

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

LCCMR ID: 216-G

Project Title:

Science and Innovation from the Soudan Iron Mine

LCCMR 2010 Funding Priority:

G. Creative Ideas

Total Project Budget: \$ \$551,451

Proposed Project Time Period for the Funding Requested: 3 years, 2010 - 2013

Other Non-State Funds: \$ \$0

Summary:

The Soudan Iron Mine in Northern Minnesota is a unique and exciting opportunity for basic science, educational outreach and innovative applications impacting the areas of drug discovery, bioenergy and bioremediation.

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Sponsoring Organization: U of MN

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Fax: _____

Web Address: _____

Location:

Region: Regional

County Name: Ramsey

City / Township: St. Paul

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

MAIN PROPOSAL

PROJECT TITLE: Science and Innovation from the Soudan Iron Mine

I. PROJECT STATEMENT

There are three integrated facets of this proposal: Basic Science, Innovative Applications and Outreach and Education. Basic Science will provide a fundamental understanding of the unique environment found in the bottom of the Soudan Iron Mine. Innovative Applications will explore how novel microbes from the mine will be harnessed for drug discovery, bioenergy and bioremediation. Outreach and Education will integrate our findings to educate the general public and mine visitors.

The study of extreme or novel environments can sometimes lead to discoveries that change the world. In the early 1970's such a discovery was made in hot springs in Yellowstone National Park – bacteria thriving at temperatures previously thought to be inhospitable for life were identified. The commercialization of proteins from these bacteria revolutionized science, enabling an era of modern molecular genetics, including sequencing of the human genome, and is the foundation of a multi-billion dollar biotechnology industry. **Based on our preliminary analysis, a similar commercial opportunity may be found deep underground in Northern Minnesota at the bottom of an abandoned iron mine.**

The Soudan Iron Mine near Ely, MN closed in 1962 and later became the Soudan Underground Mine State Park. The Soudan Mine is both a historical site, offering the public an opportunity to reflect on past economic activities and technologies of the iron range, and also houses the High Energy Physics Lab, administered by the University of Minnesota. This state park attracts around 40,000 visitors per year; making it an important destination for both residents of Minnesota and surrounding states.

The lowest level of the Soudan Mine is home to an extraordinary environment where the fields of microbiology, geochemistry and mineralogy converge. The sedimentary iron-rich rock that was mined for 80 years at this site is known as a 'Banded Iron Formation' or BIF. BIFs contain a substantial portion of the iron found on the surface of our planet, and the Soudan BIF is estimated to be around 2.7 billion years old. Typically, oxygen is required to form rust (as we all know well in Minnesota), however the Soudan BIF was deposited ~ 400 million years before oxygen was present in significant amounts in our atmosphere. Microbiologists have suggested that iron-oxidizing bacteria could have been responsible for these ancient sedimentary formations and studying this site will provide insight into this process.

In the lowest level of the Soudan Iron Mine water seeping from boreholes drilled in the waning days of the mine can be found. This water is quite unusual since it is almost three times saltier than seawater and is devoid of oxygen. Associated with many of these seeps are unique iron oxide structures and throughout this strange water are poorly characterized iron minerals and thriving bacterial communities. Some of the bacteria we have analyzed from this environment appear to be distant relatives of bacteria found in the ocean. What are bacteria from the ocean doing in water found 2341 feet underground in northern Minnesota? Are descendants of organisms that helped form the Soudan BIF still living in waters trapped within the iron formation? Are there novel microbes found here?

The unique environment of the lowest level of the Soudan Mine presents many exciting opportunities directly related to LCCMR funding priorities (Creative Ideas). Result 1 will help us understand the fundamental nature of this unique environment where we will characterize the microbiology, mineralogy and geochemistry of the Level 27 brine and the formations found in the mine. This information will be used to protect this unusual and exciting ecosystem and communicate our findings (result 3). In result 2 we will explore exciting innovative applications that utilize microbes isolated from the mine. **Given the novelty of the microbes and environment, we expect this work will lead to new areas of research and potential revenue for the State of Minnesota in the areas of drug discovery, bioenergy and bioremediation.** We have already gathered preliminary results demonstrating that several of the Soudan Mine isolates are highly unusual and may produce novel compounds.

