North & South Twin Lakes
AIS Control & Prevention Project
Final Report

Vilas County, Wisconsin

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INTRODUCTION

Photo 1. North Twin Lake, Vilas County, with extensive emergent and floating-leaf plant communities. Photo taken during August 2011 comprehensive plant survey.

North and South Twin Lakes are mesotrophic drainage lakes located in eastern Vilas County, Wisconsin (Map 1, Photo 1). The 2,788-acre North Twin Lake and 642-acre South Twin Lake have reported maximum depths of 60 and 43 feet, respectively. The water levels in this system are maintained by a small dam located at the South Twin Lake outlet.

Property owners on these lakes organized the North and South Twin Lakes Riparian Association (NSTLRA) in 1995. In 1996, the NSTLRA partnered with Vilas County, the University of Wisconsin-Extension, and the Wisconsin Department of Natural Resources (WDNR) to begin the creation of a comprehensive lake management plan for the two lakes. Phase I of the comprehensive plan was completed in 2000 and included components addressing fisheries, watershed composition, water quality, geology, aquatic plant communities, and wildlife use of the lakes, along with the results of a detailed property owner survey. Although Phase I of the management plan contains a great deal of information, additional work was required to continue to the planning effort. In 2004, the Town of Phelps and the NSTLRA partnered to sponsor Phase II of the North and South Twin Lakes Comprehensive Management Planning Project (Phase II Plan). The Phase II Plan was completed during the summer of 2006 (Onterra, 2006) and contained an expanded analysis of current and past water quality data, modeling of watershed phosphorus inputs from surface flows and septic systems, and a comprehensive analysis of each lake's aquatic plant community.

During the surveys completed by Vilas County during the summers of 1996 and 1997, the non-native aquatic plant Eurasian water milfoil (EWM) was not located within either lake. However, in 2001, EWM was located in both North and South Twin Lakes by NSTLRA members and by staff members of the WDNR and Vilas County during 2002 surveys. Based upon guidance in the sensitive area survey and directly from WDNR specialists, multiple 2,4-D treatments were completed between 2001 and 2004, with the largest area, approximately 50 acres, being treated in August 2003. The treatments in 2001 and 2002 were basically spot treatments, with the 2001 treatments totaling 7.0 acres and the 2002 treatments totaling 6.35 acres. Anecdotal reports indicated that the smaller treatments were met with only limited success, while the final, larger treatment was more successful. Unfortunately, no structured monitoring was completed in conjunction with these treatments and little is truly known about their success of failure.

In February of 2008, the NSTLRA successfully applied for an Aquatic Invasive Species (AIS) Control Grant to conduct a four-year project with a chief goal of minimizing the negative impact that AIS, in this case EWM, can have on the ecology of North and South Twin Lakes. The objective of this control project was not to eradicate EWM from North and South Twin Lakes, as that would be impossible; the objective was to bring EWM down to more easily controlled levels. In an attempt to meet these objectives, a cyclic series of steps was used to plan and implement the treatment strategies. The series included:

1. A lake-wide assessment of Eurasian water milfoil completed while the plant is at peak-biomass (July or August).
2. Creation of treatment strategy for the following spring.
3. Verification and refinement of treatment plan immediately before treatments are implemented (spring before treatment).
4. Completion of treatments (spring).
5. Assessment of treatment results (summer after treatment).

The herbicide treatments that took place on North and South Twin Lakes in 2008, 2009, 2010, and 2011 were all quantitatively and qualitatively monitored, and the details of these treatments and their effectiveness will be discussed in the Eurasian water milfoil Section. In addition to annual treatment monitoring, comprehensive aquatic plant surveys were conducted on both lakes in 2011 to assess the aquatic plant communities following four years of large-scale herbicide treatments, the results of which will also serve as an update to the Aquatic Plant Section of the North and South Twin Lakes Management Plan. This report discusses the results of the 2011 comprehensive aquatic plant surveys, the fourth and final year of treatment under this grant-funded project, as well as full project summary. Specific details regarding treatments completed in 2008, 2009, and 2010 can be found in their respective reports.

AQUATIC PLANTS

2011 Aquatic Plant Sampling Methodology and Data Analysis

As discussed previously, in 2011, the final year of this project, comprehensive aquatic plant surveys were conducted on North and South Twin Lakes to assess their aquatic plant communities following annual large-scale herbicide treatments. Aquatic plants are an important element in every healthy aquatic ecosystem. Changes in these ecosystems are often first observed in the plant community. Whether these changes are positive, like variable water levels or negative, like increased shoreland development or the introduction of an exotic species, the plant community will respond. Plant communities respond in a variety of ways; there may be a loss of one or more species, certain life forms, such as emergents or floating-leaf communities may disappear from certain areas of the waterbody, or there may be a shift in plant dominance between species. With periodic monitoring and proper analysis, these changes are relatively easy to detect and provide very useful information for management decisions.

The point-intercept method as described Wisconsin Department of Natural Resources Bureau of Science Services, PUB-SS-1068 2010 (Hauxwell et al 2010) was used to complete the studies on North and South Twin Lakes in August of 2011. Based upon guidance from the WDNR, a point spacing of 100 meters was used on North Twin Lake, while a spacing of 63 meters was used on South Twin Lake (Map 1).

At each point-intercept location within the littoral zone, information regarding the depth, substrate type (muck, sand, or rock), and the plant species sampled along with their relative abundance (Figure 1) on the sampling rake was recorded. A pole-mounted rake was used to collect the plant samples, depth, and sediment information at point locations of 13 feet or less. A rake head tied to a rope (rope rake) was used at sites greater than 13 feet. Depth information was collected using graduated marks on the pole of the rake or using an onboard sonar unit at depths greater than 13 feet. Also, when a rope rake was used, information regarding substrate type was not collected due to the inability of the sampler to accurately feel the bottom with this sampling device.

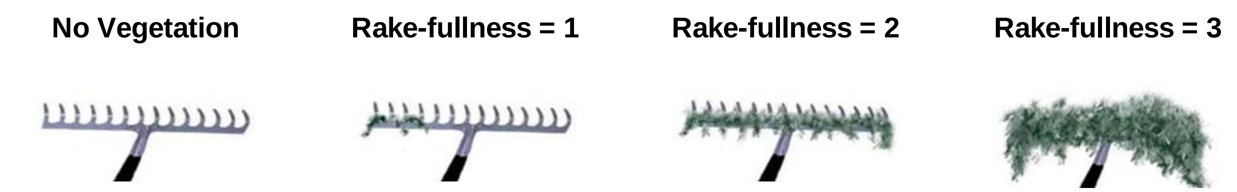

Figure 1. Aquatic plant rake-fullness ratings. Adapted from Hauxwell et al (2010).

The point-intercept survey produces a great deal of information about a lake's aquatic vegetation and overall health. These data are analyzed and presented in numerous ways; each is discussed in more detail the following section.

Primer on Data Analysis & Data Interpretation

Species List

The species list is simply a list of all of the species that were found within each lake, both exotic and native. The list also contains the life-form of each plant found, its scientific name, and its coefficient of conservatism. The latter is discussed in more detail below. Changes in this list over time, whether it is differences in total species present, gains and losses of individual species, or changes in life-forms that are present, can be an early indicator of changes in the health of the ecosystem.

