Environment and Natural Resources Trust Fund (ENRTF)
2010 Work Program

Date of Report: November 24, 2009
Date of Next Progress Report: N/A
Date of Work Program Approval:
Project Completion Date: July 1, 2013

I. PROJECT TITLE: Sustainable Biofuels

Project Manager: G. David Tilman
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Location: Cedar Creek Ecosystem Science Reserve
2660 Fawn Lake Drive Northeast
East Bethel, MN 55005 (Anoka/Isanti Counties)

Total ENRTF Project Budget:	**ENRTF Appropriation**	**$ 221,000**
	Minus Amount Spent:	**$ 0**
	Equal Balance:	**$ 221,000**

Legal Citation: M.L. 2010, Chp. 362, Sec. 2, Subd. 7b

Appropriation Language:
$221,000 is from the trust fund to the Board of Regents of the University of Minnesota to determine how fertilization and irrigation impact yields of grass monoculture and high diversity prairie biofuel crops, their storage of soil carbon, and susceptibility to invasion by exotic species. This appropriation is available until June 30, 2013, by which time the project must be completed and final products delivered.

II. PROJECT SUMMARY AND RESULTS:

Perennial grassland ecosystems have the potential to provide Minnesota with locally grown energy sources that reduce greenhouse gas emissions, improve water quality, and provide other important services. It seems likely that such perennial grassland crops will experience climate change in the coming decades, and that in attempts to improve yields farmers would try fertilizing and/or irrigating their grasslands. However the effects of these factors on the potential benefits of alternative biomass crops, including switchgrass monocultures, *Miscanthus* monocultures and high-diversity prairies, are unstudied and unknown. Soils are the largest storehouse of carbon in Minnesota, and soil carbon sequestration may become a marketed item as part of a carbon cap and trade system. The net effects of warming, fertilization and irrigation on

soil carbon storage, though, have not been tested. Similarly, we do not know how either the biomass yields of alternative crops or their susceptibility to invasion by exotic plant species might be impacted by these factors.

This project will use a Fertilization-Irrigation Experiment (consisting of 96 plots) and a Climate Experiment (consisting of 114 plots) to determine how irrigation, fertilization, and climate warming impact yields, carbon sequestration, plant biodiversity, water quality and susceptibility to invasion in grasslands with varying diversity (1, 4, 16 or 32 plant species). An additional Invasion Experiment nested within the first two experiments would also monitor whether *Miscanthus*, an exotic perennial grass species, poses a threat as pernicious invasive. Our results will be synthesized to find methods for optimizing biofuel production, carbon storage, and habitat restoration.

III. PROGRESS SUMMARY AS OF *[11/24/09]***:** N/A

IV. OUTLINE OF PROJECT RESULTS*:*

RESULT/ACTIVITY 1: Effects of Agricultural Inputs and Warming on Biomass Production and Sustainability

Description:

Fertilization-Irrigation Experiment

The Fertilization-Irrigation Experiment consists of ninety-six 9x9 meter plots, and is designed to determine how irrigation and fertilization would impact the yields and ecosystem functioning of four potential cellulosic biofuel crops. The four crops are switchgrass monoculture, *Miscanthus* monocultures, diverse prairie polycultures (sixteen species) and highly diverse prairie polycultures (thirty-two species).

These Fertilization-Irrigation Experiment plots will be located within the same grid of plots that contains the thirty-eight Climate Experiment plots. This grid as a whole contains a total of 342 plots, of which 168 are in the Biodiversity Experiment (which provides control plots for the experiments discussed here) and thirty-five are in an LCCMR/ USGS supported study of the abilities of different vegetation types to prevent various agrochemicals from entering and polluting the groundwater.