II. DESCRIPTION OF PROJECT RESULTS

Result 1: Basic Science – Microbiology, Mineralogy and Geochemistry

Budget: \$379,201

The microbiology section will focus on identifying and categorizing the microbial populations found in the mine. The mineralogy portion will focus on synchrotron-enabled analysis of both structure and composition of minerals found in the Level 27 brine and on samples from iron formations. The geochemistry analysis will focus on chemical and isotopic characterization of the Level 27 brine, in addition to heavy metal analysis on Level 10 (and elsewhere in the mine as directed by mine staff).

Deliverables:	Completion Date
1. Molecular phylogenetic analysis and characterization of microbes	June 30, 2013
2. Mineralogical, speciation and elemental analysis	June 30, 2013
3. Isotope analysis of level 27 brine, gas composition, geochemistry	June 30, 2013

Result 2: Innovative Applications – Drug Discovery, Bioenergy, Bioremediation Budget: \$162,250

In this section we will determine the feasibility of utilizing microorganisms isolated from the Soudan Iron Mine in three specific commercial areas. Our priority here is to identify potential commercial assets deriving from microbes cultivated from the Soudan Mine.

Deliverables:	Completion Date
1. Screen novel isolates for production of anticancer and antimicrobial drugs	June 30, 2013
2. Demonstrate the use of iron oxidizing bacteria in microbial fuel cells	June 30, 2013
3. Isolate and characterize bacteria that promote oxidation of toxic metals	June 30, 2013

Result 3: Public Outreach and Education Budget: \$10,000

We will collaborate to develop training for DNR tour guides to describe the features, microorganisms, biogenic mineralogy and geochemistry within the mine and collaborate with staff to generate educational displays and to develop best practices for protecting this unique environment.

Deliverables:	Completion Date
1. DNR microbiology training module, best practices assessment, display	June 30, 2012

III. PROJECT STRATEGY

A. Project Partners

Dr. Brandy Toner (Co-PI) is responsible for all mineralogical analyses. She is an Assistant Professor at the University of Minnesota (Soil, Water and Climate) with extensive experience working with synchrotron-based analysis of biogenic and natural minerals and characterizing microbial populations.

Dr. Christine Solomon (University of Minnesota) of the Center for Drug Design will isolate and screen microbes for production of medically relevant compounds. Her group will be involved in screening for antibacterial and anticancer activities, and will purify and characterize novel compounds.

Dr. E. Calvin Alexander, Jr. is a Morse-Alumni Professor at the University of Minnesota (Geology and Geophysics) and will be responsible for all geochemical and isotopic analyses.

James Essig (DNR, Park Manager – Soudan Underground Mine State Park) will help coordinate research trips to the mine, outreach activities on site and future commercialization possibilities.

Dr. Daniel Bond (University of Minnesota) is an Assistant Professor of Microbiology and member of the BioTechnology Institute. Dr. Bond is an expert in microbial fuel cell technology and metal reduction.

B. Timeline Requirements

Three years of funding will be sufficient to accomplish the basic, fundamental research (Result 1), explore innovative applications (Result 2) and education / outreach (Result 3) proposed here.

C. Long Term Strategy

At the end of the three year project, we expect to have obtained results that will support funding from national agencies (such as the National Science Foundation, Department of Energy and National Institutes of Health) and we will begin partnering with organizations (such as Natural Resources Research Institute) and local companies to utilize novel compounds identified, for potential bioenergy applications and for bioremediation using microbes isolated from the mine.

Project Budget

IV. TOTAL PROJECT REQUEST BUDGET (3 years)

BUDGET ITEM	AMOUNT	% FTE
Personnel:		
Brandy Toner (Co-PI - 1 month summer salary) - Result 1 - Years 1-3	\$ 35,280	8%
UM Scientist (Scott Alexander, 1 month salary / year) - Result 1.3 - Years 1-3	\$ 13,740	8%
1 Postdoctoral Research Associate (Center for Drug Design) - Result 2.1 - Years 1-3	\$ 68,250	50%
1 Graduate Research Assistant (Soil, Water, Climate, PhD) - Result 1.2 - Years 1-3	\$102,381	50%
1 Graduate Research Assistant (Microbiology, PhD) - Result 1.1 - Years 1-3	\$114,000	50%
1 Graduate Research Assistant (Microbial Engineering, MS) - Results 2.2, 2.3 - Years 2-3	\$ 64,000	50%

