Les Cheneaux Islands Eurasian Watermilfoil Control

Final Report

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1. Table of Contents

1. Table of Contents	1
2. Executive Summary	2
3. Project Overview/Background	5
4. Project Tasks	10
4.1 Task 1: Planning and Permitting	10
4.1.1 Permits for management activities	10
4.1.2 Quality Assurance Project Plan	12
4.2 Task 2: Treatment	13
4.3 Task 3: Monitoring	16
4.3.1 Task 3a. Remote sensing-based monitoring and mapping	16
4.3.1.1 Satellite mapping	17
4.3.1.2 Spectral profiles	19
4.3.1.3 UAS mapping	24
4.3.2 Task 3b. Field surveys, including collecting ecological and macrophyt data	e community 34
4.3.2.1 EnviroScience vegetation surveys	34
4.3.2.2 Michigan Tech vegetation and water sampling	37
4.4 Task 4: Development/improvement of Mt biocontrol methods	40
4.5 Task 5: Reporting and Communication of Results	45
5. Conclusions	52
6. References	53

2. Executive Summary

This 30-month project, a collaboration between Michigan Technological University and the Les Cheneaux Watershed Council, consisted of deploying a fungal plant pathogen indigenous to the Great Lakes, *Mycoleptodiscus terrestris* (Mt), that reduced the biomass and growth of invasive Eurasian watermilfoil (*Myriophyllum spicatum*, or EWM) in the Les Cheneaux Islands (LCI). Detailed field methods documented densities and biomass of EWM and other submerged aquatic vegetation at sampling sites. The use of multi-scale remote sensing methods, from satellite imagery to unmanned aerial systems (UAS), were demonstrated and documented to be useful to mapping EWM extent and monitoring multiple treatment methods, including the Mt application plus mechanical harvesting and diver-assisted suction harvesting (DASH). While permitting requirements meant that Mt fungus could only be applied on areas totalling one acre, UAS-enabled mapping of EWM was demonstrated for six areas, and satellite-based mapping was completed for an area of over 1730 acres in the LCI.

The project was organized into an approach comprised of five task areas, as described in the selected proposal, with the following major outcomes:

Approach Area 1: Planning and Permitting established that all biocontrol applications, mechanical harvesting and dredging aimed at EWM control was performed with appropriate permits, and that field and UAS surveys were planned and approved in advance to ensure that the project goals would be met. This included the creation and approval of a Quality Assurance Project Plan, approved on September 12, 2016, which was revised in December 2017 to reflect project changes. These changes included 1) the application of Mt under a USDA APHIS permit rather than an EPA Experimental Use Permit, 2) a change in the source of Mt from the USDA Agricultural Research Service to Wisconsin BioProducts, and 3) additional work to leverage a related project by using UAS imaging to evaluate the the effectiveness of Diver-Assisted Suction Harvesting (DASH) for controlling EWM in the Keweenaw Waterway. These changes were approved by EPA on December 21, 2017.

Approach Area 2: Treatment built on previous research and testing of Mt formulations for EWM control in the Les Cheneaux Islands and elsewhere to ferment a liquid culture of microsclerotia of the USDA TX-05 strain of Mt fungus. Whole-culture Mt was diluted with local lake water and applied via gravity feed from a mixing tank. The application protocol was based on the best information available from previous trials and lab work. The results of the 2017 pilot application of a liquid Mt culture will inform and improve future use of Mt for EWM biocontrol, which have been documented in an updated list of best management practices for use of Mt. Mechanical harvesting of EWM was also demonstrated to be complementary to biocontrol in areas where Mt application would be difficult or unwanted.

Approach Area 3: Monitoring, which was divided into two areas:

3a. Remote sensing-based monitoring and mapping

Multiscale remote sensing-based mapping included peak growth (2012) and recent (2016) maps of aquatic vegetation cover across a large swath of the Les Cheneaux waterways (1730 acres),

while finer-scale maps derived from UAS imagery allowed for more detailed monitoring of treatment response across multiple sites in the Les Cheneaux Islands in 2016, 2017 and 2018 (see Figures 11-19 below for examples from six locations in the LCI).

3b. Field surveys, including collecting ecological and macrophyte community data An integrated field assessment included surveys of aquatic macrophyte abundance, species composition and biomass, surveys for milfoil weevils, and water quality measurements (dissolved organic carbon (DOC) concentration and composition, water clarity, conductivity, pH, and water chemistry) to better understand treatment response and to serve as field truth for algorithm development for classifying the satellite and UAS imagery.

Approach Area 4: Development/improvement of Mt biocontrol methods

The GLRI "Arresting the spread of Eurasian watermilfoil in Lake Superior" grant started a centralized, web-based clearinghouse of reliable information on EWM control and management. This information is available at http://www.mtri.org/eurasian watermilfoil.html and includes information on biology, invasive properties and ecological impacts, development of mapping and modeling tools, spread, and further web resources. This leveraging of previous work and extending it through work took advantage of this project taking place in Les Cheneaux Islands, where an active community represented by the Les Cheneaux Watershed Council has been working to implement effective, safe, and economical biocontrol programs. LCWC has been posting information on its Mt biocontrol work to serve as information for updated best management practices for use of this treatment method. For examples, please see http://lescheneauxwatershed.org/projects/mycoleptodiscus-terrestris and especially their final report at http://www.lescheneauxwatershed.org/library/nuisance-species/eurasianwatermilfoil/lcwc-ewm-research/310-wc6-use-of-mycoleptodiscus-terrestris-as-a-mycoherbicidefor-myriophyllum-spicatum-eurasian-watermilfoil-management-in-the-open-water-system-of-theles-cheneaux-islands-michigan (Smith et al. 2018a). The LCWC final report serves as a detailed description of the Mt treatment methods and impacts, and are described in further detail below in Section 4.

Approach Area 5: Reporting and Communication of Results

The project has included an active outreach program focused on communicating results to both local stakeholders and the scientific community. This has included sharing results with the Les Cheneaux Watershed Council in person and through their newsletter, sharing information at the FrogFest annual community festival, giving presentations at the Les Cheneaux Community Library and to science students at Cedarville High School, and presenting at scientific meetings including International Association of Great Lakes Research (IAGLR) Annual Conferences, the Society for Freshwater Science Conference, and the Ecological Society of America (ESA) conference. In addition, the EWM resource page from the project team's previous "Arresting the spread..." GLRI project focused on EWM was updated, a dedicated project web page was created and maintained at http://www.mtri.org/ewmlci.html, educational signage was posted at treatment sites, and the Great Lakes Echo reported on the project ("Fighting invaders with drones and fungi" - http://greatlakesecho.org/2016/09/30/fighting-invaders-with-drones-and-fungi/).

Sharing invasive species biocontrol knowledge, experience and methods is critically important for others in the biological control community to understand what is working and what is not. Case studies of relatively new biocontrol vectors in field conditions can be especially useful. By applying Mt at a pilot scale in a Great Lake coastal zone, updating the available information on EWM biocontrol information, demonstrating a detailed EWM monitoring field protocol, and applying remote sensing tools, this project has contributed to the development of an integrated EWM management strategy that includes flexible options for sites where herbicide application is inadvisable or unwanted.

3. Project Overview/Background

Since the mid-2000s, several inland lakes and sheltered Great Lakes bays in and along Michigan's Upper Peninsula have developed populations of the aquatic invasive species Eurasian watermilfoil (*Myriophyllum spicatum*, or EWM). This invader has been especially prolific in the Les Cheneaux Islands (LCI, Figure 1), where as early as 2003, a Michigan Department of Environmental Quality (MDEQ) survey of Cedarville Bay found that EWM had colonized 225 of 289 surveyed acres. Point intercept surveys of Cedarville and Sheppard Bays found EWM present at 46% of survey points in 2014 (approx. 350 acres) and 18% in 2015 (approx. 135 acres). In the peak EWM growth years of 2006 and 2012, local fish catches declined notably, and boaters were unable to navigate nearshore waters without the weed fouling their propellers. EWM has continued to be a problem in the years since, with visible infestations during summer surveys in the docks and other high-traffic areas in the main local communities of Cedarville and Hessel.



Figure 1: Map of the Les Cheneaux Islands, with circles showing the main study locations of the towns of Hessel (left top) and Cedarville (center near top).

The 36-island LCI archipelago includes almost 200 miles of Great Lakes shoreline and shelters an intricate complex of shallow bays and channels that represents important aquatic habitat, particularly for fish species. The north shore of Lake Huron, including the LCI, was identified in *State of the Great Lakes 1999* as "a significant biodiversity investment area". The islands' recreational opportunities make the area popular for tourists, anglers, boaters, and homeowners, including over 4,000 seasonal and full-time residents in Clark Township, which includes Cedarville and Hessel. Small communities such as these along northern Great Lakes

shorelines rely on the nearshore waterways, ecosystems, and fish and bird habitat that enable the region's tourism industry. Left unmanaged, EWM can severely impact both the ecosystems and financial viability of local communities. Dense surface weed canopies can suppress desirable native plants, indirectly impacting fish and other aquatic organisms that are important for local tribal subsistence fishing and commerce as well as recreation and tourism. For these reasons, the 2012-2017 Michigan Tourism Strategic Plan identified invasive species as a primary threat to tourism in the State of Michigan (Nicholls 2012). EWM growth also directly impacts property values; in the LCI, 34% of waterfront properties have been adversely affected by dense EWM growth, and the estimated taxable value of twenty percent of township properties has been reduced due to degraded aesthetic values (total taxable value of EWM-impacted properties; estimates from Clark Township Supervisor in 2015).

