

## 2017 Project Abstract

For the Period Ending June 30, 2021

### **PROJECT TITLE: Preserving Minnesota Prairie Plant Diversity – Phase II**

**PROJECT MANAGERS:** Ruth G. Shaw, Georgiana May

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**FUNDING SOURCE:** Environment and Natural Resources Trust Fund

**LEGAL CITATION:** M.L. 2017, Chp. 96, Sec. 2, Subd. 03c as extended by M.L. 2020, First Special Session, Chp. 4, Sec. 2

**APPROPRIATION AMOUNT:** \$900,000

**AMOUNT SPENT:** \$900,000

**AMOUNT REMAINING:** \$0

### **Sound bite of Project Outcomes and Results**

We gathered seeds of prairie plants and shared them with producers who are expanding seed availability for restorations. We collected, identified and studied many microbes that prairie plants harbor, documenting their effects on their hosts. Our experiments have clarified the geographic scale of plant adaptation and genetics underlying ongoing adaptation.

### **Overall Project Outcome and Results**

Minnesota prairies harbor extraordinary diversity of plants and microbes, while also nurturing wildlife, retaining water and topsoil, and beautifying landscapes. Yet habitat loss threatens the persistence of the once vast prairies and their stunning biotic diversity. Limited understanding of this diversity and insufficient seed availability hinder sustainable management of this iconic Minnesota biome. We conducted Healthy Prairies (HP) Phase II to expand availability of seeds for prairie restorations and study approaches to increase success of restorations. Building on our prior accomplishments under ENRTF funding, we have:

1. Preserved diverse seed from 57 rarer prairie species, gathering them from widely separated locations.
2. Obtained, archived, and studied 2,600 naturally occurring microbial partners from two species.
3. Gathered data to assess the geographic scale important to plant survival and reproduction in MN.

Our extensive collections of source-identified seeds and microbes across a wide range of MN's prairie region help to conserve the diversity of MN prairies. We have provided seeds to seed producers, who have, in turn, used them in establishing fields and are seeking certification of the seeds that they obtain from them.

Our studies of effects of microbial associates on prairie plants have indicated that the bacteria providing nitrogen to prairie clover (*Dalea purpurea*, *D. candida*) disperse widely across MN prairies. Consequently, we can recommend to growers an inoculum that need not be site-specific. In contrast, the communities of fungi associated with roots of *S. scoparium* are spatially restricted, indicating that a regionally-based inoculum may be preferable.

We continued our large-scale experiment to elucidate the geographic scale of adaptation of six prairie species. We gathered extensive data from this experiment and began analyses of the data. We implemented experiments to investigate genetic structure of two populations of little bluestem (*Schizachyrium scoparium*), including genetic variance for fitness and the fitness consequences of inbreeding and of crossing between populations.

### **Project Results Use and Dissemination**

HP team members have participated in varied opportunities to disseminate findings from this project. These include informal events to communicate with members of the public who are not all well-versed in science and may not be aware of prairies (Market Science), as well as workshops involving other scientists and land managers (Nature Conservancy 'Science Slams', Local Adaptation Workshop, held at UM-TC, March 2019, discussions of seed sourcing guidelines led by staff of MN DNR).

A paper providing an overview of the Local Adaptation Workshop has been published in *New Phytologist* (2020) 225:2246–2248. A manuscript reporting findings about geographic scale of local adaptation has been submitted to *Restoration Ecology* and has received positive reviews. A second manuscript reporting on a study that used focus groups to identify impediments to use of source-identified seeds for prairie restorations has been submitted to *Restoration Ecology* and has received positive reviews. Both manuscripts are under revision and will be resubmitted soon.



## Environment and Natural Resources Trust Fund (ENRTF)

### M.L. 2017 LCCMR Work Plan Final Report

**Date of Submission:** March 17, 2021

**Final Report**

**Date of Work Plan Approval:** 06/07/2017

**Project Completion Date:** June 30, 2021

**Does this submission include an amendment request?** Yes

**PROJECT TITLE:** Preserving Minnesota Prairie Plant Diversity – Phase II

**Project Managers:** Ruth G. Shaw, Georgiana May

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**Location:** WCentral, SE,SW,NW

**Total ENRTF Project Budget:**

<b>ENRTF Appropriation:</b>	<b>\$900,000</b>
<b>Amount Spent:</b>	<b>\$900,000</b>
<b>Balance:</b>	<b>\$0</b>

**Legal Citation:** M.L. 2017, Chp. 96, Sec. 2, Subd. 03c as extended by M.L. 2020, First Special Session, Chp. 4, Sec. 2

**Appropriation Language:**

\$900,000 the first year is from the trust fund to the Board of Regents of the University of Minnesota to continue collecting and preserving germplasm of plants throughout Minnesota's prairie region, study the microbial effects that promote plant health, analyze local adaptation, and evaluate the adaptive capacity of prairie plant populations. This appropriation is available until June 30, 2020, by which time the project must be completed and final products delivered.



## **Environment and Natural Resources Trust Fund (ENRTF)**

### **M.L. 2017 LCCMR Work Plan**

M.L. 2020 - Sec. 2. ENVIRONMENT AND NATURAL RESOURCES TRUST FUND; EXTENSIONS. [to June 30, 2021]

## **I. PROJECT TITLE: Healthy Prairies II: Preserving Prairie Plant Diversity**

### **II. PROJECT STATEMENT:**

Minnesota prairies harbor an extraordinary diversity of plant and microbial life, while also nurturing wildlife, retaining water and topsoil, and beautifying rural landscapes. Yet habitat loss and environmental variability threaten the persistence of the once immense prairie landscape and its stunning biotic diversity. Moreover, limited understanding of this diversity and insufficient seed availability hinder cost-effective and sustainable management of this iconic Minnesota biome.

Healthy Prairies (HP) Phase II will build on the accomplishments under current funding (2014-2017). Our team and volunteers spent over 1000 hours scouting 27 prairie remnants and cataloging locations over MN prairie regions for 40 of the more common and widespread native prairie species. We collected seed from thousands of individuals, retaining extensive genetic variation while tracking locality. For experimental work, we have cultured over 5000 plant-associated microbes. We established seed-increase plots for 6 plant species (from 12 sites) and used these in experimental plantings at three locations spanning the latitudinal range of MN prairies. To realize this investment in the preservation of MN prairie plant diversity, while providing essential resources and information for prairie restoration, we will:

- Preserve diverse seed from 20 of the rarer prairie species.
- Obtain and maintain cultures of an additional 5000 naturally occurring microbial partners for grasses.
- Determine the geographic scale important to plant survival and reproduction in a varying environment.

Four major MN geographic regions across the native prairie will be served. Providing locally-sourced seed, the project will help restore and conserve the diversity of MN prairies and their associated wildlife, pollinator and microbial diversity.

### **III. OVERALL PROJECT STATUS UPDATES:**

#### **Amendment Request (8/14/17):**

We request approval of the attached budget and the Summary Budgets for each Activity in this workplan. These items adjust our proposed budget to the amount allocated (\$900,000).

