

## **M.L. 2015 Project Abstract**

For the Period Ending June 30, 2017

**PROJECT TITLE:** Habitat Mitigation for Goblin Fern Conservation

**PROJECT MANAGER:** Bobby Henderson

**AFFILIATION:** Leech Lake Band of Ojibwe

**MAILING ADDRESS:** 115, 6th St. NW, Suite E

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

**LEGAL CITATION:** M.L. 2015, Chp. 76, Sec. 2, Subd. 03s

**APPROPRIATION AMOUNT:** \$61,000

**AMOUNT SPENT:** \$39,276

**AMOUNT REMAINING:** \$21,724

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

Goblin fern (*Botrychium mormo*) is a tiny, cryptic species of fern from the Great Lakes region of North America. The species once occurred throughout rich sugar maple and basswood forests of Minnesota, Michigan, and Wisconsin, but in recent times has become exceedingly rare and vulnerable throughout the entirety of its historic range. The primary goals of this project were to (1) evaluate habitat conditions and environmental factors influencing the decline of goblin fern populations; (2) quantify projected population extirpation rates for all recorded populations across Leech Lake Reservation, including Chippewa National Forest.

1) Habitat conditions were assessed by the degree of worm damage at each location and assigned an IERAT rank of 1-5 (1 = non-wormed, 5 = extremely wormed). Each site was also assigned a habitat ranking of 1-5 (1 = prime, 5 = non-extant); which directly correlates with the probability of goblin fern presence/absence at each location.

2) Our estimates of Minnesota goblin fern extirpation are consistent with previous publications; in fact our estimates indicate a significant increase in extirpation rates when compared to older publications addressing habitat issues and concerns.

Though startling, our study provides current published information about the loss of critical habitat, and subsequent decrease in occurrence and abundance of this state threatened species across its native range within Minnesota. With exception to the driftless area in southeastern Minnesota, there are no earthworm species native to the state, especially the rich maple and basswood forests of northern Minnesota. Contrary to long held belief, earthworms, especially those known as "crawlers", cause irreparable damage to the forest floor and soil. As a result, much of the vital habitat required for the survival of goblin fern has become seriously degraded and fragmented across the north woods of Minnesota.

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

Our abstract was submitted for peer review in June 2018. Upon receiving comment, review and revisions were made to the abstract, which was submitted in July 2018, and ultimately accepted for publication. The published article has been disseminated amongst select individuals within Minnesota Department of Natural Resources, Chippewa National Forest, Superior National Forest, and Ottawa National Forest for the purpose of developing and implementing improved habitat conservation

measures. Additionally, all data collected from the project were shared with the USFS, Chippewa National Forest for the purposes of updating database records.



# Environment and Natural Resources Trust Fund (ENRTF) M.L. 2015 Work Plan Final Report

**M.L. 2015, Chp. 76, Sec. 2, Subd. 03s Project Abstract**  
For the Period Ending June 30, 2017

**PROJECT TITLE:** Habitat Mitigation for Goblin Fern Conservation  
**PROJECT MANAGER:** Bobby Henderson  
**AFFILIATION:** Leech Lake Band of Ojibwe  
**MAILING ADDRESS:** 115, 6th St. NW, Suite E  
**CITY/STATE/ZIP:** Cass Lake, MN 56633  
**PHONE:** (218) 784-8620  
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**WEBSITE:** [If applicable]  
**FUNDING SOURCE:** Environment and Natural Resources Trust Fund  
**LEGAL CITATION:** M.L. 2015, Chp. 76, Sec. 2, Subd. 03s

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Chippewa National Forest, Superior National Forest, and Ottawa National Forest for the purpose of developing and implementing improved habitat conservation measures. Additionally, all data collected from the project were shared with the USFS, Chippewa National Forest for the purposes of updating database records.

- 1)** Zlonis, K.J., Henderson, B.W., 2018. Invasive earthworm damage predicts occupancy of a threatened forest fern: Implications for conservation and management. *Forest Ecology and Management* 430, 291-298



# Environment and Natural Resources Trust Fund (ENRTF) M.L. 2015 Work Plan Final Report

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**Date of Report:** December 18, 2018 (Final Report)

**Final Report**

**Date of Work Plan Approval:** June 11, 2015

**Project Completion Date:** June 30, 2018

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**PROJECT TITLE:** Habitat Mitigation for Goblin Fern Conservation

**Project Manager:** Bobby Henderson

**Organization:** Leech Lake Band of Ojibwe

**Mailing Address:** 115, 6th St. NW, Suite E

**City/State/Zip Code:** Cass Lake, MN 56633

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**Email Address:** bwh1940355@gmail.com

**Web Address:**

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**Location:** Beltrami, Cass, Itasca (Leech Lake Reservation/Chippewa National Forest)

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**Total ENRTF Project Budget:**

**ENRTF Appropriation:** \$61,000

**Amount Spent:** \$39,276

**Balance:** \$21,724

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**Legal Citation:** M.L. 2015, Chp. 76, Sec. 2, Subd. 03s

**Appropriation Language:**

\$61,000 the first year is from the trust fund to the commissioner of natural resources for an agreement with the Leech Lake Band of Ojibwe to examine goblin fern populations, a threatened species in Minnesota, in relation to habitat degradation and to develop long term habitat mitigation and species conservation strategies. This appropriation is available until June 30, 2018, by which time the project must be completed and final products delivered.

## I. PROJECT TITLE: Habitat Mitigation for Goblin Fern Conservation

### II. PROJECT STATEMENT:

In 2013 Minnesota made updates to the Endangered and Threatened Species list, Statement of Need and Reasonableness (SONAR). As a result the status for Goblin fern (*Botrychium mormo*) was raised from a species of Special Concern to a Threatened species in Minnesota. This rare fern commonly referred to as a moonwort or Goblin fern, is a small discreet fern endemic to the rich Northern Hardwood forests of Minnesota, Wisconsin, Michigan, with a single historic record from Ontario, Canada. Currently this species is threatened in Minnesota (total records unknown), threatened in Michigan (13 records), endangered in Wisconsin (89 records), and receives a Global rank of G3 (vulnerable). Within this restricted range it occurs only within specific habitats defined by forest community type and soils. These communities in Minnesota are often dominated by *Acer saccharum*, *Tilia americana*, *Fraxinus nigra*, *Betula alleghaniensis*, with occasional upland *Thuja occidentalis* and *Ulmus spp...* The soils tend to be sandy loams – loam, with an intact thick organic layer (O and A soil horizons) Outside Minnesota the habitats and canopy composition slightly differ, but *Acer* and *Tilia* continue to be a dominant associate species.

Since 1975 there have been over 600 Goblin fern observations on Leech Lake Reservation/Chippewa NF; which makes this area the heart of Goblin fern distribution. But it is highly suspected many of the populations prior to 2000 have been extirpated as a result of severe habitat degradation. The scourge most responsible for Goblin fern extirpation throughout its range has been the introduction of non-native earthworms. All earthworms found in Minnesota are non-native; with the majority being European species of the family *Lumbricidae*. The initial arrival of earthworms came with European settlers around the mid-1800s, but since that time their spread has been expedited through human activities such as the dumping of unused fishing bait, forest management activities, recreation, development, and potentially anything that moves soil from a contaminated area to areas unaffected by earthworms. As a result the continual rapid loss of habitat is extirpating Goblin fern populations at an alarming rate.

This is a multifaceted project with multiple goals. The **overall goal** is to review historic record locations to examine populations in order to quantify Goblin fern abundance, decline, and possible extirpations at the local scale. These crucial steps in the project will be accomplished through surveying and monitoring a random sampling of sample units (EO records).

The **second goal** is to collect earthworm data utilizing the Invasive Earthworm Rapid Assessment Tool (IERAT) and assign each specific location a ranking as defined by IERAT. This exercise will also give us the opportunity to assess and document the adverse effects earthworms cause to Goblin fern habitat. Ultimately the earthworm data will be submitted to a much larger earthworm study being conducted by Great Lakes Worm Watch and UMD NRRI.

The **third goal** is to analyze the data with the assistance of Forest Service botanist Kirk Larson in order to determine:

- 1) Is Goblin fern abundance and distribution diminishing on the Leech Lake Reservation/Chippewa National Forest?
- 2) Identify the factors responsible for diminishing population abundance and/or extirpations at the local scale.
- 3) Identify conservation strategies/plans which will need to be addressed and/or developed in order to minimize further negative effects to Goblin fern habitat and populations.

### III. OVERALL PROJECT STATUS UPDATES:

#### Project Status as of October 30, 2015:

In early July Kirk Larsen and I began field activities to relocate and establish the probable goblin fern plots based on available coordinates for each location. The oldest 40 locations proved to be the greatest challenge as many

of these locations were observed pre GPS days. Consequently, I was forced to seek out the closest available habitat and/or forest community matching the descriptions documented in the original field forms.

As a result of poor or inadequate field notes, timber management activities, and/or ecological issues, some locations were not relocated in 2015. This forced us to establish the most probable location within a reasonable distance of the inferred coordinate. Unfortunately, there were a number of instances where little or no suitable goblin fern habitat remained within a 300' radius of the given UTM and in some instances there was no suitable goblin fern habitat within 1000' of the original coordinates. But one could observe that there had been goblin fern habitat within the past 20 years.

For 2015, Goblin fern phenology appeared to be approximately 2 weeks behind schedule compared to previous 6 years phenology monitoring I've conducted. Kirk and I strongly suspect this was the result of environmental factors (winter and spring drought conditions) we observed during early 2015. For a phenology reference we visited a known location where hundreds goblin fern are typically observed, but struggled to find a dozen plants this past season. These environmental factors possibly explain why we encountered a number of locations where habitat continues to exist, but no plants were observed.

With the use of the Invasive Earthworm Rapid Assessment Tool (IERAT), earthworm damage is ranked on a scale of 1-5; 1 being a fully intact forest floor and 5 representing no forest floor. For each location visited the habitat was examined and assigned a rank using the IERAT scale. Although the data has not been compiled, our preliminary field observations strongly suggest less than 8% of our sites have a fully intact forest floor.

**Project Status as of April 15, 2016:**

Since our initial 2015 relocation efforts of the 80 goblin fern sites, I began to compile and enter the 2015 field data in regards to target species presence, population viability, habitat conditions, earthworm activity per IERAT, and canopy cover.

Currently we are preparing for the 2016 field season. Field activities will commence late June/early July provided phenology and environmental conditions allow.

**Project Status as of October 30, 2016:**

The second year of site revisits and data collection began the third week in June and concluded the mid-August 2016. Phenology appeared to be a little earlier than the 2015 field season, plus environmental conditions appeared more favorable as the spring was not as droughty compared to spring 2015.

Being a somewhat subterranean species, small populations of goblin fern can be difficult to observe from year to year as this species is capable of completing all necessary biological functions without emerging during years where environmental conditions are less than favorable. However, this should not be mistaken for a habitat which no longer exists as a result of extensive and severe earthworm degradation.

Overall the field work is going well and will resume June 2017.

**Project Status as of April 15, 2017:**

Currently we are preparing for the 2017 field season. Field activities will commence late June/early July provided phenology and environmental conditions allow. 2016 field data was entered over the course of winter.

**Project Status as of October 30, 2017:**

The third and final year of site revisits and data collection began the third week in June and concluded early September 2017. Phenology appeared to be odd this year compared to the previous two field seasons. Environmental conditions appeared favorable as spring began with adequate moisture, but quickly became dry and remained that way from mid-June through August.

Being a somewhat subterranean species, small populations of goblin fern can be difficult to observe from year to year as this species is capable of completing all necessary biological functions without emerging during years where environmental conditions are less than favorable.

Overall, the project has been time consuming during the summer months, but the data collected is beginning to answer questions surrounding the species requirement for quality habitat. We are currently planning to extend the project additional years beginning June 2018. The protocol will be refined to collect data only from extant sites vs. all eighty locations, with a similar focus of monitoring habitat and population density. This will be important data collection as a number of sites are in serious jeopardy of extirpation as a result of advancing worm fronts. To capture an extirpation while it is occurring is extremely valuable research.

#### **Retroactive Amendment Request (04/09/2018):**

We are seeking an amendment to 1) retroactively add Katie Zlonis as project personnel to the budget and 2) extend the project end date to June 30, 2018. Bobby Henderson accepted a position with the USFS in 2016 and has continued to work on the project as the Project Manager. In March 2017, Katie began working as the Plant Resources Program Manager at Leech Lake DRM and has since worked on data analysis, synthesis, and writing the resulting manuscript for the project. Her work on the project prior to 2018 includes approximately 1 day of work in May 2017, which we understand won't be reimbursed, and approximately 60 hours of work from November 20, 2017–December 15, 2017 (approximately \$1,500). Her work on the project continued after January 1, 2018 (approximately 150 hours to date, ~\$3,600) and is ongoing.