The Fertilization-Irrigation Experiment employs a full-factorial design, with each plot receiving one of three levels of nitrogen addition (none, 7 $g/m^2/yr$, 14 $g/m^2/yr$) and one of two irrigation treatments (no irrigation or addition of 2.4 cm/week of water every week from mid-May through August), for a total of six possible treatment combinations. There will be four replicates of each irrigation/fertilization treatment combination applied to each of the four biofuel crops. Thus:

3 nitrogen treatments x 2 irrigation treatments x 4 replicates per biofuel crop type x 4 biofuel crops = 96 plots total

Climate Experiment

The Climate Experiment consists of thirty-eight 9x9 meter plots designed to determine the effects of climate warming, plant diversity, and plant community functional composition on the functioning of prairie-like ecosystems. The Climate Experiment also has a full factorial design, with each 9x9 meter plot being maintained at one of the four levels of diversity (1, 4, 16 and 32 species), and all three warming treatments (ambient temperature, low warming and high warming) nested as smaller subplots within. The "high" subplot is warmed by +3.0ºC using a 1500 watt Kal-Glo® infrared heat lamp, the "low" subplot is warmed by +1.5ºC using an 800 watt Kal-Glo® infrared heat lamp, while the control plot is unwarmed but has a sham (empty) "heat lamp" erected over it to control for non-warming effects, if any, such as shading. An additional fourth subplot was established, also un-warmed, and lacking any heat lamp structure.

Sampling

By weighing vegetation samples clipped from our Climate Experiment and Fertilization-Irrigation Experiment plots at peak biomass, we will determine how biomass yield (dry weight of above-ground plant matter) depends on plant diversity (the number of species the samples contained), plant composition (which species were in the plot and in what proportion), soil temperature, nitrogen fertilization and irrigation. We will discover if yields of specific plant communities are harmed or helped by warming, and how diversity is impacted by fertilization and irrigation. Statistical analysis of this data will entail a combination of regression and analysis of variance (the generalized linear models approach), taking our experimentally imposed treatments as independent variables for which we will examine both direct and interactive effects.

Summary Budget Information for Result/Activity 1:

ENRTF Budget:	**$ 52,500**
Amount Spent:	**$ 0**
Balance:	**$ 52,500**

Deliverable/Outcome	Completion Date	Budget
1. Summer 2010: Interns (3.5 undergraduates) establish the Fertilization-Irrigation Experiment. After first clearing and seeding its new plots, all 96 plots are regularly watered and fertilized. Interns maintain the 114 Climate Experiment plots as well, weeding and collecting temperature data. At the end of the summer, interns clip, sort and weigh aboveground biomass samples from the Climate plots.	8/31/10	$16800
2. Summer 2011: Interns (3.5 undergraduates) maintain all Fertilization-Irrigation Experiment and Climate Experiment plots: Fertilization-Irrigation plots are watered and/or fertilized and Climate plots are weeded and have their temperature data collected. At the end of the summer, interns clip, sort and weigh	8/31/11	$17500

aboveground biomass samples from the Fertilization-Irrigation and Climate plots.		
3. Summer 2012: Interns (3.5 undergraduates) maintain all Fertilization-Irrigation Experiment and Climate Experiment plots: Fertilization-Irrigation plots are watered and/or fertilized and Climate plots are weeded and have their temperature data collected. At the end of the summer, interns clip, sort and weigh aboveground biomass samples from the Fertilization-Irrigation and Climate plots.	8/31/12	$18200

Result Completion Date: August 31, 2012

Result Status as of *(*August 2010*)*:

Result Status as of *(*February 2011*)***:**

Result Status as of *(*August 2011*)***:**

Result Status as of *(*February 2012*)***:**

Final Report Summary:

RESULT/ACTIVITY 2: Agricultural Input and Warming Effects on Invasions by Exotic Species

Description: By carefully mapping the spatial locations of the plants in all Invasion Experiment plots (a subset of plots nested within the Fertilization-Irrigation Experiment), we will be able to observe which species invade our experimental plots each year and determine how warming, plant diversity, plant composition, fertilization and irrigation impact invasion susceptibility. Each Invasion Experiment plot will explicitly test the invasion potential of *Miscanthus* by vegetative spread, by planting a wide strip of *Miscanthus* down the center of the each plot, and a series of *Miscanthus* rhizome plugs down one of the side strips (which contain the regular prairie mixtures or monocultures). In addition, we will study the invasion potential of specific non-native invasive plant species, including *Miscanthus*, in Climate Experiment plots by adding seed (or rhizome plugs in the case of *Miscanthus*) of those species to a specific area of a subset of the plots, and measuring their survival, growth and spread.