Responding to the local community's strong preference for avoiding commercial herbicide use at the scale that would be required to control the area's nuisance EWM (and the approx. 120 potable water intakes currently used by area residents, which present an obstacle to chemical herbicide application), and building on previous work testing and developing EWM biocontrol methods at this site, a collaborative team from Michigan Technological University ("Michigan Tech") and the Les Cheneaux Watershed Council (LCWC) aimed to continue implementing and evaluating EWM biocontrol techniques. We proposed an adaptive management approach that evaluated the performance of the treatment technique established for control of EWM using the fungal pathogen *Mycoleptodiscus terrestris* (Gerd.), or "Mt", an indigenous organism not altered by genetic engineering. The use of adaptive management builds from the GLRI project "Arresting the spread of Eurasian watermilfoil in Lake Superior" (GL-00E01291-0), led by Dr. Casey Huckins and completed in December 2016 (Huckins et al. 2018); Dr. Huckins was a Co-Investigator for this project.

EWM management in the LCI began in 2007, with support also provided by a 2011 GLRI grant to the Les Cheneaux Watershed Council (GL-00E00809, "Eurasian Watermilfoil Strategic Biological Control Program"). Initial management techniques consisted of using a mechanical harvester to cut EWM beds and artificially augmenting the population of a milfoil weevil native to North America (*Euhrychiopsis lecontei*). Weevil stocking was very effective at decreasing the relative density of EWM in quiet and shallow bays but was less successful where boat traffic was heavy, which corresponds to a large proportion of the LCI nearshore zone as well as priority areas for EWM control. In the summer of 2012, EWM growth in the LCI became so prolific that mechanical harvesting and weevil stocking became virtually ineffective. The community responded by forming a task force chaired by the LCWC, which met with Michigan DEQ aquatic invasive species experts and representatives from the Michigan DNR Fisheries Division, the U.S.Army Corps of Engineers (USACE), and two lake management companies in November 2012 to gather input on the development of a milfoil management plan (now part of LCWC's Dynamic Aquatic Adaptive Management Plan,

http://lescheneauxwatershed.org/library/lcwc-management/119-lcwc-dynamic-aquatic-adaptivemanagement-plan/file). The management plan was developed based on those discussions and after reviewing the EWM control methods used by dozens of lake management associations across the US.

Also in 2012, LCWC began to evaluate Mycoleptodiscus terrestris (Mt), a native fungal pathogen that has been under study as a potential biocontrol agent for EWM since the 1980s (Shearer and Jackson 2006). Mt is considered to be indigenous to the LCI and was recovered from watermilfoil growing in the archipelago in 2012, as confirmed by a Research Plant Pathologist at the USACE). Mt is not considered to pose a human health risk, as it does not produce toxins or grow at mammalian body temperatures (Briggs 1991). An early formulation of the fungus was determined to be ineffective at inoculating plants in field trials (Shearer 1994), but Agricultural Research Service (ARS) microbiologist Mark Jackson and USACE plant pathologist Judy Shearer collaborated to develop an improved formulation for commercial bioherbicide applications. Their work has led to the development of a technique for cultivating Mt microsclerotia—small, filamentous clumps that tolerate drying and storage and perform better at adhering to plant surfaces (Shearer and Jackson 2006). In 2013 and 2014, LCWC collaborated with the USDA ARS to conduct test applications of the new formulation to small (0.5 to 1 acre) plots of EWM. The 2013 trials established the appropriate dosage rate and provided data on the effect of pump shear forces on the microsclerotia. Following these trials, a low-shear pump system was developed that can be mounted on a boat and applies an optimal dose of Mt at an even dilution rate, which was available and used for this project. Further smallscale treatment in 2014 resulted in a 77% reduction in EWM biomass compared with untreated control areas 35 days after treatment. At the same time, native vegetation cover in the treated areas has increased significantly since 2012. Table 1 summarizes the history of EWM invasion and management in the LCI by the beginning of this project.

2003	MDEQ vegetation survey in Cedarville Bay identified EWM in 78% of surveyed area, representing 16% of overall bay
2006	EWM has expanded throughout Cedarville Bay
2007	LCWC plants over 15,000 milfoil weevils (Euhrychiopsis lecontei) at two sites in Cedarville Bay. LCWC acquires a mechanical harvester to cut EWM.
2008	EWM density is dramatically lower within both stocking sites with concomitant increases in bare substrate and native species
2011	LCWC is awarded a 3-year GLRI grant (GL-00E00809) allowing them to plant 100,000 additional weevils

Table 1: Timeline of EWM invasion and management in LCI

2012	Rampant EWM growth throughout LCI, EWM control by weevils is limited to the plots where they were stocked, an intensive and expensive process Task force meets with government scientists and lake management experts to draft a management plan for EWM in the LCI
2013	Working with Mark Jackson at the USDA ARS, LCWC treats two one-acre test plots of EWM in Cedarville Bay with the fungal pathogen <i>Mycleptodiscus terrestris</i> (Mt).
2014	Two new half-acre EWM test plots in Sheppard Bay and John Smith Bay treated with Mt

Through this project, we implemented the Mt biopesticide treatment, used multi-scale remote sensing to help document EWM presence and treatment responses, deployed detailed field sampling to characterize EWM locations and growth conditions, presented on the project at Great Lakes meetings, and worked closely with the Les Cheneaux Watershed Council for community outreach. A detailed Quality Assurance Project Plan (QAPP) was developed by the project team and approved in September 2016 by the EPA project officer (then Rajen Patel) and Kevin O'Donnell, delegate for the Quality Assurance Manager at the US EPA Great Lakes National Program Office (GLNPO). It was revised by the project team and approved by the EPA in December of 2017 to reflect project changes. An APHIS permit (P526P-16-01796) was obtained by the LCWC in 2016 to apply Mt, with updated directions provided by the EPA in 2017 on where it could be applied. The terms of the permit restricted application to harbor sites with a total application area of less than one acre. This change was reflected in the approved QAPP revision.

While the application permit that was approved reduced the original scope of Mt fungus treatment from up to 10 acres per year to one acre total, it did enable application during the project period, and the application sites were well documented through fieldwork and remote sensing. Comparing one treatment site in the Hessel Marina harbor to two untreated control sites, there was a 60% reduction in biomass 50 days after treatment, and greater than 70% decrease in biomass 70 days after treatment. This compared favorably to the previous Mt treatment testing by the LCWC in 2014, which saw an 85% decrease in biomass. In addition, the 2017 treatment results showed the Mt shifting from healthy growth pre-treatment to darkened stems with discolored, brittle, and missing leaflets and shafts after 70 days, whereas untreated areas continued to show healthy EWM growth 70 days later.

To help address the reduced treatment area, and to take advantage of other projects and local treatment efforts, the QAPP revision documented how remote sensing could be used to not only track the Mt treatment area, but also document the impacts of mechanical harvesting and diver assisted suction harvesting (DASH). The harbor managers in Cedarville performed mechanical harvesting treatments in the summer of 2017, and those areas could be seen and quantified in UAS-collected imagery. For a Michigan Department of Natural Resources (MDNR) funded project (# IS14-2005, "Innovative and Multifaceted Control of Invasive Eurasian and Hybrid

Watermilfoil using Integrative Pest Management Principles"), DASH treatment areas in the Portage Waterway that bisects the Keweenaw Peninsula (see Figure 2) were documented with UAS imagery, and the change in biomass could clearly be identified. When combined with UAS data and satellite imagery documentation of EWM extent for the LCI, these helped demonstrate that high-resolution remote sensing can be valuable tool for mapping and monitoring of Eurasian watermilfoil. These remote sensing methods would likely be applicable and useful for monitoring and tracking other invasive aquatic plants as well throughout Great Lakes nearshore areas.



Figure 2: Primary project location in the Les Cheneaux Islands, with the extended treatment documentation area in the Keweenaw Peninsula.

The five tasks that correspond to the project's approach areas, summarized in the Executive Summary (planning/permitting, treatment, monitoring, development of application method recommendations, and reporting/outreach), were the main focus of the project work. These are presented in greater detail in Section 4, Project Tasks. Section 5 presents overall conclusions and recommendations stemming from the project, and Section 6 lists the references cited throughout this report.

4. Project Tasks

4.1 Task 1: Planning and Permitting

4.1.1 Permits for management activities

As noted, permitting was focused on obtaining an appropriate permit for application of a liquid culture of Mt as a biocontrol agent. Based on their previous experience with applying the Mt fungus, the project collaborators at the Les Cheneaux Watershed Council travelled to EPA headquarters in July 2017 to get more information on what was permissible for Mt application, building from an APHIS permit obtained in 2016 (APHIS permit P526P-16-01796). The LCWC partners presented on the need for Mt application during the critical summer time period when EWM would be most susceptible to Mt, how it was target specific, highly effective against EWM (and Hydrilla), the planned application areas in Hessel Harbor and Cedarville Bay, details of the field tests, and the application method. Specifically requested was permission to apply Mt in four plats of up to ¼ acre each. Figure 3 shows the areas of Mt application proposed for the permit (in red polygons). Based on this information, permission was granted to apply the native fungus in the restricted areas of harbors in Hessel and/or Cedarville, MI, with the total treatment area not to exceed the one acre approved under the APHIS permit. The permit noted that repeat applications to the same areas within the same season would not add to the total acreage.

6

HESSEL HARBOR APPLICATION AREA 7040 SQ FT



Three Cedarville Bay application sites:





-Breezeswept Basin at 9590 sq ft, lower left - green lines indicate emergent bulrush beds

- Clark Twp Dock at 10,500 sq ft, upper left

- Cedarville Marine at 9500 sq ft, upper right

Figure 3: Planned application areas of Mt fungus treatment at the Hessel Marina and Cedarville Bay areas in red, as presented to the EPA for approval.

The materials provided to support the application noted that Mt has been shown to be an effective control organism for EWM since the 1980s, that Mt is native to local waters, and has been demonstrated to be safe for other vegetation, animals, and humans. The application method was described as having the following key attributes:

- 45.8 gal/surface acre (Active ingredient rate of 11.4 lbs)
- 11.5 gal per ¼ acre application area (Active ingredient rate of 2.9 lbs)
- Whole culture Mt would be diluted 20:1 using local lake water
- It would be applied via gravity feed from mix tank through a PVC manifold, with the manifold position one foot below the surface, perpendicular to the bow of the pontoon boat.

