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)							
	Applicat	ion 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 Tre	atment, DAT = L	Days After Treati	ment			

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, logistic	Project specifics, logistics and sampling methods				
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2.0 EAGLE LAKE 2022 PROCELLACOR™ TREATMENT

2.5 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.

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 (SX-1552-A) which is the immediate by-product that it breaks down into. Studies have indicated the acid derivative of florpyrauxifen-benzyl to be active on EWM at some concentrations albeit to a lesser degree than the primary active ingredient.

Figures 1-2 and Table 1 display 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 1. Figure 2 shows the same data as Figure 1, 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 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 herbicide movement. 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.



202	2022 ProcellaCOR™ herbicide treatment in Eagle Lake.									
	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

Table 1. Florpyrauxifen-benzyl (SX-1552) concentrations at four monitoring locations following a June







Concentrations of the acid metabolite (SX-1552-A) are displayed on Table 2 and Figure 3. The measured concentrations of the acid metabolite were variable and ranged from below detection limits to approximately 0.235 ppb in all samples. Note that the y-axis on Figure 2 extends to 0.1 ppb so that the data can be more easily viewed and is a different axis height than Figure 1 (0.5ppb).

locations within Eagle Lake and Flynn Lake following a June 2022 ProcellaCOR™ herbicide treatment.									
Florpyrauxifen-benzyl acid metabolite (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









2590-X549	E3 - 21 DAT	2	1	0.0120	0.0108	<lod< th=""></lod<>
2590-X550	F1 - 21 DAT	2	1	0.000	0.000	<lod< td=""></lod<>
2590-X551	E3 - 28 DAT	2	1	0.114	0.102	LOD <loq< td=""></loq<>
2590-X552	F1 - 28 DAT	2	1	0.000	0.000	<lod< td=""></lod<>
Recovery Calculati						
Recovery Galculat	Client	Valuma	Culture Colo	Fartification	1	Moon
Lab	Client	Volume Spiking	Spiking Soln.	Fortification	Recovery	Mean Recovery
Lab Sample ID	Client Sample ID	Volume Spiking Soln (mL)	Spiking Soln. Concn. (ng/mL)	Fortification Level (ng/mL)	Recovery (%)	Mean Recovery %
Lab Sample ID 2590-X527-S1	Client Sample ID E1 - Pre	Volume Spiking Soln (mL) 0.020	Spiking Soln. Concn. (ng/mL) 10.000	Fortification Level (ng/mL) 0.200	Recovery (%) 116	Mean Recovery %

EPL Bio-Analytical Services (EPL-BAS) EPL-BAS Study No. 625N2590 Study Title: Determinatinon of SX-1552 and SX-1552 Acid Metabolite in Surface Water

Analyte:	SX-1552-Acid	Location:	Eagle Lake
Extraction Set:	W046	Matrix:	Water
Analyst Set:	W046	Method LOQ:	0.500 ng/mL
Internal Standard (IS):	IS-SX-1552-Acid	IS Concentration:	0.500 ng/mL
Instrument:	UPLC #23	MRM Transition:	349/268

Standard Information	tion							
		Nominal	Analyte	Internal	Calculated	Relative	Standard	
Injection	Lab	Standard	Peak	Standard	Concn.	Error	Excluded	Reason
#	Standard ID	Concn. (ng/mL)	Area	Peak Area	(ng/mL)	Accuracy (%)	(x)	(High/Low)
1	2022-1008	0.050	273.716	119.998	0.055	10.0		
6	2022-1009	0.150	373.109	133.533	0.149	-0.667		
14	2202-1010	0.500	623.091	156.863	0.364	-27.2		
23	2022-1011	1.000	932.063	130.952	0.937	-6.30		
29	2022-1012	10.000	7357.809	134.616	9.61	-3.90		
35	2022-1013	50.000	34555.355	124.070	50.449	0.898		
Linear Regression	Equation:	2.74077 * x + 0.989569						
Weighting:		1/x						
Correlation Coeffic	Correlation Coefficient (r): 0.							
Coefficient of Dete	rmination (r ²) =	0.99961						