We also request approval for an additional item under the "Other" line, Activity 1, on the attached budget, namely "Fees for independent contractors collecting seeds". Utilizing services offered by professional native-seed collectors in distant parts of Minnesota will result in more efficient, cost-effective, and diverse seed collection.

Finally, we request approval of revised deadlines for Project Status updates. The previous version of this workplan included due dates of Dec 1, 2018, Dec 1, 2019, and Dec. 1, 2020. These appear to be errors; we have changed these dates to Dec. 1, 2017, Dec. 1, 2018, and Dec. 1, 2019.

#### **Amendment Approved by LCCMR 8/14/2017**

**Project Status as of Dec. 1, 2017:** Report deferred per communication from LCCMR staff on July 24, 2017.

#### **Project Status as of June 1, 2018:**

We have staffed the project with highly capable individuals who have the necessary expertise to collect and conserve seeds, carry out the experiments, and maintain the research infrastructure established during Healthy Prairies I (July 2014-June 2017). We have scouted seed collection sites, obtained necessary permits, and are collaborating with partners and citizen groups to obtain collections of seeds that represent much of the geographic extent of MN prairie, as well as the genetic variability of target species. We are currently installing field experiments to test conclusions reached during Healthy Prairies I; namely, that use of seed sourced at the regional scale (e.g., southwestern MN) could improve prairie restoration outcomes and that there is sufficient variation in the rhizobial populations within sites to support healthy growth of prairie clovers (*Dalea* species). In July 2017, we installed 800 individuals from 2 populations of little bluestem into a field site in St. Paul; these individuals form the basis of our experiment on the adaptive capacity of these populations. In fall 2017, we collected data and samples related to local adaptation from over 7,000 individuals of 6 species at our 3 field experiments; data analysis is proceeding. We are maintaining the field experiments installed during Healthy

Prairies I that assess the geographic scale of local adaptation in perennial plant species and evaluate the extent of adaptive potential to environmental change. Additional data and samples will be collected from these experiments in July – October 2018.

**Amendment request (June 1, 2018)**

We request approval for transfer of funds as follows: \$10,000 from Activity 1 Personnel, \$4,000 from Activity 2 Personnel, and \$3,000 from Activity 3 Equipment/Tools/Supplies to cover previous and anticipated future Activity 3 Travel costs. We will be working with citizen groups and external partners to meet our Activity 1 goals despite this decrease in the Personnel line. Similarly, we have recruited undergraduate employees to help us meet Activity 2 goals more cost-efficiently than anticipated. Activity 3 Equipment costs are lower than expected, as we have been able to use materials purchased under Healthy Prairies Phase I to a greater degree than we anticipated.

**Amendment Approved by LCCMR 6/7/2018**

**Project Status as of January 18, 2019:**

With the help of volunteers and UM-Morris undergraduates and in cooperation with The Nature Conservancy, we made substantial collections of 34 species of prairie plants from 23 sites in 9 counties in western and south-central Minnesota. We are in early stages of conveying these seeds to native seed producers, for use in establishing seed-increase populations to support prairie restoration. We completed a field experiment with prairie clovers (*Dalea* spp.) finding that when seed is sourced closer to the planting site, the rate of association with beneficial nitrogen-fixing rhizobia is increased. We are currently designing a greenhouse experiment to test whether beneficial bacteria from local plant populations results in healthier plants. During summer 2018, 2000 new microbes were collected from little bluestem (*Schizachyrium scoparium*) with the plan to test their effect on plant drought resistance. We also collected pedigreed seed from 400 little bluestem plants at our field site in St. Paul; these seeds will form the basis of greenhouse and field experiments in 2019. In fall 2018, we collected data from over 7,000 individuals of 6 species at our 3 field sites in western Minnesota. These data are being jointly analyzed with data collected in 2015-17.

**Amendment request (January 18, 2019)**

We request approval for transfer of funds as follows: \$2,000 from Activity 1 Travel, \$4,000 from Activity 1 Other, and \$3,000 from Activity 3 Other to cover anticipated future Activity 3 Travel costs. We will be transitioning our Activity 1 efforts from collection of seed to distribution, requiring less travel money and maximizing efficiency of the time of employees working on both Activities 1 and 3. As Activity 3 requires little new infrastructure at this stage, resources in the “Other” line may now be used for travel expenses.

**Amendment Approved by LCCMR 2/1/2019**

**Project Status as of June 1, 2019**

We continue to make strong progress toward the project goals, which are on-track for completion June 2020. Our goal of enhancing availability of source-identified seed for prairie restoration advanced through drafting of material transfer agreements (MTAs) and related documents, which we circulated to select native seed producers, revised in response to their comments, and re-issued to additional external partners for further input. The 2019 seed collection plan has been finalized; staff and collaborators have been recruited to harvest seeds at 14 sites in 7 western MN counties.

Results from an Activity 2 field experiment with prairie clovers (*Dalea* sp.) and beneficial nitrogen-fixing bacteria show that plant and bacterial growth responses are predicted both by spatial distance to the source and differences specific to source sites. A greenhouse experiment to evaluate the impacts of differing combinations of plant and bacteria from the same and different source sites is in progress. Diverse fungi have been isolated from little bluestem (*Schizachyrium scoparium*) in conjunction with quantitative genetic studies of the host grass to evaluate its capacity for ongoing evolutionary adaptation. A greenhouse study to evaluate the contribution of

these symbiotic fungi to drought tolerance of host plants is planned. For Activity 3, entry of data collected during the 2018 censuses at the 3 field sites in western Minnesota has been completed. Joint analysis of data from 2015-2018 is completed for thimbleweed (*Anemone cylindrica*) and is underway for the other 5 species. Preliminary results suggest local adaptation of southwest-origin populations of *D. candida*; size declines with northward distance from their origin. At this early stage of our experiments, we do not detect local adaptation in the other 5 species. *Outreach and dissemination*: Our findings were presented at a workshop on local adaptation attended by staff from BWSR, DNR, The Nature Conservancy, USFW, and USGS, as well as researchers from UM-TC, UM-Duluth, UM-Morris, NDSU, SDSU, the Chicago Botanic Garden, the U. of Missouri-Columbia, and Michigan State U.

#### **Project Status as of Dec. 1, 2019**

- Activity 1: We successfully transferred 77 collections of seed from 41 native prairie species to four native-seed producers and as we continue these outreach efforts, source-identified seed for use in Minnesota prairie restorations will be increasingly available. In 2019, seed collections from 40 species (seven entirely new) across 15 sites in seven western Minnesota counties were made.
- Activity 2: Results of field studies show that prairie clovers (*Dalea* species) are adapted to a broad range of local conditions and that the critical seedling-establishment phase depends on the availability of beneficial nitrogen-fixing bacteria. Our results suggest that the addition of beneficial bacteria from regional sources to restoration sites will increase establishment and pose few risks.
- Activity 3: Data were collected from 7,000+ individuals of 6 species at our 3 outstate field sites. Results to date suggest that like *Dalea* spp above, these slow-growing perennial species, are able to survive a broad range of environmental conditions. Further, our genetic analysis of variation in little bluestem (*Schizachyrium scoparium*), suggest significant genetic variation in the capacity of populations to adapt to new environments. *Together* our results to date suggest that prairie plants have the capacity to adapt to new environments in which they find themselves (e.g. restorations) and emphasize the critical importance of both maintaining diversity and expanding seed resources.