The EPA recommended that following this approved field trial, an EUP could be obtained in the future that would require including costs of skin irritation studies, obtaining a tolerance exemption for open water studies, and possibly additional data. The LCWC is planning on obtaining an EUP once funding is available to deploy the Mt fungus over a larger area. The reduction in planned treatment area was documented in quarterly reporting and formalized in the QAPP revision. Finally, Clark Township continued to utilize mechanical harvesting and dredging to control aquatic plant growth around public docks and marinas, under permits from the Michigan DEQ Water Resources Division (#13-49-0077-P) and USACE (#LRE-2013-00695-16-S13, exp. 2023).

4.1.2 Quality Assurance Project Plan

At the beginning of the project, a Quality Assurance Project Plan (QAPP) was developed based on the approved statement of work from the project proposal, using the team's best scientific knowledge and experience from previous GLRI and related projects. The approved QAPP development was led by PI Colin Brooks with input from Co-Investigators Dr. Amy Marcarelli, Dr. Casey Huckins, and Amanda Grimm. The QAPP, approved in 2016, served as the overall guide to the project to ensure collection of quality data that would meet the project's needs. Its five tasks matched those in the project proposal and helped ensure that the quality of collected information met the needs of the project. Copies of the QAPP are available by contacting PI Brooks, and is on record with the US EPA GLNPO and the Michigan Tech Project Quality Assurance Manager, Joanne Polzien.

The 115 pages of the QAPP covered task descriptions (Element A) and data generation/acquisition (Element B). Among the highlights of Element A were descriptions of key project personnel, additional background on the EWM problem in the area, a listing of potential test locations for Mt treatment, reviews of previous benthic response monitoring, a review of remote sensing-based methods for monitoring, vegetation survey methods, water chemistry data collection methods, project documentation plans, and instrument calibration methods. Additional attachments covered the original planned participation of the USDA Agricultural Research Service (before they unexpectedly announced they would no longer be growing Mt), previous survey results show methods from 2014, the planned data sheet covering water and vegetation surveys, YSI sonde calibration specifics, and standard operating procedures for Enviroscience milfoil/vegetation survey transects.

Most of this QAPP served through the duration of the project. A six page revision was drafted in 2017 and approved by GLNPO to reflect needed changes. Two revisions to the original QAPP were included. One reflected a scaled-down plan to deploy the Mt fungus that meet the requirements of the APHIS permit that was approved after the project was proposed, where Mt fungus deployment would be limited to harbor sites (not open water areas that would include swimming areas or drinking water intakes) and would need to be less than once acre total. The other revision reflected that an alternative source for Mt fungus being grown had to be obtained from Wisconsin Bioproducts (<u>http://wisbio.com/</u>) after the USDA ARS was no longer able to provide it, and that the <one acre total area meant that small plots in the Hessel and Cedarville harbor areas were implemented instead of the larger area of up to 10 acres per year for two

years that had originally been planned. Two new sections in the QAPP revision captured the addition of using remote sensing to document the impacts of two additional treatment methods, the previously mentioned mechanical harvesting being done under the auspices of the local township, and the diver assisted suction harvesting (DASH) that was done under the MDNR invasive species grant. While these were not tasks completed for this project, the project team was able to use the fact that they were happening to demonstrate additional value in using high-resolution UAS-enabled remote sensing to monitor treatment types.

4.2 Task 2: Treatment

The most common treatment method for invasive watermilfoils is chemical herbicide. However, in some invaded areas, herbicide application is infeasible, prohibited or just undesirable. The Les Cheneaux communities are not open to aquatic herbicide application in their waterways due to the perceived risk of drinking water contamination (many lakefront properties have private water intakes for household water supplies). Thus, EWM management in Les Cheneaux waterways has been a combination of biocontrol and manual removal activities. The treatment component of this project focused on biocontrol with Mt fungus as previous efforts in the area had led to it being close to practical deployment.

In the proposal for this project, the U.S. Department of Agriculture Agricultural Research Service provided a letter of support from Research Biologist Dr. Mark Jackson, who was going to ferment and provide the needed amounts of Mt and transport it under refrigeration from the fermenter at a USDA facility in Peoria, Illinois. This would have provided the ability to apply Mt fungus in both 2016 and 2017, as originally planned. However, the project team learned in the summer of 2016 that Dr. Jackson's office would no longer be providing Mt growth capabilities to anyone, so a new source of Mt fungus had to be found, as documented in the project semi-annual report that covered April - September 2016. Over the winter of 2016-2017, a new source was found in Wisconsin BioProducts (<u>http://www.wisbio.com/</u>); LCWC staff negotiated with them to ferment Mt for the 2017 treatment season. The USDA lab did provide WisBio with the initial culture, so the Mt strain intended for treatment (USDA TX-05) remained the same.

In October of 2018, Bob Smith and Mark Clymer of the LCWC submitted a final report to PI Brooks that detailed the Mt fungus application, entitled "Use of Use of *Mycoleptodiscus terrestris* as a mycoherbicide for *Myriophyllum spicatum* (Eurasian watermilfoil) management in the openwater system of the Les Cheneaux Islands, Michigan". The full report is available from PI Brooks and is being posted to the LCWC webpage at <u>http://lescheneauxwatershed.org/</u>.

For the treatment effort, Mt culturing was complete with a whole culture harvest at Wisconsin Bioproducts on July 26, 2017. The liquid culture was chilled to $4^{\circ}F$, transferred into five gallon (20L) carboys, and stored in Styrofoam containers placed inside 16 protective cardboard shipping boxes. These boxes were loaded into a pickup truck, layered with dry ice, covered by plastic tarp, and driven from Milwaukee to the Les Cheneaux Islands. The boxes were stored on the truck until the application day of July 28th, 2017. The dry ice was still present between the shipping boxes, and Mt culture temperatures ranged from 39 to $44^{\circ}F$ (3.9 to $6.7^{\circ}C$) at application time.

Figure 4 shows the Mt being applied using gravity feed from a mix tank on board an available LCWC vessel, as planned. Also as planned, it was applied at a rate of 45.6 gal/acre with dilution with local lake water at a 20:1 ratio per volume of Mt, which improved Mt distribution through the plot. Mt was applied at the Cedarville launch ramp (CRAMP) area totalling 37,865 ft² (0.8692 acres) and at Hessel Marina area totalling 7,040 ft² (0.1616 acres). Additional areas that received Mt were at the Cedarville Marine marina (corresponding to the FDS sampling site) at 0.2181 acres, Breezeswept at 0.2202 acres), and Bumpa's waterfront (corresponding to our Court East sampling site) at 0.2410 acres (Figure 5). Only the Hessel Marina site was monitored for quantitative analysis due all other areas being comprised for monitoring by mechanical harvesting by the local business responsible for them. The CRAMP site was monitored on a qualitative basis because the EWM growth was so dense at the time of Mt application that quantitative monitoring was not possible.



Figure 4: Example photos of the Mt fungus being applied on July 28, 2017 from the LCWC vessel with the customized boom.



Figure 5: Locations of the project data collection sites: Les Cheneaux Islands (Hessel and Cedarville) and the Keweenaw Waterway.

At the Hessel Marina site, plant biomass surveys were performed on the day of treatment, 25 days after treatment (DAT), 35 DAT, 47 DAT, and 70 DAT by the LCWC. Also, the Michigan Tech team performed monitoring of the area June 19-23, 2017; July 13-15, 2017; and August 21-25, 2017 to help with pre- and post-treatment assessment during the treatment year. Follow up surveys by the combined team also were completed in 2018, and initial pre-treatment surveys were completed in 2016 (see Task 3).

The application of Mt fungus to the Hessel Harbor marina location resulted in over 70% biomass loss at 70 days after treatment as compared to two different untreated control sites. To achieve efficacy this similar for field trials conducted three years apart was a very positive result. Having the Mt culture produced by two different laboratories and achieving the observed degree of EWM control was also encouraging. One year after treatment, the LCWC found that the EWM biomass in the previously treated area was one-half the biomass recorded for an untreated area. This limited evaluation suggests that EWM vigor might be reduced during the season following Mt treatment, based upon EWM biomass in previously treated and untreated areas. If annual reduction in EWM vigor were to occur during successive annual Mt treatments, it is possible that multiple Mt applications could reduce EWM growth to a "minimum nuisance macrophyte" relative to aquatic ecology and recreational activities. Moreover, there continues to be appear to be no obvious impact on non-target macrophytes.

4.3 Task 3: Monitoring

4.3.1 Task 3a. Remote sensing-based monitoring and mapping

Remote sensing was used as a tool to help map the extent of EWM, and to show how highresolution imagery can be used to track the effects of management efforts. The remote sensing work used several components to meet these needs, including:

- Using commercially-available high resolution satellite imagery to map the extent of EWM and other other SAV.
- Developing spectral profiles to document how EWM can look different than other vegetation and bottom types, to help with the mapping process.
- Using unmanned aerial systems (UAS, also unmanned aerial vehicles / UAVs, or "drones") to show how very high-resolution, rapidly deployable imaging systems can help with mapping mapping and monitoring.

Both the satellite- and UAS-enabled remote sensing activities built on previous work supported by GLRI and NASA, such as the GL-00E01291-0 project (see the Huckins et al. 2018) and methods documented in Brooks et al. 2015 and Shuchman et al. 2013 developed under GLRI funding, among other sources (<u>http://www.mtri.org/cladophora.html</u>). Current UAS technology provides up to approximately 20 minutes of flight time in systems costing less than \$10,000. Also, standard rules from the Federal Aviation Administration (FAA) that went into effect in 2016 (called "Part 107") provide clear direction that UAS flights must be limited to within line-of-sight and no higher than 400 feet (122 m). This limits the area that can be covered by a low-cost UAS operating under Part 107. To map larger areas, satellite imagery is more practical, although at

lower resolution. The project proposal had the goal of using satellite imagery to map at least an 800-acre area for SAV including EWM, which was completed. While species-level identification is more difficult with commercial multispectral satellite imagery, it can provide at least a screening tool to identify areas of higher- or lower-density SAV that can then be surveyed with higher-solution UAS imagery for more detailed mapping.