Sample Information

		Client			Analyte	Internal	Amount
Injection	Lab	Sample	Date(s)	Date(s)	Peak	Standard	Found
#	Sample ID	ID	Extracted	Analyzed	Area	Peak Area	(ng/mL)
2	Blank-W046	NA	8/9/2022	8/9/2022	239.411	134.858	0.000
3	2590-X527	E1 - Pre	8/9/2022	8/9/2022	277.016	142.287	0.000
4	2590-X527-S1 1	E1 - Pre	8/9/2022	8/9/2022	433.979	124.820	0.273
5	2590-X527-S2 2	E1 - Pre	8/9/2022	8/9/2022	2044.801	119.500	2.761
7	2590-X528	E1 - 3 HAT	8/9/2022	8/9/2022	419.419	131.753	0.220
8	2590-X529	E2 - 3 HAT	8/9/2022	8/9/2022	384.858	132.102	0.170
9	2590-X530	E1 - 9 HAT	8/9/2022	8/9/2022	416.949	137.199	0.193
10	2590-X531	E2 - 9 HAT	8/9/2022	8/9/2022	298.369	133.880	0.046
11	2590-X532	E3 - 9 HAT	8/9/2022	8/9/2022	291.213	139.529	0.020
12	2590-X533	E1 - 24 HAT	8/9/2022	8/9/2022	365.242	137.083	0.125
13	2590-X534	E2 - 24 HAT	8/9/2022	8/9/2022	293.177	132.910	0.041
15	2590-X535	E3 - 24 HAT	8/9/2022	8/9/2022	284.072	139.570	0.010
16	2590-X536	E1 - 48 HAT	8/9/2022	8/9/2022	333.683	150.267	0.044
17	2590-X537	E2 - 48 HAT	8/9/2022	8/9/2022	334.361	123.406	0.133
18	2590-X538	E3 - 48 HAT	8/9/2022	8/9/2022	279.413	138.962	0.006
19	2590-X539	E1 - 4 DAT	8/9/2022	8/9/2022	315.963	121.518	0.113
20	2590-X540	E2 - 4 DAT	8/9/2022	8/9/2022	353.201	124.165	0.158
21	2590-X541	E3 - 4 DAT	8/9/2022	8/9/2022	335.531	132.528	0.101
22	2590-X542	E1 - 7 DAT	8/9/2022	8/9/2022	319.652	144.529	0.042
24	2590-X543	E2 - 7 DAT	8/9/2022	8/9/2022	337.476	134.226	0.098
25	2590-X544	E3 - 7 DAT	8/9/2022	8/9/2022	317.776	109.452	0.169
26	2590-X545	F1 - 7 DAT	8/9/2022	8/9/2022	275.997	131.523	0.022
27	2590-X546	E1 - 14 DAT	8/9/2022	8/9/2022	300.628	117.956	0.104
28	2590-X547	E2 - 14 DAT	8/9/2022	8/9/2022	323.998	130.018	0.094
30	2590-X548	E3 - 14 DAT	8/9/2022	8/9/2022	328.837	129.226	0.103
31	2590-X549	E3 - 21 DAT	8/9/2022	8/9/2022	298.186	145.411	0.013
32	2590-X550	F1 - 21 DAT	8/9/2022	8/9/2022	274.165	119.281	0.058
33	2590-X551	E3 - 28 DAT	8/9/2022	8/9/2022	308.503	123.098	0.096
34	2590-X552	F1 - 28 DAT	8/9/2022	8/10/2022	270.406	146.954	0.000

