Herbicide Treatment Analysis for *Potamogeton crispus*

Bone Lake, Polk County WI

2017

Surveys, mapping and analysis provided by: Ecological Integrity Service, LLC Amery, Wisconsin

Abstract

An herbicide treatment targeting the AIS *Potamogeton crispus* (curly leaf pondweed or CLP) was conducted in Bone Lake, Polk County on May 3 and May 9 of 2017. The treatment occurred in 7 beds totaling 30.66 acres. The treatment was evaluated by conducting a pretreatment survey on April 8, 2017 and a post treatment survey on June 9, 2017. These surveys were then compared (as well as the previous year's survey results) to evaluate the effectiveness using a chi-square analysis. There was a significant reduction in frequency of occurrence of the CLP comparing the pretreatment survey in April 2017 to the post treatment survey June 2017 (P<0.001). There was a rather large increase in frequency of occurrence of CLP from 2016 post treatment to 2017 post treatment, with an increase in mean density. The pretreatment frequency of CLP in 2017 was slightly lower than the pretreatment frequency in 2016. This decrease was not significant (P=0.54) There was also a significant reduction in 10 native plants from 2016 to 2017 within the treatment beds. A turion analysis showed a small increase in mean turion density from 2016 to 2017 in the more recently treated beds (205.5 turions/m² in 2016 to 218.5 turions/m² in 2016 to 23.1 turions/m² in 2017).

Introduction

An herbicide treatment was conducted on Bone Lake on May 3 and May 9, 2017 to reduce *Potamogeton crispus*-curly leaf pondweed (CLP). There were 7 beds treated, totaling 30.66 acres. This was the 10th year of treatment on 3 of the 7 beds (Beds 2,3,4) and the 9th year in Bed 5. Beds 6-8 were treated in 2013 for the first time (so this is the 5th year of treatment). Figures 1-3 are maps showing the location of each bed treated and the acreage. Beds 6 and 7 were separated into parts to better determine mean depths for the applicator concentration calculation. Also, beds 7 and 8 were delineated into shallow and deep sub-polygons in order to target acceptable herbicide concentration. The difference in color in those beds on the map shows the depth distinction.

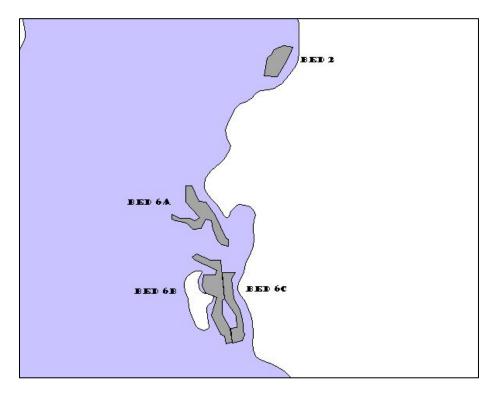


Figure 1: Bone Lake 2017 CLP treatment beds 2 and 6.

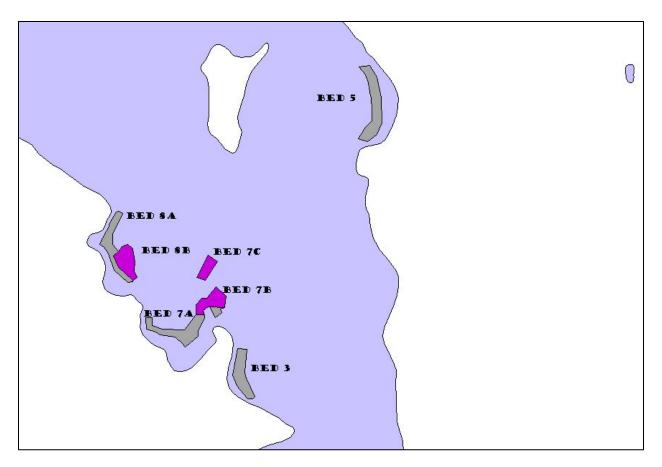


Figure 2: Bone Lake 2017 CLP treatment beds 3, 5, 7 and 8.

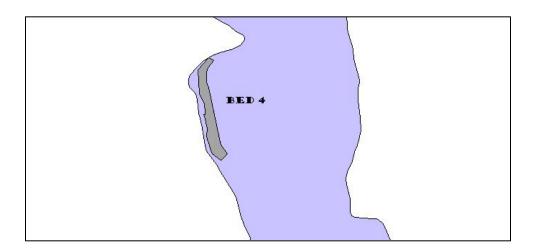


Figure 3: Bone Lake 2017 CLP treatment bed 4.

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Bed	Bed Acres		Acre feet	Date treated	Target Concen- tration(ppm)	Wind (mph)	Water Temp
2	1.81	9.9	17.92	5-9-17	2.5	Calm	55
3	2.30	7.4	17.02	5/3/17	2.0	2-8	49
4	3.96	8.6	34.06	5/3/17	2.0	2-8	49
5	3.10	8.2	25.42	5-9-17	2.0	Calm	55
6A	3.40	6.6	22.44	5-9-17	2.0	Calm	55
6B	3.74	7.5	28.05	5-9-17	2.0	Calm	55
6C	2.32	7.7	17.86	5-9-17	2.0	Calm	55
7A	3.07	7.4	22.72	5/3/17	2.0	2-8	49
7B	1.77	10.7	18.94	5-9-17	2.5	Calm	55
7C	0.97	11.4	11.06	5-9-17	2.5	Calm	55
8A	2.22	6.2	13.76	5/3/17	2.0	2-8	49
8B	2.00	9.6	19.20	5/3/17	2.5	2-8	49
Total	30.66		248.45				

 Table 1: Bone Lake 2017 CLP treatment bed data summary (from treatment record provided by applicator).