4.3.1.1 Satellite mapping

Given that 2012 was the year of peak EWM growth and therefore should be the year for which EWM is easiest to map, a spectral-based classification method was developed using a summer 2012 Quickbird satellite image (resolution 2 m / 6.6 ft). However, as the more intensive annual EnviroScience vegetation surveys were initiated in 2013, no field data were available, so an unsupervised method was utilized. The final classification (Figure 6) covers 1730 acres and includes four spectrally separable submerged aquatic vegetation (SAV) classes as well as a deep-water/dark SAV class, a sparse SAV class, and a floating aquatic vegetation class. Based on field and aerial photos and qualitative information on vegetation growth and distribution in 2012 provided by the LCWC, it is likely that the class 'SAV 2' represents a dense monoculture of EWM, 'SAV 1' represents a mixture of lower-density EWM and other SAV species, 'SAV 4' represents mixed SAV and floating-leaved vegetation with an EWM component, and 'SAV 3' primarily represents benthic algae. This map demonstrates that satellite imagery can be used for initial mapping of surface aquatic vegetation vs. submerged aquatic vegetation even in the absence of field data.



Figure 6: Classified SAV map of Cedarville and Sheppard's Bays in Les Cheneaux, summer 2012.

More recently, a second satellite-based pre-treatment map of Cedarville Bay and Sheppard's Bay was classified for a 750-acre area from a cloud-free WorldView-3 image (resolution 2 m) collected June 30, 2016. This second map reflects the lower-density EWM conditions present in the Les Cheneaux waterways just before Mt treatment, and utilizes the point-intercept data

collected by EnviroScience in summer 2016 to inform the class names. This map was created using the Maximum Likelihood Classification tool in ESRI ArcGIS, using the point intercept data to create the input class signatures. An accuracy assessment of this map (Figure 7), performed using a subsample of this field data, indicated an overall map accuracy of 87.4% (Table 2). The performance of this approach demonstrates that spectral-based unsupervised classification tuned with field data can be an effective technique for mapping EWM. The map results indicate dense EWM growth in the northwest corner of Cedarville Bay and inner Sheppard's Bay, which agrees with the field data, as well as selected areas along La Salle Island that were not specifically sampled during the fieldwork.





Table 2. Error matrix for the classified map shown in Figure 7, based on coincident field truth data.



4.3.1.2 Spectral profiles

To help understand how EWM can be identified in aerial and satellite imagery, spectral profiles showing the remote sensing reflectance (Rrs) of different submerged aquatic vegetation (SAV) types were collected by the Michigan Tech team using two types of spectroradiometers. These spectroradiometers were an ASD FieldSpec 3 and a MTRI-built Lightweight Portable Radiometer (LPR) which recorded the amount of reflected light in wavelengths from 350 nm to 1000 nm (ultraviolet to near-infrared, including visible light). The spectral data collections were completed at three different scales of spectral profiles: SAV species removed from the water to obtain a direct vegetation profile without water column influences (out-of-water or "OOW data"), profiles of submerged vegetation collected from the side of the boat with the radiometer held approximately three feet above the water ("boatside" data), and spectral profiles collected from the LPR radiometer flown onboard a UAS at approx. 10–15 m above the water ("LPR UAS" data). Figure 8, from Brooks et al. (under review) shows the methods used to collect the spectral data, including the LPR system.



Heads-up display camera

Spectrometer optics -



Camera for simultaneous field-of-view reference photos

Figure 8. Images demonstrating spectral data collection methods. (a) Single-species plants on a black tarp about to have their spectral profile recorded for out-of-water (OOW) scale data. From left to right: Chara sp. (stonewort), Potamogeton richardsonii (clasping-leaf pondweed), and EWM. (b) Collection of OOW scale data using the LPR spectroradiometer during an August, 2017 data collection (c) Collection of spectral profile data at the boatside scale using the LPR spectroradiometer over an area of predominantly EWM. (d) Initial aerial photo test from 2015 at a site in Keweenaw Waterway showing visible submerged aquatic vegetation, emergent vegetation, shoreline vegetation along with the Michigan Tech research vessel used for launch and recovery of a DJI Phantom UAS. (e) Aerial photo taken from the Bergen hexacopter UAS with the LPR's five mp camera, with EWM visible near the water's surface at a boat slip in the

Hessel Marina site in the Les Cheneaux Islands study area. (f) The LPR mounted underneath the Bergen hexacopter UAS, about to collect spectral data over an area of EWM (including hardware notations).

Table 3 lists all the places that spectral data were collected, helping to identify the spectral signatures of EWM and other macrophytes (See Figure 5 for their locations).

Table 3. Collection sites listing types of spectral data collected by time period and data
collection scale. Scale and method of spectral data collection: OOW = out-of-water; Boatside = from
side of boat; LPR UAS = Light Weight Portable Radiometer from an unmanned aerial system

Collection	Collection		July	August	June	July	August
Sites	Region	2015	2016	2016	2017	2017	2017
Pike	Portage						
Bay	Waterway	OOW		LPR UAS			
Torch	Portage						
Bay	Waterway	OOW					
Breeze-	Les Cheneaux					Boatside,	Boatside,
swept	Islands					LPR UAS	OOW
	Les Cheneaux						
Cedar	Islands						OOW
Court	Les Cheneaux						
Dock	Islands		LPR UAS				
Court	Les Cheneaux		Boatside,		Boatside, LPR		Boatside,
East	Islands		LPR UAS	Boatside	UAS	Boatside	OOW
Court	Les Cheneaux						
West	Islands					Boatside	OOW
	Les Cheneaux						Boatside,
Chappell	Islands				Boatside		OOW
	Les Cheneaux		Boatside,	Boatside,	Boatside, LPR	Boatside,	
FDS	Islands		LPR UAS	LPR UAS	UAS	LPR UAS	OOW
Hessel	Les Cheneaux					Boatside,	Boatside,
Marina	Islands					LPR UAS	OOW
Howells	Les Cheneaux			Boatside,	OOW, Boatside,		Boatside,
Dock	Islands			LPR UAS	LPR UAS		OOW
	Les Cheneaux				Boatside, LPR	Boatside,	Boatside,
Neil	Islands				UAS	LPR UAS	OOW

Under the right water and light conditions, certain spectral bands, and depending on the growth patterns, the spectral profiles of EWM did appear to be distinct from those of other aquatic vegetation species and bottom types. This was particularly true when the Normalized Difference Vegetation Index (NDVI) was included as a predictor. NDVI is a reflectance ratio of near-infrared to red light that is able to indicate different amounts of vegetative biomass. These methods have been documented in Brooks et al. (under review), the first journal article in PI Brooks' PhD dissertation that focuses on SAV mapping methods and applications.

Figure 9 shows an example of spectral profiles of EWM and other vegetation types when the plants were taken out of the water to get the strongest spectral signature possible. These use all 651 one-nanometer(nm)-wide spectral bands from 350 to 1000 nm. The two-sample

Kolmogorov-Smirnov (K-S) test (Sokal and Rolf 1995) used in R (versions 3.4.0 to 3.4.3) reveals that when using all available spectral bands, EWM does look different than other vegetation species. However, this does not provide a practical imaging system for EWM mapping, as available multispectral and hyperspectral systems more typically from four to 80 bands.



Figure 9. Spectral profiles for out of water vegetation. (a) Spectral profiles of eight OOW aquatic plant species from June 2015, showing ultraviolet to near-infrared (350–1000 nm) wavelengths for all 651 bands. (b) Spectral profiles for nine aquatic plant species, plus a reference tarp, from June 2017, showing all 651 one nm wide bands.

Figure 10 shows spectral data resampled to two levels: one representing six spectral bands that correspond to a Tetracam MCA-6 imaging camera that was available for summer field work in

(12a & 12c), and other for eight bands that correspond to those found useful for wetlands mapping by Becker et al. (12 b & 12 d) (Becker et al. 2005, 2007). 12a and 12b show 2015 OOW data, while 12c & 12d show 2017 data.



Figure 10. Resampled spectral reflectance values for Tetracam and Becker spectral bands: (a) Resampled to approximate the Tetracam bands for the eight OOW species samples collected in 2015. (b) Resampled to the Becker bands for the 2015 OOW data. (c) Spectral reflectance values for the Tetracam wavelengths for the nine out-of-water species collected in June, 2017. (d) Spectral reflectance values for the Becker wetland bands for the nine out-of-water species collected in June 2017.

Using just the spectral data equivalent to the Tetracam and Becker bands did not result in reliable differentiation of EWM spectra from other vegetation types when using the K-S test. However, average all 651 bands to just 65 10-nm wide bands did result in EWM being different from nearly all other vegetation types when using June of 2017 OOW data. This supports the concept of deploying a hyperspectral imaging system with a number of bands similar to the 65 averaged bands tested here as a reliable way of identifying EWM from other vegetation types.

Also analyzed were 62 spectral profiles representing all the boatside-scale data collected in 2016 and 2017. A two-way ANOVA mixed model was the analysis showed that NDVI values were significantly different among dominant vegetation groups. Two aquatic vegetation indices were investigated as well (the Normalized Difference Vegetation Index and the Water Adjusted Vegetation Index, see Villa et al. 2014) but did not help differentiate EWM from other SAV.

When comparing the LPR UAS, boatside, and OOW spectral data for EWM samples, the LPR UAS data only averaged 13.3% of the boatside remote sensing reflectance and 27.4% of the OOW values. The lower values for the LPR UAS data are most likely caused by the greater distance to the spectroradiometer sensor when it was been flown 30-45 feet in the air vs. the boatside and OOW spectral data collection. Maximizing the amount of information reaching a UAS-based camera by collecting on sunny days, near solar noon, and with relatively calm waters should help strengthen the signal of vegetation profiles when using this more distant method of vegetation profiling.