Residue Calculation

	Client		Sample	Sample	Corrected	
Lab	Sample	Dilution	Volume	Concn.	Sample	
Sample ID	ID	Factor	(mL)	(ng/mL)	Concn. (ng/mL)	Flags
2590-X527	E1 - Pre	2	1	0.000	0.000	<lod< td=""></lod<>
2590-X527-S1 1	E1 - Pre	2	1	0.546	-	
2590-X527-S2 2	E1 - Pre	2	1	5.52	-	
2590-X528	E1 - 3 HAT	2	1	0.440	0.401	LOD <loq< td=""></loq<>
2590-X529	E2 - 3 HAT	2	1	0.340	0.310	LOD <loq< td=""></loq<>
2590-X530	E1 - 9 HAT	2	1	0.386	0.351	LOD <loq< td=""></loq<>
2590-X531	E2 - 9 HAT	2	1	0.0920	0.0838	<lod< td=""></lod<>
2590-X532	E3 - 9 HAT	2	1	0.0400	0.0364	<lod< td=""></lod<>
2590-X533	E1 - 24 HAT	2	1	0.250	0.228	LOD <loq< td=""></loq<>
2590-X534	E2 - 24 HAT	2	1	0.0820	0.0747	<lod< td=""></lod<>
2590-X535	E3 - 24 HAT	2	1	0.0200	0.0182	<lod< td=""></lod<>
2590-X536	E1 - 48 HAT	2	1	0.0880	0.0801	<lod< td=""></lod<>
2590-X537	E2 - 48 HAT	2	1	0.266	0.242	LOD <loq< td=""></loq<>
2590-X538	E3 - 48 HAT	2	1	0.0120	0.0109	<lod< td=""></lod<>
2590-X539	E1 - 4 DAT	2	1	0.226	0.206	LOD <loq< td=""></loq<>
2590-X540	E2 - 4 DAT	2	1	0.316	0.288	LOD <loq< td=""></loq<>
2590-X541	E3 - 4 DAT	2	1	0.202	0.184	LOD <loq< td=""></loq<>
2590-X542	E1 - 7 DAT	2	1	0.0840	0.0765	<lod< td=""></lod<>
2590-X543	E2 - 7 DAT	2	1	0.196	0.178	LOD <loq< td=""></loq<>
2590-X544	E3 - 7 DAT	2	1	0.338	0.308	LOD <loq< td=""></loq<>
2590-X545	F1 - 7 DAT	2	1	0.0440	0.0401	<lod< td=""></lod<>
2590-X546	E1 - 14 DAT	2	1	0.208	0.189	LOD <loq< td=""></loq<>
2590-X547	E2 - 14 DAT	2	1	0.188	0.171	LOD <loq< td=""></loq<>
2590-X548	E3 - 14 DAT	2	1	0.206	0.188	LOD <loq< td=""></loq<>
2590-X549	E3 - 21 DAT	2	1	0.0260	0.0237	<lod< td=""></lod<>
2590-X550	F1 - 21 DAT	2	1	0.116	0.106	<lod< td=""></lod<>
2590-X551	E3 - 28 DAT	2	1	0.192	0.175	LOD <loq< td=""></loq<>
2590-X552	F1 - 28 DAT	2	1	0.000	0.000	<lod< td=""></lod<>

Recovery Calculation

Lab Sample ID	Client Sample ID	Volume Spiking Soln (mL)	Spiking Soln. Concn. (ng/mL)	Fortification Level (ng/mL)	Recovery (%)	Mean Recovery %
2590-X527-S1 1	E1 - Pre	0.050	10.000	0.500	109	110
2590-X527-S2 2	E1 - Pre	0.005	1000.000	5.00	110	110