Summary Budget Information for Result/Activity 2:

ENRTF Budget:	**$ 54,500**
Amount Spent:	**$ 0**
Balance:	**$ 54,500**

Deliverable/Outcome	Completion Date	Budget
1. Summer 2010: Interns (2 undergraduates) work	8/31/10	$9,600

with the Result 1 interns to establish the Invasion Experiment within the Management Experiment, and make initial maps and surveys of the Invasion Experiment plots. May also assist with trait and variable measurement of invasion studies within the Climate Experiment.		
2. Summer 2010: Graduate student provides expert guidance in the establishment and surveying of plots, and begins analyses of invasion data. May contribute to the invasion studies in the Climate Experiment.	8/31/10	$7,700
3. Summer 2011: Interns (2 undergraduates) survey and map the Invasion Experiment plots multiple times throughout the summer. May assist with trait and variable measurement of invasion studies within the Climate Experiment.	8/31/11	$10,000
4. Summer 2011: Graduate student continues to analyze invasion data and provide guidance to the interns working on the experiments. Begins research	8/31/11	$8,100
5. Summer 2012: Interns (2 undergraduates) survey and map Invasion Experiment plots multiple times throughout the summer. May assist with trait and variable measurement of invasion studies within the Climate Experiment.	8/31/12	$10,400
6. Summer 2012: Graduate student continues to analyze invasion data and provide guidance to the interns working on the experiments.	8/31/12	$8,700

Result Completion Date: July 1, 2013

Result Status as of *(*August 2010*)*:

Result Status as of *(*February 2011*)***:**

Result Status as of *(*August 2011*)***:**

Result Status as of *(*February 2012*)***:**

Result Status as of *(*August 2012*)***:**

Result Status as of *(*February 2013*)***:**

Final Report Summary:

RESULT/ACTIVITY 3: Effects of Agricultural Inputs and Warming on Soil Carbon

Description: The amount of soil carbon stored in or lost from soils depends on the balance between the carbon sequestered through plant growth and the carbon lost through microbial decomposition in the soil. Chemical analyses of plant biomass and soil samples can be used to characterize these dynamics for a given locale. Carbon content of soil and of above- and below-ground plant biomass provides an estimate of the total carbon within the current system. The changes in these quantities indicate if the soil is releasing or sequestering carbon and nitrogen. Prairie grasslands tend to be limited in their growth by nitrogen, so the concentration of biologically available nitrogen (nitrate and ammonium) in the soil indicates the potential for plant growth. The nitrogen mineralization analyses indicate how quickly microbes are able to convert dead or dying plant material into biologically available nitrogen, which has feed-back effects on plant growth. In addition, we will directly monitor the amount of carbon released from the soil by decomposition (referred to as soil respiration). By compiling these analyses with the data collected for Result 1, we will be able to determine the reasons why warming, fertilization or irrigation may impact the carbon balance.

Summary Budget Information for Result/Activity 3:

ENRTF Budget:	**$71,000**
Amount Spent:	**$ 0**
Balance:	**$71,000**

Deliverable/Outcome	Completion Date	Budget
1. Summer 2010: Interns (2 undergraduates) collect and prepare 3186 soil/root samples for analysis and measure soil respiration in the field monthly.	8/31/10	$7,200
4. Summer 2010: Graduate student begins analyses with previous biomass and soil carbon and nitrogen data, and collects background information from other studies.	8/31/10	$7,700
2. Winter 2010: Chemical analysis of soil and below- and above-ground biomass samples (3396 samples total - aboveground biomass samples collected as part of Result 1).	2/31/11	$9,000
3. Summer 2011: Interns (2 undergraduates) collect and prepare 2346 soil/root samples for analysis and measure soil respiration in the field monthly.	8/31/11	$7,500
7. Summer 2011: Graduate student continues analyses with the 2010 biomass and soil carbon and nitrogen data.	8/31/11	$8,100
5. Winter 2011: Chemical analysis of soil and below- and above-ground biomass samples (2556 samples total – aboveground biomass samples collected as part of Result 1).	2/31/12	$6,000
6. Summer 2012: Interns (2 undergraduates) collect and prepare 3186 soil/root samples for	8/31/12	$7,800

