**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**

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| **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.

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| **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 1st, 2020 |
| 2. Confirm the ability to reintroduce natural nitrogen-fixing bacteria as endophytes into selected target plants through established delivery techniques. | June 1st, 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 15th, 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 31st, 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.