In addition, we are requesting an extension in the project end date to June 30, 2018 so that we can conclude publication of a peer-reviewed manuscript that is nearly ready to be submitted and complete management plans.

#### **Amendment Approved: [04/11/2018]**

##### **Project Status as of April 15, 2018:**

All data collected over the past three field seasons have been collected, entered, checked, and analyzed.

As of April 15, a manuscript detailing methods and results of this project were nearly ready for submission to a peer-reviewed journal. The manuscript was submitted after this reporting period on May 1, 2018.

##### **Project Status as of September 14, 2018:**

As of August 2018 our manuscript was accepted and ultimately published by *Forest Ecology and Management* 430 (2018) 291–298. Along with the updated report and budget is the journal article titled “Invasive earthworm damage predicts occupancy of a threatened forest fern: Implications for conservation and management”.

Though data collection for the initial goblin fern project was not planned beyond three years, Kirk Larson and I continued monitoring the extant 27 locations during August/September 2018. We feel the project should continue for an additional seven years in order to collect a long term data set for the remaining sites. Though I'm now employed with US Forest Service, I continue to consult with LLBO representatives Steve Mortensen and Kate Hagsten regarding the long term plans and goals of this project. Currently we are in the process of planning a meeting with representatives from other natural resource management agencies to discuss the long term implications of current forest management uses and practices. At this time, the initial three year phase of the project has been completed.

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

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Wisconsin, but in recent times has become exceedingly rare and vulnerable throughout the entirety of its historic range. The primary goals of this project were to (1) evaluate habitat conditions and environmental factors influencing the decline of goblin fern populations; (2) quantify projected population extirpation rates for all recorded populations across Leech Lake Reservation, including Chippewa National Forest.

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Though startling, our study provides current published information about the loss of critical habitat, and subsequent decrease in occurrence and abundance of this state threatened species across its native range within Minnesota. With exception to the driftless area in southeastern Minnesota, there are no earthworm species native to the state, especially the rich maple and basswood forests of northern Minnesota. Contrary to long held belief, earthworms, especially those known as “crawlers”, cause irreparable damage to the forest floor and soil. As a result, much of the vital habitat required for the survival of goblin fern has become seriously degraded and/or fragmented across the north woods of Minnesota.

#### **IV. PROJECT ACTIVITIES AND OUTCOMES:**

##### **ACTIVITY 1: Identify Experimental Units, Finalize Project Design, Survey and Assess Locations.**

CNF botanist Kirk Larson and I will examine the CNF Goblin fern records to identify four sub-sets of experimental units. The sub-sets will be based on five year increments from 1992 – 2011 giving us a sample period of 20 years. It is important to note that CNF Forest Service lands are divided into districts, compartments, and timber stands, so each experimental unit has a unique identifier (District - Compartment/Stand number). For each five year period we will randomly identify our 20 experimental units for a total of 80 unique experimental units.

Sample units will be based on the element of occurrence (Goblin fern record) within the experimental unit. Each experimental unit will contain a sample unit identified by a unique corporate number for the EO (example: BOMO3001) There will be a total of 80 sample units to coincide with the 80 experimental units. For timber stands containing more than one Goblin fern EO, a sample unit will be randomly selected for survey and monitoring.

Sub-set 1: 1992-1996 – 20 experimental units – 20 sample units

Sub-set 2: 1997-2001 – 20 experimental units – 20 sample units

Sub-set 3: 2002-2006 – 20 experimental units – 20 sample units

Sub-set 4: 2007-2011 – 20 experimental units – 20 sample units

Once the experimental and sample units are identified, CNF botanist Kirk Larson and I will design the survey and data collection protocol for the three year project. Much of the information we wish to obtain is related to habitat quality and population viability (*goblin fern presence/persistence, canopy closure, soil moisture, associated species, soil organic layer, earthworm presence and effects*). This simple base data will be important for decisions regarding future Forest Plan updates.

The first year of surveys will be most labor intensive as the location coordinates from early observations tend to be highly inaccurate. If ground truthing the original EO proves to be difficult, then the experimental unit (timber stand) will be fully surveyed to help establish the probable EO coordinates. If the original location goes without

detection, the original field notes will be utilized to identify the best habitat within proximity of the given EO coordinates. Once the probable habitat is identified it will be designated as the sample unit.

For sample plots where Goblin fern continues to persist, there will be pin flags placed in the ground to identify a meter square sample unit at the site. At EO locations where Goblin fern is absent, there will be a single pin flag placed in the vicinity of the coordinates or habitat to identify the location as a sample unit. If at a later date Goblin fern is observed in the vicinity, then there will be a meter square sample unit placed around the EO to identify it as the probable location.

By mid-August most Goblin fern have senesced, so intensive field work is required as there is approximately a six week time window from late June – early August for conducting quality Goblin fern surveys. Kirk Larson and I are highly experienced in *Botrychium* surveys and identification so this accounts for the decision to utilize only two surveyors for the project. Data consistency and thoroughness are crucial to reduce inaccuracy which also contributes to the decision for only two surveyors.

**Summary Budget Information for Activity 1:**

**ENRTF Budget: \$ 37,400**  
**Amount Spent: \$ 27,188**  
**Balance: \$ 10,212**

| <b>Outcome</b>  | <b>Completion Date</b> |
|---|------------------------|
| 1. Establish project and survey protocol and identify experimental and sample units | July 2015              |
| 2. Survey experimental and sample units late June-mid August 2015, 2016, 2017       | August 2017            |
| 3. Compile and analyze data from survey and monitoring efforts                      | December 2017          |

**Project Status as of October 30, 2015:**

In early March we identified our random sample of locations and began preliminary data collection for the goblin fern sites to be relocated and monitored over the next three years. After the 80 random locations were identified, GIS specialist Devona Berndt with the USDA Forest Service offered up her services to produce maps along with copies of existing documentation for each of the 80 locations. Between March and June 2015 Kirk Larson and I met on a number of occasions to discuss parameters of the project and establish relocation protocol.

Between early July through early September, Kirk and I completed our field work activities. This included relocation efforts, assigning and marking the population (plot), collecting all relevant data (described in activity 2) as it pertains to the project, plus collecting photos of the actual plants. A total of approximately 412 hours have been contributed to preliminary data gathering and compiling, cartography, relocation and survey efforts.

**Project Status as of April 15, 2016:**

Although it's too early in the project to draw conclusions, there have been some interesting but not unforeseen correlations between species absence and earthworm activity. As the project progresses I expect recurring patterns in the data where earthworm activity is ranked 4 or higher.

**Project Status as of October 30, 2016:**

Between late June to early August, Kirk and I conducted and completed the 2016 field activities. This included relocation efforts, assigning and marking the population (plot) for 12 populations which were not observed during the 2015 field visit, collecting all relevant data (described in activity 2) as it pertains to the project, plus collecting photos of the actual plants at the locations.

Compared to the 2015 field season, April and May 2016 offered greater early season soil moisture which carried into June when goblin fern begins to emerge. As field work progressed we observed a number of locations which exhibited greater numbers of plants vs. the 2015 field season, plus 12 populations which were not observed during the 2015 field visit.

For summer 2016, Kirk and I contributed a combined total of 321 hours for survey efforts and data collection.

**Project Status as of April 15, 2017:**

Although it's too early in the project to draw conclusions, there have been some interesting but not unforeseen correlations between species absence and severe earthworm activity. As the project progresses I expect recurring patterns in the data where earthworm activity is ranked 4 or higher. There is a direct correlation between earthworm activity and habitat integrity, which comes as no surprise, but is still valuable data to record.

**Project Status as of October 30, 2017:**

Between late June to early September, Kirk and I completed the 2017 field data collection. This included revisiting efforts, population counts, habitat assessment, collecting all relevant data (described in activity 2) as it pertains to the project, locating and marking populations that have not been observed the previous two field seasons, and ensuring all the locations are well marked for additional revisits which will begin summer 2018.

Compared to the previous two field seasons, 2017 field season was very dry during the emergence period, so population numbers were either low, or in a number of instances plants were absent altogether.

**Project Status as of April 15, 2018:**

As of April 15, 2018, all collected field data were entered, checked, and sent to Katie Zlonis at LLBO for analysis.

**Final Report Summary:**

During the course of the project, goblin fern was observed during at least one of three survey seasons at 27 locations (33.75%). Abundance across extant sites was relatively low with 42 plants observed in 2015, 62 plants in 2016, and 54 plants in 2017; range equals 1 - 12 plants with a mean of 3.

The Invasive Earthworm Rapid Assessment Tool (IERAT) was used at all locations to gather baseline information on soil and habitat condition. IERAT assessments range from 1-5; 1 being non-impacted, 5 representing severely degraded by invasive earthworms, especially *Lumbricus spp.* "night crawlers".

Goblin fern was documented at 27 of 80 sites (33.75%), with IERAT scores of 1-4; IERAT 1: 1 location, IERAT 2: 5 locations, IERAT 3: 16 locations, IERAT 4: 5 locations. Conversely, 53 locations (66.25%) displayed zero plants over the course of three field seasons. The locations where goblin fern was absent received IERAT assessments of 3-5; IERAT 3: 9 locations, IERAT 4: 21 locations, and IERAT 5: 22 locations.

Forest canopy conditions, including sub canopy, were monitored at all locations to document any significant changes that may occur in the canopy closure between survey periods. The majority of locations have a canopy closure range of 62% to 98%. Years of repeated field observation suggests canopy closure and structure play a complex role in maintaining environmental and microclimate conditions within viable goblin fern habitat. But the range of canopy closure alone does not appear to contribute to existence, absence, or abundance of goblin fern colonies. In summary, the range of canopy closure does not add any explanatory power for whether goblin fern remains present or absent above and beyond habitat degradation resulting from invasive earthworm activity.

**ACTIVITY 2: Data Analysis, Submit Results to CNF, Assist in Development of Mitigation Plan for Updated CNF Forest Plan**

For each sample unit, we will monitor and collect data for three consecutive seasons in order to document population health for our random selection of sample units. The data collection protocol will focus on target species presence, population viability, probable extirpations, current habitat conditions and earthworm activity. From this project we can potentially conduct long term monitoring of the locations which continue to contain viable Goblin fern populations.

Specific data we wish to collect includes:

- 1) Target species presence
- 2) Conduct plant count at sample units
- 3) Population viability (based on plant count, habitat conditions, earthworm activity)
- 4) Canopy composition
- 5) Canopy density
- 6) Associate vegetation species
- 7) Habitat conditions
- 8) Soil moisture
- 9) Evaluate Earthworm activity with the assistance of IERAT

Within the data analysis we will search for correlations between population abundance, habitat conditions and earthworm activity. Through repeated field observations I've observed what appear to be Goblin fern extirpations in the wake of earthworm invasions, but there has been very little work on the CNF to document or prove these events.

The major goal of this project is to extract enough information from data to support proposal for the development of improved Goblin fern habitat mitigation strategies and species conservation plans.

The intensive three year survey and monitor project will help in identifying the most detrimental issues surrounding Goblin fern habitat and populations. From this data, Leech Lake Band of Ojibwe will be able to develop appropriate Goblin fern habitat mitigation strategies and species conservation plans to insure appropriate protection for Goblin fern and Goblin fern habitat on tribally owned and managed lands.

As a co-managing agency, Leech Lake Band of Ojibwe must collaborate with Chippewa National Forest to insure habitat mitigation strategies and species conservation plans are developed at the forest wide level. This will help insure the species and habitat on Forest Service managed lands within the Leech Lake Reservation boundaries are adequately protected.

The Chippewa National Forest, Forest Management Plan is scheduled for review and revision on a 10 year cycle, but has not been revised since 2004. This indicates the Forest Plan is up for review and revision this year. As a result, the current forest plan is probably inadequate for the long term conservation of Goblin fern and the required habitat for this species. As a co-management agency, Leech Lake Reservation will request review of the current CNF Forest Plan with the intentions of co-developing a current and appropriate species conservation plan for Goblin fern and all struggling moonwort species residing on Leech Lake Reservation/Chippewa National Forest.

**Summary Budget Information for Activity 2:**

**ENRTF Budget: \$ 23,600**  
**Amount Spent: \$ 12,088**  
**Balance: \$ 11,512**

| <b>Outcome</b>  | <b>Completion Date</b> |
|---|------------------------|
| 1. Predict probable extirpations within our experimental and sample units                             | October 2017           |
| 2. Updating CNF records to account for extirpations   | October 2017           |
| 3. Submit data to support a larger earthworm project  | December 2017          |
| 4. Coordinate with CNF to develop and implement new habitat mitigation and species conservation plans | April 2018             |
| 5. Develop and implement new habitat mitigation plans for Leech Lake Tribal Lands                     | April 2018             |
| 6. Disseminate results to interested management agencies  | April 2018             |
|   |                        |
|   |                        |
|   |                        |

**Project Status as of October 30, 2015:**

No data analysis at this time as 2015 data has yet to be compiled and analyzed.

