MTSU Clean Energy Initiative Project Funding Request

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There are five (5) sections of the request to complete before submitting. See http://www.mtsu.edu/sga/cleanenergy.shtml for funding guidelines. Save completed form and email to cee@mtsu.edu or mail to MTSU Box 57.

1. General Information	
Name of Person Submitting Request **Dr. James Brian Robertson**	
Department/Office **Biology / Jones Hall 334**	Phone # (Office) **898-2066**
MTSU Box # **60**	Phone # (Cell)
E-mail **james.robertson@mtsu.edu**	Submittal Date 1-17-2012

2. Project Categories (Select One)			
Select the category that best describes the project.			
	Energy Conservation/Efficiency		Sustainable Design
X	Alternative Fuels		Other
	Renewable Energy		

3. Project Information
a. Please provide a brief descriptive title for the project. **b.** The project cost estimate is the expected cost of the project to be considered by the committee for approval, which may differ from the total project cost in the case of matching funding opportunities. **Any funding request is a 'not-to-exceed' amount. Any proposed expenditure above the requested amount will require a resubmission.** c. List the source of project cost estimates. d. Provide a brief explanation in response to question regarding previous funding.
3a. Project Title **Development of a Biosensor to Detect Hydrogen Production in Photosynthetic Microbes**
3b. Project Cost Estimate **$4,250 – for consumables / genetic engineering materials** **$4,737 – for student summer salary and benefits** **$8,987 – Total (Price breakdown is below)** $ 80 Petri dishes, $ 124 disposable tips $ 124 microcentrifuge tubes

$ 550 growth media chemicals
$ 600 PCR primers
$ 680 DNA modifying enzymes
$ 86 filter units
$ 297 DNA extraction kits
$ 242 DNA purification kit
$ 68 Ampicillin
$ 82 Spectinomycin
$ 57 SybrSafe DNA stain
$ 49 Autoclave tray
$ 46 Sample storage boxes
$ 180 Agar
$ 165 PCR tubes
$ 300 glassware
$ 300 p20 Micropipette
$ 100 DNA sequencing services
$ 120 Fluorescent grow lights and chamber
$4400 Summer Salary for Master's Student @ $10/hr for 40h/week for 11 weeks
$ 337 Master's student Fringe Benefits for Summer work

3c. Source of Estimate
Itemized costs for materials are found in catalogs from Fisher Scientific, New England Biolabs, Invitrogen, Promega, Sigma-Aldrich.

3d. If previous funding from this source was awarded, explain how this request differs? **N/A**

4. Project Description

(Completed in as much detail as possible.)

a. The scope of the work to be accomplished is a detailed description of project activities.
b. The benefit statement describes the advantages of the project as relates to the selected project category.
c. The location of the project includes the name of the building, department, and/or specific location of where the project will be conducted on campus.
d. List any departments you anticipate to be involved. Were any departments consulted in preparation of this request? Who? A listing may be attached to this form when submitted.
e. Provide specific information on anticipated student involvement or benefit.
f. Provide information for anticipated future operating and/or maintenance requirements occurring as a result of the proposed project.
g. Provide any additional comments or information that may be pertinent to approval of the project funding request.

4a. Scope: Work to be accomplished

The work proposed here has ONE major objective: to develop a biological hydrogen sensor for strains of microbes that are capable of using the energy from sunlight to produce hydrogen. As a result, conditions and strains that are better at producing hydrogen can be rapidly identified for future biofuel production.

Some microbes like the soil bacterium *Ralstonia eutropha* are capable of consuming hydrogen as a food source, so they possess the ability to sense hydrogen in the environment. The genes that code for this hydrogen sensing ability are known. Other microbes like photosynthetic cyanobacteria can use energy from the sun to split water into hydrogen and oxygen; however, they do not possess a hydrogen sensor. Although it is often not in cyanobacteria's best interests to make hydrogen in this way, they can produce hydrogen when certain environmental conditions are just right. In addition, cyanobacteria can be genetically modified (mutated) so that they produce more hydrogen.

The key to identifying strains of mutated cyanobacteria that produce more hydrogen than their unaltered counterparts is to develop a detection system that is sensitive to hydrogen so that the more hydrogen the culture produces, the more "signal" it gives off. The light emitted from the glow-in-the-dark enzymes of bioluminescent organisms like fireflies and jellyfish are good biological reporters because the genes for these enzymes (called luciferases) can be produced by microbes and the products of these enzyme reactions glow as a result of the luciferase gene being activated (for example, activated by the hydrogen sensor from *Ralstonia eutropha*).

