Pike Chain of Lakes Bayfield County, Wisconsin 2022 EWM Management & Monitoring Report January 2023

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1.0 INTRODUCTION

The Pike Chain of Lakes is comprised of 9 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 portage (shown in pink).

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 as 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).

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.

The IRPCLA understands that the small size and exposed/off-shore nature of the EWM colonies on the chain make them difficult scenarios to hold sufficient herbicide <u>c</u>oncentrations and <u>exposure times</u> (CETs) to result in multi-year control. Specific to the Pike Chain, the *Plan* included additional guidance on using herbicides that have reported short CET requirements (e.g. florpyrauxifen-benzyl) as well as manipulating conditions to hold higher and longer CETs such as deploying barrier curtains.

1.2 2022 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 2022 control and monitoring plan was outlined within the *2021 Control Strategy Development Report* distributed in early February 2022. This strategy was also incorporated into a successful WDNR AIS Large-Scale Population Control Grant application, providing state-share assistance in carrying out the effort. This report marks the first report deliverable of ACEI-291-22, as well as provides the control and monitoring plan for 2023 (Section 6.0).

The IRPCLA aimed to conduct a series of trial treatments in 2022 with follow-up hand-harvesting/DASH in 2023. Eagle Lake had the highest EWM population and was slated to be targeted with a series of ProcellaCORTM spot treatments that would likely have the potential to produce lake -wide EWM impacts. A problematic site in an area of flow on Hart Lake was to be targeted with ProcellaCORTM in a spot treatment scenario. One site in Twin Bear Lake was selected for liquid 2,4-D amine treatment within a temporary barrier curtain enclosure, and a separate site in Twin Bear to be targeted with ProcellaCORTM spot treatment. The herbicide treatments were scheduled to occur in mid-June 2022 to allow for the collection of meaningful pretreatment quantitative assessment of the native aquatic plants while fisheries managers and Tribal partners also favored the slightly later treatment date to avoid overlap with sensitive stages of larval fish development. The WDNR put a condition on the permit such that the herbicide treatment could not occur until after June 15 to avoid overlap with sensitive life stages of certain fish species (primarily 0-14 days post hatch).

1.3 Pretreatment Confirmation and Refinement Survey

Onterra ecologists conducted the Pretreatment Confirmation and Refinement Surveys on the Pike Chain on Tuesday, June 14, 2022. 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 between 65-69°F. Using an optical probe, the pH was measured at 8.2-8.5 dependent on depth and location. 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.



Photograph 1.3-1. Actively growing EWM observed during Pretreatment Survey. Photo credit 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 24, 2022 by Northern Aquatic Services. The applicator noted northwest winds between 2-6 mph during the time of the applications.

2.0 2022 MONITORING METHODS

It is important to note that two types of surveys are discussed in the subsequent materials: 1) pointintercept surveys and 2) EWM mapping surveys. 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. It is common to see a particularly plant species, such as EWM, very near the sampling location but not yield it on the rake sampler. Particularly in low-density colonies such as those designated by Onterra as *highly scattered* and *scattered*, large gaps between EWM plants may exist resulting in EWM not being present at a particular pre-determined point-intercept sampling location in that area. 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 (Photograph 2.0-1). 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.



Photograph2.0-1.EWMmapping survey on a Wisconsinlake.Photo credit Onterra.

Overall, each survey has its strengths and weaknesses, which is why both are utilized in different ways as part of this project.

Sub-sample Point-Intercept Survey

A quantitative monitoring plan involved the collection of a total of 171 sub-sample pointintercept sampling locations contained within the five trial treatment sites (Figure 2.0-1). The quantitative assessment is completed through the comparison of the sub point-intercept survey from mid-June 2022 (year of pretreatment), late-season 2022 (year of post-treatment), and late-season 2023 (year after treatment). The 2022 herbicide treatment was planned for roughly the middle of June and ultimately occurred on June 22. 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 below, the pretreatment data were collected on June 14, 2022 and the post-treatment data were collected on September 7, 2022.



EWM Mapping Surveys

Onterra conducted the Late-Summer EWM Mapping Survey on August 30-31, 2021 to qualitatively assess the peak growth (peak-biomass) of the EWM population throughout the lake and to aid in the development of management options for 2022. A replication of the Late-Summer EWM Mapping Survey in 2022 serves to evaluate the efficacy of the management strategy.

