

**Herbicide Treatment Analysis for  
*Potamogeton crispus* (Curly-leaf pondweed)**

**Deer Lake, Polk County Wisconsin**

**WBIC: 2619400**

**October, 2016**

***Survey and analysis conducted by: Ecological Integrity Service, LLC  
Amery, WI***

## *Abstract*

On April 29, 2016 22.6 acres of *Potamogeton crispus*-curly leaf pondweed (CLP) were treated with endothall(broad spectrum herbicide) to reduce the frequency and density of the CLP within 5 different beds. An April 21, 2016 pretreatment survey revealed CLP present in 47.5% of the sample points within the delineated beds. A June 10, 2016 post treatment survey revealed a frequency of 13.7% of the same sample points within the delineated beds. The treatment resulted in a frequency reduction of 71.2%. This reduction was statistically significant ( $P=9.5 \times 10^{-10}$ ). Comparing the pretreatment survey results in 2015 to the pretreatment survey results in 2016, a slight increase in CLP frequency of occurrence in the five beds occurred. The frequency of occurrence after treatment (post treatment survey) in 2015 compared to 2016 showed an increase from 2.1% to 13.7%. The mean density increased from 0.03 in 2015 to 0.2 in 2016 (scale of 0-3). There was a statistically significant reduction (chi-square analysis) in two native species and a statistically significant increase in two other native species from 2015 to 2016. The turion analysis showed an increase in turion density in three of the five treatment beds. Some increase is likely due to increased CLP growth after treatment. It also demonstrates that CLP growth will likely be returning with fairly high density in these beds in 2017.

## Introduction

On April 29, 2016 an herbicide treatment targeting curly-leaf pondweed (*Potamogeton crispus*) was conducted using endothall. This analysis will outline the areas treated, describe the treatment protocol, and analyze the effectiveness of the treatment.

The treatment areas for Deer Lake were made up of five beds, labeled A-E (totaling 22.6 acres). Those beds, with their areas, are shown in figures 1 and 2. Portions or all of beds B, C and D have been treated annually since 2006, while beds A and E have been treated annually since 2010.

The herbicide endothall was used in the treatment of the CLP. The water temperature was 49 degrees F and winds were 0-3 mph from the north at application.

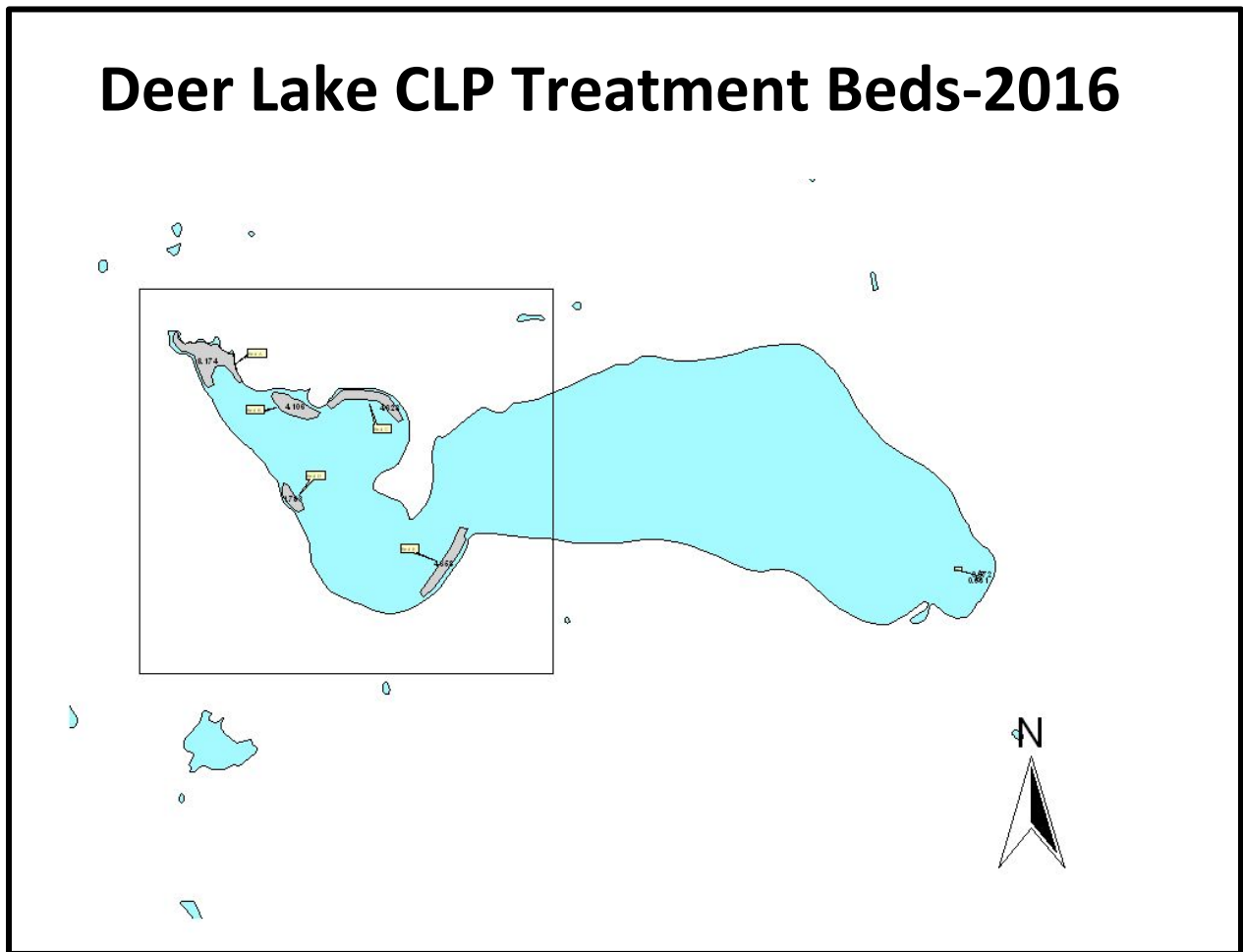


Figure 1: Large map showing the location of the treatment beds relative to the remaining lake in 2016.

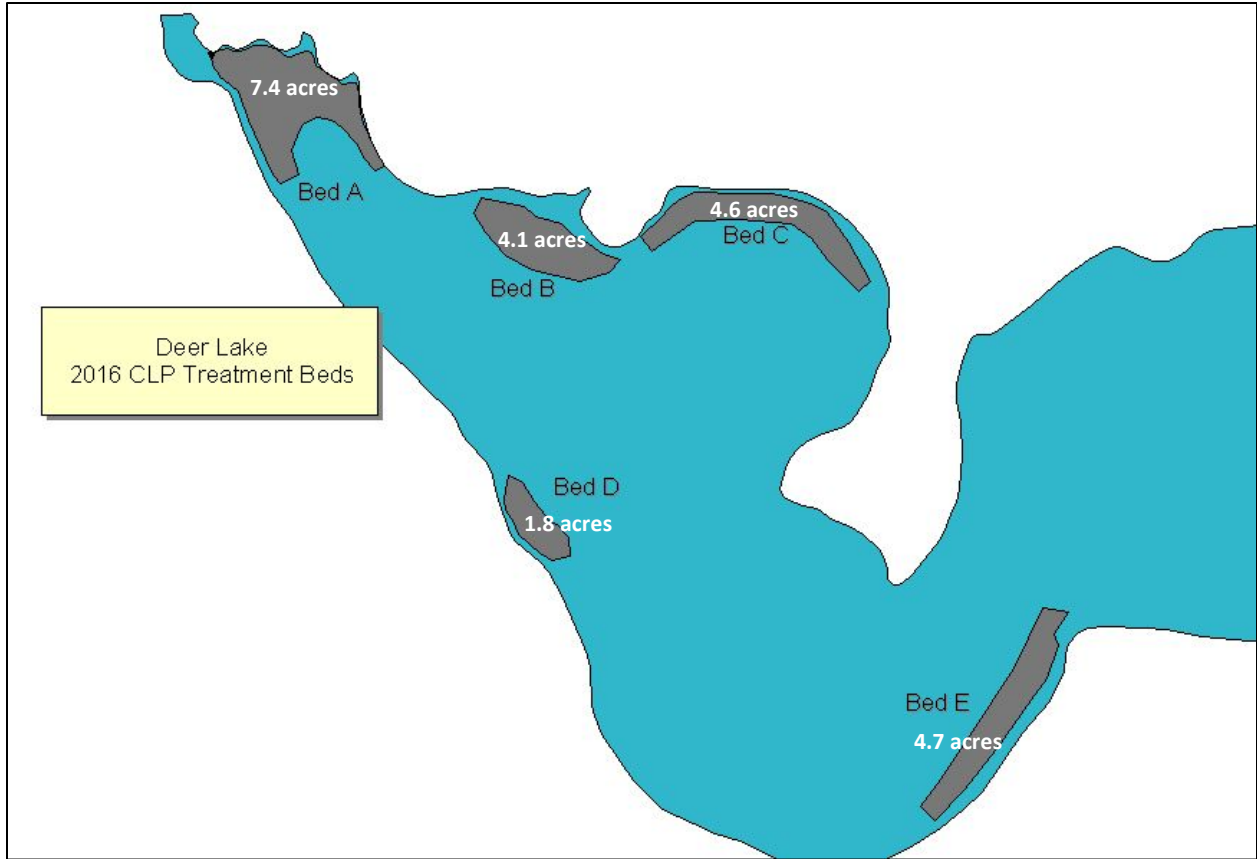


Figure 2: Close map of 2016 CLP treatment beds.

<b>Deer Lake CLP Beds</b> (Treatment on April 29, 2016)						
<b>2016 Beds</b>	<b>Area (acres)</b>	<b>Mean Depth</b>	<b>Acre-Feet</b>	<b>Application Rate*(ppm)</b>	<b>Wind conditions*</b>	<b>Water Temp*(°F)</b>
<b>Bed A</b>	7.4	5.2	38.48	1.5	0-3 N	49
<b>Bed B</b>	4.1	6.8	27.88	1.5	0-3 N	49
<b>Bed C</b>	4.6	7.8	35.88	1.5	0-3 N	49
<b>Bed D</b>	1.8	7.9	14.22	1.5	0-3 N	49
<b>Bed E</b>	4.7	8.1	38.07	1.5	0-3 N	49
<b>Total</b>	22.6		154.53			

*\*Reported from applicator treatment records.*

**Table 1: Summary of treatment beds, 2016.**

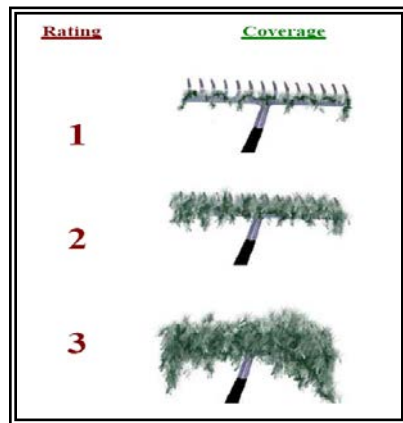
<b>Treatment Bed</b>	<b>Description</b>
<b>Bed A</b>	Bed A is near the landing and extends out from the landing quite a distance. The area in the middle is too deep causing the CLP to spit the bed into two forks. The CLP has been quite dense except for the area just near the landing. The eastern fork of the bed has quite a large amount of floating vegetation. The bed had successful treatment in 2012, 2013, 2014 and 2015. It has been treated since 2010. The bed was reduced in 2016 adjacent to boat landing as no CLP has been found for 2 years.
<b>Bed B</b>	Bed B is located on the east shoreline just south of Bed A. This bed has been notoriously dense and has been treated since 2006. The bed has white-stem pondweed, forked duckweed and coontail in fairly high frequency. The bed gets quite scattered with CLP in the more shallow areas and is then quite dense in deeper water. The boundary has been very well defined. The treatment was successful in 2012, 2013, 2014 and 2015.
<b>Bed C</b>	This bed is south and east of Bed B. The bed is quite long curving along the shoreline to the north and west. This bed is narrow but long, bordered on the lake side by deeper water, creating a well defined boundary. The bed has been very dense in the 6-8 ft depths, with less density on the shore side of the bed. The ends have been sporadic, but very dense just inside. The treatment was successful on Bed C in 2012-2015. This bed has been treated since 2006 in half of the bed and then the bed was increased in size and treated in 2010.
<b>Bed D</b>	This is a small bed on the western shore, just south of the landing. It changes in depth greatly over a rather short distance across the bed. It has been very dense in the middle and toward the north portion of the bed. The treatment was successful in 2012, 2013, 2014 and 2015. This bed was one of the original beds treated starting in 2006.
<b>Bed E</b>	Bed E is a long and very narrow bed that changes from 2.5 feet to 12+ feet on the lake side boundary. The highest density has been on the eastern ½ of the bed, but it is quite dense throughout. This bed has a fairly large amount of northern milfoil present throughout the bed. This bed had successful 2012, 2013, 2014 and 2015 treatments and has been treated since 2010.

**Table 2: Treatment bed descriptions.**

## Methods

To conduct and analyze the treatment, two surveys are conducted following the Wisconsin DNR treatment protocol outlined in 2009 by the Wisconsin DNR. The first survey is referred to 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 that sample point. Density is not measured as the plants are typically very small and density is subjective, but is rated low/high density based upon relative number of CLP plants. The presence of CLP is simply determined. There are many points checked outside of the bed delineation to assure the boundary is correct.

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 3 for reference). All other species are also recorded from the rake sample in order to verify no damage to the native plants.



**Figure 3: 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 as well as 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 ( $p < 0.05$ ). The goal is to find the chi-square analysis show that the frequency of CLP is significantly reduced and the native plants are not significantly reduced.

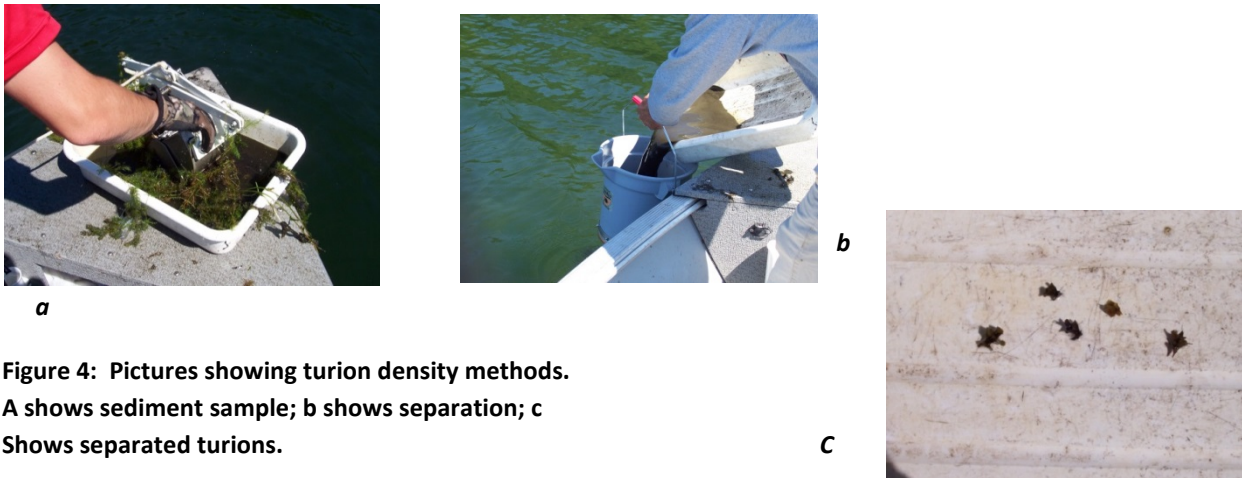
The comparison for reduction can involve three evaluations. First, the result from the previous year's post treatment survey is compared to the present year post treatment survey. This reflects a long-term effectiveness. As more treatments are done in annual succession, these frequency values can become 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, new turions can germinate in the fall/winter and create new growth. The result from turion germination is a low frequency in the post treatment survey, but in the next spring the CLP has grown immensely, and results in a high frequency.

In order to reflect that new growth and the effect the treatment has on it, 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 method is to evaluate the pretreatment survey frequency from year to year. Since the pretreatment survey frequency reflects new growth from turion germination, a reduction from year to year in this frequency can show long-term reduction since it reflects the new CLP growth resulting from turions. If the CLP frequency goes down each year, there must be less turions germinating each year.

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 the pre and post treatment surveys in 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 not be realistic, but is the goal. Comparing the pretreatment surveys from year to year can show the progress being made as it reflects growth after turion germination, thus reflecting potential overall reduction. Turions can remain viable for several years, which can affect reduction amounts achieved.

In order to further reflect potential future growth and the cumulative success of treatments, a turion analysis is conducted. 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 separated 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 4: Pictures showing turion density methods. A shows sediment sample; b shows separation; c Shows separated turions.**

## Results

### CLP Reduction

The results of the pretreatment and post treatment surveys from 2016 are summarized in table 3. The pretreatment survey was conducted on April 26 and the post treatment survey was conducted on June 10, 2016. CLP was dense in other area lakes when the post treatment survey was completed, thus demonstrating post survey was near peak CLP growth. The tables also contain information from 2015 to show changes between years of treatment.

Treatment Bed	Pre-treat freq (2015)	Post treat freq (2015)	Pre treat freq (2016)	Post treat freq (2016)	Mean density 2015	Mean density 2016
Bed A	57.4%	6.4%	60.4%	4.6%	0.11	0.05
Bed B	58.3%	0.0%	45.8%	20.8%	0.0	0.38
Bed C	32.2%	0.0%	38.7%	19.4%	0.0	0.32
Bed D	33.3%	0.0%	50.0%	0.0%	0.0	0.00
Bed E	36.7%	0.0%	37.9%	20.7%	0.0	0.28
All beds	45.8%	2.1%	47.5%	13.7%	0.03	0.2

**Table 3: Summary of CLP growth frequency pre and post treatment 2015-2016.**

As stated in the methods, a chi-square analysis is conducted on the frequency data. The results of this are summarized in table 4 (all beds combined).

Survey Comparison	Statistically significant reduction?	Chi-square result (reduction)
2016 pretreatment freq/2016 post treatment freq.	Yes	$P=9.5 \times 10^{-10}$
2015 post treatment freq/2016 post treatment freq.	Increase	n/a
2015 pretreatment freq/2016 pretreatment freq.	Increase	n/a

**Table 4: Summary of frequency reduction and significance after treatment.**

The chi-square analysis shows a statistically significant reduction from before treatment to after treatment in 2016. There was a increase from the post treatment frequency 2015 to post treatment frequency 2016. There was a slight increase from pretreatment frequency 2015 to pretreatment frequency 2016. The overall density from 2015 to 2016 increased. Most of the CLP growth after treatment occurred in Beds B, C and E.