Frequency of Occurrence

Frequency of occurrence describes how often a certain species is found within a lake. Obviously, all of the plants cannot be counted in a lake, so samples are collected from pre-determined areas. In the case of the comprehensive point-intercept surveys conducted in 2011 on North and South Twin Lakes, plant samples were collected from plots laid out on a grid that covered the entire system (Map 1). Using the data collected from these plots, an estimate of occurrence of each plant species can be determined. In this section, two types of data are displayed: littoral frequency of occurrence and relative frequency of occurrence. Littoral frequency of occurrence is used to describe how often each species occurred in the plots that are less than the maximum depth of plant growth (littoral zone). Littoral frequency is displayed as a percentage.

Relative frequency of occurrence uses the littoral frequency for occurrence for each species compared to the sum of the littoral frequency of occurrence from all species. These values are presented in percentages and if all of the values were added up, they would equal 100%. For example, if water lily had a relative frequency of 0.1 and we described that value as a percentage, it would mean that water lily made up 10% of the population.

In the end, this analysis indicates the species that dominate the plant community within the lake. Shifts in dominant plants over time may indicate disturbances in the ecosystem. For instance, low water levels over several years may increase the occurrence of emergent species while decreasing the occurrence of floating-leaf species. Introductions of invasive exotic species may result in major shifts as they crowd out native plants within the system.

Species Diversity

Species diversity is probably the most misused value in ecology because it is often confused with species richness. Species richness is simply the number of species found within a system or community. Although these values are related, they are far from the same because diversity also takes into account how evenly the species are distributed within the system. A lake with 25 species may not be more diverse than a lake with 10 if the first lake is highly dominated by one or two species and the second lake has a more even distribution.

An aquatic system with high species diversity is much more stable than a system with a low diversity. This is analogous to a diverse financial portfolio in that a diverse aquatic plant community can withstand environmental fluctuations much like a diverse portfolio can handle economic fluctuations. For example, a lake with a diverse plant community is much better suited to compete against exotic infestation than a lake with a lower diversity.

Floristic Quality Assessment

The floristic quality of a lake is calculated using its species richness and average species conservatism. As mentioned above, species richness is simply the number of species that occur in the lake, for this analysis, only native species are utilized. Average species conservatism utilizes the coefficient of conservatism values for each of those species in its calculation. A species coefficient of conservatism value indicates that species' likelihood of being found in an undisturbed (pristine) system. The values range from one to ten. Species that are normally found in disturbed systems have lower coefficients, while species frequently found in pristine systems have higher values. For example, cattail, an invasive native species, has a value of 1, while common hard and softstem bulrush have values of 5, and Oakes pondweed, a sensitive and rare species, has a value of 10.

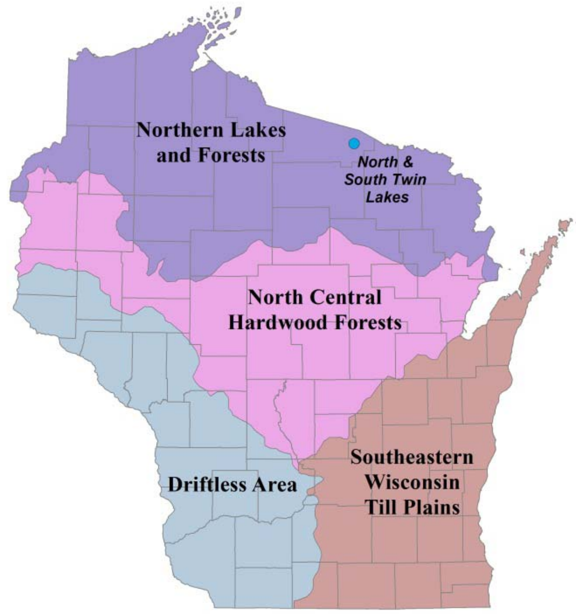

Figure 2. Location of North and South Twin Lakes within the ecoregions of Wisconsin. After Nichols (1999).

On their own, the species richness and average conservatism values for a lake are useful in assessing a lake's plant community; however, the best assessment of the lake's plant community health is determined when the two values are used to calculate the lake's floristic quality. The floristic quality is calculated using the species richness and average conservatism value of the aquatic plant species that were solely encountered on the rake during the point-intercept survey and does not include incidental species or those encountered during other aquatic plant surveys.

North and South Twin Lakes fall within the Northern Lakes and Forests Ecoregion of Wisconsin, and their floristic qualities will be compared to other lakes within this ecoregion as well as the entire state (Figure 2). Ecoregions are areas related by similar climate, physiography, hydrology, vegetation and wildlife potential. Comparing ecosystems within the same ecoregion is sounder than comparing systems within manmade boundaries such as counties, towns, or states.

Community Mapping

A key component of the aquatic plant survey is the creation of an aquatic plant community map. The map represents a snapshot of the important plant communities in the lake as they existed during the survey and is valuable in the development of the management plan and in comparisons with surveys completed in the past and future. A mapped community can consist of floating-leaf or emergent plants, or a combination of these life-forms. Examples of floating-leaf plants include watershield and species of pond lilies; while emergents may include cattails, bulrushes, and arrowheads. Emergents and floating-leaf communities lend themselves well to mapping because there are distinct boundaries between communities. Submergent species are often mixed throughout large areas of the lake and are seldom visible from the surface; therefore, mapping of submergent communities is more difficult and often impossible. If necessary, the point-intercept survey can be used to display the locations of the submergent species of interest.

2011 Comprehensive Aquatic Plant Survey Results

The whole-lake aquatic plant point-intercept surveys and community mapping surveys were conducted by Onterra on North and South Twin Lakes on August 11, 15, and 16, 2011. During these surveys, a total of 40 aquatic plant species were located; two of which are considered to be non-native and invasive: Eurasian water milfoil (*Myriophyllum spicatum*) and reed canary grass (*Phalaris arundinacea*) (Table 1). A type of *Phragmites* spp. was located along the shoreline in several areas as well. Morphologically, this species was confirmed as the native strain of common reed grass. Discussion of these species can be found in the final sections of this report.

Table 1. Aquatic plant species located in North and South Twin Lakes during August 2011 surveys.

Life Form	Scientific Name	Common Name	Coefficient of Conservatism (C)	2011 North Twin	2011 South Twin
Emergent	*Carex lacustris*	Lake sedge	6	I	I
	Eleocharis palustris	Creeping spikerush	6	X	
	Iris versicolor	Northern blue flag	5	I	
	Juncus effusus	Soft rush	4	I	
	Phalaris arundinacea	Reed canary grass	Exotic	I	
	Phragmites australis subsp. americanus	Giant reed (Native)	N/A	I	
	Sagittaria rigida	Stiff arrowhead	8	I	I
	Schoenoplectus acutus	Hardstem bulrush	5	X	X
	Scirpus cyperinus	Wool grass	4	I	I
	Typha spp.	Cattail spp.	1	I	
FL	*Nuphar variegata*	Spatterdock	6	I	
FL/E	*Sparganium angustifolium*	Narrow-leaf bur-reed	9	I	I
Submergent	*Ceratophyllum demersum*	Coontail	3	X	X
	Chara spp.	Muskgrasses	7	X	X
	Elodea canadensis	Common waterweed	3	X	X
	Heteranthera dubia	Water stargrass	6	X	X
	Isoetes sp.	Quillwort species	8	X	X
	Megalodonta beckii	Water marigold	8	X	X
	Myriophyllum alterniflorum	Alternate-flowered water milfoil	10	X	X
	Myriophyllum sibiricum	Northern water milfoil	7	X	
	Myriophyllum spicatum	Eurasian water milfoil	Exotic	X	X
	Najas flexilis	Slender naiad	6	X	X
	Nitella sp.	Stoneworts	7	X	X
	Potamogeton amplifolius	Large-leaf pondweed	7	X	X
	Potamogeton friesii	Fries' pondweed	8	X	X
	Potamogeton gramineus	Variable pondweed	7	X	X
	Potamogeton hybrid 1	Pondweed hybrid 1	N/A	X	X
	Potamogeton hybrid 2	Pondweed hybrid 2	N/A		X
	Potamogeton praelongus	White-stem pondweed	8	X	X
	Potamogeton pusillus	Small pondweed	7	X	X
	Potamogeton richardsonii	Clasping-leaf pondweed	5	X	X
	Potamogeton robbinsii	Fern pondweed	8	X	X
	Potamogeton strictifolius	Stiff pondweed	8	X	X
	Potamogeton zosteriformis	Flat-stem pondweed	6	X	X
	Ranunculus aquatilis	White water-crowfoot	8	X	
	Sagitaria sp. (rosette)	Arrowhead rosette	N/A	X	
	Vallisneria americana	Wild celery	6	X	X
S/E	*Eleocharis acicularis*	Needle spikerush	5	X	X
	Juncus pelocarpus	Brown-fruited rush	8	X	X
	Sagittaria cuneata	Arum-leaved arrowhead	7	I	