**Project Status as of April 15, 2016:**

2015 data has been compiled and entered, but as a result of project infancy no conclusions can be drawn based on a single year of data.

**Project Status as of October 30, 2016:**

2015 data has been compiled and entered and 2016 data entry is in progress. As a result of project infancy no conclusions can be inferred based on a two years of data collection.

**Project Status as of April 15, 2017:**

2016 data was compiled and entered.

**Project Status as of October 30, 2017:**

2017 data entry is almost completed. We have not begun data analysis yet, but that is planned to begin in November.

**Retroactive Amendment Request (04/09/2018):**

**Project Status as of April 15, 2018:**

As of April 15, 2018 Katie Zlonis has conducted data analysis, synthesis, and nearly completed a draft manuscript for the project. This includes predictions of probability of occupancy/extirpation for each goblin fern study site. Her work on the project prior to 2018 includes approximately 60 hours from November 20, 2017-December 15, 2017. Since January 1, 2018 to date, Katie has contributed approximately 150 hours to the project and her work is ongoing.

Leech Lake DRM is organizing a meeting with stakeholders, which includes Minnesota DNR biologists, US Forest Service biologists, and TNC biologists. The purpose of this meeting is to raise awareness of the severe and rapid loss of goblin fern habitat as a direct result of forest management activities which perpetuate invasive earthworms invading these fragile ecosystems. At the current rate of extirpation, goblin fern could very well become petitioned for Federal listing within 10 years. The time has come for all parties to seriously discuss and develop comprehensive conservation strategies for the future of goblin fern within Leech Lake Reservation/Chippewa National Forest.

The Chippewa National Forest has shown an interest in continuation of the project for an additional 7-10 years in order to monitor and collect data on the remaining 27 populations within the original project. This will continue to be a collaborative project between LLBO and CNF.

**Final Report Summary:**

Data collected from the project, along with observations and data collected outside the project indicate the presence or absence of goblin fern within its habitat has a direct correlation with IERAT. We conducted occupancy modeling, taking detectability across years into account, to assess probability of presence and absence of goblin fern, and environmental factors that may influence that probability. The best model for explaining goblin fern occupancy, evaluated by Akaike's Information Criterion (AIC), included only IERAT score. We found that probability of occurrence decreased significantly as IERAT score increased. At IERAT stage 2, which is characterized by worm species that do not completely consume the duff layer, goblin fern was present at least one year in every site, suggesting these sites are still good habitat for goblin fern. We identified IERAT 3 as the tipping point at which habitat is rapidly becoming unsuitable for goblin fern due to invasion of *Lumbricus* spp. At IERAT 4, occupancy probability decreased below 50%, and at IERAT 5 we found no evidence that goblin fern can still be supported (i.e. no plants were found during the three-year survey period at any sites classified as IERAT 5).

The Leech Lake Reservation/Chippewa National Forest appears to be the final stronghold for this species. Based on the analysis performed for this project, we estimate that at the current rate of extirpation, goblin fern could soon become a candidate for federal listing, so if goblin fern is to persist, a strict and conscious conservation plan must be developed for the species.

## **V. DISSEMINATION:**

### **Description:**

Once we compile all relevant data, the results will be shared with interested agencies and organizations who express similar interest in the conservation of Goblin fern. All relevant earthworm data will be shared with NRRI, University of Minnesota, Duluth to help with their ongoing research. This information will include coordinates, and severity of infestation. Since this Goblin fern is considered a Threatened, Endangered, Sensitive species by Leech Lake Band of Ojibwe and the Chippewa National Forest, data and results will be shared at the discretion of the two co-management agencies.

### **Project Status as of October 30, 2015:**

Data and findings will not be disseminated until project is completed.

### **Project Status as of April 15, 2016:**

Data and findings will not be disseminated until project is completed.

### **Project Status as of October 30, 2016:**

Data and findings will not be disseminated until project is completed.

### **Project Status as of April 15, 2017:**

Data and findings will not be disseminated until project is completed.

### **Project Status as of October 30, 2017:**

Data and findings will not be disseminated until project is completed.

### **Retroactive Amendment Request (04/09/2018):**

### **Project Status as of April 15, 2018:**

Project results have been informally shared with relevant DNR, USFS, and LLBO biologists. Once the manuscript is accepted for publication, we will disseminate it widely across agencies to all relevant staff. In addition, as stated previously, LLBO is arranging a meeting across agencies to disseminate the results of this research and discuss conservation measures to take going forward for this species.

### **Project Status as of September 14, 2018:**

Dissemination of field data has been completed with US Forest Service and is currently being entered into the corporate data base. The journal article has recently been distributed to interested parties including LLBO, US Forest Service, Minnesota DNR, and other interested natural resource management and academia professionals. IERAT data has been compiled and will be shared with NRRI within the month. This concludes obligations outlined in the grant proposal.

### **Final Report Summary:**

Our abstract was submitted for peer review in June 2018. Upon receiving comment, review and revisions were made to the abstract, which was submitted in July 2018, and ultimately accepted for publication. The published article has been disseminated amongst select individuals within Minnesota Department of Natural Resources, Chippewa National Forest, Superior National Forest, and Ottawa National Forest for the purpose of developing and implementing improved habitat conservation measures. Additionally, all data collected from the project were

shared with the USFS, Chippewa National Forest for the purposes of updating database records. The publication and IERAT data will be shared with NRRI before 2019.

- 1) Zlonis, K.J., Henderson, B.W., 2018. Invasive earthworm damage predicts occupancy of a threatened forest fern: Implications for conservation and management. Forest Ecology and Management 430, 291-298

**VI. PROJECT BUDGET SUMMARY:**

**A. ENRTF Budget Overview:**

| Budget Category            | \$ Amount       | Overview Explanation   |
|----------------------------|-----------------|--|
| Personnel:                 | \$42,091        | Bobby Henderson, project manager, botanist, data analysis (80% salary, 20% benefits) 35% FTE for 2 years; 20% FTE for 1 year |
|                            | \$10,109        | Katie Zlonis, data analysis, manuscript preparation and submission (80% salary, 20% benefits) 15% FTE for 1 year             |
| Equipment/Tools/Supplies:  | \$2050          | High precision GPS for establishing sites, Densimeters, Field supplies: collection bags, batteries, flagging,                |
| Travel Expenses in MN:     | \$6750          | Travel to, between, and from survey and data collection sites. Mileage:  |
| <b>TOTAL ENRTF BUDGET:</b> | <b>\$61,000</b> |  |

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

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

**Number of Full-time Equivalents (FTE) Directly Funded with this ENRTF Appropriation:** 1.05

**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>                            | \$0                | \$0             |   |
| <b>State</b>                                | \$0                | \$0             |   |
| <b>In-kind Services To Be Applied</b>       |                    |                 |   |
| USDA Forest Service: Botanist Kirk Larson.  | \$11,100           | \$12,321        | Survey and data collection support.   |
| Leech Lake Division of Resource Management. | \$40,000           | \$19,973        | IDC, Administrative support, office space, computers, GIS programs, vehicles and maintenance. |
|   |                    |                 |   |
| <b>TOTAL OTHER FUNDS:</b>                   | <b>\$51,000</b>    | <b>\$32,204</b> |   |

**VII. PROJECT STRATEGY:**

**A. Project Team/Partners**

**Funded:**

Bobby Henderson, Leech Lake Band of Ojibwe Botanist

Katie Zlonis, Leech Lake Band of Ojibwe, Plant Resources Program Manager

**Non-funded/in-kind partners:**

- Kirk Larson (Chippewa National Forest, Botanist/rare plants specialist, assist in survey and data collection).
- Dr. Don Farrar professor emeritus (Iowa State University, leading moonwort expert), support for Botrychium species.
- Dr. Cindy Johnson-Groh (Gustavus Adolphus College, Professor of Biology and Environmental Studies, moonwort expert) support for project design and protocol.

**B. Project Impact and Long-term Strategy:**

1. Provide data to support the degree of imperilment Goblin fern may be currently facing in Minnesota.
2. Facilitate the development of long term mitigation strategies and conservation plans to protect remaining Goblin fern habitat and populations.
3. Facilitate revisions to the current CNF Forest Plan.
4. Invoke more conscious decisions for updating and implementing BMPs in northern hardwood forests.
5. Provide data to support updating records in the CNF corporate database.
6. Provide Earthworm data to a much larger earthworm study being conducted by Great Lakes Worm Watch and UMD NRRRI.
7. Neighboring state agencies continue to express interest in the results of this project as Goblin fern faces extinction within neighboring states.

**C. Funding History:**

| Funding Source and Use of Funds | Funding Timeframe | \$ Amount |
|---------------------------------|-------------------|-----------|
| N/A                             |                   |           |

**IX. VISUAL COMPONENT or MAP(S):** See attached figures

**X. RESEARCH ADDENDUM:** N/A

**XI. REPORTING REQUIREMENTS:**

**Project Status as of October 30, 2015:**

Year one of project field work was completed in September 2015.

**Project Status as of April 15, 2016:**

Year one of project data entry was completed in February 2016.

**Project Status as of October 30, 2016:**

Year two of project field work was completed in August 2016.

**Project Status as of April 15, 2017:**

Year two of project data entry was completed in April 2017.

**Project Status as of October 30, 2017:**

Year three of project field work concluded in September 2017.

**Project Status as of April 15, 2018:**

**Retroactive Amendment Request (04/09/2018):**

**Project Status as of September 14, 2018:**

**The final report is being compiled at this time (final report scheduled for December 18 2018)**



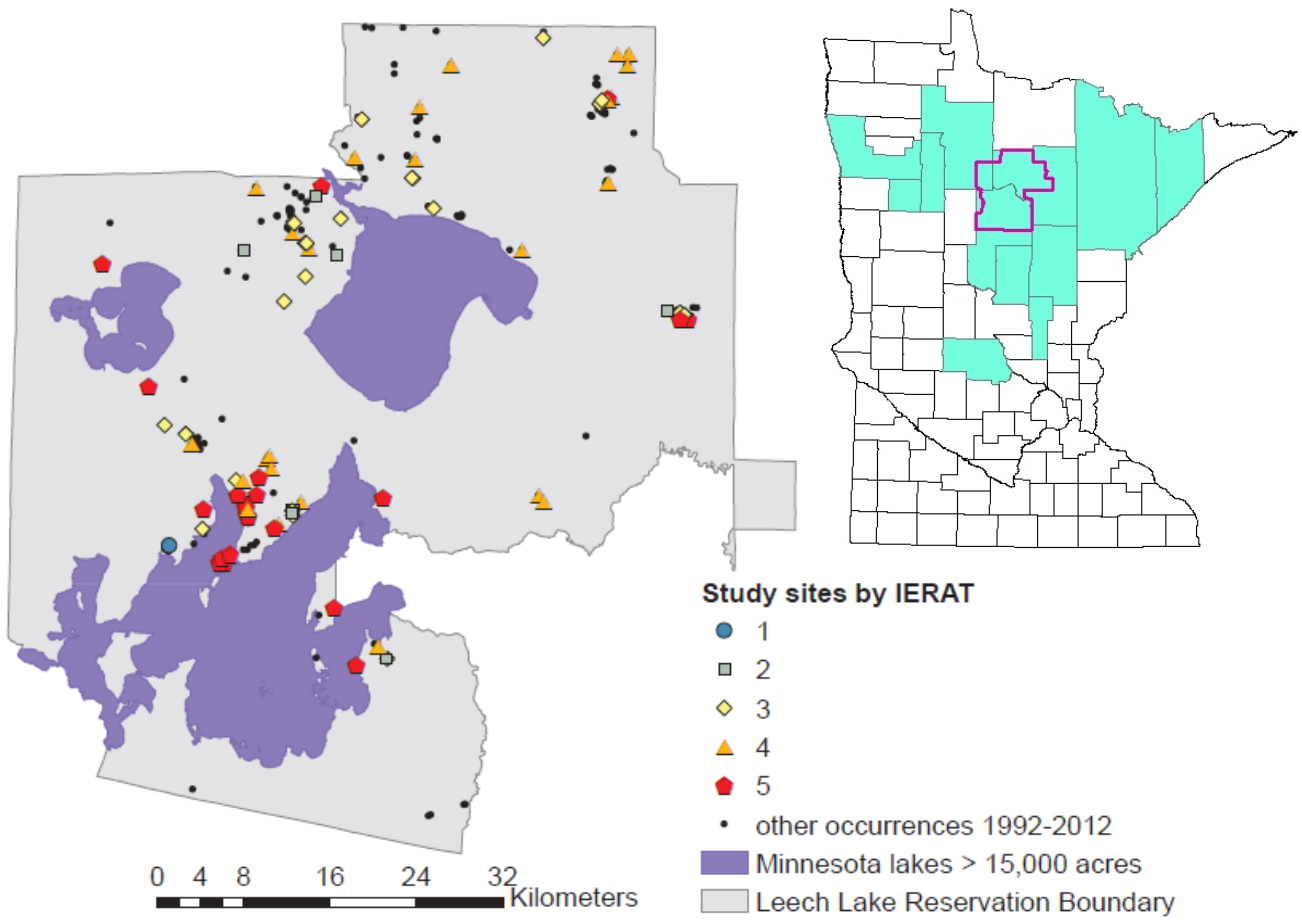


Figure 1. Map displaying general location of Chippewa National Forest/Leech Lake Reservation within Minnesota, with historic goblin fern range highlighted in blue. Leech Lake Reservation goblin fern study sites represented by IERAT stages: 1 – blue circle; 2 – green square; 3 – yellow diamond; 4 – orange triangle; 5 – red pentagon), additional historic occurrences documented from 1992 to 2012 (black dots).



