

**Environment and Natural Resources Trust Fund
2020 Request for Proposals (RFP)**

Project Title:

ENRTF ID: 228-F

Lowering Nitrogen Requirements for Agricultural Crops

Category: F. Methods to Protect, Restore, and Enhance Land, Water, and Habitat

Sub-Category:

Total Project Budget: \$ 271,000

Proposed Project Time Period for the Funding Requested: June 30, 2023 (3 yrs)

Summary:

We will bestow upon plants the ability to obtain nitrogen through sustainable biological processes by improving associations between plants and nitrogen-fixing microbes that live with the above-ground plant tissues.

Name: Brett Barney

Sponsoring Organization: U of MN

Job Title: Professor

Department: Bioproducts and Biosystems Engineering

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Location:

Region: Statewide

County Name: Statewide

City / Township:

Alternate Text for Visual:

Graphic illustrating the concept of nitrogen-fixing endophyte enhancement

_____ Funding Priorities	_____ Multiple Benefits	_____ Outcomes	_____ Knowledge Base
_____ Extent of Impact	_____ Innovation	_____ Scientific/Tech Basis	_____ Urgency
_____ Capacity Readiness	_____ Leverage	_____ TOTAL	_____ %



PROJECT TITLE: Lowering Nitrogen Requirements for Agricultural Crops

I. PROJECT STATEMENT

CONCEPT. The goal of this project is to bestow upon plants the ability to obtain nitrogen through sustainable biological processes by improving associations between plants and targeted nitrogen-fixing bacteria that live within the above-ground tissues of the plant. Our laboratory has made substantial progress in recent years toward attaining this goal that could provide revolutionary solutions to feeding a growing population while minimizing losses of nitrogen applied through conventional fertilizers. We are currently addressing the final remaining technological hurdle toward realizing the potential of this technology.

BACKGROUND. Prior to the introduction of industrially derived nitrogen fertilizers, farmers understood that rotating crops such as soybeans, alfalfa and clover on alternating years resulted in improved yields of crops such as wheat or corn the following year. Decades of research have taught us that the reason why certain crops improve soils is due to a symbiotic relationship between these plants and specific bacteria that live within or around the root systems of these plants. These symbiotic relationships have several benefits. The plant is fed a continuous supply of nitrogen from the bacterium as long as it supplies food (such as sugar) to the bacterium, which minimizes or eliminates the need for direct human intervention via nitrogen fertilizer application. However, this root-nodule based symbiosis generally occurs only within the roots of specific crops, while other key crops (corn and wheat) and native plants important to Minnesota still require external application of nitrogen to achieve desired yields.

RELATED ISSUES. Agriculture requires substantial resources to produce the crops that meet the needs of our modern society. Nitrogen is a primary component of fertilizers, and while industrial processes have enabled decades of increased agricultural production, this comes at both an environmental and an economic cost;

- **Excessive nitrogen application results in high runoff and downstream water contamination** leading to eutrophication and high nitrate levels in wells found across Minnesota, as has been highlighted in reports by the Minnesota Pollution Control Agency.
- Industrial nitrogen fixation is the main route to the production of ammonia. It consumes 3-5% of natural gas production and **requires about 1-2% of all worldwide energy consumption**, releasing massive amounts of CO₂ into the atmosphere due to the dependence of this process on petroleum derived fuels.
- Transportation of nitrogen from industrial production sites to geographically dispersed farms adds costs, and **storage of ammonia represents a danger to farmers, their communities and the environment**, as illustrated by recent accidents resulting in evacuations and hospitalizations across Minnesota.

BENEFITS. Complementing or replacing industrial nitrogen production with natural nitrogen-fixation processes will lower emissions from industrial processes, providing a sustainable and locally produced commodity product with commercial value. This will establish Minnesota as a global leader in sustainable agriculture. In addition, because the nitrogen that is produced is released directly within the plant, there will be minimal runoff of excess nitrogen into lakes and streams. Localization of the bacterium within the plant also provides the bacterium with direct access to sugars produced by the plant to fuel this biologically driven process. **Successful examples of these above-ground endophyte relationships are already found in commodity crops such as sugarcane and rice.**

LONG-TERM GOALS. Our project goal is to overcome barriers to the efficient production of inexpensive nitrogen through the natural biological process of nitrogen-fixation. Our recent research efforts have already resulted in nitrogen-fixing bacteria that produce high yields of nitrogen. The next step to our long-term goals is to better understand how natural bacteria that reside within the plant tissues (endophytes) establish themselves and serve as a valuable symbiont without eliciting a disease response (pathogenicity). **Once this task is accomplished and merged with our existing technologies, we will be able to pursue longer-term goals of replacing industrial fertilizers and requirements for nitrogen application altogether.** This current project allows us to approach this initial task by identifying natural strains native to Minnesota that are able to function as nitrogen-fixing endophytes for the development of optimal biofertilizer seed cultures.