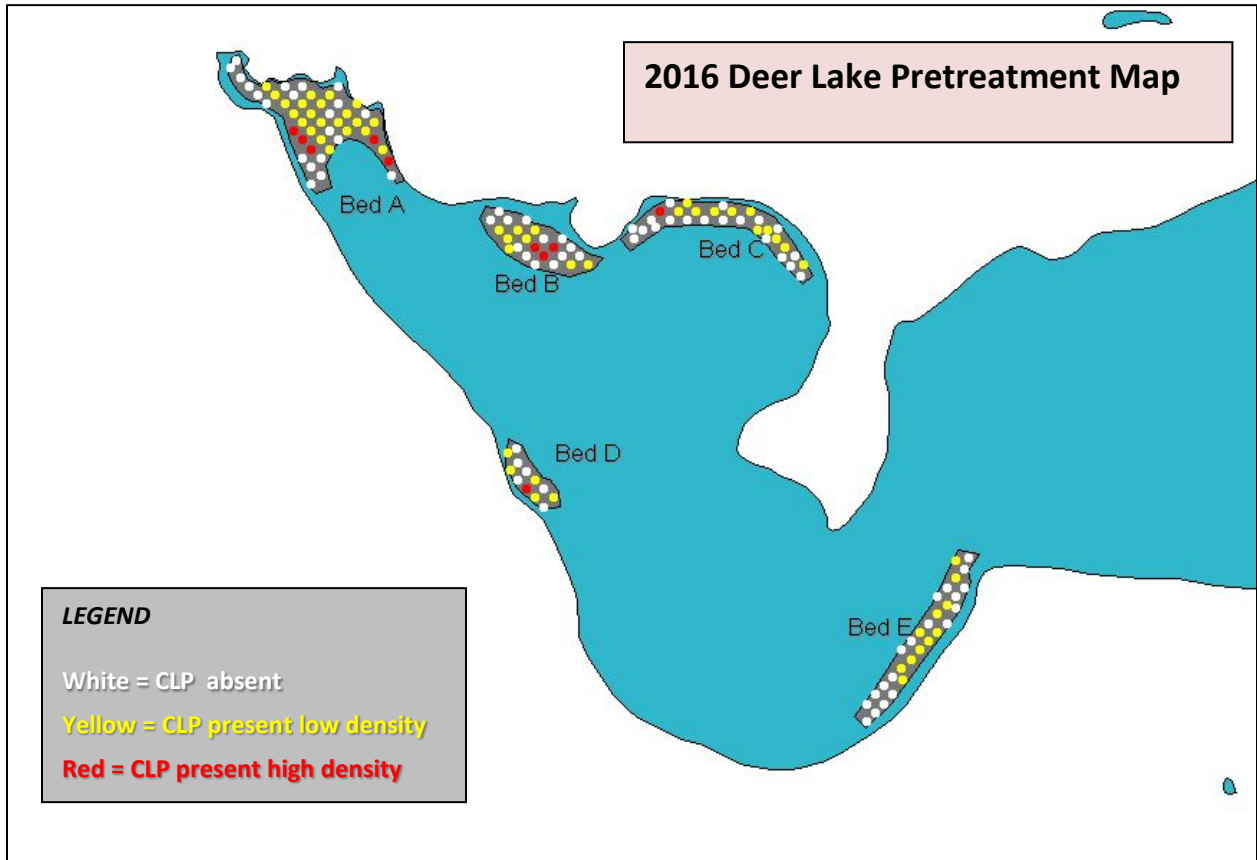


Figure 5: Pre-treatment map from 2016 pretreatment survey showing presence/absence of CLP .

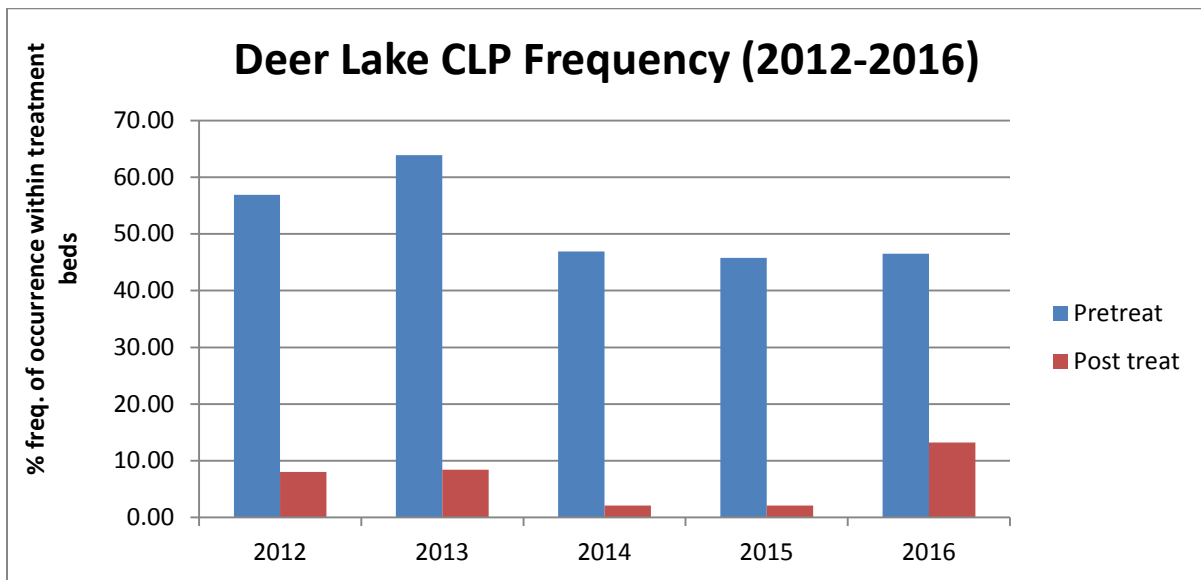


Figure 6: Graph showing the frequency comparison from 2012 and 2016-all beds treated.

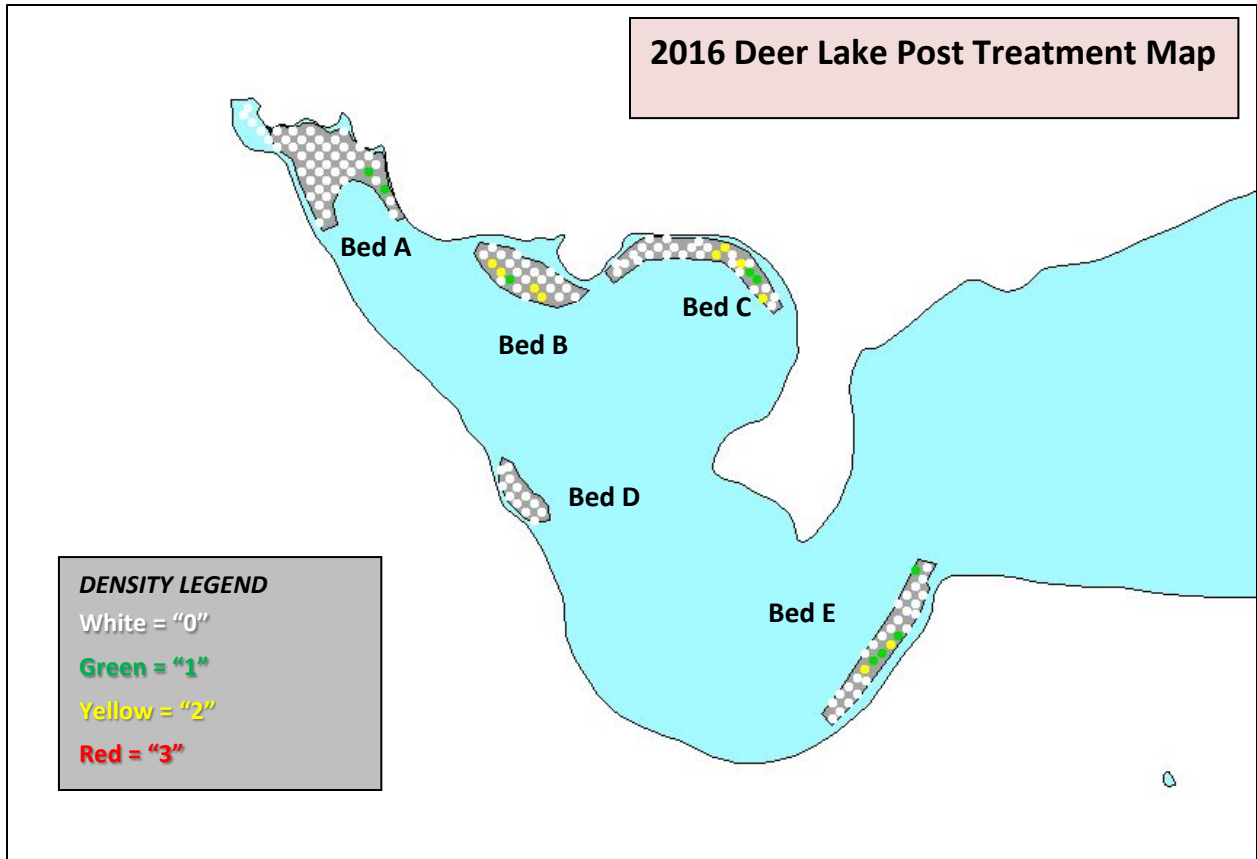


Figure 7: Map showing CLP sampled and density in 2016 post treatment survey.

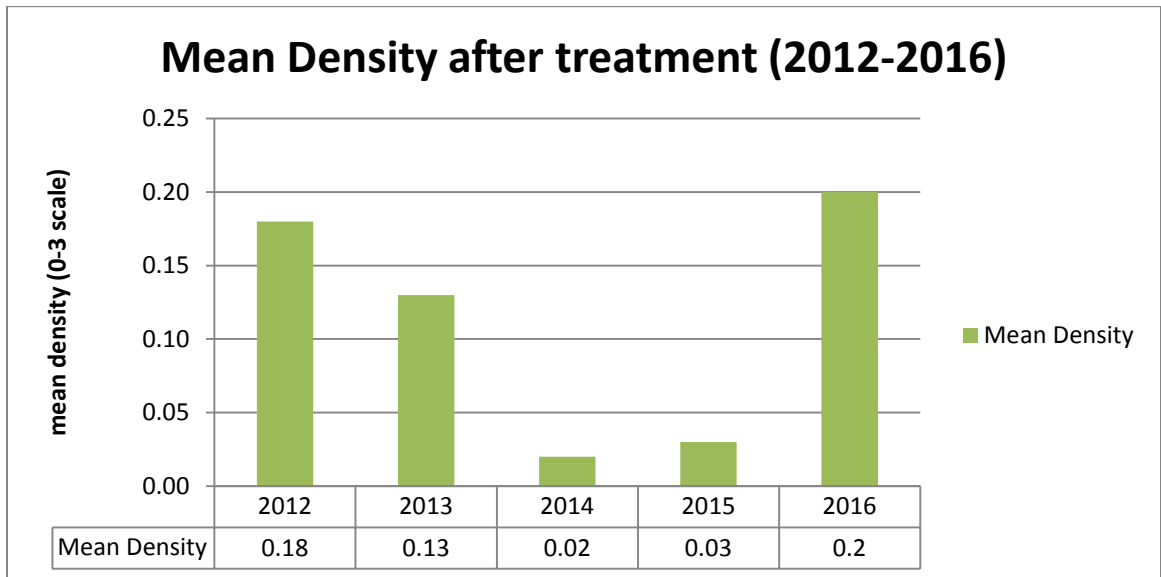
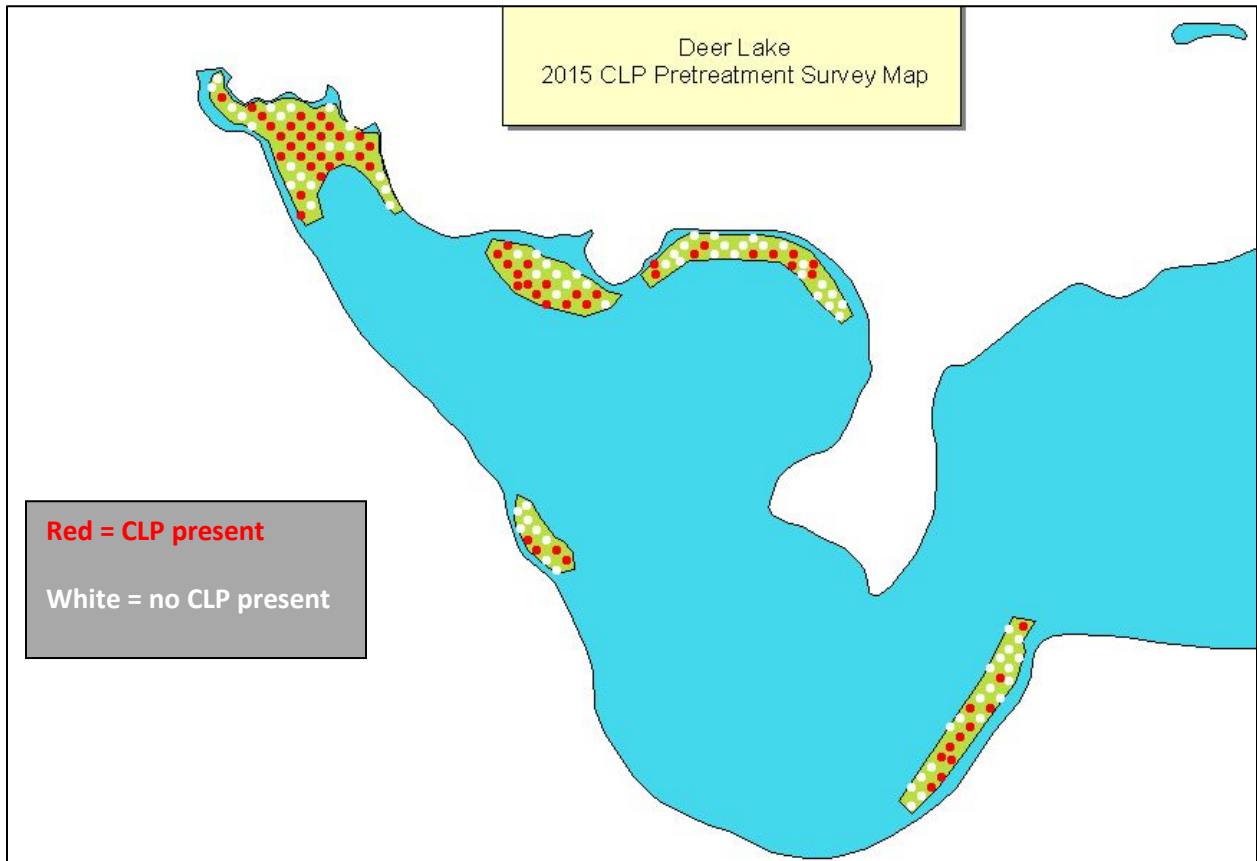


Figure 8: Comparison of post treatment mean density all beds, 2012-2016.

The mean density increased significantly from 2015 to 2016. Figure 8 shows that the mean density rose from 0.03 to 0.2 (density rated from 0-3). This occurred predominately in Beds B, C and E. Figure 8

shows that the mean density has declined since 2012, with a dramatic decline from 2013 to 2014 and small increase in 2015 and finally a large increase in 2016.

Figures 9 and 10 show the maps of the pretreatment and post treatment surveys from 2015 for comparison to 2016 maps.



**Figure 9: Pretreatment survey map, 2015**

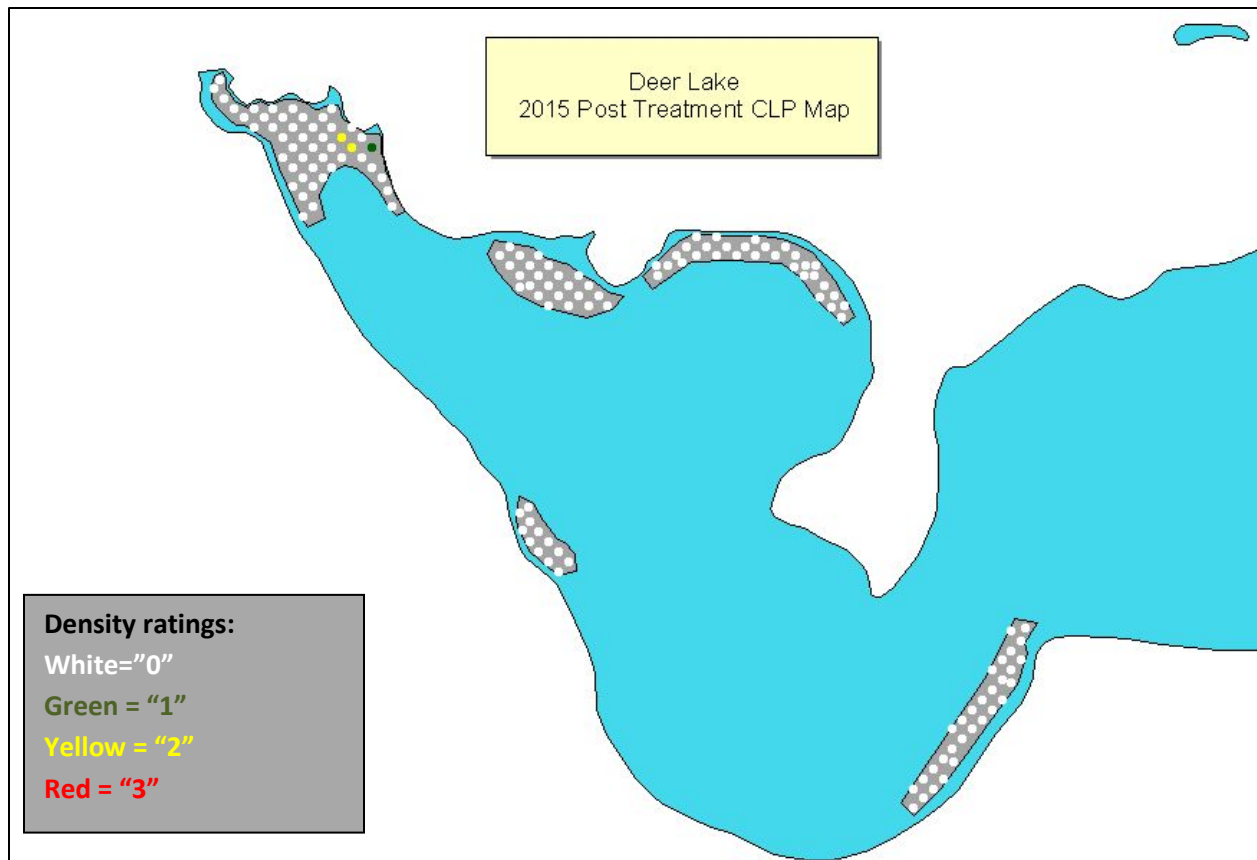


Figure 10: Post treatment survey map, 2015.

### Native Plant Changes

The native plant frequencies were evaluated during the post treatment survey. Table 5 summarizes those results and the chi-square analysis that determines the significance of any reductions, potentially to herbicide exposure from the 2016 treatment.

Native species	Frequency 2015	Frequency 2016	P value	Significant reduction	Change
<i>Lemna trisulca</i> , forked duckweed	0.24	0.12	0.009	yes	-
<i>Potamogeton. robbinsii</i> , Fern pondweed	0.01	0	0.32	no	-
<i>Potamogeton praelongus</i> , White-stem pondweed	0.12	0.03	0.004	yes	-
<i>Ceratophyllum demersum</i> , Coontail	0.55	0.51	0.52	no	-
<i>Myriophyllum sibiricum</i> , Northern milfoil	0.21	0.43	5.5X10 <sup>-5</sup>	n/a	+
<i>Potamogeton richardsonii</i> , Clasping pondweed	0.11	0.24	0.003	n/a	+
<i>Vallisneria americana</i> , Wild celery	0.01	0	0.16	no	-
<i>Elodea canadensis</i> , elodea	0.28	0.22	0.23	no	-
<i>Heteranthera dubia</i> , water stargrass	0.17	0.17	n/a	n/a	nc
<i>Ranunculus aquatilis</i> , stiff water crowfoot	0.14	0.11	0.43	no	-
<i>Chara sp.</i> , muskgrasses	0.10	0.09	0.61	no	-

Native species	Frequency 2015	Frequency 2016	P value	Significant reduction	Change
<i>Nymphaea odorata</i> , white lily	0.08	0.05	0.27	no	-
<i>Stuckenia pectinata</i> , sago pondweed	0.03	0.05	0.51	n/a	+
<i>Nitella sp.</i> , Stonewort	0.01	0.01	n/a	n/a	nc
<i>Bidens beckii</i> , Water marigold	0.07	0.05	0.50	no	-
<i>Potamogeton illinoensis</i> , Illinois pondweed	0.01	0.0	0.33	no	-
<i>Potamogeton gramineus</i> , variable pondweed	0.01	0.0	0.33	no	-
<i>Eleocharis acicularis</i> , needle spikerush	0.01	0.0	0.33	no	-
<i>Potamogeton epihydrous</i> , ribbon pondweed	0.01	0.0	0.33	no	-

**Table 5: Native species frequency and chi-square analysis-2015 to 2016.**

The native plant survey data shows a reduction in two native species, which were significant (*Potamogeton praelongus*-white stem pondweed and *Lemna trisulca*-forked duckweed). The source of this reduction is unknown. It could be due to natural variation, sampling variation or herbicide application. There was also a statistically significant increase in two native species so this may indicate it isn't due to herbicide since the broad spectrum herbicide used can kill all plants. If the native plants are out of dormancy at the time of application, they are more susceptible to the herbicide.