Travel: In-state travel to/from mine (+ lodging), in/out of mine - estimate 5-6 trips / year	\$ 12,000
Travel: Out of state travel to national meetings to present results from this research and to learn from colleagues in other states. American Society for Microbiology General Meeting, American Geophysical Union, Applied and Environmental Microbiology Gordon Conference (1 trip / year for 2 @ \$1000 / person)	\$ 6,000

Result 1:

Molecular biology reagents (PCR reagents, DNA extraction kits, plasmid purification kits (\$250 ea., ~12/year), enzymes, chemicals, microbiology consumables (agar, media), general lab supplies (tubes, tips, gloves etc.), cultivation supplies, sterile sampling supplies - Micro PhD Student - Years 1-3	\$ 30,000
Laboratory supplies and consumables (chemicals, sample storage, sample preparation, general lab supplies) - SWC PhD Student - Years 1-3	\$ 15,000
Microscopy (SEM, Light - User fees at CBS Biological Imaging Facility)	\$ 2,500
Sequencing for phylogenetic analysis of microbial communities - bacteria, fungi and archaea (AGAC Sequencing facility on UM campus \$3.50 / reaction, estimate 600 reactions / year over 3 years)	\$ 6,300
Advanced Photon Source at Argonne National Labs (Chicago - travel, lodging, user fees) for mineralogical analyses (years 1-3)	\$ 10,500
Chemical, isotopic and gas analysis (reagents for extraction / preservation, measurements, user facility fees) Years 1-3	\$ 30,000
Publication fees (~ 3 total, \$500/publication - page charges required to make scientific discoveries available to other scientists and the public)	\$ 1,500

Result 2:

Bioremediation experiments - Heavy metal quantitation, pure and mixed culture screening for bioreduction, characterization of strains, laboratory consumables. Bioenergy experiments - electrode maintenance, new reactor design for Fe oxidizers, media preparation for MS student - Years 2-3	\$ 15,000
Lab supplies for Center for Drug Design - chemicals and glassware for culturing microbes, DNA isolation and sequencing for strain identification, solvents for compound isolation, HPLC and MS time for compound identification and structural characterization, laboratory consumables for Postdoc (50% time) - Years 1-3	\$ 15,000

Result 3:

Outreach development (display, content development, education, implementation, content updates)	\$ 10,000
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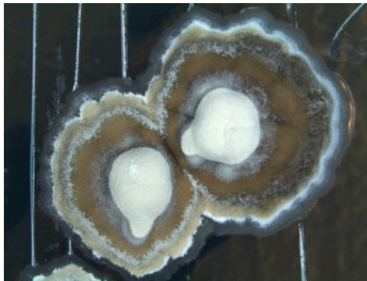
TOTAL PROJECT BUDGET REQUEST TO LCCMR	\$551,451
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V. OTHER FUNDS

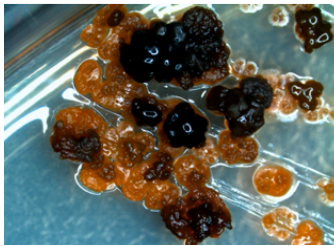
SOURCE OF FUNDS	AMOUNT	Status
Other Non-State \$ Being Leveraged During Project Period:	N/A	
Other State \$ Being Spent During Project Period:	N/A	

Basic Science

Discover Novel Drugs



Novel bacterial isolates of the genus *Streptomyces* cultivated from the Level 27 brine. These kind of bacteria are known to produce over 2/3 of natural antibiotics used clinically. More isolates will be obtained and tested for production antibiotics and other bioactive compounds.



LEVEL NO. 27
2341 FEET BELOW THE SURFACE,
689 FEET BELOW SEA LEVEL

Questions Our Research Will Answer

- ✦ Microbiology: What bacteria, fungi and archaea exist in this extreme environment? What do they eat? What do they breathe? Who are they related to? How can we culture them in the laboratory?
- ✦ Mineralogy: What kinds of minerals are formed in the Level 27 brine? Are the minerals formed by activity of microbes? What is the composition of iron oxide formation? How does the brine chemistry influence dam formation (see center picture)?
- ✦ Geochemistry: How ancient is the water found on Level 27? Is the water composition the same in all areas of Level 27?