Table 2: Bone Lake CLP treatment bed descriptions.

Bed	Description
2	Bed 2 lies to the north on the east shore. It is the smallest bed. This bed has been treated for
	7 years prior to 2014. The depth drops off quickly on the lake side and gets quite shallow near
	shore. The CLP has been most dense in the southern portion of the bed and quite sporadic in
	coverage on the north. The effectiveness of treatment has been most consistent in this bed,
	but only very effective the last four years. The northern portion of this bed was eliminated for
	treatment in 2015 as no CLP has been found there for some time. Some CLP has returned to
	the eliminated portion after treatment in June 2016 and should be noted for future.
3	Bed 3 lies on the western shore just north of Bed 4. This bed is the smallest but has a history
	of being very dense, especially in the northern half of the bed. Treatment results have been
	inconsistent, but effective the last four years.
4	Until 2013, Bed 4 was the largest bed, but has been shrinking due to reduced CLP. This bed
	borders deep water and has been very inconsistent in response to treatment. It has been
	speculated that wind may be a factor in the results so treatment has been limited to winds
	below 10 mph. The middle portion of the bed has been the most dense, with the north and
	south end being more sporadic. There are a large number of piers bordering this bed on the
	west shore. The south portion of this bed was reduced in 2016 due to no CLP growth for last
	few years.
5	This bed has been treated one year less than beds 2,3 and 4. It lies on the east shore, south of
	bed 2. Bed 5 has been very dense in the southern half of the bed. The treatment has been
	very inconsistent in terms of effectiveness including 2012, in which it responded the least of
	all beds to the herbicide. The 2013 and 2014 treatments were more effective. It also borders

	deep water.
6	Bed 6 is made up of three separate areas. This is one of the densest areas of CLP on Bone
	Lake. Historically navigation channels have been treated within the area of this bed. Also, the
	bed borders a sensitive habitat area established many years ago by the Wisconsin DNR. To
	reduce adverse effects on this area, the bay to the north of the bed has been avoided. This
	bed lies adjacent (east) of Chaffee Island. Much of the bed reaches the surface each year.
	Treatment has been quite effective in 2013 and 2014. In 2015 the treatment was marginal.
7	Bed 7 is divided into two parts. One is a small portion to the north of the main part of the
	bed. The bed lies on the west shore north of bed 3. The bed is most dense in the middle
	portion with the west arm being very narrow in coverage and reduced in density as moving
	west. The small north portion that stands alone is dense, but in deep water (8-10 feet). Bed 7
	historically has been very dense with CLP reaching the surface every year.
8	Bed 8 is just north of bed 7 on the west shore. It is relatively shallow and flat, but borders
	deep water on the lake side. This bed has been extremely dense in past years with CLP
	reaching the surface in most of the bed. Beds 6, 7 and 8 were treated first in 2013. Bed 8
	had quite a large amount of CLP growing after treatment in 2014, mainly in deep water. This
	bed has been divided into a deep and shallow region to increase the effectiveness of the
	herbicide.

Methods

To conduct and analyze the treatment, two surveys are conducted following the protocol outlined in 2009 by the Wisconsin DNR. The first survey is referred to as a pretreatment survey. This involves going to predetermined GPS coordinates within the proposed treatment area. A high definition underwater camera as well as a rake is used to determine the presence of CLP at each sample point. Density is not measured as the plants are typically very small and density is very subjective and variable. The presence of CLP is simply determined. There are also points checked outside of the bed delineation to assure the boundary is correct (not recorded unless CLP was present).

The second survey is referred to as the post treatment survey. This survey involves going to the same GPS coordinates as the pre-treatment survey and doing a rake sample at the point. If any CLP is on the rake, the density of the CLP is recorded (see Figure 2 for reference). All other species are also recorded from the rake sample in order to verify no damage to the native plants.

Rating	Coverage
1	farter the second
2	Manadaranana
3	MARTIN



Figure 4: Density rating system and example CLP rake sample.

When the surveys are complete, the frequency of occurrence is determined as well as the mean density for each bed and all beds combined. The frequency of occurrence for each native plant species sampled is also calculated. A chi-square analysis is then used to determine if the change in frequency is statistically significant (p<0.05). With a successful treatment, the chi-square analysis shows that the frequency of CLP is significantly reduced and the native plants are not significantly reduced.

The comparison for reduction is evaluated three ways. First, the result from the previous year's post treatment survey is compared to the present year post treatment survey. This reflects a long-term effectiveness. As more treatments are done in annual succession, these frequency values can become very similar since the CLP growth is reduced so much. This can make it appear the treatment is not progressing successfully since the frequency appears to not be reduced. Each year, turions which germinate in the fall/winter create new growth. The result may be a low frequency in the post treatment survey, but with turion germination, high frequency CLP growth can result the following spring.