### CLP mapping

After the post treatment survey is completed, the entire lake is surveyed looking for CLP beds. A bed is defined as an area of CLP that is dominated by CLP, has a mean CLP density >2, and can be delineated by sight. In order to be delineated by sight, the CLP must be growing at or near the lake surface. There were four CLP beds seen outside of treatment and one bed within treatment area in 2016. Figures 11 and 12 shows the location of the CLP observed out of the treatment beds. Figure 13 is a map of the CLP beds observed.

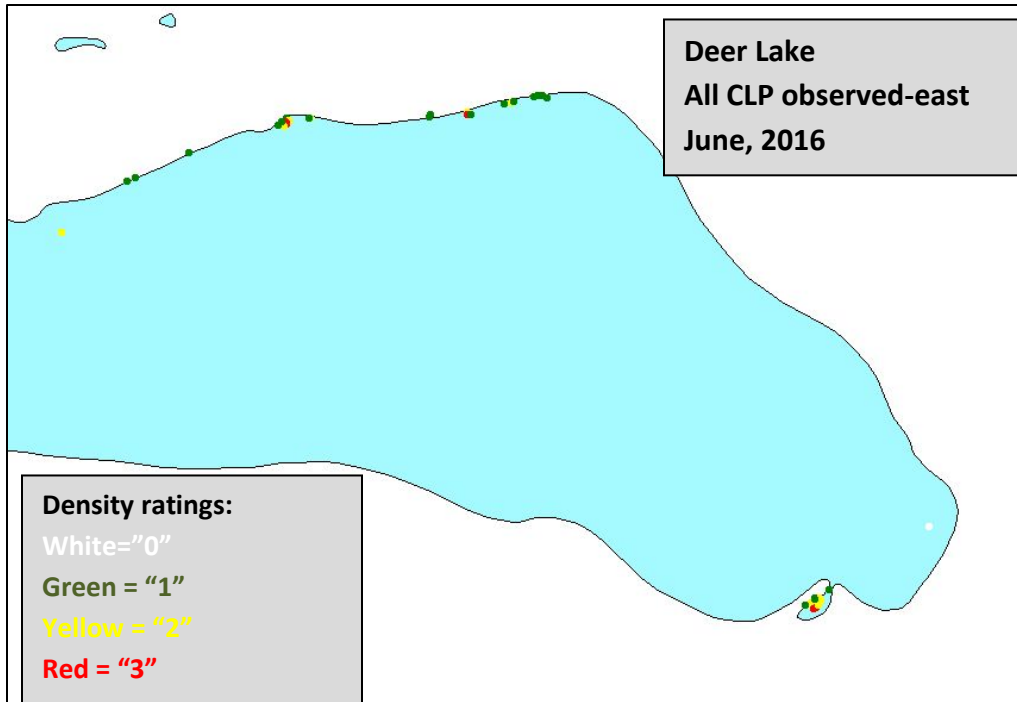


Figure 11: All CLP observed with density in the eastern portion of Deer Lake-June, 2016.

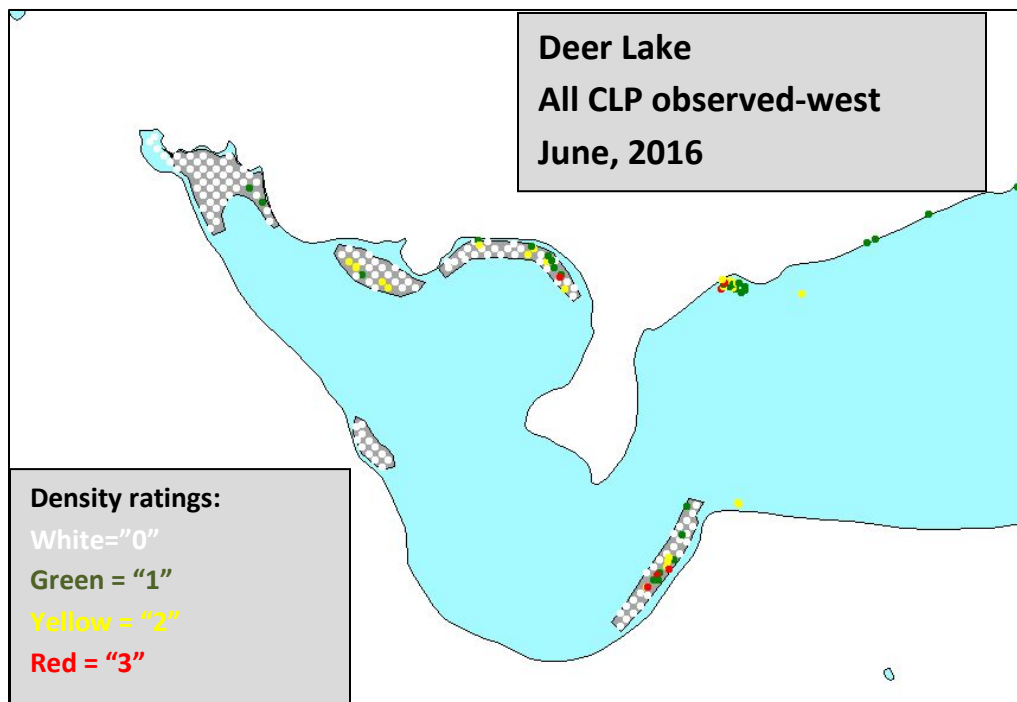
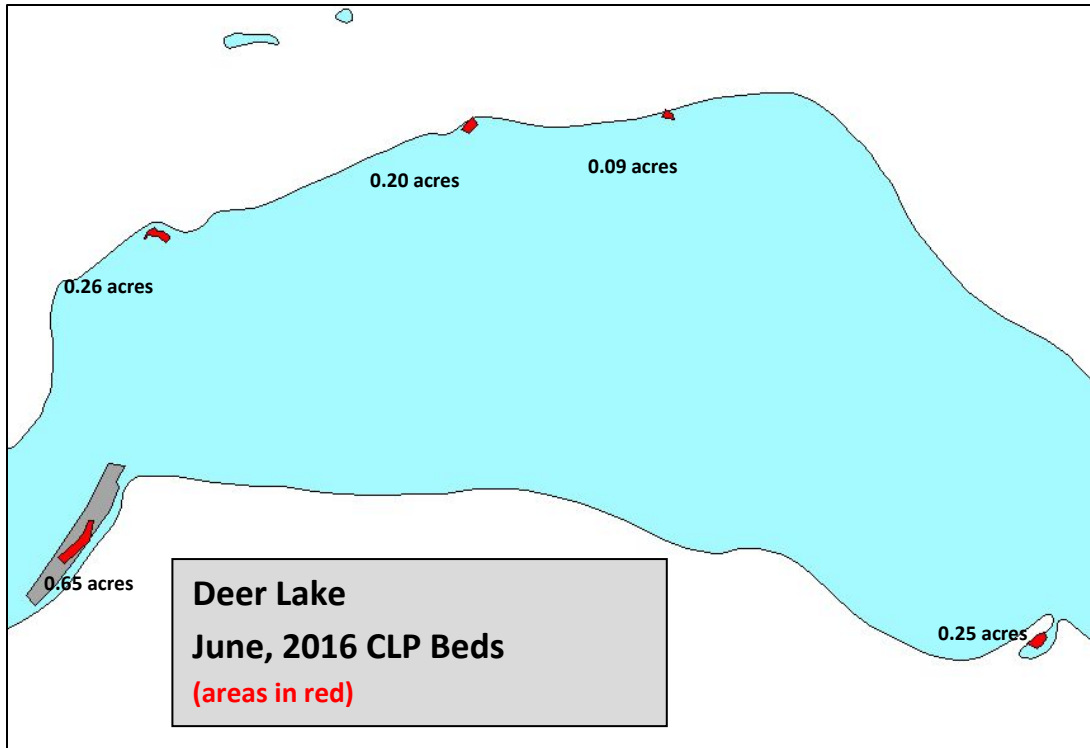


Figure 12: All CLP observed with density in the western portion of Deer Lake-June, 2016.



**Figure 13: Areas that constitute a bed of CLP in Deer Lake-June, 2016.**

### Turion Analysis

The 2016 turion analysis showed an increase in the turion density in beds A, B, and C. The increase in beds B and C may reflect the presence of CLP growing in these beds after treatment. The CLP plants likely created new turions into the beds after they died off in July. In bed A, the increase is unknown since little CLP was sampled or observed in the post treatment survey. Bed A has historically had high density of CLP and it may be that clumps of turions are present and sampling variation didn't reveal this density in previous years. In all, the overall turion density increased in 2016, which is not desirable in CLP management. It is assumed that due to these densities, CLP growth will return quite dense in these beds in 2017.

Table 6 summarizes the turion density comparison from 2013 to 2016. Figures 14 and 15 graphically show the changes.

Bed	2013 Mean (T/m <sup>2</sup> )	2014 Mean (T/m <sup>2</sup> )	2015 Mean (T/m <sup>2</sup> )	2016 Mean (T/m <sup>2</sup> )
A	77.7	63.1	39.1	83
B	153.6	46.1	96.75	122
C	91.8	89.5	75.25	136
D	15.0	16.3	32.25	5
E	71.0	18.6	55.3	31
All Beds	88.8	52.0	61.1	84.7

**Table 6: Turion density in each bed 2013 through 2016.**

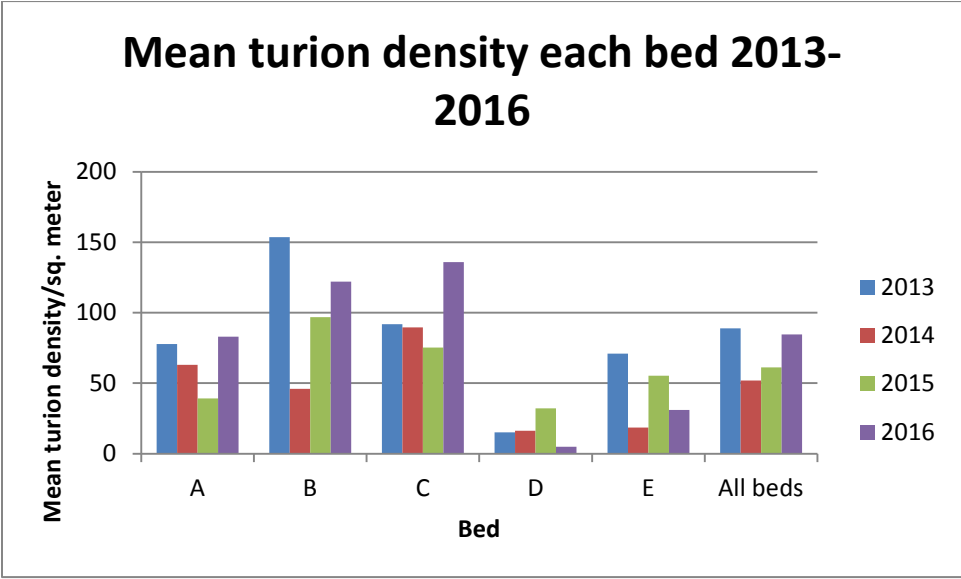


Figure 14: Turion densities by bed for comparison 2013 through 2016.

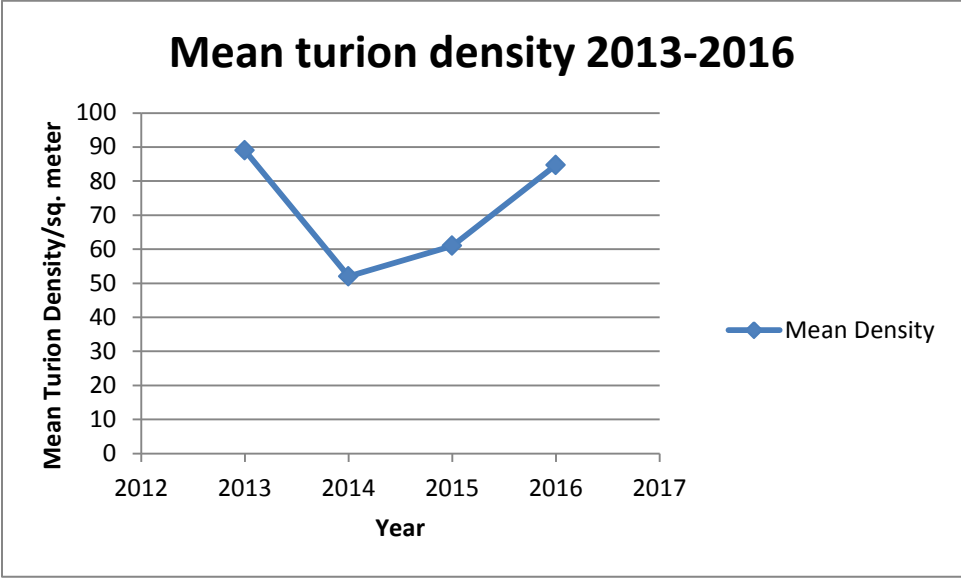
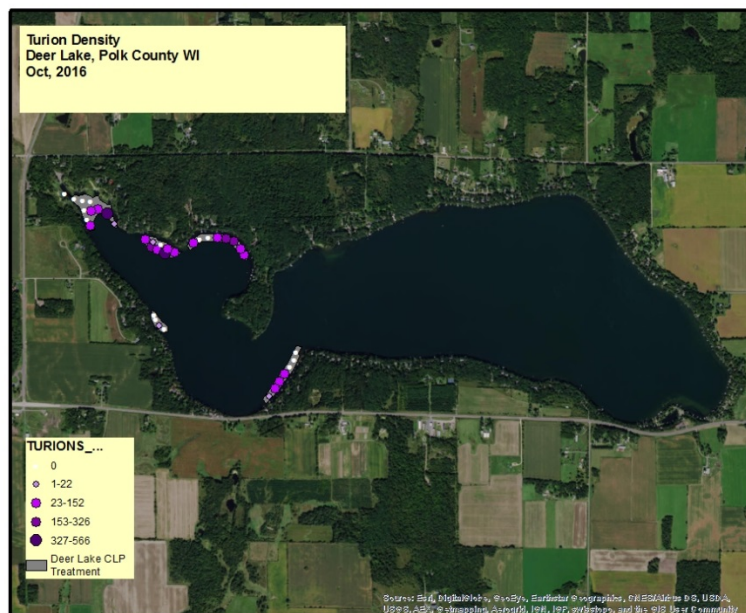


Figure 15: Change in turion density, all beds 2013 through 2016.





**Figure 16: Map on turion density by bed, 2016.**

## Discussion

The 2016 CLP herbicide treatment was successful at reducing CLP growth occurring in 2016. The frequency of occurrence of CLP was significantly reduced according to a chi-square analysis. When comparing the frequency of CLP just before treatment (pre-treatment survey 2016) it was much higher than after treatment (post treatment survey 2016) with a reduction of 72.1%. See Figure 6 for graphic representation.

When comparing the post treatment frequency in 2015 to the post treatment frequency in 2016, there was an increase. This shows that overall reduction from the previous year did not occur, with more CLP growing after treatment in 2016 than in 2015. The density also increased from 0.03 in 2015 to 0.3 in 2016. The frequency increase and density increase is a concern because these plants can produce turions which can remain viable for several years. The portions of the beds with CLP growth in 2016 may now contribute to new CLP growth for many more years, increasing treatment time.

Since the beds tend to fill in from turion germination, comparing the pretreatment surveys from year to year can reflect the progress that is being made. If the CLP frequency is reduced from pretreatment to pretreatment survey, then overall reduction of CLP is occurring. The pretreatment frequency in 2016 was slightly increased from 2015 and was not found to be statistically significant. The pretreatment frequency is getting small in the beds, with the overall frequency less than 50% in 2015 and 2016.

The native plant species did show a reduction in two species. This reduction could be due to the herbicide application, sample location variation or natural variation. It was the only reduction of native species and

there was a statistically significant increase in two native species from 2015 to 2016 which may indicate herbicide reduction was not the cause of the reduction.

Following the post treatment survey of the treatment beds, the CLP was mapped in all areas. Any areas that constituted a bed, the area was delineated. In years past, very little to no CLP was observed outside of the treatment area. In June, 2016, more CLP was observed than in previous years, resulting in the delineation of four beds outside of the treatment areas. There was one area within treatment Bed E that also was dense enough to delineate a bed.

The 2016 turion analysis revealed an increase in turion density compared to 2015. This may be due to the increased CLP growth after treatment. In CLP management, turion density reduction is desired. More CLP growth in 2017 can be expected as these turions germinate in the winter/spring.

## References

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Crow, Garrett E. and C. Barre Hellquist. *Aquatic and Wetland Plants of Northeastern North America*. The University of Wisconsin Press. Madison, Wisconsin. Volumes 1 and 2. 2000. 880p.

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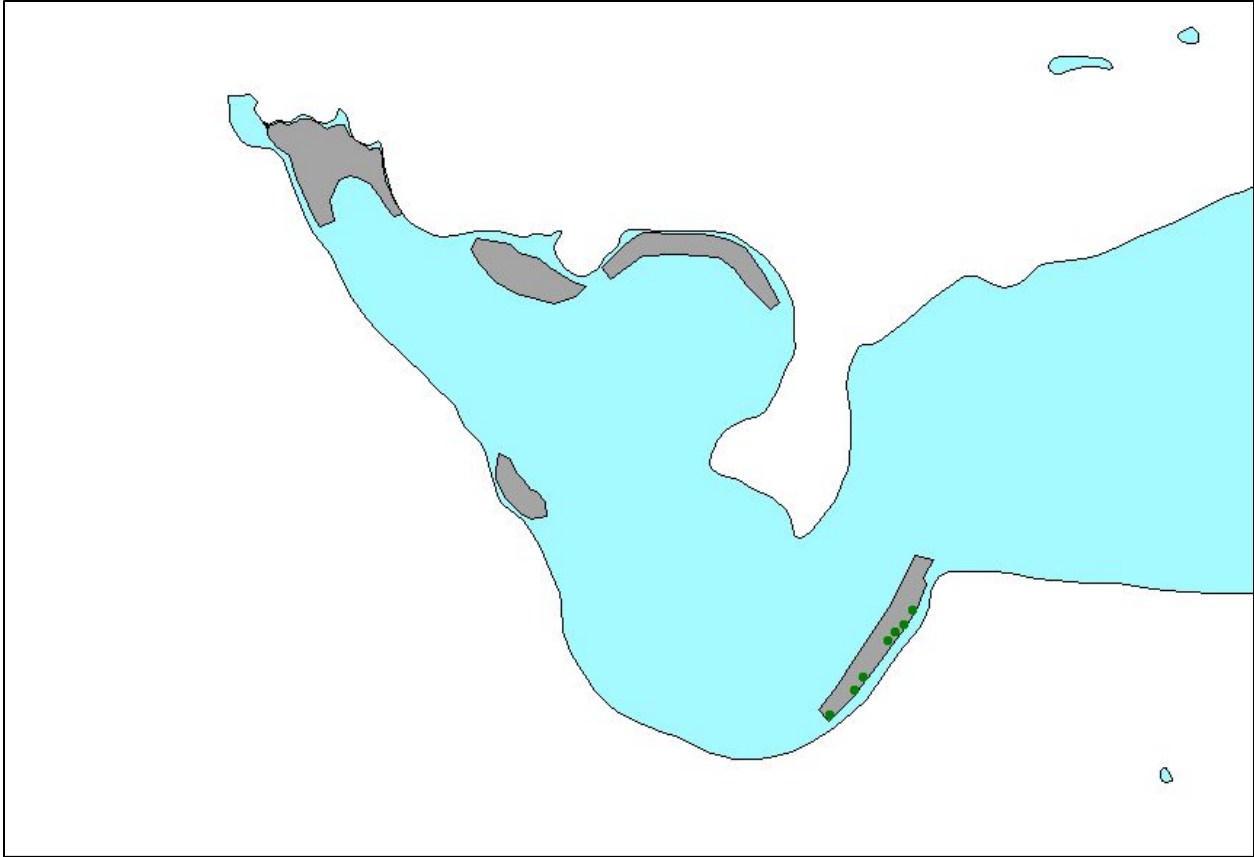
UW-Extension. Aquatic Plant Management website.