#### **Amendment request (December 10, 2019)**

We request approval to transfer \$2,432 from Activity 1 Other to cover unanticipated expenses as follows: \$15 (Activity 1, Equipment/Tools/Supplies), \$738 (Activity 1, Travel) and \$1,679 (Activity 3, Travel). Because the Project Completion Date falls well before the height of the seed-collection season, we expect to minimally engage the services of our collection partners during the remainder of this Work Plan; the Activity 1 Other funds can thus be reallocated without compromising project goals.

**Project extended to June 30, 2021 by LCCMR 6/18/20** as a result of M.L. 2020, First Special Session, Chp. 4, Sec. 2, legislative extension criteria being met.

#### **Project Status as of July 1, 2020:**

- **Activity 1:** Since December, 2019, the Healthy Prairies Project has distributed seed to two seed producers. Due to COVID restrictions and uncertainty in funding, seed collections planned for Spring and early Summer 2020 have been rescheduled for the same period in 2021.
- **Activity 2:** We obtained over 7000 cultures for fungi potentially beneficial to little bluestem (LBS) (*Schizachyrium scoparium*) from prairie sites across a water availability gradient from Western to Eastern MN. We used DNA sequencing to identify these and preliminary analyses show that these fungal communities associated with LBS may be responsive to drought. For this project, we were able to complete Outcome 2 but not Outcome 3 before COVID delays. The status for the project with prairie clovers (*Dalea* sp.) is the same as reported Dec. 1, 2019 and a publication is in preparation by Dr. Pozzi. However, Outcome 3 in which we planned to evaluate the role of beneficial microbes in *Dalea*, and LBS

early seedling establishment and survival could not be carried out due to COVID restrictions, and have been rescheduled to greenhouse studies (LBS) Winter 2021, and local field studies (Dalea) Spring 2021.

- **Activity 3:** Analysis of data has continued. Plans have been developed for field research in August. This planning has included design of experiments to evaluate effects of inbreeding and of crossing between populations of little bluestem plants (*Schizachyrium scoparium*).

#### **Project Status as of February 1, 2021:**

- **Activity 1:** Due to COVID restrictions and uncertainty in funding, seed collections intended to be completed in late summer and fall of 2020 could not be made.
- **Activity 2:** Graduate student Cedric Ndinga-Muniania identified over 1200 fungal cultures obtained from roots of little bluestem. In addition, he has shown that these beneficial fungal communities change across a gradient of water availability from western to eastern MN using Next-Generation sequencing methods. We obtained additional funding from the U. Minnesota to allow Mr. Muniania to screen these fungi for beneficial effects on plant growth in drought conditions during Winter 2021.
- **Activity 3:** Through July and August, graduate student Wes Braker carried out experimental crosses between little bluestem plants (*Schizachyrium scoparium*) growing in field plants on the St. Paul campus of UMN-TC. Mr. Braker gathered the resulting seeds and has proceeded to prepare them for germination to evaluate their viability. A small team (Shaw, May, Braker, and technician Em Daily) traveled in mid-August to the Lake Bella field site to gather data on the plants in the experimental garden there, taking extreme precautions against Covid-19 and completing the census in four days. Data have been prepared for analysis.

#### **Amendment request (March 17, 2021)**

We request approval to transfer \$1,056 from Activity 2 Other and \$467 from Activity 2 Travel to Activity 2 Personnel (\$132) and to Activity 2 Equipment/Tools/Supplies (\$1,391). These small shifts are needed to cover expenses for lab supplies and personnel involved in this activity. The pandemic led to reductions in travel and greenhouse use from the original plan.

#### **Amendment Approved by LCCMR on 4/1/2021**

#### **Overall Project Outcomes and Results: through February 1, 2021**

Minnesota prairies harbor extraordinary diversity of plants and microbes, while also nurturing wildlife, retaining water and topsoil, and beautifying landscapes. Yet habitat loss threatens the persistence of the once vast prairies and their stunning biotic diversity. Limited understanding of this diversity and insufficient seed availability hinder sustainable management of this iconic Minnesota biome. We conducted Healthy Prairies (HP) Phase II to expand availability of seeds for prairie restorations and study approaches to increase success of restorations. Building on our prior accomplishments under ENRTF funding, we have:

1. Preserved diverse seed from 57 rarer prairie species, gathering them from widely separated locations.
2. Obtained, archived, and studied 2,600 naturally occurring microbial partners from two species.
3. Gathered data to assess the geographic scale important to plant survival and reproduction in MN.

Our extensive collections of source-identified seeds and microbes across a wide range of MN's prairie region help to conserve the diversity of MN prairies. We have provided seeds to seed producers, who have, in turn, used them in establishing fields and are seeking certification of the seeds that they obtain from them.

Our studies of effects of microbial associates on prairie plants have indicated that the bacteria providing nitrogen to prairie clover (*Dalea purpurea*, *D. candida*) disperse widely across MN prairies. Consequently, we can recommend to growers an inoculum that need not be site-specific. In contrast, the communities of fungi



associated with roots of *S. scoparium* are spatially restricted, indicating that a regionally-based inoculum may be preferable.

We continued our large-scale experiment to elucidate the geographic scale of adaptation of six prairie species. We gathered extensive data from this experiment and began analyses of the data. We implemented experiments to investigate genetic structure of two populations of little bluestem (*Schizachyrium scoparium*), including genetic variance for fitness and the fitness consequences of inbreeding and of crossing between populations.

#### IV. PROJECT ACTIVITIES AND OUTCOMES:

##### ACTIVITY 1: Preserving prairie plant diversity for conservation and restoration

**Description:** We will increase availability of source-identified seed for use in MN prairie restorations by working with partners to increase seed collection, distribution, and to develop transfer agreements. Twenty of the less common but important prairie species, in addition to the 40 species obtained in 2014-17, are targeted, and these will entail greater time and scouting to collect. Efforts will be evaluated via the amount and diversity of seed collected and by the level and quality of partner involvement.

##### Summary Budget Information for Activity 1:

<b>Revised ENRTF Budget:</b>	<b>\$102,821</b>
<b>Amount Spent:</b>	<b>\$118,374</b>
<b>Balance:</b>	<b>-\$15,553</b>

Outcome	Completion Date
1. Increase availability of diverse, source-identified seed for prairie restorations by expanding our network of collectors and collection locations. Collect seed for 20 additional, relatively common species.	October, 2019
2. Implement material transfer agreements with producers.	December, 2019
3. Collect source-identified seed for 20 rarer prairie plant species. Deposit voucher specimens at UM herbaria, deposit seed at USDA facility for long-term storage, transfer seed to producers.	June, 2020

**Activity 1 Status as of Dec. 1, 2017:** Report deferred per communication from LCCMR staff on July 24, 2017.

##### Activity 1 Status as of June 1, 2018:

By leveraging our support from LCCMR, we were able to obtain funding from the University of Minnesota's Institute on the Environment to conduct a series of focus groups with native seed producers and consumers. One outcome of these conversations was a list of less-common but important prairie species of which producers are interested in developing commercially viable, source-identified populations for use in restorations. We developed our list of target species for 2018 collections by combining the producers' suggestions with input from other collaborators. In addition to Dr. Kuchenreuther at UMM, who collected populations in west central MN in 2017, we have partnered with a seed collector based in northwestern Minnesota; by subcontracting to him seed collections in that region, we can make most efficient use of project resources. In 2017, he collected seeds of 19 species from populations in NW MN. In addition, we have established a partnership with the Minnesota Master Naturalists in anticipation of multiple seed collection events scheduled for June – October 2018. We have completed collection of one early-flowering species, pasque flower.

