Pike Chain of Lakes Bayfield County, Wisconsin 2023 EWM Management & Monitoring Report April 2024

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1.0 INTRODUCTION

The Pike Chain of Lakes is comprised of nine lake basins located near the Town of Iron River in Bayfield County, Wisconsin (Figure 1.0-1). The chain includes over 1,000 acres of surface water, and forms the headwaters of a drainage system that leads to the White River which flows through the Bad River Indian Reservation on its way to Lake Superior. Six of the lakes, sometimes referred to as the main lakes, are able to be boated between (colored blue on Figure 1.0-1). The other three lakes are hydrologically connected but cannot be reached by watercraft without a portage (shown in green).

All lakes within the chain are considered Priority Navigable Waterways by the Wisconsin Department of Natural Resources (WDNR), primarily for having waters with self-sustaining walleye and/or muskellunge populations. The six main lakes and Pike Lake are classified as Areas of Special Natural Resource Interest due to their outstanding or exceptional resource waters.

One non-native submergent plant species has been identified within the Pike Chain, Eurasian watermilfoil (*Myriophyllum spicatum*, EWM). EWM was first documented in the Twin Bear – Hart Channel in 2004. EWM populations were identified in Eagle Lake in 2005, Buskey Bay in 2007, and Millicent in 2008. Flynn Lake was the last lake for EWM to be identified within during surveys in 2014. The Iron River Pike Chain of Lakes Association (IRPCLA) and partners have historically managed EWM with spatially targeted



herbicide spot treatments, whole-lake 2,4-D treatments, and hand-harvesting efforts (volunteer and contracted). More recent management strategies have included 2,4-D treatments with the aid of a barrier curtain as well as spot and whole-lake treatments with florpyrauxifen-benzyl, the primary active ingredient in ProcellaCORTM.

1.1 Historic AIS Management & Planning

The IRPCLA's *Comprehensive Management Plan* (Dec 2008) for the Pike Chain of lakes outlines an EWM management strategy that primarily uses herbicide spot treatments. An official addendum to the *Plan* was made in January 2016 that incorporated whole-lake treatment philosophies, following the completion of a 5-year AIS-Established Population Control Grant-funded project. The IRPCLA was awarded a proceeding WDNR AIS Established Population Control Grant in February 2016 (ACEI-180-16) that ultimately funded EWM management and monitoring from 2016-2020. As a part of that project, the IRPCLA revisited their aquatic plant management-related Implementation Plan and updated its



content based on the lessons learned during the EWM control project. The *Aquatic Plant Management Plan (Plan)* was completed in November 2021 following the collaboration of multiple state, county, and tribal partners.

Within the *Plan*, the IRPCLA outlined a management goal to "Manage Aquatic Invasive Species and Prevent Establishment of New Aquatic Invasive Species." This goal includes a management action to "conduct management actions towards Eurasian watermilfoil" including a density-based trigger of when herbicide use would be applicable. The *Plan* outlines herbicide formulation recommendations, treatment design constraints, and likely monitoring strategies that are consistent with current Best Management Practices (BMPs) at the time of the *Plan* construction.

1.2 2023 EWM Management Strategy

While the Aquatic Plant Management Plan (Plan) provides a framework to guide the overall management direction, the specific control and monitoring plan for a given year are outlined in the preceding annual control plan. As technology and BMPs evolve, this allows incorporation of these facets during the lifespan of the *Plan*. The annually-produced control and monitoring plan is useful for WDNR and tribal regulators when considering approval of the action, as well as to convey the control plan to IRPCLA members for their understanding. The preliminary 2023 control and monitoring plan was outlined within the 2022 EWM Management & Monitoring Report distributed in January 2023. This strategy was also incorporated into a successful WDNR AIS Large-Scale Population Control Grant application (ACEL 22223), providing state share assistance in control and monitoring for the state share assistance in control and successful within the 2022 Bart of the state share assistance in control within the state share assistance in control within the state share assistance in control within the state share assistance in control within the state share assistance in control within the state share share assistance in control within the state share share assistance in control within the state share share state share assistance in control within the state share share assistance in control within the state share state state share state share state share state state state share state stat

application (ACEI-33223), providing state-share assistance in carrying out the effort.

The IRPCLA conducted a series of treatments in 2023 with follow-up hand-harvesting/DASH mostly for lakes which had an herbicide treatment in 2022 (Table 1.2-1). Buskey Bay contained the highest EWM population and was targeted with ProcellaCOR spot treatments that may have potential lake/basin-wide impacts (Map 1). A specific area of Millicent also contained a high EWM population and was conducive to an enclosure with a barrier curtain and 2,4-D while other sites were targeted with ProcellaCOR spot treatments (Map 2).

Table 1.2-1. R management ac	ecent tivitie	EWM s.
Lake	2023	2022
Buskey Bay		
Lake Millicent		
Hart Lake		
Twin Bear Lake		
Eagle Lake		
Flynn Lake		
Herbicide treat	nent	
Hand harvesting	g w/ D	ASH

1.3 Pretreatment Confirmation and Refinement Survey

Onterra ecologists conducted the Pretreatment Confirmation and Refinement Surveys on the Pike Chain on June 12 and 13, 2023. Aside from the collection of the pretreatment sub-sample point-intercept aquatic plant data, the survey evaluated the growth stage of the EWM population in the treatment areas as well as confirmed the average depth of the sites for dosing purposes. The survey was conducted using a combination of survey methods, but largely consisted of visual observations as the EWM was visible from the surface. Water temperatures at mid-depth were about 68°F. Using an optical probe, the pH was measured at 8.4 in Buskey Bay and 8.2 in Lake Millicent. New EWM growth was apparent on the target plants and appeared to be in an active growth stage ideal for treatment (Photo 1.3-1). The relative accuracy of the average depth of the treatment sites were confirmed. No alterations were recommended to the treatment plan as a result of the pretreatment survey.



Onterra.

Onterra delivered the post-treatment herbicide concentration monitoring supplies to a volunteer member of IRPCLA. A video sampling instructional tutorial was made available to the IRPCLA along with spatial data for use with smartphone applications and handheld GPS to guide the volunteers to the exact sampling locations. The herbicide treatments were completed on June 30, 2023 by Northern Aquatic Services. The applicator noted south-southwest winds between 5-7 mph during the time of the applications.

2.0 MONITORING METHODOLOGIES

It is important to note that two types of surveys are discussed in the subsequent materials: 1) pointintercept surveys and 2) EWM mapping surveys. Overall, each survey has its strengths and weaknesses, which is why both are utilized in different ways as part of this project.

The point-intercept survey provides a standardized way to gain quantitative information about a lake's aquatic plant population through visiting predetermined locations and using a rake sampler to identify all the plants at each location. The survey methodology allows comparisons to be made over time, as well as between lakes. The point-intercept survey can be applied at various scales. The point-intercept survey is most often applied at the whole-lake scale.

If a smaller area is being studied, a modified and finer-scale point-intercept sampling grid may be needed to produce a sufficient number of sampling points for comparison purposes. The <u>sub-sample point-intercept survey</u> methodology is often applied over management areas such as herbicide application sites. This type of sampling is used within this project as a part of the spot-herbicide treatment monitoring.

While the point-intercept survey is a valuable tool to understand the overall plant population of a lake, it does not offer a full account (census) of where a particular species exists in the lake. During the EWM mapping survey, the entire littoral area of the lake is surveyed through visual observations from the boat.



Field crews supplement the visual survey by deploying a submersible camera along with periodically doing rake tows. The EWM population is mapped using sub-meter GPS technology by using either 1) point-based or 2) area-based methodologies. Large colonies >40 feet in diameter are mapped using polygons (areas) and are qualitatively attributed a density rating based upon a five-tiered scale from *highly scattered* to *surface matting*. Point-based techniques were applied to AIS locations that were considered as *small plant colonies* (<40 feet in diameter), *clumps of plants*, or *single or few plants*. Annual late-summer EWM mapping surveys serve to evaluate the management occurring on the system.





3.0 2023 HERBICIDE TREATMENT MONITORING RESULTS

3.1 Buskey Bay

ProcellaCOR Herbicide Concentration Monitoring

The herbicide concentration monitoring plan associated with the treatment was developed by Onterra and the WDNR, with the intent of gaining sufficient data to aid in understanding the concentrations of herbicide that were achieved after treatment. Samples were collected at three sites following treatment – two within application areas, and one site located in the deep hole area of the lake. Past herbicide concentration monitoring in the state has shown the primary active ingredient in ProcellaCOR, florpyrauxifen-benzyl, typically falls below detection limits by 14 days after treatment (DAT), however the first breakdown product, florpyrauxifen acid, has been found to persist considerably longer in some case studies. Therefore, the monitoring for this project included samples to be collected at 28, 42, 56, and 70 DAT to gain further understanding on the persistence of florpyrauxifen acid. A copy of the herbicide concentration monitoring plan is included as Appendix A.

All samples were collected by a volunteer member of the association and upon completion of the sampling, were shipped to EPL Bio Analytical Services for analysis. This lab has the ability to detect florpyrauxifen-benzyl and the florpyrauxifen acid at levels below 1 part per billion (ppb). The lab reports the concentration in parts per billion (ppb) of florpyrauxifen-benzyl, as well as florpyrauxifen acid.

For reference, the dosing rate of 4.0 PDU (prescription dosing units) equates to approximately 7.7 ppb of florpyrauxifen-benzyl. Concentrations of florpyrauxifen-benzyl were highest in the early sampling intervals at site B2 and measured 1.856 ppb at 3 HAT (Figure 3.1-1). Concentrations at B1 were lower than B2 and B3 in intervals between 9 HAT and 2 DAT. Concentrations at the untreated site (B3) were initially near 0.5 ppb and declined to below detection limits at 4 DAT and were 0.042 at 7 DAT. All samples that were collected at 14 DAT or later were below detection limits for florpyrauxifen-benzyl which is consistent with expectations as the active ingredient is broken down to acid form and is typically below detection limits within the first week or two after application.

The primary breakdown product of florpyrauxifen-benzyl is florpyrauxifen acid. Florpyrauxifen acid has been shown to persist in the lake longer than the active ingredient. This chemical metabolite is reported to have activity as an herbicide on aquatic plants, albeit to a lower degree than the active ingredient. It is unclear at this time the exact role that the acid metabolite may play in contributing to EWM reductions, particularly in areas not located directly within the herbicide application area.

Concentrations florpyrauxifen acid are displayed on Figure 3.1-2. Acid concentrations were higher at sites within application areas (B1 & B2) compared to the untreated site (B3) during samples collected out to 7 DAT. All samples collected after 7 DAT were from site B3 and were above detection limits at around 0.1 ppb at 14 DAT and 56 DAT while being below detection limits at 28 DAT, 42 DAT, and 70 DAT.







Figure 3.1-2. Buskey Bay 2023 Florpyrauxifen acid concentration monitoring results.

Qualitative Monitoring: EWM Mapping Surveys

Four sites comprising of 11.7-acres in Buskey Bay were treated with ProcellaCORTM in June 2023 (Map 1). All sites harbored colonized EWM in the September 2022 pretreatment survey (Figure 3.1-3 - left frame). After treatment, no EWM was located in application area BB C-23, while the other three application areas were found to contain sparse EWM occurrences consisting of either *single plants* or *clumps of plants* (Figure 3.1-2-right frame). The posttreatment mapping survey also showed EWM reductions throughout the entire waterbody including areas not located within direct application areas. This indicates that the herbicide treatment likely had lake-wide impacts.





Quantitative Monitoring: Subsample Point-Intercept Surveys

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 75 sampling locations within all four application areas in Buskey Bay. The quantitative assessment is completed through the comparison of the sub pointintercept survey from mid-June 2023 (year of pretreatment), late-season 2023 (year of posttreatment), and late-season 2024 (year after treatment). The 2023 herbicide treatment was planned for roughly the middle of June and ultimately occurred on June 20. This slight delay in implementation allows the pretreatment sub-sample point-intercept survey to take place after many native plants have emerged from winter dormancy. In the analysis within the following sections, the pretreatment data were collected on June 12-13, 2023 and the post-treatment data were collected on September 6, 2023.

Site BB A-23

Monitoring from Site BB A-23 included the

completion of a sub-sample point-intercept survey from 26 sampling locations within the application area. Analysis of these data show that the occurrence of EWM was reduced from 30.8% prior to treatment to 3.8% in the post-treatment survey. The occurrence of variable-leaf pondweed and white-stem pondweed showed statistically higher occurrences in the post-treatment survey; however, these increases may be due to the seasonality of the survey timing. Northern watermilfoil is known to be highly susceptible to ProcellaCORTM treatments and was present in the post-treatment survey (7.7%) which was slightly higher than the pre-treatment survey. Most native species showed no statistical change in occurrence between the two surveys.







Site BB B-23

Monitoring from Site BB B-23 included the completion of a sub-sample point-intercept survey from 29 sampling locations within the application area. Analysis of these data show that the occurrence of EWM was reduced from 24.1% prior to treatment to 3.4% in the post-treatment survey (Figure 3.1-6). The occurrence of stiff pondweed and wild celery showed statistically higher occurrences in the post-treatment survey; however, these increases may be due to the seasonality of the survey timing. Coontail, fern-leaf pondweed, and common waterweed were very common in the pre-treatment survey and all showed no changes in occurrence in the posttreatment survey.



Site BB C-23

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 10 sampling locations within the application area. Table 3.1-1 lists the aquatic plants that were present before and after treatment. EWM was sampled on one point before treatment and was not encountered in the post treatment replication of the survey. Many native species were identified in the post treatment survey. The sample size from this site is too small for meaningful statistical analysis.

Table 3.1-1. Species	sampled before and	d after Proce	llaCOR treatr	nent at site BB C-23 (n=10)
		Number of S	ampling Points	
Scientific Name	Common Name	Pre-Treatment	Post-Treatment	
Ceratophyllum demersum	Coontail	2	2	
Chara spp.	Muskgrasses	3	0	
Eleocharis acicularis	Needle spikerush	1	0	
Elodea canadensis	Common waterweed	3	3	
Heteranthera dubia	Water stargrass	1	2	
Myriophyllum spicatum	Eurasian watermilfoil	1	0	
Najas flexilis	Slender naiad	0	2	
Najas guadalupensis	Southern naiad	0	2	
Potamogeton gramineus	Variable-leaf pondweed	0	2	
Potamogeton pusillus	Small pondweed	0	2	
Potamogeton richardsonii	Clasping-leaf pondweed	0	1	
Potamogeton robbinsii	Fern-leaf pondweed	2	1	
Potamogeton strictifolius	Stiff pondweed	0	1	
Vallisneria americana	Wild celery	1	2	

Site BB D-23

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 10 sampling locations within the application area. Table 3.1-2 lists the aquatic plants that were present before and after treatment. EWM was sampled on two points before treatment and was not encountered in the post treatment replication of the survey. The sample size from this site is too small for meaningful statistical analysis.