<http://www4.uwsp.edu/cnr/uwexplakes/ecology/apmguide.asp> appendix d.

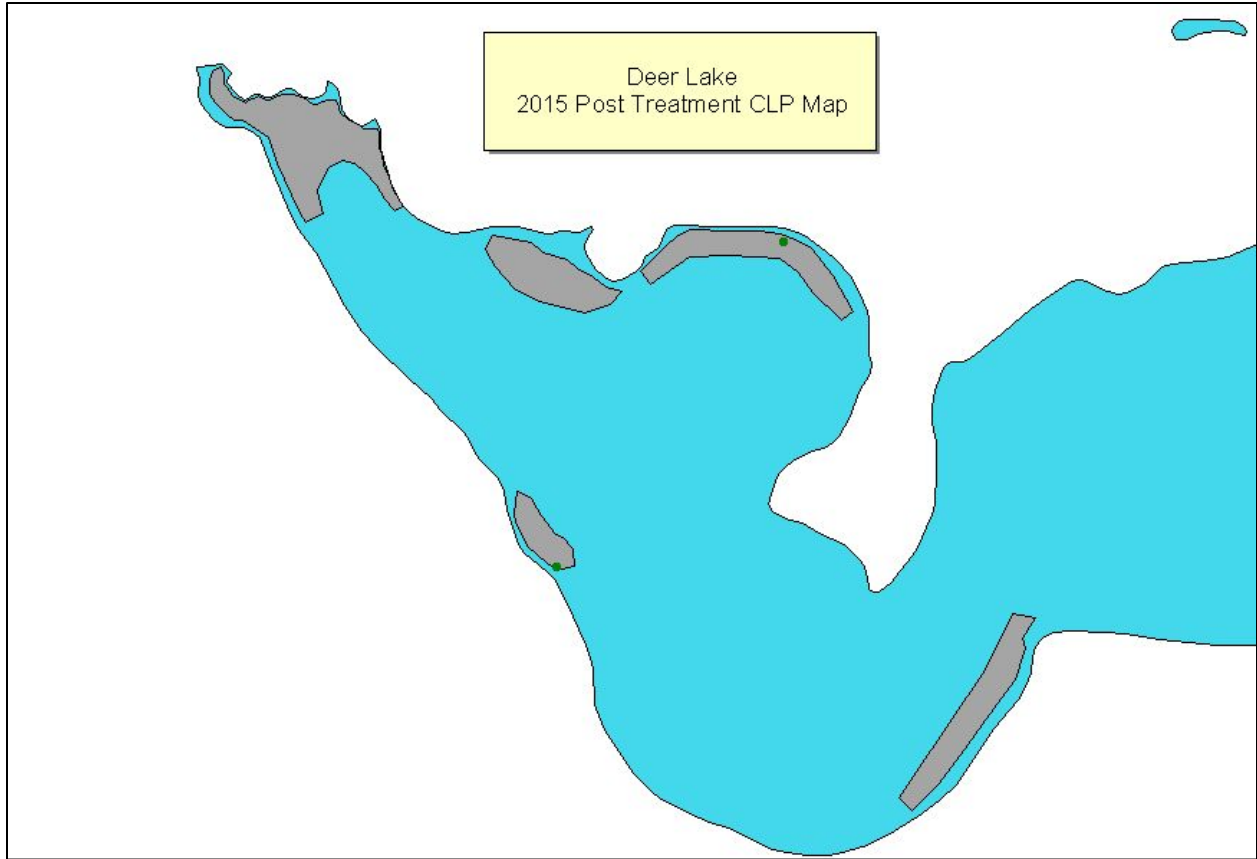
**Appendix-Native plant maps from 2015 and 2016.**