Compound Summary Report

Tue Aug 23 07:33:56 2022

npound 1: IS-SX1552

	File Name	Sample ID	Sample Type	Std. Conc
1	080922-01	2022-1008	Standard	0.5
2	080922-02	Blank-W046	Blank	0.5
3	080922-03	2590-X527	Analyte	0.5
4	080922-04	2590-X527-S1	Analyte	0.5
5	080922-05	2590-X527-S2	Analyte	0.5
6	080922-06	2022-1009	Standard	0.5
7	080922-07	2590-X528	Analyte	0.5
8	080922-08	2590-X529	Analyte	0.5
9	080922-09	2590-X530	Analyte	0.5
10	080922-10	2590-X531	Analyte	0.5
11	080922-11	2590-X532	Analyte	0.5
12	080922-12	2590-X533	Analyte	0.5
13	080922-13	2590-X534	Analyte	0.5
14	080922-14	2022-1010	Standard	0.5
15	080922-15	2590-X535	Analyte	0.5
16	080922-16	2590-X536	Analyte	0.5
17	080922-17	2590-X537	Analyte	0.5
18	080922-18	2590-X538	Analyte	0.5
19	080922-19	2590-X539	Analyte	0.5
20	080922-20	2590-X540	Analyte	0.5
21	080922-21	2590-X541	Analyte	0.5
22	080922-22	2590-X542	Analyte	0.5
23	080922-23	2022-1011	Standard	0.5
24	080922-24	2590-X543	Analyte	0.5
25	080922-25	2590-X544	Analyte	0.5
26	080922-26	2590-X545	Analyte	0.5
27	080922-27	2590-X546	Analyte	0.5
28	080922-28	2590-X547	Analyte	0.5
29	080922-29	2022-1012	Standard	0.5
30	080922-30	2590-X548	Analyte	0.5
31	080922-31	2590-X549	Analyte	0.5
32	080922-32	2590-X550	Analyte	0.5
33	080922-33	2590-X551	Analyte	0.5
34	080922-34	2590-X552	Analyte	0.5
35	080922-35	2022-1013	Standard	0.5

ipound 2: SX-1552 (Q)

	File Name	Sample ID	Sample Type	Std. Conc
1	080922-01	2022-1008	Standard	0.05

2	080922-02	Blank-W046	Blank	
3	080922-03	2590-X527	Analyte	
4	080922-04	2590-X527-S1	Analyte	
5	080922-05	2590-X527-S2	Analyte	
6	080922-06	2022-1009	Standard	0.15
7	080922-07	2590-X528	Analyte	
8	080922-08	2590-X529	Analyte	
9	080922-09	2590-X530	Analyte	
10	080922-10	2590-X531	Analyte	
11	080922-11	2590-X532	Analyte	
12	080922-12	2590-X533	Analyte	
13	080922-13	2590-X534	Analyte	
14	080922-14	2022-1010	Standard	0.5
15	080922-15	2590-X535	Analyte	
16	080922-16	2590-X536	Analyte	
17	080922-17	2590-X537	Analyte	
18	080922-18	2590-X538	Analyte	
19	080922-19	2590-X539	Analyte	
20	080922-20	2590-X540	Analyte	
21	080922-21	2590-X541	Analyte	
22	080922-22	2590-X542	Analyte	
23	080922-23	2022-1011	Standard	1
24	080922-24	2590-X543	Analyte	
25	080922-25	2590-X544	Analyte	
26	080922-26	2590-X545	Analyte	
27	080922-27	2590-X546	Analyte	
28	080922-28	2590-X547	Analyte	
29	080922-29	2022-1012	Standard	10
30	080922-30	2590-X548	Analyte	
31	080922-31	2590-X549	Analyte	
32	080922-32	2590-X550	Analyte	
33	080922-33	2590-X551	Analyte	
34	080922-34	2590-X552	Analyte	
35	080922-35	2022-1013	Standard	50
וpound 3: Sx-155	2 (C)			
	File Name	Sample ID	Sample Type	Std Cor

	File Name	Sample ID	Sample Type	Std. Conc
1	080922-01	2022-1008	Standard	0.05
2	080922-02	Blank-W046	Blank	
3	080922-03	2590-X527	Analyte	
4	080922-04	2590-X527-S1	Analyte	
5	080922-05	2590-X527-S2	Analyte	
6	080922-06	2022-1009	Standard	0.15
7	080922-07	2590-X528	Analyte	
8	080922-08	2590-X529	Analyte	
9	080922-09	2590-X530	Analyte	