Table 3.1-2. Species s	ampled before and aft	er ProcellaCO	OR treatment a	t site BB D-23 (n=10)
		Number of Sa	ampling Points	
Scientific Name	Common Name	Pre-Treatment	Post-Treatment	[]
Nymphaea odorata	White water lily	2	2	
Ceratophyllum demersum	Coontail	2	2	
Ranunculus aquatilis	White water crowfoot	0	1	
Myriophyllum spicatum	Eurasian watermilfoil	2	0	
Vallisneria americana	Wild celery	1	2	4
Heteranthera dubia	Water stargrass	3	1	6 1
Chara spp.	Muskgrasses	1	2	
Potamogeton gramineus	Variable-leaf pondweed	1	1	
Sagittaria sp. (rosette)	Arrowhead sp. (rosette)	0	1	
Najas flexilis	Slender naiad	0	1	
Elodea canadensis	Common waterweed	0	1	
Potamogeton robbinsii	Fern-leaf pondweed	1	0	

3.2 Lake Millicent (2,4-D barrier curtain)

Part of the 2023 treatment plan targets a 1.5 acres site on the northern end of Lake Millicent with the aid of a barrier curtain (Map 2). Along with a few other stipulations, the WDNR does not require any additional permits, aside from normal NR 107 Herbicide Treatment Permit, to implement a barrier curtain so long as access is not denied to any part of the system and the curtain is in place for no more than 96 hours. The curtain is typically deployed the day prior to treatment, then held in place for 72 hours after the herbicide applicator conducts the treatment the following morning. Volunteer members of the IRPCLA constructed and placed the barrier curtain in advance of the treatment. The 2,4-D application occurred on June 20, 2023 at an application area dosing rate of 4.0 ppm.

2,4-D Concentration Monitoring

The herbicide concentration monitoring plan associated with the treatment was developed by Onterra and the WDNR, with the intent of gaining sufficient data to aid in understanding the concentrations of the herbicide 2,4-D that were achieved in the hours after treatment. The herbicide was applied as liquid 2,4-D amine, with herbicide concentration analysis occurring by the Wisconsin State Laboratory of Hygiene (WSLH) and reporting the results as 2,4-D acid equivalent (ae). A copy of the Herbicide Concentration Sample Plan is included in Appendix A.

The 2023 herbicide concentration samples were collected by volunteers at four separate sites - two within the barrier (M4 and M5), and two outside of the barrier (M6 and M7). Samples were collected beginning at one hour after treatment (HAT), with additional samples collected at 6, 24, 48, and 72 HAT. At 72 HAT, the curtain barrier was removed and additional samples were collected at 1, 3, 6, 12, and 24 hours after curtain removal. All samples were preserved and sent to the WSLH for analysis.

Figure 3.2-1 displays the concentration of 2,4-D at the four monitoring locations in parts per million (ppm) to be consistent with the units of the dosing strategy (4.0 ppm ae). Concentrations of the herbicide were approximately half of the target rate from samples collected within the barrier curtain with the highest concentration measured at 1.7 and 1.9 ppm from sites within the barrier curtain at 1 HAT. Concentrations gradually decreased from samples collected within the barrier to approximately 0.6-0.7 ppm at 72 HAT just prior to curtain removal. By comparison, minimal 2,4-D was detected in either of the two sampling sites located outside the barrier while the barrier was in place. This indicates that the barrier curtain functioned as intended with minimal herbicide loss. Once the barrier curtain was removed, concentrations decreased rapidly within the treatment site as the herbicide dissipated within surrounding waters. At the final sampling interval 24 hours after the curtain removal, detectable levels of 2,4-D were measured at all four monitoring sites, with the highest concentration of 0.19 ppm collected from site M4.





Qualitative Monitoring: EWM Mapping Surveys

The pretreatment EWM population consists of colonized EWM including highly scattered, scattered, and highly dominant densities (Figure 3.2-2, left frame). After treatment, no EWM was located in the area that was contained by the barrier curtain (Figure 3.2-2, right frame).



Quantitative Monitoring: Subsample Point-Intercept Survey

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 17 sampling locations within the application area. For this monitoring, the pretreatment dataset was collected on June 13, 2023 and the posttreatment dataset was collected on September 6, 2023. Analysis of these data show that the occurrence of EWM was reduced from 11.8% prior to treatment to 0% in the post-treatment survey (Figure 3.2-3). The occurrence of wild celery showed statistically higher occurrences in the post-treatment survey; however, these increases may be due to the seasonality of the survey timing. No statistically significant declines were observed following the treatment.



3.3 Lake Millicent (ProcellaCOR Treatments)

ProcellaCOR Herbicide Concentration Monitoring

The herbicide concentration monitoring plan associated with the treatment was developed by Onterra and the WDNR, with the intent of understanding the concentrations of herbicide that were achieved after treatment. Samples were collected at three sites following treatment – two within application areas, and one site located in the deep hole area of the lake. The monitoring for this project included samples to be collected at 28, 42, 56, and 70 DAT to gain further understanding on the persistence of florpyrauxifen



acid. All samples were collected by a volunteer member of the association and upon completion of the sampling, were shipped to EPL Bio Analytical Services for analysis. A copy of the herbicide concentration monitoring plan is included as Appendix A.

For reference, the dosing rate of 4.5 PDU equates to approximately 8.67 ppb of florpyrauxifen-benzyl. Concentrations of florpyrauxifen-benzyl measured below 1.0 ppb in all samples (Figure 3.3-1). The earliest intervals measured higher concentrations in the application area sites compared to the untreated site, which is consistent with expectations. Concentrations at site M1 were measured at 0.071 ppb at 7 DAT, and were below detection limits in all other intervals which indicates that uniform lake-wide mixing of florpyrauxifen-benzyl likely did not occur with this treatment. All samples that were collected at 14 DAT or later were below detection limits for florpyrauxifen-benzyl which is consistent with expectations.



Concentrations of the florpyrauxifen acid are displayed on Figure 3.3-2. Acid concentrations were higher at sites within application areas (M2 & M3) compared to the untreated site (M1) during samples collected out to 4 DAT. Acid concentrations were near 0.06 ppb at site M3 at 7 DAT, and were below detection limits at the other two sites at the same interval. Samples collected beyond 14 DAT from site M1 were largely intended to measure acid persistence and measured near 0.01 ppb at 28 DAT and 42 DAT and was below detection limits at 56 DAT and 70 DAT.





Qualitative Monitoring: EWM Mapping Surveys

Four sites comprising of 5.9-acres in Lake Millicent were treated with ProcellaCORTM in June 2023 (Map 2). All sites harbored colonized EWM in the September 2022 pretreatment survey (Figure 3.3-3 - left frame). After treatment, no EWM was located within any of the four application areas (Figure 3.3-3-right frame). The posttreatment mapping survey also showed EWM reductions throughout the entire waterbody including areas not located within direct application areas. The only EWM located in the posttreatment survey was several single or few plants occurrences on the southern end of the lake and furthest away from the application areas. This indicates that the herbicide treatment likely had lake-wide impacts.





Quantitative Monitoring: Subsample Point-Intercept Surveys

Quantitative monitoring consisted of the completion of a subsample point-intercept survey from 56 sampling locations within all four application areas in Lake Millicent (Figure 3.3-4). The quantitative assessment is completed through the comparison of the sub point-intercept survey from mid-June 2023 (*year of pretreatment*), late-season 2023 (*year of post-treatment*), and lateseason 2024 (*year after treatment*). In the analysis within the following sections, the pretreatment data were collected on June 12-13, 2023 and the post-treatment data were collected on September 6, 2023.

Site M B-23

Monitoring from Site M B-23 included the completion of a subsample point-intercept survey from nine sampling locations within the application area. EWM was present at five sampling sites before treatment compared to one site after treatment (Table 3.3-1). The number of points with wild celery increased between the two surveys likely due to the seasonality of the survey timing. The sample size from this site is too small for meaningful statistical analysis.



Quantitative Monitoring Plan for 2023 Herbicide Treatments. 25m spacing, n=56

		Number of S	ampling Points
Scientific Name	Common Name	Pre-Treatment	Post-Treatment
Vallisneria americana	Wild celery	0	4
Myriophyllum spicatum	Eurasian watermilfoil	5	1
Potamogeton gramineus	Variable-leaf pondweed	2	2
Eleocharis acicularis	Needle spikerush	2	2
Chara spp.	Muskgrasses	3	1
Potamogeton pusillus	Small pondweed	0	2
Najas flexilis	Slender naiad	0	2
Elodea canadensis	Common waterweed	2	1
Potamogeton robbinsii	Fern-leaf pondweed	1	1
Ceratophyllum demersum	Coontail	1	1
Potamogeton praelongus	White-stem pondweed	0	1
Potamogeton amplifolius	Large-leaf pondweed	0	1

Site M C-23

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 12 sampling locations within the application area. EWM was present at seven sampling sites before treatment compared to one point in the post treatment survey (Table 3.3-2). Many native species were present in the pre and post treatment surveys. The sample size from this site is too small for meaningful statistical analysis.



		Number of S	ampling Points
Scientific Name	Common Name	Pre-Treatment	Post-Treatment
Elodea canadensis	Common waterweed	2	5
Vallisneria americana	Wild celery	1	5
Chara spp.	Muskgrasses	4	3
Myriophyllum spicatum	Eurasian watermilfoil	7	1
Potamogeton pusillus	Small pondweed	0	3
Ceratophyllum demersum	Coontail	2	2
Potamogeton gramineus	Variable-leaf pondweed	0	2
Eleocharis acicularis	Needle spikerush	2	1
Potamogeton robbinsii	Fern-leaf pondweed	2	0
Myriophyllum tenellum	Dwarf watermilfoil	0	1
Heteranthera dubia	Water stargrass	2	0
Potamogeton amplifolius	Large-leaf pondweed	1	0

Site M D-23

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 17 sampling locations within the application area. Analysis of these data show that the occurrence of EWM was reduced from 41.2% prior to treatment to 0% in the post-treatment survey (Figure 3.3-5). The occurrence of small pondweed showed statistically higher occurrences in the post-treatment survey. With the exception of EWM, no statistically significant declines of native plants were observed.





Site M E-23

Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 18 sampling locations within the application area. Analysis of these data show that the occurrence of EWM was reduced from 27.8% prior to treatment to 0% in the post-treatment survey (Figure 3.3-6). The occurrence of wild celery showed statistically higher occurrences in the post-treatment survey; however, these increases may be due to the seasonality of the survey timing. Once native species, coontail, observed a statistically valid decline in occurrence.



Figure 3.3-6. Littoral frequency of occurrence of aquatic plants Site M E-23 in Lake Millicent. Asterisk represents statistically valid change from June 2023 to September 2023 (Chi-Square α = 0.05). n=18.

4.0 PROFESSIONAL HAND-HARVESTING ACTIVITIES

The IRPCLA contracted with a professional firm to conduct hand harvesting efforts within the Chain during 2023. Divers from Aquatic Plant Management, LLC conducted four days of removal efforts that resulted in a harvest of 184.5 cubic feet of EWM over five sites (Table 4.0-1). Details of the professional harvesting activities are included within Appendix B.

ble 4.0-1. 2023 professional DASH hand-harvesting activities in Pike Chain. Table extracted fro M Summary Report.							
Dive Location	Avg. Water Depth	# of Dives	Underwater Dive Time	AIS Removed (cubic feet			
Eagle HH	6.0	1	3.3	10.0			
Flynn HH	5.5	1	3.3	5.5			
Hart-A-23	12.0	1	0.6	1.5			
Hart-B-23	8.8	10	9.3	85.5			
TB-A-23	12.0	5	7.0	82.0			
Grand Total	9.5	18	23.4	184.5			

As has occurred in past years, the EWM population in the Pike Chain of Lakes was mapped professionally during Onterra's 2022 and 2023 Late-Season EWM Mapping Surveys. The results from these surveys can be used to compare hand harvesting efforts each year. Each lake is assessed individually below for the hand harvesting efforts completed in 2023. The 2023 harvesting efforts were guided by the results of the late-summer 2022 EWM mapping survey. It is acknowledged that in some cases, EWM population expansion may have occurred between the time of the late-summer 2022 survey and the start of the hand harvesting activities in summer 2023. If EWM expansion occurred prior to the start of the harvesting efforts, the pre- harvesting data presented in the figures below may under-represent the true extent of the population before harvesting efforts began.

Hart Lake

Hand harvesting in Hart Lake was completed on the southern shoreline where EWM has historically held high populations in the past. This site was treated with ProcellaCOR in 2022 and the 2023 hand harvesting strategy was to target the remaining EWM population that was identified during the 2022 late-summer mapping survey. The majority of the 2023 professional hand harvesting efforts were focused in Hart Lake with approximately 87 cubic feet of EWM harvested over the course of 11 dives. The EWM population before and after harvesting efforts in the hand harvesting area is highlighted in Figure 4.0-1. The EWM population decreased in the area when compared to the late-summer 2022 results. Hand harvesting in this site met expectations of the strategy in that the EWM population did not increase in the targeted area.





Twin Bear Lake

Hand harvesting in Twin Bear Lake was completed in one area on the western shoreline. Approximately 82 cubic feet of EWM was harvested during five dives. The EWM population before and after harvesting efforts in the site is highlighted in Figure 4.0-2. The EWM population remained about the same in the area when compared to the late-summer 2022 results. Professional harvesting assisted in maintaining the current population level in this area while inhibiting population expansion from occurring.





Eagle Lake & Flynn Lake

Hand harvesting in Eagle Lake was completed within the channel leading towards Twin Bear Lake. Approximately 10 cubic feet of EWM was removed during one dive (Appendix B). Hand harvesting in Flynn Lake was completed near the outlet channel and targeted known *single or few plants* in the area and yielded a harvest of approximately 5.5 cubic feet of EWM during one dive (Appendix B). The EWM population before and after harvesting efforts in the hand harvesting area remained at similar levels with only *single or few plants* occurrence present in the site.