##### Activity 1 Status as of January 18, 2019:

We have produced a preliminary report summarizing and analyzing the findings of the focus groups; this report has been submitted to focus group participants for their review and further revision. We met our 2018 seed collection goals: through the combined efforts of Margaret Kuchenreuther, 2 UM-Morris undergraduates, 9 community volunteers, and Healthy Prairies staff, we collected 34 species from 21 sites in south-central and west-central MN. In addition, we again partnered with a seed collector based in northwestern MN, allowing us

to make efficient use of project resources. In 2018, he collected 8 species from an additional 2 sites. We are in the process of drafting material transfer agreements that will facilitate conveyance of these seeds to native-seed producers, who will use them to establish seed-increase fields from which additional generations of seeds can be harvested and used for prairie restoration in MN.

**Activity 1 Status as of June 1, 2019:**

Recent Ph.D. recipient Nicholas Goldsmith has prepared a report on the results of the focus groups for submission to *Restoration Ecology* in June 2019. With the collaboration of Dr. Goldsmith, we drafted material transfer agreements (MTAs) that were informed by the focus group report and reviewed by external partners. We have distributed the draft MTAs and associated documents to other partners for additional feedback. We finalized the 2019 seed collection plan; in this last full collection season under current funding, we aim to supplement and broaden geographic representation of collections from previous years as well as to increase the number of species available for conveyance to seed producers. Three UM-Morris undergraduates supervised by Dr. Kuchenreuther, and a collaborator in northwest Minnesota have been recruited to support our goal of collecting 38 species from 14 sites in 7 western MN counties.

**Activity 1 Status as of Dec. 1, 2019:**

The manuscript on the results of the focus groups was submitted to *Restoration Ecology* and is currently in revision. Material transfer agreements and associated documents were finalized in September 2019. Using these documents, we executed agreements with 4 native seed producers, resulting in the transfer of 77 seed lots representing 41 species collected from 9 counties in western Minnesota. Producers will use these seed lots to establish commercial-scale, source-identified populations that will supply geographically and genetically diverse seed to Minnesota prairie restorations. We are in the process of contacting additional producers with the aim of disbursing additional seed lots. The 2019 seed collection team (3 undergraduates from UM-Morris, 2 undergraduates from UM-Twin Cities, Dr. Kuchenreuther, and a collaborator from northwest Minnesota) collected seeds from 40 species (7 of them new to the Healthy Prairies inventory) from 15 sites in 7 western Minnesota counties.

**Activity 1 Status as of July 1, 2020:**

Since December, 2019, the Healthy Prairies Project has distributed seed to two seed producers. Each of these producers responded to email contact about available seeds in the Healthy Prairies Project collection. The requests from each were fulfilled in full and mailed to the respective producers with no competing requests. As part of the permit that allows the Healthy Prairies Project to collect seeds on property that The Nature Conservancy owns and manages, ½ of all seed collected must be returned to The Nature Conservancy for their own use. In January 2020, the Healthy Prairies Project returned 57 species from 9 locations, with a total of 147 collections. These collections included seed that was collected in the summer of 2019 directly from Nature Conservancy sites as well as seed from plants that were originally collected as seed at Nature Conservancy sites and have been grown at the University of Minnesota as part of the Healthy Prairies Project. Due to COVID restrictions and uncertainty in funding, seed collections planned for Spring and early Summer 2020 have been rescheduled for the same period in 2021.

**Activity 1 Status as of February 1, 2021:**

Due to COVID restrictions and uncertainty in funding, seed collections planned for late Summer and Fall 2020 could not be made.

**Final Report Summary:**

By leveraging our support from LCCMR, we obtained funding from the University of Minnesota's Institute on the Environment to conduct a series of focus groups with native seed producers and consumers. One outcome of these conversations was a list of less-common but important prairie species of which producers are interested in

developing commercially viable, source-identified populations for use in restorations. Based on this and other considerations, we made extensive seed collections in a wide range of MN's prairie region. We have offered these collections to seed producers and distributed seed to those who requested them. These producers have established fields from these seeds and are seeking certification of the seeds that they obtain from them. In this way, this project will have substantively increased availability of source-identified seeds for use in prairie restorations in Minnesota.

## **Activity 2: Finding your friends in unlikely places – beneficial microbes for prairie plants**

**Description:** We will assess the diversity and effect of naturally occurring plant-associated microbes for two types of plants essential to healthy prairies – legumes and grasses. Results will inform land managers about the use of microbes to improve prairie plant establishment in restorations, a practice common in agriculture but not widely applied to natural systems.

<b>Summary Budget Information for Activity 2:</b>	<b>ENRTF Budget:</b>	<b>\$387,680</b>
	<b>Amount Spent:</b>	<b>\$352,561</b>
	<b>Balance:</b>	<b>\$35,119</b>

<b>Outcome</b>	<b>Completion Date</b>
<b>1.</b> <i>Use previously collected microbes to determine beneficial microbes' potential for enhancing prairie clover (Dalea spp.) survival and reproduction in experimental plantings and greenhouse studies.</i>	November, 2019
<b>2.</b> <i>Determine the diversity of microbial communities associated with little bluestem grass (Schizachyrium scoparium) and collect 5000 new microbes. Store living cultures at UM and USDA.</i>	December, 2019
<b>3.</b> <i>Determine effects of plant-associated microbes on little bluestem establishment and reproduction in experimental plantings and in greenhouse studies.</i>	June, 2020

### **Activity 2 Status as of Dec. 1, 2017:**

Report deferred per communication from LCCMR staff on July 24, 2017.

### **Activity 2 Status as of June 1, 2018:**

In Activity 2, we ask the role of microbial symbionts in the ability of prairie plants to live in the varied environments of MN prairies. Specifically, we are conducting experiments with *Dalea purpurea*, the legume purple prairie clover, and its nitrogen-fixing bacterial partner, rhizobium. Results of a greenhouse experiment and genotyping of rhizobium isolates from 15 prairie sites in MN in Phase I demonstrated that each site harbors extensive variation in rhizobia and that plants have growth patterns characteristic of sites and regions (Kane Keller, postdoc in Phase I). The results suggest that locally sourced seed (on a regional scale) could improve prairie restoration efforts and that there is sufficient variation in the rhizobial populations within sites to support healthy growth of *Dalea* plants. We are currently testing these conclusions with plantings of *Dalea* from different source sites into 2 of the 3 experimental sites established under Phase I of the Healthy Prairies project (Adrien Pozzi, postdoc Phase II). To accomplish goal 2 (above), Cedric Ndinga-Muniania (UM graduate student) will conduct extensive collections and culturing from little bluestem in summer 2018.

