

**Environment and Natural Resources Trust Fund  
2010 Request for Proposals (RFP)**

**LCCMR ID: 016-A2**

**Project Title:**

Use of Woodchip Bioreactors to Maintain Minnesota's Resources

**LCCMR 2010 Funding Priority:**

A. Water Resources

**Total Project Budget: \$** \$244,826

**Proposed Project Time Period for the Funding Requested:** 2 years, 2010 - 2012

**Other Non-State Funds: \$** \$0

**Summary:**

The overall goals of the research proposed here are to develop and utilize a microbial-based woodchip bioreactor system to remediate surface- and soil-water contaminated with agrochemicals and industrial pollutants.

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**Sponsoring Organization:** U of MN

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**Location:**

**Region:** SW

**County Name:** Cottonwood

**City / Township:** Windom

_____ Knowledge Base	_____ Broad App.	_____ Innovation
_____ Leverage	_____ Outcomes	
_____ Partnerships	_____ Urgency	_____ TOTAL

## I. PROJECT STATEMENT

The pervasive use of industrial chemicals, including pharmaceuticals, pesticides, and plant growth enhancers, has resulted in the deterioration of water quality throughout the State of Minnesota. Additionally, many of these compounds make their way into surface and ground water where they ultimately jeopardize human health and harm flora and fauna. This is especially true for some pharmaceutical and agricultural chemicals that have potential hormone-mimicking effects on humans and animals. It is not possible, or even desirable, to curtail agriculture in Minnesota as the need to supply food and feedstocks for biofuels will only increase. In light of this pressure, we need to devise more effective agricultural practices that maintain our natural resources. In this proposed research, our twenty years of experience in using microbes to degrade agricultural wastes will be used in a novel design that integrates new technology with widespread agricultural practices.

Recently, a relatively inexpensive treatment technology has been developed, a Woodchip Bioreactor, to remediate surface and soil water contaminated with fertilizer leaching from agricultural fields. Woodchip bioreactors are frequently coupled to tile-drained agricultural fields and consist of a lined trench containing woodchips of various size classes and porosities (Figure 1). The wood chips are rapidly colonized by microorganisms, which act as the engines driving biodegradation of the chemical compounds present in water. Water flow through the bioreactor is controlled by a weir at each end of the bioreactor, that controls 1) the height of the water in the bioreactor, 2) in-field water table management, and 3) the biodegradation rate of the chemicals suspended in the water (see Figure 1). While originally developed to promote the microbial-mediated removal of nitrate, this same technology holds great promise as a remediation tool to clean surface and soil water contaminated with many agrochemicals and pharmaceuticals.

The overall goals of the research proposed here are to develop and utilize a microbial-based woodchip bioreactor system to remediate surface- and soil-water contaminated with agrochemicals, and prevent surface and ground water contamination from occurring in the first place. Our initial studies will concentrate on the remediation of water contaminated with atrazine and related triazine herbicides, some of which have been reported to act as hormone mimics (endocrine disruptors). Atrazine is the most widely used herbicide in the U.S., and it frequently makes its way into surface-, soil-, and ground-water. However, the system we propose to implement can be used with a wide range of other chemicals and pharmaceuticals; it will have widespread application throughout Minnesota for a range of pollution issues. We currently have at our disposal several microorganisms that rapidly degrade atrazine, having extensively characterized these microbes at the genetic, biochemical, and ecological levels over the last 15 years. We also have at our disposal many other bacteria degrading other chemical compounds. This system we propose serves two purposes: (1) it will allow us to continue to use agrochemicals to increase crop productivity, which will provide continued economic enhancement for Minnesota's farmers, provide a lasting food supply, and allow for extensive growth of plants for biofuel production; and, at the same time, (2) allow us to improve surface and ground water quality throughout the state of Minnesota.

To achieve our goals, we propose to utilize a woodchip bioreactor, initially installed by the Minnesota Department of Agriculture at the Thompson farm in Windom, MN (Cottonwood County), to degrade atrazine contaminated water originating from an agricultural field. The Windom bioreactor is approximately 50 feet long, 10 feet wide, and 8 feet deep and processes subsurface tile water from approximately 58 acres of tile-drained land. While the bioreactors themselves are relatively cheap to install and maintain (~\$6,400), and thus offer a cost effective approach to improve water quality, in our studies we will leverage the use of an existing bioreactor that has been in place for one year, and has the ability to be continually supplied with water from an adjacent lake (within 100 yds). In these studies we

will inoculate the wood chips within the bioreactor with naturally-occurring atrazine degrading bacteria and evaluate how well the system performs in degrading atrazine over time. We will also determine over two cropping seasons the rate and duration of bacteria growth, appropriate reactor conditions, and if the bioreactor simultaneously removes nitrate and other bacteria from soil leachates.