FL = Floating-leaf; FL/E = Floating-leaf and Emergent; S/E = Submergent and Emergent
X = Located on rake during point-intercept survey; I = Incidental species

Before discussing the distribution of aquatic plants within North and South Twin Lake, it is important to understand the composition of the substrate types as this can limit where aquatic plants are able to grow. Aside from a handful of species that favor course substrates like sand, most aquatic plants favor soft and nutrient-rich sediments – muck. During the point-intercept surveys conducted in 2011, the dominant substrate type at each sampling location was classified into three different, albeit general, categories: 1) muck, 2) sand, and 3) rock. It is important to note that a continuum exists between these categories, and while the dominant substrate type was recorded, some sampling locations contained more than one type.

The dominant substrate type within littoral areas (pole-mounted rake method only) as determined from the point-intercept sampling survey is sand in both lakes (Map 2, Map 3, Figure 3). No sampling locations with muck substrate where encountered in North Twin Lake, while 16% of sampling locations contained muck in South Twin Lake. Rock was the dominant substrate type at 15% and 3% of the locations sampled on North Twin Lake and South Twin Lake, respectively.

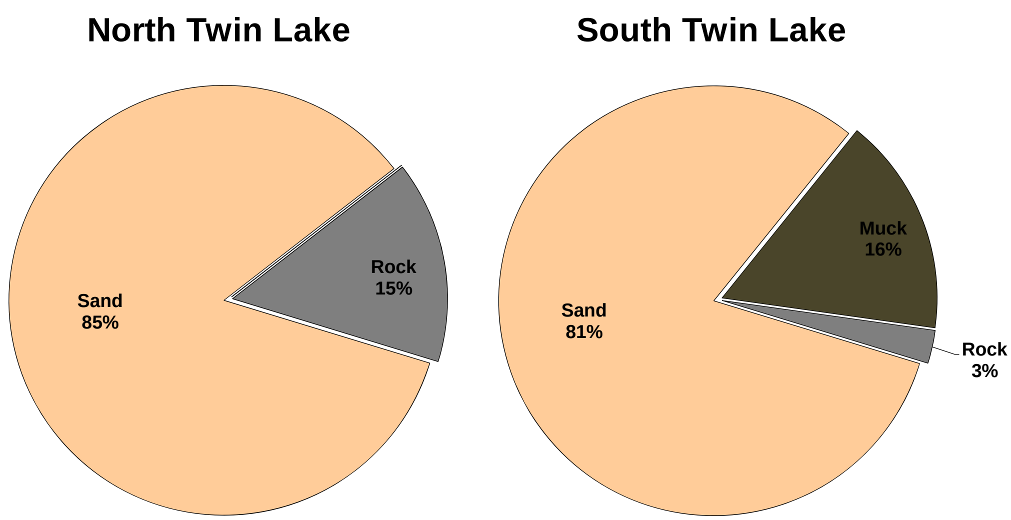

Figure 3. North and South Twin Lakes proportion of substrate types. Created using data from August 2011 aquatic plant point-intercept surveys.

During the 2011 surveys, aquatic plants were located growing to a maximum depth of 20 feet in North Twin Lake and 18 feet in South Twin Lake. Of the sampling points that fell within the maximum depth of plant growth (littoral zone), 67% in North Twin Lake and 94% in South twin Lake contained aquatic vegetation. Map 4 and 5 display the distribution and abundance of aquatic vegetation within the lakes' littoral zones. With a littoral zone comprised mainly of sand, most areas in North Twin Lake had lower densities of aquatic plant growth, and the majority of vegetated sampling locations in North Twin Lake had rake-fullness ratings of 1 or 2 (Map 4). The northeast portion of the lake was sparsely vegetated and contained rockier substrates.

Looking at Map 3 and Map 5, the areas with the highest aquatic plant abundance in South Twin Lake (rake-fullness = 3) fall in the same areas that contain mucky substrates. The aquatic plant species composition between these areas was also markedly different; wild celery, slender naiad, variable pondweed, and muskgrasses dominated the sandy areas and were growing at lower

densities, while fern pondweed, coontail, clasping-leaf pondweed, and flat-stem pondweed were observed growing in higher densities within areas of muck.
Of the 39 aquatic plant species located during the surveys on North and South Twin Lakes, 28 species were common to both lakes. Figure 4 and Figure 5 display the littoral frequencies of occurrence of aquatic plant species in both systems. Wild celery, variable pondweed, and slender naiad were the three most frequently encountered species in both lakes during the 2011 surveys.

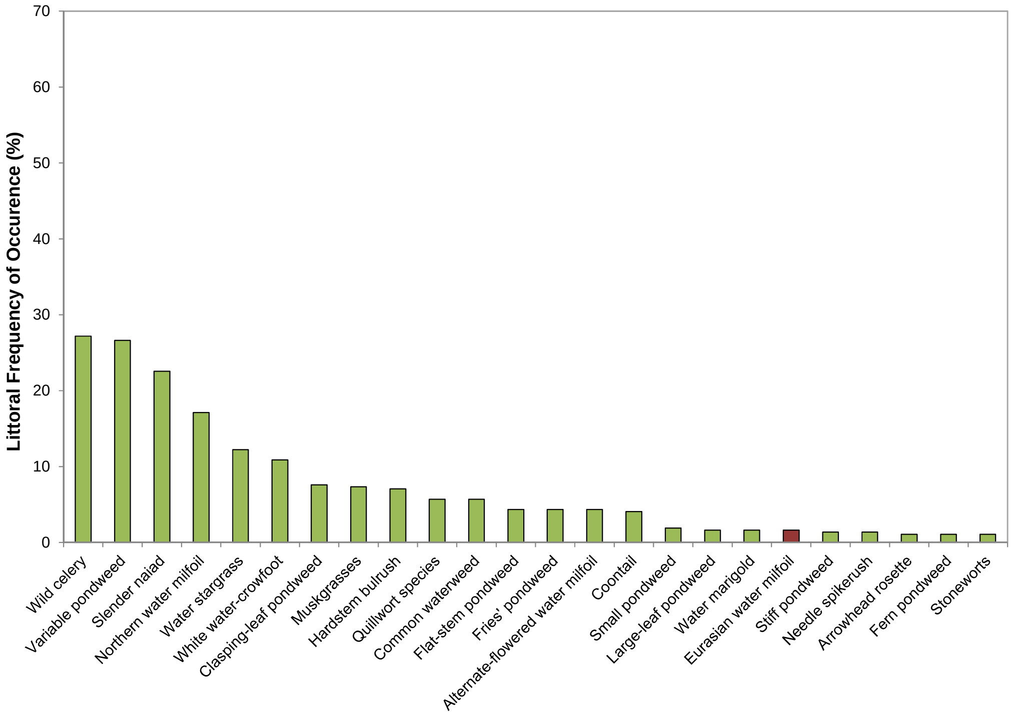

Figure 4. North Twin Lake aquatic plant littoral occurrence analysis. Created using data from August 2011 aquatic plant surveys. Species with littoral frequency <1% are not included. Exotic species indicated with red.