*Figure 2. Goblin fern (Botrychium mormo); plant on left approximately 2.5cm tall, plant on right approximately 10cm tall. Typical size range for this species varies from .25cm – 12cm, with an average height of only 4cm. In locations where leaf duff is intact and healthy, the species often only peaks through the duff layer.*



Figure 3. Left: (IERAT 1) thick duff layer (O) horizon atop the mineral soil (E) horizon. The duff layer or humus layer greatly differs from “leaf litter”; duff forms a thick mat of leaf mould which holds moisture, creates a cool and stable microclimate for goblin fern and other organisms including mycorrhiza, plus the decomposing material is a consistent nutrient release. Right: (IERAT 5) duff layer (O) horizon has been completely removed and incorporated into the mineral soil. At IERAT 4 and 5 the distinct soil layers have been incorporated into a single dark layer (A) horizon as seen in the photo. At this stage of worm damage a number of forest health issues begin to occur; loss of habitat for organisms beneficial to forest health, microclimates at the soil level disappear, soils become highly compacted, dry, and erosion begins to occur.



*Figure 4. A healthy sugar maple and basswood forest floor complete with intact duff layer, abundant forest dwelling perennial plants, and sugar maple regeneration.*



*Figure 5. A severely earthworm damaged forest floor exhibiting greatly diminished plant diversity, abundance, and absence of tree seedling regeneration. At this stage, the forest soils are greatly altered and inhibitive of supporting a healthy forest.*

**Environment and Natural Resources Trust Fund  
FINAL M.L. 2015 Project Budget**



**Project Title: Habitat Mitigation for Goblin Fern Conservation**

**Legal Citation: M.L. 2015, Chp. 76, Sec. 2, Subd. 03s**

**Project Manager: Bobby Henderson**

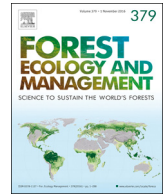
**Organization: Leech Lake Band of Ojibwe**

**M.L. 2015 ENRTF Appropriation: \$61,000**

**Project Length and Completion Date: 3 years, June 30, 2018**

**Date of Report: June 30, 2018**

| <b>ENVIRONMENT AND NATURAL RESOURCES TRUST<br/>FUND BUDGET</b>  | <b>Activity 1<br/>Budget</b>                                      | <b>Amount Spent</b> | <b>Activity 1<br/>Balance</b>   | <b>Activity 2<br/>Budget</b> | <b>Amount Spent</b> | <b>Activity 2<br/>Balance</b> | <b>TOTAL<br/>BUDGET</b> | <b>TOTAL<br/>BALANCE</b> |
|---|---|---------------------|---|------------------------------|---------------------|-------------------------------|-------------------------|--------------------------|
| <b>BUDGET ITEM</b>  | <b>Identify, Project Design, Survey, and Assess<br/>Locations</b> |                     | <b>Data Analysis, Submit Results to CNF,<br/>Assist in Development of Mitigation Plan for<br/>Updated CNF Forest Plan</b> |                              |                     |                               |                         |                          |
| <b>Personnel (Wages and Benefits)</b>   | \$28,600  | \$25,096.73         | \$3,503   | \$23,600                     | \$12,088.14         | \$11,512                      | \$52,200                | \$15,015                 |
| Bobby Henderson, project manager, botanist, data analysis<br>(80% salary, 20% benefits ) 35% FTE for 2 years; 20% FTE<br>for 1 year |   |                     |   |                              |                     |                               | \$42,091                |                          |
| Katie Zlonis, data analysis, manuscript preparation (80%<br>salary, 20% benefits) 15% FTE for 1 year                                |   |                     |   |                              |                     |                               | \$10,109                |                          |
| <b>Equipment/Tools/Supplies</b>   | \$2,050   |                     | \$1,557   |                              |                     |                               | \$2,050                 | \$1,557                  |
| 2 High precision GPS for relocating sites (\$1300)  |   |                     |   |                              |                     |                               |                         |                          |
| 2 Densimeters (\$220)   |   | \$225               |   |                              |                     |                               |                         |                          |
| Field supplies: collection bags, batteries, flagging, (\$530)   |   | \$268               |   |                              |                     |                               |                         |                          |
| <b>Travel expenses in Minnesota</b>   |   |                     |   |                              |                     |                               |                         |                          |
| Travel to, between, and from survey and data collection sites.<br>Mileage: \$6750   | \$6,750   | \$1,598             | \$5,152   |                              |                     |                               | \$6,750                 | \$5,152                  |
| <b>COLUMN TOTAL</b>   | <b>\$37,400</b>   | <b>\$27,188</b>     | <b>\$10,212</b>   | <b>\$23,600</b>              | <b>\$12,088</b>     | <b>\$11,512</b>               | <b>\$61,000</b>         | <b>\$21,724</b>          |



# Invasive earthworm damage predicts occupancy of a threatened forest fern: Implications for conservation and management



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## ARTICLE INFO

### Keywords:

Invasive earthworms  
Northern hardwood  
Occupancy modeling  
Detectability  
*Botrychium mormo*

## ABSTRACT

Adequate detection and monitoring of threatened, endangered, and sensitive species presents a challenge to forest managers seeking to balance management activities with conservation and forest health. This is especially true for rare, cryptic plant species that are difficult to detect, like goblin fern (*Botrychium mormo*), which is small and does not emerge from the duff layer of the rich hardwood forests it inhabits every year, even when present. Imperfect detection of this species makes it difficult to monitor, because lack of plants detected at a specific site does not necessarily indicate that the species has been extirpated there. In this study, 80 historic locations of *B. mormo* were surveyed for occurrence over three consecutive years to assess probability of occupancy and environmental factors expected to impact occupancy, including earthworm damage and canopy closure, while accounting for detectability. We found that probability of occurrence is most strongly related to earthworm damage and were able to identify levels of earthworm damage at which the species is more likely to remain present or be extirpated. These results suggest that use of a simple metric for quantifying ecological impact of earthworm damage can be used during monitoring to assess the likelihood that *B. mormo* is still present. With this information, forest managers can prioritize sites for habitat preservation and better shape policy and management decisions to protect and enhance habitat for this species. In addition, our study demonstrates the utility of occupancy modeling for management and conservation of rare and elusive plant species.

## 1. Introduction

Consideration of management impacts to threatened, endangered, and sensitive (TES) species is critical to forest management, especially for agencies that are under regulatory obligation to protect them (USDA). Developing appropriate policies and management strategies related to TES and other vulnerable species depends on precise knowledge of population locations, population viability, and factors contributing to any population declines. Inventory and monitoring programs are often designed to answer questions related to where populations are and how they are trending, but typically cannot identify causal agents of those trends (Elzinga et al., 1998). Further, traditional inventory and monitoring approaches may not be sufficient to adequately address any of these issues for elusive species that are difficult to detect.

Increasingly, imperfect detectability is recognized as an important factor to account for in research and monitoring (Kéry and Schmid, 2004). The assumption that a species is not present or has been extirpated at a site where it isn't detected often does not hold, since populations or individuals can be missed during surveys (Kéry, 2004).

Detectability estimates are now routinely incorporated into analysis of occupancy and abundance data for animals (MacKenzie et al., 2017). Although this approach has been used less frequently in plants and other sessile organisms (but see: Alexander et al., 2009; Berberich et al., 2016; Emry et al., 2011), detectability in plant surveys is typically less than one, and therefore important to account for (Chen et al., 2013). Without perfect detection, “false zeros” recorded when a species is present but not detected in a monitoring dataset can lead to biased under-estimates of occupancy (and over-estimates of extirpation) and misleading or incorrect results regarding the factors influencing these occupancy states (Chen et al., 2013; MacKenzie et al., 2017). Thus, using inferences from analyses based on inventory and monitoring programs that don't incorporate detectability to inform policy and management decisions may be highly problematic (Guillera-Arroita et al., 2014). Utilizing occupancy models that incorporate presence/absence data, while accounting for imperfect detectability and factors contributing to heterogeneity in detectability and occupancy, can be a valuable tool for overcoming these common obstacles in analysis of rare plant monitoring data.

*Botrychium mormo* (goblin fern), is an example of a rare species of

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conservation concern that is of particular importance to forest management in the Great Lakes states, but is difficult to monitor due to its small size and the fact that it does not emerge from the duff every year. It is endemic to the rich northern hardwoods of the western Great Lakes region including Minnesota, Wisconsin, and Michigan in the USA, and has one recorded occurrence in Quebec, Canada (USDA NRCS, 2018; Wagner Jr. and Wagner, 1993). The majority of known extant populations are concentrated in north-central Minnesota within the Chippewa National Forest, which overlaps 90% of the Leech Lake Reservation (Johnson-Groh and Lee, 2002; Mortensen and Mortensen, 1998). In the Chippewa National Forest, the rich northern hardwood forests this species depends on account for approximately less than 1% of forested land. Surveyors have searched this habitat extensively over the past twenty years for *B. mormo*, and over 600 element occurrences have been documented in the Chippewa National Forest. The species has state conservation ranks assigned to it throughout its entire range designating it as vulnerable to extirpation in Minnesota, imperiled in Michigan, and critically imperiled in Wisconsin and Quebec (NatureServe, 2017). It has not been reviewed for the Endangered Species Act in the United States, but is listed as endangered in Wisconsin (WI DNR, 2017) and threatened in Minnesota and Michigan (Michigan Natural Features Inventory, 2017; MN DNR, 2013).

The life history of *B. mormo* makes it particularly difficult to detect and verify occupancy. It is a small perennial fern that is known to lie dormant or fail to emerge from the duff in some years, likely due to unsuitable conditions such as drought (Wagner and Wagner, 1981), even when there is a thriving population of immature and mature plants below-ground (Johnson-Groh, 1998). Because of this, abundance of above-ground plants can vary significantly from year to year, which does not necessarily correlate with total abundance of above- and below-ground plants, and individuals or all plants from a population may fail to emerge in a given year (Johnson-Groh and Lee, 2002; Johnson-Groh et al., 2002). Thus, monitoring above-ground plant number, especially for only one year, would yield poor estimates of abundance and underestimates of occupancy. Incorporating detectability estimates derived from surveys over multiple years into occupancy models is one way to obtain unbiased estimates of probability of occurrence within sites, and can also be used to assess the effect of factors that may influence probability of occurrence (MacKenzie et al., 2017).

One factor known to negatively impact *B. mormo* is the invasion of non-native earthworms in the northern hardwood forests that harbor this species. *Botrychium mormo* requires a thick duff layer and organic soil horizon over mineral soil that is well to moderately well-drained and has loamy to silty texture (Casson et al., 2001). Personal field observation (Henderson) and Natural Resource Conservation Service (NRCS) soils mapping further indicate goblin fern hot spots typically have well-drained fine sandy loam to fine loamy sand soil textures, and are nearly level to undulating with 1–8 percent slopes (Soil Survey Staff). Within the humus layer, this species is dependent on associations with mycorrhizal fungi to obtain nutrients and water, as are other *Botrychium* species (Berch and Kendrick, 1982; Whittier, 1984; Winther and Friedman, 2007). Earthworms, especially *Lumbricus* spp., rapidly reduce organic soil layers to the detriment of *B. mormo* and other plants dependent upon intact soil O horizons in northern hardwood systems (Gundale, 2002; Hale et al., 2005a, 2006, 2008; Mortensen and Mortensen, 1998). While we know that earthworms have a significant adverse effect on this species, the level of worm damage necessary to make habitat unsuitable for *B. mormo* has not yet been quantified. This information would be highly valuable for managers to help determine when the species is likely extirpated from sites, as not finding plants on a given monitoring visit does not necessarily mean the species is absent.