*Bidens beckii*-Water marigold-2015

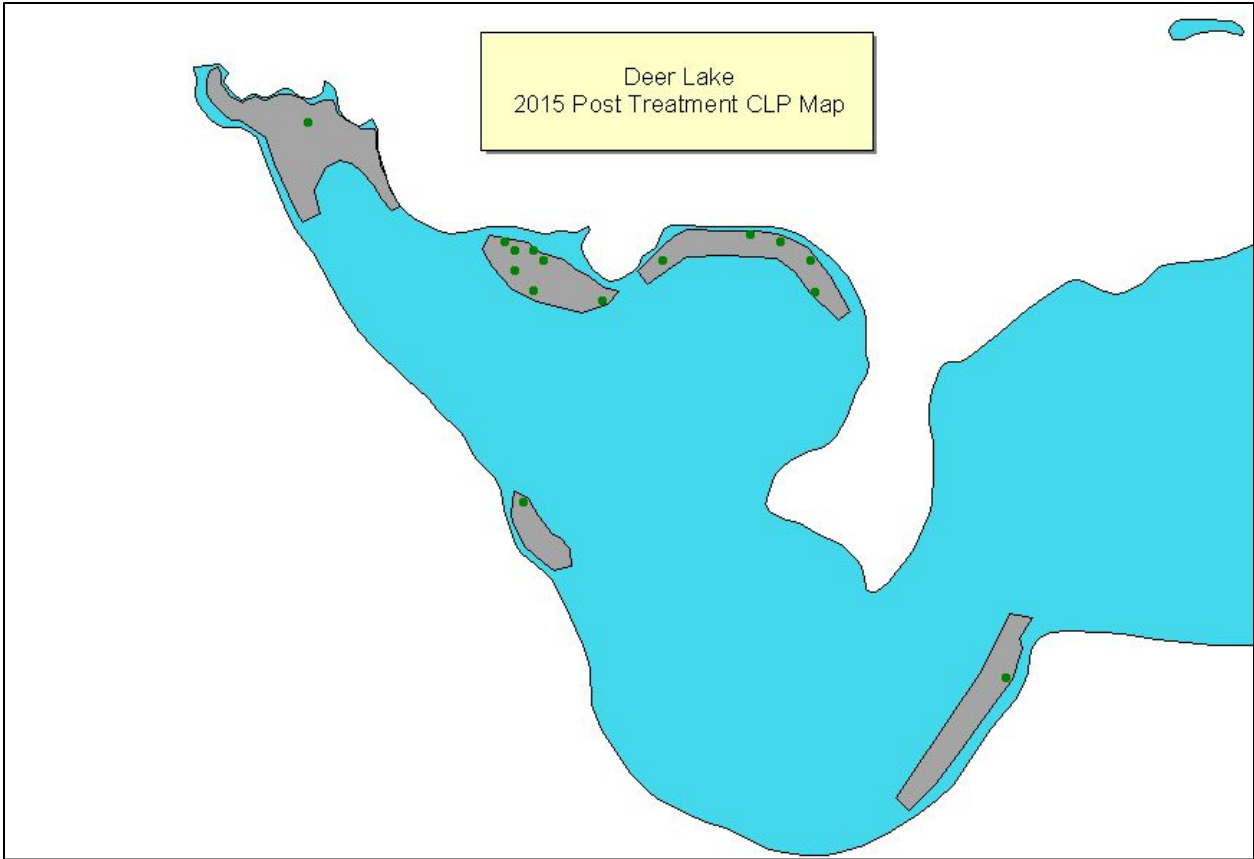


*Bidens beckii*-Water marigold-2016

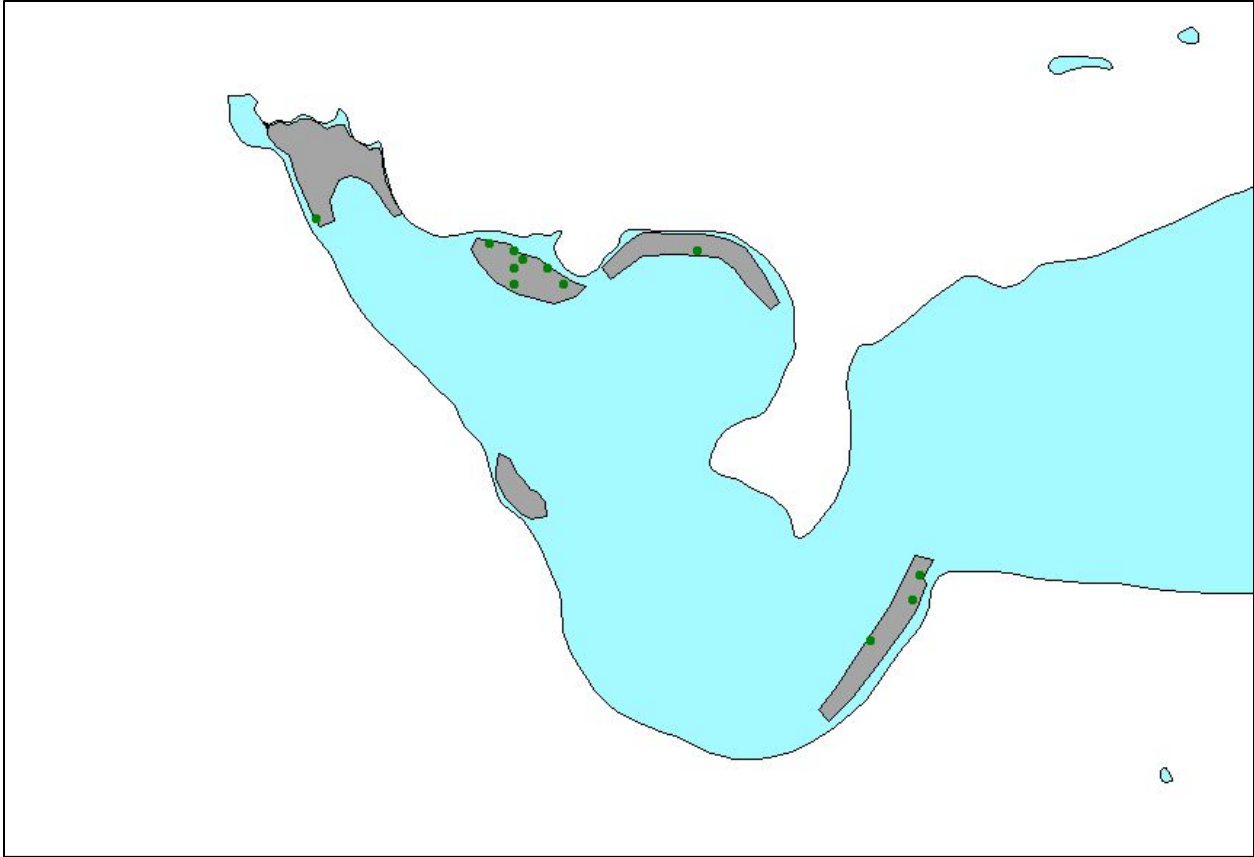


*Vallisneria americana*-Wild Celery-2015

*Vallisneria americana*-Wild Celery not sampled in 2016

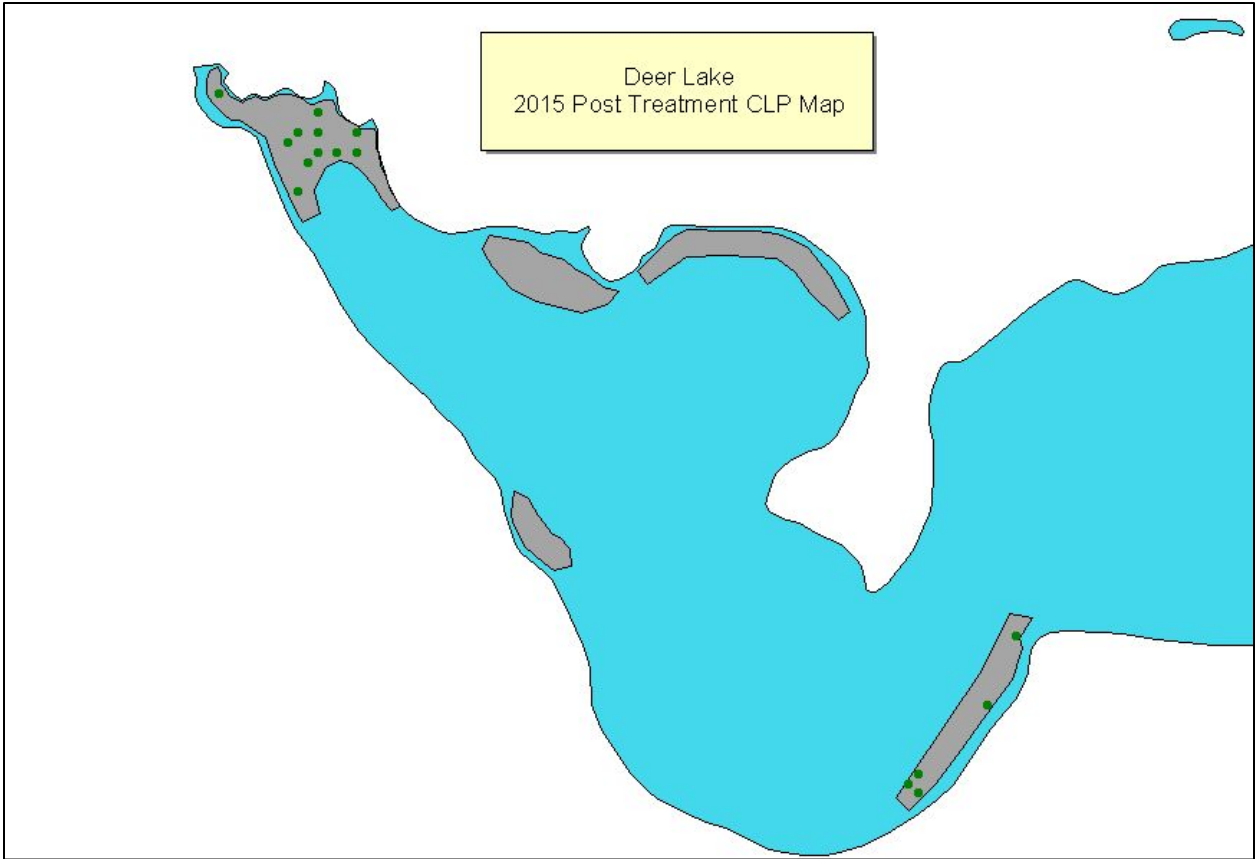


*Chara sp.*-Muskgrass 2015

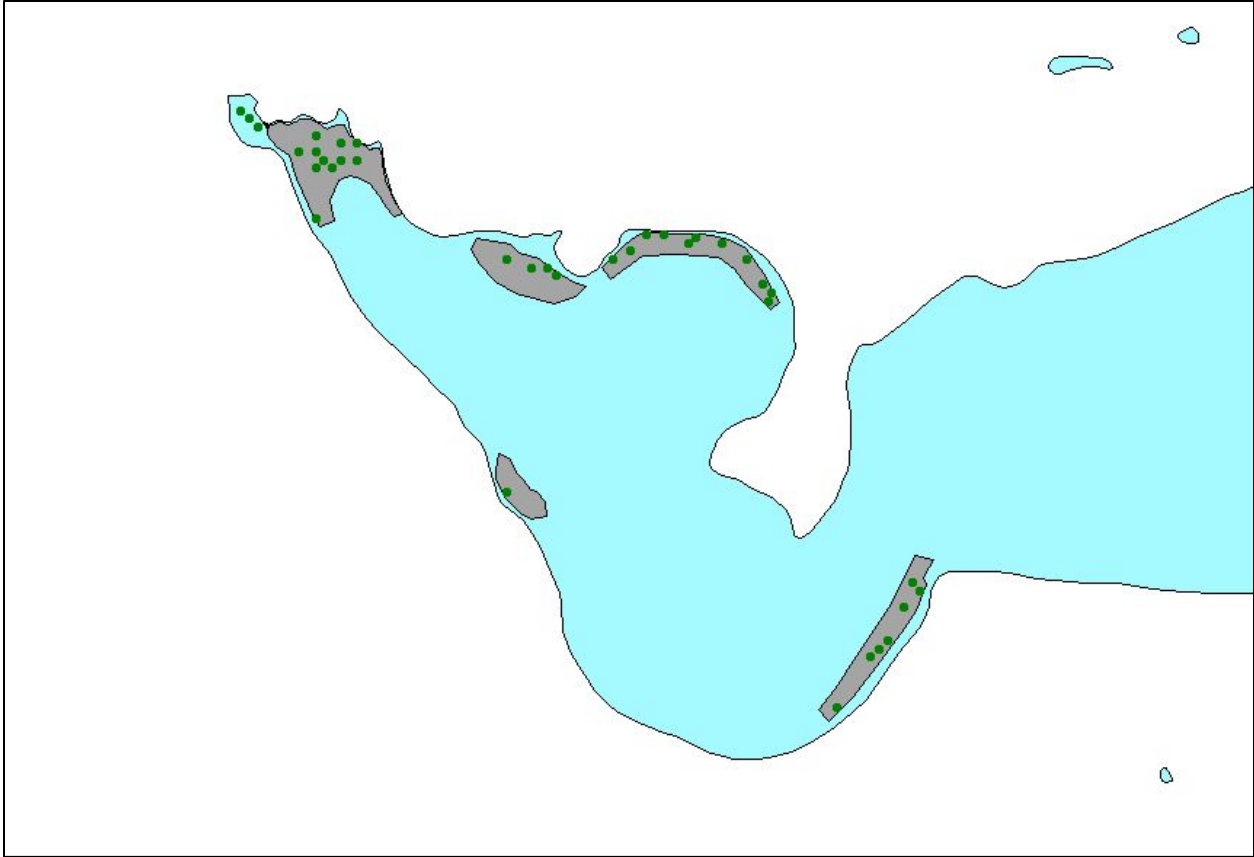


*Chara sp.*-Muskgrass-2016

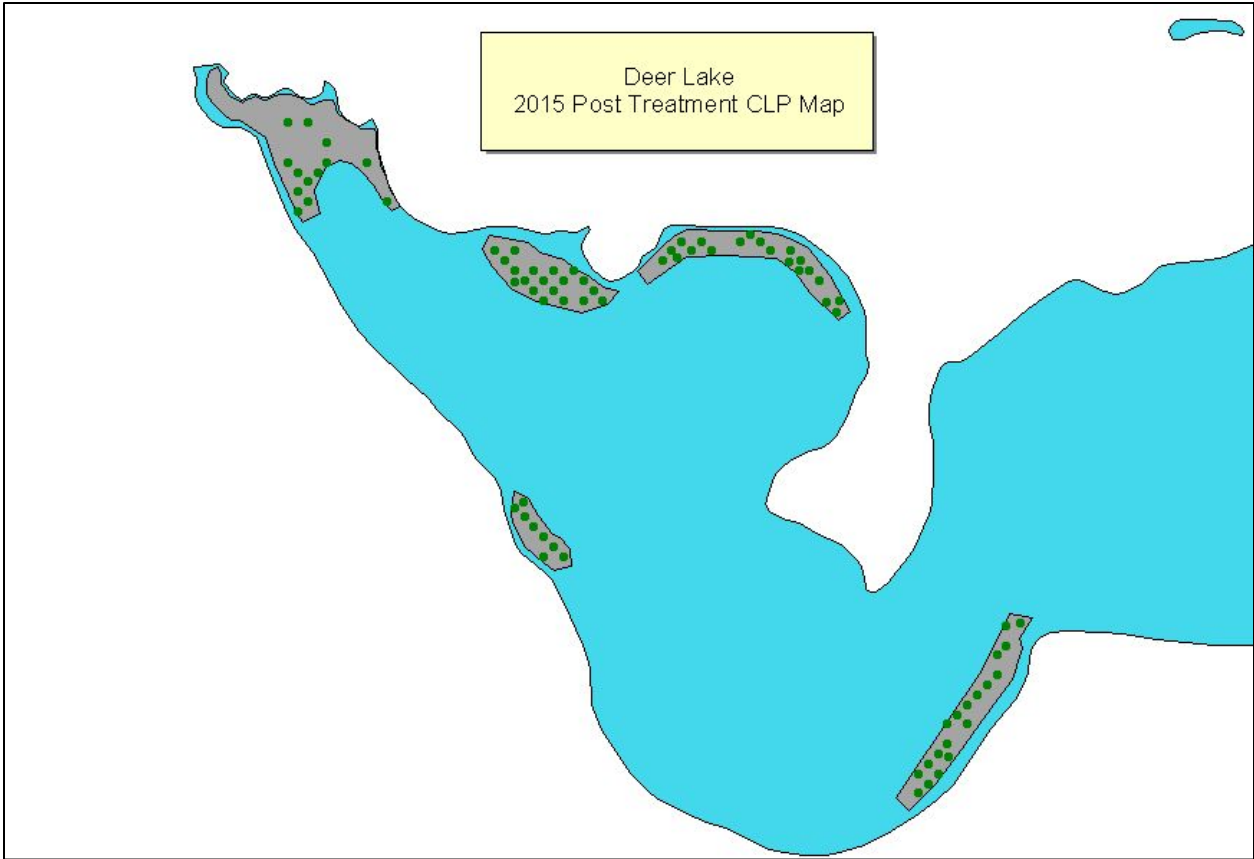




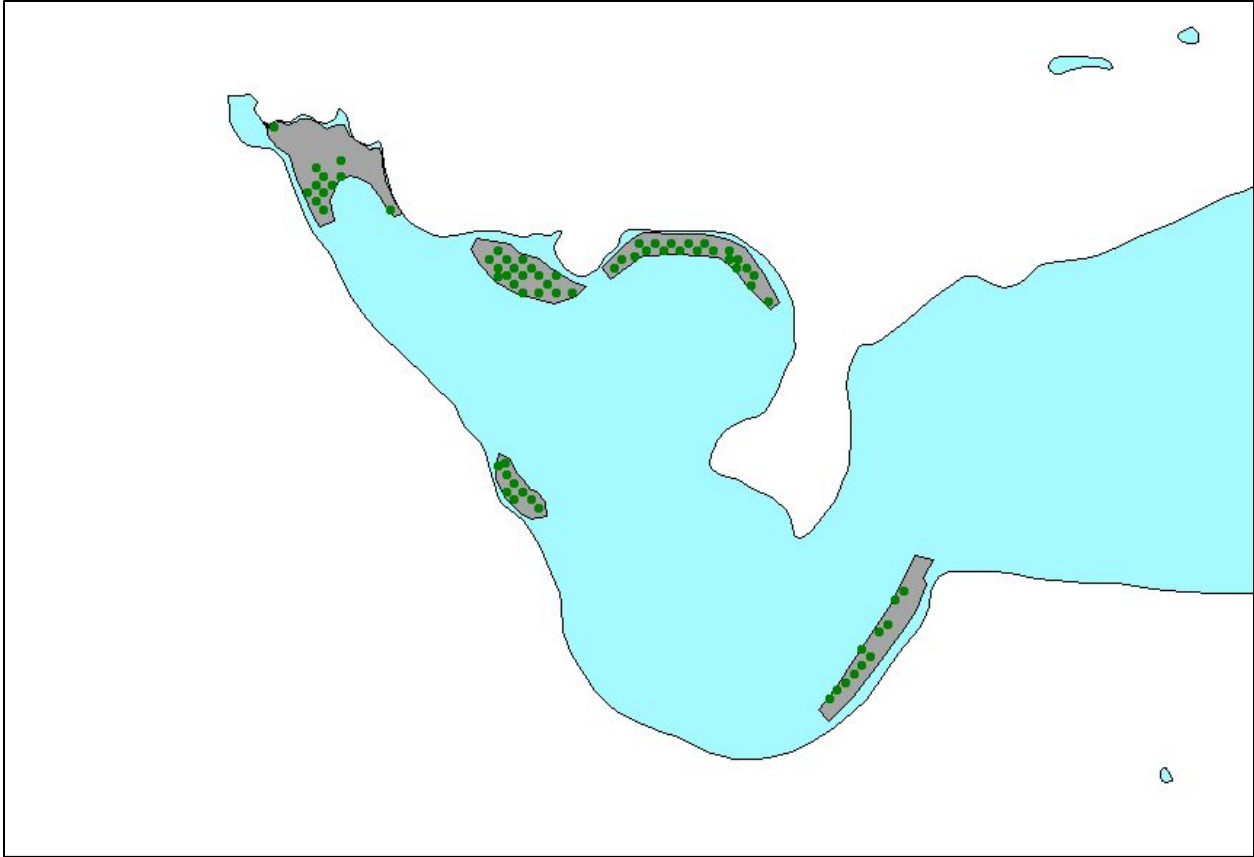
*Potamogeton richardsonii*-Clasping pondweed-2015



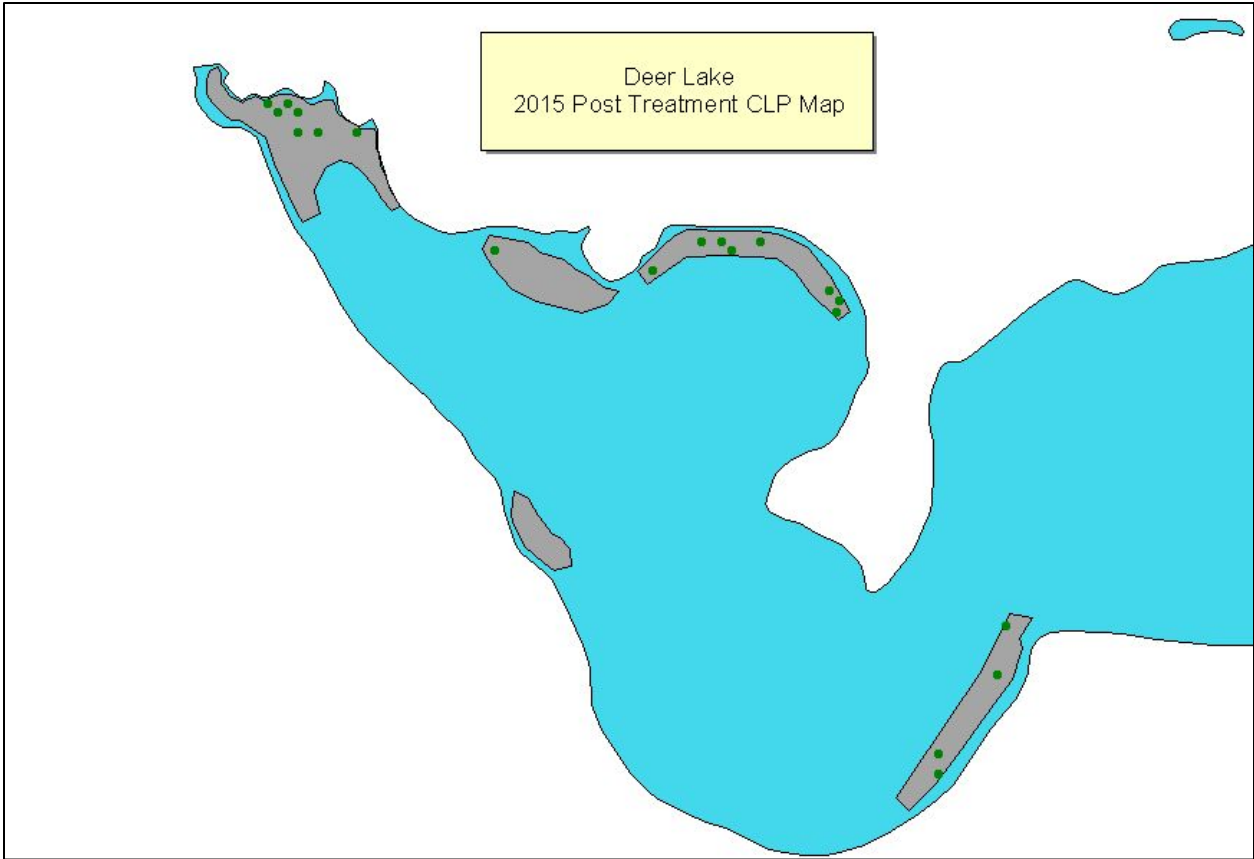
*Potamogeton richardsonii*-Clasping pondweed-2016



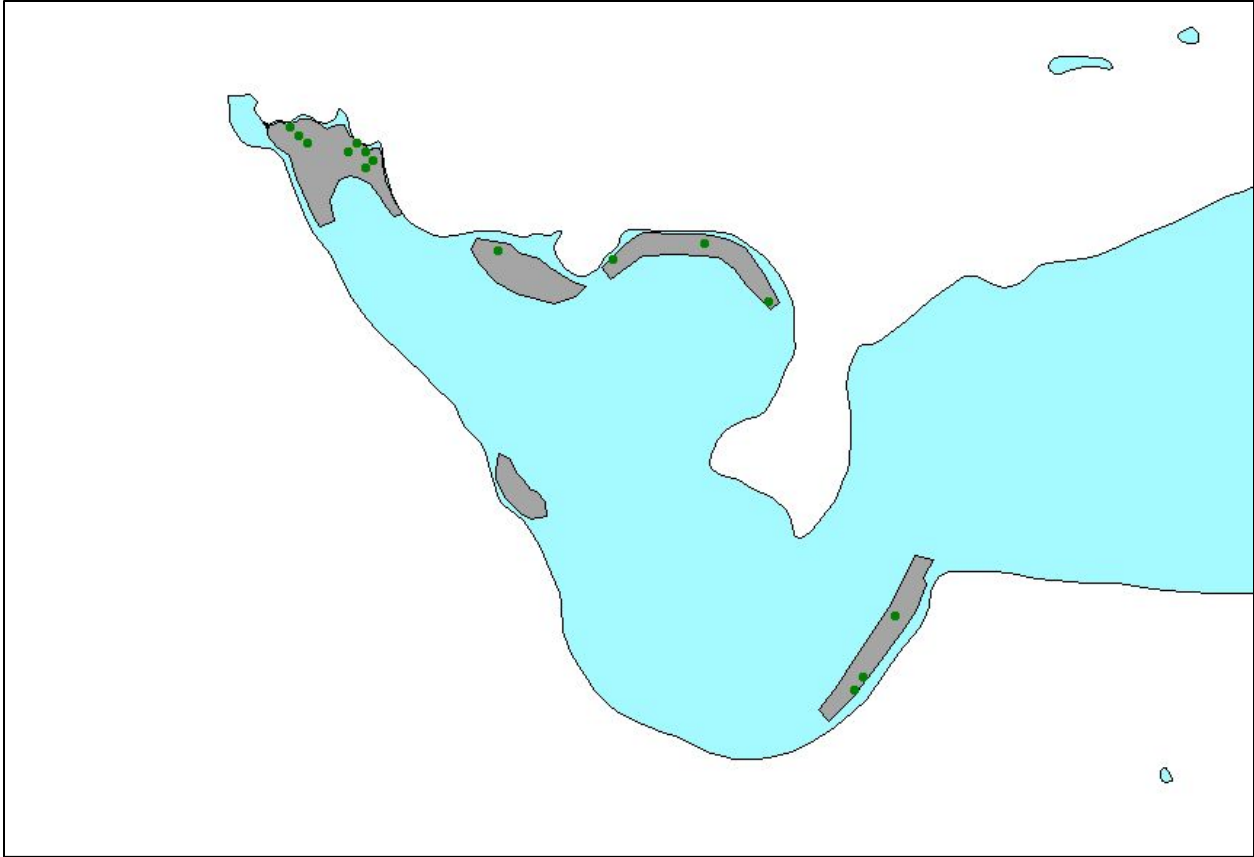
*Ceratophyllum demersum*-Coontail-2015



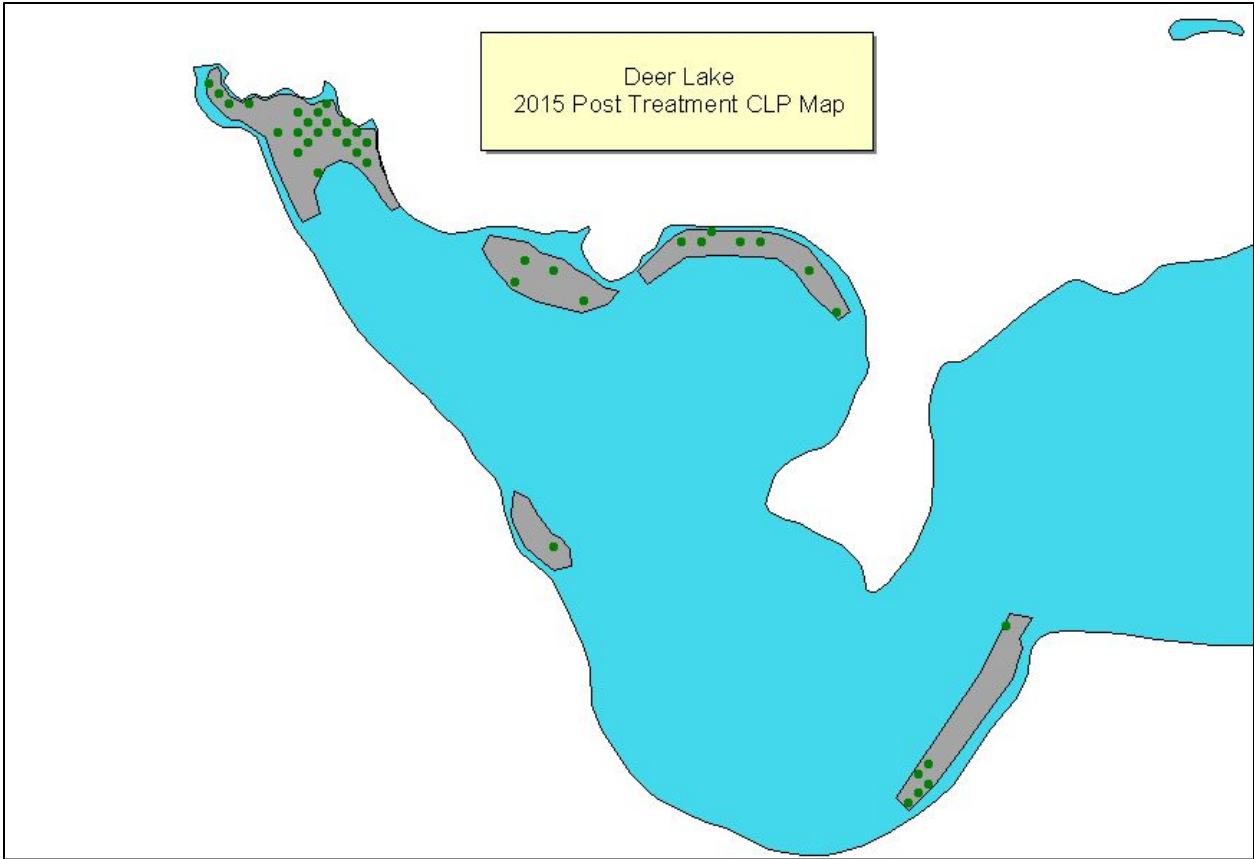
*Ceratophyllum demersum*-Coontail-2016



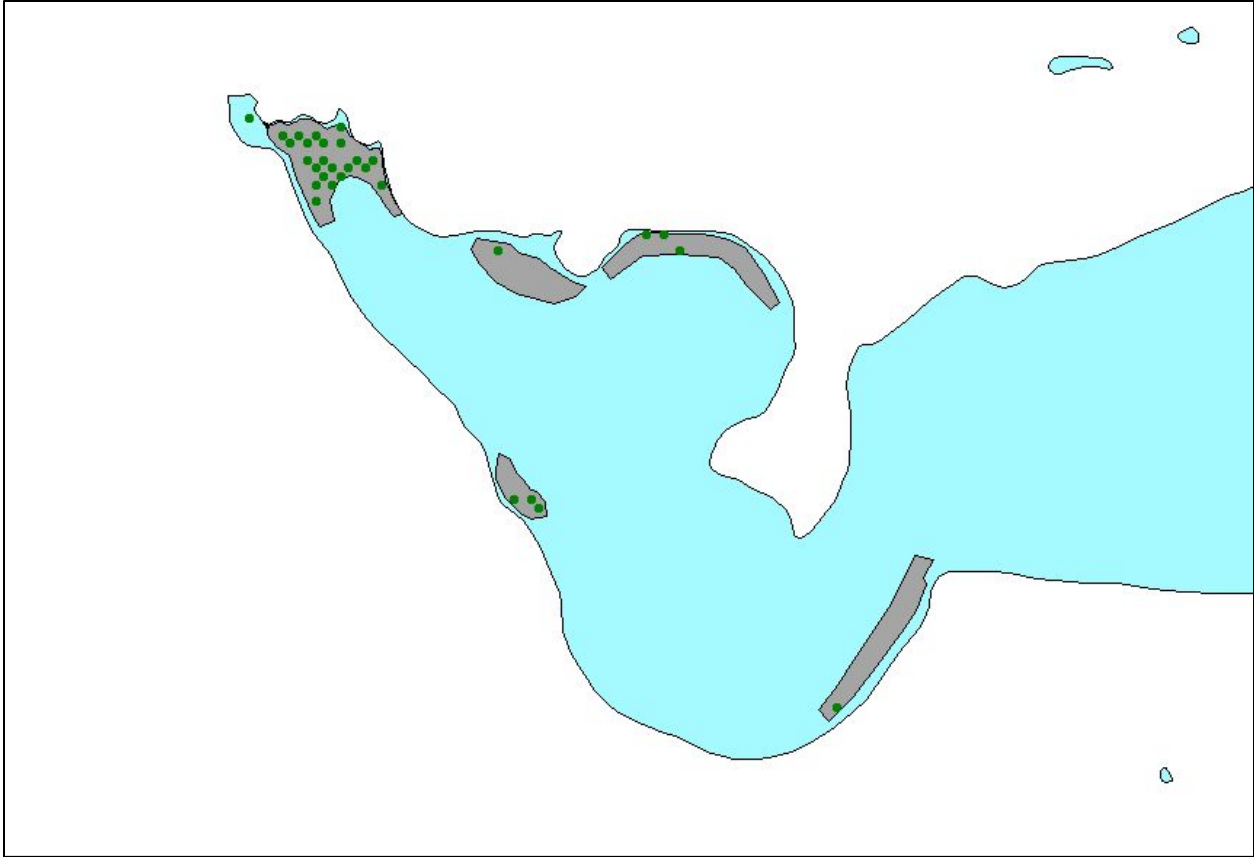
*Rununculus aquatilis*-Whitewater Crowfoot-2015