Second, in order to reflect the effect the treatment has on new spring growth, a second comparison is done. This compares the frequency of CLP in the spring, pre-treatment survey to the post treatment results in that same year. This shows what the CLP growth was just before treating and the result after treatment.

The third evaluation is comparing the pretreatment survey of the previous year(s). Since the spring growth will reflect CLP growth after turion germination, a reduction in the pretreatment frequency can show an overall reduction in CLP due to reduced turion germination, thus reflecting an overall reduction in CLP and its turions.

In the end, we want to see a statistically significant reduction when comparing the pre-treatment frequency to the post treatment frequency. We would also like to see a consistent frequency reduction from year to year, depending on how low it is in both the pre and post treatment surveys of successive years. If the frequency in any post treatment survey is very low (less than 10% as an example), then lowering it even more may be difficult. Turions can remain viable for several years, which can affect reduction amounts achieved.

In order reflect potential future growth and the cumulative success of treatments, a turion analysis is conducted in the fall following treatment. This analysis involves going to sample points near the middle of the CLP bed (assuming this will reflect the highest density). At each sample point a sediment sampler is lowered to the lake sediment and a sediment sample is obtained. Two samples are obtained from each side of the boat at each location. The samples are then sifted with a screened bucket to isolate the turions. The turions are then counted and the density of turions is calculated in turions/square meter. Consistently successful treatments should show a trend of reduced turion density each year. This way we know the treatments are killing plants prior to turion production, resulting in overall reduction in CLP in those beds.



Figure 5: Pictures showing turion density methods. A shows sediment sample; b shows separation; c Shows separated turions.



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Results

A pre-treatment survey was conducted on April 8, 2017 when water temperatures ranged from 44-46°F. The post treatment survey was conducted on June 9, 2017 which was 31 and 36 days respectively after herbicide was applied. CLP was dense and near surface in untreated beds at the time of the post treatment survey. The comparison of the surveys in 2017 indicate reduction in CLP occurred (statistically significant) from the 2017 treatment, but increases occurred in comparison to 2016. Table 3 and Figures 6 and 7 summarize the frequency of occurrence from the various surveys. Table 4 summarizes the reductions and/or increases in frequency with chi-square results.

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Bed	2017 pretreat frequency	2017 posttreat frequency	2016 pretreat frequency	2016 post treat freq.	2017 Post Treat density
2	60.0%	10.0%	68.8%	6.2%	0.10
3	68.2%	27.3%	59.1%	22.7%	0.27
4	63.6%	22.7%	60.0%	11.4%	0.25
5	64.7%	26.5%	78.8%	18.2%	0.41
6	75%	15.0%	87.3%	1.8%	0.20
7	65.9%	34.1%	72.1%	20.9%	0.52
8	84.8%	81.8%	93.9%	12.1%	1.52
All beds	69.6%	30.4%	71.9%	11.5%	0.46

Table 3: 2017 (and 2016) CLP survey statistic summary.

2017 Pre to 2017 Post	Decrease (statistically significant)	$P=1.0 \times 10^{-19}$
2016 Post to 2017 Post	Increase (statistically significant)	$P=4.8 \times 10^{-7}$
2016 Pre to 2017 Pre	Decrease (not significant)	P= 0.57

Table 4: Comparison and chi-square results of frequency in 2016 and 2017 surveys.

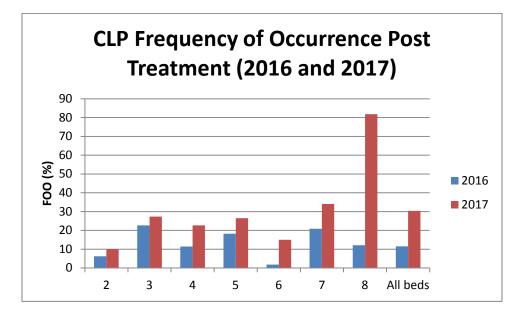


Figure 6: Frequency of occurrence of CLP after treatment 2016 to 2017 in each treatment bed.

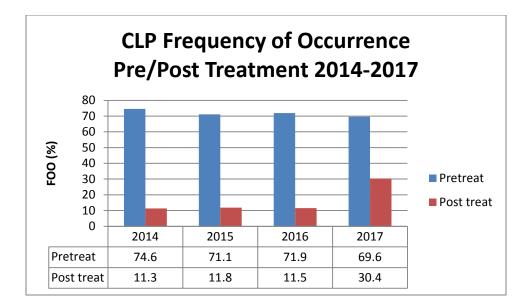


Figure 7: Frequency of occurrence of CLP in all beds pre and post treatment 2013-2017.

Density is also determined in the post treatment surveys only. Table 5 and Figure 8 show changes in mean density from the last three post treatment survey years.

Mean Density-All Beds	2013	2014	2015	2016	2017
Post treatment mean	0.1	0.14	0.16	0.17	0.46
density (0-3 rating)					

Table 5: Mean density statistics, 2013-2017, all beds.