3.0 2022 TREATMENT MONITORING RESULTS

3.1 Twin Bear Lake - Site TB A-22 (2,4-D barrier curtain)

The preliminary 2022 treatment plan was to target an area of 3.4 acres within a barrier curtain, requiring approximately 950 feet of curtain. Based upon logistical hurdles encountered on the day of deployment, the decision was made, with WDNR support, to reduce the treatment area to 2.0 acres, requiring a more-manageable 400 linear feet of curtain.

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. As was the case for the 2022 treatment on Twin Bear, 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 a barrier curtain on June 23, 2022 (Photo 3.1-1) with site A-22 in Twin Bear Lake being treated with liquid 2,4-D on June 22, 2022 at an application area dosing rate of 4.0 ppm.



Photograph 3.1-1. Barrier curtain construction & placement in Twin Bear Lake. Photo credits IRPCLA.

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 3.1-1, left frame). Following the treatment, one *single or few* EWM plants occurrence was located within the treated area (Figure 3.1-1, right frame).





Due to the reduction of the size of the final treatment site, the intensity of sub-sample point-intercept monitoring was also reduced. 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 3.1-1). 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 3.1-1. Pre/Post an	alysis of aquation	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	
Cha	ara spp.	Muskgrasses	2	4	
Cei	ratophyllum demersum	Coontail	3	2	
Ler	mna trisulca	Forked duckweed	1	2	
Pot	tamogeton pusillus	Small pondweed	0	2	
Naj	ijas flexilis	Slender naiad	1	1	
My	riophyllum spicatum	Eurasian watermilfoil	3	0	
Elo	odea canadensis	Common waterweed	1	1	
Pot	tamogeton robbinsii	Fern-leaf pondweed	0	1	
Pot	tamogeton gramineus	Variable-leaf pondweed	0	1	
Pot	tamogeton amplifolius	Large-leaf pondweed	0	1	
Nu	ıphar variegata	Spatterdock	0	1	



2,4-D 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 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 and reporting the results as 2,4-D acid equivalent (ae). A copy of the final herbicide concentration monitoring plan is included as Appendix A. Please note that that only sites TB1 and TB3 were retained following the logistical need to reduce the final size of this treatment site.

The application rate of the reduced treatment site remained constant at 4.0 ppm ae, with the gallons of product required to reach that concentration being reduced to 45 gallons compared with the original estimate of 77 gallons.

The 2022 herbicide concentration samples were collected by volunteers at two separate sites - one within the barrier (TB1), and one outside of the barrier (TB3). 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 73, 75, 78, 84, and 96 HAT; which in terms of post curtain removal relate to 1 HAT, 3 HAT, 6 HAT, 12 HAT, and 24 HAT. All samples were preserved and then sent to the State Lab of Hygiene for analysis.

Figure 3.1-2 and Table 3.1-2 displays the concentration of 2,4-D at the two 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 measured at 7.9 ppm at site TB1 and was not detected outside of the barrier at one HAT. At 24 HAT, the concentration at site TB1 measured 5.1 ppm and 0.07 ppb at site TB3. At three hours after the curtain removal, 75 HAT, the concentration measured 0.26 ppb at site TB1 and 0.06 ppb at site TB3. The measured concentrations exceeded the targets, and remained above target for 24 HAT.

	Hours after Curtain Removal									
	1	6	24	48	72	1	3	6	12	24
TB1	7.90	5.40	5.10	2.80	1.50	1.10	0.26	0.76	0.20	0.03
TB3	ND	0.01	0.07	0.07	0.04	0.04	0.06	0.03		0.02

 Table 3.1-2. Twin Bear Lake 2022 2,4-D Concentration Monitoring Results.
 TB1 within curtain, TB3 outside curtain.

 Curtain.
 Values in parts per million (ppm).





3.2 Twin Bear Lake - Site TB B-22 (ProcellaCOR Spot Treat)

Site B-22 comprises 5.6-acres in Twin Bear Lake and was treated with ProcellaCORTM in June 2022 at a dosing application rate of 4.5 PDU's (Map 1). The pretreatment EWM population consisted of a dominant colony in the north end of the site with many single plants and clumps of plants, as well as a small plant colony elsewhere in the site (Figure 3.2-1, left frame). After treatment, no colonized areas of EWM were present, but two small plant colonies and other point-based occurrences were present within the treated area (Figure 3.2-1, right frame).





Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 24 sampling locations within the application area. Analysis of these data show that the occurrence of EWM was reduced from 16.7% prior to treatment to 0% in the post-treatment survey. The occurrence of variable-leaf pondweed, forked duckweed, and slender 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 (4.2%).



3.3 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 (Map 2). 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, single or few plants (Figure 3.3-1, left frame). 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 (Figure 3.3-1, right frame).





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. The occurrence of small pondweed, variable-leaf pondweed, and slender naiad showed statistically higher occurrences in the post-treatment survey.





3.4 Eagle Lake – Site E-22 & F-22 (ProcellaCOR Spot Treat)

Sites E-22 (7.4 acres) and F-22 (8.0 acres) were each treated with ProcellaCORTM in June 2022 with an application area dosing rate of 3.0 PDU's (Map 3). The pretreatment mapping survey indicated a combination of colonized EWM and point-based occurrences within each site (Figure 3.4-1, left frames). After treatment, no EWM was marked within site E-22, while two single or few plants occurrences were mapped in the western end of site F-22 (Figure 3.4-1, right frame).



Quantitative monitoring consisted of the completion of a sub-sample point-intercept survey from 34 sampling locations within the application area E-22 and another 38 sampling locations within application area F-22. Each site is analyzed individually. Within Site E-22, the occurrence of EWM was reduced from 38.2% prior to treatment to 0% in the post-treatment survey (Figure 3.4-2). The occurrence of



small pondweed, wild celery, and spatterdock showed statistically higher occurrences in the posttreatment survey. Analysis of the data collected from site F-22 show that the occurrence of EWM was reduced from 28.9% to 0% (Figure 3.4-3). Large-leaf pondweed exhibited a statistically valid decrease in occurrence between the two surveys, while wild celery, clasping-leaf pondweed, and small pondweed all showed statistically valid increases in occurrence.







represents statistically valid change from June 2022 to September 2022 (Chi-Square $\alpha = 0.05$). n=38.



Florpyrauxifen-benzyl 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 florpyrauxifen-benzyl that were achieved in the hours and days after treatment. Samples were collected four total sites following treatment – two within application areas, one site located in the deep hole area of the lake, and one located in the center of downstream Flynn Lake. Samples were collected at nine time intervals after treatment beginning at 3 hours after treatment (HAT), with additional samples collected at 9, 24, and 48 HAT as well as 4, 7, 14, 21, and 28 days after treatment (DAT). Samples were collected by a volunteer member of the association and upon completion of the sampling, were shipped to EPL Bio Analytical Services in Niantic Illinois for analysis. This lab was identified by the WDNR as being able to detect florpyrauxifen-benzyl at lower levels than the herbicide manufacturer's facility – 1 part per billion (ppb). A copy of the herbicide concentration monitoring plan is included as Appendix B. All samples were tested for florpyrauxifen-benzyl and florpyrauxifen acid.

The EPL Lab reports the concentration in parts per billion (ppb) of the initial parent active ingredient in ProcellaCORTM (florpyrauxifen-benzyl, SX-1552), as well as an acid metabolite (florpyrauxifen acid, SX-1552-A) which is the immediate by-product that it breaks down into.

Figures 3.4-5 & 3.4-6 and Table 3.4-1 displays the concentrations of florpyrauxifen-benzyl from the three monitoring locations. For reference, the dosing rate of 3.0 PDU (prescription dosing units) equates to 5.8 ppb of florpyrauxifen-benzyl.

Site E1 was placed in application area E-22 and site E2 was placed in application area F-22. The active ingredient was measured at 2.90 ppb at site E1 and 5.22 ppb at site E2 at 3 HAT, which can be best observed on Figure 3.4-6. Figure 3.4-6 shows the same data as Figure 3.4-5, but reduced the horizontal axis by a power of 10. Concentrations measured at 9 HAT decreased to 0.949 ppb at E1 and 0.463 ppb at E2. By 24 HAT, the active ingredient was measured at 0.251 ppb at site E1 and 0.319 ppb at site E2. By 21 DAT, the last sample interval for sites E1 and E2, the active ingredient measured 0.0120 ppb at site E1 and was not detected at site E2.