### **Activity 2 Status as of January 18, 2019:**

For Outcome 1, seed representing different populations of *Dalea purpurea* and *Dalea candida* were planted in 2 experimental sites established under Phase I of the Healthy Prairies project and were harvested in September 2018 (Adrien Pozzi, postdoc Phase II). Preliminary analyses of data for plant growth and the rate at which these plants associate with nitrogen-fixing bacterial partners suggest that locally sourced seed could favor beneficial

associations between plant and soil microbes. In addition, 600 new bacterial partners were cultured and will be used in a greenhouse experiment in Spring 2019 to test whether more beneficial associations and better plant growth result from matching source microbes and plants. For Outcome 2, Cedric Ndinga-Muniania (UM graduate student) collected and cultured over 2,000 new fungal endophytes from little bluestem collected in 5 remnant prairie sites across southern MN.

**Activity 2 Status as of June 1, 2019:**

For Outcome 1, field experiments conducted by postdoc Adrien Pozzi were completed Fall 2018. Results of analyses to date suggest that growth and survival of prairie clovers (*Dalea* spp.) and of beneficial nitrogen-fixing bacteria depend not only on distance to the source site, but also on source identity. A manuscript is being drafted. Greenhouse studies are underway to evaluate the impact of differing plant and bacterial source populations on plant growth (to be completed Fall 2019). For Outcome 2, fungal endophytes have been isolated from little bluestem (*Schizachyrium scoparium*) (UM graduate student Cedric Ndinga-Muniania). Results will inform differences in fungal communities in *S. scoparium* across a drought gradient (completion Dec 2019). For Outcome 3, Ndinga-Muniania will evaluate the fungi isolated under Outcome 2 for their tendency to confer drought tolerance to *S. scoparium* (greenhouse studies completed by June 2020).

**Activity 2 Status as of Dec. 1, 2019:**

Results from the research of Adrien Pozzi (postdoc) and the May Lab show that *Dalea purpurea* and *D. candida* populations are adapted to a broad range of local conditions. In addition, we find that the seedling establishment phase is critical and dependent on the availability of beneficial nitrogen-fixing bacteria in the soils. Along with previous information on the geographic scale of diversity in these bacteria, we conclude that there are few risks of adding beneficial bacteria to restorations sites from regional sources. We hypothesize that adding beneficial bacteria to soils at the time of seed germination will increase recruitment success in *Dalea* plantings. We plan to test this question in Spring 2020 using our collections of beneficial bacteria made under LCCMR funding.

**Activity 2 Status as of July 1, 2020:**

We obtained over 7000 cultures for fungi potentially beneficial to little bluestem (LBS) (*Schizachyrium scoparium*) from prairie sites across a water availability gradient from Western to Eastern MN (Ph.D. student Cedric Ndinga-Muniania). We used DNA sequencing to identify these and preliminary analyses show that these fungal communities associated with LBS may be responsive to drought. For this project, we were able to complete Outcome 2 but not Outcome 3 before COVID delays. The status for the project with prairie clovers (*Dalea* sp.) is the same as reported Dec. 1, 2019 and a publication is in preparation by Dr. Pozzi. However, Outcome 3 in which we planned to evaluate the role of beneficial microbes in *Dalea*, and LBS early seedling establishment and survival could not be carried out due to COVID restrictions, and have been rescheduled to greenhouse studies (LBS) Winter 2021, and local field studies (*Dalea*) Spring 2021.

**Activity 2 Status as of February 1, 2021:**

Graduate student Cedric Ndinga-Muniania identified over 1200 fungal cultures obtained from roots of little bluestem. In addition, he has shown that these beneficial fungal communities change across a gradient of water availability from western to eastern MN using Next-Generation sequencing methods. Given that our ENRTF funding is exhausted, we obtained funding from the U. Minnesota to allow Mr. Muniania to screen these fungi for beneficial effects on plant growth in drought conditions during Winter 2021.

**Final Report Summary:**

We accomplished the goals outlined above. Because we find that the bacteria providing nitrogen to prairie clover (*Dalea purpurea*, *D. candida*, legume) are widely dispersed across the varied environments of MN prairies, we can recommend an inoculum to growers that need not be site-specific. In contrast, the communities of fungi

associated with roots of *S. scoparium* are more spatially structured, and, pending the results of the above screens, it may be best to develop a regionally-based inoculum.

### ACTIVITY 3: Adaptive genetic diversity of prairie plants

**Description:** Continue field experiments established under Phase I to characterize the spatial scale of local adaptation for 6 prairie perennials. Evaluate genetic variation for survival and reproduction of little bluestem grass. Results will inform methods of prairie conservation and healthy prairie restoration that maintain diversity of prairie plant species.

#### Summary Budget Information for Activity 3:

**Revised ENRTF Budget:** **\$409,499**  
**Amount Spent:** **\$429,065**  
**Balance:** **-\$19,556**

Outcome	Completion Date
1. Monitor survival, growth, and reproduction in established experiments with 6 species and over 6000 plants to evaluate effect of seed source on establishment and success of prairie plants in restorations.	November 30, 2019
2. Plant pedigreed little bluestem seed into field experiments to assess its capacity to adapt to varied environmental conditions, and the role of microbes (identified in Activity 2) in that process.	November 30, 2019

**Activity 3 Status as of Dec. 1, 2017:** Report deferred per communication from LCCMR staff on July 24, 2017.

#### Activity 3 Status as of June 1, 2018:

To assess the adaptive capacity of little bluestem, which is a key component of upland prairies, we installed 800 little bluestem individuals into a field site in St. Paul in July 2017. These individuals will form the basis of formal genetic crosses to produce pedigreed seeds from which we can evaluate the extent of genetic variation that is present and could support the species' ongoing adaptation to environmental change. Also in fall 2017, as part of our effort to estimate the geographic scale of local adaptation in important prairie species, we collected data & samples from over 7,000 individuals of 6 species that were installed at 3 field sites during Healthy Prairies Phase I. 2017 was the 2<sup>nd</sup> year of data for 2 grass species and the 1<sup>st</sup> year of data for 2 forb and 2 legume species. Sample analyses (seed germination trials) were conducted in Dec 2017 – February 2018. Data analysis is under way. We are currently maintaining our field sites, including the common garden plots (Rosemount Research & Outreach Center) that provide seed for Activity 2 and Activity 3 experiments.

#### Activity 3 Status as of January 18, 2019:

In August 2018, we performed formal genetic crosses on 400 little bluestem plants at our field site in St. Paul. Seeds were harvested in October 2018; these will form the basis of greenhouse and field experiments to evaluate the extent of standing genetic variation. Also in fall 2018, we collected data from over 7,000 individuals of 6 species at our 3 field sites in western Minnesota. These data have been combined with data collected in 2015-2017 and analysis is under way. During May – August 2018, we continued maintenance of our common garden plots at the Rosemount Research and Outreach Center; these provide seed for Activity 2 and Activity 3 experiments. This work included installation of substantial fencing at and below ground level to deter gopher herbivory.

