

**Environment and Natural Resources Trust Fund  
2015 Request for Proposals (RFP)**

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**Project Title:**

**ENRTF ID: 034-B**

A Novel Biofilm Technology for Water Nutrient Removal

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**Category:** B. Water Resources

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**Total Project Budget:** \$ 281,270

**Proposed Project Time Period for the Funding Requested:** 3 years, July 2015 - June 2018

**Summary:**

This project will develop a novel simulated lichen biofilm system to remove pollutants and possibly recycle nutrients from storm water runoff as well as polluted lakes, ponds, and lagoons.

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**Sponsoring Organization:** U of MN

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**Location**

**Region:** Statewide

**County Name:** Statewide

**City / Township:**

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**Alternate Text for Visual:**

A brief description of the proposed technology

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



**Project Title: A novel biofilm technology for water nutrient removal**

**I. Project statement**

Non-point source pollution is the leading cause of water quality impairment in Minnesota. Excessive loading of nutrient from stormwater runoff at urban and agricultural landscapes results in eutrophication and encourages the growth of invasive species. We propose a novel biofilm technology to remove and possibly recover nutrients such as nitrogen and phosphorus from water and the process will also remove pollutants such as heavy metal ions (lead, zinc, copper, arsenic etc.) and pesticides. This new technology will be developed based on the concept of a “simulated lichen biofilm”, mimicking the natural symbiotic lichen ecosystem, for efficiently removing and recovering nutrients and pollutants, by introducing a supporting matrix, binding filamentous fungal strains and microalgae. The project will develop this technology through the lab experiments and evaluate its effectiveness on the Sarita wetland close to UMN St Paul campus.

Conventional practices use sedimentation methods to remove suspended pollutants; whereas dissolved nutrients (phosphorous and nitrogen), organics, pesticides and heavy metals (lead, zinc, copper, arsenic) require more complicated approaches to remove them from the runoff. Lichen, a natural ecosystem with phototrophic algae and heterotrophic fungi symbiotically growing on the solid surface of rock or roof, is not readily applied in engineering field due to their low growth rate. A concept of “simulated lichen system” is recently created by our UMN research group that we can select different desired microalgae and fungal combinations that will be growing on the surface of some specific polymers to form the biofilm. We are proposing to study this concept and apply to remove and even possibly recover nutrients from water. Microalgae are naturally growing on the surface of the nutrient-rich water; however, biological treatment of polluted waters using microalgae is limited by problems associated with the settling and separation of algae downstream of the treatment site. The proposed methodology using bioaugment filamentous fungi in lichen biofilms overcomes this limitation, by efficiently retaining algae and recovering the nutrients and heavy metals with possible recycling of useful nutrients. The simulate lichen biofilm will also grow much faster than aquatic plants in removing nutrients.

Our preliminary research has shown that filamentous fungi and fresh water microalgae can naturally be grown attached on some specific bio-based polymers to form the “simulated lichen biofilm”. Both types of cells can accumulate phosphorous, nitrogen and toxic heavy metals, and pollutants will be removed by removing this biomass from water. The development of a stable lichen biofilm will have multiple benefits over the current available technologies. This composite will have the capacity to remove the pollutants in a wide concentration range, and to possibly recycle valuable and non-renewable nitrogen and phosphorous. The technology can be incorporated into current practices for protecting our water bodies. A successful on-site demonstration of the prototype would transform the storm water runoff treatment, in polluted lakes, pond, and lagoon.

**II. PROJECT ACTIVITIES AND OUTCOMES**

**Activity 1: Develop the biofilm technology through lab experiments**

**Budget: \$141,645**

The objective of this activity will include laboratory-scale studies on methods and conditions to form the simulated lichen biofilm and process optimization for nutrients and pollutants removal and recuperation. The chemical and physicochemical characteristics of storm water runoff at Sarita wetland close to UMN St Paul campus as well as the manure lagoon storage water at UMN-Waseca center will be done. The composition of the storm water will be quantified and a suitable lichen system will be established for the specific runoff, after passing through the standard sand fillings or infiltration systems for total suspended solids (TSS) removal. The Laboratory-scale prototypes will be constructed for studying the recovery capacity for nutrients (phosphorous and nitrogen) and heavy metals, with the influent and effluent characteristics analyzed. The lab-scale evaluation of the operating parameters will be carried out using a robust statistical methodology and mathematical models will be developed for the system under study.