4.3.1.3 UAS mapping

UAS-collected imagery provided an important resource for documenting EWM extent. UAS imagery was used both for EWM mapping and more generally for documenting treatment locations. Figure 11 shows the treated Hessel marina area on August 24, 2017 (27 days after Mt treatment) in a natural color image, where EWM is clearly identifiable as the tall, feathery stems reaching near the water's surface. The MTRI team created several natural color orthomosaics using DJI Mavic Pro, Phantom 3 Advanced, Nikon D800 and Nikon D810 camera systems. After obtaining the imagery, the team used Agisoft Photoscan to create orthomosaic base maps of each site. To create these, Agisoft takes individual image frames and merges them together using their GPS locations and GPS ground control points. It creates a 3D point cloud of each site that is used to create the 2D orthomosaic.



Figure 11: Example UAS-collected image from 8/24/2017 of the primary Mt treatment location in the Hessel marina. The dense feathery vegetation near the water's surface is EWM, which can be clearly identified in the UAS imagery.

Once the orthomosaics have been created, the next step is to create supervised classifications using eCognition Developer software. Once the imagery has been loaded into the program, we execute a command called multiresolution segmentation on each image. This command groups similar pixels into polygons based on spectral similarities to neighboring pixels. Once this step has been completed, we next create all of the image classes and begin identifying regions where examples of each class exist in the imagery. After supplying sufficient training data to the program, we next execute the classify step. This command uses the supplied training data to assign each pixel a class based on pixel parameters such as color and brightness. Once complete, the generated classification is exported to create figures of the classification. Figure 12 below is a classification example at Howells Dock. This natural color orthomosaic (top) was taken in 2016 using a Nikon D800 digital camera with 36-mp resolution. One of the dominant vegetation types for this site was Eurasian watermilfoil (indicated in yellow in the classification, bottom).



Figure 12: Classified map (bottom) derived from an orthomosaic (top) of imagery collected in August 2016 using an UAS-flown Nikon D800 at the Howells Dock site (see Fig. 5).

In addition to natural color images (also known as red/green/blue or RGB images), multispectral images were also collected using two systems that could image in the both the visible and near-infrared ranges. These were a six-band Tetracam multispectral camera and a system that combined two Canon cameras. For the two Canon camera system, called the VISNIR system, one was a normal camera sensitive to visible RGB light, and the second camera was sensitive only to near-infrared (NIR) light. Collecting imagery in this way made it possible to generate a NDVI layer using both the NIR and RGB imagery. Once created, the RGB, NIR and NDVI images were layered together in order to create the classification in eCognition. The near-infrared light, along with the NDVI, were useful for identifying areas of heavy vegetative biomass near the water's surface, which was often EWM.

Figure 13 presents an example of this method for the Court East project site (see site map in Fig. 5). The left map panel shows the VISNIR data displayed as an RGB image, and the right panel shows the results of object-based classification performed on the VISNIR bands and derived NDVI. Instead of using Agisoft Photoscan to process this imagery, we used ESRI ArcMap to georeference individual image tiles to their respective locations. We used GPS points and the ESRI basemap imagery to align these images correctly.



Figure 13: Example classification based off VISNIR imagery and NDVI for the Court East site with June, 2017 UAS images.

The other multispectral system used was the six-channel multispectral camera manufactured by Tetracam (Chatsworth, California; <u>http://tetracam.com/</u>), the MCA-6 system, whose bands were investigated with spectral data. This was selected for its ability to cover the 400–1000 nm (visible to near-infrared) spectral range, the availability of different spectral filters within that range, company reputation, and low weight (<one kg / 2.2 lbs with battery). The system was available for three one-week data collection periods in 2016 and 2017 via rental. After demonstrating its value, it was purchased by Michigan Tech for use in 2018. The Tetracam Micro MCA-6 was configured with the following bands: 490 nm (blue), 530 nm (green 1), 550 nm (green 2), 600 nm (orange), 680 nm (red), and 720 nm (red edge). Preliminary data collected in 2015 in the Keweenaw Peninsula was used to help select these specific bands as potentially informative. An example of classified Tetracam imagery is shown below in Figure 14. Here, georeferenced Tetracam multispectral imagery has been laid over Nikon D800 and DJI Phantom imagery. Through the use of multispectral imagery, we were able to differentiate between Eurasian watermilfoil and Northern watermilfoil. Northern watermilfoil appears bright green, while Eurasian watermilfoil appears reddish-brown in the multispectral images.

Howell Dock Multispectral Classification



Figure 14: Classified Tetracam multispectral imagery comparing coverage of Northern to Eurasian watermilfoil at Howell's Dock.

UAS imaging was focused on monitoring the Hessel marina treatment site and nearby untreated control sites as a reference. During the project, the opportunity to document two other treatment types became available. Mechanical harvesting by local marina operators took place in Cedarville in early July 2017, and the effects of this mechanical removal of EWM could be seen in RGB UAS imagery collected on July 19, 2017 and in multispectral imagery collected on August 23, 2017 (Figure 15). Specific quantitative data could be calculated from the Tetracam image in Figure 15, where 83.3% of the mechanically harvested area (1211.3 ft²) was open water after treatment, but 16.7% (243.4 ft²) of the 1454.7 ft² (0.03 acres) imaged area was still in EWM. This shows how UAS-enabled multispectral sensing can help with monitoring the mechanical harvesting treatment type by documenting remaining vegetation after treatment.



Figure 15: Demonstration of using UAS multispectral Tetracam imagery to document effectiveness of mechanical harvest treatment method.

Additionally, the "Innovative and Multifaceted Control of Invasive Eurasian and Hybrid Watermilfoil using Integrative Pest Management Principles" project funded by Michigan DNR provided the opportunity to image areas of diver-assisted suction harvesting (DASH) treatment immediately before and after the treatment took place. There were five DASH plots, totalling 1141 ft² or 0.0262 acres. Figure 16 show how changes in EWM near-surface biomass due to DASH removal can clearly be identified in the near-infrared (NIR) imagery collected by UAS of

the DASH plots. Figure 17 shows classification analysis results using the post-DASH imagery, where the lack of EWM biomass after treatment can clearly be seen after treatment within the four DASH plots covered by the UAS imagery. Using pre- and post-DASH classifications, the amount of EWM within those four plots dropped from 25.1% of the plot areas to only 2.7% after DASH treatment (Table 4). These results also show how quantitative data on DASH treatment effectiveness can be obtained using UAS imagery.



Figure 16: Examples of NIR imagery collected via UAS showing changes in EWM extent before and after DASH treatment in July of 2017.



Figure 17: Mapping results for the four DASH treatment plots covered by VISNIR UAS imagery collected immediately before and after treatment in July, 2017.

Cover type	Pre-DASH	% of area	Post-DASH	% of area		
Water	640.59	73.0%	882.77	96.8%		
EWM	220.61	25.1%	24.52	2.7%		
Buoy	2.41	0.3%	2.65	0.3%		
Lillypads / Emergent Veg	14.10	1.6%	2.08	0.2%		
	877.70		912.02			
	877.70		912.02			

Table 4: Area in ft ² of EWM and other type	in pre-DASH and post-DASH treatment plo	ots.
--	---	------

Area units are in ft²

Figures 18 and 19 illustrate another use of UAS imagery to track EWM treatment, this time using July 2017 and August 2018 natural color (RGB) imagery collected with DJI Phantom 3 Advanced and Mavic Pro UAS. EWM appears dense before treatment in July 2017, but less dense one year later, within a matrix of other SAV species.

Hessel Marina July 2017 Classification



Figure 18: July 2017 images and classification results of Hessel Marina shortly before Mt fungus treatment. EWM is dense within the marina area.

Hessel Marina August 2018 Classification

Figure 19: August 2018 UAS images and classification results of Hessel Marina one year after Mt fungus treatment. EWM appears lense dense, and within a matrix of other SAV.

Table 5 shows that based on classification of UAS imagery for Hessel Marina, EWM appears to have become less prominent from 2017 to 2018 (one year after treatment), with other SAV making up the largest area a year later. In 2017, EWM was 59.6% of the approximate Mt fungus treatment area, but was only 16.6% of that same area in 2018. Other SAV appears to make up the difference, along with a reduction in open water. While there is the potential for error in such classifications, these figures illustrate that within the limits of image analysis, the UAS imagery can help monitor changing EWM extent for this biocontrol treatment method.

July, 2017	% of area	Aug., 2018	% of area
1918.39	30.3%	566.96	9.0%
3769.85	59.6%	1041.906	16.6%
109.10	1.7%	4175.084	66.4%
529.24	8.4%	501.89	8.0%
6326.59		6285.84	
	July, 2017 1918.39 3769.85 109.10 529.24 6326.59	July, 2017 % of area 1918.39 30.3% 3769.85 59.6% 109.10 1.7% 529.24 8.4% 6326.59	July, 2017% of areaAug., 20181918.3930.3%566.963769.8559.6%1041.906109.101.7%4175.084529.248.4%501.896326.596285.84

Table 5: Comparison of 2017 vs. 2018 EWM extent within treatment area based on UAS imagery analysis.

Area units are in ft²

4.3.2 Task 3b. Field surveys, including collecting ecological and macrophyte community data

This project included assessments of aquatic vegetation, environmental characteristics, and ecological processes (i.e., water chemistry, phytoplankton chlorophyll-a, water depth, light transmission, etc.) to gain data needed to understand EWM, the Mt fungus treatment, and provide information to inform the remote sensing-based monitoring. This included bay-scale aquatic vegetation surveying by EnviroScience, Inc., continuing a time series of monitoring data that they initiated in 2013 for LCI. Complementary sampling by Michigan Technological University was performed at a spatial scale that could be easily related to the spectral profile and UAS imagery data collected to identify plants using remote sensing tools and to obtain more detailed information about biomass, water quality, and treatment sites. These methods were described in the QAPP, with some changes informed by practical field experience to adjust the spatial distribution of our sampling from that described in the QAPP.