10	080922-10	2590-X531	Analyte	
11	080922-11	2590-X532	Analyte	
12	080922-12	2590-X533	Analyte	
13	080922-13	2590-X534	Analyte	
14	080922-14	2022-1010	Standard	0.5
15	080922-15	2590-X535	Analyte	
16	080922-16	2590-X536	Analyte	
17	080922-17	2590-X537	Analyte	
18	080922-18	2590-X538	Analyte	
19	080922-19	2590-X539	Analyte	
20	080922-20	2590-X540	Analyte	
21	080922-21	2590-X541	Analyte	
22	080922-22	2590-X542	Analyte	
23	080922-23	2022-1011	Standard	1
24	080922-24	2590-X543	Analyte	
25	080922-25	2590-X544	Analyte	
26	080922-26	2590-X545	Analyte	
27	080922-27	2590-X546	Analyte	
28	080922-28	2590-X547	Analyte	
29	080922-29	2022-1012	Standard	10
30	080922-30	2590-X548	Analyte	
31	080922-31	2590-X549	Analyte	
32	080922-32	2590-X550	Analyte	
33	080922-33	2590-X551	Analyte	
34	080922-34	2590-X552	Analyte	
35	080922-35	2022-1013	Standard	50

mpound 4: IS-1552-A

	File Name	Sample ID	Sample Type	Std. Conc
2	080922-02	Blank-W046	Blank	0.5
3	080922-03	2590-X527	Analyte	0.5
4	080922-04	2590-X527-S1	Analyte	0.5
5	080922-05	2590-X527-S2	Analyte	0.5
7	080922-07	2590-X528	Analyte	0.5
8	080922-08	2590-X529	Analyte	0.5
9	080922-09	2590-X530	Analyte	0.5
10	080922-10	2590-X531	Analyte	0.5
11	080922-11	2590-X532	Analyte	0.5
12	080922-12	2590-X533	Analyte	0.5
13	080922-13	2590-X534	Analyte	0.5
15	080922-15	2590-X535	Analyte	0.5
16	080922-16	2590-X536	Analyte	0.5
17	080922-17	2590-X537	Analyte	0.5
18	080922-18	2590-X538	Analyte	0.5
19	080922-19	2590-X539	Analyte	0.5
20	080922-20	2590-X540	Analyte	0.5

080922-21	2590-X541	Analyte	0.5
080922-22	2590-X542	Analyte	0.5
080922-24	2590-X543	Analyte	0.5
080922-25	2590-X544	Analyte	0.5
080922-26	2590-X545	Analyte	0.5
080922-27	2590-X546	Analyte	0.5
080922-28	2590-X547	Analyte	0.5
080922-30	2590-X548	Analyte	0.5
080922-31	2590-X549	Analyte	0.5
080922-32	2590-X550	Analyte	0.5
080922-33	2590-X551	Analyte	0.5
080922-34	2590-X552	Analyte	0.5
	080922-21 080922-22 080922-24 080922-25 080922-26 080922-27 080922-28 080922-30 080922-31 080922-32 080922-33 080922-34	080922-212590-X541080922-222590-X542080922-242590-X543080922-252590-X544080922-262590-X545080922-272590-X546080922-282590-X547080922-302590-X548080922-312590-X549080922-322590-X550080922-332590-X551080922-342590-X552	080922-212590-X541Analyte080922-222590-X542Analyte080922-242590-X543Analyte080922-252590-X544Analyte080922-262590-X545Analyte080922-272590-X546Analyte080922-282590-X547Analyte080922-302590-X548Analyte080922-312590-X549Analyte080922-322590-X550Analyte080922-332590-X551Analyte080922-342590-X552Analyte

npound 5: 1552-A (Q)