In an effort to understand the lake-wide herbicide concentration following dispersion and dissipation away from the herbicide application area, samples were collected from the deep hole location in the central part of Eagle Lake (site E3). Concentrations at site E3 are expected to be reflective of the lake-wide concentration following treatment. Herbicide concentrations at 9 HAT at site E3 were 0.36 ppb compared with the whole-lake potential concentration of 0.41 ppb. Studies of this nature conducted to date indicate the herbicide mixes and reaches equilibrium within the mixing water volume by approximately 24-48 HAT. For ProcellaCORTM, this herbicide quickly degrades into the acid metabolite version, potentially before dissipating into a lake-wide volume occurs. Concentrations of the active ingredient was still detected at the final sample interval 28 DAT.

Additionally, a sampling site was placed in Flynn Lake (F1), downstream of Eagle Lake, to try and capture any directional herbicide movement downstream. Samples were collected 7 DAT and 21 DAT from this location. At 7 DAT, the parent ingredient was measured at 0.12 ppb. At 21 DAT, the parent ingredient was not detected.

 Table 3.4-1. Eagle Lake 2022 Florpyrauxifen-benzyl concentration monitoring results.
 Values in parts

 per billion (ppb).
 Values in parts
 Values in parts



	Florpyrauxifen-benzyl (SX-1552) ppb HAT										
	3	9	24	48	96 (4 DAT)	168 (7 DAT)	336 (14 DAT)	504 (21 DAT)	672 (28 DAT)		
E1	2.900	0.949	0.251	0.154	0.072	0.000	0.010	0.012			
E2	5.22	0.463	0.319	0.097	0.016	0.000	0.000	0.000			
E3		0.355	0.129	0.196	0.020	0.025	0.000	0.114	0.102		
F1						0.118			0.000		





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 of the acid metabolite (florpyrauxifen acid, SX-1552-A) are displayed on Table 3.4-2 and Figure 3.4-7 The measured concentrations of the acid metabolite were variable with a peak around 4-7 DAT. Concentrations were maintained above detection limits in many of the sites out to the last sampling interval at 28 DAT.

Table 3.4-2. Eagle Lake 2022 Florpyrauxifen acid concentration monitoring results.Values in parts perbillion (ppb).

Florpyrauxifen acid (SX-1552-A) ppb HAT										
	3	9	24	48	96 (4 DAT)	168 (7 DAT)	336 (14 DAT)	504 (21 DAT)	672 (28 DAT)	
E1	0.401	0.351	0.228	0.080	0.2058	0.076	0.189	0.0237		
E2	0.31	0.084	0.075	0.242	0.2877	0.178	0.171	0.106		
E3		0.036	0.018	0.011	0.184	0.3078	0.1876		0.175	
F1						0.0401			0.000	



4.0 LATE-SUMMER 2022 EWM MAPPING SURVEYS

Multiple Onterra field survey crews conducted a late-summer EWM mapping survey on the six main bodies of water comprising the Pike Chain of Lakes on September 6-7, 2022. Field survey notes indicated excellent conditions during the survey with very good water clarity, light winds and mostly sunny skies. The results of mapping surveys are displayed on Maps 4-9.

Buskey Bay

The largest concentration of EWM in the system was documented in Buskey Bay. Most of the littoral areas of the lake harbored EWM at various densities (Map 4). Most of the eastern shoreline of the lake contained colonized EWM ranging in density from *highly scattered* to *highly dominant*. Many point-based occurrences including *single or few plants*, *clumps of plants*, and *small plant colonies* were mapped throughout the littoral areas of the lake.



Lake Millicent

EWM was relatively common in littoral areas of Lake Millicent in the late-summer survey with several colonized areas in the lake as well as numerous *single plants*, *clumps of plants* and *small plant colonies* (Map 5). Some of the larger colonies were located in the northern half or so of the lake with slightly lower densities in the southern portion of the lake.

<u>Hart Lake</u>

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 two *small plant colonies*, and several *clumps of plants* and *single or few plants* occurrences. One isolated *single or few plants* occurrence was marked on the north end of the lake. No colonized areas of EWM that required polygon-based mapping were present in the lake.

<u>Twin Bear Lake</u>

Two *small plant colonies*, and several *clumps of plants* and *single or few plants* of EWM were marked on the western shoreline of Twin Bear Lake, including within a 2022 ProcellaCOR treatment site (Map 7). A few other isolated occurrences were marked around the lake as well. Just one *single or few plants* occurrence was located within the extents of the 2022 2,4-D barrier curtain treatment site. No colonized areas of EWM that required polygon-based mapping were present in the lake.