II. PROJECT ACTIVITIES AND OUTCOMES

Activity 1: Boosting Nitrogen Fixation through Minnesota Endophytes

Budget: \$271,000

This activity will focus on determining methods to enhance the natural processes associated with beneficial endophyte bacteria that colonize various plants, and in many cases, enhance the general health of the plant through these associations. With a focus on symbiotic bacteria native to Minnesota, we aim to obtain a large catalog of bacteria, with the goal of studying the potential to expand their distribution, developing methods and practices that supplement or replace the requirement for industrially-provided fertilizers. By producing the nitrogen directly within the plant, we will eliminate the potential migration of this nitrogen into our soils and groundwater, lakes and streams, and combat eutrophication in these waters.

Outcome	Completion Date
1. Develop a collection (~100 strains) of natural nitrogen-fixing bacterial endophytes from various native plants and commodity crops across Minnesota.	Dec 1 st , 2020
2. Confirm the ability to reintroduce natural nitrogen-fixing bacteria as endophytes into selected target plants through established delivery techniques.	June 1 st , 2020
3. Down-select several target nitrogen-fixing bacteria with the highest potential to displace the need for externally provided industrial fertilizers and develop natural consortia for application by farmers and organic gardeners.	August 15 th , 2020
4. Sequence strains of several down-selected target nitrogen-fixing bacteria (~10 strains) to better understand optimal features of these strains, and assure that wide-scale introduction would not result in any detrimental effects (harm to the plants or the environment).	Jan 31 st , 2021

III. PROJECT PARTNERS:

The research team includes Professor Brett Barney from the Department of Bioproducts and Biosystems Engineering and the Biotechnology Institute at the University of Minnesota, who will oversee the project. Professor Barney is an expert in the field of biological nitrogen fixation and has studied this process for 20 years. Neil Olszewski from the Department of Plant and Microbial Biology is a plant expert with more than 30 years of research experience in the field, and has also recently developed a plant line that can serve as an important screen for nitrogen-producing endophytes, which would be ideal for this project.

IV. LONG-TERM IMPLEMENTATION AND FUNDING:

Minnesota is a major agricultural state and requires long-term solutions to environmental issues associated with farming. ***Sustainable production of internally-produced nitrogen with minimal runoff potential through a biologically derived process would build the local economy and save farmers money while lowering the impact of farming on water quality.*** Success from this project would be truly transformative, replacing an antiquated process that has been responsible for enormous quantities of carbon added to the atmosphere, and damage to our lakes and streams related to nitrogen over-application and severe weather events. Previous funding of this project through the MnDRIVE program at the UMN has already overcome a major hurdle toward the production of biologically derived nitrogen. Successful demonstration of the goals set here would draw private support.

V. TIME LINE REQUIREMENTS:

This project has a target for completion of 3 years. As preliminary work, a small number of bacteria that behave as model endophytes have been collected along with additional bacteria from Minnesota. Preliminary studies are underway, but would be substantially expanded once the project is funded. Further support would be sought through additional funding sources based on the overall success of the project.

Attachment A: Project Budget Spreadsheet
 Environment and Natural Resources Trust Fund
 M.L. 2020 Budget Spreadsheet



Legal Citation:

Project Manager: Brett Barney

Project Title: Lowering Nitrogen Requirements for Agricultural Crops

Organization: University of Minnesota

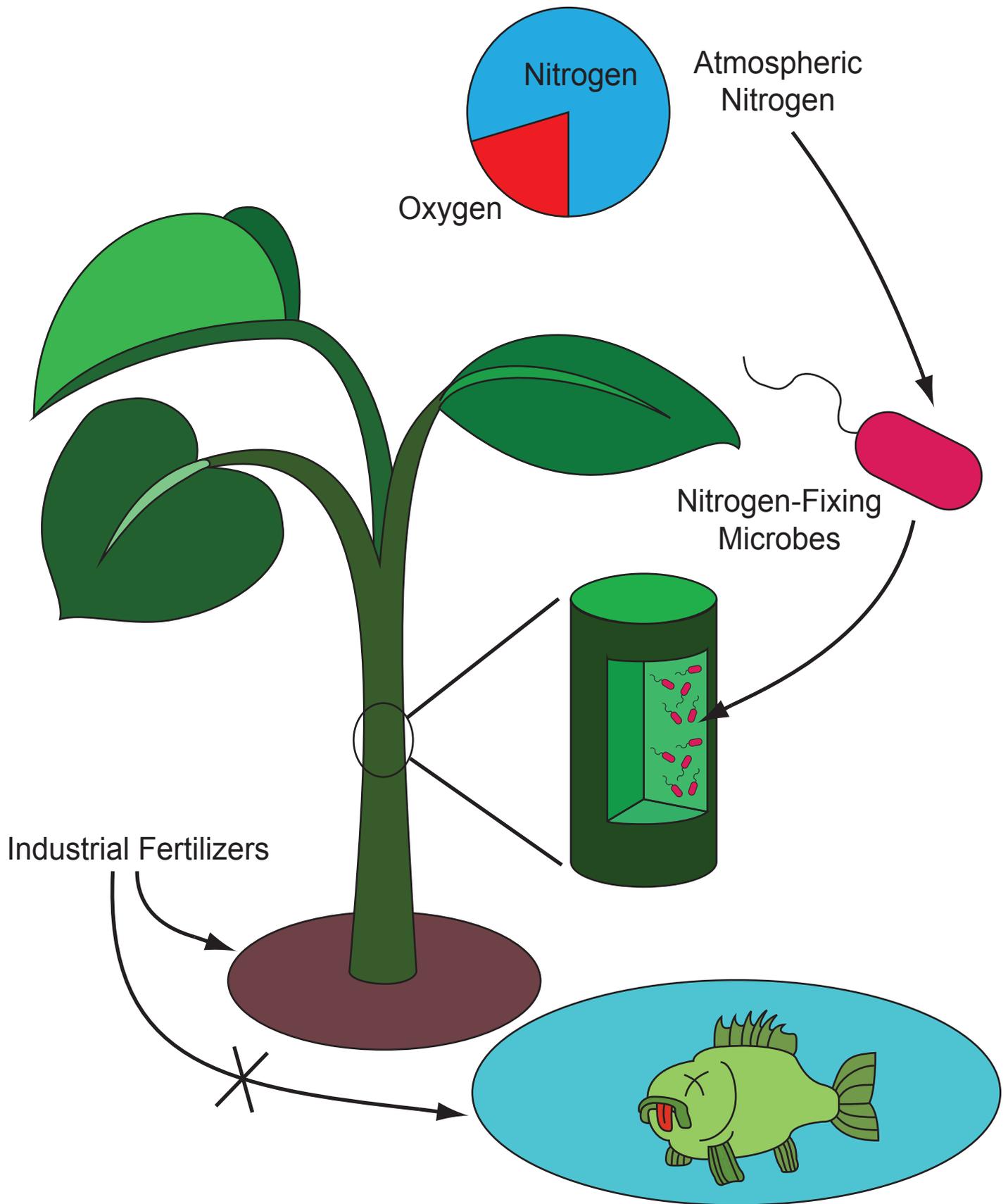
Project Budget: \$271,000

Project Length and Completion Date: 3 years - 06/30/2023

Today's Date: 4/11/2019

ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET		Budget	Amount Spent	Balance
BUDGET ITEM				
Personnel (Wages and Benefits)		\$ 223,000	\$ -	\$ 223,000
Brett Barney, Project Manager (73.5% salary, 26.5% benefits), Associate Professor, 9 Month Appointment, Summer Salary; 4% FTE (0.5 month) for 3 years, \$24,000				
1 Graduate Research Assistant, UMN (Twin Cities), Laboratory Experiment Data Analysis, supervised by Barney and Olszewski (65% salary/35% fringe); 50% FTE for 3 years, \$148,000				
2 Undergraduate Technicians, Laboratory and Field Data Collection (100% salary, 0% benefits); 10% FTE for 3 years (generally rotating 1 year appointments), \$51,000				
Professional/Technical/Service Contracts				
DNA Sequencing Analysis, Sequencing of Endophyte Microbes for analysis and Characterization, Locally sourced through either the University of Minnesota Sequencing Center or Local Companies. (10 Strains at ~\$1,000 per strain)		\$ 10,000	\$ -	\$ 10,000
Equipment/Tools/Supplies				
Laboratory Supplies: General Laboratory Chemicals for Media and Reagents (\$200 per month) and Kits for Performing Routine Molecular Biology (\$400 per kit), Analytical Reagents, DNA Synthesis of Primers (\$200 per month), Liquid Nitrogen for Strain Storage (\$400 per year).		\$ 36,000	\$ -	\$ 36,000
Capital Expenditures Over \$5,000				
		\$ -	\$ -	\$ -
Fee Title Acquisition				
		\$ -	\$ -	\$ -
Easement Acquisition				
		\$ -	\$ -	\$ -
Professional Services for Acquisition				
		\$ -	\$ -	\$ -
Printing				
		\$ -	\$ -	\$ -
Travel expenses in Minnesota				
Travel across state of Minnesota for bacterial strain collection by project participants (4 trips per year at ~\$160 per trip), to be reimbursed by the University Compensation Plan.		\$ 2,000	\$ -	\$ 2,000
Other				
		\$ -	\$ -	\$ -
COLUMN TOTAL		\$ 271,000	\$ -	\$ 271,000
SOURCE AND USE OF OTHER FUNDS CONTRIBUTED TO THE PROJECT				
	Status (secured or pending)	Budget	Spent	Balance
Non-State:		\$ -	\$ -	\$ -
State:		\$ -	\$ -	\$ -
In kind:		\$ 121,000	\$ -	\$ 121,000
Other ENRTF APPROPRIATIONS AWARDED IN THE LAST SIX YEARS				
	Amount legally obligated but not yet spent	Budget	Spent	Balance
		\$ -	\$ -	\$ -