Wild celery, also known as tape or eel grass, was the most common native aquatic plant species encountered in both North and South Twin Lakes. This plant is common in Wisconsin and can be found growing in many differing lake habitats, making it an excellent source of food for waterfowl, muskrats, and other wildlife. Often found growing particularly well in sandy substrates, wild celery’s long leaves also provide excellent structural habitat for numerous aquatic organisms while its extensive root systems stabilize bottom sediments.

Variable pondweed, as its name suggests, can differ in its appearance depending on where it’s growing. For example, in North and South Twin Lakes, when variable pondweed was observed growing in sandy areas it had smaller, more crowded leaves and was more compact. On the other hand, when growing in areas of softer substrate, this species was long and tapering, with larger, less crowded leaves. The seeds and tubers of variable pondweed provide sources of food for numerous species of wildlife, while its branching foliage provides structurally diverse habitat for many aquatic organisms.

Slender naiad, a common annual species in Wisconsin, is considered to be one of the most important food sources for a number of migratory waterfowl species (Borman et al. 1997). Their numerous seeds, leaves, and stems all provide sources of food. The small, condensed network of leaves provide excellent habitat for aquatic invertebrates.

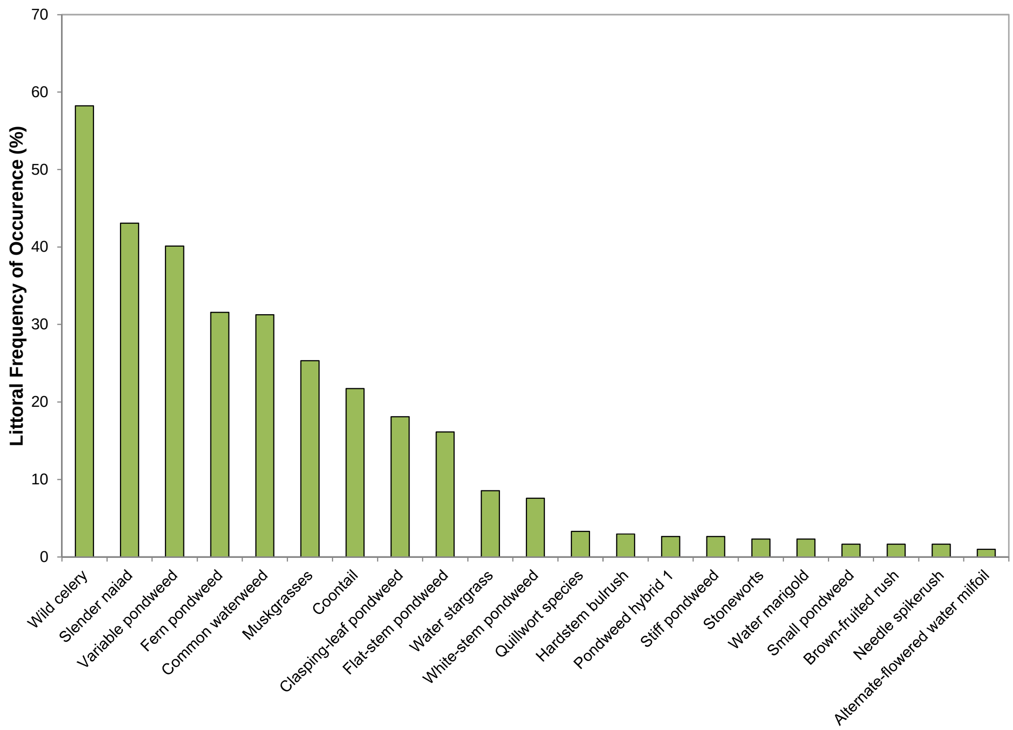

Figure 5. South Twin Lake aquatic plant littoral occurrence analysis. Created using data from August 2011 aquatic plant surveys. Species with littoral frequency <1% are not included.

As discussed previously, the calculations used for the Floristic Quality Index (FQI) for a lake's aquatic plant community are based on the aquatic plant species that were encountered on the rake during the point-intercept survey and does not include incidental species. For example, while 37 native aquatic plant species were located in North Twin Lake during the 2011 surveys, only 26 were encountered on the rake during the point-intercept survey. These 26 native species and their conservatism values were used to calculate the FQI of North Twin Lake's aquatic plant community in 2011 (equation shown below).

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

Figure 6 compares the FQI of North Twin Lake's aquatic plant community from a survey conducted in 2004 to the one conducted in 2011. It should be noted that the survey conducted in 2004 was transect-based, a commonly accepted methodology at that time. Since then, the new point-intercept methodology developed by the WDNR has become accepted method of assessing a lake's aquatic plant community. The 2004 transect-based survey targets near-shore areas of the lake more so than the point-intercept survey performed in 2011. The point-intercept survey covers more of the lake and has a higher focus on deeper areas.

As Figure 6 displays, native species richness was similar between the two surveys with 24 in 2004 and 26 in 2011, both well above the Northern Lakes Ecoregion and Wisconsin State medians. The average conservatism values in 2004 (6.6) and 2011 (6.7) indicate that the quality of North Twin Lake's aquatic plant community has been maintained over this time period and is comparable to other lakes within the ecoregion and of higher quality than most lakes in the state. Combining native species richness and the plant community's average conservatism value from 2011 results in an exceptionally high FQI value of 34.1; well above the ecoregion and state medians (Figure 6).