<b>Outcome</b>	<b>Completion Date</b>
1. Characterization of runoff streams at Sarita wetland	Sep 2015
2. Reactor configuration coupled with TSS removal system/bio-retention system	March 2016
3. Laboratory systems for the evaluation	Feb 2017

**Activity 2: On-site evaluation of the biofilm technology**

**Budget: \$94,228**

The objective of this activity is the prototype development and onsite evaluation. The Sarita wetland in University of Minnesota will be selected as a model system for tracking patterns in field parameters for the design of prototypes and implementing the scaled-up model for evaluating the effectiveness of lichen composites. A “floating island” system will be designed and constructed to use fungal and microalgae species instead of native plants. The treatment capacities of the systems will be estimated and effects of system design parameters will be evaluated. The kinetic and model parameters evaluated from the previous bench-top studies will be used to scale-up the process.

<b>Outcome</b>	<b>Completion Date</b>
1. Floating island design and construction	April 2017
2. Data collection and evaluation at Sarita	May 2018

**Activity 3: Develop a universal application strategy and cost estimation**

**Budget: \$45,398**

The objective of this activity is to compare the runoff samples from different geographic regions (urban and rural) to determine an effective treatment strategy for various runoffs. The sites for samplings will include two urban runoff sites at Twin Cities Metro, the swine manure wastewater lagoon at Waseca, a heavily polluted lake at Albert Lee and a possible site at Northern MN. Cost estimation will be developed onto a spreadsheet calculation model in order to determine the capital investment, the useful life; and operations and maintenance costs of the system, considering size the pollution level of different water body.

<b>Outcome</b>	<b>Completion Date</b>
1. Water sample collection and analysis for the application	May, 2018
2. Cost estimation	June, 2018

**III. PROJECT STRATEGY**

**A. Project Team/Partners**

The team includes Professor Bo Hu, Dr. Aravindan Rajendran, Professor Bruce Wilson and his postdoc researcher, all from the Department of Bioproducts and Biosystems Engineering, University of Minnesota. Professor Hu is specialized in bioprocess development, and has extensively studied the fungal/microalgae pelletization and co-culture. Aravindan Rajendran, a post doc researcher, specialized in bioprocessing technologies and bioprocess modeling, will execute the research activities and provide technical expertise. Professor Bruce Wilson has extensive experience in reducing nutrient loading from agricultural and urban watersheds.

**B. Timeline Requirements**

The project will be completed in 3 years, with two years for lab-scale study and the remaining one year for on-site implementation and evaluation as well as cost estimation.

**C. Project Impact and Long-Term Strategy**

The outcomes of the project will provide a sustainable solution to capture and recycle the reusable resources from runoffs by protecting rivers, lakes, and vital landscape along with protecting terrestrial and aquatic life. The long term strategy is to transform the storm water runoff treatment facility for revenue generation by recovering and recycling the resources, and also to incorporate social responsibility for completely eliminating the runoff pollution, decimating the adverse effects of storm water on water resources.