4.3.2.1 EnviroScience vegetation surveys

The LCWC has contracted the environmental consulting firm EnviroScience Inc. to monitor aquatic vegetation in inner waterways of the Les Cheneaux Islands on an annual basis since 2007. Continuation of this monitoring program, including point-intercept and aquatic vegetation assessment sites (AVAS) surveys, was funded in part by this project for 2016-2018. The methods and results for the EnviroScience annual survey work are summarized here; annual reports providing greater detail are available on the LCWC website (http://www.lescheneauxwatershed.org/library/nuisance-species/aquatic-vegetation-and-weevil-surveys).

Surveying was performed using the Michigan DEQ guidance contained in "Standard Procedures for Surveying Aquatic Plants" (available at https://www.michigan.gov/documents/deq/wrd-illm-surveyprocedure_445615_7.pdf). For consistency, the same survey areas have been monitored annually around the same time of the year since 2013. Plant community data were collected through visual and rake tow surveys along evenly-spaced transects of the littoral zone. In each of these transect zones, the presence and relative density of each aquatic plant species was determined and the information was recorded on the Standard Aquatic Vegetation Assessment Site Species Density Sheet developed by the State of Michigan. Visual and rake surveys were performed at each site until no new species were encountered and the biologists conducting the

survey were confident that adequate information had been obtained to estimate the density of each species encountered. Species of unknown identity were placed in a sample bag, appropriately labeled, and identified using taxonomic keys at the completion of the survey. The approximate percentage of cumulative cover (%CC) was reported as cover codes A, B, C, and D to describe the approximate coverage of each plant between each transect and within each AVAS.

Point-intercept (PI) surveys were conducted annually in the LCI areas of Cedarville and Sheppards Bays

following the methods outlined in "Point Intercept and Line Intercept Methods for Aquatic Plant Management" (Madsen, 1999). This survey method was chosen based on the relatively shallow depths and larger areas of both bays. A grid of evenly-spaced point intercepts was created using GPS technology, and the surveyors navigated to each point along the grid. At each PI, the presence and relative density of each aquatic plant species was determined based on a single rake tow. Once the rake was retrieved from a point, each species found on the rake was identified and assigned a density code for rake cover similar to the AVAS method. Species of questionable identity were identified at the completion of the survey.

The EnviroScience survey areas included the main project areas of Cedarville Bay and Hessel Harbor in addition to several additional sites. In Cedarville Bay, EWM density and cover as measured by the point-intercept surveys decreased fairly consistently from the alarming peak growth seen in 2012 through 2017, with EWM present at 51, 44, 28, 11, and 14 out of 146-148 survey points in 2013-2017 respectively (Figure 20). The dominant species observed in Cedarville Bay beginning in 2013 were consistently eelgrass and *Chara*. In contrast, the Cedarville Bay AVAS transect data, which covers only the westernmost portion of the point-intercept survey area, indicated a significant increase in EWM cover in the area from the public boat launch to the FDS marina, from 1% cumulative cover in 2014 to 40% CC in 2017.

The Hessel Harbor survey area was consistently dominated by *Chara*, with EWM increasing slightly from 2015 (1.0% of cumulative cover) to 2017 (5.5% CC) but remaining at a low density compared to 2013-14 (17-40% CC). A new invasive species, curlyleaf pondweed, was identified at Hessel for the first time in 2016 but not seen in 2017. Similar patterns of EWM cover declining steeply from 2012 to 2015 and then remaining relatively low were observed for all of the other EnviroScience-surveyed areas, though small increases in cover were observed from 2016 to 2017 in 16 of the 18 areas surveyed in both years.



Figure 20: Point intercept survey map comparing 2013, 2015 and 2017 in Cedarville and Sheppard's Bays.

New survey points were added within the experimental Mt application areas in 2017. The EnviroScience crew, surveying August 22-24, 2017, approximately 1 month after the Mt treatment, did not observe signs of damage to the stems or leaflets in the treated areas. The four Hessel Harbor points were scored a B (3%-20%) in EWM density, and the four Cedarville Bay points scored 'no EWM observed', A (1-2%), B, and C (21%-60%) at one point each. Of two points at Breezeswept, no EWM was observed at one point and EWM density at the other point was scored a B. The last two locations were within the boat slips of Cedarville Marina and both were scored an A.

The 2018 EnviroScience survey work indicated that EWM continued its slow increase in abundance along the shorelines of Islington Channel, Snows Channel and Sheppards Bay, while desirable, low-growing native species continued to dominate Cedarville Bay and central Sheppard's Bay. The second-year evaluation of the MT sites revealed mixed results. The milfoil appeared healthy and green during the August 2018 survey at all the locations, but density changes were noted. Two of the four treatment sites decreased in Hessel Harbor. A third site (HHMt4) may have also decreased but a yacht obstructed the survey. Five of the eight treatment sites in Cedarville Bay increased in density, while two decreased (Table 6).

Table 6. EWM density scores assigned by EnviroScience to Mt treatment locations shortly after treatment (August 2017) and 13 months after treatment (August 2018).

Scores are N (no EWM observed), A (1-2% density), B (3-20% density), C (21-60% density) or D (>60% density). HH = Hessel Harbor, CB = Cedarville Bay, BS = Breezeswept. See maps in Appendices B and C for exact sampling locations.

	HHMt1	HHMt2	HHMt3	HHMt4	CBMt5	CBMt6	CBMt7	CBMt8	BSMt9	BSMt 10	CBMt 11	CBMt 12
2017	В	В	В	В	N	С	В	А	N	В	А	А
2018	В	А	А	N?	А	N	В	С	В	В	В	В

4.3.2.2 Michigan Tech vegetation and water sampling

For our vegetation data collection, instead of using a line-transect method as originally planned, we modified our Michigan Tech sampling plan to characterize set points that could be directly matched in space with marked locations in aerial imagery, in the process developing a robust sampling protocol that can be applied to any aquatic monitoring program integrating point samples and remotely collected imagery in aquatic environments.

At each sampling site on each date, we used marker buoys for areas where vegetation, chemistry, and imagery data were collected. Between two and four marker buoys were deployed at points of interest in the area to be sampled using UAS-based imagery (Figure 21 shows these marker buoys as seen from the side of the boat, and from above in a UAS image). We collected a GPS location for each buoy using a Trimble GeoExplorer GPS unit running in "code phase", meaning it was collecting with accuracy in the typical range of 50 cm to 1 m (1.6 to 3.3 feet). As previously described we collected boatside spectral profiles of submerged aquatic vegetation using a spectroradiometer, either an ASD FieldSpec3 or the LPR that using Ocean Optics radiometer sensors. Aerial imagery was collected via UAS. At each site for one these marker buoy locations, we also sampled water chemistry and physical characteristics (light extinction, Secchi depth, water depth conductivity, temperature, pH, water samples for dissolved and total nutrients, underwater and surface photos) following methods detailed in the approved QAPP.



Figure 21: Marker buoys deployed for an example sampling site, Howells Dock. (a) Marker buoys visible from the sampling boat, (b) close-up to one of the marker buoys, (c) the same buoys visible in a UAS image of Howells Dock (yellow arrows point to the three buoys placed temporarily for this site).

Following collection of physical and chemical characteristics and all boatside and UAS-based spectral data, we characterized the macrophyte assemblage surrounding each marker buoy using three approaches: 1. Visual observations of percent cover of different macrophyte species in a 3-m (10 foot) radius and depth below water surface facing forward, port and starboard from the bow of the boat, which was tied to the marker buoy for sampling; 2. Relative abundance of macrophytes using three sampling rake tosses forward, port and starboard from the bow and classified using aquatic macrophyte assessment site (AMAS) procedures recommended by the Michigan Department of Environmental Quality (MDEQ 2005); 3. Twist samples for standing crop estimates collected forward, port and starboard from the bow by lowering a 16.5 cm (6.5") diameter double sided rake vertically to the lake bottom (Johnson and Newman 2011) and

spinning one revolution to collect a 0.214 m² (2.3 ft²) sample of macrophytes. Biomass samples were sorted, identified and analyzed following procedures detailed in the QAPP.

This sampling revealed that EWM comprised 15-80% of the macrophyte assemblage sampled using visual estimates on most sampling dates (Figure 22), which is unsurprising as these sampling sites were selected to focus on collection of UAS imagery to determine the feasibility of classifying and mapping EWM. The native macrophytes were diverse across all sites and dates. The greatest species richness was observed at the three marina sites (Breezeswept, Hessel Marina, FDS), which were the sites of greatest boat traffic and disturbance (boat props and mechanical harvesting) (Table 7). All macrophyte samples have been processed to date.