#### Activity 3 Status as of June 1, 2019:

To advance our assessment of local adaptation, we have completed entry of the 2018 census data collected on over 7,000 plants at our 3 field sites in western Minnesota. Analysis of all data collected to date for *Anemone cylindrica* is complete; joint analysis of data collected 2016-2018 is underway for the other 5 species. Preliminary

results suggest local adaptation of southwest-origin populations of *D. candida*; size declines with planting distance further northward from their origin. At this early stage in the lives of these long-lived species, we do not detect local adaptation in the other 5 species. We advanced our study of capacity for ongoing adaptation of little bluestem by establishing plantings of the pedigreed seed obtained from crosses during summer 2018. In early May, we initiated routine maintenance of our nursery plots at the Rosemount ROC. We have hired 2 highly qualified UM-TC undergraduates to assist with field and greenhouse work. A third UM-TC undergraduate will earn academic credit working with our team this summer.

**Activity 3 Status as of Dec. 1, 2019:**

We collected census data on 7,000 plants from our 6 focal species at our 3 field sites in western Minnesota. Entry of the 2019 data is complete. Joint analysis of the 2016-2019 data is under way. To date, we have not detected evidence of local adaptation in these slow-growing perennial species, which take several years to reach reproductive maturity. We maintained and collected data on seedlings that were established from seed produced by the 2018 crosses among little bluestem plants (*Schizachyrium scoparium*). In July 2019, we transplanted these 1,100 individuals into a field site in St. Paul. We also sowed additional seed produced by the 2018 crosses into an adjacent field site; this resulted in over 780 individuals on which we collected survival and morphological data. These data have been entered and analyzed. Results from the juvenile stage of this perennial grass indicate significant genetic, as opposed to environmental, influence on survival and differences between the two populations in adaptive capacity.

**Activity 3 Status as of July 1, 2020:**

Analysis of data, as described above, has continued. Plans have been developed for field research in August. This planning has included design of experiments to evaluate effects of inbreeding and of crossing between populations of little bluestem plants (*Schizachyrium scoparium*).

**Activity 3 Status as of February 1, 2021:**

Through July and August, graduate student Wes Braker carried out experimental crosses between little bluestem plants (*Schizachyrium scoparium*) growing in field plants on the St. Paul campus of UMN-TC. Mr. Braker gathered the resulting seeds and has proceeded to prepare them for germination to evaluate their viability. A small team (Shaw, May, Braker, and technician Em Daily) traveled in mid-August to the Lake Bella field site to gather data on the plants in the experimental garden there, taking extreme precautions against Covid-19 and completing the census in four days. Data have been prepared for analysis.

**Final Report Summary:**

As planned, we have continued our large-scale experiment to elucidate the geographic scale of adaptation of six prairie species. We have gathered extensive data from this experiment, and Dr. Shelby Flint has made progress on analyses of the data. Mr. Braker plans to complete analyses of part of this dataset in his dissertation work. We have also implemented experiments to investigate several aspects of the genetic structure of two populations of little bluestem (*Schizachyrium scoparium*), including genetic variance for fitness and the fitness consequences of inbreeding and of crossing between populations. Further work on these experiments will contribute to Mr. Braker's doctoral dissertation.

**V. DISSEMINATION:**

**Description:** Information and materials gained in Healthy Prairies II will be disseminated as follows. Seed collected from 20 prairie species will be deposited at UM and NCGRP (Activity 1). Information on microbial collections and their effects on prairie plant survival and reproduction information on the establishment will be communicated as written reports. Microbial collections will be maintained at UM and USDA (Activity 2). Information on the survival, and reproduction of 6 prairie plants in 3 outstate locations will be communicated to the MN-DNR, The Nature Conservancy, private land managers, and seed companies as written reports (Activity



3). The research findings will be disseminated through peer-reviewed papers published in major journals of evolution and ecology. A publicly accessible website giving collection locations and approximate population densities for prairie species will be maintained. In addition, public outreach will be conducted for all 3 Activities via Market Science, a program of UM presenting results at farmers markets throughout the Twin Cities.

**Status as of Dec. 1, 2017:** Report deferred per communication from LCCMR staff on July 24, 2017.

**Status as of June 1, 2018:**

Accession of 20 species have been deposited at UM.

**Status as of January 18, 2019:**

Dr. Ruth Shaw, Dr. Adrien Pozzi, John Benning and Anna Peschel (UMN graduate students) designed and took part in a Market Science session, a 3-hour UMN science outreach initiative where 20-25 people (children and adults) were made aware of prairie fragmentation and the need for restoration to reconnect habitat patches, as well as how fungi facilitate acquisition of plant nutrients (9/27/18, Tiny Diner farmers market, Minneapolis, MN). Accessions of 34 species have been deposited at UM. A preliminary technical report on the focus groups (discussion summaries and analysis) has been sent to participants for their review. Dr. Adrien Pozzi presented an overview of Healthy Prairies Project objectives and preliminary findings to staff and academic attendees (1/17/19, TNC office, Minneapolis, MN).

**Status as of June 1, 2019:**

Preliminary results from the local adaptation experiment (Activity 3) were presented at a workshop on local adaptation (March 21, 2019, on the St. Paul campus of UM-TC). Participants included staff from BWSR, DNR, The Nature Conservancy, and USGS, as well as researchers from UM-TC, UM-Duluth, UM-Morris, NDSU, SDSU, the Chicago Botanic Garden, the U. of Missouri, and Michigan State. Manuscript reporting on the results of focus groups identifying and relieving impediments to production and use of source-identified seeds will be submitted to *Restoration Ecology* June 2019.

Market Science session planned at Tiny Diner 7/18/19.

A manuscript on Activity 2 – Outcome 1 expected submission Fall 2019.

**Status as of Dec. 1, 2019:**

77 seed lots, representing 41 species, have been delivered to four commercial producers of native seeds. Results to date from all Activities were presented at a symposium (November 19, 2019, St. Paul campus of UM-TC). Attendees included collaborators from UM-Morris, past and current undergraduate, graduate, and professional project staff, and potential collaborators from other UM-TC research groups. A manuscript that reports on the focus group discussions aimed at identifying and relieving impediments to production and use of source-identified seeds was submitted to *Restoration Ecology* and is currently in revision. A manuscript on Activity 2, Outcome 1, is in progress. A Market Science session held on July 18, 2019 involved 88 members of the public participating in 3 activities that showcased MN's tallgrass prairies as well as their extreme fragmentation and its consequence of severely restricting seed dispersal. Visitors were also shown microbial partners of prairie plants with emphasis on their role in nutrient acquisition.

**Status as of July 1, 2020:**

A manuscript titled "Factors limiting the availability of native seed for reconstructing Minnesota's prairies: Stakeholder perspectives" was resubmitted, following revision, to *Restoration Ecology*. Two additional manuscripts are nearing completion.

**Status as of February 1, 2021:**

A manuscript titled “Factors limiting the availability of native seed for reconstructing Minnesota’s prairies: Stakeholder perspectives” was further revised and resubmitted. It is under consideration for publication in the journal, *Restoration Ecology*. Graduate student Naomi Rushing submitted a manuscript, "Latitude of seed source impacts flowering phenology and fitness in translocated plant populations", to be considered for publication in *Restoration Ecology*.