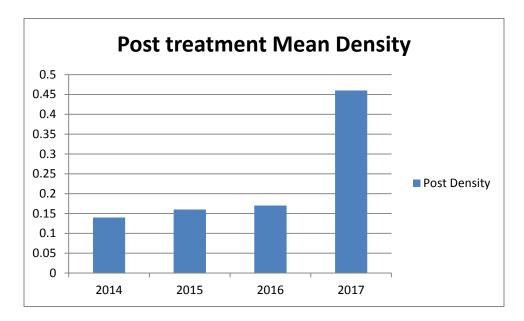
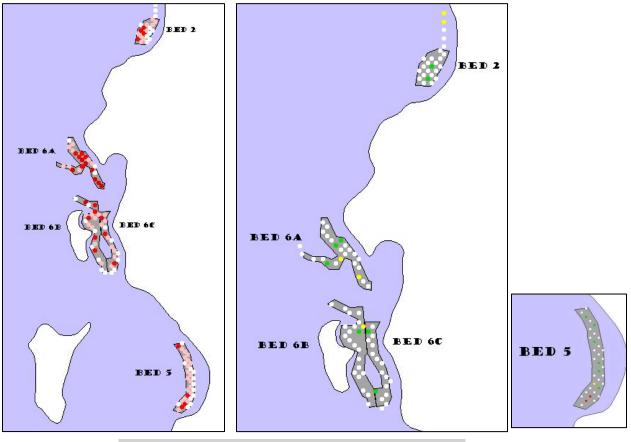


Figure 8: CLP mean density 2013-2017, all treatment beds (scale of 0-3).

Figures 6 and 7 show the frequency of occurrence in each bed in the pretreatment and post treatment surveys 2017. The specific density is also displayed in the post treatment survey, while relative density is shown in the pretreatment survey only, since plants are so small in early spring.



Pretreatment survey: Red = higher density Pink = lower density White = CLP absent Post treatment survey: White=0 Green = 1 Yellow = 2 Red = 3

Figure 9: Pre and post treatment CLP distribution maps for Beds 2.5 and 6 2017.

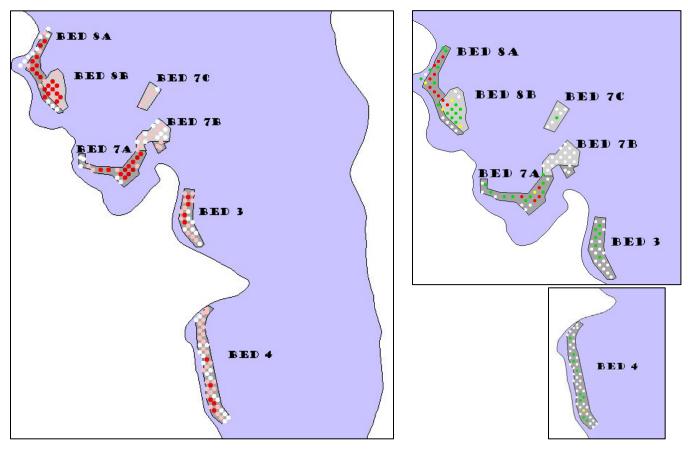
As the data shows, there was a significant reduction in CLP when comparing the pretreatment survey (April) of 2017 and the post treatment survey (June) of 2017 (69.6% to 30.4%). After the 2016 treatment, the CLP growth returned filling in much of the treatment polygons leading to a high frequency in the April pretreatment survey. After treatment occurred, the post treatment survey in June showed the frequency was significantly reduced.

A comparison between the 2016 post treatment frequency/density to the 2017 post treatments shows an increase in frequency in all beds from 11.5% in 2016 to 30.4% in 2017, which was significant. There was varying increases in frequency in each bed from 2016 to 2017.

Comparing the pretreatment survey in previous years is arguably the best method to show true long term reduction since this survey is conducted after the CLP has grown back in the winter/spring. This

comparison shows a slight decrease in frequency (71.9% in 2016 to 69.6% in 2017). This reduction was not significant statistically. Long-term the desire is for a reduction in pretreatment frequency as this indicates overall reduced CLP growth. The frequencies have remained stable over the past several years.

A few beds had more growth than would be desired after treatment. Bed 8 appeared have little to no reduction with dense, widespread CLP growing in June 2017. Bed 7A also had quite a large amount of CLP growing after treatment. Beds 7A and 8 (as well as Beds 3 and 4) occurred on a day when there was a wind up to 8 mph. This may have been a factor in the reduced results.



Pretreatment survey: Red = higher density Pink = lower density White = CLP absent Post treatment survey: White=0 Green = 1 Yellow = 2 Red = 3

Figure 10: Pre and post treatment CLP distribution Beds 3,4 7 and 8.

Native Plant Response

In addition to the evaluation of the CLP changes, the native plant species are evaluated for any reductions, potentially due to herbicide application. Table 5 shows the frequency of native species in 2016 and 2017. A chi-square analysis is calculated to determine if the changes are statistically significant. Ten native species had a significant reduction. This is cause for concern if caused by herbicide use. However, the CLP reduction from herbicide was less than typical and so to have less effect on CLP and more effect on natives is somewhat strange. It is possible this reduction is due to herbicide but it is also possibly due to seasonal variation and dense CLP growth.