Through this work, we will use genetic engineering to construct a molecular biosensor for hydrogen that uses the hydrogen-sensing genes from *Ralstonia eutropha* to control the light production from the glow-in-the-dark gene from fireflies. This hydrogen sensor gene can be inserted into cyanobacteria DNA so that by measuring light emitted from the cyanobacteria, researchers can quickly identify conditions and strains of cyanobacteria that produce more hydrogen. Once this hydrogen reporter system is made, it will be tested both with application of environmental hydrogen as well as in strains of cyanobacteria that produce their own hydrogen.

4b. Scope: Benefit Statement

Hydrogen is a potential biofuel so developing strains of cyanobacteria that can readily produce it is a valuable research endeavor. Since cyanobacteria do not normally want to waste energy splitting water into hydrogen and oxygen, identifying mutant strains that produce more hydrogen than others can be like finding a needle in a haystack (that is, unless you can make the needle glow in the dark). By splicing the hydrogen sensor of *Ralstonia eutropha* to the glow-in-the-dark gene of fireflies, students at MTSU can make strains of cyanobacteria that glow in the dark depending on the amount of hydrogen the strain produces. Once strains are identified that produce more hydrogen, further mutations can be made to the cyanobacteria to improve their hydrogen production even more. Through this process, hydrogen production can be ratcheted up at each step. What is lacking for this process to work is a way to quickly and easily tell which mutants make the most hydrogen. That is why this research would be valuable toward production of hydrogen from cyanobacteria.

Results of this research will be published in one or more reputable science journals and MTSU's Clean Energy Initiative will be acknowledged for its financial support of this work. Once published, other researchers that are working to develop hydrogen as a biofuel (including labs at MTSU) can have access to the benefits that our research produced.

4. Project Description (continued)

4c. Location of Project (Building, etc.)
This research will be conducted in Dr. Robertson's lab in Davis Science Building (room 102), where he has the equipment necessary for students to perform the required genetic manipulations and culture development.

4d. Participants and Roles
Dr. James Robertson is an expert in bioluminescence and genetic engineering and will oversee the work of students on this project.

Robbie Martin is a master's student who will be responsible for constructing the molecular hydrogen sensor and testing its effectiveness in cyanobacteria.

James Dolbow is an undergrad researcher who will assist Robbie and learn techniques for transgenic manipulation.

4e. Student participation and/or student benefit
This research project will be completely student-driven. Dr. Robertson will lend his expertise and advice to guide the project but the hands-on construction and development will be the work of masters and undergraduate students. Additional students from those listed in 4d may participate in parts of this project for educational purposes. Students will get hands-on experience with molecular biology, and so better appreciate the intricacies of developing biofuels and the organisms that produce them.

4f. Future Operating and/or Maintenance Requirements

None – this is a one-time investment in research material needed to construct the genes for protein-based hydrogen sensors. Once created, they can be copied and mass produced for virtually no cost. Cyanobacterial strains identified through this technology can also be grown for little to no cost. The proposed student summer salary will be used for the summer of 2013.

4g. Additional Comments or Information Pertinent to the Proposed Project
This proposal is for technology development with a proposed timeframe of two semesters to construct the hydrogen sensor and another two semesters for testing and implementing the system. Large scale production of hydrogen is beyond the scope of this proposal. However, should this research be successful, the goal of scaling up production may be worthy of another proposal in the future.

5. Project Performance Information

Provide information if applicable.

a. Provide information on estimated annual energy savings stated in units such as kW, kWh, Btu, gallons, etc.
b. Provide information on estimated annual energy cost savings in monetary terms.
c. Provide information on any annual operating or other cost savings in monetary terms. Be specific.
d. Provide information about any matching or supplementary funding opportunities that are available. Identify all sources and explain.

5a. Estimated Annual Energy Savings (Estimated in kW, kWh, Btu, etc.)
See Below

5b. Annual Energy COST Savings ($)
This project by itself does not produce energy cost savings, however it is the beginning of a series of steps that can identify and develop efficient hydrogen-producing microbes that utilize solar energy for hydrogen production. In the big picture, biological solar power offers advantages over photoelectric solar panels in that microbes can be grown at little expense rather than solar panels that are manufactured at great expense.

5c. Annual Operating or Other Cost Savings. Specify. ($)

5d.Matching or Supplementary Funding (Identify and Explain)

Most of the equipment needed for this research (and all of the expensive equipment) has been purchased using other funds. Additionally, graduate student labor for this project will be funded through a graduate teaching assistantship for the academic year.

Linda Hardymon

From: James Robertson [James.Robertson@mtsu.edu]
Sent: Thursday, January 26, 2012 11:36 AM
To: cee@mtsu.edu
Subject: CEI Research Proposal
Attachments: CEI Proposal Robertson 2012.doc

Dear Dr. Kelly and Sustainable Campus Fee Committee,

Attached is my research proposal for the Clean Energy Inititive.

If you have any questions, please feel free to contact me.

Sincerely,
J. Brian Robertson