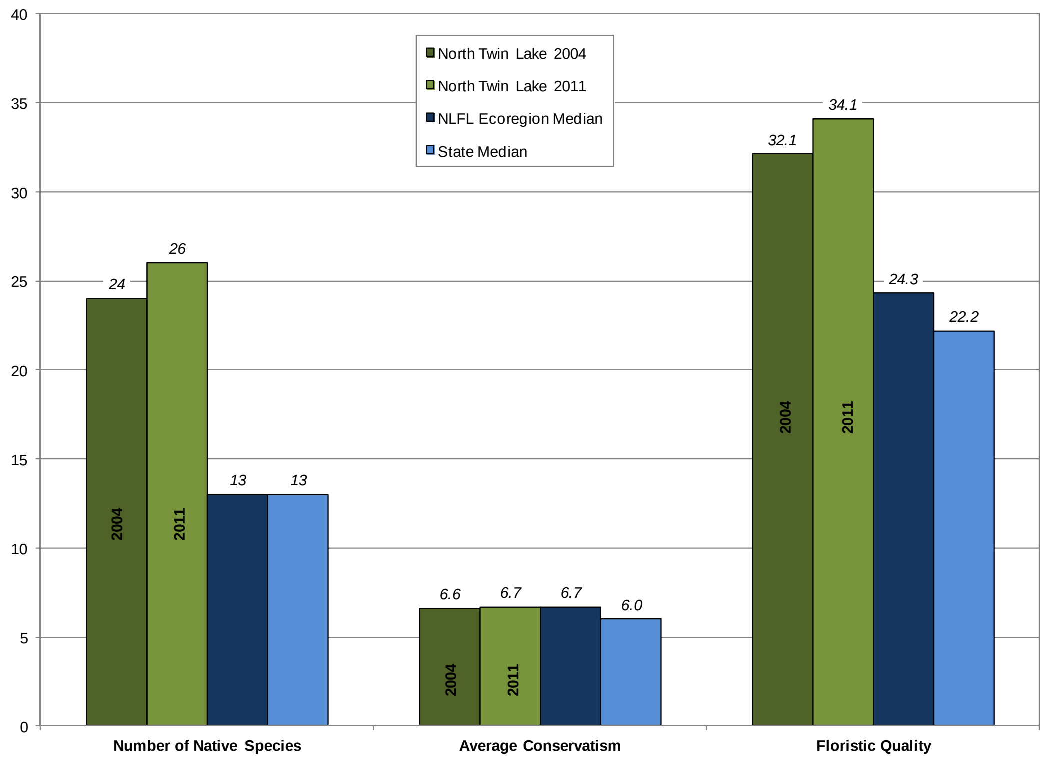

Figure 6. North Twin Lake Floristic Quality Assessment. Created using data from 2004 transect survey data and 2011 aquatic plant point-intercept survey data. Analysis follows Nichols (1999).

Like North Twin Lake, a transect-based survey was also completed on South Twin Lake in 2004. However, because the intent of the treatments in 2009 and 2010 was to disperse herbicide throughout the entire lake, whole-lake aquatic plant point-intercept surveys were also completed over the course of this project in 2008, 2009, 2010, and 2011 to monitor these treatments.

Figure 7 shows how the native species community of South Twin Lake has changed in regards to the floristic quality index. Again, the slightly lower species richness and average conservatism values in 2004 are likely due to differences in the survey methodology. The figure shows that species richness and average conservatism values between 2008 and 2011 were relatively constant, however closer analysis indicates that there were some notable differences as some plant species were impacted by the treatments.

After the whole-lake treatment that was conducted during the spring of 2009, dwarf water milfoil was not located within South Twin Lake. Stiff pondweed, which was not located in 2008, was found in 2009 and all subsequent surveys resulting in the same number of species occurring in 2008 and 2009. Because both these species have high average conservatism values, there is almost no net difference in average conservatism and floristic quality between the two years.

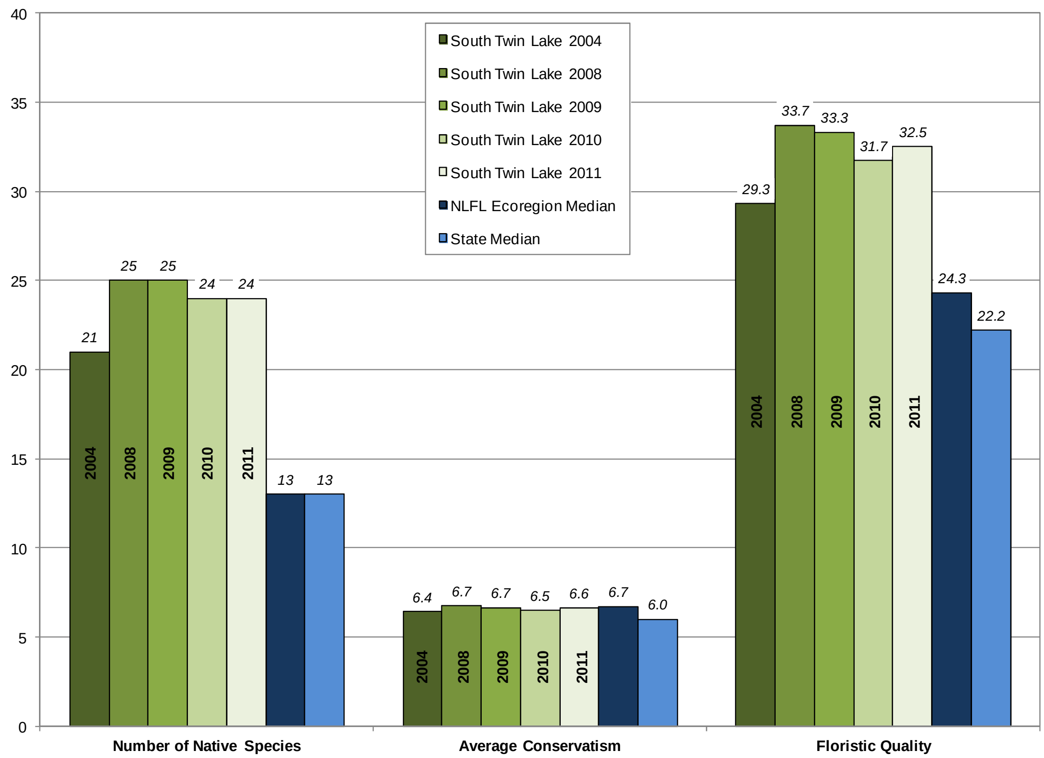

Figure 7. South Twin Lake Floristic Quality Assessment. Created using data from 2004 transect survey data and 2008-2011 aquatic plant point-intercept survey data. Analysis following Nichols (1999).

As will be discussed in greater detail in a separate section, the spring 2010 whole-lake treatment resulted in 4-5 times higher herbicide concentrations that the 2009 treatment. EWM was not detected during the summer following that treatment. However, neither was northern water milfoil nor white water-crowfoot, two important native species. While this was a decline of two species, sago pondweed was located for the first time in 2010, resulting in a net loss of only one species. However, sago pondweed has a much lower conservatism value than that of the other two species, which in the end was actually responsible for decreasing the floristic quality index value for 2010. In other words, the floristic quality was truly not decreased by losing northern water milfoil and white water-crowfoot, but by gaining a lower value species – that being sago pondweed.

No herbicide treatment was conducted on South Twin Lake in 2011. During 2011, EWM was located at one point-intercept sampling location. Sago pondweed was not located during this survey, but a new hybrid pondweed species was detected. With sago pondweed's low average conservatism value not contributing to the analysis in 2011, the floristic quality increased slightly.

Overall, subtle but important differences in the native plant community occurred between 2008 and 2011 on South Twin Lake. The floristic quality analysis only incorporates presence/absence of species and subsequent analysis of South Twin Lake's aquatic plant community shows how the frequency of some native species changed over this timeframe. It is important to note that the floristic quality of South Twin Lake continues to remain of higher quality than most lakes in the ecoregion and in the state (Figure 7). Also, EWM frequency of occurrence was reduced by over 98% on South Twin Lake during the same timeframe.

Lakes with diverse aquatic plant communities have higher resilience to environmental disturbances and greater resistance to invasion by non-native plants. A plant community with a mosaic of species with differing morphological attributes provides zooplankton, macroinvertebrates, fish and other wildlife with diverse structural habitat and various sources of food. Because North and South Twin contain a high number of native aquatic plant species, one may assume their aquatic plant communities have high species diversity. As discussed earlier, species diversity is also influenced by how evenly the plant species are distributed within the community.

An ecological tool called Simpson's Diversity Index (1-D) is commonly used to determine a habitat's diversity. Simpson's diversity is calculated as:

$$D = \sum (n/N)^2$$

where: n = the total number of instances of a particular species
N = the total number of instances of all species
D is a value between 0 and 1

Using the data collected from the 2011 aquatic plant point-intercept surveys, North and South Twin Lakes' plant communities were shown to be highly diverse, with respective Simpson's Diversity values of 0.91 and 0.90 (Figure 7). These values correspond with the likelihood that the next plant sampled will be a different species. For example, a Simpson's Diversity Index of 0.91 means that there is a 91% chance that the next species encountered is different from the previous. As discussed earlier, how evenly the species are distributed throughout the system and species richness together influence species diversity. These values indicate that the plant communities of the lakes are not overly dominated by a single or few species, but have a more even distribution of species.