Eagle Lake

Several *singe or few* EWM plants were mapped in the channel leading from Twin Bear Lake into Eagle Lake (Map 8). Just two *single plants* were located within the 2022 ProcellaCOR application area F-22. No EWM was located within site E-22. 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 around 15 *single or few plants* occurrences (Map 9). Several plants were located near the outlet where they have often been found in past surveys. No colonized areas of EWM that required polygon-based mapping were present in the lake.

5.0 CONCLUSIONS & DISCUSSION

Monitoring results showed EWM reductions in the ProcellaCORTM spot treatment sites in Hart Lake and Twin Bear Lake; however, Onterra expected a little less EWM to be present in the application areas during the *year or treatment*. Most often the limiting factor in meeting control expectations in a spot treatment design is achieving sufficient herbicide concentration exposure times (CET's) needed to kill the target plants. Relatively small treatment areas often fail to meet CET's when rapidly dissipating herbicides like 2,4-D are used, however ProcellaCORTM is thought to be more effective in these scenarios.

There are a number of factors that may impact herbicide dissipation rates (e.g. flow, underwater currents, groundwater inlets, etc.), with wind conditions potentially being one that can be controlled for. For this reason, Onterra recommends spot treatment applications take place during a period of minimal wind if logistically possible. Wind speed and direction data recorded from a weather station near the Pike Chain



of Lakes is investigated in Figure 5.0-1. These data show that winds were relatively low during and immediately after the treatment and likely did not con tribute to quicker herbicide dissipation rates.



The two large herbicide spot treatments in Eagle Lake collectively appeared to have functioned as anticipated as a whole-lake treatment based on the documented EWM reductions lake-wide and the measured herbicide concentrations. The early interval mixed herbicide concentration of the active ingredient came close to the whole-lake calculation of 0.41 ppb and reduced greatly within the first four days after treatment to less than 0.1 ppb As the active ingredient is metabolized and degraded into the acid metabolite, levels increase and peak at roughly 1-7 DAT, where this chemical is degraded over time. At the final monitoring interval on 28 DAT, the concentration of the acid metabolite was around 0.175 ppb. It is unknown how long the acid metabolite remained above detection limits after the final monitoring interval. The initial EWM control results appear extremely promising, but the *year after treatment* results in 2023 will allow for an understanding if the plants were greatly injured for a season (i.e. seasonal impacts) or if the root crowns were indeed controlled and rebound does not occur.

The 2,4-barrier curtain treatment met control expectations with only one EWM plant located within the site during the post-treatment evaluation. It is unclear why the measured 2,4-D concentrations exceeded the target, potentially a result of incomplete mixing into shallow waters and the fact that only one sampling location was detecting this hot spot. 2,4-D concentrations remained above the target for 24 HAT and trended down to about 2.0 ppm ae by the time the curtain was removed at 72 HAT, potentially indicating leakage from the contained area. Following the removal of the curtain, the concentrations greatly were reduced, indicating the curtain functioned as intended to and aided in holding herbicide concentration exposure times longer than would otherwise be achieved. Limited aquatic plant monitoring data collected from within the barrier treatment site did not indicate any detectable negative impacts to native aquatic plant species.

6.0 2023 EWM MANAGEMENT & MONITORING STRATEGY

Few lakes in Bayfield County contain EWM, and the local WDNR has historically supported aggressive management of existing populations assuming this may lessen the chance of EWM spreading within the lake and to other nearby waterbodies. The IRPCLA understands that EWM is established within at least



the six main lakes of the Pike Chain, but wants to continue managing with the goal of maintaining a low lake-wide population within the system. The IRPCLA also wants to minimize areas of dense vegetation that are preferred by largemouth bass species and promote more habitat for walleye and smallmouth bass.

The 2021 *Aquatic Plant Management Plan (Plan)* indicates that when a Late Season EWM Mapping Survey documents colonized EWM populations that are *dominant* or greater in density, an herbicide spot treatment would be considered for the following early-spring. Herbicide spot treatment techniques would be implemented if the colonies have a size/shape/location where management is anticipated to be effective. These treatment design parameters are admittedly vague to account for the evolving research on the subject. While some herbicide spot treatments show promise, the unpredictability of spot treatments state-wide has resulted in less favorability of this strategy with some WDNR regulators and lake managers. This is particularly true in areas of increased water exchange via flow, exposed and offshore EWM colonies, or when traditional weak-acid herbicides like 2,4-D are used. The length of exposure time required for herbicides like 2,4-D are unachievable in most spot treatment scenarios.