analysis and measure soil respiration in the field monthly..		
3. Summer 2012: Graduate student continues analyses with the 2011 biomass and soil carbon and nitrogen data.	8/31/12	$8,700
7. Winter 2012: Chemical analysis of soil and below- and above-ground biomass samples (3396 samples total – aboveground biomass samples collected as part of Result 1).	2/31/13	$9,000

Result Completion Date: July 1, 2013

Result Status as of *(*August 2010*)*:

Result Status as of *(*February 2011*)***:**

Result Status as of *(*August 2011*)***:**

Result Status as of *(*February 2012*)***:**

Result Status as of *(*August 2012*)***:**

Result Status as of *(*February 2013*)***:**

Final Report Summary:

RESULT/ACTIVITY 4: Sustainable Restoration Practices

Description: Results 1-3 will be synthesized to report the optimal methods for combining biofuel production, carbon storage, and habitat restoration.

Summary Budget Information for Result/Activity 4:

ENRTF Budget: $43,000
Amount Spent: $ 0
Balance: $43,000

Deliverable/Outcome	Completion Date	Budget
Winter 2010: Dr. Lehman begins synthesis of biofuel restoration sustainability	2/31/11	$9,300
Winter 2010: Research assistant (1 undergraduate) works with Dr. Lehman, assisting with research and data analysis	2/31/11	$4,800
Winter 2011: Dr. Lehman continues synthesis of biofuel restoration sustainability	2/31/12	$9,300
Winter 2010: Research assistant (1 undergraduate) works with Dr. Lehman, assisting with research and data analysis	2/31/12	$5,000

Winter 2011: Dr. Lehman continues synthesis of biofuel restoration sustainability	2/31/13	$9,400
Winter 2011: Research assistant (1 undergraduate) works with Dr. Lehman, assisting with research and data analysis	2/31/13	$5,200

Result Completion Date: July 1, 2013

Result Status as of *(*August 2010*)*:

Result Status as of *(*February 2011*)***:**

Result Status as of *(*August 2011*)***:**

Result Status as of *(*February 2012*)***:**

Result Status as of *(*August 2012*)***:**

Result Status as of *(*February 2013*)***:**

Final Report Summary:

V. TOTAL ENRTF PROJECT BUDGET: *$221,000*

Personnel: $197,000
Additional Budget Items: $24,000

Breakdown of Additional Budget Items: Chemical Analyses

Analyses *	**Plant Carbon-Nitrogen Content**	**Soil Carbon-Nitrogen Content**	**Soil Active Nitrogen Content**	**Soil Nitrogen Mineralization Rate**
Experiment(s)	*Climate & Management*	*Climate & Management*	*Climate & Management*	*Climate only*
Plots	210	210	210	114
Samples required per analysis	1	1	2	2
Number of depths analyzed	**2	2	2	1
Replicates per plot	1	2	2	1
Measurements per year	1	1	1	***2
Number of years measured	3	2	3	3
Total samples	1260	1680	5040	1368
Estimated price per sample	$4	$4	$2	$2
Subtotals	$5040	$6720	$10080	$2736

Total	****$24,576

* Analyses will be performed by the Ecosystem Analysis Lab at the University of Nebraska-Lincoln. The Cedar Creek LTER has a long-standing partnership with the EAL to obtain laboratory services unavailable at the University of Minnesota.