Endothall is a broad spectrum herbicide so it can affect most plant species. Especially low frequency native species could have changed due to late growth or sampling variation. Bed 8 had very dense CLP growth with many sample points only having CLP present and limited native species. This dense growth could keep natives from growing. Also, in many areas, very dense samples of dead CLP were sampled, with very little native species present. This may demonstrate that the CLP was quite tall when application of herbicide occurred and may show that this dense, taller CLP affected the native species growth and there was too little time for the native species to respond with growth after application. Regardless, this is a large number of native species to have significant reductions.

YEAR	Potamogeton zosteriformis	Nitella sp.	Lemna triscula	Heterantehra dbuia	Ceratophyllum demersum	Bidens beckii	Potamogeton praelongis	Najas flexilis	Vallisneria americana	Myriophyllum sibiricum	Stuckenia pectinatus	Ranunculus aquatilis	Chara sp.	Elodea canadensis	Nymphaea odorata	Potamogeton gramineus
2016	0.01	0.03	0.29	0.06	0.24	0.07	0.06	0.03	0.09	0.13	0.00	0.05	0.10	0.03	0.01	0.004
2017	0.00	0.01	0.05	0.02	0.09	0.00 3	0.02	0.00	0.04	0.04	0.01	0.01	0.12	0.0	0.0	0.003
Change	-	-	-	-	-	-	-	-	-	-	+	-	+	-	-	-
Reduction Sig.	no	no	Yes	yes	yes	yes	yes	yes	yes	yes		yes		yes	no	n/c

Table 6: Native plant frequency of occurrence, 2016 and 2017 with change and significance based upon chisquare analysis.

Turion Analysis

The turion analysis data collection was conducted on Oct. 6, 2017. Table 6 shows the turion density for each treatment bed in 2017, and previous years the analysis was conducted in all treatment beds.

Bed	2011 (T/m ²)	2012 (T/m ²)	2013 (T/m ²)	2014 (T/m ²)	2015 (T/m ²)	2016 (T/m ²)	2017 (T/m ²)
2	75	27	34.7	10.9	0	16.3	0
3	269	65	79.4	48.8	0	119.6	54.25
4	512	47	29.8	36.2	28.7	18.1	14.5
5	274	161	64.5	76	75.25	65.2	16.25
All (2-5)	296	75	49.6	43.0	25.99	60.6	23.1
6	n/a	n/a	421.6	384.1	303.2	214.7	267
7	n/a	n/a	165.3	178.4	38.2	55.6	48.3
8	n/a	n/a	489.4	271.3	258	395.1	304
All(6-8)	n/a	n/a	358.8	277.9	199.8	205.5	218.55

 Table 7: Turion density data all treatment beds, 2011-2017.

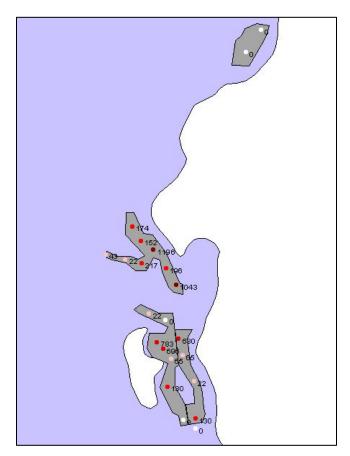


Figure 13: Turion density map for beds 2 and 6.

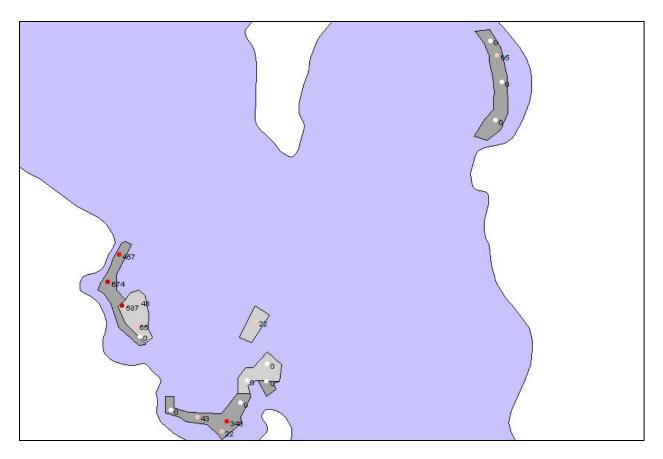


Figure 14: Turion density map for beds 5, 7 and 8.

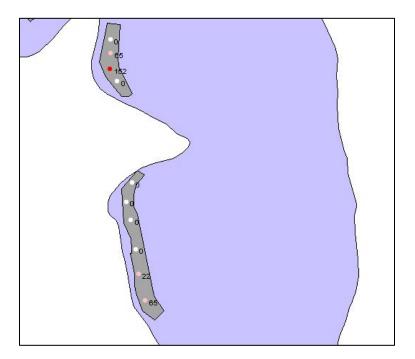


Figure 15: Turion density map for beds 3 and 4.

Figure 16 is a bar graph of the turion density by bed and all beds combined (separated as beds 2-5 and 6-8 due to total years of treatment).

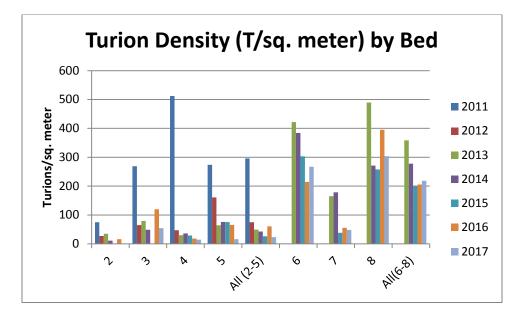


Figure 16: Turion density by bed 2011-2017.