#### **Final Report Summary:**

HP team members have participated in varied opportunities to disseminate findings from this project. These include informal events to communicate with members of the public who are not all well-versed in science and may not be aware of prairies (Market Science), as well as workshops involving other scientists and land managers (Nature Conservancy ‘Science Slams’, Local Adaptation Workshop, held at UM-TC, March 2019, discussions of seed sourcing guidelines led by staff of MN DNR).

A paper providing an overview of the Local Adaptation Workshop has been published in *New Phytologist* (2020) 225:2246–2248. A manuscript reporting findings about geographic scale of local adaptation has been submitted to *Restoration Ecology* and has received positive reviews. A second manuscript reporting on a study that used focus groups to identify impediments to use of source-identified seeds for prairie restorations has been submitted to *Restoration Ecology* and has received positive reviews. Both manuscripts are under revision and will be resubmitted soon.

## **VI. PROJECT BUDGET SUMMARY:**

### **A. Preliminary ENRTF Budget Overview:**

**\*This section represents an overview of the preliminary budget at the start of the project. It will be reconciled with actual expenditures at the time of the final report.**

<b>Budget Category</b>	<b>\$ Amount</b>	<b>Overview Explanation</b>
Personnel:	\$ <b>818,500</b>	Labor intensive field work, lab analyses
Professional/Technical/Service Contracts:	\$ <b>31,000</b>	Sequencing (microbes, Activity 2), greenhouse, seed collection by local harvesters
Equipment/Tools/Supplies:	\$ <b>36,000</b>	Field and lab supplies, postage
Capital Expenditures over \$5,000:	\$	
Fee Title Acquisition:	\$	
Easement Acquisition:	\$	
Professional Services for Acquisition:	\$	
Printing:	\$	
Travel Expenses in MN:	\$ <b>14,500</b>	Travel to experimental and seed collection sites
Other:	\$	
<b>TOTAL ENRTF BUDGET:</b>	<b>\$ 900,000</b>	



**Explanation of Use of Classified Staff: N/A**

**Explanation of Capital Expenditures Greater Than \$5,000: N/A**

**Total Number of Full-time Equivalents (FTE) Directly Funded with this ENRTF Appropriation:**  
**14.1**

**Total Number of Full-time Equivalents (FTE) Estimated to Be Funded through Contracts with this ENRTF Appropriation: N/A**

**B. Other Funds:**

Source of Funds	\$ Amount Proposed	\$ Amount Spent	Use of Other Funds
<b>Non-state</b>			
	\$	\$	
<b>State</b>			
Indirect costs – In kind services 53%/54% of total direct costs	\$477,000	\$460,492	Office, lab, and meeting space, accounting and secretarial services, phone & office equipment, security, and library access, for all project personnel.
<b>TOTAL OTHER FUNDS:</b>	\$	\$	

**VII. PROJECT STRATEGY:**

**A. Project Partners:**

**Partners receiving ENRTF funding**

- UMN-TC faculty and Project Managers - Drs. R. Shaw (\$52,000, summer salary), G. May (\$49,000, summer salary); Collaborator Dr. Margaret Kuchenreuther, UM Morris (\$37,000, outstate seed collections); 2 post-doctoral fellows (\$157,500 each; plant adaptation, beneficial microbes); 2 graduate students (\$91,500 each, plant adaptation, beneficial microbes); 4 undergraduate students (\$32,000, field assistance, lab and greenhouse studies of plant – microbe interactions); Coordinator of Personnel (\$103,000, recruit, train, and work with volunteers, field assistance); Technical assistant (\$81,000, conduct lab and field research, maintain cultures, ordering, equipment management). *Amounts shown reflect 3 years funding.*

**Partners NOT receiving ENRTF funding**

- UMN-TC faculty Drs. D. Wyse, D. Moeller, P. Tiffin; UM-D faculty Dr. J. Etterson; MN-DNR; The Nature Conservancy. USDA NCGRP (Drs. C. Walters, C. Richards).

**B. Project Impact and Long-term Strategy:** The impact of the proposed work will be to preserve MN prairie plant diversity, and to provide a knowledge base for restoration and maintenance of prairie plant diversity, for future generations' use. The project will enhance land management efforts that maintain prairie lands for wildlife, provide sources of new plant and microbial products, and provide databases on distributions and abundances of many iconic prairie plant species.

The strategy for accomplishing these goals is to:

- Collect seed from 20 plant species, additional to those collected in the previous project and to include those considered more rare. This will be accomplished by the Healthy Prairies team, and our outstate collaborators.
- Determine survivorship, growth, and reproduction of 6 prairie plant species at experimental plots established under previous funding at three outstate locations. These locations represent a north-south gradient across the western prairie area of MN (see Visual). We will continue seed increase plots at the Rosemount Research and Outreach Center (UM)
- Investigate the role of beneficial microbes in plant survival and reproduction. Make collections of microbial isolates, identify these, and use in experimental greenhouse studies. Beneficial microbes will be deposited at UM and USDA culture collections for public use.
- Determine the scale of genetic variation for plant survival and reproduction across the varied MN landscape as represented by the experimental plots. The results will be communicated in publications, to the public such as native plant groups, and to prairie seed companies.

Together, the results and information generated in Healthy Prairies II will have the intended impacts as we work with land managers, seed companies, and collections resources to increase the production and success of seed sources for prairie plantings across Minnesota.

#### C. Funding History:

Funding Source and Use of Funds	Funding Timeframe	\$ Amount
Healthy Prairies I: Seed storage, beneficial microbes, and adaptation	7/1/14 - 6/30/2017	\$ 600,000
		\$
		\$

#### VIII. REPORTING REQUIREMENTS:

- The project is for 4 years, will begin on 07/01/2017, and end on 06/30/2021.
- Periodic project status update reports will be submitted *Dec. 1* and *June 1* of each year.
- A final report and associated products will be submitted between June 30 and August 15, 2021.

#### IX. VISUAL COMPONENT or MAP(S):

#### X. FEE TITLE ACQUISITION/CONSERVATION EASEMENT/RESTORATION REQUIREMENTS:

##### A. Parcel List:

##### B. Acquisition/Restoration Information:

##### Fee Title Acquisition

4. Describe the selection process for identifying and including proposed parcels on the parcel list, including explanation of the criteria and decision-making process used to rank and prioritize parcels.
5. List all adopted state, regional, or local natural resource plans in which the lands included in the parcel list are identified. Include a link to the plan if one is available.
6. For any parcels acquired in fee title, a restoration and management must be prepared. Summarize the components and expected outcomes of restoration and management plans for parcels acquired by your organization, how these plans are kept on file by your organization, and overall strategies for long-term plan implementation, including how long-term maintenance and management needs of the parcel will be financed into the future.
7. For each parcel to be conveyed to a State of Minnesota entity (e.g., DNR) after purchase, provide a statement confirming that county board approval will be obtained.
8. If applicable (see M.S. 116P.17), provide a statement confirming that written approval from the DNR Commissioner will be obtained 10 business days prior to any final acquisition transaction.