	File Name	Sample ID	Sample Type	Std. Conc
1	080922-01	2022-1008	Standard	0.05
2	080922-02	Blank-W046	Blank	
3	080922-03	2590-X527	Analyte	
4	080922-04	2590-X527-S1	Analyte	
5	080922-05	2590-X527-S2	Analyte	
6	080922-06	2022-1009	Standard	0.15
7	080922-07	2590-X528	Analyte	
8	080922-08	2590-X529	Analyte	
9	080922-09	2590-X530	Analyte	
10	080922-10	2590-X531	Analyte	
11	080922-11	2590-X532	Analyte	
12	080922-12	2590-X533	Analyte	
13	080922-13	2590-X534	Analyte	
14	080922-14	2022-1010	Standard	0.5
15	080922-15	2590-X535	Analyte	
16	080922-16	2590-X536	Analyte	
17	080922-17	2590-X537	Analyte	
18	080922-18	2590-X538	Analyte	
19	080922-19	2590-X539	Analyte	
20	080922-20	2590-X540	Analyte	
21	080922-21	2590-X541	Analyte	
22	080922-22	2590-X542	Analyte	
23	080922-23	2022-1011	Standard	1
24	080922-24	2590-X543	Analyte	
25	080922-25	2590-X544	Analyte	
26	080922-26	2590-X545	Analyte	
27	080922-27	2590-X546	Analyte	
28	080922-28	2590-X547	Analyte	
29	080922-29	2022-1012	Standard	10
30	080922-30	2590-X548	Analyte	
31	080922-31	2590-X549	Analyte	

32	080922-32	2590-X550	Analyte	
33	080922-33	2590-X551	Analyte	
34	080922-34	2590-X552	Analyte	
35	080922-35	2022-1013	Standard	50

npound 6: 1552-A (C)

	File Name	Sample ID	Sample Type	Std. Conc
1	080922-01	2022-1008	Standard	0.05
2	080922-02	Blank-W046	Blank	
3	080922-03	2590-X527	Analyte	
4	080922-04	2590-X527-S1	Analyte	
5	080922-05	2590-X527-S2	Analyte	
6	080922-06	2022-1009	Standard	0.15
7	080922-07	2590-X528	Analyte	
8	080922-08	2590-X529	Analyte	
9	080922-09	2590-X530	Analyte	
10	080922-10	2590-X531	Analyte	
11	080922-11	2590-X532	Analyte	
12	080922-12	2590-X533	Analyte	
13	080922-13	2590-X534	Analyte	
14	080922-14	2022-1010	Standard	0.5
15	080922-15	2590-X535	Analyte	
16	080922-16	2590-X536	Analyte	
17	080922-17	2590-X537	Analyte	
18	080922-18	2590-X538	Analyte	

RT	Area	Detection Flags	Conc.	%Dev
5.26	2556.755	MM	0.51	2.1
5.26	2397.435	MM	0.478	-4.3
5.26	2356.757	MM	0.47	-5.9
5.26	2249.988	MM	0.449	-10.2
5.26	1989.039	bb	0.397	-20.6
5.26	2442.734	MM	0.488	-2.5
5.26	2438.916	MM	0.487	-2.6
5.26	2344.3	MM	0.468	-6.4
5.26	2373.423	MM	0.474	-5.3
5.26	2355.257	bb	0.47	-6
5.25	2360.722	MM	0.471	-5.8
5.25	2412.459	MM	0.481	-3.7
5.26	2461.426	MM	0.491	-1.7
5.26	2405.257	MM	0.48	-4
5.26	2378.653	MM	0.475	-5.1
5.26	2421.394	bb	0.483	-3.3
5.26	2170.231	bb	0.433	-13.4
5.26	2080.186	bb	0.415	-17
5.25	2431.575	MM	0.485	-2.9
5.26	2408.965	MM	0.481	-3.8
5.25	2190.437	bb	0.437	-12.6
5.25	2372.844	bb	0.474	-5.3
5.26	2419.802	bb	0.483	-3.4
5.25	2353.105	bb	0.47	-6.1
5.25	2274.581	bb	0.454	-9.2
5.25	2359.167	bb	0.471	-5.8
5.25	2259.885	bb	0.451	-9.8
5.25	2250.812	bb	0.449	-10.2
5.26	2465.959	MM	0.492	-1.6
5.25	2279.111	bb	0.455	-9
5.25	2294.784	bb	0.458	-8.4
5.25	2281.822	bb	0.455	-8.9
5.25	2215.134	bb	0.442	-11.6
5.25	2308.617	bb	0.461	-7.8
5.25	2740.967	bb	0.547	9.4