Figure 17 graphically shows the turion density that has been occurring in all beds. As successful treatments continue, the turion density should continue to decrease, resulting in less CLP growing each spring within these treatment beds.

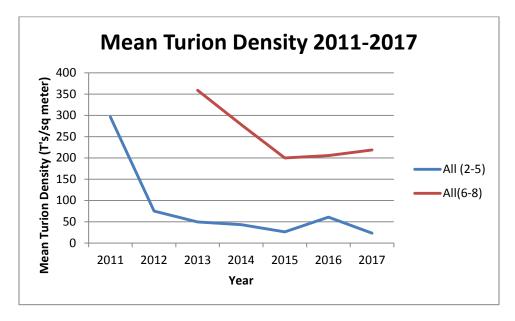


Figure 17: Mean turion density change, 2011-2017.

CLP Bed Mapping

On June 19, the CLP outside of the treatment beds was mapped. The 2017 mapping survey resulted in similar areal coverage of CLP compared to 2016. There was a slight increase in the area of higher density beds. CLP was mapped based upon being either higher density (at (or very near) the surface and mean density estimated >2) or lower density (not at surface and mean density estimated <2).

Figures 18 and 19 show the CLP beds mapped in 2017. The beds that had high density CLP totaled 41.5 acres (red on map). The total area for low density was 9.6 acres (pink on map).

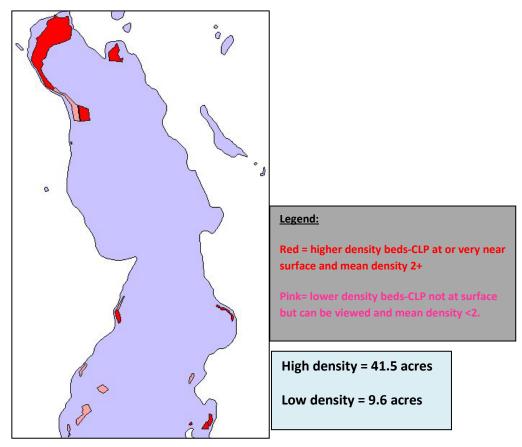


Figure 18: CLP map of dense, surface beds north.



Figure 19: CLP map of dense, surface beds south.

Discussion

The data shows that the CLP treatment was successful in 2017 at reducing the CLP that was growing in April (before treatment). However, the treatment cannot be considered effective when comparing the surveys to 2016. The post treatment frequency of CLP in 2017 was higher in all beds than in 2016, as was the mean density, which had a marked increase. The least effective treatment beds were Bed 8 and Bed 7A. The cause of the lack of effectiveness is unknown but could be wind related as these two beds were treated when the wind speed reached 8 mph (and may have exceeded this velocity after treatment). The pretreatment 2016 survey compared to pretreatment 2017 survey, had a slight decrease but was very small and therefore does not really demonstrate long term CLP reduction this past year.

The native plants changed significantly, with a significant reduction in 10 native species. This is cause for concern and could be the result of the herbicide, but there could be other factors as well. One may be that dense CLP growth reduced the ability for native plants to grow. This could be variation in growth from one year to the next.

In the turion analysis, it was revealed that the turion density declined in the beds 2-5, which have been treated more years. Beds 6-8 showed a small increase, likely due to CLP growth from ineffective herbicide reduction in portions of these beds.

There remains a rather large area of dense CLP beds growing in Bone Lake. A survey of the CLP outside of the treatment areas at or near peak growth resulted in the delineation of 38.2 acres of CLP that is dense and at or near the surface. There was 9.6 acres of low density (not near the surface and more sporadic coverage) delineated.

It is recommended that treatment continue in all treatment beds, if the goal remains for CLP reduction. Turion density has been decreasing in beds 2,3,4 and 5, with an exception this past year in beds 2 and 3. The spring pretreatment survey may lead to reduction in the treatment area within these beds. It should be noted that Bed 2 had some CLP in an area that was not treated due to lack of CLP for 2-3 years. This area will be monitored in terms of a need to treat that area again. Beds 6, 7 and 8 have not been treated as many years and so the treatment area should continue as the same size until further reductions are made. The turion density remains quite high in these areas.