5.0 2022 HERBICIDE TREATMENTS – YEAR AFTER TREATMENT MONITORING

Extended monitoring of the 2022 herbicide treatment sites in Twin Bear Lake, Hart Lake and Eagle Lake took place during 2023 to evaluate the treatment efficacy during the *year after treatment*. All sites showed initial decrease in EWM during the post-treatment survey conducted a few months after treatment in late-summer 2022, but an evaluation of the sites in 2023 is used to understand whether the treatments met control expectations of EWM reductions lasting through the year after treatment.



5.1 Twin Bear Lake

2,4-D Barrier Curtain Site TB A-22

Before the treatment, a *highly dominant* EWM colony was present near the center of the site along with several *single or few plant* occurrences and a *clump of plants* (Figure 5.1-1). Following the 2,4-D treatment, one *single or few* EWM plants occurrence was located within the treated area in late-summer 2022 and two *single or few plants* were present during the year-after-treatment survey completed in September 2023. This treatment met success criteria for the site with EWM reductions extending through the year-after-treatment.





Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from seven sampling locations within the application area. These data show that EWM was present at three of the seven sampling locations before treatment and was not present at any of the sampling locations after treatment (Table 5.1-1). Native aquatic plant species that were present before the treatment were all still detected post-treatment. Several additional native species were sampled in the post-treatment survey that were not sampled before the treatment. This is likely a reflection of the timing of the surveys in which some species may have been dormant during the mid-June pretreatment survey timing compared with the September post treatment survey.

Table	Table 5.1-1. Pre/Post analysis of aquatic plants in A-22. 2022 2,4-D treatment in Twin Bear Lake. n=7									
	Number of Sampling Points									
	Scientific Name	Common Name	Pre Treatment	Post Treatment	1-Year Post Treatment					
	Chara spp.	Muskgrasses	2	4	4					
	Ceratophyllum demersum	Coontail	3	2	2					
	Lemna trisulca	Forked duckweed	1	2	1					
	Potamogeton pusillus	Small pondweed	0	2	0					
	Najas flexilis	Slender naiad	1	1	1					
	Myriophyllum spicatum	Eurasian watermilfoil	3	0	0					
	Elodea canadensis	Common waterweed	1	1	3					
	Potamogeton robbinsii	Fern-leaf pondweed	0	1	1					
	Potamogeton gramineus	Variable-leaf pondweed	0	1	2					
	Potamogeton amplifolius	Large-leaf pondweed	0	1	0					
	Nuphar variegata	Spatterdock	0	1	0					
	Heteranthera dubia	Water stargrass	0	0	2					
	Potamogeton richardsonii	Clasping-leaf pondweed	0	0	1					

ProcellaCOR spot-treatment site TB B-22

Site TB B-22 comprised 5.6 acres and was treated with ProcellaCORTM in June 2022 at a dosing application rate of 4.5 PDU's. The EWM population was reduced after treatment to consist of *single plants*, *clumps of plants* and *small plant colonies*. The EWM reductions during the year of treatment did not meet initial expectations because in most cases little no EWM is present in the treated site during the year of treatment. Follow up hand harvesting efforts took place during 2023 which are highlighted in Figure 4.0-2 above. These efforts resulted in the EWM population remaining at approximately the same levels between 2022-2023. Management efforts in this site over the past few years have resulted in a reduced EWM population; however, the overall level of control has been somewhat lower than other managed sites around the Pike Chain of Lakes system in recent years.





Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 24 sampling locations within the application area collected pretreatment, posttreatment, and year after treatment. Analysis of these data showed EWM initially showed a 100% decrease in occurrence during the year of treatment, while the occurrence in the year after treatment represents a 75% decrease compared to pretreatment (Figure 5.1-3). No native species showed statistically valid decreases in occurrence, while a few species showed valid increases in occurrence over the period of study.





5.2 Hart Lake

Site Hart C-22 (ProcellaCOR Spot Treat)

Site C-22 comprises 13.3-acres in Hart Lake and was treated with ProcellaCORTM in June 2022 at a dosing application rate of 3.5 PDU's. The pretreatment EWM population in the site included *dominant* density colonies in the eastern portions of the site as well as other small colonized areas and several *small plant colonies, clumps of plants,* and *single or few plants* (Figure 5.2-1). The post-treatment mapping survey indicated a reduction of EWM in many areas of the site, notably on the eastern end, although EWM was still present in the northern and central portions of the treatment site.

This site was targeted with professional hand harvesting efforts during 2023 as a part of the IRPCLA's integrated pest management strategy. The 2023 professional harvesting efforts served to prolong the longevity of control stemming from the 2022 herbicide treatment, potentially delaying the need for a future herbicide treatment in this area. The late-summer 2023 mapping survey shows an overall similar EWM population compared to 2022 which confirms the EWM population reductions extended through the year after treatment and no large EWM population recovery took place. Details of the 2023 professional harvesting efforts in this site are discussed in Section 4.0 of this report.



Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 61 sampling locations within the application area. Analysis of these data show that the occurrence of EWM was reduced from 8.2% prior to treatment to 0% in the post-treatment survey (Figure 5.2-2). The occurrence of small pondweed, variable-leaf pondweed, and slender naiad showed statistically higher occurrences in the post-treatment survey during the year of treatment. Coontail and muskgrasses each showed valid decreases in occurrence between 2022-2023, but these species did not show changes in occurrence comparing pretreatment (2021) to year of treatment (2022).





5.3 Eagle Lake

Sites E-22 & F-22 (ProcellaCOR Spot Treat w/ whole lake potential)

The two large herbicide spot treatments in 2022 in Eagle Lake collectively functioned as anticipated as a whole-lake treatment based on the documented posttreatment EWM reductions lake-wide and the measured herbicide concentrations. Little to no EWM has been located within either of the two application areas during 2022 and 2023 post-treatment surveys (Map 3). Many occurrences of EWM that were present in Eagle Lake before treatment and were outside of direct application areas, have not rebounded for the most part as of the year after treatment assessment. The late-summer 2023 EWM mapping survey documented a modest population in Eagle Lake consisting entirely of *single or few plants* or *clumps of plants*.

Quantitative monitoring of this treatment included collection of subsample point-intercept survey data during pretreatment (June 2022) and posttreatment (August 2022) which was reported on within the 2022 reporting. Since the treatment resulted in whole-lake impacts to EWM, the whole lake point-intercept survey data was used to evaluate any potential changes to the aquatic plant community. These data are discussed in Section 6.5 below.



6.0 WHOLE-LAKE POINT-INTERCEPT SURVEYS

Additional aquatic plant monitoring occurred in 2023 through the completion of a whole-lake pointintercept surveys on the six main lakes. The whole-lake point-intercept survey is valuable in assessing the lake-wide aquatic plant population and results are compared to previous or future surveys to monitor aquatic plant populations in the lake. Point-intercept surveys were last completed on Eagle and Flynn in 2020 as part of a whole-lake treatment monitoring. Point-intercept surveys were last completed on Buskey Bay, Millicent, Hart, and Twin Bear in 2018.

The point-intercept method as described in the WDNR publication (WDNR PUB-SS-1068 2010) was used to complete this study. This survey allows for a quantitative analysis of the aquatic plant community in the lake and is directly comparable to past or future surveys completed with the same methodology. Onterra ecologists completed a whole-lake point-intercept survey on the Pike Chain of Lakes on August 8-9, 2023 (Photograph 6.0-1). The results of the 2023 point-intercept survey are highlighted below as well as a comparison of the past surveys that have been completed to date.

Chain-wide Species List

In total, approximately 87 species have been recorded from the Pike Chain Lakes over the course of the aquatic plant surveys that have taken place to date (Table 6.0-1



Photograph 6.0-1. Point-intercept survey on a WI lake. Photo credit Onterra.

and 6.0-2). Species encountered during point-intercept surveys are shown with an "X" on the tables while species that were visually observed on the lakes but were not physically encountered during point-intercept surveys are shown with an "I" on the tables referring to incidental species. The list also contains the species' scientific name, common name, status in Wisconsin, and its coefficient of conservatism. Changes in this list over time, whether it is differences in total species present, gains and losses of individual species, or changes in growth forms that are present, can be an early indicator of changes in the ecosystem. The large number of species present around the Pike Chain Lakes indicate an ecosystem of high species richness.



Table 6	Table 6.0-1. Floating-leaf & Emergent Aquatic plant species located in Pike Chain of Lakes.									
Growth Form	Scientific Nam e	Com m on Nam e	Status in Wisconsin	Coefficient of Conservatism	Buskey Bay	Millicent	Hart	Twin Bear	Eagle	Flynn
	14/ /		NL C	2	×				V	V
	vvater arum	Calla palustris	Native	6	X				Х	X
	Bristiy sedge		Native	7	I			1		-
	Cypress-like sedge	Carex pseudocyperus	Native	7	X				V	
	Unidentified Sedge	Carex sp.	Native	N/A	X	1			X	
	Common tussock sedge	Carex stricta	Native	(I
	Three-way sedge	Dulichium arundinaceum	Native	9				I	X	X
	Creeping spikerush	Eleocharis palustris	Native	/	X	1		х	X	Х
	Water horsetail	Equisetum fluviatile	Native	6	1	1	1		Х	
	Northern blue flag	Iris versicolor	Native	N/A	I	T				1
	Soft rush	Juncus effusus	Native	6	Ι					
ц.	Purple loosestrife	Lythrum salicaria	Non-Native - Invasive	6	Ι	Ι				
gen	Giant reed	Phragmites australis	Non-Native - Invasive	N/A					I	
mer	Pickerelweed	Pontederia cordata	Native	N/A		Т				
ш	Common arrowhead	Sagittaria latifolia	Native	7	х			Ι	Х	Х
	Hardstem bulrush	Schoenoplectus acutus	Native	10	Т	Х	Х	Т	Х	Х
	Three-square rush	Schoenoplectus pungens	Native	9	Х					
	Water bulrush	Schoenoplectus subterminalis	Native	6	Х	Х			Х	Х
	Softstem bulrush	Schoenoplectus tabernaemontani	Native	6	Т	Х			Х	
	Wool grass	Scirpus cyperinus	Native	N/A	Т					
	American bur-reed	Sparganium americanum	Native	8	Х	Х	Т	Т	Х	Х
	Common bur-reed	Sparganium eurycarpum	Native	7		T	Т			
	Narrow-leaved cattail	Typha angustifolia	Non-Native - Invasive	N/A	х					
	Broad-leaved cattail	Typha latifolia	Native	1	Х	Т	1	Т	Х	Х
	Cattail spp.	Typha spp.	Unknown (Sterile)	6	Ι	Х	Х		Х	Х
	Watershield	Brasenia schreberi	Native	6	Х	Х	Ι		Х	Х
	Spatterdock	Nuphar variegata	Native	N/A	х	Х	Х	Х	Х	х
_	White water lily	Nymphaea odorata	Native	6	Х	Х	Х	Х	Х	Х
Ē	Water smartweed	Persicaria amphibia	Native	6	х	T			Х	
	Narrow-leaf bur-reed	Sparganium angustifolium	Native	N/A					1	Х
	Floating-leaf bur-reed	Sparganium fluctuans	Native	0			I			



L I	Short-stemmed bur-reed	Sparganium emersum	Native	6	ХХІІХ
	Water marigold	Bidens beckii	Native	8	x x x x x x
	Coontail		Native	3	
	Spiny Hornwort	Ceratophyllum echipatum	Native	10	
	Muskarasses	Chara sp	Native	7	
	Common waterwood	Elodea canadensis	Native	3	
	Slander waterward	Elodea puttallii	Native	7	
	Water stargrass		Native	6	
			Native	0	
			Native	0	
	Quiliwort spp.	isoetes spp.	Native	8	
	Northern water miltoil		Native	1	
	Eurasian water milfoil	Myriophyllum spicatum	Non-Native - Invasive	N/A	XXXXX
	Dwarf water milfoil	Myriophyllum tenellum	Native	10	XXXXX
	Whorled watermilfoil	Myriophyllum verticillatum	Native	8	Х
	Slender naiad	Najas flexilis	Native	6	X X X X X
	Southern naiad	Najas guadalupensis	Native	7	ХХ
	Stoneworts	Nitella sp.	Native	7	XXXXX
	Large-leaf pondweed	Potamogeton amplifolius	Native	7	x x x x x
	Slender pondweed	Potamogeton berchtoldii	Native	7	X X X X X
	Ribbon-leaf pondweed	Potamogeton epihydrus	Native	8	ххххх
	Leafy pondweed	Potamogeton foliosus	Native	6	x x x x x
	Fries' pondweed	Potamogeton friesii	Native	8	x x
lent	Variable pondweed	Potamogeton gramineus	Native	7	x x x x x
Jerç	Illinois pondweed	Potamogeton illinoensis	Native	6	ххххх
ndu	Floating-leaf pondweed	Potamogeton natans	Native	5	хххіх
S	Oakes pondweed	Potamogeton oakesianus	Native - Special Concern	10	1.1
	White-stem pondweed	Potamogeton praelongus	Native	8	$\mathbf{X} \mathbf{X} \mathbf{X} \mathbf{X} \mathbf{X}$
	Small pondweed	Potamogeton pusillus	Native	7	XXXXX
	, Clasping-leaf pondweed	Potamogeton richardsonii	Native	5	XXXXX
	Fern pondweed	Potamogeton robbinsii	Native	8	x x x x x x
	Spiral-fruited pondweed	Potamogeton spirillus	Native	8	X
	Stiff pondweed	Potamogeton strictifolius	Native	8	XXXXX
	Havnes' pondweed	Potamogeton X havnesii	Native	N/A	XX
	Flat-stem pondweed	Potamogeton zosteriformis	Native	6	XXXXX
	White water-crowfoot	Rapupculus aquatilis	Native	8	
	Creeping spearwort	Ranunculus flammula	Native	9	x
	Arrowhead Sp	Sagittaria sp. (Rosette)	Native	N/A	XXXX
	Sago pondweed	Stuckenia pectinata	Native	3	X X
	Twin-stemmed bladderwort	Utricularia geminiscana	Native	9	
	Creeping bladderwort		Native	9	X X X X
	Elat-leaf bladderwort		Native	9	X X
	Small bladdenuort		Native	10	X X X
	l arge purple bladderwort		Native	9	XXX
	Northoastern bladdenvort		Native Special Concern	0	×
	Common bladdenuort		Native - Opecial Concern	7	
	Wild celery	Vallisneria americana	Native	6	
	Needle spikerush	Eleocharis acicularis	Native	5	X I X X
Ж	Brown-fruited rush	Juncus pelocarpus	Native	8	ххх
	Crested arrowhead	Sagittaria cristata	Native	9	
	Grass-leaved arrowhead	Sagittaria graminea	Native	9	
	Lesser duck weed	Lemna minor	Native	5	ХІ
	Forked duckweed	Lemna trisulca	Native	6	х хх
	Turion duck weed	Lemna turionifera	Native	2	X X
tt.	Slender riccia	Riccia fluitans	Native	7	
	Riccia sp.	Riccia sp.	Native	7	х
	Greater duck weed	Spirodela polyrrhiza	Native	5	XIXX
			NL (1		


Some of the most common native aquatic plant species in the Pike Chain Lakes include: coontail, common waterweed, fern-leaf pondweed, muskgrasses, and wild celery (Photo 6.0-2).



