 2023 American Chemical Society National Meeting

Chesley, A.J., S.L. Clausen, S.M. Berg, H. He, W.A. Arnold, K.H. Wammer. "Determination of quantum yield values of antibiotics in Minnesota waterways." Poster presentation at American Chemical Society National Meeting, Indianapolis, March 26-30, 2023.

Clausen, S.L., A.J. Chesley, S.M. Berg, H. He, W.A. Arnold, K.H. Wammer. "Antibiotic concentrations within Minnesota surface waters." Poster presentation at American Chemical Society National Meeting, Indianapolis, March 26-30, 2023.

Status Update December 1, 2022

Date Submitted: December 23, 2022

Date Approved: December 27, 2022

Overall Update

We have sampled specific sites and are currently working on laboratory analysis for quantifying antibiotic release. We have almost finished a literature review that is helping design the transformation process of antibiotic compounds. This review will be submitted for publication.

In Activity 2, we updated the sites to be sampled based on two factors. One was site access, which led to seeking similar sites where boats could be launched and picked up readily. The other was samples of opportunity. Dr. Arnold has other projects conducting field sampling of rivers, and we leveraged those events to collect additional samples for this project. Based on the measurements obtained from the 2022 data, we expect a more focused campaign in Summer 2023.

Activity 1

While laboratory analysis of samples is ongoing, we are using the literature review to help inform our modeling efforts. We are collecting data on water management practices for points of interest to help inform the user-friendly interface we will be creating.

Point sources of interest have been chosen and sampled for 2022. Further investigation into additional sampling sites for 2023 is currently being assessed.

Waste management practices are still being collected from all point sources and will continue to be collected until field sampling is completed. Further sampling sites for 2023 are being assessed which will add to the data collection of waste management practices from point sources of interest. The hospital survey is developed and undergoing edits currently with the intention of being used in early 2023. The majority of hospitals are connected to the community wastewater treatment plant systems, and thus the hospitals are not 'point sources' per se as the WWTP are. The hospital survey was going to be focused on finding out specific details about potential pre-treatments the hospitals do before the waste gets released into the WWTP, plus other information.

Activity 2

We chose seven rivers and one lake that receive wastewater effluent for our 2022 sampling efforts. Cannon River, Crow River, Lake Winona, Minnesota River, Red River, St. Louis River, St. Croix River, and Zumbro River. The Cannon River, Minnesota River, and St. Croix River were sampled monthly from April through October. Lake Winona and the Zumbro river were sampled three times spanning these months. The Crow River was sampled twice, and both the Red and St. Louis rivers were sampled once. Upstream of the discharge point, near the discharge point, and 3-5 samples downstream of the discharge point were collected for each site. In lab, the samples were split into triplicates, and antibiotics were extracted and the cartridges frozen. Once these extracts are reconstituted into a suitable matrix, the antibiotics will be quantified with liquid chromatography-mass spectrometry. A literature review compiling information about antibiotic photolysis, biodegradation, and sorption is in preparation. Based on identified knowledge gaps, studies of photolysis of compounds with inadequate literature data have begun. Sorption and biodegradation experiments will commence in 2023.

Dissemination

A manuscript containing the scoping review results will be submitted for publication.

Status Update June 1, 2022

Date Submitted: August 9, 2022

Date Approved: August 9, 2022

Overall Update

We have selected specific sampling sites associated with individual point sources and have started field sampling. For those sites, we have also started data collection on waste management interventions. We have almost finished conducting a scoping literature review on the persistence of antibiotics in the environment which we will submit for publication in the next two months. This literature review will help us design several laboratory experiments to obtain data on transformation processes for antibiotic compounds of interest.

Activity 1

We have selected most of the specific point sources to investigate the release and dissemination of antibiotics temporally and spatially, and have started field sample collection (as of May 1st, 2022). Thus far, three specific waterbodies will be sampled monthly (the Minnesota river, the St. Croix river, and Lake Byllesby, and three additional sites quarterly (the Crow river, and two lakes in Alexandria and Rochester respectively). All these sites (monthly and quarterly sites) have wastewater treatment plant influence. We are currently assessing the feasibility of additional sampling sites.

We are using a new field app from ESRI called ArcGIS Field Maps which allows the collection of data with mobile devices in the field including distance measurement between sampling points, and to visualize the data collection in real-time in ArcGIS online (Fig. 1).

We have also collected data on waste management practices for some of the point sources of interest. Specifically, information about wastewater treatment for the plants of interest and hospital waste management have been retrieved through the Minnesota Pollution Control Agency. We are also developing a survey to be sent to the main hospitals in the state to understand pre-treatment of hospital waste.

Activity 2

We have conducted a scoping review of the literature to understand the persistence of antibiotics in the natural environment. We have followed rigorous and reproducible methodology for this literature review, which included the publication of a protocol prior to starting the review. The protocol was published online at the University of Minnesota Libraries Digital Conservancy (https://conservancy.umn.edu/handle/11299/221594) and at the systematic reviews for animals & food website (https://www.syreaf.org/protocol/). We are now in the process of summarizing the data from all the studies included in the review which comprises the following transformation processes: sorption, biodegradation, and photolysis. We will be submitting a manuscript to a peer-reviewed journal with the results of this review by July 2022. Through this review we are also identifying data gaps about the transformation processes, which we will use to conduct laboratory experiments starting during the summer of 2022. We have also started our field measurements of antibiotic levels (as of May 1st, 2022) and will continue that work in the coming months.

Dissemination

To date, we have published a protocol for our scoping review of the literature (Activity 2) online at the University of Minnesota Libraries Digital Conservancy (https://conservancy.umn.edu/handle/11299/221594) and at the systematic reviews for animals & food website (https://www.syreaf.org/protocol/). We have also presented preliminary results for the scoping literature review through a scientific poster at the American Chemical Society Spring Conference 2022-

Bonding Through Chemistry in San Diego (March 20-24, 2022). Finally, we will be submitting a manuscript containing the scoping review results for peer-review publication during the Summer of 2022.					