In addition to soil characteristics, another factor that may influence *B. mormo* occupancy is canopy closure. This species has only been observed in forests with a relatively closed canopy (Casson et al., 2001). Shade is hypothesized to be important to *B. mormo* spores, which only

germinate in the dark (Whittier, 1973), and to maintain cool, moist conditions in the duff layer (Casson et al., 2001). *Botrychium mormo* is known to be sensitive to drought conditions, and can fail to emerge during drought years (Wagner and Wagner, 1981). An open canopy can create or exacerbate dry conditions on the forest floor, impacting species dependent on moisture in the duff layer (Harpole and Haas, 1999; Semlitsch et al., 2009). Factors such as earthworm invasion and canopy closure may contribute to heterogeneity in occupancy probability, and thus are important to include in occupancy models for this species.

During this three-year study, data on *B. mormo* occupancy and covariates, including earthworm invasion stage and canopy closure, were collected from historic *B. mormo* sites to answer the following three questions: (1) How do various levels of earthworm damage influence probability of occupancy? We expect that as earthworm damage increases, occupancy probability will decrease. (2) Does canopy closure influence probability of occupancy, and if so, how? We expect that decreased canopy closure may have a negative relationship with occupancy probability. (3) Do earthworm damage and canopy closure interact? We expect that the negative effects of earthworm damage may be buffered by increased canopy closure or may be exacerbated by decreased canopy closure. Results are discussed in the context of using occupancy modeling, including detectability, in monitoring efforts to inform conservation and forest management.

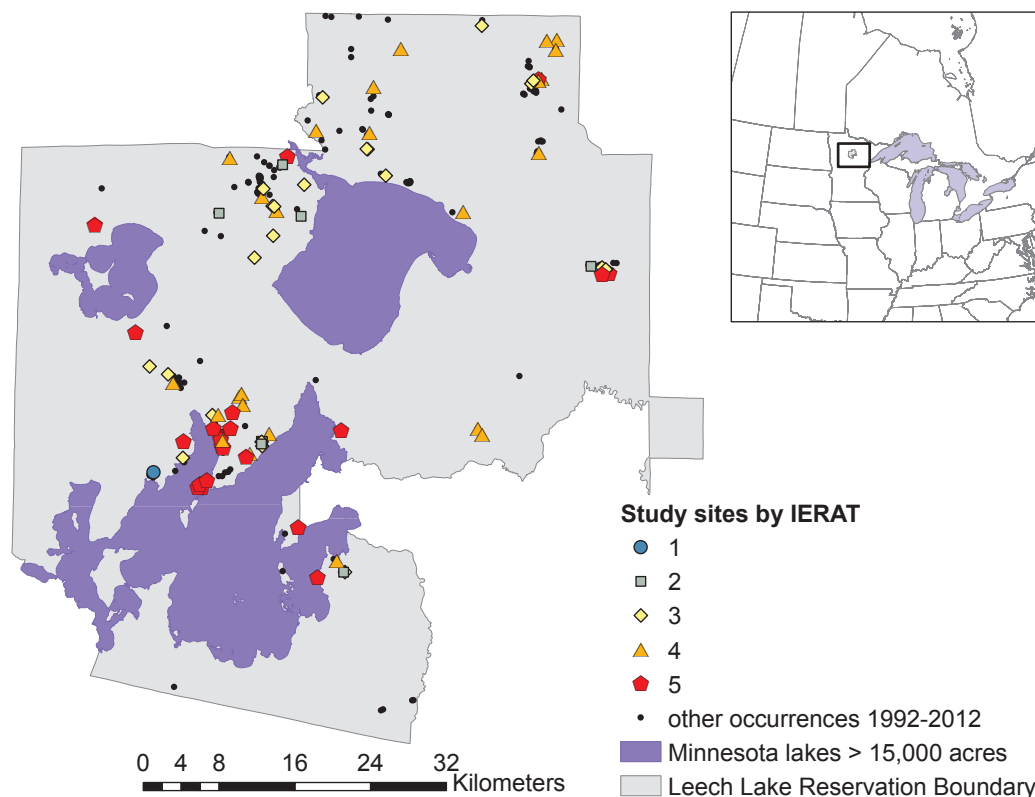
## 2. Methods

### 2.1. Data collection

To assess factors that may influence occurrence of *B. mormo*, locations where the species was historically documented were monitored in 80 randomly selected sites within the Leech Lake Reservation and Chippewa National Forest. For this experiment, a “site” is defined as the area where a population of a species has been documented. Site selection was stratified by choosing 20 random sites within four time periods of original date of detection (1994–1997, 1998–2002, 2003–2007, 2008–2011; Fig. 1). Because historical sites were used, our results apply to the population of sites where this species has been documented rather than the population as a whole. Sites were divided between two observers experienced in *Botrychium* surveys, who visited the same sites once per year over three years to increase likelihood of detection if the species was indeed occupying the site. Sites were relocated from original records based on GPS coordinates or coordinates inferred from field notes that identified sites based on unique features and bearings and distances from specific features (for records that originated before GPS was widely used). During initial relocation visits, a hybrid spiral/random meaner method was used for sites with coordinates. If historic occurrence sites had inferred coordinates, or had GPS coordinates but were not initially located through this method, a parallel transect method was used to more thoroughly search sites. Initial relocation efforts ranged from 1 to 6 h per site, with an average of 4 h per site (shorter times are associated with sites where the species was easily found near given coordinates). Surveys occurred between July and August, the time period during which above-ground abundance and individual plant size peak (Johnson-Groh and Lee, 2002).

During surveys, the following variables were recorded: presence/absence of above-ground *B. mormo*, abundance of *B. mormo* plants if detected, Invasive Earthworm Rapid Assessment Tool (IERAT) stage (Loss et al., 2013), and canopy closure (foliar volume measured with a densiometer). The IERAT stage was determined using a protocol developed by the Natural Resources Research Institute Great Lakes Worm Watch program, which takes into account several facets of worm damage to forest understories, including loss of duff layer and organic soil horizons, plant community diversity, and direct evidence of earthworms including castings, middens, and earthworms themselves, to rank ecological impact of worm invasion on a site (1–5, with 5 indicating the greatest impact; Loss et al., 2013). Canopy closure was





**Fig. 1.** Map showing general location of Leech Lake Reservation within Minnesota, USA and *Botrychium mormo* study sites by Invasive Earthworm Rapid Assessment Tool (IERAT) stage (stages: 1 – blue circle; 2 – green square; 3 – yellow diamond; 4 – orange triangle; 5 – red pentagon), with additional historic occurrences documented from 1992 to 2012 (black dots). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

measured using a model C concave spherical crown densiometer, with a cross-shaped grid of 24 quarter-inch squares.

## 2.2. Statistical analysis

Single-season occupancy models were used to model detectability of *B. mormo* and test effects of covariates on site occupancy (MacKenzie et al., 2017). Because *B. mormo* is a perennial plant that can have up to five years' worth of pre-formed leaf buds (Wagner, 1998), it was assumed that over a three-year period the occupancy state for this species is closed within survey sites (MacKenzie et al., 2017). After initial data exploration, several observations were removed from the final analyzed dataset. The lone site with an IERAT stage of one had *B. mormo* detected in all three surveys and was removed from analysis due to insufficient sampling of sites of this quality, which are rare across the landscape (Zuur et al., 2010). In addition, all 22 stage 5 sites were excluded from occupancy modeling, as *B. mormo* was not detected at any of these sites during any survey over the three-year survey period. Because no plants were observed at any site or visit for IERAT stage 5, analysis of this stage would yield inestimable standard errors for its occupancy probability estimate of 0. However, to ensure that excluding these observations did not impact results for stages 2–4, we also ran the analysis including all stage 5 observations. Results for model selection and beta coefficients of covariates were qualitatively the same, and results for occupancy estimates and their standard errors were the same, as the results when these observations were excluded. For this reason, we focus on occupancy analysis and results for IERAT stages 2–4 for the rest of the paper. Two additional sites were removed due to missing data for site-specific covariates. The final dataset included presence/absence and covariate data for 55 sites with the following distribution of IERAT stages: 2–7 sites, 3–22 sites, 4–26 sites.

Factors hypothesized to impact *B. mormo* occupancy were used to build a general occupancy model in PRESENCE (v. 12.7; Hines, 2006) to model detection probability. The initial general occupancy model included IERAT stage (IER; categories 2–4), canopy closure (CC), an interaction between IER and CC, and the original detection date (categories 1–4 based on dates site selection was stratified by). Canopy closure was transformed by dividing by 100 to improve model convergence, as suggested in the PRESENCE manual (Hines and MacKenzie). During initial model runs, original detection date was dropped from analysis because large standard errors, standard errors that were not estimable, and poor resolution on maximum likelihood estimates indicated that the model was likely over-parameterized given the sample size. In addition, there was no indication based on AIC that original detection date was a useful parameter for modeling occupancy, and original detection date is a difficult variable to interpret since it contains no information on how long the species has actually occupied a site, or what the population history (extirpation, colonization) has been at the site since the original detection. The final general model for occupancy was:

$$\text{logit}(\psi_i) = \alpha_0 + \alpha_1 \text{IER}_i + \alpha_2 \text{CC}_i + \alpha_3 \text{IER}_i \times \text{CC}_i$$

This general model was used to assess factors that may influence *B. mormo* detectability. Four models were compared including one indicating whether *B. mormo* had been previously detected at a site (PD), the year of the survey (YR), and the minimum number of plants recorded in the original detection (ODA, standardized by subtracting each value from the mean and dividing by the standard deviation to generate Z scores) (Table 1). The variable indicating whether the species had been previously detected was important to include because, since the same surveyor visited each site every year, the assumption of independent site visits was violated. Including a variable indicating

**Table 1**

Summary of detectability model results for goblin fern (*Botrychium mormo*) data.  $\Delta AIC$  is the difference in AIC between the model with lowest AIC (top ranked model),  $w$  is the AIC model weight,  $N_{par}$  is the number of parameters in the model, and  $-2ll$  is the  $-2$  log likelihood of the model. For all models the global occupancy model  $\psi(IER + CC + IERxCC)$  was used, where IER is the Invasive Earthworm Rapid Assessment Tool (IERAT) stage and CC is canopy closure.

| Model      | $\Delta AIC$ | $w$    | $N_{par}$ | $-2ll$ |
|------------|--------------|--------|-----------|--------|
| $p(PD)$    | 0.00         | 0.6667 | 7         | 145.43 |
| $p(ODA)$   | 2.89         | 0.1572 | 7         | 148.32 |
| $p(YR)$    | 3.57         | 0.1119 | 9         | 145.00 |
| $p(\cdot)$ | 4.68         | 0.0642 | 7         | 150.11 |

PD = species detected during previous survey; ODA = abundance from original detection; YR = year of survey.

whether the species had been previously detected allows this dependency to be modeled for detectability (MacKenzie et al., 2017). Combinations of variables with PD were modeled, but removed from the candidate model set because there was not sufficient data to model multiple detection parameters (as indicated by the same problems discussed above for modeling original detection date). The final general detectability model, best supported by AIC was:

$$\text{logit}(p_{ij}) = \beta_0 + \beta_1 PD_i$$

Using the above detectability model, all combinations of the general occupancy model were tested, including a model holding occupancy constant across sites. However, before testing simpler models, the global model (IER + CC + IERxCC as site covariates and PD as covariate on detectability) was used to assess model fit using 5000 bootstrap samples ( $X^2 = 3.58$ ,  $P$ -value = 0.8204,  $\hat{c} = 0.4267$ ). Results of the goodness-of-fit test show that the model is a reasonable fit for the data, that standard errors for model parameters do not require adjustment, and that simpler models derived from the global model will also be adequate to describe the data (MacKenzie and Bailey, 2004; MacKenzie et al., 2017). Final model selection was done using AIC to select the best supported model.