Photograph 6.0-2. Common native aquatic plant species found during the Pike Chain of Lakes. Photo credit Onterra.

Coontail has whorls of leaves which fork into two to three segments, and provides ample surface area for the growth of periphyton and habitat for invertebrates. Unlike most of the submersed plants found in Wisconsin, coontail does not produce true roots and is often found growing entangled amongst other aquatic plants or matted at the surface. Since it lacks true roots, coontail derives most of its nutrients directly from the water (Gross, Erhard and Ivanyi 2003). This ability in combination with a tolerance for low-light conditions allows coontail to become more abundant in eutrophic waterbodies with higher



nutrients and low water clarity. Coontail has the capacity to form dense beds that can float and mat on the water's surface.

Fern-leaf pondweed, as its name suggests, has an arrangement of leaves along the stem give this plant a fern-like appearance. Fern-leaf pondweed typically develops large colonies over soft sediments which grow close to the lake bottom, and it is one of the deepest-growing vascular plants in Wisconsin. Large beds of fern-leaf pondweed provide excellent structural habitat for aquatic wildlife and help to prevent the suspension of the soft bottom sediments in which they grow.

Muskgrasses are a genus of macroalgae, of which there are ten documented species that occur in Wisconsin. Dominance of the aquatic plant community by muskgrasses is common in hardwater lakes and these macroalgae have been found to be more competitive against vascular plants (e.g., pondweeds, milfoils, etc.) in lakes with higher concentrations of calcium carbonate in the sediment (Kufel and Kufel 2002); (Wetzel 2001). Muskgrasses require lakes with good water clarity, and their large beds stabilize bottom sediments. Studies have also shown that muskgrasses sequester phosphorus in the calcium carbonate encrustations which form on these plants, aiding in improving water quality by making the phosphorus unavailable to phytoplankton (Coops 2002). Muskgrasses can be easily identified by their strong skunk-like odor. As well as providing a food source for waterfowl, muskgrasses often serves as a sanctuary for small fish and other aquatic organisms.

Common and slender waterweed is found in waterbodies across Wisconsin, is tolerant of high-nutrient, low-light conditions, and can grow to nuisance levels under ideal conditions. Common waterweed has blade-like leaves in whorls of three produced on long, slender stems. Like other submersed aquatic plants, common waterweed helps to stabilize bottom sediments and provides structural habitat and food for wildlife.

Wild celery produces long, ribbon-like leaves which emerge from a basal rosette, and it prefers to grow over harder substrates and is tolerant of low-light conditions. Its long leaves provide valuable structural habitat for the aquatic community while its network of roots and rhizomes help to stabilize bottom sediments. In mid- to late-summer, wild celery often produces abundant fruit which are important food sources for wildlife including migratory waterfowl.

Frequency of Occurrence

Littoral frequency of occurrence (LFOO) is used to describe how often each species occurred in the points that are within the maximum depth of plant growth (littoral zone), and is displayed as a percentage. The littoral frequency of occurrence of aquatic plants from the 2023 point-intercept surveys from each of the Pike Chain Lakes is detailed below. Additional analysis includes comparisons to past surveys, particularly for lakes in which recent whole-lake herbicide management strategies have occurred (Eagle Lake 2022, Buskey Bay and Lake Millicent – 2023).

Figure 6.0-1 highlights the littoral frequency of occurrence of EWM from the six main lakes of the Pike Chain and demonstrates the population dynamics over the course of a time period of active EWM management. These data indicate that some of the highest occurrences of EWM were recorded during 2016, particularly in Buskey Bay (6.3%), Millicent (13.7%), Hart Lake (10.0%), and Twin Bear Lake (9.4%). The occurrence of EWM in 2023 was 2.5% or below in all lakes, and was 0% in Hart Lake and Eagle Lake.



Iron River Pike Chain Of Lakes Association





Floristic Quality Assessment

The floristic quality of a lake's aquatic plant community is calculated using its native *species richness* and their *average conservatism*. Species richness is the number of native aquatic plant species that were physically encountered on the rake during the point-intercept survey. Average conservatism is calculated by taking the sum of the coefficients of conservatism (C-values) of the native species located and dividing it by species richness. Every plant in Wisconsin has been assigned a coefficient of conservatism, ranging from 1-10, which describes the likelihood of that species being found in an undisturbed environment. Species which are more specialized and require undisturbed habitat are given higher coefficients, while species which are more tolerant of environmental disturbance have lower coefficients. Higher average conservatism values generally indicate a healthier lake as it is able to support a greater number of environment, one that is only able to support disturbance-tolerant species.

On their own, the species richness and average conservatism values for a lake are useful in assessing a lake's plant community; however, the best assessment of the lake's plant community health is determined when the two values are used to calculate the lake's floristic quality. The floristic quality is calculated using the species richness and average conservatism value of the aquatic plant species that were solely encountered on the rake during the point-intercept surveys (equation shown below). This assessment allows the aquatic plant community of a lake to be compared to other lakes within the region and state.

FQI = Average Coefficient of Conservatism *
$$\sqrt{\text{Number of Native Species}}$$

Data collected during the aquatic plant surveys was also used to complete a Floristic Quality Assessment (FQA) which incorporates the number of native aquatic plant species recorded on the rake during the point-intercept survey and their average conservatism. The data used for these calculations does not include any incidental species (visual observations) but only considers plants that were sampled on the rake during the survey.

The native aquatic plant species located on the survey rake during the point-intercept surveys from 2005/07 to 2023 and their conservatism values were used to calculate the FQI for each year. While species richness is well above the median species richness for lakes in the NLF ecoregion and lakes throughout Wisconsin, most lakes in the Pike Chain have shown declining trends over the period from 2005/07-2023 (Figure 6.0-2).

Average species conservatism in the Pike Chain of Lakes has remained relatively consistent with little variability over the period from 2005/07-2023 (Figure 6.0-3). In most years, the average conservatism value has fallen near the NLF ecoregion median for each lake. Buskey Bay and Twin Bear Lake have averaged slightly lower values than other lakes in the Chain with typical values near or below the ecoregion median.

Using the species richness and average conservatism to calculate the Floristic Quality Index for the Pike Chain of Lakes reveals exceptionally high values for all lakes (Figure 6.0-4). Given the decline in chainwide species richness, chain-wide Floristic Quality Index values have also declined. The largest declines in floristic quality have occurred in Buskey Bay, Lake Millicent, and Flynn Lake.









aquatic plant species physically encountered on the rake during the point-intercept survey and does not include incidentally-located species.



Species Diversity

Species diversity is often confused with species richness. Species richness is simply the number of species found within a given community. While species diversity utilizes species richness, it also takes into account evenness or the variation in abundance of the individual species within the community. For example, a lake with 10 aquatic plant species that had relatively similar abundances within the community would be more diverse than another lake with 10 aquatic plant species were 50% of the community was comprised of just one or two species.

An aquatic system with high species diversity is more stable than a system with a low diversity. This is analogous to a diverse financial portfolio in that a diverse aquatic plant community can withstand environmental fluctuations much like a diverse portfolio can handle economic fluctuations. Some managers believe a lake with a diverse plant community is also better suited to compete against exotic infestations than a lake with a lower diversity. However, in a recent study of 1,100 Minnesota lakes, researchers concluded that more diverse communities were not necessarily more resistant or resilient to invaders (Muthukrishnan et al. 2018).

If a lake has a diversity index value of 0.90, it means that if two plants were randomly sampled from the lake there is a 90% probability that the two individuals would be of a different species. The Simpson's Diversity Index value is compared to data collected by Onterra and the WDNR Science Services on lakes within the Northern Lakes and Forests ecoregion and on lakes throughout Wisconsin. While a method for characterizing diversity values of fair, poor, etc. does not exist, lakes within the same ecoregion may be compared to provide an idea of how its diversity values rank.



The average Simpson's diversity index value declined from 0.94 in the 2005/2007 surveys to 0.91 in 2018 and averaged 0.90 in the 2023 surveys which falls at the 75th percentile for lakes in the NLF ecoregion (Figure 6.0-5).









6.1 Buskey Bay

A total of 30 species were physically encountered on the survey rake during the 2023 survey with coontail, fern-leaf pondweed, and common waterweed being the most-frequently encountered native aquatic plant species. The maximum depth of plant growth in the 2023 survey was 25 feet which has been about the average in the past.

Eurasian watermilfoil was found at six of the sampling locations during the 2023 point-intercept survey resulting in a littoral frequency of occurrence of 2.0%.



Table 6.1-1 displays the littoral frequency of occurrence of aquatic plants from all point-intercept surveys that have taken place on Buskey Bay. The 2023 survey indicates statistically valid decreases compared to the last survey (2018) for fern-leaf pondweed and white-stem pondweed. Five native species exhibited statistically valid increases in occurrence between 2018-2023 including northern watermilfoil and four native pondweed species. Many other native species did not show statistically valid changes in occurrence between the two surveys.

Since the 2023 herbicide treatment resulted in lake-wide impacts to EWM, susceptible native plants may have also been impacted. Northern watermilfoil is highly impacted by ProcellaCOR treatments and this species was present in the 2023 survey at 3.4%. Coontail is susceptible to ProcellaCOR treatments, often



decreasing by around 50% during the year of treatment. The 2023 survey indicated an occurrence of 32.1% for coontail, which is the lowest of any past survey on the lake. Coontail remains prevalent within Buskey Bay and was the most frequently encountered species in the 2023 survey. Other species that are present in Buskey Bay and believed to be susceptible to ProcellaCOR include water marigold and water stargrass, both which were present in the lake in the 2023 survey and have typically been present at relatively low frequencies of occurrence in the past.

		LFOO (%)				2018-2023		
Scientific Name	Com m on Nam e	2013	2016	2017	2018	2023	% Change	Direction
Ceratophyllum demersum	Coontail	37.6	38.5	41.9	39.1	32.1	-18.0	
Potamogeton robbinsii	Fern-leaf pondw eed	37.3	40.5	45.0	35.9	23.2	-35.4	▼
Elodea canadensis	Common waterweed	42.1	43.1	40.6	18.5	21.8	18.0	
Chara spp.	Muskgrasses	21.2	22.7	25.8	22.1	16.4	-25.8	V
Vallisneria americana	Wild celery	13.9	17.1	17.8	15.3	14.0	-8.6	
Potamogeton pusillus & P. berchtoldi	i Small & Slender pondw eed	10.6	13.2	17.1	10.7	9.2	-13.7	
Potamogeton amplifolius	Large-leaf pondw eed	15.2	10.5	14.4	1.8	8.5	379.5	▲
Potamogeton gramineus	Variable-leaf pondw eed	9.1	10.9	14.8	4.6	9.9	113.9	A
Potamogeton pusillus	Small pondw eed	5.5	8.6	9.1	10.7	9.2	-13.7	
Potamogeton zosteriformis	Flat-stem pondw eed	19.1	10.5	5.4	5.0	3.1	-38.3	
Potamogeton praelongus	White-stem pondw eed	7.0	11.8	6.7	10.0	3.4	-65.7	▼
Najas flexilis	Slender naiad	5.2	5.6	4.0	9.6	8.9	-7.6	V
Myriophyllum sibiricum	Northern w atermilfoil	11.5	10.2	4.0	0.4	3.4	859.0	
Heteranthera dubia	Water stargrass	2.7	7.2	2.7	2.8	4.4	55.8	
Potamogeton berchtoldii	Slender pondw eed	5.2	4.6	8.4	0.0	0.0		-
Nymphaea odorata	White water lily	4.2	3.0	3.4	1.1	3.1	187.7	
Potamogeton strictifolius	Stiff pondw eed	9.1	3.6	1.7	0.0	0.3		
Myriophyllum spicatum	Eurasian w atermilfoil	0.6	6.3	2.3	1.4	2.0	43.9	
Potamogeton illinoensis	Illinois pondw eed	2.4	2.0	1.7	0.0	2.7		
Potamogeton foliosus	Leafy pondw eed	1.8	3.6	2.7	1.4	1.0	-28.1	V
Bidens beckii	Water marigold	3.0	3.0	0.3	0.4	1.4	283.6	
Ranunculus aquatilis	White water crow foot	3.9	2.3	1.3	0.0	0.3		
Potamogeton friesii	Fries' pondw eed	0.0	0.3	0.0	0.0	3.4		
Nitella spp.	Stonew orts	4.8	0.3	0.3	0.0	0.0		-
Eleocharis acicularis	Needle spikerush	0.6	1.6	2.0	1.1	0.3	-68.0	
Potamogeton richardsonii	Clasping-leaf pondw eed	0.3	0.7	2.0	0.0	0.7		
Utricularia gibba	Creeping bladderw ort	0.6	0.7	1.0	1.1	0.0	-100.0	
Potamogeton X haynesii	Haynes' pondw eed	0.0	0.0	3.4	0.0	0.0		-
Potamogeton natans	Floating-leaf pondw eed	0.9	0.3	0.3	0.4	0.7	91.8	
Utricularia vulgaris	Common bladderw ort	0.9	0.7	1.0	0.0	0.0		-
Persicaria amphibia	Water smartw eed	0.6	0.7	0.3	0.4	0.3	-4.1	
Nuphar variegata	Spatterdock	0.6	0.7	0.3	0.4	0.3	-4.1	V
Stuckenia pectinata	Sago pondw eed	0.6	0.7	0.0	0.4	0.3	-4.1	
Fissidens spp. & Fontinalis spp.	Aquatic Moss	0.0	0.3	1.3	0.4	0.0	-100.0	
Isoetes spp.	Quillw ort spp.	0.0	0.3	0.0	0.0	0.7		
Mvriophyllum tenellum	Dw arf w atermilfoil	0.0	0.7	0.3	0.0	0.0		-
Lemna trisulca	Forked duckw eed	0.0	0.0	0.0	0.4	0.3	-4.1	
Schoenoplectus acutus	Hardstem bulrush	0.0	0.0	0.0	0.0	0.3		
Brasenia schreberi	Watershield	0.0	0.0	0.3	0.4	0.0	-100.0	
Spirodela polyrhiza	Greater duckw eed	0.0	0.0	0.0	0.4	0.0	-100.0	V
Sparganium americanum	American bur-reed	0.3	0.0	0.0	0.0	0.0		-
Riccia sp.	Riccia sp.	0.3	0.0	0.0	0.0	0.0		-
Potamogeton epihvdrus	Ribbon-leaf pondw eed	0.3	0.0	0.0	0.0	0.0		-
Myriophyllum verticillatum	Whorled w atermilfoil	0.3	0.0	0.0	0.0	0.0		-
Lemna turionifera	Turion duckw eed	0.0	0.0	0.3	0.0	0.0		-
Ceratophyllum echinatum	Spiny hornwort	0.0	0.3	0.0	0.0	0.0	1	-
						2 C		

 Table 6.1-1. Littoral Frequency of Occurrence of Aquatic Plants from Point-Intercept Surveys in Buskey

 Bay. Arrows indicate statistically valid change in occurrence between 2023 survey and previous survey.