6.1 2023 Management Strategy

The IRPCLA is generally pleased with the results of the 2022 treatment strategy, integrating a new herbicide (ProcellaCORTM) and a new technique (deploying a barrier curtain) for the first time. The association intends to build upon lessons learned from this effort for 2023. The IRPCLA has applied for a dovetail WDNR AIS Large Scale Control Grant during the recent cycle to potentially assist with costs associated with the control and monitoring strategy outlined in the remaining portion of this report.

Buskey Bay is currently the lake with the highest EWM population based on the late-summer 2022 EWM mapping survey and is proposed to be targeted with ProcellaCOR[™] spot treatments in 2023 that have the potential for lake/basin-wide impacts. This would be a similar effort to what was employed in 2022 for Eagle Lake. The proposed treatment strategy includes four application areas that total 11.7 acres with a dosing rate of 4.0 PDU's (Map 10). All application area dosing rates are consistent with typical dosing rates being used in EWM spot-treatment designs in Wisconsin. The proposed dosing rate has been confirmed by experts from SePRO, the manufacturer of ProcellaCOR[™]. At the proposed application area, a theoretical lake-wide concentration of 0.47 ppb is calculated, slightly higher than the 2022 Eagle Lake treatment (0.41 ppb). This means that along with the upfront high concentration in the application area, the entire lake would likely reach an equilibrium concentration that at an extended exposure are anticipated to impact EWM and sensitive native species throughout the entire lake. However, measured whole-lake concentrations of florpyrauxifen-benzyl typically fall a little short of predicted levels as the herbicide converts into its acid metabolite form (florpyrauxifen acid) during the time it takes to mix evenly within the lake.

Four sites in Lake Millicent that total 8.9 acres are included in the ProcellaCORTM spot treatment strategy for 2023 (Map 11). Each of these sites have a proposed dosing application rate of 4.5 PDU's, which is slightly higher to account for the relatively small size of the sites and to obtain a little higher level of control than was achieved in the 2022 spot treatments. A theoretical lake-wide concentration of 0.21 ppb is calculated with the treatment design, approximately half the levels of what Onterra would employ for a planned whole-lake treatment. But due to the location of the sites being all on the eastern lobe of Lake Millicent, the potential for this area to also have EWM impacts are high.

A specific area on the north east end of Lake Millicent appears conducive to enclosure with a barrier curtain and treatment with 2,4-D. This proposed treatment site includes a 1.5-acre application area that would be treated with 2,4-D at 4.0 ppm (Map 11). Approximately 400 linear feet of barrier curtain would be required to accommodate this strategy, a similar curtain length that was employed on Twin Bear in 2022. The IRPCLA will apply the knowledge and skills gained from the deployment of a barrier curtain in association with a 2022 treatment to the proposed 2023 strategy.

The professional manual-removal program devised for 2023 will primarily target the 2022 treatment sites for rebounding EWM that was detected, consistent with the IPM framework outlined in the IRPCLA's APM Plan. The IRPCLA has had mixed results with past hand-harvesting efforts, but feels targeting the low population and density of rebounding EWM is better aligned for success than past efforts targeting large and dense colonies. As a part of the current grant-funded project (ACEI-291-22), a budget of 40 hours of removal with Diver Assisted Suction Harvesting (DASH) methods is in place for 2023 and additional efforts could potentially be funded out of pocket if deemed appropriate. There is also the potential that traditional contracted hand-harvesting may be more appropriate for the targeted site, allowing closer to 60 hours of effort for the same budgeted amount. Map 12 displays a preliminary hand harvesting strategy for 2023 that includes targeting two sites in Hart Lake and one site in Twin Bear Lake. All three sites are located within the extents of the 2022 herbicide application areas and are targeting known EWM located in the late-summer 2022 mapping survey or where larger colonies were present prior to the herbicide treatment.