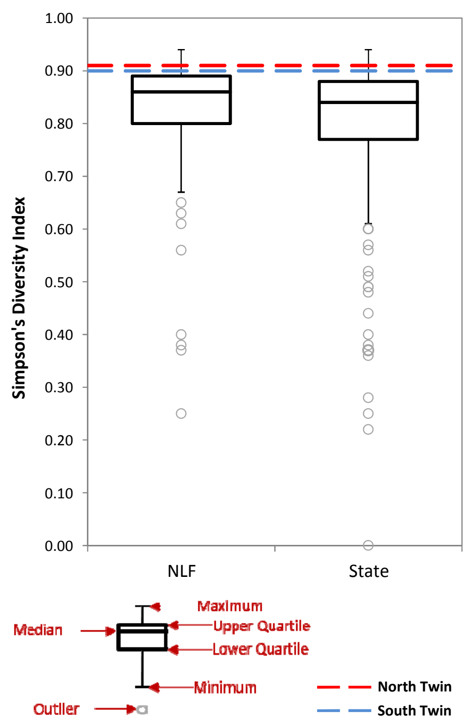

Figure 8. North and South Twin Lakes species diversity index. Created using data from 2011 point-intercept surveys.

Larger lakes tend to have a larger suite of habitat types (e.g. calm back water bays, sand bars) that can support many different species. While a method of characterizing diversity values as "Fair" or "Poor", etc. does not exist, lakes within the same ecoregion may be compared to provide an idea of how North and South Twin Lakes' scores rank. Using data obtained from WDNR Science Services, median values and upper/lower quartiles were calculated for 109 lakes within the Northern Lakes and Forests ecoregion (Figure 8). Both lakes rank above the upper quartile for both the ecoregion and the state, indicating the diversity values of North and South Twin Lakes' plant communities fall within the top 25% of lakes sampled.

As explained above in the Primer on Data Analysis and Data Interpretation Section, the littoral frequency of occurrence analysis allows for an understanding of how often each of the plants is located during the point-intercept survey. Because each sampling location may contain numerous plant species, relative frequency of occurrence is one tool to evaluate how often each plant species is found in relation to all other species found (composition of population). For instance, while wild celery was found at almost 30% of the sampling locations in North Twin Lake, its relative frequency of occurrence is 15%. Explained another way, if 100 plants were randomly sampled from North Twin Lake, 15 of them would be wild celery. Figure 9 displays the relative frequency of occurrence of aquatic plant species from North and South Twin Lakes and illustrates the even distribution of species in these diverse plant communities.

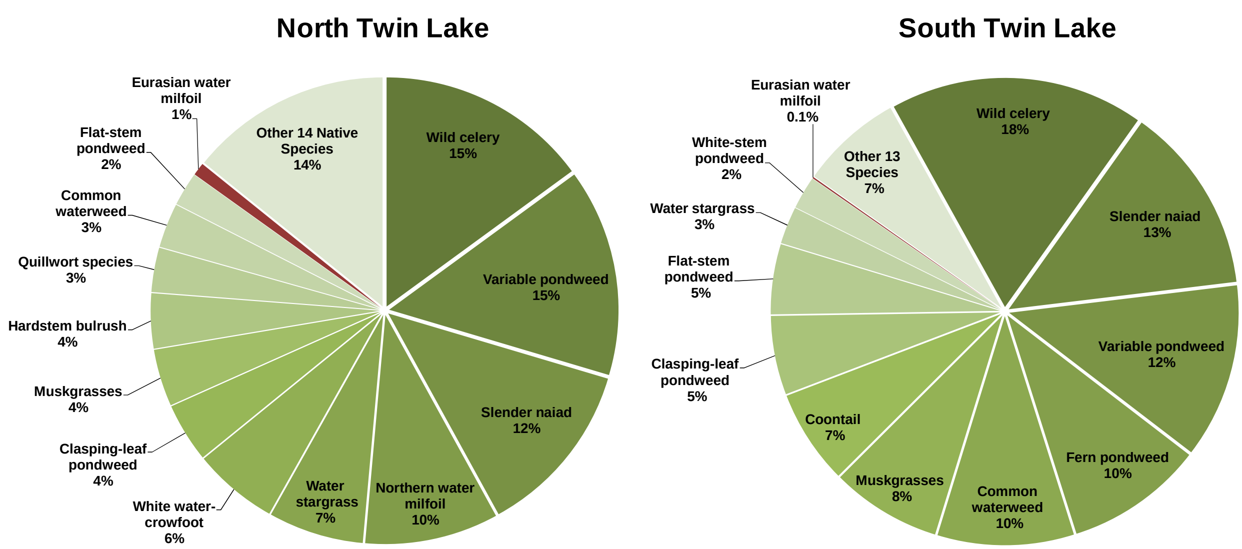

Figure 9. North and South Twin Lakes relative occurrence analysis. Created using data from August 2011 point-intercept surveys. Exotic species indicated with red.

The 2011 community mapping surveys on the North and South Twin Lakes were the second surveys of this type to be conducted, the first being conducted by Onterra in 2004. The quality of these lakes' plant communities is also indicated by the high incidence of emergent and floating-leaf aquatic plant communities that occur in many areas. Ten emergent and floating-leaf aquatic plant species were located in 2011 (Table 1). The 2011 community maps indicate that approximately 82 and 13 acres of North and South Twin Lakes, respectively, contain these types of plant communities (Map 6, Map 7, Table 2).

Table 2. North and South Twin Lakes acres of emergent and floating-leaf plant communities from the 2004 and 2011 community mapping surveys.

	Acres			
	North Twin Lake		South Twin Lake	
Plant Community	2004	2011	2004	2011
Floating-leaf	2.1	0.3	0.0	0.4
Emergent	74.0	73.6	11.4	12.9
Floating-leaf/Emergent	3.9	8.0	0.0	0.0
Total	**80.0**	**81.9**	**11.4**	**13.3**

The total acreage of floating-leaf and emergent plant communities in both North Twin and South Twin Lake has remained virtually unchanged since 2004 (Table 2). Some initial concerns were brought forth regarding the possibility that the whole-lake 2,4-D treatments on South Twin would have a negative impact on the bulrush communities within this lake. Based on these data, the bulrush communities have not declined, but actually increased slightly during this timeframe.

Continuing the analogy that the community map represents a snapshot of the emergent and floating-leaf plant communities, replications of this survey through time will provide a valuable understanding of the dynamics of these communities within North and South Twin Lakes. This is important, because these communities are often negatively affected by recreational use and shoreland development. Radomski and Goeman (2001) found a 66% reduction in vegetation coverage on developed shorelines when compared to undeveloped shorelines in Minnesota Lakes. Furthermore, they also found a significant reduction in abundance and size of northern pike (*Esox lucius*), bluegill (*Lepomis macrochirus*), and pumpkinseed (*Lepomis gibbosus*) associated with these developed shorelines.

Overall, the aquatic plant communities of North and South Twin Lake are of excellent quality, and this quality has been maintained over the course of this project. Their plant communities contain a high number of native species and are highly diverse. With the use of annual herbicide treatments, the non-native EWM constitutes a very small portion of the plant populations in North and South Twin Lakes.