6.2 Lake Millicent

A total of 25 species were physically encountered on the survey rake during the 2023 survey with muskgrasses, wild celery, and common waterweed being the most-frequently encountered native aquatic plant species. The maximum depth of plant growth in the 2023 survey was 25 feet which has been about the average in the past.

Eurasian watermilfoil was found at five of the sampling locations during the 2023 point-intercept survey resulting in a littoral frequency of occurrence of 2.5%.



Figure 6.2-1. 2023 Littoral Frequency of Occurrence of aquatic plant species in Lake Millicent.

Table 6.2-1 displays the littoral frequency of occurrence of aquatic plants from all point-intercept surveys that have taken place on Lake Millicent. Three native species exhibited statistically valid increases in occurrence between 2018-2023 including slender naiad, slender pondweed, and water stargrass. Many other native species did not show statistically valid changes in occurrence between the two surveys.

Since the 2023 herbicide treatment resulted in lake-wide impacts to EWM, susceptible native plants may have also been impacted. Northern watermilfoil was present in the 2023 survey at 0.5% and was not sampled in the previous two point-intercept surveys from 2017-2018. The occurrence of coontail in 2023 was similar to past surveys.



Table 6.2-1. Littoral Frequency of Occurrence of Aquatic Plants from Point-Intercept Surveys in Lake Millicent. Arrows indicate statistically valid change in occurrence between 2023 survey and previous survey.

		LFOO (%)					2018	-2023
Scientific Name	Common Name	2013	2016	2017	2018	2023	% Change	Direction
Chara spp.	Muskgrasses	26.2	21.7	29.2	26.5	32.5	22.6	
Potamogeton gramineus	Variable-leaf pondw eed	33.0	20.8	24.7	15.3	13.0	-14.8	
Elodea canadensis	Common waterweed	19.5	16.8	20.6	11.2	16.5	46.7	
Vallisneria americana	Wild celery	11.8	15.5	18.1	14.1	19.0	35.2	
Potamogeton pusillus & P. Berchtoldi	Small & Slender pondw eed	15.4	12.8	14.8	10.8	15.5	42.9	
Potamogeton pusillus	Small pondw eed	15.4	12.8	5.3	10.8	10.5	-3.2	V
Eleocharis acicularis	Needle spikerush	9.0	9.7	7.4	8.0	9.0	12.1	
Najas flexilis	Slender naiad	6.3	5.3	6.2	6.0	14.5	140.7	
Ceratophyllum demersum	Coontail	9.0	9.3	9.5	6.0	8.5	41.1	
Potamogeton robbinsii	Fern-leaf pondw eed	6.3	6.2	6.2	4.0	7.5	86.8	
Myriophyllum tenellum	Dw arf w atermilfoil	3.6	4.9	5.8	1.6	2.0	24.5	
Potamogeton berchtoldii	Slender pondw eed	0.0	0.0	9.5	0.0	5.0		
Nitella spp.	Stonew orts	0.0	3.5	8.6	1.6	2.5	55.6	
Myriophyllum spicatum	Eurasian w atermilfoil	0.9	13.7	0.0	0.0	2.5		A
Myriophyllum sibiricum	Northern w atermilfoil	7.2	8.4	0.0	0.0	0.5		
Heteranthera dubia	Water stargrass	5.4	5.3	0.0	0.0	3.0		A
Ranunculus aquatilis	White water crow foot	5.0	7.1	0.8	0.0	1.5		
Potamogeton illinoensis	Illinois pondw eed	1.4	7.1	0.8	0.0	1.0		
Potamogeton richardsonii	Clasping-leaf pondw eed	3.2	0.0	0.8	1.6	2.5	55.6	
Potamogeton amplifolius	Large-leaf pondw eed	1.8	1.3	3.3	1.6	1.0	-37.8	
Potamogeton praelongus	White-stem pondw eed	0.0	2.7	0.4	1.2	1.5	24.5	
Potamogeton foliosus	Leafy pondw eed	0.0	4.0	0.4	1.6	0.0	-100.0	
Potamogeton strictifolius	Stiff pondw eed	3.2	1.8	0.0	0.0	0.0		-
Nuphar variegata	Spatterdock	1.8	0.0	0.4	0.0	1.5		
Fissidens spp. & Fontinalis spp.	Aquatic Moss	2.7	0.0	0.0	0.4	1.0	149.0	
Nymphaea odorata	White w ater lily	0.0	0.4	1.6	0.8	0.5	-37.8	
Bidens beckii	Water marigold	1.4	1.8	0.0	0.0	0.5		
Utricularia vulgaris	Common bladderw ort	0.9	0.0	1.6	0.4	0.0	-100.0	
Schoenoplectus subterminalis	Water bulrush	0.9	0.4	1.2	0.4	0.0	-100.0	$\overline{\mathbf{v}}$
Potamogeton zosteriformis	Flat-stem pondw eed	0.9	0.9	0.0	0.0	0.5		
Ceratophyllum echinatum	Spiny hornw ort	0.0	2.2	0.4	0.0	0.0		-
Utricularia intermedia	Flat-leaf bladderw ort	0.5	0.4	0.8	0.4	0.0	-100.0	
Potamogeton alpinus	Alpine pondw eed	1.8	0.0	0.0	0.0	0.0		-
Brasenia schreberi	Watershield	0.9	0.0	0.4	0.4	0.0	-100.0	
Utricularia gibba	Creeping bladderw ort	0.9	0.4	0.0	0.0	0.0		-
Schoenoplectus acutus	Hardstem bulrush	0.5	0.0	0.8	0.0	0.0		-
Utricularia minor	Small bladderw ort	0.0	0.0	0.8	0.0	0.0		-
Schoenoplectus tabernaemontani	Softstem bulrush	0.0	0.9	0.0	0.0	0.0		-
Potamogeton natans	Floating-leaf pondw eed	0.5	0.0	0.4	0.0	0.0		-
Najas guadalupensis	Southern naiad	0.0	0.0	0.0	0.0	0.5		
Ranunculus flammula	Creeping spearw ort	0.5	0.0	0.0	0.0	0.0		-
Potamogeton epihydrus	Ribbon-leaf pondw eed	0.5	0.0	0.0	0.0	0.0		-
Juncus pelocarpus	Brow n-fruited rush	0.5	0.0	0.0	0.0	0.0		-



6.3 Hart Lake

A total of 28 species were physically encountered on the survey rake during the 2023 survey with muskgrasses, fern-leaf pondweed, and slender naiad being the most frequently encountered native aquatic plant species. The maximum depth of plant growth in the 2023 survey was 27 feet which is consistent with the last few surveys on the lake.

Eurasian watermilfoil was not encountered at any of the sampling locations during the 2023 point-intercept survey (0% occurrence).



Figure 6.3-1. 2023 Littoral Frequency of Occurrence of aquatic plant species in Hart Lake.

Table 6.3-1 displays the littoral frequency of occurrence of aquatic plants from all point-intercept surveys that have taken place on Hart Lake. The 2023 survey indicates five native species showed statistically valid decreases compared to the last survey (2018) including muskgrasses, variable-leaf pondweed, stoneworts, coontail, and leafy pondweed. Slender naiad as well as the combined occurrences of slender and small pondweed showed statistically valid increased in occurrence from 2018-2023. Many other native species did not show statistically valid changes in occurrence between the two surveys.



Table 6.3-1. Littoral Frequency of Occurrence of Aquatic Plants from Point-Intercept Surveys in Hart Lake. Arrows indicate statistically valid change in occurrence between 2023 survey and previous survey.

		LFOO (%)					2018	-2023
Scientific Name	Common Name	2013	2016	2017	2018	2023	% Change	Direction
Chara spp.	Muskgrasses	27.9	28.4	38.3	36.3	25.9	-28.6	•
Potamogeton robbinsii	Fern-leaf pondw eed	10.0	11.1	15.9	13.7	17.1	24.9	A
Potamogeton gramineus	Variable-leaf pondweed	14.2	15.0	20.2	10.7	6.5	-39.6	▼
Elodea canadensis	Common w aterw eed	13.3	15.9	12.2	7.9	9.9	25.5	A
Najas flexilis	Slender naiad	8.9	12.3	4.1	8.8	15.4	74.6	
Potamogeton berchtoldii & P. pusillus	Slender & Small pondw eed	14.4	11.2	4.9	4.9	10.1	106.6	A
Potamogeton pusillus	Small pondw eed	14.0	6.6	4.1	4.9	6.7	36.4	A
Nitella spp.	Stonew orts	5.1	8.4	11.4	7.3	1.1	-84.4	•
Ceratophyllum demersum	Coontail	5.9	2.7	6.7	5.3	1.7	-67.4	•
Vallisneria americana	Wild celery	0.0	3.6	5.3	4.9	4.2	-14.3	•
Potamogeton amplifolius	Large-leaf pondw eed	4.2	3.0	3.5	1.9	1.7	-8.8	
Myriophyllum spicatum	Eurasian w atermilfoil	0.9	10.0	0.0	1.9	0.0	-100.0	•
Eleocharis acicularis	Needle spikerush	1.1	1.7	2.8	2.4	3.0	24.7	A
Potamogeton berchtoldii	Slender pondw eed	0.3	4.7	0.8	0.0	3.4		
Potamogeton praelongus	White-stem pondw eed	2.3	2.7	2.6	1.7	1.5	-9.9	
Nuphar variegata	Spatterdock	2.2	1.2	1.2	0.8	1.5	102.7	A
Potamogeton strictifolius	Stiff pondw eed	1.1	5.0	0.0	0.0	0.2		A
Myriophyllum sibiricum	Northern w atermilfoil	1.6	3.9	0.2	0.0	0.4		A
Myriophyllum tenellum	Dw arf w atermilfoil	0.6	1.2	1.0	1.1	1.3	18.2	
Potamogeton zosteriformis	Flat-stem pondw eed	1.4	1.7	0.6	0.2	1.0	406.7	A
Heteranthera dubia	Water stargrass	1.6	1.2	0.4	0.4	0.6	52.0	
Ceratophyllum echinatum	Spiny hornw ort	0.9	1.9	1.6	0.0	0.2		
Nymphaea odorata	White w ater lily	1.4	0.5	0.8	1.1	0.4	-66.2	
Potamogeton richardsonii	Clasping-leaf pondw eed	0.8	0.6	0.2	0.8	1.0	26.7	A
Utricularia vulgaris	Common bladderw ort	2.5	0.2	0.8	0.2	0.0	-100.0	
Potamogeton foliosus	Leafy pondw eed	0.2	1.4	0.4	0.9	0.0	-100.0	▼
Potamogeton illinoensis	Illinois pondw eed	0.5	1.4	0.2	0.0	0.2		A
Schoenoplectus acutus	Hardstem bulrush	0.6	0.3	0.6	0.4	0.2	-49.3	•
Brasenia schreberi	Watershield	0.6	0.8	0.2	0.2	0.2	1.3	
Bidens beckii	Water marigold	0.6	0.6	0.2	0.0	0.0		-
Potamogeton epihydrus	Ribbon-leaf pondw eed	0.6	0.2	0.0	0.0	0.2		
Isoetes spp.	Quillw ort spp.	0.3	0.6	0.0	0.0	0.0		-
Fissidens spp. & Fontinalis spp.	Aquatic Moss	0.2	0.0	0.0	0.0	0.4	1	
Sagittaria sp. (rosette)	Arrow head sp. (rosette)	0.0	0.6	0.0	0.0	0.0		-
Ranunculus aquatilis	White water crow foot	0.2	0.2	0.0	0.0	0.2		
Utricularia gibba	Creeping bladderw ort	0.3	0.2	0.0	0.0	0.0		-
Potamogeton natans	Floating-leaf pondw eed	0.2	0.0	0.2	0.2	0.0	-100.0	
Juncus pelocarpus	Brow n-fruited rush	0.0	0.2	0.0	0.0	0.2		
Utricularia resupinata	Northeastern bladderw ort	0.3	0.0	0.0	0.0	0.0		-
Utricularia purpurea	Large purple bladderw ort	0.0	0.0	0.0	0.2	0.0	-100.0	•
Utricularia minor	Small bladderw ort	0.2	0.0	0.0	0.0	0.0		-
Typha spp.	Cattail spp.	0.2	0.0	0.0	0.0	0.0		-
Spirodela polyrhiza	Greater duckw eed	0.0	0.0	0.2	0.0	0.0		-
Ranunculus flammula	Creeping spearw ort	0.2	0.0	0.0	0.0	0.0		-

6.4 Twin Bear Lake

A total of 27 species were physically encountered on the survey rake during the 2023 survey with coontail, fern-leaf pondweed, and common waterweed being the most-frequently encountered native aquatic plant species. The maximum depth of plant growth in the 2023 survey was 19 feet which is slightly shallower than past surveys where plants have been documented between 22-28 feet.

Eurasian watermilfoil was found at two of the sampling locations during the 2023 point-intercept survey resulting in a littoral frequency of occurrence of 0.9%.



Figure 6.4-1. 2023 Littoral Frequency of Occurrence of aquatic plant species in Twin Bear Lake.