6.1 2023 Monitoring Strategy

Both quantitative and qualitative surveys are incorporated into the IRPCLA's EWM management and monitoring strategy. These data will be collected *prior to treatment, year of treatment*, and *year after treatment*. Onterra believes comparing data from *prior to treatment* to the *year after treatment* allows for the best assessment of the treatment outcome. Many treatment impacts during the *year of treatment* may be short-lived, so understanding how populations stabilize during the *year after treatment* is important within evaluations.

Quantitative Aquatic Plant Monitoring

A preliminary quantitative monitoring plan is being considered for the proposed treatment sites in which a total of 148 sub-sample point-intercept sampling locations are contained within the nine treatment sites (Figure 6.1-1). The quantitative assessment would be completed through the comparison of the sub pointintercept survey from mid-June 2023 (*year of postpretreatment*), late-season 2023 (*year of post-*



treatment), and late-season 2024 (*year after treatment*). The 2023 herbicide treatment is planned for roughly the middle of June. This slight delay in implementation will allow the pretreatment sub-sample



point-intercept survey to take place after many native plants have emerged from winter dormancy and be documented by the pretreatment survey. As previously discussed, this slightly delayed implementation is also favored by WDNR and tribal regulators to avoid sensitive life stages of spawning fish species.

Qualitative EWM Monitoring

A Late Season EWM Mapping Survey would be conducted each year on the six main lakes to produce the mapping data to document a census of the EWM population within the chain at the perceived peak growth stage. Comparing these data to previous surveys will help lake stakeholders understand management outcomes. The EWM mapping data are also valuable to direct follow-up management, such as contracted hand-harvesting, aimed to maintain the gains made. McCarry and Muskellunge Lakes will be surveyed in this fashion periodically, with 2023 being the next scheduled event. The IRPCLA is currently working to secure private access opportunities for Onterra to get their specialized boats on these waterbodies.

Herbicide Concentration Monitoring

IRPCLA volunteers would collect herbicide concentration monitoring during the hours/days following treatment following a sampling regime created through collaborative efforts of the WDNR and Onterra. The 2023 monitoring structure would investigate concentrations within direct application areas as well as lake-wide to understand whether the cumulative ProcellaCORTM spot treatments function as whole-lake treatments.

Whole-Lake Point-Intercept Survey

Additional aquatic plant monitoring is planned in 2023 through the completion of a whole-lake pointintercept surveys on the six main lakes. The whole-lake point-intercept survey will be 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. This would be a 5-year interval between surveys, which is outlined as a target within the IRPCLA's Plan and required for future eligibility for grant funds.















Priet Location in Wiscomit Exem of large map down in ref	
Notes 370 L S Feet Eurasian watermilfoil Su Notes Highly Scattered Sis Prosper Rd Bathymetry: Onerra De Pere, WI 54115 Sources Surces Nources Aquatic Plants: Onerra Dominant Aquatic Plants: Onerra Acco.com Map Date: October 14th, 2022 KLW Weitename: PKChar_EWMPB_BuskeyBay_Sept22.mxd Surface Matting (No	egend rvey: September 6th - 7th, 2022 ● Single or Few Plants ● Clumps of Plants ● Small Plant Colony ■ Public Boat Landing ne)

A

APPENDIX A

Twin Bear Lake Final 2,4-D Sampling Plan

Twin Bear Lake, Pike Chain, Bayfield County (WBIC:2903100) 2022 Herbicide Sample Plan Onterra, LLC

Twin Bear Lake, located within the Pike Chain of Lakes in Bayfield County, is a 157-acre drainage lake that has a maximum depth of 59 feet. Liquid 2,4-D is proposed to be applied to approximately 3.4 acres on the west end of the lake in spring of 2022 to control Eurasian watermilfoil. A barrier curtain will be used around the perimeter of the treatment area in an attempt to maintain desired herbicide concentration levels. Herbicide concentration sampling will be conducted in order to monitor the herbicide concentrations in the days following the application.

Water samples will need to be collected at the sites and depths listed below. Data are in decimal degrees and the datum is WGS84. Locations of each sampling site are displayed with green squares on the image below.