## 2015 Detailed Project Budget

**Project Title:** [A novel biofilm technology for water nutrient removal ]

### IV. TOTAL ENRTF REQUEST BUDGET [3] years

<u>BUDGET ITEM</u>	<u>AMOUNT</u>
<b>Personnel:</b>	
Project director, Bo Hu will be paid to manage the project, design the experiments and write the project report. The payment will cover his one month summer salary and fringe benefits. 75.32% of payment will be the salary and 24.68% will be the fringe benefits.	\$ 37,067
Postdoc researcher, Dr. Aravidan Rajendran will be paid to execute the activities and provide technical expertise. 100% of time employment will be covered for this position by the project for the first two years and then 50% of the time on the third project year. The budget includes 82.30% for the salary and 17.70% for the fringe benefits.	\$ 124,438
Dr. Bruce Wilson's postdoc research will be paid 100% for the third year to assist with the onsite evaluation. The budget includes 82.30% for the salary and 17.70% for the fringe benefits.	\$ 54,675
<b>Equipment/Tools/Supplies:</b>	
Large scale "floating islands" will be constructed containing the supporting structure, the polymer matrix and necessary fungal attachment. The large scale fungal attachment will need to be cultured at Bioconversion Resource Center at UMN using their pilot scale equipment. The cost includes the material and labor for constructing the floating island as well as the equipment rental fees to be paid to the Bioresource Center	\$ 5,000
A HPLC will be purchased to analyze the intermediate signal chemicals generated by the microalgae/fungal ecosystem as well as the water quality parameters such as nitrogen, phosphorus and heavy metals.	\$ 40,000
Supply and chemicals to be used to work on the experiments in the lab and field	\$ 18,545
<b>Travel:</b> Travel to the agricultural lagoon at Waseca MN to obtain wastewaters. Two travels are planned per each project year and \$250 is budgeted per travel for the first year with 3% increase for the following years.	\$ 1,545
<b>Additional Budget Items:</b>	\$ -
<b>TOTAL ENVIRONMENT AND NATURAL RESOURCES TRUST FUND \$ REQUEST =</b>	<b>\$ 281,270</b>

### V. OTHER FUNDS

<u>SOURCE OF FUNDS</u>	<u>AMOUNT</u>	<u>Status</u>
<b>Other Non-State \$ To Be Applied To Project During Project Period:</b>	\$ -	
<b>Other State \$ To Be Applied To Project During Project Period:</b>	\$ -	
<b>In-kind Services During Project Period: Unrecovered F&amp;A at 52% MTDC</b>	\$ 125,461	Secured
<b>Funding History:</b>	\$ -	
<b>Remaining \$ From Current ENRTF Appropriation:</b>	\$ -	

# A Novel Biofilm Technology for Water Nutrient Removal

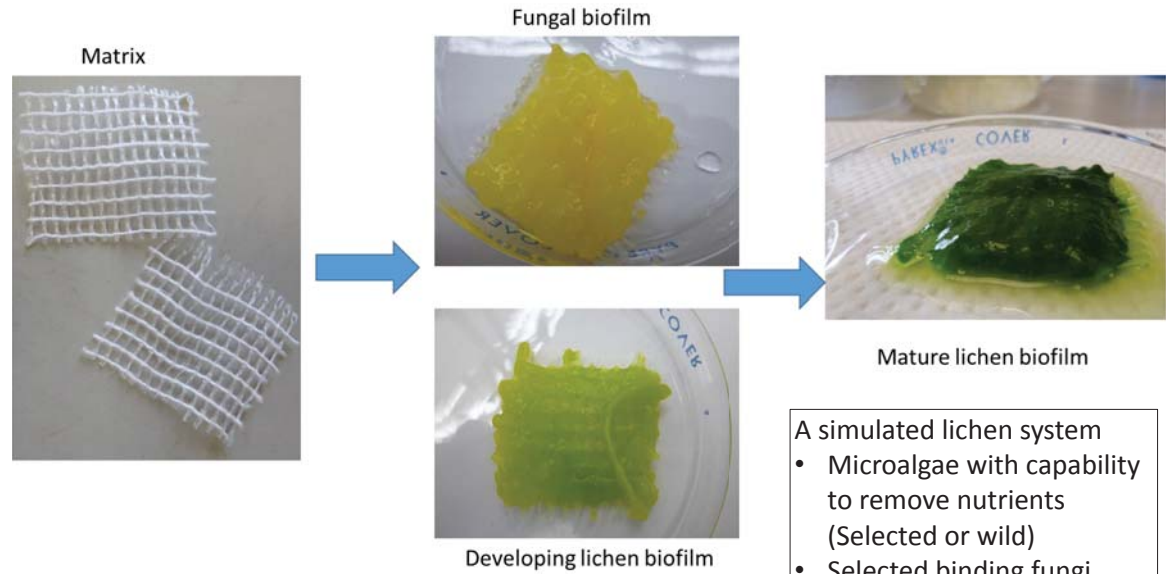
Bo Hu, Aravindan Rajendran and Bruce Wilson  
University of Minnesota

Environment and Natural Resources Trust Fund

**Project summary** - This project proposes to develop a novel simulated lichen biofilm system to remove and recycle nutrients from storm water runoff as well as polluted lakes, ponds, and lagoons.