Develop New Technology

Bioremediation: We have cultured iron oxidizing bacteria from the Level 27 brine. These bacteria accelerate rust (iron oxide) formation, and could be used to precipitate toxic metals, facilitating remediation.

Renewable Energy: Bacteria will be tested for their ability to be used in microbial fuel cells to increase electrical output.



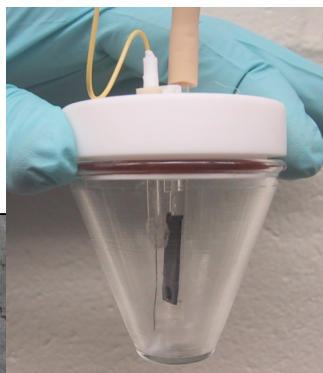
Biogenic iron oxide (ferrihydrate) 'speleothem' deposit from Level 27 of the Soudan Iron Mine.



Scott Alexander, UMN

Outreach and Education

- ✦ About 40,000 people visit the Soudan Underground Mine and Park each year, including tourists visiting the Iron Range and student groups from Minnesota and surrounding states.
- ✦ We will work with the DNR to develop a microbiology, mineralogy and geochemistry training module to incorporate results from our research into the historical mine tour.
- ✦ We will develop an interactive display for the Park's Visitor Center that will provide an up-to-date learning experience about research specifically derived from this proposal.



Electrobioreactor (above), Soudan *Marinobacter* SEM image (left).

VI. PROJECT MANAGER QUALIFICATIONS & ORGANIZATION DESCRIPTION

Project Manager Qualifications

Dr. Jeffrey A. Gralnick, Project Manager and Principle Investigator

Dr. Gralnick is an Assistant Professor of Microbiology at the University of Minnesota and a member of the BioTechnology Institute, located on the St. Paul Campus. Gralnick was trained in Bacteriology at the University of Wisconsin – Madison where he specialized in microbial physiology. He then spent three years at the California Institute of Technology (Caltech) in the Division of Geological and Planetary Sciences where he studied Geomicrobiology – specifically how bacteria directly influence the fate of minerals. His training in both microbiology and geomicrobiology will provide cohesiveness for our interdisciplinary team. Please visit his website for further description of research interests and for images from the Soudan Iron Mine:

http://cbs.umn.edu/labs/gralnick/Gralnick_Lab/

Dr. Gralnick is among the first group of Associate Fellows in the University's new Institute on the Environment, based primarily on his vision for educational outreach, dedication to environmental microbiology and ability to blend both basic and applied science in his work. His commitment to education and outreach is demonstrated by his participation in the College of Biological Sciences Nature of Life program for incoming freshmen. He also co-teaches a course for freshman entitled 'Happy in Hell: Microbes in Extreme Environments,' where examples from the Soudan Iron Mine are discussed.

Dr. Gralnick is the Project Manager of this proposal. Through four years at the University, he has established a solid record of both internal (Institute on the Environment, Initiative for Renewable Energy and the Environment, Graduate School, Academic Health Center, Cargill Initiative for Higher Education) and external funding (Office of Naval Research). Importantly, Dr. Gralnick is a member of the University of Minnesota's BioTechnology Institute – an organization designed to help interface academia with industry (see below). These relationships will be key to any future commercialization of technologies or discoveries made by research proposed here.

Dr. Gralnick is on a 12-month tenure-track appointment through the University of Minnesota Academic Health Center (Microbiology) and has therefore not requested any salary support.

Organizational Description

The University of Minnesota's Mission is threefold: 1) Research and Discovery, 2) Teaching and Learning, and 3) Outreach and Public Service.

The University of Minnesota's BioTechnology Institute (BTI) provides advanced research, training, and university-industry interaction in biological process technology, a major area of biotechnology research. The Institute is the central University of Minnesota vehicle for coordinated research in the biological, chemical, and engineering aspects of biotechnology.

The Minnesota Department of Natural Resources – State Parks Mission: We will work with the people of Minnesota to provide a state park system that preserves and manages Minnesota's natural, scenic, and cultural resources for present and future generations, and provides appropriate recreational and educational opportunities.