### 3. Results

*Botrychium mormo* was observed during at least one of three surveys at 27 out of 80 sites (33.75%), and the proportion of sites differed by IERAT stage. Of the randomly selected sites, 27.5% were assessed as IERAT stage 5, 32.5% stage 4, 27.5% stage 3, 8.75% stage 2, and 1.25% stage 1 (Table 2). *Botrychium mormo* was observed in at least one year at all sites with IERAT stages of 1 or 2, at 59% of sites with IERAT stage 3, 19% of sites with IERAT stage 4, and none of the sites with IERAT stage 5. For sites where the species was detected, all possible detection histories were represented, including plants being observed in 1/3 years,

**Table 2**

Summary data for all sites, excluding one site with missing data for IERAT stage, including abundance from historical occurrence records, abundance recorded across three-year study, and number of historical sites where species was or was not observed in at least one year during three-year study, by IERAT stage. Note historical abundance is reported from original detection records and is a minimum estimate of population size at the time of the observation.

| IERAT stage | Historical Data |      |      | Data from current three years of surveys |      |     |                          |    |                              |    |
|-------------|-----------------|------|------|--|------|-----|--------------------------|----|------------------------------|----|
|             | abundance       |      |      | abundance                                |      |     | # sites species observed |    | # sites species not observed |    |
|             | min             | mean | max  | min                                      | mean | max |                          |    |                              |    |
| 1           | 12              | 12   | 12+  | 1  | 3    | 4   | 1                        | 0  |                              | 1  |
| 2           | 1               | 5    | 10+  | 0  | 3    | 9   | 8                        | 0  |                              | 8  |
| 3           | 1               | 10   | 50+  | 0  | 1    | 12  | 13                       | 9  |                              | 22 |
| 4           | 1               | 28   | 500+ | 0  | < 1  | 5   | 5                        | 21 |                              | 26 |
| 5           | 2               | 23   | 500+ | 0  | 0    | 0   | 0                        | 22 |                              | 22 |

2/3 years, and 3/3 years (9 sites, 12 sites, and 6 sites respectively).

Abundance across all sites was relatively low with 42 plants observed across all sites in the first year of surveys, 62 plants in the second year, and 54 plants the third year. Several sites had only one plant observed, the maximum number observed at a site was 12, and the mean was three (Table 2). Number of plants observed also fluctuated among years at the same site, for example, at one site three plants were observed the first year, 9 the next, and six in the third year. As another example, no plants were observed at one site the first year, one was observed the second year, and five the last year. Plants were not observed in any of the five sites with the greatest original abundance (three sites with over 100 and two with over 500 plants minimum originally observed). Two of these sites were scored as stage 4 and three as stage 5 on the IERAT scale. Minimum abundance from the historical detection was not correlated with abundance during any of the three years of surveys for this study, whether all sites were considered (2015:  $r = -0.07$ ,  $P = 0.580$ ; 2016:  $r = -0.10$ ,  $P = 0.394$ ; 2017:  $r = -0.11$ ,  $P = 0.355$ ) or only those where plants were found during that year (2015:  $r = 0.31$ ,  $P = 0.277$ ; 2016:  $r = -0.12$ ,  $P = 0.623$ ; 2017:  $r = -0.19$ ,  $P = 0.410$ ).

For sites included in occupancy analysis (IERAT stage 2–4), there was no significant difference in canopy closure between sites where *B. mormo* was observed and not observed. Canopy closure ranged from 63% to 98% in sites where *B. mormo* was observed and 62–96% at sites where *B. mormo* was not detected. Mean canopy closure did not differ between sites where the species was observed compared to sites where the species was not observed (86% vs 81%;  $t = 1.39$ ,  $P = 0.17$ ).

The occupancy model best supported by AIC model selection modeled occupancy based solely on IERAT stage, and detectability based on whether the species had been previously found at a given site (Table 3). Although CC was in the second-best ranked model, and this model was within two AIC points of the top-ranked model, this variable is considered uninformative. Akaike’s Information Criterion penalizes models by increasing AIC 2 units for each additional model parameter (Johnson and Omland, 2004). Thus, if a model has one additional parameter and is within 2 units of the top model, as in this case, that additional parameter is considered uninformative (Arnold, 2010).

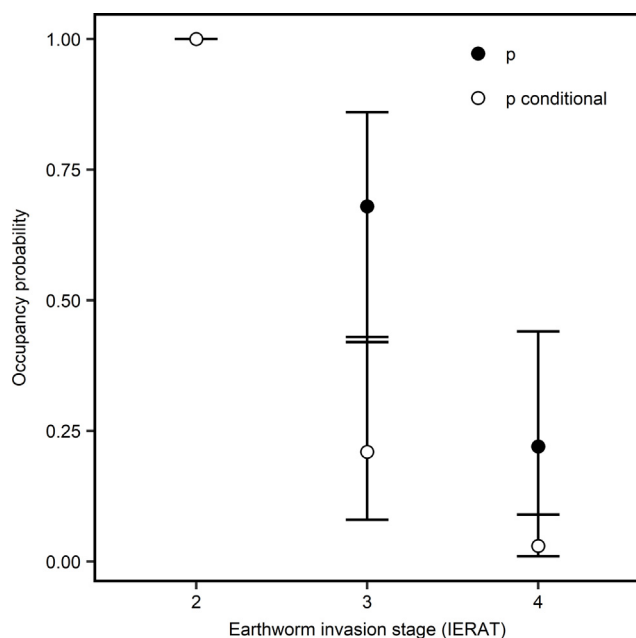
The naïve overall occupancy estimate, not incorporating detectability, for sites at IERAT stages 2, 3, or 4 was 0.45. Modeled occupancy probability for each site differed based on the IERAT stage of that site (Fig. 2). Sites at IERAT stage 2 had a probability of 1 that the species is still occupying the site. As IERAT stage increases, estimated probability that the site is still occupied decreases to 0.68 (95% CI 0.42–0.86) for sites at IERAT stage 3 and to 0.22 (0.09–0.44) for sites at IERAT stage 4. Using these estimates to predict the proportion of extant sites within each IERAT stage, and assuming that the species still exists at the few sites with IERAT stage 1 and is extirpated on all IERAT stage 5 sites, we estimate that the proportion of extant sites across all IERAT stages is 38% (25–49%). When previous detection history is taken into account (whether the species was found during at least one survey over the

**Table 3**

Summary of occupancy models fit to goblin fern (*Botrychium mormo*) data.  $\Delta AIC$  is the difference in AIC between the model with lowest AIC (top ranked model),  $w$  is the AIC model weight,  $Npar$  is the number of parameters in the model, and  $-2ll$  is the  $-2$  log likelihood of the model. For all models the detection model  $p$  (PD) was used, where PD indicates whether the species was detected at a site on a previous visit.

| Model                     | $\Delta AIC$ | $w$    | $Npar$ | $-2ll$ |
|---------------------------|--------------|--------|--------|--------|
| $\psi(IER)$               | 0.00         | 0.4827 | 4      | 147.53 |
| $\psi(IER + CC)$          | 0.15         | 0.4479 | 5      | 145.68 |
| $\psi(IER + CC + IERxCC)$ | 3.90         | 0.0687 | 7      | 145.43 |
| $\psi(CC)$                | 14.38        | 0.0004 | 3      | 163.91 |
| $\psi(\bullet)$           | 14.54        | 0.0003 | 2      | 166.07 |

IER = Invasive Earthworm Rapid Assessment Tool (IERAT) stage, CC = canopy closure,  $\psi$  = probability of occupancy,  $\psi(\bullet)$  = model with no covariates



**Fig. 2.** Probability of occurrence for *B. mormo* by Invasive Earthworm Rapid Assessment Tool (IERAT) stage, showing both probability ( $p$ ) and probability conditional on having found the species at least once at a site. Bars represent 95% confidence intervals. Note  $p$  and  $p$  conditional are the same at IERAT stage 2.

three-year study period), these occupancy probabilities decrease for sites where the species was not observed. For sites at IERAT stage 3 where *B. mormo* was not observed during any surveys over three years, estimated probability of occupancy dropped to 0.21 (95% CI: 0.08–0.43), and for sites at IERAT stage 4, estimated probability of occupancy plummets to 0.03 (0.01–0.09).

Detection probability in the first year, and for sites where the species had not yet been observed during the study period, was 0.50. Once

**Table 4**

Summary of regression coefficients (SE) for occupancy models fit to goblin fern (*Botrychium mormo*) data;  $w$  is the AIC model weight.

| Model                     | $w$  | Intercept    | IER3          | IER4          | CC           | IER3xCC       | IER4xCC       |
|---------------------------|------|--------------|---------------|---------------|--------------|---------------|---------------|
| $\psi(IER)$               | 0.48 | 29.40 (2.70) | –28.67 (2.70) | –30.66 (2.71) | –            | –             | –             |
| $\psi(IER + CC)$          | 0.45 | 30.46 (2.90) | –33.88 (2.77) | –35.74 (2.77) | 4.83 (3.08)  | –             | –             |
| $\psi(IER + CC + IERxCC)$ | 0.07 | 16.49 (5.19) | –21.90 (5.93) | –20.58 (5.56) | 18.85 (7.83) | –11.62 (9.59) | –15.42 (8.09) |
| $\psi(CC)$                | 0.00 | –3.43 (2.40) | –             | –             | 4.23 (2.88)  | –             | –             |
| $\psi(\bullet)$           | 0.00 | 0.09 (0.31)  | –             | –             | –            | –             | –             |

IER = Invasive Earthworm Rapid Assessment Tool (IERAT) stage, CC = canopy closure,  $\psi$  = probability of occupancy,  $\psi(\bullet)$  = model with no covariates holding occupancy constant across sites.

the species was observed at a site (by the same observer), mean detection probability increased to 0.73 (95% CI 0.55–0.86).

Negative coefficients for IERAT stages 3 and 4, relative to IERAT stage 2, show decreasing log odds that *B. mormo* occupies a site as earthworm damage progresses (Table 4). The positive intercept term indicates that the log odds of occupancy are high at sites with IERAT stage 2. As IERAT stage increases to 3 and 4, the magnitude of negative coefficients with relatively small standard errors indicates an enormous negative effect on occupancy. This results in odds ratios near 0 (i.e.  $3.5 \times 10^{-13}$  for IERAT stage 3), which indicates occupancy is much less likely in sites with IERAT stages greater than 2. The log odds presented here are relatively large for logistic regression, and their magnitude is driven by comparison with IERAT stage 2, which had comparatively few observations (sites of this quality are rare on the landscape), and positive species detection within all sites in at least one year of surveys. Because of this, the beta coefficients comparing log odds with IERAT stages that are less likely to be occupied in this analysis are inflated. Nevertheless, these results make sense biologically, and we feel they are important to report and document due to the increasing rarity of high quality sites on the landscape and the striking effect of degradation of habitat suitability beyond this level of worm infestation.

#### 4. Discussion

##### 4.1. Earthworm effects and canopy closure

Botanists and ecologists have long known that earthworm damage to northern hardwood forest floors degrades habitat for species like *B. mormo*, a threatened plant of particular relevance to forest management in the Great Lakes states. This species depends on a relatively thick organic soil horizon and duff layer, which earthworms, especially *Lumbricus* spp., rapidly destroy in a matter of years after invasion (Gundale, 2002; Hale et al., 2006, 2008; Mortensen and Mortensen, 1998). Our results agree with other work showing that non-native earthworms strongly drive ecosystem changes resulting in declines and extirpation of *B. mormo* populations (Gundale, 2002). Further, we identify levels of worm damage most likely to negatively impact *B. mormo* occupancy and show that a simple, quick protocol, the Invasive Earthworm Rapid Assessment Tool, can be effectively used to describe and monitor worm damage to assess *B. mormo* habitat.

Most importantly, our results from three years of sampling allow us to identify a threshold in habitat quality after worm invasion for *B. mormo* (Question 1). Over 90% of sites within the Leech Lake Reservation, the stronghold for occurrences within this species' range, have degraded habitat such that *B. mormo* no longer occupies historic sites or is in peril (IERAT  $\geq 3$ ). At IERAT stage 2, there is a high probability that *B. mormo* still occupies sites. This stage is characterized by invasion of *Dendrobaena octaedra* (Loss et al., 2013), an epigeic species that lives and feeds in leaf litter without reducing forest floor thickness (Hale et al., 2005b), and does not negatively impact *B. mormo* (Gundale, 2002). Sites at IERAT stage 3 still have a relatively high occupancy probability, but between stages 3 and 4 probability of occupancy decreases below 50%, to a mean of 0.22. Especially for sites categorized as IERAT 4, it is very unlikely that *B. mormo* still occupies a

site if it hasn't been observed during three consecutive years of surveys by an experienced surveyor. At IERAT stage 5, we find no evidence that sites can continue to support *B. mormo*. This tipping point corresponds to invasion of *Lumbricus rubellus*, an epi-endogeic species which rapidly consumes litter and organic soil horizons (Hale et al., 2005b) at IERAT stage 3. The final stages of invasion correspond to initial introduction and peak of *L. terrestris* at IERAT stages 4 and 5 (Loss et al., 2013). *Lumbricus terrestris* is an anecic species that invades sites after the forest floor has been impacted by other species, such as *L. rubellus* (Hale et al., 2005b). It burrows deep within the soil and can consume all litter each year, preventing the forest floor and organic soil horizon from recovering after earthworm invasion (Hale et al. 2005b). Our results support Gundale's (2002) work showing invasion of *L. rubellus* is associated with decline in *B. mormo* populations; however, our results suggest that *L. terrestris* invasion, or a combination of species with *L. terrestris*, at stages 4 and 5 is most detrimental to *B. mormo*. Alternatively, the existing populations at stage 3 may represent a lag effect on extirpation if sites inevitably proceed to stage 4. Although earthworm species present in earlier IERAT stages can facilitate invasion of species associated with later stages (Hale et al., 2005b), introduction of those species to sites is still necessary (Holdsworth et al., 2007). At the current level of infestation, our study suggests that 60% of historical *B. mormo* sites are already unsuitable and that the species is either extirpated or in immediate peril at sites with IERAT stages 5 and 4 respectively. Continued monitoring of these sites, which is planned for the next 10 years, will help to elucidate how invaded sites progress in terms of IERAT stage and provide more precise information on the conditions that lead to population extinction, especially within IERAT stages 3 and 4.