RT	Area	Detection Flags	Conc.	%Dev
5.26	114.439	MM	0.056	11.1

5.26	80.92	bbl		
5.27	79.331	MMI		
5.27	121.848	MM	0.116	
5.26	400.561	bb	1.069	
5.26	139.361	MM	0.135	-9.9
5.26	696.963	bb	1.616	
5.26	1139.121	bb	2.911	
5.26	279.855	bb	0.529	
5.27	179.003	bb	0.258	
5.27	157.789	bb	0.198	
5.27	139.584	bb	0.14	
5.26	156.708	bb	0.178	
5.26	276.674	bb	0.51	2.1
5.26	112.373	bb	0.072	
5.26	119.833	bb	0.086	
5.26	96.606	bb	0.054	
5.27	110.227	bb	0.109	
5.27	103.073	bb	0.04	
5.26	90.618	bb	0.009	
5.28	82.847	bb	0.011	
5.26	72.168	bbl		
5.26	440.356	MM	0.944	-5.6
5.27	82.679	bbl		
5.27	87.109	MM	0.014	
5.25	109.315	bb	0.066	
5.31	83.469	bb	0.005	
5.27	72.62	bbl		
5.26	4007.545	MM	10.286	2.9
5.27	78.683	bbl		
5.27	85.157	bb	0.006	
5.26	76.19	bbl		
5.26	99.79	MM	0.057	
5.28	82.797	bbl		
5.25	21172.391	bb	49.769	-0.5

Area	Detection Flags	Conc.	%Dev
954.178	MM	0.053	6.3
672.735	bbl		
658.75	bbl		
969.763	bb	0.098	
3017.086	bb	0.944	
1201.399	bb	0.146	-2.9
5702.088	bb	1.584	
9362.893	bb	2.874	
2207.336	bb	0.487	
	Area 954.178 672.735 658.75 969.763 3017.086 1201.399 5702.088 9362.893 2207.336	AreaDetection Flags954.178MM672.735bbl658.75bbl969.763bb3017.086bb1201.399bb5702.088bb9362.893bb2207.336bb	AreaDetection FlagsConc.954.178MM0.053672.735bbl-658.75bbl-969.763bb0.0983017.086bb0.9441201.399bb0.1465702.088bb1.5849362.893bb2.8742207.336bb0.487

5.26	1546.871	MM	0.274	
5.26	1087.028	bb	0.121	
5.26	1187.214	bb	0.146	
5.26	1124.189	bb	0.118	
5.26	2367.575	bb	0.529	5.9
5.26	924.522	bb	0.065	
5.26	1068.67	bb	0.106	
5.26	807.917	bb	0.052	
5.27	844.79	bb	0.079	
5.26	678.308	bbl		
5.26	780.31	bb	0.015	
5.26	771.89	bb	0.037	
5.26	704.891	MMI		
5.26	3555.836	MM	0.907	-9.3
5.27	775.556	bb	0.019	
5.26	691.259	bbl		
5.26	747.536	bb	0.009	
5.26	635.96	bbl		
5.26	740.341	bb	0.019	
5.26	32343.334	MM	9.982	-0.2
5.26	730.117	bb	0.012	
5.26	675.51	MMI		
5.26	719.353	bb	0.008	
5.26	659.937	bbl		
5.26	778.205	bb	0.025	
5.25	177005.125	bb	50.082	0.2

RT	Area	Detection Flags	Conc.	%Dev
4.27	134.858	bb	0.506	1.1
4.26	142.287	bb	0.534	6.7
4.26	124.82	bb	0.468	-6.4
4.26	119.5	MM	0.448	-10.4
4.25	131.753	bb	0.494	-1.2
4.26	132.102	bb	0.495	-0.9
4.26	137.199	bb	0.514	2.9
4.25	133.88	bb	0.502	0.4
4.26	139.529	bb	0.523	4.6
4.26	137.083	bb	0.514	2.8
4.26	132.91	bb	0.498	-0.3
4.26	139.57	bb	0.523	4.7
4.26	150.267	bb	0.563	12.7
4.26	123.406	bb	0.463	-7.4
4.25	138.962	bb	0.521	4.2
4.25	121.518	bb	0.456	-8.9
4.26	124.165	bb	0.466	-6.9