** Analyses of above- and below-ground plant biomass

*** Samples taken in spring and fall

**** To balance the budget, only 1080 Soil Nitrogen Mineralization analyses will be paid for by this LCCMR grant ($2160); the remaining 288 ($576) will be covered by other funds (see Section VI.C).

VI. PROJECT STRATEGY:

A. Project Partners:

Dr. David Tilman (Regents' Professor and Director of Cedar Creek Ecosystem Science Reserve, U of M) will lead the research on sustainability and on effects of warming and inputs on invasion by exotic plant species (time donated in kind). Dr. Jennifer Powers (Assistant Professor, U of M) will lead work on soil carbon and nitrogen dynamics and will supervise graduate students and interns (time donated in kind). Dr. Clarence Lehman (Adjunct Faculty, U of M) will lead the synthesis of biofuel and restoration sustainability ($28,000).

B. Project Impact and Long-term Strategy:

The funds requested here are essential to allow the study of the sustainability, carbon and nitrogen dynamics, and susceptibility to invasion of the grassland biofuel ecosystems established at Cedar Creek Ecosystem Science Reserve. The scale of the prairie research field experiments located at Cedar Creek are unparalleled, both in replication and long-term research capability. These attributes are essential to gaining meaningful insights into the potential impacts of prairie biofuel agriculture and, in doing so, setting standards of analysis for other biofuel industries. Such research at Cedar Creek has been supported for the past 20 years by Dr. Tilman's continuing Long Term Ecological Research (LTER) grants from the National Science Foundation, grants provided to support basic ecological research on ecosystem functioning. Funds from LCCMR and earlier funds from the University of Minnesota's Initiative for Renewable Energy and the Environment (now fully expended) are allowing us to apply the fundamental advances in ecological science we have achieved to societally relevant issues, particularly the production of biofuels from ecosystems that offer society multiple, simultaneous energetic and environmental benefits. It is thus our strategy to continue to combine support from NSF for basic research with support from LCCMR for more applied research to develop new ways of producing biofuels that help restore prairie grassland ecosystems in Minnesota, that use soils as a site for significant

sequestration of carbon, that help improve water quality in agricultural ecosystems, and that provide a significant and sustainable new source of energy for our society,

C. Other Funds Proposed to be Spent during the Project Period:

Each year the National Science Foundation provides Cedar Creek researchers with about $800,000 in funding for our long-term ecological research projects. We used these NSF funds to establish our large Biodiversity Experiment and our Climate Experiment, both of which are the basis for the work we propose to LCCMR. NSF will continue to pay the majority of the costs associated with the operation and maintenance of these experiments, however LCCMR funds allow us to gather additional data, and to do some additional experiments (such as the watering and fertilization experiment and the invasion experiment) that would not be possible with our NSF funds. Thus, in essence, LCCMR support is providing the marginal, additional expenses needed to address the major but more applied issues articulated in our proposal, and does so by building on the expensive infrastructure that is already supported by the National Science Foundation.

D. Spending HIstory:

At present, the LTER grant alone provides approximately $800,000 annually for its research projects based at Cedar Creek, approximately $300,000 of which are allocated to Drs. Tilman and Powers. The infrastructure of the warming experiment, including buried wiring, circuit boxes, and heat lamps, was established in 2008 and 2009 with about $120,000 in LTER funds. Sampling of the 150 control plots for this research has been supported for the past 15 years by the same grant.

VII. DISSEMINATION:

As detailed by our deliverables, we plan to report our results with at least eight scientific papers published in high-impact peer-review journals.

Data collected with the support of this LCCMR grant will be included in the database supported by the Cedar Creek LTER grant and managed and distributed in accordance with LTER requirements. All such LTER data is required to be published within four years of its collection on Cedar Creek's publically accessible website: http://www.lter.umn.edu/research/data

In addition, because these results are likely to be of great interest to general audiences concerned with biofuel production in Minnesota and nationally, we will make it a priority to communicate our findings in public presentations and through direct contact with legislators, other government bodies, agricultural organizations, researchers in the field, farmers, energy businesses and the general public.