## **II. DESCRIPTION OF PROJECT RESULTS**

### **Result 1: Establishment and monitoring of sensors**

**Budget: \$50,000**

During this phase of the project we will establish and calibrate a series of sensors along the bioreactor length to monitor water temperature, pH, redox potential, flow rate, and nitrate nitrogen.

Specific deliverable include data on bioreactor performance and characterization data that will be used for subsequent results and performance testing.

### **Result 2: Development and inoculation of atrazine degrading microbial populations in bioreactor.**

**Budget: \$97,913**

In these studies we will develop stable, atrazine-degrading bacterial inoculants for placement in the bioreactor and install bacteria in the bioreactor.

Specific deliverables include data on the stability of the degrading bacteria over time and their ability to degrade atrazine within the bioreactor.

### **Result 3: Determine the efficacy of bioreactor to remove chemicals from water. Budget: \$87,913**

In this result we will examine the woodchip bioreactor for its ability degrade atrazine levels in soil leachate. We will also determine if atrazine-degrading bacteria are released from the bioreactor, and if the bioreactor simultaneously reduces levels of nitrate and fecal bacteria in the effluent.

Specific deliverables include data on the efficacy of the bioreactor to remove atrazine, nitrate and bacteria from soil leachate water, and bioreactor run parameters effecting efficient removal of these materials from water. This will be done over two field seasons.

### **Result 4: Project data dissemination.**

**Budget: \$9,000**

Specific deliverables include dissemination of reports made to the LCCMR, periodic updates made to cooperators, in seminars given throughout the state, at the Agroecology Summit, and in scientific publications in peer-reviewed journals. Results from our studies will also be posted on a website that will be specific for this project. Our data will be utilized by cooperating agencies to prioritize pollution abatement efforts, implement best management practices, and validate pollution prevention efforts.

## **III. PROJECT STRATEGY**

**A. Project Team/Partners.** The project will be carried out by Drs. Michael Sadowsky and Larry Wackett, (U. of Minnesota) who will be responsible for project design and implementation. The project will be done in collaboration with Mark Dittrich (Minn. Dept of Ag), who has oversight of the bioreactor and will direct installation of monitoring equipment for bioreactor characterization, and Mr. Tony Thompson, site owner, who will help in data gathering and sample collection.

**B. Timeline Requirements.** The project will be completed in 2 years, and data will be collected over two field seasons to ensure replication of our studies and that adequate rainfall and water flow is obtained.

## Project Budget

### IV. TOTAL PROJECT REQUEST BUDGET (two-year budget)

<u>BUDGET ITEM</u> (See list of Eligible & Non-Eligible Costs, p. 13)	<u>AMOUNT</u>
<b>Personnel:</b>	\$ -
one post-doctoral associate, 100% time, two year appt., salary is 87% and fringe rate 17% of total amount. Post doc will be working on the woodchip bioreactor.	83,826
one graduate Research Assistant, 50% time for two years. Salary is 56% and fringe is 44% of total amount. Graduate RA will be working on the woodchip bioreactor.	68,000
one undergraduate student 25-50% time for two years. Salary is 93% and fringe is 7% of total amount. Student will set-up a dedicated web site for dissemination of project activities and data.	7,000
<b>Contracts:</b> subcontract to MDA to instrument and monitor the woodchip bioreactor for performance as follows: 1. Subcontractors salaries \$12,000; 2. Farm Cooperator Fee \$1000; 3. Equipment and Materials \$25,000; 4. Travel \$5,000; 5. Other costs: Education, and outreach \$2,000; 6. Data Transmission Fees: \$5,000; Total Funds Requested: \$50,000	50,000
<b>Equipment/Tools/Supplies:</b> includes funds for genomic DNA isolation kits, general laboratory and bacteriological supplies, growth media, PCR enzymes, plasticware, other DNA isolation kits, oligonucleotide primers, laboratory reagents and supplies, atrazine detection microplates, nitrate and fecal bacteria measurement supplies, plasticware, chemicals and reagents, and fees (\$5,000 per year) for the production of atrazine-degrading bacteria at the UMN Biotechnology Resource Center	30,000
<b>Travel:</b> travel to field site in Windom, MN to obtain samples and bioreactor data	4,000
<b>Additional Budget Items:</b> sponsored publication fees for publication in peer-reviewed journals	2,000
<b>TOTAL PROJECT BUDGET REQUEST TO LCCMR</b>	244,826