4.26	132.528	bb	0.497	-0.6
4.27	144.529	bb	0.542	8.4
4.27	134.226	bb	0.503	0.7
4.26	109.452	bb	0.41	-17.9
4.25	131.523	bb	0.493	-1.4
4.25	117.956	bb	0.442	-11.5
4.26	130.018	bb	0.488	-2.5
4.25	129.226	bb	0.485	-3.1
4.26	145.411	bb	0.545	9.1
4.26	119.281	MM	0.447	-10.5
4.26	123.098	bb	0.462	-7.7
4.26	146.954	bb	0.551	10.2

RT	Area	Detection Flags	Conc.	%Dev
4.26	273.716	MM	0.055	10.1
4.26	239.411	MMI		
4.28	277.016	bbl		
4.26	433.979	bb	0.273	
4.26	2044.801	bb	2.761	
4.26	373.109	MM	0.149	-0.9
4.26	419.419	MM	0.22	
4.26	384.858	bb	0.17	
4.26	416.949	bb	0.193	
4.26	298.369	bb	0.046	
4.27	291.213	bb	0.02	
4.26	365.242	bb	0.125	
4.25	293.177	bb	0.041	
4.27	623.091	MMX	0.364	-27.3
4.27	284.072	bb	0.01	
4.26	333.683	bb	0.044	
4.26	334.361	bb	0.133	
4.27	279.413	bb	0.006	
4.26	315.963	bb	0.113	
4.26	353.201	bb	0.158	
4.27	335.531	bb	0.101	
4.25	319.652	bb	0.042	
4.26	932.063	MM	0.937	-6.3
4.25	337.476	bb	0.098	
4.26	317.776	bb	0.169	
4.26	275.997	bb	0.022	
4.25	300.628	MM	0.104	
4.26	323.998	bb	0.094	
4.26	7357.809	MM	9.61	-3.9
4.26	328.837	bb	0.103	
4.26	298.186	bb	0.013	

4.26	274.165	bb	0.058	
4.25	308.503	bb	0.096	
4.26	270.406	bbl		
4.26	34555.355	MM	50.449	0.9

RT	Area	Detection Flags	Conc.	%Dev
4.26	213.43	MM	0.078	55.1
4.28	204.137	bb	0.011	
4.29	189.397	bbl		
4.26	308.486	MM	0.251	
4.27	1504.522	MM	2.788	
4.26	268.321	MM	0.135	-9.7
4.28	296.531	bb	0.196	
4.26	307.964	bb	0.216	
4.26	315.732	bb	0.209	
4.27	235.846	bb	0.073	
4.26	212.153	bb	0.013	
4.26	244.681	bb	0.079	
4.25	226.674	bb	0.059	
4.26	433.533	MM	0.324	-35.1
4.27	211.193	bb	0.011	
4.27	243.524	MM	0.038	
4.26	243.334	bb	0.126	
4.25	230.013	bb	0.047	

2.0 TWIN BEAR LAKE 2022 2,4-D HERBICIDE TREATMENT

2.5 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).

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 400 linear feet of curtain. The application rate remained constant at 4.0 ppm ae, but the gallons of product required to reach that concentration in the reduced application area was 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 6,24,48,72, 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 sample were preserved then sent to the State Lab of Hygiene for analysis.

Figure 1 and Table 1 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.

	Hou	rs after Tre	atment			Но	ours afte	r Curtain	Remov	al
	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
ND : Not Detected							_			

 Table 1. Twin Bear Lake 2022 2,4-D Concentration Monitoring Results from two locations – one within and one outside a barrier curtain. Values in parts per million (ppm).







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	0	utside Application Area		
mervar	Site TB1	SiteTB2	Site TB3	Site TB4	Site TB5-Deep Hole	
		Herbicide App	lication Comple	ete		
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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WDNR	Support			
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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 Interval:							
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)					