Figure 22: Visual estimates of cover at all sampling sites in Aug 2017 and 2018

			SITES										
Group	Common Name	Scientific Name	Breeze- swept	Chappel	Court	Court East	Court West	CRAMP	FDS	Hessel Marina	Howells Dock	Neil	Urie Point
Aquatic Mosses	Aquatic Moss	Several genra, especially Drepanocladus and Fontinalis							x				
Arrowheads	Arum-leaved arrowhead	Sagittaria cuneata										x	
Bladderworts	Common Bladderwort	Utricularia macrorhiza							x				
Bur-reeds	Floating Bur-reed	Sparganium fluctuans	x										
Chara	Small Nitella	Nitella tenuissima	х										
Coontail	Coontail	Ceratophyllum demersum	х		х	х	х	х	х	х	х		
Nalads	Slender Naiad	Najas flexilis		x	x	х			х	х	x	x	x
Pondweeds	Algal-leaved Pondweed	Potamogeton confervoides	x		x	x			х				
Pondweeds	Clasping-leaf Pondweed	Potamogeton richardsonii	х	x		х	x			х	х		
Pondweeds	Curly-leaf Pondweed	Potamogeton crispus	х		х	х	x		х	х			
Pondweeds	Fern Pondweed	Potamogeton robbinsii							x		x		
Pondweeds	Flatstem Pondweed	Potamogeton zosteriformis	х	x	×	x	x	x	x	x	x		
Pondweeds	Floating-leaf Pondweed	Potamogeton natans	х										
Pondweeds	Fries' pondweed	Potamogeton friesil						x		х		х	
Pondweeds	Large-leaf Pondweed	Potamogeton amplifolius	x										
Pondweeds	Leafy Pondweed	Potamogeton foliosus				х			х				
Pondweeds	Sago Pondweed	Stuckenia pectinata	х									х	
Pondweeds	Sheathed Pondweed	Stuckenia vaginata							х	х			
Pondweeds	Small Pondweed	Potamogeton pusillus spp.	x		x	x	x		х	х		×	
Pondweeds	Variable Pondweed	Potamogeton gramineus	х		х								
Pondweeds	Whitestem Pondweed	Potamogeton praelongus							х				
Quillworts	Lake Quillwort	Isoetes lacustris		х						х		x	
Stoneworts	Chara	Chara spp.	х	х			x		х	х	X	х	х
Water Celery	Eel Grass/Water Celery	Vallisneria americana	х			х			х	х	x	х	х
Water Crowfoot	White Water Crowfoot	Ranunculas aquatilis	x		x				х	x		×	
Water Marigold	Water Marigold	Bidens becki	х		x	х			х	x			
Water Stargrass	Water Stargrass	Heteranthera dubia	х			х	х		х	х	х	х	
Watermilfoils	Eurasian Watermilfoil	Myriophyllum spicatum	х	X	х	х	х	х	х	х	X	х	х
Watermilfoils	Northern Watermilfoil	Mylophyllum sibiricum	x	x	×				x	x	x	x	
Watermilfoils	Various-leaved Watermilfoil	Myriophyllum heterophyllum	x		х				x		x		
Waterweeds	Elodea	Elodea canadensis	х	х	х	х	x	X	x	X	х	х	

Table 7: Macrophyte species observed across all sites and dates using rake toss, twist and visual estimate methods.

4.4 Task 4: Development/improvement of Mt biocontrol methods

The GLRI "Arresting the spread of Eurasian watermilfoil in Lake Superior" grant started a centralized, web-based clearinghouse of reliable information on EWM control and management. This information is available at http://www.mtri.org/eurasian_watermilfoil.html and includes information on biology, invasive properties and ecological impacts, development of mapping and modeling tools, spread, and further web resources. Information from Les Cheneaux Islands Eurasian Watermilfoil Control grant was used to expand the previous grant's output to include vetted, up-to-date information on biocontrol options, necessary inputs, and limitations. This leveraging of previous work and extending it through work took advantage of this project taking place in Les Cheneaux Islands, where an active community represented by the Les Cheneaux Watershed Council has been working to implement effective, safe, and economical biocontrol programs.

LCWC has been posting information on its Mt biocontrol work to serve as information for updated best management practices for use of this treatment method. For examples, please see http://lescheneauxwatershed.org/projects/mycoleptodiscus-terrestris and especially their final report at http://www.lescheneauxwatershed.org/library/nuisance-species/eurasian-watermilfoil/lcwc-ewm-research/310-wc6-use-of-mycoleptodiscus-terrestris-as-a-mycoherbicide-for-myriophyllum-spicatum-eurasian-watermilfoil-management-in-the-open-water-system-of-the-les-cheneaux-islands-michigan">http://www.lescheneauxwatershed.org/library/nuisance-species/eurasian-watermilfoil/lcwc-ewm-research/310-wc6-use-of-mycoleptodiscus-terrestris-as-a-mycoherbicide-for-myriophyllum-spicatum-eurasian-watermilfoil-management-in-the-open-water-system-of-the-les-cheneaux-islands-michigan">http://www.lescheneauxwatershed.org/library/nuisance-species/eurasian-watermilfoil/lcwc-ewm-research/310-wc6-use-of-mycoleptodiscus-terrestris-as-a-mycoherbicide-for-myriophyllum-spicatum-eurasian-watermilfoil-management-in-the-open-water-system-of-the-les-cheneaux-islands-michigan (Smith et al. 2018a). The LCWC final report serves as the main summary of the Mt treatment methods and impacts, and are described in further detail below.

The Smith et al. 2018a LCWC report documents the results of the Mt treatment at 25, 30, 45, and 70 days after treatment (DAT) with quantitative results for the Hessel Marina site, as compared to untreated control sites at Howells Dock and Point Urie (see Figure 5 for locations). Results have been posted to the LCWC site and linked to from the project information web page at http://www.mtri.org/ewmlci.html. The degree of Mt infectivity in EWM was quantified using the change in EWM biomass over time. Biomass change was calculated as mg of EWM wet weight per cm of stem length. A grapnel hook was used for collecting plant samples. Wet weights were recorded within 24 hours, and dry weights were recorded after a drying procedure that started with 72-80 hours of air drying followed by oven drying at 80°C (176°F) for 12 hours. The weight and dry weights were recorded in both weight per inch of stem and weight in mg per cm of stem.

The main result seen was that EWM biomass decreased at the Hessel Marina treatment site in the weeks after treatment, but stayed constant or increased at the two untreated control sites in the days and weeks after treatment. These results were similar to initial field trials in 2014 when the USDA was able to produce the Mt fungus. Between 25 and 30 DAT, a downward trend in EWM biomass at the Hessel Marina site vs. an increasing trend at the untreated Point Urie site could be identified (see Figure 23, which is Figure 1 from the Smith et al. 2018a report). Hessel Marina saw an almost 75% reduction in biomass 70 DAT when compared to the Point Urie and Howells Dock sites. These results were similar to the 2014 Mt trial in the LCI where an 85% biomass reduction was seen. The LCWC noted that water temperature dropped seven degrees F during the 70 DAT period but biomass loss continued. Smith et al. 2018a concluded that "Observations from these two open water trials indicate that Mt can reproducibly and significantly reduce EWM biomass in LCI waters, even when the water temperature is less than optimum.



Figure 23: Change in biomass at treated Hessel Marina site vs. untreated Urie Point site from time of treatment to 70 DAT; biomass declined at the treated site vs. the untreated site.

In the Smith et al. 2018a report, the LCWC authors briefly note that EWM growth appeared less vigorous one year after treatment. EWM biomass at Hessel Marina was less than one-half of the biomass vs. the untreated Urie Point area. No obvious impacts could be seen on non-target aquatic plants. The LCWC partners followed up this initial evaluation with a more detailed report, Smith et al. 2018b, entitled "Residual effect of *Mycoleptodiscus terrestris* (Mt) on *Myriophyllum spicatum* (Eurasian watermilfoil) one year post treatment", dated October 31, 2018. They describe EWM growth 410 DAT (one year and 45 days after the July 2017 treatment) as showing a residual effect that delayed EWM growth vigor in the early part of the 2018 growing season (Figure 24). In 2018, EWM density was at a level similar to the low density seen 70 DAT in 2017. The Hessel Marina site also saw significant growth of the native plants Coontail (*Ceratophyllum demersum*) and flatstem pondweed (*Potamogeton zosteriformis*), likely forming the "Other SAV" areas in the August 2018 UAS image and classification shown in Figure 19, whereas Urie Point was an EWM monoculture.





Independent sampling by Michigan Tech scientists prior to and one year following treatment with Mt fungus in support of UAV-based monitoring suggest that EWM persisted at the treatment sites at similar amounts as in untreated control sites. EWM comprised 16.5% of the visual cover at CRAMP one year following treatment, with no comparable pretreatment data collected. At Hessel Marina, EWM visual cover increased slightly from 26.3% to 33.6% one year following treatment within the observation areas near sampling points (note that the EWM percent cover shown in Figure 19 covers a larger area than these visual sampling locations, however). EWM cover increased 7% but decreased 14% at two different control sites, suggesting the magnitude of change observed in Hessel Marina one year post treatment could be due to natural variations in EWM populations (Figure 25) rather than necessarily being due to treatment. Similar changes in EWM abundance before and after treatment were detected using rake toss and AMAS surveys, with EWM increasing in Hessel Marina between August 2017 and 2017, and both increasing and decreasing at the control sites (Figure 26).



Figure 25: Visual estimates of cover at the treatment and control sites in August 2017 and 2018.



Figure 26: Estimates of EWM abundance using rake tosses and AMAS surveys at the treatment and control sites in July, August 2017 and August 2018.

4.5 Task 5: Reporting and Communication of Results

The project has included an active outreach program focused on communicating results to local stakeholders and the scientific community. In 2016, 2017, and 2018, results of the project were shared with the Les Cheneaux Watershed Council at their annual meeting, and through the Council newsletter. Additionally, PI Brooks presented to the LCWC annual meeting in person in July 5th, 2018, and answered questions from community members. PI Brooks and Co-I attended the community Frogfest on July 7th, 2018, and had a booth present where community members, including children, had the opportunity to ask questions of project scientists, look at the UAS and cameras used to collect data, see a UAS flight demonstration, and interact with a display with EWM and other aquatic vegetation in a fish tank. Figure 27 shows Dr. Marcarelli engaging with Frogfest attendees about the project, and Figure 28 shows Mr. Brooks discussing UAS-based imaging. The LCWC estimated the between 390 and 440 people attended Frogfest in 2018.



Figure 27: Dr. Marcarelli engaging with community members at the July 2018 Frogfest, helping explain the project and share information on submerged aquatic vegetation.



Figure 28: Mr. Brooks explaining how UAS can be used to collect EWM extent data during the 2018 LCI community Frogfest.

In addition to updating the EWM resource page from the "Arresting the spread…" GLRI project (<u>http://www.mtri.org/eurasian_watermilfoil.html</u>), a dedicated project web page was created and maintained at <u>http://www.mtri.org/ewmlci.html</u>. This enabled anyone interested in the project to get a rapid overview of the project and a rotating set of representative field photos and aerial images helping show what was going on with the project. The LCWC also shared information about the project through its webpage, including a project overview at <u>http://www.lescheneauxwatershed.org/library/grants/lci-eurasian-watermilfoil-control-glri-2016-</u>

17 and information about the previous vegetation and weevil surveys

(http://www.lescheneauxwatershed.org/library/nuisance-species/aquatic-vegetation-and-weevilsurveys).