References

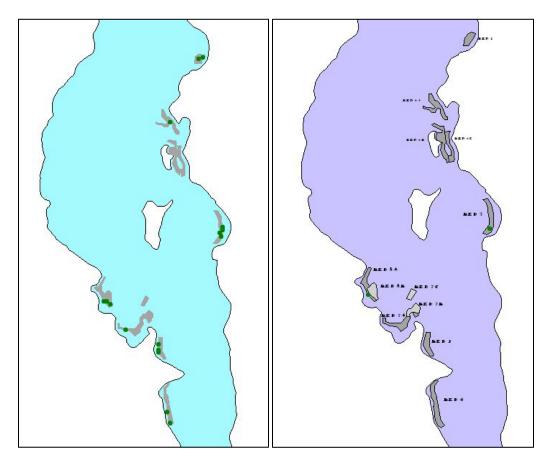
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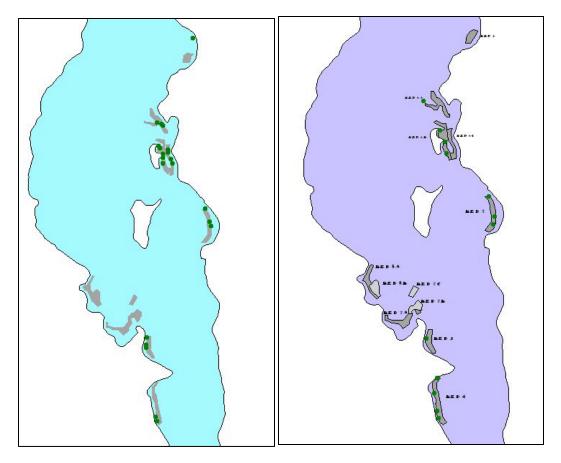
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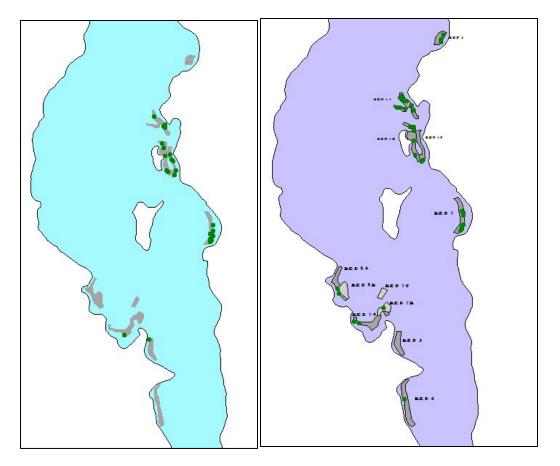
Appendix-Native Plant Distribution Maps