Aside from an intact forest floor, canopy closure is hypothesized to be important to providing necessary darkness and moisture to *B. mormo* plants, but this factor was uninformative for predicting occupancy in this study (Question 2). Similarly, there was no evidence for a significant interaction between canopy closure and IERAT on occupancy (Question 3). It is critical to realize that while canopy closure didn't provide any explanatory power beyond IERAT for these sites, that does not mean canopy closure isn't important for this species. This species has only been found in sites with relatively high canopy closure (Casson et al., 2001). The randomly selected sites in this study ranged from over 60% to over 90% canopy closure, which did not differ between sites that were and were not occupied over the three years of this study. Further, models that included canopy closure had large positive regression coefficients suggesting a positive relationship between canopy closure and occupancy. Taken together, these results suggest that the range of canopy closure for all sites in this study is within the range that defines suitable habitat for this species. This is an important issue as it impacts forest management decisions, for example, buffer size around plants of conservation concern (Casson et al., 2001). While more research is needed to determine the effect of harvest on *B. mormo* populations, anecdotal evidence suggests that clearcutting results in extirpation of populations while selective harvests that don't directly impact plants, or the duff layer and organic soil horizons they depend on, may allow populations to persist (Casson et al., 2001; Sather et al., 1998). In addition, our results provide no insight into the potential importance of different forest strata to providing canopy coverage, which can be patchy to continuous in the mesic hardwood forests where *B. mormo* is found (MN DNR, 2003).

#### 4.2. Occupancy and abundance

This study illustrates the importance of designing monitoring efforts based on species' life history attributes and including detectability in analysis of monitoring data for plants and TES species. For *B. mormo*, managers cannot count on a single survey by a naïve observer to determine whether this species is present during one monitoring visit in a single year. Our results show that even for very experienced observers,

it is just as likely that plants will be found when present than that they will not, unless the same observer has already found *B. mormo* at a given site. In addition, for a majority of sites where the species was documented as present during the three years of sampling, plants were not observed in all years. This underscores the importance of monitoring this species over several years to determine whether a site is occupied, and using habitat information, especially IERAT, to assess the likelihood that the species is still present in sites that were historically occupied for making management decisions. Our results also highlight the importance of modeling detection probability for this species in assessing site occupancy, because, at best, experienced botanists who have already found the species at a site have a probability less than 0.75 of detecting the species, which will lead to biased underestimates of occupancy if detectability is not incorporated in analysis.

While this study focused on occupancy, some notes can be made regarding population abundance. Observed abundance within populations during the three years of sampling was relatively low and variable, which agrees with other research and reports (Casson et al., 2001; Johnson-Groh and Lee, 2002). Since *B. mormo* was first officially recorded as a Regional Forest Sensitive Species (RFSS) within the Chippewa National Forest (CNF) in 1992, over 600 historic records have been documented, most of which were recorded between the late 1990's and 2012. The overwhelming majority of these historical observations had small population counts, typically ranging between 1 and 8 individuals (CNF, unpublished data), which is similar to abundances observed in our three years of contemporary surveys. In one report from the CNF in 2001 (Casson et al., 2001), 79% of 116 populations were documented as having 20 or fewer above-ground plants. Large fluctuations in countable population size from year to year and lack of correlation inhibits making inferences about abundance for this species. More sophisticated analysis of abundance would be valuable, but given the life-history of *B. mormo*, existing statistical approaches may not be valid. For example, the Royle-Nichols abundance model assumes a demographically closed population across the study period, which likely does not hold over three years, even for this perennial species (Denes et al., 2015). Also, sampling was not standardized to occur during peak population size in mid-July (Johnson-Groh and Lee, 2002), which could bias results. A more sophisticated approach may be possible (Dail and Madsen, 2011), but we don't have precise estimates of the demographic rates, especially for underground plants (Berlin et al., 1998), necessary to model abundance to overcome the assumption of a closed population between, or even within, seasons.

Greater abundance does not appear to have buffered the species from extirpation at sites with high worm impacts; the species appears to be extirpated from all of the sites with the largest historical populations, which were documented as having over 100 individuals. Sites that originally harbored these large populations are now heavily impacted by worms and are at IERAT stage 4 or 5. This point highlights the importance of historical data in this study. Without knowing that the species had once occurred, and as large populations, in sites that now have IERAT stages of 4 or 5, we may have inferred, for example, that *L. terrestris* and *B. mormo* potentially inhabit different habitat niches. Several hypotheses may help to explain the complete loss of *B. mormo* from the sites in this study that historically had large populations. Large populations may have always been rare, making up just 3% of occurrences with at least 1 verified plant between 1992 and 2012 on the Chippewa National Forest (Chippewa National Forest, unpublished data). One hypothesis is that the area where most of these populations occurred on Ottertail Point, a large peninsula that juts into Leech Lake from the north, experienced earthworm invasions relatively early (possibly through release or escape of worms used for fishing bait) and are showing the result of prolonged and successive damage. This hypothesis is supported by documentation of *Lumbricus* spp. on the peninsula for over 20 years and evidence from tree rings of earthworm fronts on the peninsula dating to the 1960s (Hale et al., 2005a; Larson et al., 2010). Further, data from this area show that below-ground

abundance of *B. mormo* decreases following decrease in above-ground abundance in response to worm invasion (Johnson, 2015). A second hypothesis is that the unique rich hardwood forests in this area contained exceptional habitat characterized by a deep duff layer that facilitated both large populations of *B. mormo* in the past and explosive growth in the worm population after invasion (Curry, 1998). Third, plant-soil feedbacks, positive or negative, could influence population abundance through potential mechanisms such as build-up of pathogens in the soil or changes to mycorrhizal communities *Botrychium* depend on to obtain nutrients (Bennett et al., 2017; Klironomos, 2002).

#### 4.3. Conservation and management recommendations

In light of the severe decline in occupancy observed in this study, we recommend a conservative approach to management within the vicinity of known *B. mormo* populations and conservation of high quality habitat. Human mediated transport of worms is the most important factor influencing earthworm spread across the landscape (Cameron et al., 2007; Hale, 2008). Use of worm species, especially *Lumbricus* spp., as fishing bait is a major vector of earthworm introduction, and cabin and boat landing locations are associated with spread of these species (Holdsworth et al., 2007). Vehicles and equipment, including ATVs and logging equipment (Hale et al., 2009), transport both live worms and cocoons, and distance to roads is a significant predictor of earthworm presence, especially for *Lumbricus* spp. (Holdsworth et al., 2007). Within the Chippewa National Forest, recommendations currently restrict forest management activity within a 250' buffer around known occurrences and limit harvest activity beyond 250' to winter harvest (Casson et al., 2001). Earthworm eggs and cocoons can survive temperatures below freezing (Holmstrup, 1994), so even if the ground is not disturbed, earthworm cocoons inadvertently picked up and transported by equipment during winter could pose a threat to *B. mormo* populations. Once a site has been invaded, worms on their own can spread 5–10 m per year, meaning that it would take approximately 7–15 years to overcome the 250' buffer (Hale, 2008). However, with continuous facilitated spread and introduction, for example along an ATV trail or road, this can occur much more rapidly (Hale et al., 2009). Thus restricting motor vehicle and equipment use, practicing stringent equipment cleaning within and around sites harboring habitat not yet significantly impacted by worms, and educating foresters and other users of forest roads regarding earthworm damage and spread may help prevent further destruction to northern hardwood forests (Hale et al., 2009).

In addition to addressing spread of worms through management, we recommend identifying and protecting sites of high quality habitat for *B. mormo*. First, an inventory of high quality mesic northern hardwood and *B. mormo* sites should be conducted to identify areas in early IERAT stages. Ideally, this information would be collected each time a northern hardwood stand is surveyed, which would allow decoupling the effects of worm damage and management, and detecting potential interactions between those factors. Based on that information, we recommend designating and protecting areas that are the appropriate native plant community (in Minnesota primarily MHn35 and MHn47, Northern Mesic Hardwood Forest and Northern Rich Mesic Hardwood Forest; J. Almendinger, personal communication; Casson et al., 2001; MN DNR, 2003) and have an IERAT stage of 2 or less as “critical habitat” for *B. mormo*. Such sites could be buffered by ecological unit rather than distance to help prevent earthworm spread. For example, management could be limited to stands buffered from *B. mormo* sites or critical *B. mormo* habitat by other plant communities that may be less prone to supporting large worm populations, including fire-dependent sites with mor humus and dry, sandy soil that is relatively poor in organic matter, or acidic wetlands (Curry, 1998; Edwards and Bohlen, 1996; Parkinson and McLean, 1998). Protecting suitable habitat, even if not yet occupied, may be extremely important to *B. mormo*'s persistence on the landscape in the future, because animals are considered the

primary dispersal agent for spores (Wagner and Wagner, 1993). If deposited on suitable habitat, even a single *B. mormo* spore has the potential to germinate and grow into a reproducing plant, since gametophytes produced from individual spores produce both sperm and eggs, and this species reproduces almost exclusively via self-fertilization in this manner (Farrar and Johnson-Groh, 1990).

#### 4.4. Conclusions

Our results showing that goblin fern is extirpated from over 50% of historic sites, and in peril at another 40% of sites, underscores the importance and urgency of implementing conservation measures for this species. Reduction in probability of occupancy is best described by earthworm damage to sites and we show that IERAT can be used as a simple and fast method of assessing habitat quality for this species. Human mediated transport of earthworms, especially at boat landings and along roads and trails, is the primary means of their dispersal and spread across the landscape, rather than natural expansion of worm fronts (Hale, 2008). Because natural earthworm spread is slow and some areas are not yet impacted by earthworms, or at least the most destructive species, policies, regulations, and education aimed at restricting movement of earthworms and their cocoons are the most likely ways in which we can protect sites from infestation for a very long time. Such policies are especially important to implement now, to prevent infestation of additional, and more destructive species of Asian worms that are beginning to invade hardwood forests in the Great Lakes (Qiu and Turner, 2017). These efforts will help protect the future integrity of the northern hardwood ecosystem as a whole, which supports many ecologically and culturally important species, including sugar maple and wildlife species, that are also negatively impacted by earthworm invasion (Hale 2004; Loss and Blair, 2011; Loss et al. 2012). Finally, our study underscores the importance of appropriate monitoring and analysis techniques to assess occupancy, even for sessile species, that take life history into account.

#### Declarations of interest

None.