*Rununculus aquatilis*-Whitewater Crowfoot-2016

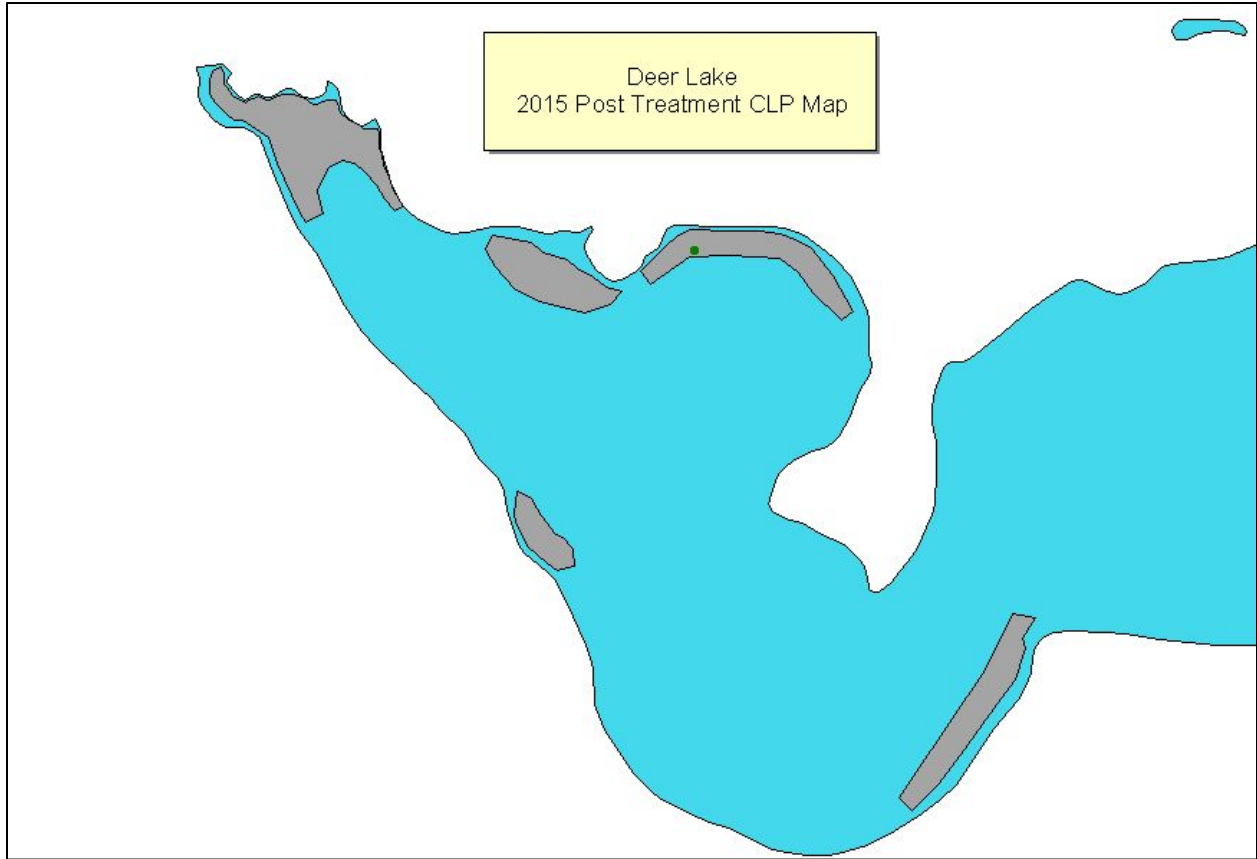


*Elodea canadensis*-Common waterweed-2015



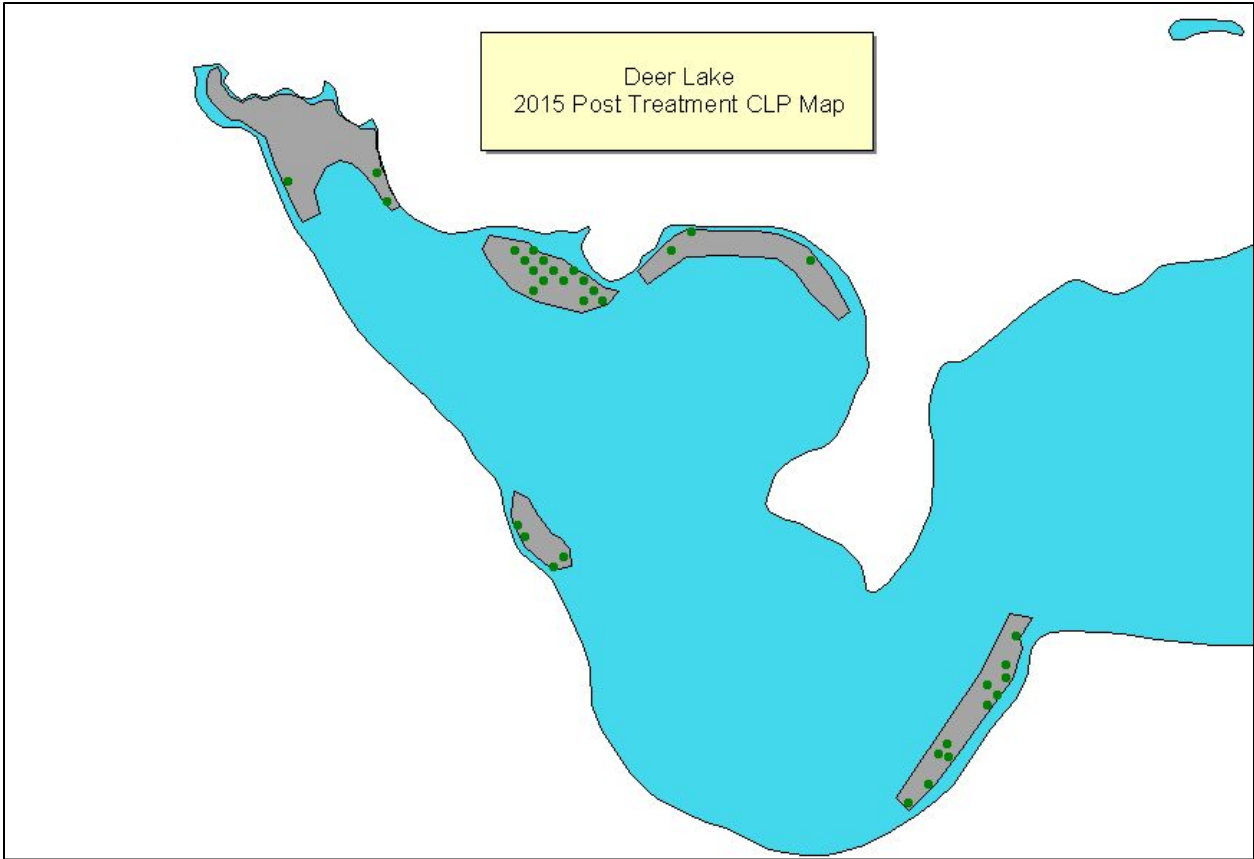
*Elodea canadensis*-Common waterweed-2016



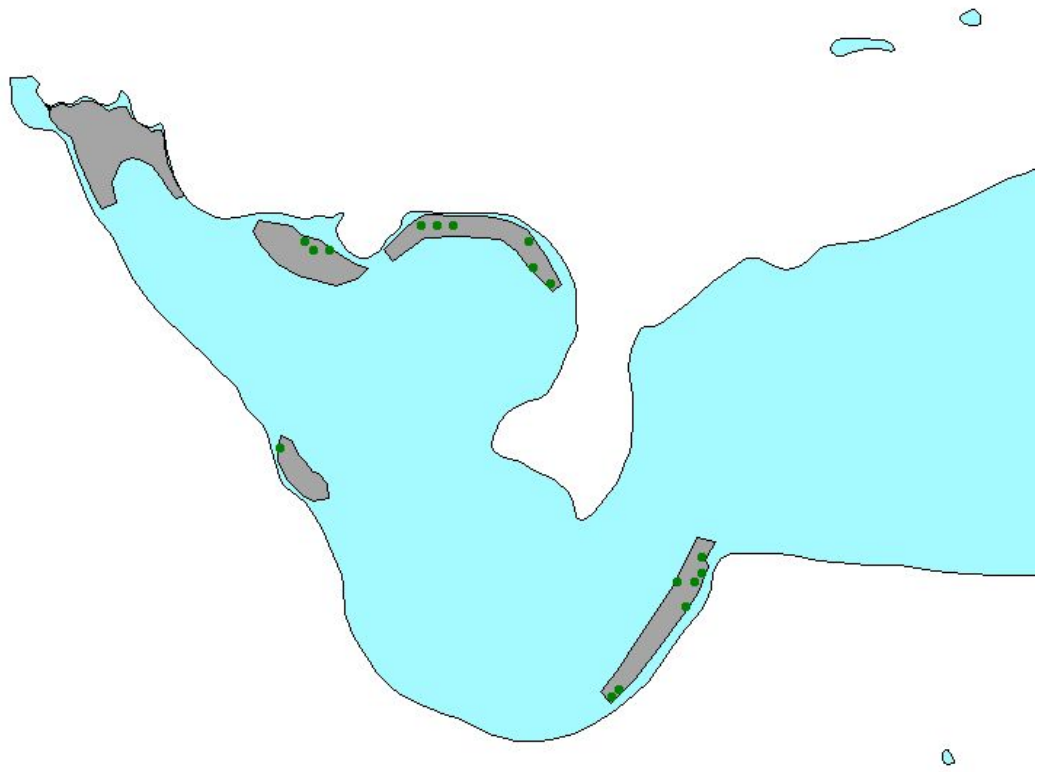


*Potamogeton robbinsii*-Fern pondweed-2015

*Potamogeton robbinsii*-Fern pondweed not sampled in 2016



*Lemna triscula*-Forked duckweed-2015

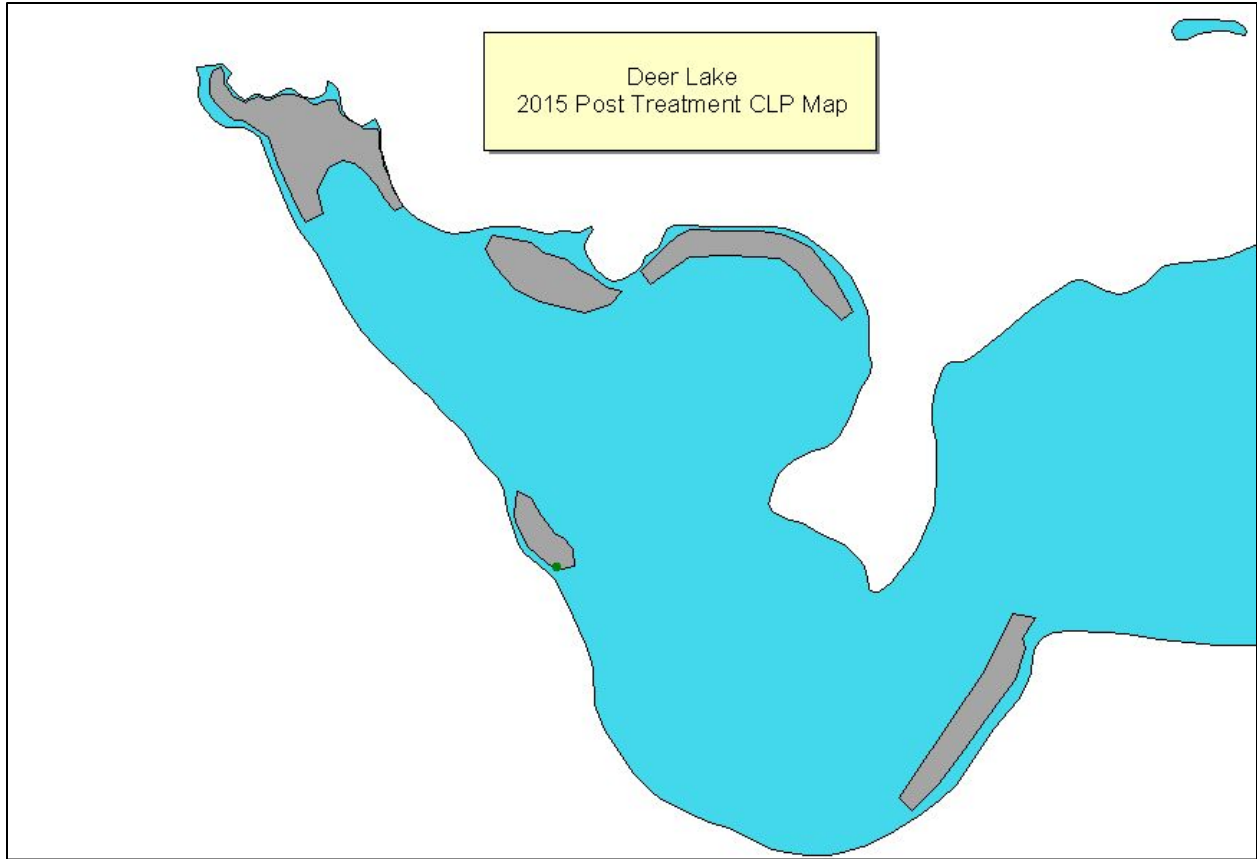


*Lemna triscula*-Forked duckweed-2016



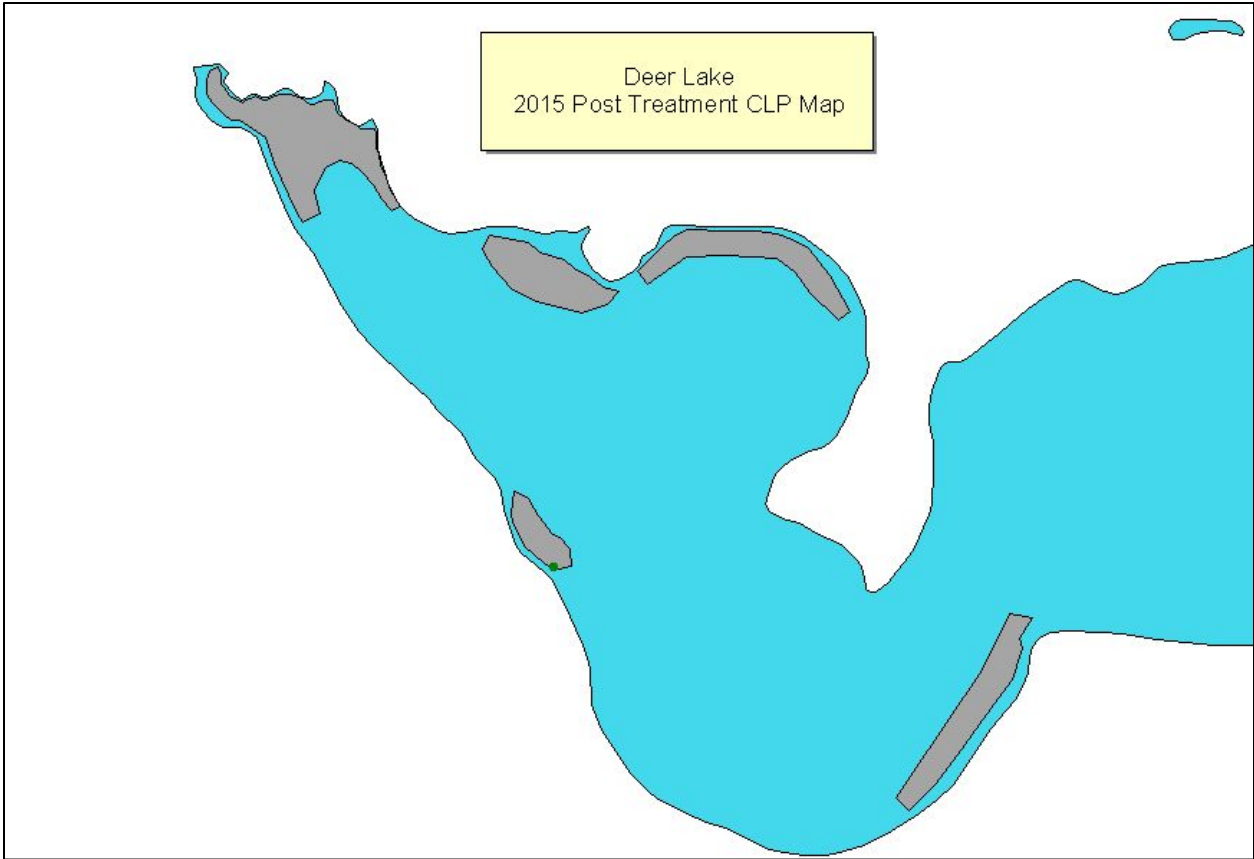
*Potamogeton illinoensis*-Illinois pondweed-2015