Emergent Plants of Concern – Reed Canary Grass and Common Reed Grass

Reed Canary Grass

Reed canary grass (*Phalaris arundinacea*) is a large, coarse perennial grass that can reach three to six feet in height. Often difficult to distinguish from native grasses, this species forms dense, highly productive stands that vigorously outcompete native species. Unlike native grasses, few wildlife species utilize the grass as a food source, and the stems grow too densely to provide cover for small mammals and waterfowl. It grows best in moist soils such as wetlands, marshes, stream banks and lake shorelines.

Reed canary grass is difficult to eradicate; at the time of this writing there is no commonly accepted control method. This plant is quite resilient to herbicide applications. Small, discrete patches have been covered by black plastic to reduce growth for an entire season. However, the species must be monitored because rhizomes may spread out beyond the plastic.

At this time, populations are not excessive, though it is recommended that continued monitoring of reed canary grass takes place. During the community mapping survey of North Twin Lake in 2011, Onterra ecologists mapped three occurrences of reed canary grass along the shoreline of the lake with sub-meter GPS technology (Map 6).

Common Reed Grass

Phragmites australis subs. americanus, or, common reed grass, is a sub species of a plant that calls every continent on Earth but Antarctica its home. It is believed that populations of common reed grass existed in pre-colonial Wisconsin, but exotic strains from Europe have been introduced and have invaded the genetic line of the native strain. Genetic identification of the plant is needed to determine whether the plant is of the native or non-native strain. A pressed specimen of this species from North Twin Lake was sent to Dr. Robert Freckman at the University of Wisconsin – Steven's Point where morphologically it appeared to be a native strain. The sub species *americanus* is native to North America and typically does not display invasive behavior, as the exotic *Phragmites australis* does. These characteristics include towering, dense colonies that overtake native vegetation and replace it with a monoculture that provides inadequate food and habitat for wildlife.

Although this plant appears to be morphologically native, it is recommended that this population be monitored for expansion. During the community mapping survey of North Twin Lake in 2011, Onterra ecologists mapped occurrences of common reed grass along the shoreline of the lake with sub-meter GPS technology (Map 6). If it appears that the plant is spreading along the shorelines of North Twin Lake, the regional WDNR Lake Specialist should be contacted to coordinate sending in plant specimens for genetic testing. If the common reed is determined to be an exotic strain, it should be removed by cutting and bagging the seed heads and applying herbicide to the cut ends. This management strategy is most effective when completed in late summer or early fall when the plant is actively storing sugars and carbohydrates in its root system in preparation for over-wintering. If this or other populations expand greatly, a management action would need to be developed to coordinate its control.

2011 EURASIAN WATER MILFOIL TREATMENT

Herbicides that target submersed plant species are directly applied to the water, either as a liquid or an encapsulated granular formulation. Factors such as water depth, water flow, treatment area size, and plant density work to dilute herbicide concentration within aquatic systems. Understanding concentration-exposure times are important considerations for aquatic herbicides. Successful control of the target plant is achieved when it is exposed to a lethal concentration of the herbicide for a specific duration of time. Much information has been gathered in recent years, largely as a result of a joint research project between the WDNR and US Army Corps of Engineers (USACE). Based on their preliminary findings, lake managers have adopted two main treatment strategies; 1) whole-lake treatments, and 2). spot treatments.

Whole-lake treatments are those where the herbicide is applied to specific sites, but when the herbicide reaches equilibrium within the entire volume of water (of the lake, lake basin, or within the epilimnion of the lake or lake basin); it is at a concentration that is sufficient to cause mortality to the target plant within that entire lake or basin. The application rate of whole-lake treatments is dictated by the volume of water in which the herbicide will reach equilibrium with. The target herbicide concentration is typically between 0.225 and 0.325 ppm acid equivalent (a.e.). when exposed to the target plants for 7-14 days or longer. However, these same rates have been shown to impact some native plant species, particularly dicot species, some thin-leaved pondweeds, and naiad species. This is the strategy implemented on South Twin Lake in 2009 and 2010.

Spot treatments are a type of control strategy where the herbicide is applied to a specific area (treatment site) such that when it dilutes from that area, its concentrations are insufficient to cause significant affects outside of that area. This is the strategy implemented on North Twin Lake. Spot treatments typically rely on a short exposure time (often hours) to cause mortality and therefore are applied at a much higher herbicide concentration than whole-lake treatments. For Eurasian water milfoil, 2,4-D is typically applied between 2.25 and 3.0 ppm a.e. in spot treatment scenarios. A newly adopted term, ‘micro-treatments’ is being used to describe very small spot treatments (working definition is less than 5 acres). Because of their small size, it is extremely difficult to predict treatment effectiveness due to rapid dilution of the herbicide. Larger treatment areas tend to be able to hold effective concentrations for a longer time.

Following the 2010 peak biomass survey, a conditional treatment permit map was created proposing 18.3 acres of spot-treatment on North Twin Lake (Map 8). No EWM was observed in South Twin Lake following the 2010 treatment and therefore no treatment was proposed for South Twin Lake. The treatment conducted on North Twin Lake in 2010 met the success criteria standards using Renovate Max G. This herbicide is a combination of granular 2,4-D and triclopyr theorized to have synergistic affects compared to the respective herbicide components alone. This herbicide was proposed for use again in 2011 at similar application rates (approximately 3.0 ppm a.e.).

On May 25, 2011, Onterra staff visited North Twin Lake to survey the proposed treatment areas and refine their boundaries as appropriate. Because EWM is low-growing at this time of year and difficult to see from the surface, transects around and through the proposed treatment areas were completed almost exclusively with submersible video technology.

During this survey, treatment sites D-11 and G-11 were expanded slightly to encompass newly discovered EWM (Map 8). Treatment sites A-11 and B-11 were not initially proposed for treatment in 2011 after no EWM was observed within these areas during the 2010 peak-biomass survey. However, it is possible that EWM within these areas was injured during the 2010 treatment and due to its scattered nature, went undetected during the peak-biomass survey completed later that year. Unfortunately, the 2011 spring pre-treatment survey revealed EWM in these areas and they were recommended for treatment. Site H-11 was removed from the 2011 treatment strategy as only a few single EWM plants were observed in this area.

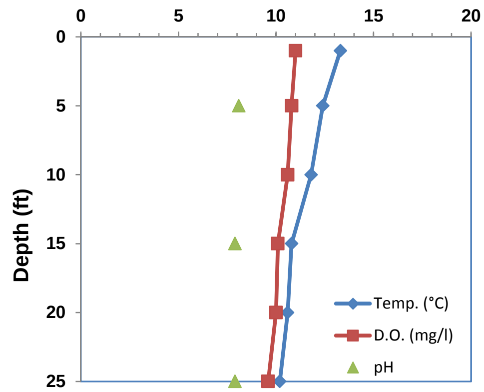

Figure 10. Temperature, dissolved oxygen, and pH profile from North Twin Lake. May 25, 2011.

Based on the pretreatment survey, 15.4 acres comprised the final 2011 treatment strategy (Map 8). During the pretreatment survey, a temperature, dissolved oxygen, and pH profile was collected in approximately 25 feet of water west of the island. Figure 1 shows that water temperatures were around 56°F at the surface and around 53°F at eight feet, the average depth of the 2011 treatment areas.

On June 6, 2011, the final treatment areas were treated with Renovate Max G by Bonestroo (now Stantec and previously Northern Environmental). They reported air temperatures of 80°F and 5-10 mph winds out of the SW.