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#### References

- Alexander, H.M., Slade, N.A., Kettle, W.D., Pittman, G.L., Reed, A.W., 2009. Detection, survival rates and dynamics of a cryptic plant, *Asclepias meadii*: applications of mark-recapture models to long-term monitoring studies. *J. Ecol.* 97, 267–276.
- Arnold, T.W., 2010. Uninformative parameters and model selection using Akaike's Information Criterion. *J. Wildl. Manage.* 74, 1175–1178.
- Bennett, J.A., Maherali, H., Reinhart, K.O., Lekberg, Y., Hart, M.M., Klironomos, J., 2017. Plant-soil feedbacks and mycorrhizal type influence temperate forest population dynamics. *Science* 355, 181–184. <https://doi.org/10.1126/science.aai8212>.
- Berberich, G.M., Dormann, C.F., Klimeczek, D., Berberich, M.B., Sanders, N.J., Ellison, A.M., 2016. Detection probabilities for sessile organisms. *Ecosphere* 7, e01546. <https://doi.org/10.1002/ecs2.1546>.
- Berch, S.M., Kendrick, B., 1982. Vesicular-arbuscular mycorrhizae of southern Ontario

- ferns and fern-allies. *Mycologia* 74, 769–776.
- Berlin, N., Miller, P., Borovansky, J., Seal, U.S., Byers, O., 1998. Population and Habitat Viability Assessment (PVHA) Workshop For The Goblin Fern (*Botrychium mormo*): Final Report. Conservation Breeding Specialist Group, Apple Valley, Minnesota.
- Cameron, E.K., Bayne, E.M., Clapperton, M.J., 2007. Human-facilitated invasion of exotic earthworms into northern boreal forests. *Ecoscience* 14, 482–490.
- Casson, J., Shackelford, I., Parker, L., Schultz, J., 2001. Conservation approach for goblin fern, *Botrychium mormo* W.H. Wagner. USDA Forest Service, Eastern Region.
- Chen, G., Kéry, M., Plattner, M., Ma, K., Gardner, B., 2013. Imperfect detection is the rule rather than the exception in plant distribution studies. *J. Ecol.* 101, 183–191.
- Curry, J.P., 1998. Factors Affecting Earthworm Abundance in Soils. In: Edwards, C.A. (Ed.), *Earthworm Ecology*, first ed. St. Lucie Press, New York, pp. 37–64.
- Dail, D., Madsen, L., 2011. Models for estimating abundance from repeated counts of an open metapopulation. *Biometrics* 67, 577–587.
- Denes, F.V., Silveira, L.F., Beissinger, S.R., 2015. Estimating abundance of unmarked animal populations: accounting for imperfect detection and other sources of zero inflation. *Methods Ecol. Evol.* 6, 543–556.
- Edwards, C.A., Bohlen, P.J., 1996. *Biology and ecology of earthworms*. Springer Science & Business Media, pp. 3.
- Elzinga, C.L., Salzer, D.W., Willoughby, J.W., 1998. *Measuring & Monitoring Plant Populations*. US Bureau of Land Management Papers, 17.
- Emry, D.J., Alexander, H.M., Tourtellot, M.K., 2011. Modelling the local spread of invasive plants: importance of including spatial distribution and detectability in management plans. *J. Appl. Ecol.* 48, 1391–1400. <https://doi.org/10.1111/j.1365-2664.2011.02050.x>.
- Farrar, D.R., Johnson-Groh, C.L., 1990. Subterranean sporophytic gemmae in moonwort ferns, *Botrychium* subgenus *Botrychium*. *Am. J. Bot.* 77, 1168–1175. <https://doi.org/10.2307/2444627>.
- Guillera-Arroita, G., Lahoz-Monfort, J.J., MacKenzie, D.I., Wintle, B.A., McCarthy, M.A., 2014. Ignoring imperfect detection in biological surveys is dangerous: a response to ‘fitting and interpreting occupancy models’. *PLoS ONE* 9, e99571.
- Gundale, M.J., 2002. Influence of exotic earthworms on the soil organic horizon and the rare fern *Botrychium mormo*. *Conserv. Biol.* 16, 1555–1561.
- Hale, C.M., 2004. *Ecological Consequences of Exotic Invaders: Interactions Involving European Earthworms and Native Plant Communities in Hardwood Forests* (PhD Thesis). University of Minnesota St. Paul, MN.
- Hale, C.M., 2008. Evidence for human-mediated dispersal of exotic earthworms: support for exploring strategies to limit further spread. *Mol. Ecol.* 17, 1165–1167.
- Hale, C.M., Frelich, L.E., Reich, P.B., Pastor, J., 2005a. Effects of European earthworm invasion on soil characteristics in northern hardwood forests of Minnesota, USA. *Ecosystems* 8, 911–927.
- Hale, C.M., Frelich, L.E., Reich, P.B., 2005b. Exotic European earthworm invasion dynamics in northern hardwood forests of Minnesota, USA. *Ecol. Appl.* 15, 848–860.
- Hale, C.M., Frelich, L.E., Reich, P.B., 2006. Changes in hardwood forest understory plant communities in response to European earthworm invasions. *Ecology* 87, 1637–1649.
- Hale, C.M., Frelich, L.E., Reich, P.B., Pastor, J., 2008. Exotic earthworm effects on hardwood forest floor, nutrient availability and native plants: a mesocosm study. *Oecologia* 155, 509–518.
- Hale, C.M., Knowles, R., Anderson, B., 2009. Non-native earthworms transported on treads of ATVs and logging equipment in northern hardwood forests of Minnesota, USA. In: *Final Report for Environment and Natural Resources Trust Fund: Prevention and early detection of Asian earthworms and reducing spread of European earthworms*.
- Harpole, D.N., Haas, C.A., 1999. Effects of seven silvicultural treatments on terrestrial salamanders. *For. Ecol. Manage.* 114, 349–356.
- Hines, J.E., 2006. PRESENCE- Software to estimate patch occupancy and related parameters. USGS-PWRC. <http://www.mbr-pwrc.usgs.gov/software/presence.html> > .
- Hines J.E., MacKenzie, D.I., Program PRESENCE ver 12.8. < [https://www.mbr-pwrc.usgs.gov/software/doc/presence/presence\\_doc.html](https://www.mbr-pwrc.usgs.gov/software/doc/presence/presence_doc.html) > .
- Holdsworth, A.R., Frelich, L.E., Reich, P.B., 2007. Regional extent of an ecosystem engineer: earthworm invasion in northern hardwood forests. *Ecol. Appl.* 17, 1666–1677.
- Holmstrup, M., 1994. Physiology of cold hardiness in cocoons of five earthworm taxa (Lumbricidae: Oligochaeta). *J. Comp. Physiol. B* 164, 222–228.
- Johnson, C., 2015. Moonwort disturbance ecology: ten year resurvey baseline study Pigeon Lake Dam. Final Report.
- Johnson, J.B., Omland, K.S., 2004. Model selection in ecology and evolution. *Trends Ecol. Evol.* 19, 101–108.
- Johnson-Groh, C.L., 1998. Population demographics, underground ecology, and phenology of *Botrychium mormo*. In: Berlin, N., Miller, P., Borovansky, J., Seal, U.S., Byers, O. (Eds.), *Population and habitat viability assessment (PHVA) for the goblin fern (Botrychium mormo)*, Final Report. Conservation Breeding Specialist Group, Apple Valley, Minnesota, USA, pp. 103–109.
- Johnson-Groh, C.L., Lee, J.M., 2002. Phenology and demography of two species of *Botrychium* (Ophioglossaceae). *Am. J. Bot.* 89, 1624–1633.
- Johnson-Groh, C., Riedel, C., Schoessler, L., Skogen, K., 2002. Belowground distribution and abundance of *Botrychium* gametophytes and juvenile sporophytes. *Am. Fern J.* 92, 80–92.
- Kéry, M., 2004. Extinction rate estimates for plant populations in revisitation studies: importance of detectability. *Conserv. Biol.* 18, 570–574.
- Kéry, M., Schmid, H., 2004. Monitoring programs need to take into account imperfect species detectability. *Basic Appl. Ecol.* 5, 65–73. <https://doi.org/10.1078/1439-1791-00194>.
- Klironomos, J.N., 2002. Feedback with soil biota contributes to plant rarity and invasiveness in communities. *Nature* 417, 67–70. <https://doi.org/10.1038/417067a>.
- Larson, E.R., Kipfmüller, K.F., Hale, C.M., Frelich, L.E., Reich, P.B., 2010. Tree rings detect earthworm invasions and their effects in northern hardwood forests. *Biol. Invasions* 12, 1053–1066.
- Loss, S.R., Blair, R.B., 2011. Reduced density and nest survival of ground-nesting songbirds relative to earthworm invasions in northern hardwood forests. *Conserv. Biol.* 25, 983–992.
- Loss, S.R., Niemi, G.J., Blair, R.B., 2012. Invasions of non-native earthworms related to population declines of ground-nesting songbirds across a regional extent in northern hardwood forests of North America. *Landscape Ecol.* 27, 683–696.
- Loss, S.R., Hueffmeier, R.M., Hale, C.M., Host, G.E., Sjøerven, G., Frelich, L.E., 2013. Earthworm invasions in northern hardwood forests: a rapid assessment method. *Natural Areas J.* 33, 21–30.
- MacKenzie, D.I., Bailey, L.L., 2004. Assessing the fit of site-occupancy models. *J. Agric. Biol. Environ. Statistics* 9, 300–318.
- MacKenzie, D.I., Nichols, J.D., Royle, J.A., Pollock, K.H., Bailey, L., Hines, J.E., 2017. *Occupancy Estimation and Modeling: Inferring Patterns and Dynamics of Species Occurrence*, second ed. Elsevier.
- Michigan Natural Features Inventory. 2017. Michigan’s special plants: endangered, threatened, special concern, and probably extirpated. < [https://mnfi.anr.msu.edu/data/special\\_plants\\_list.pdf](https://mnfi.anr.msu.edu/data/special_plants_list.pdf) > .
- MN DNR. 2003. Field guide to the native plant communities of Minnesota. St. Paul, MN.
- MN DNR. 2013. Minnesota’s list of endangered, threatened, and special concern species. < [http://files.dnr.state.mn.us/natural\\_resources/ets/endlist.pdf](http://files.dnr.state.mn.us/natural_resources/ets/endlist.pdf) > .
- Mortensen, S., Mortensen, C., 1998. A new angle on earthworms. *Minnesota Conservation Volunteer* 61, 20–29.
- NatureServe. 2017. NatureServe Explorer: An online encyclopedia of life [web application]. Version 7.1. NatureServe, Arlington, Virginia. Available < <http://explorer.natureserve.org> > . (accessed: February 13, 2018).
- Parkinson, D., McLean, M.A., 1998. Impacts of earthworms on the community structure of other biota in forest soils. In: Edwards, C.A. (Ed.), *Earthworm Ecology*, first ed. St. Lucie Press, New York, pp. 213–226.
- Qiu, J., Turner, M.G., 2017. Effects of non-native Asian earthworm invasion on temperate forest and prairie soils in the Midwestern US. *Biol. Invasions* 19, 73–88. <https://doi.org/10.1007/s10530-016-1264-5>.
- Sather, N., Kjos, C., Mortensen, C., Gallagher, J., Mortensen, S., Leible, C., Wolff, B., Trull, S., Byers, O., 1998. Threats and risk working group report. In: Berlin, N., Miller, P., Borovansky, J., Seal, U.S., Byers, O. (Eds.), *Population and habitat viability assessment (PHVA) for the goblin fern (Botrychium mormo)*, Final Report. Conservation Breeding Specialist Group, Apple Valley, Minnesota, USA.
- Semlitsch, R.D., Todd, B.D., Blomquist, S.M., Calhoun, A.J., Gibbons, J.W., Gibbs, J.P., Graeter, G.J., Harper, E.B., Hocking, D.J., Hunter Jr, M.L., Patrick, D.A., 2009. Effects of timber harvest on amphibian populations: understanding mechanisms from forest experiments. *Bioscience* 59, 853–862.
- Soil Survey Staff, Natural Resources Conservation Service, United States Department of Agriculture. Web Soil Survey. Available online at the following link: < <https://websoilsurvey.sc.egov.usda.gov/> > .
- USDA. Laws and regulations to protect endangered plants. [https://www.fs.fed.us/wildflowers/Rare\\_Plants/conservation/lawsandregulations.shtml](https://www.fs.fed.us/wildflowers/Rare_Plants/conservation/lawsandregulations.shtml). (accessed: February 21, 2018).
- USDA NRCS. 2018. The PLANTS Database (< <http://plants.usda.gov> > , 21 February 2018). National Plant Data Team, Greensboro, NC 27401-4901 USA.
- Wagner, W.H., 1998. A background for the study of moonworts. In: Berlin, N., Miller, P., Borovansky, J., Seal, U.S., Byers, O. (Eds.), *Population and habitat viability assessment (PHVA) for the goblin fern (Botrychium mormo)*, Final Report. Conservation Breeding Specialist Group, Apple Valley, Minnesota, USA, pp. 103–109.
- Wagner Jr., W.H., Wagner, F.S., 1993. Ophioglossaceae C. Agardh: Adder’s-tongue family. In: *Flora of North America Editorial Committee, eds. 1993+. Flora of North America North of Mexico. 20+ vols. New York and Oxford. vol. 2, pp. 98–99.*
- Wagner, W.H., Wagner, F.S., 1981. New species of moonworts, *Botrychium* subgenus *Botrychium* (Ophioglossaceae), from North America. *Am. Fern J.* 71, 20–30. <https://doi.org/10.2307/1546672>.
- Whittier, D.P., 1973. The effect of light and other factors on spore germination in *Botrychium dissectum*. *Can. J. Bot.* 51, 1791–1794.
- Whittier, P., 1984. The organic nutrition of *Botrychium* gametophytes. *Am. Fern J.* 74, 77–86. <https://doi.org/10.2307/1546541>.
- Winther, J.L., Friedman, W.E., 2007. Arbuscular mycorrhizal symbionts in *Botrychium* (Ophioglossaceae). *Am. J. Bot.* 94, 1248–1255.
- WI DNR. 2017. Little goblin moonwort (*Botrychium mormo*). < <https://dnr.wi.gov/topic/EndangeredResources/Plants.asp?mode=detail&SpecCode=PPOPH10N> > . (accessed: February 13, 2018).
- Zuur, A.F., Ieno, E.N., Elphick, C.S., 2010. A protocol for data exploration to avoid common statistical problems. *Methods Ecol. Evol.* 1, 3–14.