VIII. REPORTING REQUIREMENTS: **Periodic work program progress reports will be submitted not later than** August 2010, February 2011, August 2011, February 2012, August 2012, February 2013

IX. RESEARCH PROJECTS:
(See Peer Review Addendum)

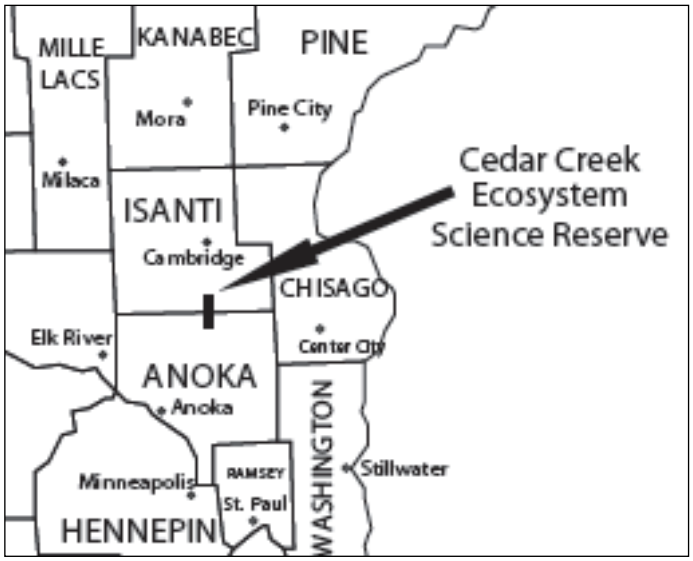

Figure 1 - Map of research location

Attachment A: Budget Detail for 2010 Projects - Summary and a Budget page for each partner (if applicable)

Project Title: Sustainable Biofuels

Project Manager Name: *G. David Tilman*

Trust Fund Appropriation: $ *221,000*

2010 Trust Fund Budget	**Result 1 Budget:**	**Amount Spent** *(11/24/09)*	**Balance** *(11/24/09)*	**Result 2 Budget:**	**Amount Spent** *(11/24/09)*	**Balance** *(11/24/09)*	**Result 3 Budget:**	**Amount Spent** *(11/24/09)*	**Balance** *(11/24/09)*	**Result 4 Budget:**	**Amount Spent** *(11/24/09)*	**Balance** *(11/24/09)*	**TOTAL BUDGET**	**TOTAL BALANCE**
	Effects of Warming and Agricultural Inputs on Biomass Production and Sustainability			*Warming and Input Effects on Invasions by Exotic Species*			*Effects of Warming and Agricultural Inputs on Soil Carbon*			*Sustainable Restoration Practices*				
PERSONNEL: Undergraduate Interns (8/summer) Average $5005 ($4580 salary + $425 fringe) per intern per summer	52,500	0	52,500	30,000	0	30,000	22,500	0	22,500	15,000	0	15,000	120,000	120,000
PERSONNEL: Graduate Students (2/summer) Average $8235 ($6500 salaray + $1625 fringe) per student per summer	0	0	0	24,500	0	24,500	24,500	0	24,500	0	0	0	49,000	49,000
PERSONNEL: Clarence Lehman (part-time) $9260 ($7000 salary + $2260 fringe) per year	0	0	0	0	0	0	0	0	0	28,000	0	28,000	28,000	28,000
OTHER: Chemical Analyses (no. of samples) Plant Carbon-Nitrogen Content (1260) - $5040 Soil Carbon-Nitrogen Content (1680) - $6720 Soil Active Nitrogen Content (5040) - $10080 Soil Nitrogen Mineralization (1080) - $2160	0	0	0	0	0	0	24,000	0	24,000	0	0	0	24,000	24,000
COLUMN TOTAL	**$52,500**	**$0**	**$52,500**	**$54,500**	**$0**	**$54,500**	**$71,000**	**$0**	**$71,000**	**$43,000**	**$0**	**$43,000**	**$221,000**	**$221,000**