*Potamogeton illinoensis*-Illinois pondweed not sampled in 2016

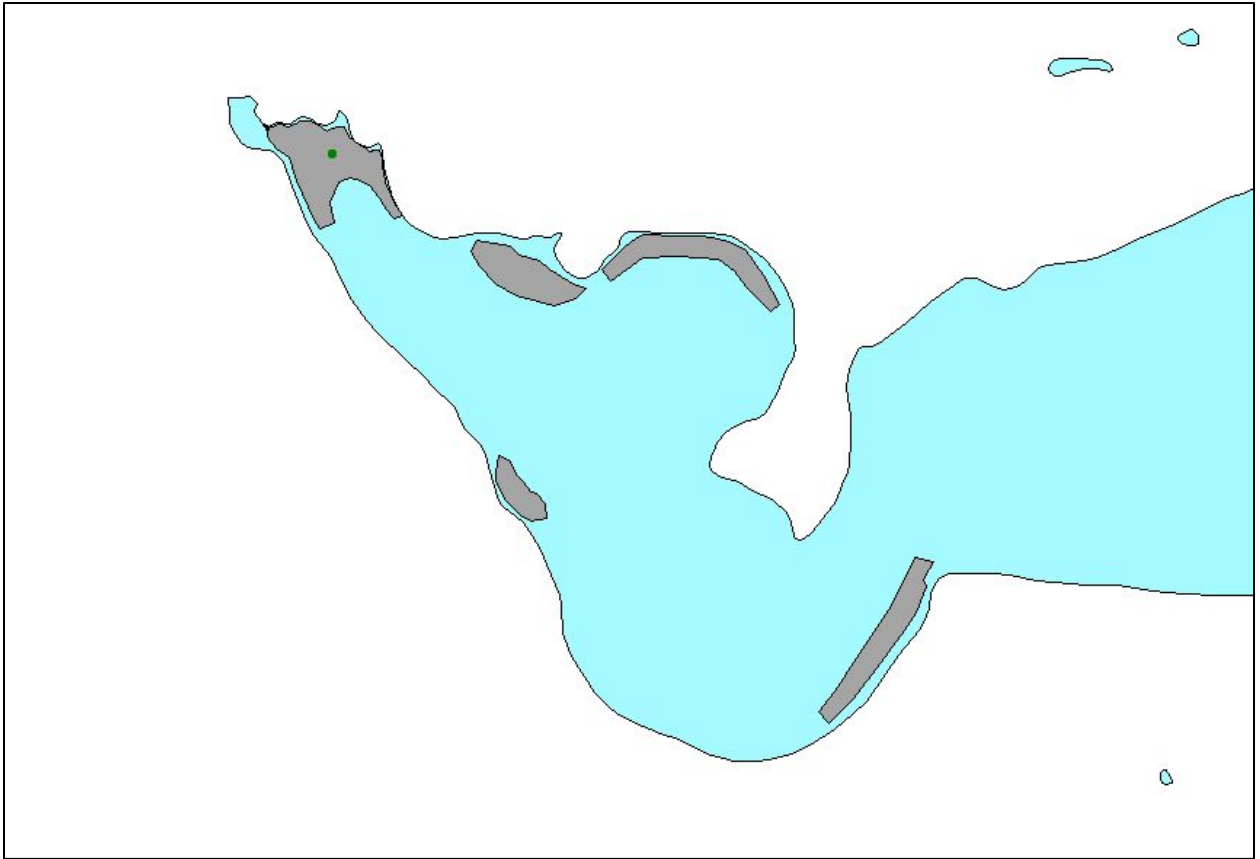


*Eleocharis acicularis*-Needle spikerush-2015

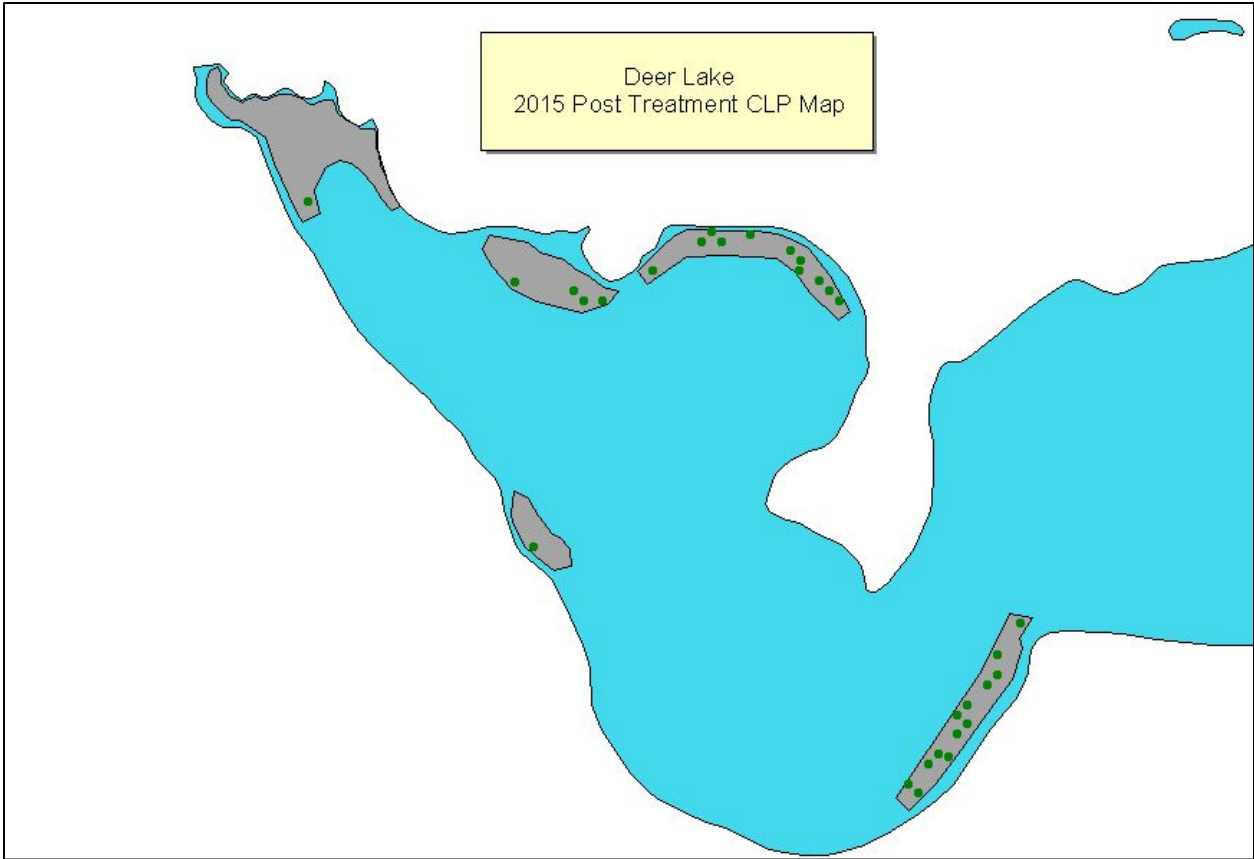
*Eleocharis acicularis*-Needle spikerush not sampled in 2016



*Nitella sp.*-Stonewort-2015

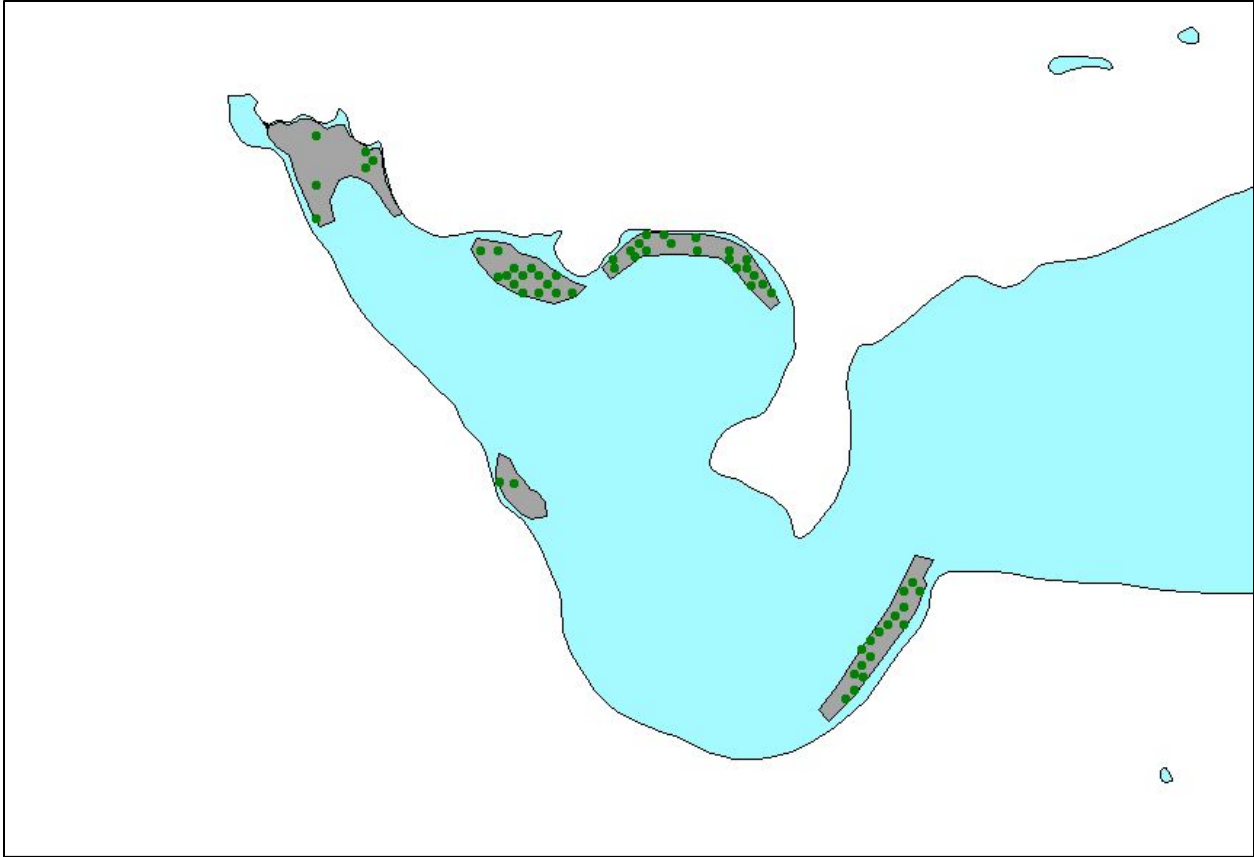


*Nitella* sp.-Stonewort-2016

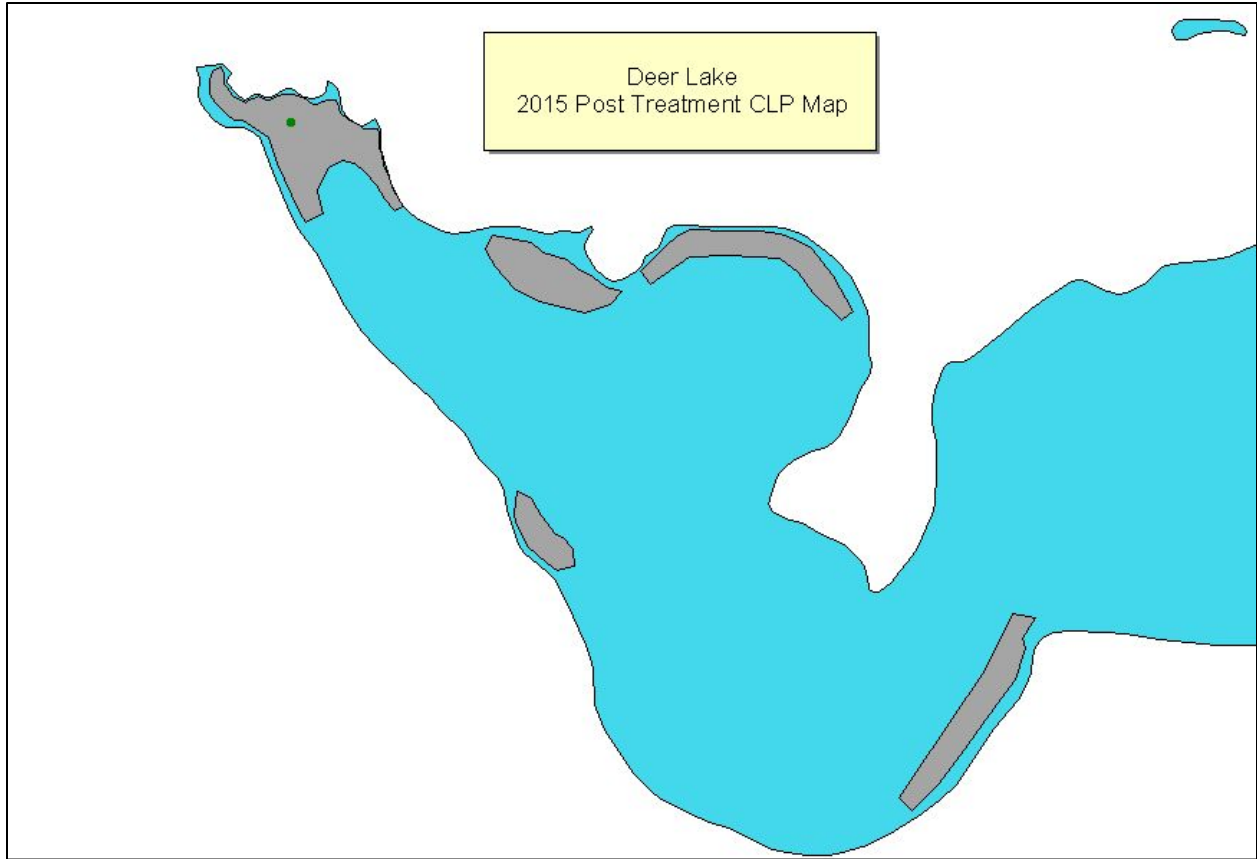


*Myriophyllum sibiricum*-Northern water milfoil-2015



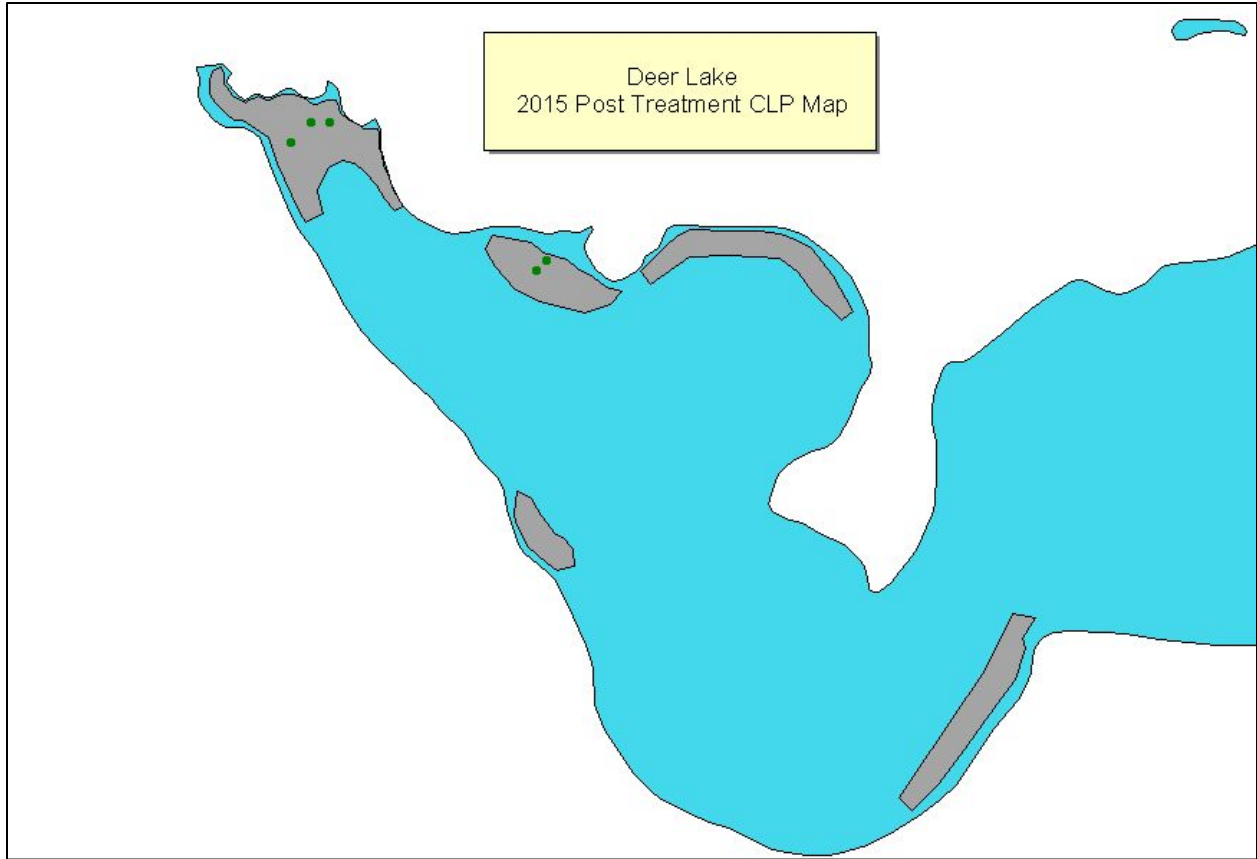


*Myriophyllum sibiricum*-Northern water milfoil-2016

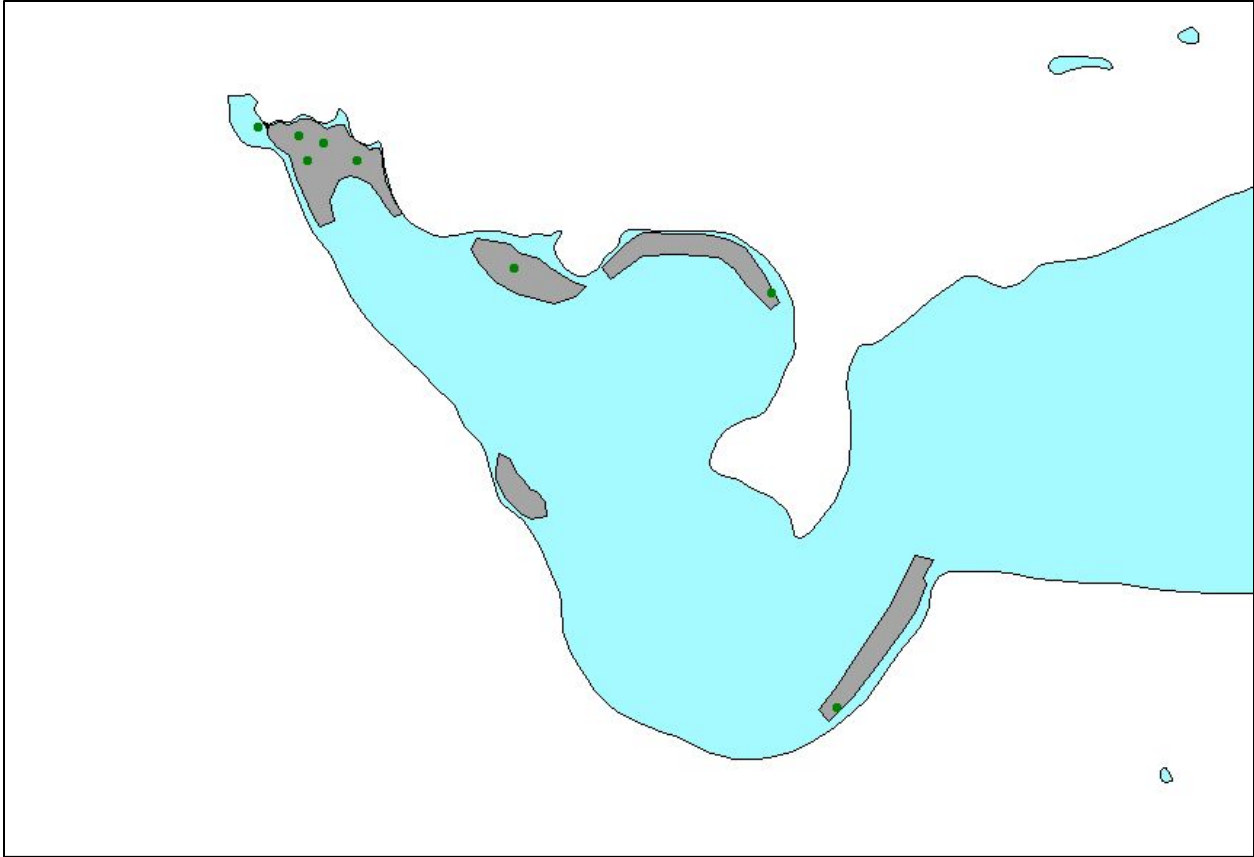


*Potamogeton epihydrous*-Ribbon leaf pondweed-2015

*Potamogeton epihydrous*-Ribbon leaf pondweed not sampled in 2016



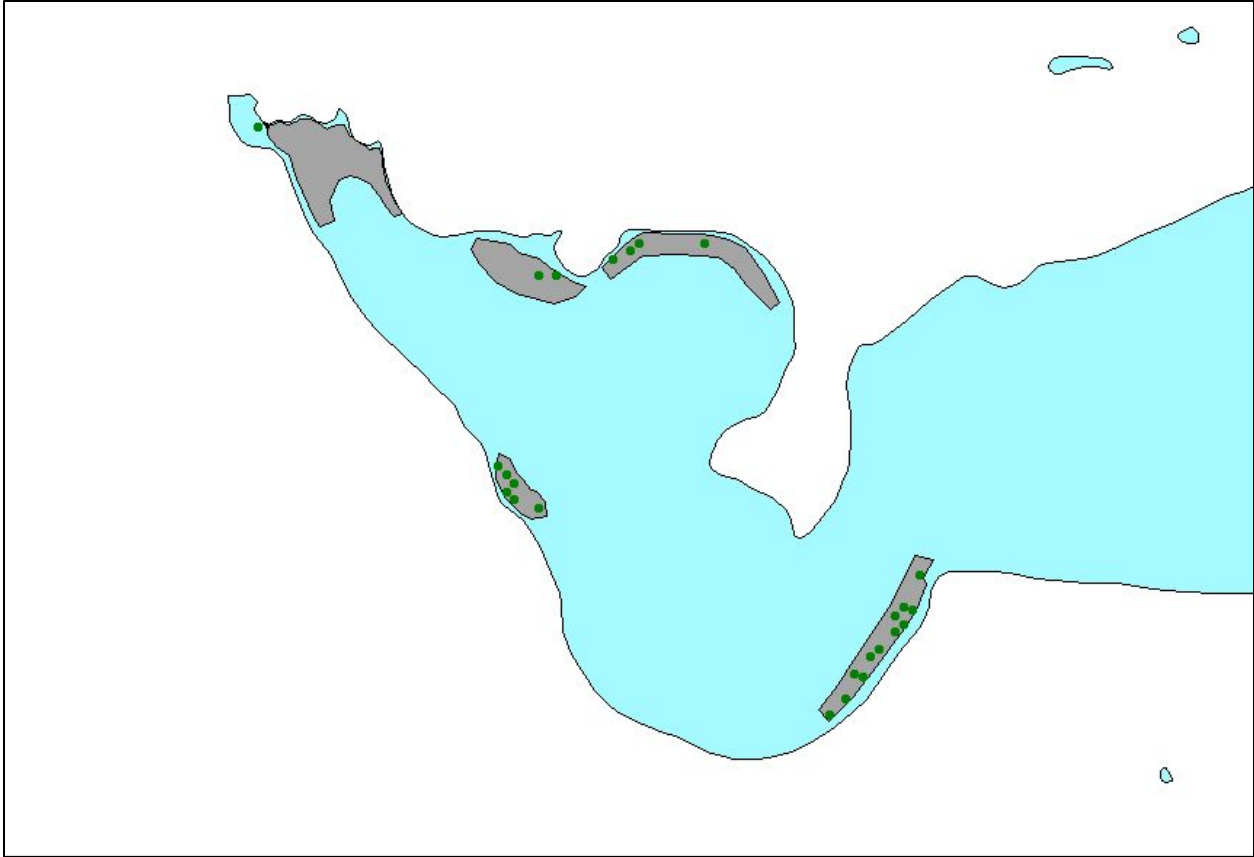
*Stuckenia pectinata*-Sago pondweed-2015