Table 6.4-1 displays the littoral frequency of occurrence of aquatic plants from all point-intercept surveys that have taken place on Twin Bear Lake. Six native species exhibited statistically valid increases in occurrence from 2018-2023 including common waterweed, small pondweed, northern watermilfoil, wild celery, flat-stem pondweed, and water marigold. No species showed a statistically valid decrease in occurrence comparing 2018 to 2023.



Table 6.4-1. Littoral Frequency of Occurrence of Aquatic Plants from Point-Intercept Surveys in Twin Bear Lake. Arrows indicate statistically valid change in occurrence between 2023 survey and previous survey.

		LFOO (%)					2018	-2023
Scientific Name	Common Name	2013	2016	2017	2018	2023	% Change	Direction
Potamogeton robbinsii	Fern-leaf pondw eed	21.0	32.0	33.8	26.7	24.8	-7.2	
Ceratophyllum demersum	Coontail	23.3	22.3	25.1	23.0	26.1	13.6	A
Elodea canadensis	Common w aterw eed	29.8	29.3	38.1	8.8	16.4	86.4	A
Chara spp.	Muskgrasses	15.9	18.4	22.1	11.1	15.0	34.9	A
Potamogeton berchtoldii & P. pusillus	Slender & Small pondw eed	15.1	19.1	23.1	7.4	11.1	48.8	
Najas flexilis	Slender naiad	9.9	13.3	11.7	11.8	14.6	23.5	A
Potamogeton gramineus	Variable-leaf pondw eed	11.4	14.5	15.7	6.4	9.3	44.8	A
Potamogeton berchtoldii	Slender pondw eed	10.2	0.0	15.7	7.4	4.9	-34.5	
Potamogeton pusillus	Small pondw eed	4.8	19.1	7.4	0.0	6.2		
Myriophyllum sibiricum	Northern w atermilfoil	6.5	12.1	4.0	0.7	3.1	358.4	
Vallisneria americana	Wild celery	1.7	3.1	4.0	3.0	7.1	132.8	
Potamogeton richardsonii	Clasping-leaf pondw eed	1.4	1.6	3.7	4.4	6.6	51.1	A
Heteranthera dubia	Water stargrass	5.4	1.6	4.3	3.0	4.0	31.0	
Potamogeton zosteriformis	Flat-stem pondw eed	5.4	2.7	2.7	1.0	4.0	292.9	
Potamogeton amplifolius	Large-leaf pondw eed	3.7	2.7	4.0	1.0	2.7	161.9	
Bidens beckii	Water marigold	1.1	4.7	0.3	0.7	4.9	620.4	
Myriophyllum spicatum	Eurasian w atermilfoil	0.3	9.4	2.0	0.3	0.9	161.9	
Potamogeton praelongus	White-stem pondw eed	0.3	2.3	3.3	2.0	2.7	31.0	A
Potamogeton strictifolius	Stiff pondw eed	1.4	4.7	1.0	0.7	0.0	-100.0	
Lemna trisulca	Forked duckw eed	0.0	0.8	0.7	0.7	2.7	292.9	A
Eleocharis acicularis	Needle spikerush	0.3	0.8	2.3	1.0	0.0	-100.0	V
Ranunculus aquatilis	White water crow foot	0.9	1.6	1.7	0.0	0.0		-
Nymphaea odorata	White w ater lily	0.6	0.0	0.7	0.7	0.9	31.0	A
Potamogeton illinoensis	Illinois pondw eed	0.3	1.2	0.3	0.0	0.4		A
Potamogeton epihydrus	Ribbon-leaf pondw eed	1.1	0.4	0.0	0.0	0.4		A
Nuphar variegata	Spatterdock	0.9	0.8	0.7	0.0	0.0		-
Nitella spp.	Stonew orts	0.0	0.0	0.7	0.7	0.4	-34.5	V
Myriophyllum tenellum	Dw arf w atermilfoil	0.0	0.4	0.7	0.3	0.4	31.0	A
Potamogeton X haynesii	Haynes' pondw eed	0.0	0.0	1.0	0.0	0.0		-
Potamogeton foliosus	Leafy pondw eed	0.0	0.0	1.0	0.0	0.0		-
Isoetes spp.	Quillw ort spp.	0.0	0.0	0.3	0.0	0.4		
Ceratophyllum echinatum	Spiny hornw ort	0.6	0.0	0.3	0.0	0.0		-
Spirodela polyrhiza	Greater duckw eed	0.0	0.0	0.0	0.0	0.4		
Najas guadalupensis	Southern naiad	0.0	0.0	0.0	0.0	0.4		A
Elodea nuttallii	Slender waterweed	0.0	0.0	0.0	0.0	0.4		
Schoenoplectus acutus	Hardstem bulrush	0.3	0.0	0.0	0.0	0.0		-
Fissidens spp. & Fontinalis spp.	Aquatic Moss	0.0	0.4	0.0	0.0	0.0		-
Eleocharis palustris	Creeping spikerush	0.3	0.0	0.0	0.0	0.0		-



6.5 Eagle Lake

A total of 35 species were physically encountered on the survey rake during the 2023 survey with fernleaf pondweed, wild celery, and common waterweed being the most-frequently encountered native aquatic plant species. The maximum depth of plant growth in the 2023 survey was 39 feet which is significantly deeper than past surveys and deeper than plants have been found in other lakes in the system. Species located at depths of 36-39 feet in the 2023 survey included coontail, small pondweed, and common waterweed.

Eurasian watermilfoil was not present at any of the sampling locations during the 2023 point-intercept survey (0% occurrence).



Table 6.5-1 displays the littoral frequency of occurrence of aquatic plants from all point-intercept surveys that have taken place on Eagle Lake. Slender and small pondweed are lumped for analysis purposes. The 2023 survey indicates nine native species exhibited statistically valid decreases in occurrence compared to the last survey conducted in 2020. The fact that the maximum depth of plant growth in the 2023 survey was much deeper than past surveys in Eagle Lake attributes to some of the statistically valid changes due to there being a larger denominator in the statistical analysis. For example, in the 2023



survey 668 sampling points were below the maximum depth of plant growth (39') compared to 530 sampling points being below the maximum depth of plant growth (25') in the 2020 survey. Despite the difference in littoral areas in recent surveys, a reduction in vegetation between 2020-2023 is evident by simply comparing the number of sampling points where specific species were present in each survey. For example, common waterweed was present at 211 points in 2020 compared to 126 in 2023 and muskgrasses were present at 150 points in 2020 and 104 in 2023.

Table 6.5-1.Littoral Frequency of Occurrence of Aquatic Plants from Point-Intercept Surveys inEagle Lake.Arrows indicate statistically valid change in occurrence between 2023 survey and previous survey.

		LFOO (%)						2020-	2023
Scientific Name	Common Name	2013	2016	2017	2018	2020	2023	% Change	Direction
Potamogeton robbinsii	Fern-leaf pondw eed	42.3	44.6	48.6	41.4	35.8	33.5	-6.5	V
Elodea canadensis	Common w aterw eed	35.7	27.6	30.4	25.0	39.8	18.9	-52.6	•
Vallisneria americana	Wild celery	17.9	22.5	26.5	23.5	35.7	25.9	-27.4	▼
Ceratophyllum demersum	Coontail	28.9	21.5	27.4	16.9	28.3	15.6	-45.0	▼
Potamogeton gramineus	Variable-leaf pondw eed	26.5	11.4	21.9	10.9	27.2	6.0	-78.0	▼
Potamogeton berchtoldii & P. pusillus	Slender & Small pondw eed	13.3	8.3	14.4	9.2	13.6	10.8	-20.7	
Chara spp.	Muskgrasses	13.2	9.9	13.2	7.7	15.1	7.8	-48.4	▼
Najas flexilis	Slender naiad	10.2	9.2	8.0	8.9	10.0	9.3	-7.2	
Potamogeton pusillus	Small pondw eed	13.3	8.3	13.0	2.3	6.6	9.9	49.6	A
Nymphaea odorata	White w ater lily	9.2	4.6	6.0	6.4	6.6	6.4	-2.5	
Potamogeton richardsonii	Clasping-leaf pondw eed	9.9	5.0	4.4	3.9	5.5	5.8	6.7	
Potamogeton amplifolius	Large-leaf pondw eed	6.6	4.4	4.6	3.0	7.0	5.1	-27.1	
Potamogeton zosteriformis	Flat-stem pondw eed	13.0	1.5	1.2	1.5	3.8	4.0	7.1	
Heteranthera dubia	Water stargrass	2.8	4.2	2.7	4.4	2.6	3.0	13.3	
Brasenia schreberi	Watershield	8.3	1.1	3.0	2.0	2.1	2.7	29.8	
Potamogeton praelongus	White-stem pondw eed	5.7	2.0	2.7	2.5	8.3	0.6	-92.8	•
Myriophyllum sibiricum	Northern w atermilfoil	6.8	4.8	3.9	2.8	0.0	0.0		-
Potamogeton berchtoldii	Slender pondw eed	0.2	0.0	1.4	6.9	7.0	1.0	-85.0	•
Eleocharis acicularis	Needle spikerush	1.9	3.1	3.2	2.8	3.0	1.3	-55.4	▼
Potamogeton strictifolius	Stiff pondw eed	2.3	3.7	1.1	1.8	6.2	0.4	-92.8	•
Potamogeton natans	Floating-leaf pondw eed	5.0	0.7	3.0	0.8	1.1	1.9	71.9	
Myriophyllum tenellum	Dw arf w atermilfoil	1.6	2.0	3.2	3.7	3.0	0.6	-80.2	•
Bidens beckii	Water marigold	2.6	4.1	2.5	2.3	1.3	0.9	-32.0	$\overline{\mathbf{v}}$
Utricularia vulgaris	Common bladderw ort	2.9	0.2	1.6	1.3	0.2	0.4	138.0	
Myriophyllum spicatum	Eurasian w atermilfoil	0.0	0.9	0.5	4.5	0.8	0.0	-100.0	▼
Ceratophyllum echinatum	Spiny hornw ort	0.0	2.9	0.0	3.0	0.2	0.0	-100.0	•
Nuphar variegata	Spatterdock	0.7	0.4	0.4	0.5	0.8	0.9	19.0	
Utricularia gibba	Creeping bladderw ort	1.0	0.0	0.2	0.0	0.0	0.0		-
Ranunculus aquatilis	White water crow foot	0.0	0.0	0.0	0.2	0.2	0.1	-20.7	$\overline{\mathbf{v}}$
Utricularia minor	Small bladderw ort	0.3	0.0	0.0	0.0	0.2	0.0	-100.0	$\mathbf{\nabla}$
Ranunculus flammula	Creeping spearw ort	0.3	0.0	0.0	0.2	0.0	0.0		-

Figure 6.5-2 investigates the average number of native plant species at each littoral point-intercept sampling location. These data show variable values between 2013-2020. The 2023 survey indicated 1.65 native species per littoral sampling site which is a decrease by one species per sampling point since the previous survey in 2020.





6.6 Flynn Lake

A total of 28 species were physically encountered on the survey rake during the 2023 survey with fernleaf pondweed, wild celery, and variable-leaf pondweed being the most frequently encountered native aquatic plant species (Figure 6.6-1). The entire lake is considered littoral with plants growing throughout all depths of the lake.

Eurasian watermilfoil was found at one of the sampling locations during the 2023 point-intercept survey resulting in a littoral frequency of occurrence of 0.9%.



Figure 6.6-1. 2023 Littoral Frequency of Occurrence of aquatic plant species in Flynn Lake.

Table 6.6-1 displays the littoral frequency of occurrence of aquatic plants from all point-intercept surveys that have taken place on Flynn Lake. The 2023 survey indicates statistically valid decreases compared to the last survey (2020) for variable leaf pondweed, large-leaf pondweed, and stiff pondweed. Statistically valid increases in occurrence between 2020-2023 included water stargrass and arrowhead. When combined for analysis purposes due to similar morphology, the change in occurrence between 2020-2023 for slender & small pondweeds is not statistically valid.



Table 6.6-1. Littoral Frequency of Occurrence of Aquatic Plants from Point-Intercept Surveys in Flynn Lake. Arrows indicate statistically valid change in occurrence between 2023 survey and previous survey.