### V. OTHER FUNDS

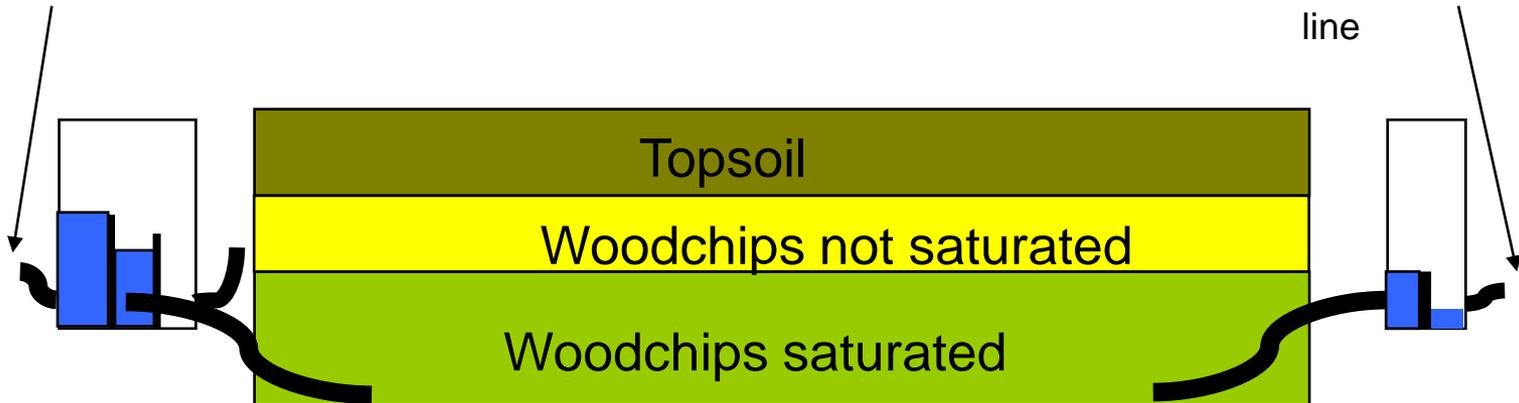
<u>SOURCE OF FUNDS</u>	<u>AMOUNT</u>	<u>Status</u>
<b>Other Non-State \$ Being Applied to Project During Project Period:</b>	\$ -	
<b>Other State \$ Being Applied to Project During Project Period:</b>	\$ -	
<b>In-kind Services During Project Period:</b>	\$ -	
<b>Remaining \$ from Current Trust Fund Appropriation (if applicable):</b>		
<b>Funding History:</b>	\$ -	

## Woodchip Bioreactor Cottonwood County Site:

- Trench is 10 feet wide, 50 feet long, 8 feet deep, at 0.1% grade.
- An eight inch tile line was intercepted, brought to a tri-chamber control structure (allows for bypass during high flows).
- Forty feet of tile was replaced with non-perf, to avoid piping around control structure, and to provide water table management in-field.
- The trench is lined with heavy plastic to insure same in-flow and out-flow measurements
- On the downstream and upstream ends,a structure will be used to control the height of the water in the bioreactor. Water passing through this control can be measured with pressure transducer and exits into an outlet.
- The trench is filled to within 1-2 feet of the surface with woodchips, and topsoil is placed over top. \
- Atrazine-contaminated water entering the bioreactors will be bioremediated by atrazine-degrading bacteria living on the surface of wood chips.



From tile line



### **Project Manager Qualifications and Organization Description:**

Project Manager: Dr. J. Michael Sadowsky

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Dr. Sadowsky will have chief management responsibilities for overseeing the proposed project. He will be responsible for working with the co-PI (Dr. Lawrence Wackett - UMN) and Cooperator (Mark Dittrich – MDA) to ensure that project goals, results and timelines are met. He will also be responsible for working with the graduate student and postdoctoral associate at UMN and MDA staff. Dr. Sadowsky is an environmental microbiologist with 16 years research experience in the use of microorganisms and enzymes for the biodegradation of atrazine and related herbicides. Dr. Sadowsky's laboratory studies the distribution and diversity of microorganisms in the environment and uses genetic, genomic, and biotechnology tools to examine how microorganism become established in new environments.

Dr. Wackett is a microbiologist and biochemist in the Department of Biochemistry, Molecular Biology, and Biophysics; and the BioTechnology Institute at the University of Minnesota. Dr. Wackett will work directly with Dr. Sadowsky and the project personnel. He will help design studies concerning the application of atrazine degrading bacteria into the bioreactors, and detection of atrazine and its metabolites in influent and treated water and help studies examining the fate of bacteria in the bioreactors. Dr. Wackett has over 25 years experience in the areas of biodegradation and bioremediation research. The Wackett laboratory studies microbial catabolic enzymology, enzyme evolution, and applications of enzyme and microorganisms for biotechnology. He helped create the widely popular and useful "University of Minnesota Biocatalysis/Biodegradation Database".

The project will be done in collaboration with Mark Dittrich at the Minnesota Department of Agriculture. Mark , who has oversight of the bioreactor at the and will direct the installation and calibration of monitoring equipment for bioreactor characterization, and will coordinate with Mr. Tony Thompson, the site owner, who will help in sample collection. The overall mission of the Minnesota Department of Agriculture is to "support the development of an agriculture that is profitable and environmentally sound." Mark Dittrich is a Senior Planner in the area of Conservation Drainage for the MDA.