#### **Conservation Easement Acquisition**

1. Describe the selection process for identifying and including proposed parcels on the parcel list, including explanation of the criteria and decision-making process used to rank and prioritize parcels.
2. List all adopted state, regional, or local natural resource plans in which the lands included in the parcel list are identified. Include a link to the plan if one is available.
3. For any conservation easement acquired, a restoration and management must be prepared. Summarize the components and expected outcomes of restoration and management plans for parcels acquired by your organization, how these plans are kept on file by your organization, and overall strategies for long-term plan implementation, including how long-term maintenance and management needs of the parcel will be financed into the future.
4. For each parcel to be conveyed to a State of Minnesota entity (e.g., DNR) after purchase, provide a statement confirming that county board approval will be obtained.

5. If applicable (see M.S. 116P.17), provide a statement confirming that written approval from the DNR Commissioner will be obtained 10 business days prior to any final acquisition transaction. A copy of the written approval should be provided to LCCMR.
6. Provide a statement addressing how conservation easements will address specific water quality protection activities, such as keeping water on the landscape, reducing nutrient and contaminant loading, protecting groundwater, and not permitting artificial hydrological modifications.
7. Describe the long-term monitoring and enforcement program for conservation easements acquired on parcels by your organization, including explanations of the process used for calculating conservation easement monitoring and enforcements costs, the process used for annual inspection and reporting on monitoring and enforcement activities, and the process used to ensure perpetual funding and implementation of monitoring and enforcement activities.

#### **Restoration**

1. Provide a statement confirming that all restoration activities completed with these funds will occur on land permanently protected by a conservation easement or public ownership.
2. Summarize the components and expected outcomes of restoration and management plans for the parcels to be restored by your organization, how these plans are kept on file by your organization, and overall strategies for long-term plan implementation.
3. Describe how restoration efforts will utilize and follow the Board of Soil and Water Resources “Native Vegetation Establishment and Enhancement Guidelines” in order to ensure ecological integrity and pollinator enhancement.
4. Describe how the long-term maintenance and management needs of the parcel being restored with these funds will be met and financed into the future.
5. Describe how consideration will be given to contracting with Conservation Corps of Minnesota for any restoration activities.
6. Provide a statement indicating that evaluations will be completed on parcels where activities were implemented both 1) initially after activity completion and 2) three years later as a follow-up. Evaluations should analyze improvements to the parcel and whether goals have been met, identify any problems with

the implementation, and identify any findings that can be used to improve implementation of future restoration efforts at the site or elsewhere.

Environment and Natural Resources Trust Fund  
M.L. 2017 Project Budget

Project Title: Healthy Prairies II: Preserving MN prairie plant diversity  
Legal Citation: M.L. 2017, Chp. 96, Sec. 2, Subd. 03c  
Project Manager: Dr. Ruth Shaw  
Organization: Regents of the University of Minnesota  
M.L. 2017 ENRTF Appropriation: \$ 900,000  
Project Length and Completion Date: 4 Years, June 30, 2021  
Date of Report: December 31, 2020



ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Activity 1 Budget (12/10/19)	Amount Spent	Activity 1 Balance	Activity 2 Budget (2/1/19)	Revised Budget 12/31/20	Amount Spent	Activity 2 Balance	Activity 3 Budget (12/10/19)	Amount Spent	Activity 3 Balance	Total Budget (12/10/19)	Revised Budget 12/31/20	TOTAL BALANCE
<b>BUDGET ITEM</b>				<i>Fill in your activity title here.</i>									
<b>Personnel (Wages and Benefits)</b>	\$98,500	\$113,640	-\$15,140	<del>\$341,180</del>	\$341,312	\$306,229	\$35,083	\$364,820	\$384,763	-\$19,943	\$804,500	\$804,632	\$0
Dr. Ruth Shaw, Co-PI: \$52,000 (75% salary, 25% benefits); 8% FTE. 1 month per year for three years.													
Dr. Georgiana May, Co-PI: \$49,000 (75% salary, 25% benefits); 8% FTE. 1 month per year for 3 years.													
Dr. Margaret Kuchenreuther, UM Morris, collaborator: \$37,000 (75% salary, 25% benefits); 8% FTE. 1 month per year, for 3 years.													
2 Postdoctoral Associates: \$315,000 (82% salary, 18% benefits); 100% FTE, 3 years													
2 Graduate Students: \$183,000 (51% salary, 49% benefits during the academic year & 85% salary, 15% benefits during the summer); 50% FTE, 2 years.													
4 Undergraduate Students: \$32,000 (100% salary, 0% benefits); 2 @ 8% FTE (UM Twin Cities) and 2 @ 15% FTE (UM Morris), 3 years.													
Coordinator of personnel: \$103,000 (79% salary, 21% benefits); 100% FTE, 2 years.													
Technical assistant: \$81,000 (79% salary, 21% benefits); 100% FTE, 2 years.													
<b>Equipment/Tools/Supplies</b>	\$15	\$15	\$0	<del>\$25,000</del>	\$26,391	\$27,442	-\$1,051	\$6,000	\$4,949	\$1,051	\$31,015	\$32,406	\$0
Lab Supplies: \$25,000. Supplies for microbial culturing and storage (~ 6000 cultures per year), microbial detection in plant materials and identification of organisms using molecular methods and microscopy.													
Field supplies and prep work: \$18,000. Envelopes and bags, blaze hats and vests, galvanized nails and landscape staples, tape measures, fencing materials, knee pads, mallets, field notebooks, etc.													
<b>Travel expenses in Minnesota</b>	\$738	\$738	\$0	<del>\$6,500</del>	\$5,033	\$4,090	\$943	\$34,679	\$35,621	-\$942	\$40,917	\$40,450	\$0
Travel to field sites for seed collection (Activity 1), and microbial sampling (Activity 2). Monitoring experimental plots (Activities 2, 3), and seed increase plots in Rosemount. Total travel estimated: 25,000 miles in MN, with 150 hotel-person overnights, over 3 years.													
<b>Other</b>	\$3,568	\$3,981	-\$413	<del>\$16,000</del>	\$14,944	\$14,800	\$144	\$4,000	\$3,732	\$268	\$23,568	\$22,512	\$0
Fees for independent contractors collecting seeds (\$8,000)													
Postage/Shipping Fees: \$2,000. Shipping seeds to Nat'l Center for Genetic Resources Preservation (NCGRP), USDA facility in Ft. Collins, CO. \$100 per shipment x 20 shipments.													
Sequencing (UMN-TC facility): \$10,000. Detection, identification, and distribution of naturally occurring microbes in native prairie plants using rapid, cutting edge "metagenomics" approaches.													
Greenhouse space rental (UMN-TC): \$13,000. Evaluating microbial effects on plant growth and reproduction (Activity 2), seedlings for outplanting, plant genetic variation analyses (Activity 3). 500 sq. ft. x \$0.81/sqft per month x 30 months. Seed increase pilot fees.													
<b>COLUMN TOTAL</b>	<b>\$102,821</b>	<b>\$118,374</b>	<b>-\$15,553</b>	<b>\$387,680</b>	<b>\$387,680</b>	<b>\$352,561</b>	<b>\$35,119</b>	<b>\$409,499</b>	<b>\$429,065</b>	<b>-\$19,566</b>	<b>\$900,000</b>	<b>\$900,000</b>	<b>\$0</b>