		LFOO (%)						2020	-2023
Scientific Name	Common Name	2013	2016	2017	2018	2020	2023	% Change	Direction
Potamogeton robbinsii	Fern-leaf pondw eed	52.1	70.8	63.7	58.3	54.5	46.9	-13 9	
Vallisperia americana	Wild celery	26.1	44.3	45.2	45.0	47.3	44.2	-6.5	
Potamogeton gramineus	Variable-leaf pondweed	40.3	19.8	37.1	30.8	40.2	26.5	-33.9	T
Potamogeton amplifolius	Large-leaf pondw eed	32.8	25.5	29.8	23.3	20.5	7 1	-65.5	, T
Flodea canadensis	Common waterweed	24.4	18.9	21.8	20.8	17.0	22.1	30.4	
Najas flevilis	Slender najad	23.5	20.8	16.1	17.5	15.2	20.4	34.1	
Nymphaea odorata	White water lilv	23.5	12.3	25.0	10.8	13.4	15.0	12.3	
Potamogeton berchtoldii & P. pusillus	Slender & Small pondweed	11.8	19.8	16.9	16.7	11.4	17.7	52.5	
Chara son	Muskarasses	17.6	12.3	18.5	14.2	10.7	14.2	32.0	
Potamogeton X havnesii & Potamoget	Havnes' pondweed & Flat-stem	8.4	12.0	14.5	11.2	63	62	-0.9	-
Potamogeton praelongus	White-stem pondw eed	7.6	8.5	16.1	67	8.9	6.2	-30.6	· ·
Brasenia schreberi	Watershield	21.8	7.5	13.7	5.8	3.6	3.5	-0.9	· ·
Potemogeton pusillus	Small pondw eed	11.8	10.8	0.0	12.5	11.6	1.8	-84.8	· ·
Potamogeton berchtoldii	Slander pondw eed	0.0	13.0	16.0	12.5	0.0	15.0	-04.0	•
Hotoropthoro dubio	Water stargrass	6.7	5.7	10.5	14.2	0.0	6.2	502.9	-
Potomogoton zostoriformis	Flat stom pondw ood	0.7	12.2	10.5	7.5	0.9	5.2	19.0	
Potamogeton patans	Flat-stell politik eeu	12.4	7.5	4.0	1.5	4.5	0.0	67.0	-
	Noodle opikerush	13.4	1.5	10.9	4.2	Z.1 A E	0.9	-07.0	· ·
	Common bladdorw ort	6.7	0.0	5.2	4.2	4.5	1.0	-00.4	×
		0.7	0.9	0.5	3.3	0.0	0.9		A
Sagittaria sp. (rosette)	Arrow flead sp. (roselle)	0.0	1.9	4.0	0.7	0.0	3.5	50.4	-
Potamogeton X naynesii	Haynes' pondwieed	0.0	0.0	11.3	4.2	1.8	0.9	-50.4	V
Schoenopiectus subterminalis	vvater buirusn	4.2	1.9	0.5	5.0	0.0	0.0	407.0	-
Bidens beckii	Water marigold	0.0	0.0	4.0	5.0	0.9	2.7	197.3	
Ceratophyllum demersum	Coontail	1.7	1.9	3.2	0.8	1.8	2.7	48.7	
Utricularia gibba	Creeping bladderw ort	1./	0.0	6.5	2.5	0.9	0.0	-100.0	V
Isoetes spp.	Quillw ort spp.	0.0	5.7	0.8	4.2	0.9	0.0	-100.0	
Potamogeton strictifolius	Stiff pondw eed	1.7	0.0	0.8	4.2	3.6	0.0	-100.0	▼
Myriophyllum tenellum	Dwarf watermilfoil	2.5	1.9	0.8	1./	0.0	1.8		
Myriophyllum sibiricum	Northern w atermilfoil	4.2	0.9	0.8	1.7	0.0	0.0		-
Potamogeton friesii	Fries' pondw eed	0.8	0.0	0.8	0.8	2.7	0.9	-67.0	V
Utricularia minor	Small bladderw ort	0.0	1.9	2.4	0.0	0.0	0.9		
Potamogeton illinoensis	Illinois pondw eed	0.0	0.0	0.0	0.0	0.0	2.7		
Nuphar variegata	Spatterdock	2.5	0.0	0.8	0.8	0.0	0.0		-
Myriophyllum spicatum	Eurasian w atermilfoil	0.0	0.9	0.0	1.7	0.0	0.9		
Sparganium sp.	Bur-reed sp.	2.5	0.9	0.0	0.0	0.0	0.0		-
Sparganium americanum	American bur-reed	0.0	0.0	2.4	0.0	0.9	0.0	-100.0	
Potamogeton foliosus	Leafy pondw eed	0.0	0.9	0.8	1.7	0.0	0.0		-
Nitella spp.	Stonew orts	0.0	0.0	0.8	0.8	0.0	0.9		
Utricularia intermedia	Flat-leaf bladderw ort	0.0	0.0	2.4	0.0	0.0	0.0		-
Schoenoplectus acutus	Hardstem bulrush	2.5	0.0	0.0	0.0	0.0	0.0		-
Potamogeton richardsonii	Clasping-leaf pondw eed	0.8	0.0	0.0	0.0	0.0	0.9		
Ceratophyllum echinatum	Spiny hornw ort	0.0	0.0	0.8	1.7	0.0	0.0		-
Sagittaria cristata	Crested arrow head	0.0	0.0	0.0	0.0	1.8	0.0	-100.0	
Ranunculus aquatilis	White water crow foot	0.8	0.0	0.0	0.0	0.9	0.0	-100.0	V
Potamogeton epihydrus	Ribbon-leaf pondw eed	0.0	0.9	0.0	0.8	0.0	0.0		-
Lemna trisulca	Forked duckw eed	0.8	0.9	0.0	0.0	0.0	0.0		-
Fissidens spp. & Fontinalis spp.	Aquatic Moss	0.0	0.0	0.0	1.7	0.0	0.0		-
Dulichium arundinaceum	Three-way sedge	0.8	0.9	0.0	0.0	0.0	0.0		-
Sparganium emersum var. acaule	Short-stemmed bur-reed	0.0	0.0	0.8	0.0	0.0	0.0		-



7.0 LATE-SUMMER 2023 EWM MAPPING SURVEYS

Multiple Onterra field survey crews conducted a late-summer EWM mapping survey on the Pike Chain of Lakes on September 5-7, 2023. Field survey notes indicated good conditions during the survey with very good water clarity, light winds and partially sunny skies. The results of mapping surveys are displayed on Maps 4-9. Overall, the EWM population in the system is low with all EWM occurrences mapped with point-based symbology's including *single or few plants, clumps of plants*, or *small plant colonies*. No colonized areas that required mapping with polygons were located anywhere in the system during the survey.

Buskey Bay

EWM was found in the form of *single or few plants* primarily with one occurrence of *clumps of plants* (Map 4). These point-based occurrences were mainly found in the southern portion of the lake. In 2022 Buskey Bay was where the majority of the EWM population was mapped. The 2023 herbicide treatment had whole-lake impacts on the EWM population.

Lake Millicent

Similar to Buskey Bay, in 2022 Lake Millicent contained multiple colonized areas of EWM. Following the 2023 herbicide treatment, EWM was only found as *single or few plant* occurrences and in the southern portion of the lake where herbicide treatments did not occur (Map 5).

Hart Lake

The EWM population in Hart Lake was relatively sparse. Most of the known occurrences were located in the southern end of the lake within the 2022 herbicide application area (Map 6). These include three *small plant colonies*, and three *clumps of plants*, and several *single or few plants* occurrences. No occurrences were marked on the north end of the lake. Hand harvesting in 2023 likely assisted in maintaining a low population in Hart Lake.

<u>Twin Bear Lake</u>

Twin Bear Lake contained most of the EWM within the chain of lakes in 2023. Numerous *small plant colonies, clumps of plants,* and *single or few plants* of EWM were marked on the western shoreline of Twin Bear Lake, including within a 2022 ProcellaCORTM treatment site (Map 7). A few other isolated occurrences were marked around the lake as well.

<u>Eagle Lake</u>

Several *singe or few* EWM plants were mapped in the channel leading from Twin Bear Lake into Eagle Lake (Map 8). Just three *clumps of plants* were located within Eagle Lake proper. No colonized areas of EWM that required polygon-based mapping were present in the lake.

<u>Flynn Lake</u>

Flynn Lake harbors a modest EWM population that consists of several *single or few plants* occurrences and one *clumps of plants* (Map 9). Several plants were located near the outlet where they have often been found in past surveys. Professional hand harvesting of this area helped to maintain the population at modest levels and inhibited further expansion. No colonized areas of EWM that required polygonbased mapping were present in the lake.



Muskellunge Lake

An Onterra field survey crew conducted a late-summer EWM Mapping Survey on Muskellunge Lake on August 7, 2023. The survey crew noted that a northern watermilfoil, a native species, was prevalent in the lake. No EWM was located within Muskellunge Lake during the survey.

Mccarry Lake

An Onterra field survey crew conducted a late-summer EWM Mapping Survey on McCarry Lake on August 7, 2023. The visual meander survey covered all littoral and navigable areas around the lake. In total, the crew marked just two *single or few plants* occurrences within the lake and noted that the combined occurrences consisted of three total individual plants (Map 6). The crew members successfully harvested all EWM plants they encountered by the roots. No EWM was observed within areas where colonies had been mapped on the eastern side of the lake in 2017-2018 before the lake was treated with 2,4-D.

Annual late-summer EWM mapping surveys on the six main chain lakes has occurred since 2007. Figure 7.0-1 shows the acreage of EWM colonies mapped on the system over the course of time. These data show that acreage of EWM was below 1.0 acre from 2007-2013. A rapid increase in colonized areas occurred from 2014-2016 with over 45 acres of EWM delineated in the 2016 survey. Aggressive herbicide management actions in 2017 resulted in reducing the overall acreage of EWM below 2.0 acres. Much of the acreage delineated in 2018 was treated in 2019 again lowering the overall EWM population to low levels. A gradual increase in EWM was measured from 2019-2022 and the 2023 management strategy targeted much of the colonized areas in the system. The 2023 mapping survey found no areas that required area-based mapping methods (0 acres) which was the lowest amount of colonized EWM since 2010.



8.0 CONCLUSIONS & DISCUSSION

The 2023 herbicide treatments that took place on the system were met with a high level of initial efficacy with little to no EWM located within any of the application areas. The Buskey Bay and Lake Millicent ProcellaCOR spot treatments appeared to have resulted in lake wide impacts to EWM as expected. The 2,4-D barrier curtain treatment was successfully implemented and met initial control expectations. All of the 2023 managed sites will be evaluated in 2024 to determine whether the EWM control extends through the year after treatment which would then meet control expectations for the strategies.

Extended monitoring of the 2022 herbicide treatments showed that the initial EWM reductions that were observed last year were maintained through 2023 in part as a result of additional management efforts through a professional hand harvesting effort.

Native aquatic plant monitoring showed minimal negative impacts within lakes treated during 2023. Whole-lake point-intercept surveys also show the Pike Chain continues to harbor a high quality aquatic plant population, although many lakes are showing a declining trend in species richness and floristic quality over time. Declines in occurrence of several native species in Eagle Lake between the 2020-2023 surveys is believed to be related to a combination of natural population variability, differences in littoral depths between the two surveys, and potentially impact from the 2022 herbicide treatment strategy for select susceptible species such as coontail and native milfoils.

8.1 2024 EWM Management & Monitoring Strategy

As a result of EWM reductions from recent active management strategies, no herbicide management is planned to occur during 2024. In summer 2024, a replication of the subsample point-intercept survey will occur in the 2023 herbicide management sites to evaluate whether the strategy meets control expectations with EWM reductions extending through the year after treatment. A late-season EWM mapping survey on the six main chain lakes is also planned which is used to assess past management activities as well as to plan for potential EWM management strategies the following year.





















A

APPENDIX A

2023 Herbicide Concentration Monitoring Plans

Buskey Bay, Bayfield County (WBIC: 2903800) 2023 Herbicide Sample Plan Onterra, LLC

Buskey Bay, located in Bayfield County, is an approximately 88-acre seepage lake that has a maximum depth of 51 feet. Florpyrauxifen-benzyl (commercially as ProcellaCORTM) is proposed to be applied to four separate application areas totaling 11.7 acres in early-summer 2023 to control Eurasian watermilfoil. Herbicide concentration sampling will be conducted in order to monitor the herbicide concentrations in the hours and days following the application.

Water samples will need to be collected at the sites and depths listed below. Coordinates are in decimal degrees. Locations of each sampling site are displayed with green squares on the image below.



Buskey Bay Herbicide Sample Sites								
Site Label	Site Description	Station ID	Latitude	Longitude	Sample Depth			
B1	Application area	10057529	46.524552	-91.380909	Integrated (0-6 feet)			
B2	Near center	10048393	46.52601	-91.37538	Integrated (0-6 feet)			
B3	Deep hole	10020502	46.53147	-91.37155	Integrated (0-6 feet)			

Please note that a single sample is to be collected before the treatment as a 'control' for the lab analysis. Please collect the pre-treatment sample from site B1 at a time that is most convenient for the volunteer but as close to the treatment date as possible. After the herbicide application is completed, 22 additional samples will need to be collected at eleven different time intervals throughout the project and are listed in the table below. Sample collection intervals are listed either as <u>Hours After Treatment (HAT) or Days After Treatment (DAT)</u>. Direct communication Finalized 5/1/2023

between the water sample collector and the herbicide applicator is necessary to ensure the collector is prepared to begin three hours after treatment is completed. If a sample cannot be collected at the interval listed below, please collect the sample as soon as reasonably possible and record the change.

Sampling Interval Matrix (X indicates sample to be collected)							
	Applicat	tion Area	Deep Hole				
Interval	Site B1	Site B2	Site B3				
Pre-Treatment	Х						
3 HAT	Х	Х	Х				
9 HAT	Х	Х	Х				
24 HAT	Х	Х	Х				
2 DAT	Х	Х	Х				
4 DAT	Х	Х	Х				
7 DAT	Х	Х	Х				
14 DAT			Х				
28 DAT			Х				
42 DAT			Х				
56 DAT			Х				
70 DAT			Х				
HAT = Hours After Treatment, DAT = Days After Treatment							

All water samples will be collected using a six-foot integrated sampler (Photo 1). A video tutorial demonstrating the proper sample collection methodology is available on Onterra's YouTube web page: <u>click here</u>



Due to the extremely low concentrations being measured at the laboratory (<1 part per billion), it is very important to thoroughly rinse the integrated sampler device and the custom mixing bottle with the water from each sampling site upon arrival at the site. Water is collected by pushing the integrated sampler straight down to a depth of six feet; or in water shallower than six feet, down to approximately one foot above the bottom sediment. The sampler is brought to the surface and emptied into a customized mixing bottle by pushing open the stop valve at the end of the integrated sampler (Photo 2). Water should be poured from the custom mixing bottle to triple rinse the clear glass bottle. After the clear glass bottle is triple rinsed, it is to be filled for a fourth time with the water from the custom mixing bottle and then carefully poured into the brown glass bottle which has a preservative solution already inside (Photo 3).

Please use a fine-tipped permanent marker to record the date and time the sample is collected on the sticker label of the brown glass bottle. The final sample (in the brown bottle) as well as the emptied clear glass bottle should be carefully placed back within the bubble wrapped pouch to protect from accidental breakage. While the samples are being collected, they should be kept cold and out of direct sunlight by keeping them in a small cooler on the boat. After collection, all samples should be stored in a refrigerator until shipping.



Onterra will provide all of the necessary supplies to complete the sampling and provide training to the volunteer(s) collecting the samples. Onterra has a supply of handheld GPS units and integrated sampler devices available to loan out for the duration of the sampling upon request. All other materials, including sampling bottles with labels, a customized mixing bottle and necessary paperwork will be provided.

Please fill out the yellow highlighted fields on the Chain of Custody forms including:

- Sampler: (Volunteer Name)
- Client Sample ID: (example: B1, B2 or B3)
- Date sample is collected

Shipping Instructions

- 1) When all sampling is complete, make sure all sample vials are placed in bubble wrap within the provided soft cooler.
- 2) Put an ice pack into the soft cooler. This can also be a frozen water bottle (contained in an unlabeled zip lock bag). Do not place loose ice in the cooler.
- 3) Find a cardboard box that will fit the soft cooler for transport. If needed, pack empty space with packing material so the soft cooler is secure within the cardboard box.
- 4) Place the completed Chain of Custody forms in the cardboard box.
- 5) <u>Only ship Monday Thursday.</u> The lab will not be open to receive the samples on a Saturday.
- 6) We recommend utilizing *FedEx Standard Overnight* so the samples can be received the next day by the lab before 4:30PM (when the lab closes).
- 7) Shipping costs are expected to be \$150-\$200 for next day delivery.
- 8) Ship the cardboard box containing the soft-sided cooler bag, water samples, and Chain of Custody forms to the address below:

EPL Bio Analytical Services 9095 W. Harristown Blvd. Niantic, IL 62551
If you have any questions, please reach out to one of the contacts listed below.