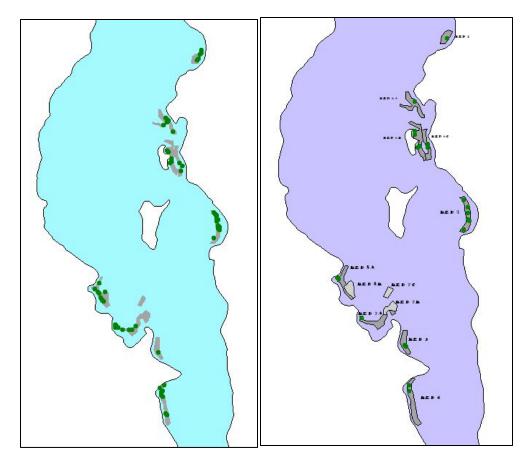
Bidens beckii-Water marigold 2016



Vallisneria americana-Wild celery 2016



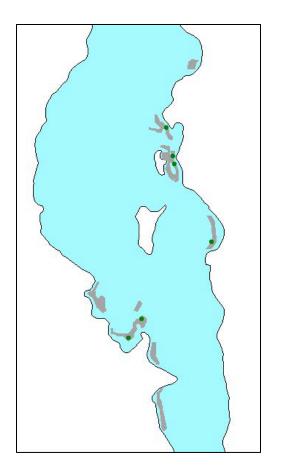
Chara sp.-muskgrass 2016



Ceratophyllum demersum-Coontail 2016

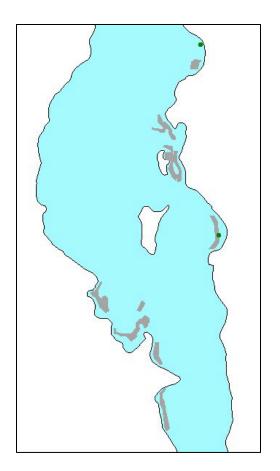


Rununculus aquatilis-Crowfoot 2016



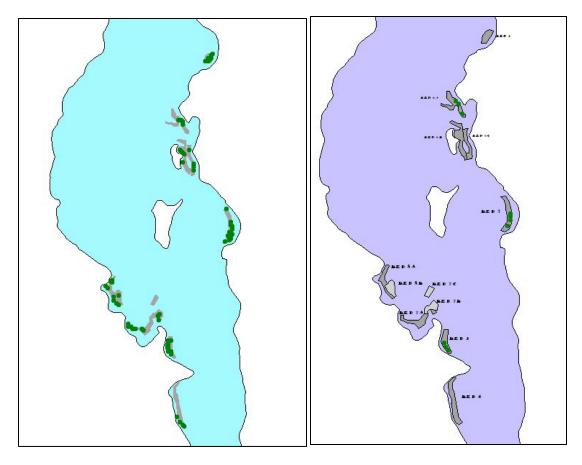
Elodea canadensis-Common waterweed 2016

Not sampled in 2017

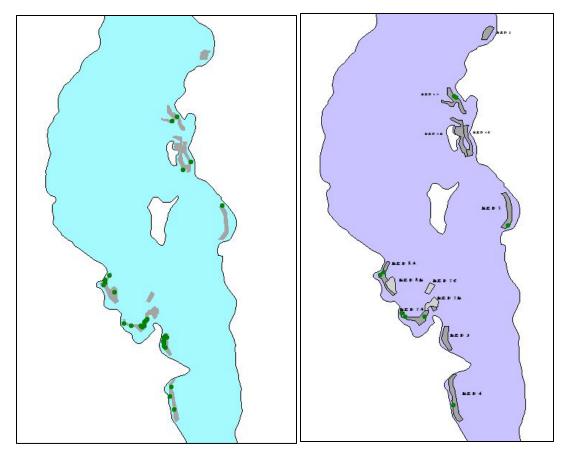


Potamogeton zosteriformis-Flatstem pondweed 2016

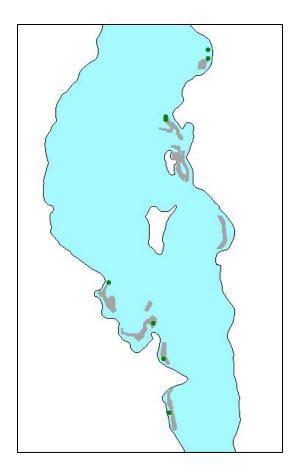
Not sampled in 2017



Lemna triscula-Forked duckweed 2016

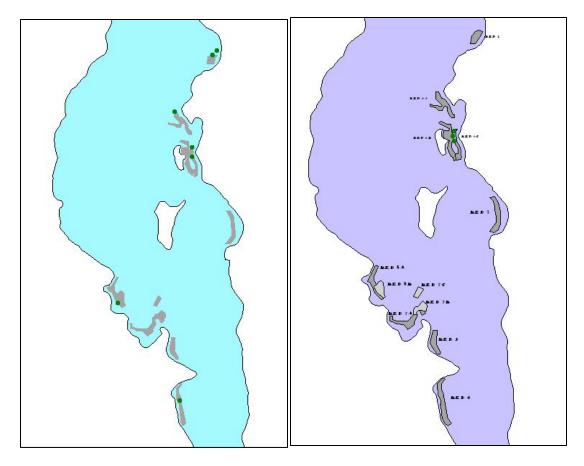


Myriophyllum sibiricum-Northern water milfoil 20162017

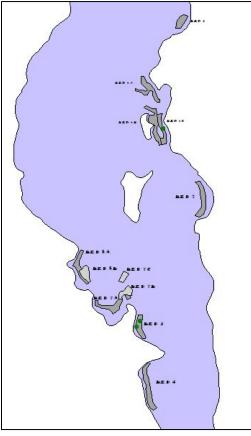


Najas flexilis-Slender naiad 2016

Not sampled in 2017



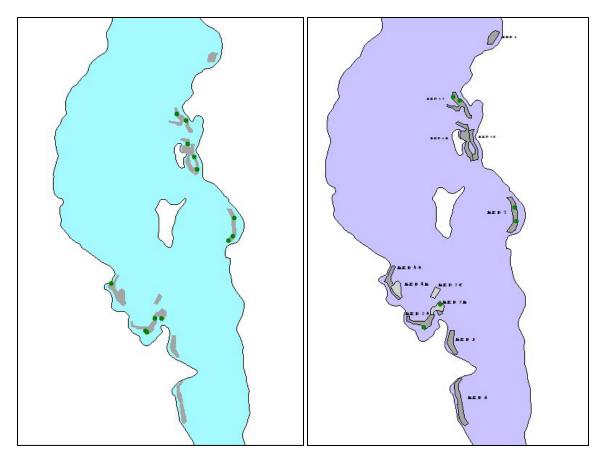
Nitella sp.-stonewort 2016



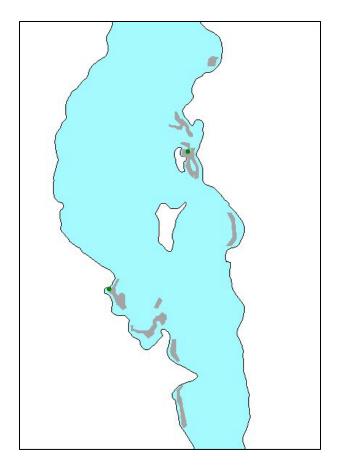
Potamogeton pectinata-Sago pondweed 2016(not sampled)2017



Heteranthera dubia-Water stargrass 2016

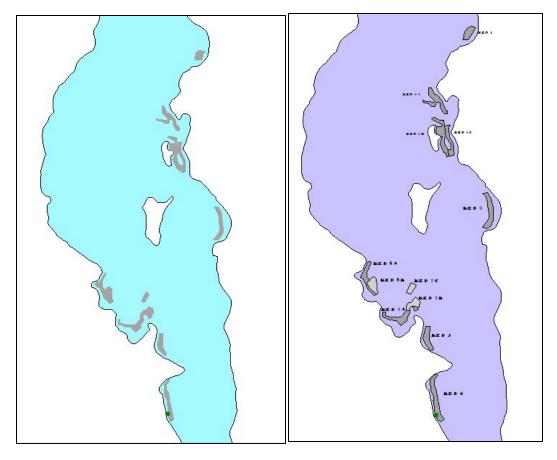


Potamogeton praelongus-Whitestem pondweed 2016



Nymphaea odorata-White water lily 2016

Not sampled in 2017



Potamogeton gramineus-Variable pondweed-2016