Lowering Nitrogen Requirements for Agricultural Crops





Environment and Natural Resources Trust Fund (ENRTF)
2020 Project Manager Qualifications
Project Title: Lowering Nitrogen Requirements for Agricultural Crops

Project Manager Qualifications

Brett Barney, Project Manager

Education:

- Ph.D. Biochemistry, Arizona State University, 2003
- B.S. Professional Chemistry, Utah State University 1993

Work and Research Experience:

- 2015 – Present Associate Professor, Bioproducts and Biosystems Engineering (UMN)
- 2017 – Present Director of Graduate Studies, BBSEM Program (UMN)
- 2015 – Present Director of Graduate Studies, Microbial Engineering Program (UMN)
- 2010 – Present Faculty Member, BioTechnology Institute and Microbial and Plant Genomics Institute (UMN)
- 2009 – 2015 Assistant Professor, Bioproducts and Biosystems Engineering (UMN)
- 2003 – 2009 Research Assistant Professor and USDA Postdoctoral Fellow (USU)
- 1999 – 2003 Research Assistant and NSF Fellow, Department of Chemistry and Biochemistry (ASU)
- 1993 – 1999 Fiber Laboratory Manager, Research Chemist, Senior Laboratory Technician and Associate Chemist, Fresenius Medical Care, Ogden, Utah
- 1991 – 1993 Research Technician, Utah Water Research Laboratory (USU)

Neil Olszewski, co-Project Manager, Professor, Department of Plant and Microbial Biology (UMN)

Neil brings expertise in plant management that are important components of agriculture in Minnesota.

Dr. Barney's laboratory is focused on biological nitrogen fixation for minimizing costs and environmental impacts associated with biofuels and agriculture. Dr. Barney has 30 years of experience in both basic and applied research in both academia and industry, including experience managing projects and laboratories in a range of settings. Previous research funding has come from the National Science Foundation (NSF), the United States Department of Agriculture (USDA), the United States Department of Energy (DOE), the Defense Advanced Research Projects Agency (DARPA), Minnesota's Discover, Research and Innovation Economy (MnDRIVE) and the Initiative for Renewable Energy and the Environment (IREE).

The Barney laboratory is housed in the Cargill building for Microbial and Plant Genomics at the University of Minnesota. The Cargill building was designed with the intention to promote interdisciplinary collaborations and provide a shared lab space for each floor, which facilitates flexible group sizes. This large laboratory space is designed around a shared communal format, with various rooms available for utilization for specific experiments. The laboratory contains the primary equipment to perform this research project, including facilities to cultivate various bacteria, autoclaves, analytical instrumentation for analysis (gas chromatography, spectrophotometers, and balances), thermocyclers for PCR reactions, centrifuges, electrophoresis equipment and various incubators. Additional facilities include the Biotechnology Resource Center, the Genomic Sequencing Center and a broad range of additional analytical laboratories which are available as pay services.

Organization Description

Dr. Brett Barney (PI) has been a professor with the Department of Bioproducts and Biosystems Engineering at the University of Minnesota since 2009. The Bioproducts and Biosystems Engineering Department serves as a core department combining Agricultural Engineering, Biological Engineering and Environmental and Ecological Engineering. The University of Minnesota provides a range of facilities and sufficient laboratory space to perform each of the activities described in this proposal. Additionally, controlled environments including greenhouse space sufficient for this work is conveniently located next door to Dr. Barney's laboratory space. UMN Sponsored Projects Administration (SPA) is the entity authorized by the Board of Regents to manage project agreements with the LCCMR program.