Project specifics, logistics and sampling methods					
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Lake Millicent, Bayfield County (WBIC: 2903700) 2023 Herbicide Sample Plan Onterra, LLC

Lake Millicent, located in Bayfield County, is an approximately 183-acre drainage lake that has a maximum depth of 53 feet. Liquid 2,4-D is proposed to be applied to approximately 4.3 acres on the north end of the lake in spring of 2023 to control Eurasian watermilfoil. Herbicide concentration sampling will be conducted in order to monitor the herbicide concentrations in the hours and days following the application.

Water samples will need to be collected at the sites and depths listed below. Coordinates are in decimal degrees. Locations of each sampling site are displayed with green squares on the image below.





Extent of large map shown in red.

Lake Millicent Herbicide Sample Sites									
Site Label	Site Description Station ID Latitude Longitude Sample Depth								
M4	Application Area M A-23	10057532	46.53896	-91.36197	Integrated (0-6 feet)				
M5	Application Area M A-23	10057533	46.53800	-91.36254	Integrated (0-6 feet)				
M6	Outside Application Area	10057534	46.53736	-91.36332	Integrated (0-6 feet)				
M7	Outside Application Area	10057535	46.53722	-91.36245	Integrated (0-6 feet)				

Typically, when structures are placed in a navigable waterway, a permit issued under NR 329, Wis. Adm. Code is required. However, when the temporary use of curtains is used to segregate invasive plant beds for chemical control, and is demonstrated to be a benefit to the public resource and protect the public rights in navigable waterways, the Department has made a determination to allow for the temporary placement of these structures without a NR 329 permit. Barriers must be placed no sooner than 24 hours before treatment and must be removed no later than 72 hours after treatment, not to exceed a total of 96 hours.

This sampling plan was created under the assumption the barrier curtain will be removed at the 72-hour after treatment limit. The table below separates the sampling intervals as either before or after curtain removal. Samples will need to be collected at ten total intervals. Five sampling intervals are scheduled to take place before curtain removal and are referred to as Hours After Treatment (HAT). The remaining four sampling intervals are referred to as Hours After Curtain (HAC) and indicate the number of hours after the curtain has been removed. If a sample cannot be collected at the interval listed below, please collect the sample as soon as reasonably possible and record the change.

Sampling Interval Matrix (X indicates sample to be collected)								
Intorval	Application	Area M A-23	Outside App	olication Area				
Interval	Site M4	Site M5	Site M6	Site M7				
	Herbicide	e Application Co	omplete					
1 HAT	Х	Х	Х	Х				
6 HAT	Х	Х	Х	Х				
24 HAT	Х	Х	Х	Х				
48 HAT	Х	X	Х	Х				
72 HAT	Х	Х	Х	Х				
	Barrie	r Curtain Rem	oved					
1 HAC	Х	Х	Х	Х				
3 HAC	Х	X	Х	Х				
6 HAC	Х	Х	Х	Х				
12 HAC	Х	Х	Х	X				
24 HAC X X X X								
HAT =	HAT = Hours After Treatment, HAC = Hours After Curtain							

All water samples will be collected using a six-foot integrated sampler (Photo 1). A video tutorial demonstrating the proper sample collection methodology is available on Onterra's YouTube web page: <u>click here</u>



Photo 1. Six-Foot Integrated Sampler (top) & custom transfer bottle (Bottom).

Water is collected by pushing the integrated sampler straight down to an approximate depth of six feet; or in water less than six feet, down to approximately one foot above the bottom sediment. The sampler is brought to the surface and emptied into a customized mixing bottle by pushing open the stop valve of the integrated sampler. The mixing bottle should be given a brief stir to mix the contents, and then emptied from the mixing bottle into the appropriately labeled final 60 mL sampling bottle. Once in the final sampling bottle, the water sample must be completely preserved by adding 3-4 drops of sulfuric acid with an eye dropper.

Onterra will provide all of the necessary supplies to complete the sampling and provide training to volunteers collecting the samples. Onterra has a supply of GPS units, temperature probes, and integrated sampler devices available to loan out for the duration of the sampling upon request. All other materials including pre-labeled sampling bottles, datasheets and a shipping container will be provided.

While the samples are being collected, they should be kept cold and out of direct sunlight by keeping them in a small cooler on the boat. After collection, all samples should be stored in a refrigerator until shipping.

It is important to use a separate data sheet for each day that is monitored. Please fill out one data sheet for each sample interval and fill in the highlighted boxes. Store the preserved samples in a refrigerator. After the completion of the final sampling interval, please ship all of the samples and the data sheets to the Wisconsin State Lab of Hygiene (WSLH) within the insulated shipping box. Please review the attached Herbicide Sampling Handling Instructions for specific shipping instructions.

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If you have any questions, please call or email one of the contacts listed below. If you have any questions, please reach out to one of the contacts listed below.

Lake Millicent, Bayfield County (WBIC: 2903700) 2023 Herbicide Sample Plan Onterra, LLC

Lake Millicent, located in Bayfield County, is an approximately 183-acre drainage lake that has a maximum depth of 53 feet. Florpyrauxifen-benzyl (commercially as ProcellaCORTM) is proposed to be applied to five separate application areas totaling 8.9 acres in early-summer 2023 to control Eurasian watermilfoil. Herbicide concentration sampling will be conducted in order to monitor the herbicide concentrations in the hours and days following the application.

Water samples will need to be collected at the sites and depths listed below. Coordinates are in decimal degrees. Locations of each sampling site are displayed with green squares on the image below.



Millicent Lake Herbicide Sample Sites									
Site Label Site Description Station ID Latitude Longitude Sample Depth									
M1	Deep hole	10057530	46.52477	-91.36941	Integrated (0-6 feet)				
M2	Application area	10057531	46.529765	-91.36886	Integrated (0-6 feet)				
M3	Application area	43086	46.53507	-91.364984	Integrated (0-6 feet)				

Please note that a single sample is to be collected before the treatment as a 'control' for the lab analysis. Please collect the pre-treatment sample from site M1 at a time that is most convenient for the volunteer but as close to the treatment date as possible. After the herbicide application is completed, 23 additional samples will need to be collected at eleven different time intervals throughout the project and are listed in the table below. Sample collection intervals are listed either as <u>Hours After Treatment (HAT) or Days After Treatment (DAT)</u>. Direct communication

between the water sample collector and the herbicide applicator is necessary to ensure the collector is prepared to begin three hours after treatment is completed. If a sample cannot be collected at the interval listed below, please collect the sample as soon as reasonably possible and record the change.

Sampling Interval Matrix (X indicates sample to be collected)							
	Applica	tion Area	Deep Hole				
Interval	Site M1	Site M2	Site M3				
Pre-Treatment	Х						
3 HAT	Х	Х	Х				
9 HAT	Х	Х	Х				
24 HAT	Х	Х	Х				
2 DAT	Х	Х	Х				
4 DAT	Х	Х	Х				
7 DAT	Х	Х	Х				
14 DAT			Х				
28 DAT			Х				
42 DAT			Х				
56 DAT			X				
70 DAT			Х				
HAT = Hours After Treatment, DAT = Days After Treatment							

All water samples will be collected using a six-foot integrated sampler (Photo 1). A video tutorial demonstrating the proper sample collection methodology is available on Onterra's YouTube web page: <u>click here</u>



Due to the extremely low concentrations being measured at the laboratory (<1 part per billion), it is very important to thoroughly rinse the integrated sampler device and the custom mixing bottle with the water from each sampling site upon arrival at the site. Water is collected by pushing the integrated sampler straight down to a depth of six feet; or in water shallower than six feet, down to approximately one foot above the bottom sediment. The sampler is brought to the surface and emptied into a customized mixing bottle by pushing open the stop valve at the end of the integrated sampler (Photo 2). Water should be poured from the custom mixing bottle to triple rinse the clear glass bottle. After the clear glass bottle is triple rinsed, it is to be filled for a fourth time with the water from the custom mixing bottle and then carefully poured into the brown glass bottle which has a preservative solution already inside (Photo 3).

Please use a fine-tipped permanent marker to record the date and time the sample is collected on the sticker label of the brown glass bottle. The final sample (in the brown bottle) as well as the emptied clear glass bottle should be carefully placed back within the bubble wrapped pouch to protect from accidental breakage. While the samples are being collected, they should be kept cold and out of direct sunlight by keeping them in a small cooler on the boat. After collection, all samples should be stored in a refrigerator until shipping.



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Please fill out the yellow highlighted fields on the Chain of Custody forms including:

- Sampler: (Volunteer Name)
- Client Sample ID: (example: M1, M2, or M3)
- Date sample is collected

Shipping Instructions

- 1) When all sampling is complete, make sure all sample vials are placed in bubble wrap within the provided soft cooler.
- 2) Put an ice pack into the soft cooler. This can also be a frozen water bottle (contained in an unlabeled zip lock bag). Do not place loose ice in the cooler.
- 3) Find a cardboard box that will fit the soft cooler for transport. If needed, pack empty space with packing material so the soft cooler is secure within the cardboard box.
- 4) Place the completed Chain of Custody forms in the cardboard box.
- 5) <u>Only ship Monday Thursday</u>. The lab will not be open to receive the samples on a Saturday.
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B

APPENDIX B

Pike Chain EWM Removal Report 2023 – Aquatic Plant Management LLC



Pike Chain of Lakes EWM Removal Report 2023

PO Box 1134 Minocqua, WI 54548



Pike Chain of Lakes EWM Removal Summary 2023

Dive Background: In August, Aquatic Plant Management LLC (APM) conducted four (4) days of Diver Assisted Suction Harvesting (DASH) and Hand Harvesting for Eurasian Watermilfoil (EWM) on Pike Chain of Lakes in Bayfield County, WI. The team focused their efforts at 5 sites as prioritized by the Pike Chain of Lakes Association. In total APM was able to remove **184.5 cubic feet of EWM** from Pike Chain of Lakes.

Date	Weather Conditions	Water Temp (F)	Underwater Dive Time (hrs)	AIS Removed (cubic ft)
8/14/2023	Cloudy	67	5.9	57.0
8/15/2023	Sunny	67	4.8	48.0
8/16/2023	Cloudy	70	6.3	64.0
8/17/2023	Cloudy	67	6.5	15.5
Grand Total			23.4	184.5

Dive Location	Avg. Water Depth	# of Dives	Underwater Dive Time	AIS Removed (cubic feet)
Eagle HH	6.0	1	3.3	10.0
Flynn HH	5.5	1	3.3	5.5
Hart-A-23	12.0	1	0.6	1.5
Hart-B-23	8.8	10	9.3	85.5
TB-A-23	12.0	5	7.0	82.0
Grand Total	9.5	18	23.4	184.5

Dive Highlights and Recommendations: The DASH team spent ~40% of their time on Hart Lake primarily at site Hart-B-23. Next, the team moved to Twin Bear lake, before shifting to hand harvesting at two additional sites in Eagle and Flynn lakes. Overall, the Pike Chain of Lakes should continue to take an Integrated Pest Management (IPM) approach and evaluate different strategies to manage the EWM population on the lake. Continued monitoring and management efforts are important to prevent the spread of EWM throughout the Pike Chain of Lakes.



Map of Pike Chain of Lakes Dive Sites



Aquatic Plant Management LLC



Detailed Diving Activities

Date	Dive Location	Latitude	Longitude	Underwater Dive Time (hrs)	AIS Removed (cubic ft)	AIS Density	Avg Water Depth (ft)	Native Species	Native By- Catch	Substrate Type
8/15/2023	Hart-B-23	46.51375	-91.36649	0.67	5.0	Clumps	9.0	Pondweeds	0.5	Organic
8/15/2023	Hart-B-23	46.51442	-91.36728	1.00	7.5	Clumps	9.0	Coontail	1.5	Organic
8/15/2023	Hart-B-23	46.51517	-91.36756	0.58	2.0	Clumps	8.0	Coontail	1.0	Organic
8/15/2023	Hart-B-23	46.51528	-91.36750	0.67	7.0	Clumps	8.0	Pondweeds	0.5	Organic
8/15/2023	Hart-B-23	46.51528	-91.36750	1.08	8.5	Clumps	8.0	Coontail	0.5	Organic
8/15/2023	TB-A-23	46.50777	-91.37050	0.75	18.0	Surface Matting	14.0	None	0.0	Organic
8/14/2023	Hart-A-23	46.51249	-91.36353	0.58	1.5	Clumps	12.0	Pondweeds	0.5	Organic
8/14/2023	Hart-B-23	46.51281	-91.36533	1.33	8.0	Small Plant Colony	10.0	Coontail	0.5	Organic
8/14/2023	Hart-B-23	46.51272	-91.36602	1.67	14.0	Highly Dominant	10.0	Coontail	1.0	Organic
8/14/2023	Hart-B-23	46.51272	-91.36604	1.42	22.5	Highly Dominant	10.0	Coontail	2.5	Organic
8/14/2023	Hart-B-23	46.51365	-91.36645	0.58	5.5	Clumps	8.0	Coontail	0.5	Organic
8/14/2023	Hart-B-23	46.51379	-91.36649	0.33	5.5	Small Plant Colony	8.0	Coontail	0.5	Organic
8/16/2023	TB-A-23	46.50800	-91.37052	1.42	18.5	Surface Matting	12.0	Coontail	2.0	Organic
8/16/2023	TB-A-23	46.50800	-91.37052	1.50	9.5	Surface Matting	12.0	Coontail	6.0	Organic
8/16/2023	TB-A-23	46.50800	-91.37052	1.75	24.5	Surface Matting	12.0	Coontail	10.0	Organic
8/16/2023	TB-A-23	46.50800	-91.37050	1.58	11.5	Dominant	10.0	Coontail	0.5	Organic/Sand
8/17/2023	Eagle HH	46.50457	-91.36140	3.25	10.0	Dominant	6.0	Pondweeds	2.0	Organic
8/17/2023	Flynn HH	46.49047	-91.34589	3.25	5.5	Scattered	5.5	Grasses	1.0	Organic
Total	18			23.41	184.5					

Aquatic Plant Management LLC