A natural lichen system:

- Phototrophic microalgae
- Fungi
- Supporting surface



A simulated lichen system

- Microalgae with capability to remove nutrients (Selected or wild)
- Selected binding fungi
- Selected polymer easy to be harvested and reused

Proposed Process Flowchart

Stormwater runoff

Nutrients (phosphorous and nitrogen),  
organics, pesticides, heavy metals  
(lead, zinc, copper, arsenic)

Lichen Biofilm system

Treated stormwater

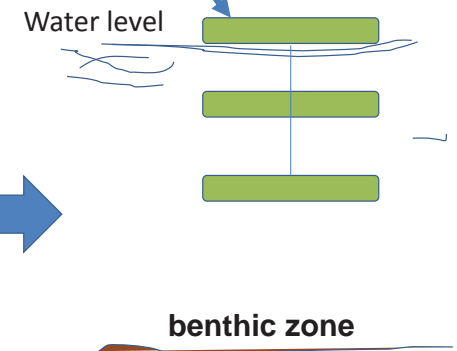
Biomass harvest and nutrient recycle

A floating island with simulated lichen systems instead of plants

- Grow much faster
- Can use native microalgae
- No need for soil and easy to harvest



Current floating island with plants



Proposed floating island with simulated lichen system

### **Project Manager Qualifications**

The research team will include Dr. Bo Hu, Dr. Aravindan Rajendran, and Dr. Bruce Wilson and his postdoc researcher, all from the Department of Bioproducts and Biosystems Engineering, University of Minnesota. Dr. Aravindan Rajendran, a post doc researcher, specialized in bioprocessing technologies and bioprocess modeling, will execute the research activities and provide technical expertise. Professor Bruce Wilson has extensive experience in reducing nutrient loading from agricultural and urban watersheds and his postdoc researcher will assist the onsite evaluation.

With regard to technical expertise, **Dr. Bo Hu** is an Assistant Professor at the Department of Bioproducts and Biosystems Engineering of UMN. He is also a joint faculty member of Biotechnology Institute of UMN. With over 10 years of active research experience specifically in biomass utilization, fermentative conversion, and molecular biology, he has led projects on microbial oil production from waste materials via mixotrophic microalgae and oleaginous fungal fermentation, and projects to develop the modified anaerobic digestion system for biohydrogen production and its microbial community change by using 16s rDNA based microbial analysis. Hu's team at UMN has set up several standard procedures such as 16s rDNA fingerprint screening for microbial species in the wastewater treatment facilities, ITS sequences to identify oleaginous fungal species; and several conversion platforms such as pelletized fungal fermentation, solid and hemi- SolidSF to accumulate oil from lignocellulosic materials. His research ideas have been funded by many programs, especially local funding agencies such as MN Pork Board, IOWA Pork Board, MN Rapid Agricultural Response Program, etc. to tackle regional issues.

Dr. Hu's lab is located at BAE 320B, adjacent to Dr. Hu's office. The lab space is around 1000 sqft and it is equipped with two laminar flow hoods and one clean bench. The lab has all the necessary equipment and facilities for this project, including a refrigerated shaker, two open air shakers, one incubation shaker, two incubators, one fermentation bioreactor, GC-FID-TCD, PCR thermal cycler, several electrophoresis, centrifuge, and ovens. The research group can also utilize facilities and equipment at the **Biotechnology Resource Center (BRC)**, on a pay-per-sample base. BRC is a 4,000 square-foot laboratory/pilot plant facility with state-of-the-art equipment for research and development in fermentation, animal cell culture technology, molecular biology, protein expression, and separation of a wide range of biological molecules.

### **Organization Description**

Dr. Bo Hu joined the faculty at Department of Bioproducts and Biosystems Engineering of UMN in August 2009. As the core department of UMN to tackle Agricultural engineering and environmental engineering issues, Bioproducts and Biosystems Engineering Department has very dynamic research activities and numerous excellent scientific researchers have received grant supports from LCCMR program. The collaborative partner Sara Heger works with the Onsite Sewage Treatment Program at the Water Resource Center of UMN. The program seeks to protect public health and the environment by improving wastewater treatment through research-based education and outreach for homeowners, small communities, professionals and policy-makers. UMN Sponsored Projects Administration (SPA) will be the entity authorized by the Board of Regents to manage the project agreements with LCCMR program.