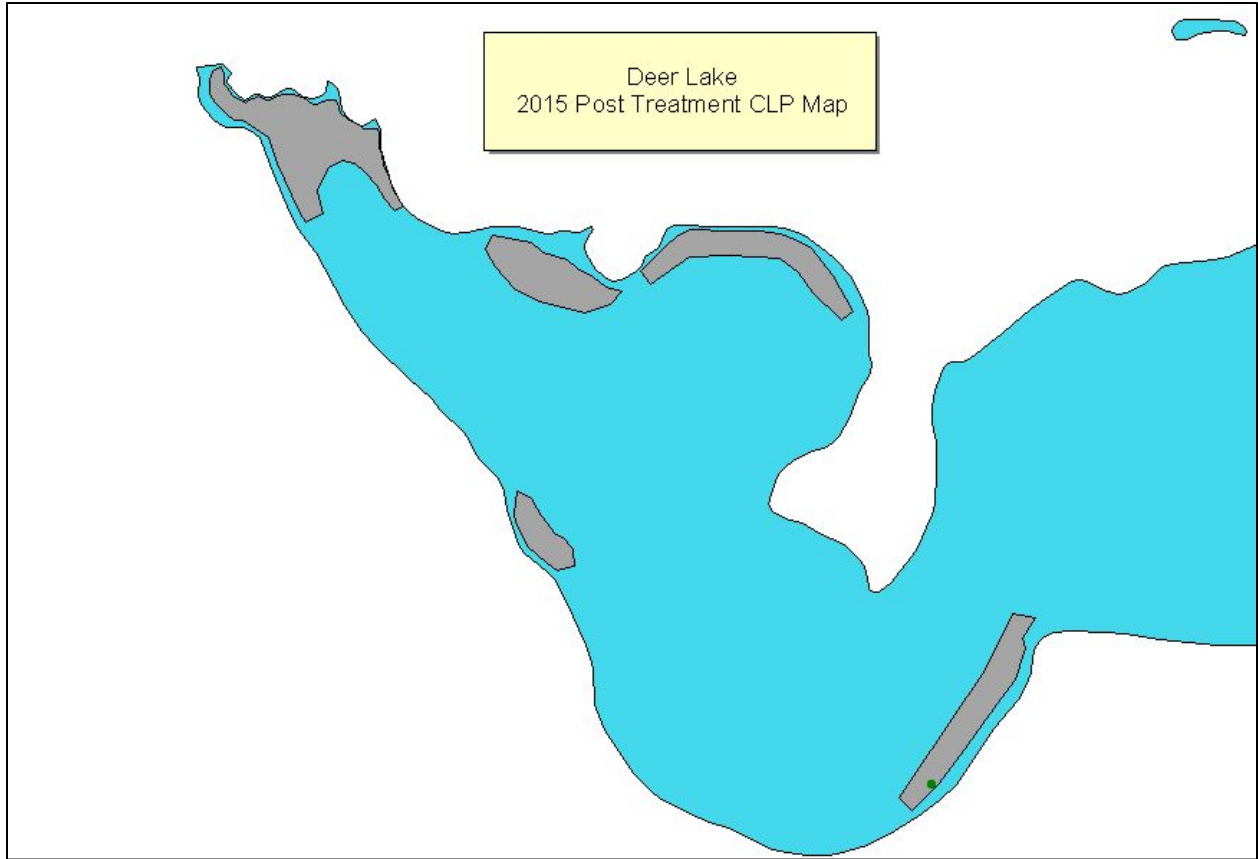
*Stuckenia pectinata*-Sago pondweed-2016



*Heteranthera dubia*-Water Stargrass-2015

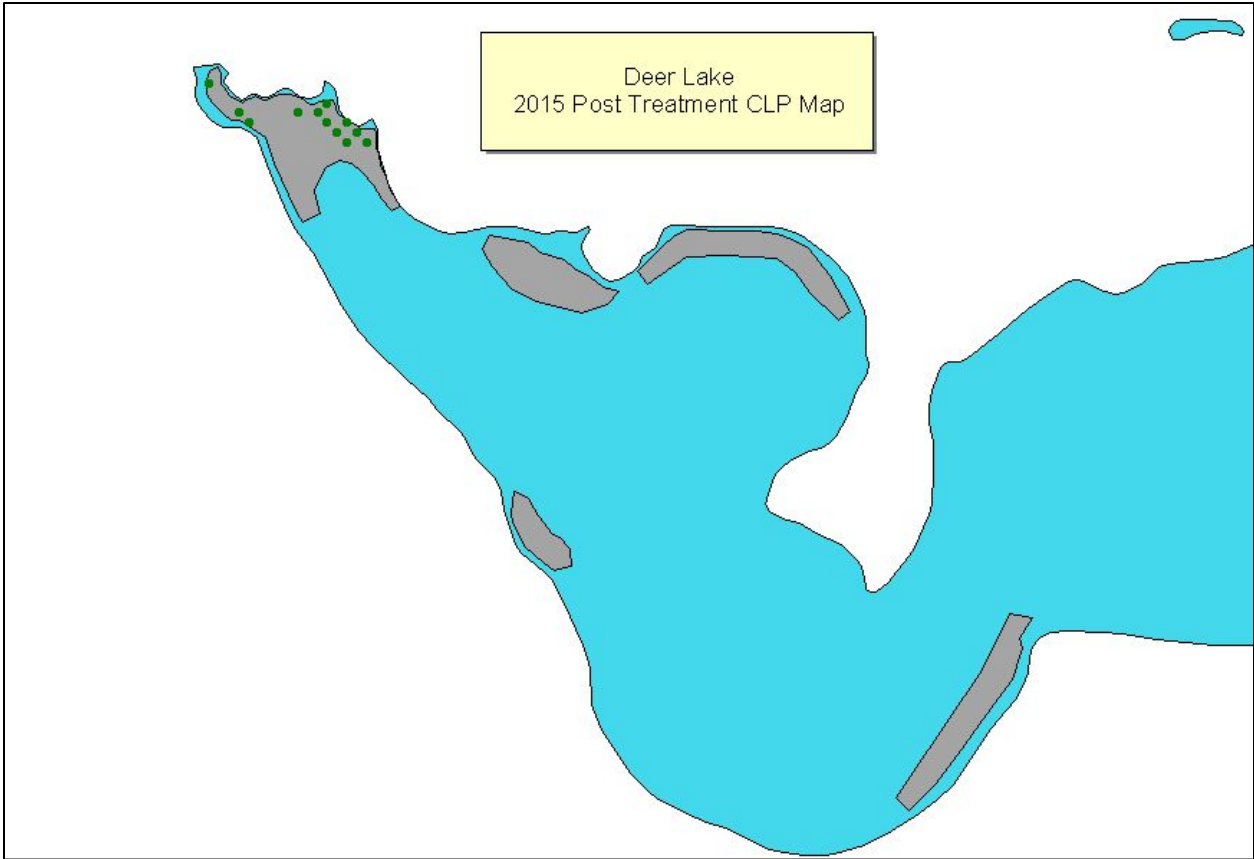


*Heteranthera dubia*-Water Stargrass-2016



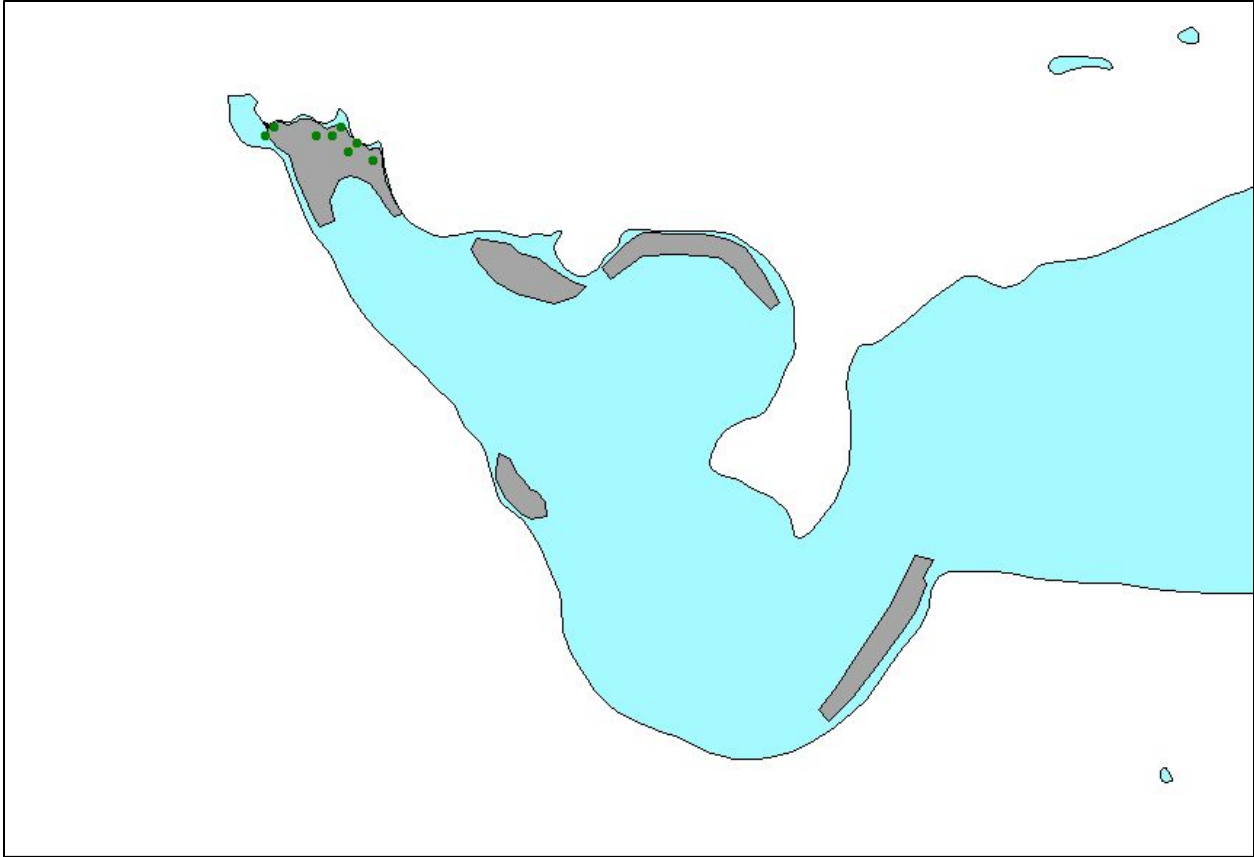
*Potamogeton gramineus*-Variable pondweed-2015

*Potamogeton gramineus*-Variable pondweed not sampled in 2016

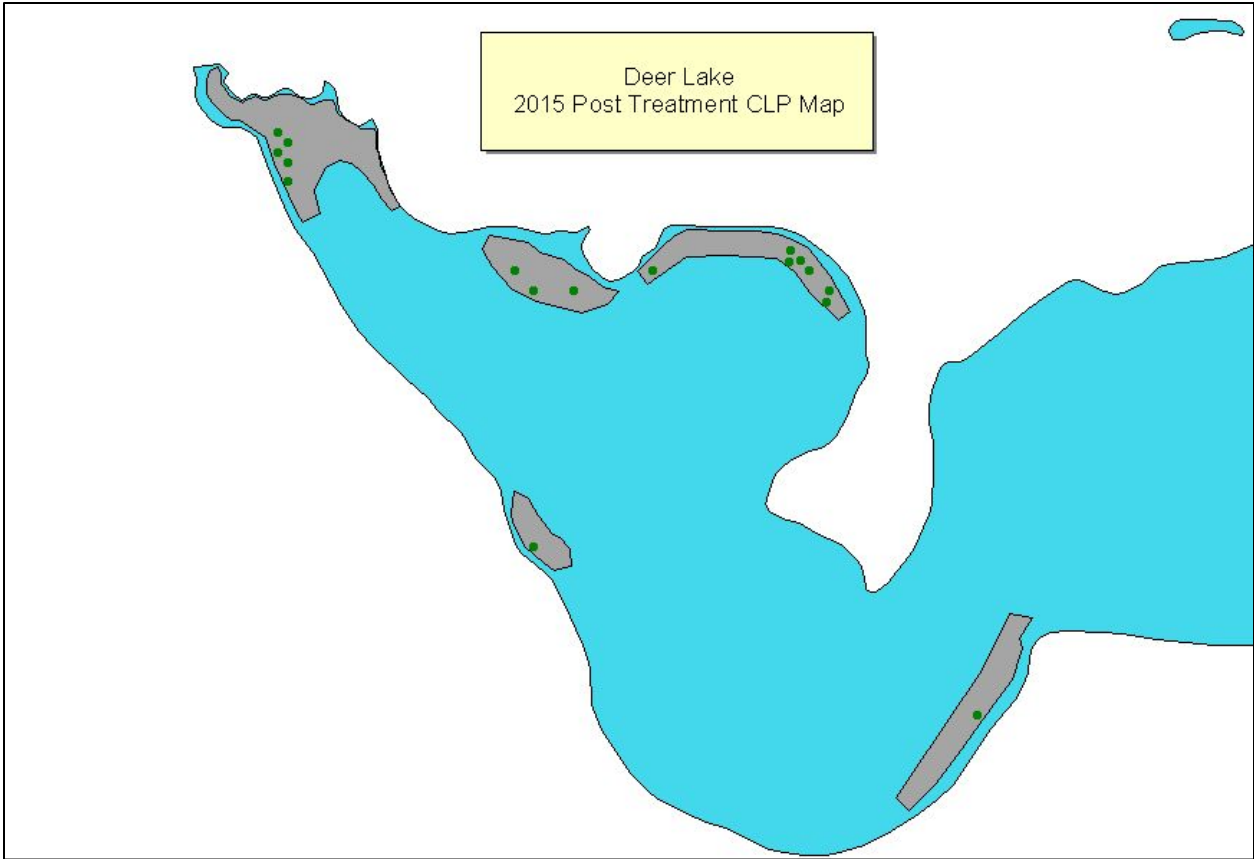


*Nymphaea odorata*-White water lily-2015

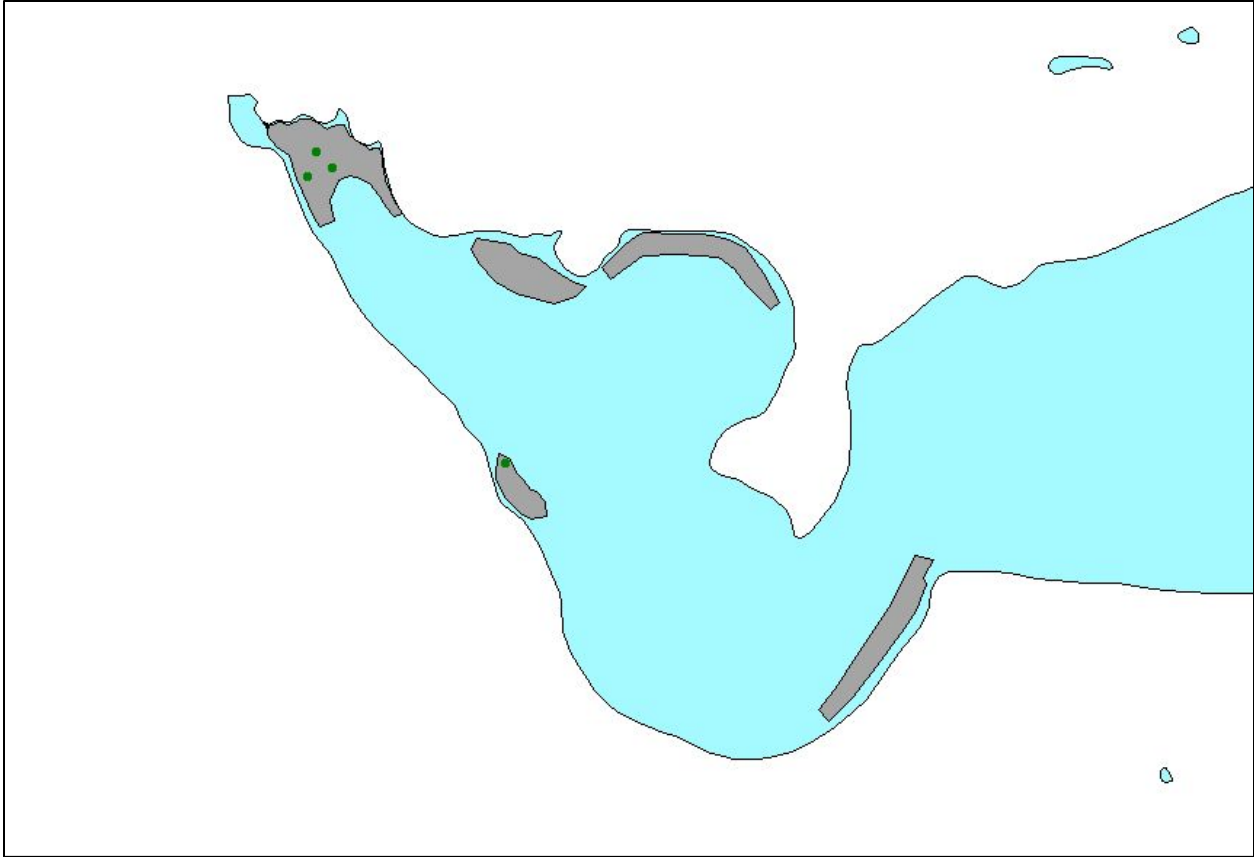




*Nymphaea odorata*-White water lily-2016



*Potamogeton praelongus*-Whitestem pondweed-2015



*Potamogeton praelongus*-Whitestem pondweed-2016