Also, Mark Clymer of the LCWC recently completed an update to EWM best management practices, entitled "Best Management Practices Enhancement: Les Cheneaux Islands Eurasian Watermilfoil Control". After five seasons of Mt field work since 2013 by the LCWC, the updated BMPs describe Mt fermentation, transportation, application, safety, and permitting practices. This is being posted to the LCWC website; the main points are shown below as Table 8.

Table 8: Main points on Mt fungus treatment Best Management Practices, as updated through this project.

- 7. Mt BMP Summary
 - a. Fermentation process and formulation have not changed since the Mt Patent was registered in 2003.
 - i. Patent available at: <u>http://pdfpiw.uspto.gov/.piw?docid=06569807&PageNum=1&&IDKey=4D2</u> <u>6889BBEB4&HomeUrl=http://patft.uspto.gov/netahtml/PTO/patimg.htm</u>
 - b. Mt can be stored safely at 4C for up to one week. Further refinement of the Mt formulation's stability will be necessary to bring an Mt product to market.
 - c. Sheer issues were successfully addressed prior to this grant project by previous LCWC research and development. Future refinement of the LCWC proprietary application equipment is not necessary, except as may be competitively desired to apply larger volumes at a faster rate.
 - d. Used at full strength directly from the fermentor, Mt has been shown to be effective when applied at a rate of 40 gallons per acre.
 - e. Optimally, Mt will be applied when water temperatures are at or close to 22C.
 - f. A consistent Mt efficacy rate of over 85% has so far been demonstrated on ¼ acre open water, northern climate areas. Further demonstration projects on larger acreages are warranted based on this research.
 - g. A drift effect was recorded in the 2014 field work, but there were not enough data points to create a BMP.
 - h. Similarly, a carry-over effect was observed during 2018 monitoring, but samples collected were not dried & weighed due to the mix of EWM with other aquatic vegetation, yet this in itself showed signs of a newfound ability to compete with Mt 1 year after treatment. More research is needed in this area.
 - i. The safety of Mt was born out by the USDA genetic testing providing a positive correlation between the toxicity and pathogenicity data collected by LCWC, and Ecoscience data on file with EPA.
 - j. Mt does not grow at mammalian temperatures (37C).
 - k. This GLRI grant has supported efforts in applied research & application development of Mt as an effective Biopesticide for use in EWM control a step closer to commercial scale-up and EPA Registration.

Two project signs were also erected in the project area, near the primary project locations - one near the Hessel Marina boat ramp, and other one near the community boat ramp in Cedarville. Each sign provides project information, includes the GLRI logo, and credits the Great Lakes Restoration Initiative and EPA for funding. Figure 29 shows PI Brooks standing by the sign in the Hessel Marina that was right by the main treatment location.



Figure 29: PI Brooks standing near the project information sign placed in Hessel Marina right by the primary Mt treatment site.

Robert Smith from the LCWC involved high schools students from the Les Cheneaux Community Schools in understand EWM management in the area. In June of 2018, he presented to Cedarville High School science and Environmental Studies classes on the project and EWM management to about 24 students per class (about 48 total). Please note that each year-class at our HS is comprised of 25-30 students. The students then went on a field trip to the Cedarville boat ramp (CRAMP site) to see one of the active areas of EWM management. Mr. Smith made second presentation to six professors and eight graduate students at the University of Michigan Biological Station at Douglas Lake the evening of 24 July, 2018. He made a third presentation that summer on the project to the general public at theLes Cheneaux library in Cedarville on the evening of 26 July, 2018 which was attended by about 28 community members.

PI Brooks presented on the project at the International Association of Great Lakes Research (IAGLR) Annual Conferences in 2017 (Detroit) and 2018 (Scarborough/Toronto), as well as the 2018 Society for Freshwater Science Conference (Detroit). Mr. Brooks took the opportunity to share project results at other fortuitous outreach opportunities. He shared information on SAV mapping with UAS through a webinar hosted by the Electric Power Research Institute (EPRI) at

their February, 2018 Cooling Water Intake online meeting. At the 2017 Michigan Drone Day hosted by Eastern Michigan University, he featured the project work as one of eight UAV-enabled projects that he has worked on to help promote advanced technology implementation in Michigan and beyond. At the Ecological Society of America (ESA) conference in Ft. Lauderdale in August of 2016, he shared project plans and initial results on how remote sensing could help with SAV monitoring and management. An article about this and a colleague's Phragmites project was published in the Great Lakes Echo ("Fighting invaders with drones and fungi" - http://greatlakesecho.org/2016/09/30/fighting-invaders-with-drones-and-fungi/) which was shared over 90 times. Dr. Marcarelli and her graduate students also presented on the project at appropriate science conferences.

Publications (Peer-Review):

- Brooks, CN, Grimm, AG, Marcarelli, AM, Dobson, RJ. Multi-Scale Collection of Submerged Aquatic Vegetation Spectral Profiles for Eurasian Watermilfoil Detection. Submitted to and under review by the Journal of Applied Remote Sensing, December 2018.
- Van Goethem, RR. Effects of invasive watermilfoil and seasonal dynamics on primary production in littoral zones of north-temperate lakes. Masters of Science Thesis, Michigan Technological University, Houghton MI. Submitted December 2018.

Presentations (Science Conferences):

- Brooks, C. N., Grimm, A. G., Huckins, C. J., Marcarelli, A. M., Van Goethem, R., Dobson, R. J., Annual Conference on Great Lakes Research, "*Evaluating the spread and control of Eurasian watermilfoil through remote sensing technologies*" International Association for Great Lakes Research, Guelph, ON, Canada. (June 2016).
- Brooks, C., Grimm, A., Huckins, C. J., Marcarelli, A. M. (Presenter & Author), Annual meeting, "Development of a spectral-based algorithm for mapping and monitoring of Eurasian watermilfoil (Myriophyllum spicatum) in the Great Lakes region from an unmanned aerial vehicle platform" Ecological Society of America, Ft Lauderdale, FL. (August 2016).
- Brooks, C. N., Grimm, A. G., Huckins, C. J., Marcarelli, A. M., Van Goethem, R., Dobson, R., Annual Conference on Great Lakes Research, "Using Advanced Mapping Tools to Help Monitor Eurasian Watermilfoil for Improved Treatment Options" International Association for Great Lakes Research, Detroit, MI. (May 2017).
- Brooks, C., Marcarelli, A. M., Grimm, A. G., Dobson, R. J., Huckins, C. J., Van Goethem, R., Smith, R., Clymer, M., Marion, N., Annual Meeting, "ANALYZING EURASIAN WATERMILFOIL EXTENT AND TREATMENT EFFICACY USING UNMANNED AERIAL SYSTEM (UAS) MULTISPECTRAL IMAGERY" Society for Freshwater Science, Detroit, MI. (May 2018).
- Brooks, C., Marcarelli, A. M., Grimm, A. G., Dobson, R. J., Huckins, C. J., Van Goethem, R., Smith, R., Clymer, M., Annual Conference on Great Lakes Research, "*Demonstrating Unmanned Aerial System multispectral analysis of Eurasian watermilfoil treatments*" International Association for Great Lakes Research, Toronto, Canada. (June 2018).
- Marcarelli, A. M., Huckins, C. J., Juneau, K., Brooks, C., Chimner, R. A., Hersch-Green, E., Meadows, G. A., Midwest Aquatic Plant Management Society 36th Annual Meeting, "Integrated management of nonnative and hybrid Eurasian Watermilfoil in the Portage Waterway of the Upper Peninsula of Michigan" Midwest Aquatic Plant Management Society, Grand Rapids, MI. (March 8, 2016).

- Van Goethem RR, Marcarelli AM, Huckins CJ, Juneau JJ. Legacy disturbance in a lake littoral zone: effects of mining residue on the composition of macrophyte communities. Society for Freshwater Science Annual Meeting, Raleigh NC. (June 2017).
- Van Goethem, R. (Presenter & Author), Marcarelli, A. M., Huckins, C. J., Annual Meeting, "Effects of Invasive Macrophytes on Littoral Primary Producers in North Temperate Lakes" Midwest Aquatic Plant Management Society, Cleveland, OH. (February 2018).
- Van Goethem, R. (Presenter & Author), Marcarelli, A. M., Huckins, C. J., Annual Meeting, "EFFECTS OF INVASIVE MACROPHYTES ON LITTORAL PRIMARY PRODUCERS IN NORTH-TEMPERATE LAKES" Society for Freshwater Science, Detroit, MI. (May 2018).

5. Conclusions

Cumulative results achieved

The main cumulative results achieved through this GLRI-sponsored project were three-fold:

- 1) Taking a relatively new treatment method using the native Mt fungus to the point of practical deployment in the Great Lakes.
- 2) Demonstrating how UAS-enabled sensing can provide quantitative mapping information that helps monitor treatment methods such as mechanical harvesting, DASH treatment, and Mt biocontrol.
- 3) Deployment of a robust field sampling protocol that provides the needed information to document changes in EWM extent.

All three of these have been documented here, presented to local community members and other stakeholders, presented at scientific conferences, and form the main information for one Master's thesis and one dissertation paper (with two more related dissertation papers planned). A close partnership with the Les Cheneaux Watershed Council meant that community engagement, local employment, and science outreach were all possible. The Mt fungus treatment resulted in lower Mt biomass in the weeks after treatment relative to two untreated control sites, and there may be residual effects of the treatment one year later.

For post-completion activities, these will focus on writing and submitting additional peerreviewed papers, along with continuing to work with the LCWC on pursuing opportunities to treat larger areas with the Mt fungus. Dissertation papers focused on an EWM-specific mapping algorithm, and on application of this algorithm to EWM treatment monitoring, will be submitted by PI (and PhD candidate) Brooks in 2019. Michigan Tech and the LCWC are investigating possible funding sources to continue their partnership in expanding methods of practical EWM treatment and extending results to the Great Lakes community.

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