Twin Bear Lake Herbicide Sample Sites									
Site Label	Site Description	Station ID	Latitude	Longitude	Sample Depth				
TB1	Application Area TB A-22	10056181	46.50356	-91.37323	Integrated (0-6 feet)				
TB2	Application Area TB A-22	10056183	46.50209	-91.37391	Integrated (0-6 feet)				
TB3	Outside Application Area	10056184	46.50336	-91.37279	Integrated (0-6 feet)				
TB4	Outside Application Area	10056185	46.50207	-91.37340	Integrated (0-6 feet)				
TB5	Deep Hole	043127	46.50590	-91.36727	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 12 total intervals. Five sampling intervals are scheduled to take place before curtain removal and are referred to as Hours After Treatment (HAT). The remaining seven 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)										
Interval	Application	Area TB A-22	Outside Application Area							
mervar	Site TB1	SiteTB2	Site TB3	Site TB4	Site TB5-Deep Hole					
Herbicide Application Complete										
1 HAT	Х	Х	Х	Х						
6 HAT	Х	Х	Х	Х						
24 HAT	Х	Х	Х	Х						
48 HAT	Х	Х	Х	Х						
72 HAT	Х	Х	Х	Х						
		Barrier Cu	tain Removed							
1 HAC	Х	Х	Х	Х						
3 HAC	Х	Х	Х	Х						
6 HAC	Х	Х	Х	Х						
12 HAC	Х	Х	Х	Х	Х					
24 HAC	Х	Х	Х	Х	Х					
48 HAC					Х					
72 HAC					Х					
	HAT = Hou	irs After Treatme	ent, HAC = Hour	s 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>

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.

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

Project specifics, logistics and sampling methods			
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Twin Bear Lake, Bayfield County Herbicide Sampling Data Sheets, 2022

Account number:	349452	
DNR User ID:	TOSHNP	

Sample Matrix:Surface Water (SU)Project:Grant #

WBIC:

2903100

Collector Name:	
Phone Number:	

Test Requested: 2,4-D herbicide

Sample Inter	val:					
Site	Station ID	Sample Depth	Date	Time (24:00)	Water Temp in C (3 foot depth)	Wind Direction and Speed
TB1	10056181	Integrated (0-6 ft)				
TB2	10056183	Integrated (0-6 ft)				
TB3	10056184	Integrated (0-6 ft)				
TB4	10056185	Integrated (0-6 ft)				
TB5	043127	Integrated (0-6 ft)				

B

APPENDIX B

Eagle Lake Final ProcellaCOR™ Sampling Plan

Eagle Lake, Bayfield County (WBIC:2902900) 2022 Herbicide Sample Plan Onterra, LLC

Eagle Lake, a lake within the Pike Chain of Lakes in Bayfield County, is an approximately 163acre drainage lake that has a maximum depth of 52 feet. Florpyrauxifen-benzyl (commercially as ProcellaCORTM) is proposed to be applied to 15.3 non-contiguous acres in early-summer 2022 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. Data are in decimal degrees and the datum is WGS84. Locations of each sampling site are displayed with green squares on the image below.

Eagle Lake Herbicide Sample Sites					
Site Label	Site Description	Station ID	Latitude	Longitude	Sample Depth
E1	Application Area E-22	10056175	46.49733	-91.35830	Integrated (0-6 feet)
E2	Application area F-22	10052499	46.50113	-91.35544	Integrated (0-6 feet)
E3	Deep Hole	043077	46.49831	-91.35918	Integrated (0-6 feet)
F1	Flynn Lake-Deep Hole	43078	46.49094	-91.34872	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 E1 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, 25 additional samples will need to be collected at nine 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)				
	Application Area		Deep Hole	Flynn Lake
Interval	Site E1	Site E2	Site E3	Site F1
Pre-Treatment	Х			
3 HAT	Х	Х		
9 HAT	Х	Х	Х	
24 HAT	Х	Х	Х	
48 HAT	Х	Х	Х	
4 DAT	Х	Х	Х	
7 DAT	Х	Х	Х	Х
14 DAT	Х	Х	Х	
21 DAT			Х	Х
28 DAT			Х	X
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: E1, E2, or E3)
- Date sample is collected

When all sampling is complete, the water samples **and** Chain of Custody Datasheets should be shipped by overnight currier to:

EPL Bio Analytical Services 9095 W. Harristown Blvd. Niantic, IL 62551

Samples should <u>not</u> be shipped on loose ice. Ice packs or frozen water bottles (contained in a zip bag) may be shipped with the samples to keep them cool. Samples should not be shipped on a Friday, but rather refrigerated and shipped on the following Monday.

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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