2011 Treatment Monitoring

The goal of herbicide treatments is to maximize target species (EWM) mortality while minimizing impacts to valuable native aquatic plant species. Monitoring herbicide treatments and defining their success incorporates both quantitative and qualitative methods. As the name suggests, quantitative monitoring involves comparing number data (or quantities) such as plant frequency of occurrence before and after the control strategy is implemented. Qualitative monitoring is completed by comparing visual data such as EWM colony density ratings before and after the treatments.

EWM treatment quantitative evaluation methodologies follow WDNR protocols in which point-intercept data are collected within treatment areas before and after the treatment. On North Twin Lake, data of this type was collected at over 130 point-intercept sub-sample locations (Figure 11). However, not all of those points were located within the areas where herbicide was directly applied and therefore not all points are used in the analysis.

The most comparative sub-sample data are those collected both the summer before and the summer immediately following the herbicide treatment. On North Twin Lake, 40 point-intercept sub-sample locations fell into this category. At these sampling locations, EWM and native aquatic plant species presence and rake-fullness were documented along with water depth and substrate type. Specifically, these surveys aim to determine if significant differences in frequencies of occurrence of EWM and native species occur following the herbicide application.

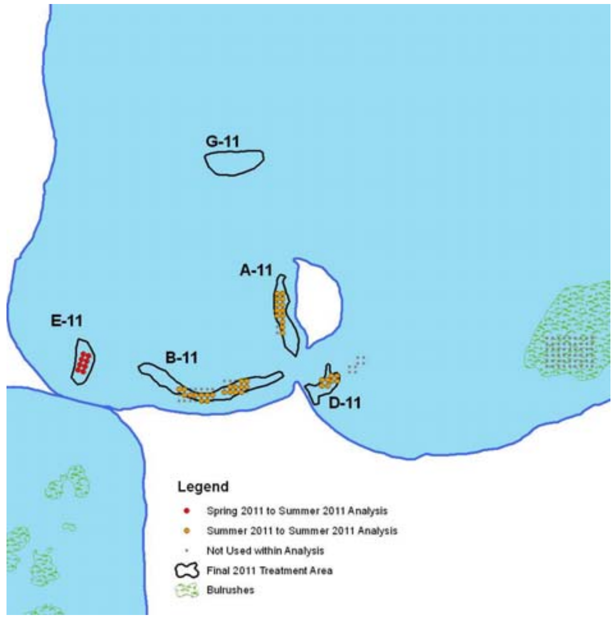

Figure 11. 2011 Quantitative monitoring plan for North Twin Lake

No sub-sampling locations were sampled within E-11 during the summer of 2010. However, 8 sub-sample locations were sampled during spring before the treatment allowing for a spring-to-summer comparison to be made. While not as appropriate as summer to summer comparisons, these data were collected to provide some level of quantitative evaluation for this treatment sites. Because this data differs from those discussed above, they will be discussed separately and not included within the main analysis.

Quantitatively, a specific treatment is deemed to be successful if the EWM frequency following the treatment exhibits a statistically valid reduction by at least 50%. On North Twin Lake, the effectiveness of each treatment method (herbicide/dosage) was quantitatively evaluated. Evaluation of treatment-wide effectiveness follows the same criteria based upon pooled sub-sample data from all of the treatment sites. Further, a noticeable decrease in rake-fullness ratings within the fullness categories of 2 and 3 should be observed and preferably, there would be no rake tows exhibiting a fullness of 2 or 3 during post treatment surveys.

Spatial data reflecting EWM locations were collected using a sub-meter Global Positioning System (GPS) during the late summers of 2010 and 2011, when EWM is assumed to be at its peak biomass or growth stage. Comparisons of these surveys are used to qualitatively evaluate the 2011 herbicide treatment on North Twin Lake. Qualitatively, a successful treatment on a particular site would include a reduction of EWM density as demonstrated by a decrease in density rating (e.g. highly dominant to dominant). In terms of a treatment as a whole (lake-wide), at least 75% of the acreage treated that year would decrease by one level of density as described above for an individual site.

Although it is never the intent of the treatments to impact native species, it is important to remember that in spot treatment scenarios, these non-target impacts can only be considered in the context of the areas treated and not on a lake-wide basis. In other words, the impact of the treatments on a non-target species in the treatment areas cannot be extrapolated to the entire population of that plant within the lake, unless the plant species is only found in locations where the herbicide applications took place.

2011 Treatment Results

Post treatment surveys were completed on North Twin Lake by Onterra on September 2, 2011. Map 9 shows the results of the early-September 2011 peak-biomass survey. Forty percent of the treatment site acreage (sites E-11, G-11, D-11, and I-11) was observed to have reductions in EWM density following the 2011 treatment, falling short of the qualitative success (75% of treatment acreage). However, it must be noted that because no EWM was observed in treatment sites A-11 and B-11 in 2010, qualitative data from that year does not exist for comparison. Two small colonies of EWM were newly discovered during the 2011 survey, one just south the northwest boat landing (E-12) and the other just north of treatment site A-11 (F-12).

Summer-to-summer quantitative analysis was conducted on three of the five treatment sites (Figure 11; Sites A-11, B-11, & D-11). During the summer of 2010, 15.0% of the point-intercept locations within the 2011 treatment areas contained EWM compared to 12.5% in 2011. Though there was a slight reduction in 2011, this change in occurrence is not statistically valid and does not meet the quantitative success criteria (50% reduction in occurrence).

Data concerning native aquatic plant species were collected at the same 40 point-intercept locations during the summers of 2010 and 2011. Table 3 shows that within the 2011 treatment areas, muskgrasses and slender naiad exhibited statistically valid reductions in occurrence following the 2011 treatment. One species, large-leaf pondweed, displayed a statistically valid increase in occurrence within the 2011 treatment areas in 2011 (Table 2).

Table 3. Statistical comparison of aquatic plant frequency data within 2011 treatment areas on North Twin Lake from 2010 pre- and 2011 post treatment surveys. Only species with greater than 5.0% frequency of occurrence in at least one of the two surveys are applicable for analysis.

	Scientific Name	Common Name	2010 FOO	2011 FOO	Percent Change	Direction	Chi-square Analysis Statistically Valid	p-value
Dicots	*Myriophyllum spicatum*	Eurasian water milfoil	15.0	12.5	-16.7	▼	No	0.745
	Ceratophyllum demersum	Coontail	60.0	70.0	16.7	▲	No	0.348
	Myriophyllum sibiricum	Northern water milfoil	7.5	0.0	-100.0	▼	No	0.077
	Myriophyllum alterniflorum	Alternate-flowered water milfoil	0.0	7.5	100.0	▲	No	0.077
Non-dicots	*Chara spp.*	Muskgrasses	12.5	0.0	-100.0	▼	Yes	0.021
	Najas flexilis	Slender naiad	20.0	2.5	-87.5	▼	Yes	0.013
	Potamogeton amplifolius	Large-leaf pondweed	0.0	10.0	100.0	▲	Yes	0.040
	Elodea canadensis	Common waterweed	80.0	70.0	-12.5	▼	No	0.302
	Vallisneria americana	Wild celery	50.0	37.5	-25.0	▼	No	0.260
	Potamogeton zosteriformis	Flat-stem pondweed	27.5	12.5	-54.5	▼	No	0.094
	Potamogeton robbinsii	Fern pondweed	15.0	10.0	-33.3	▼	No	0.499
	Potamogeton pusillus	Small pondweed	12.5	2.5	-80.0	▼	No	0.090
	Potamogeton gramineus	Variable pondweed	5.0	2.5	-50.0	▼	No	0.556
	Potamogeton richardsonii	Clasping-leaf pondweed	5.0	15.0	200.0	▲	No	0.136

2010 N = 40, 2011 N =40
FOO = Frequency of Occurrence
▲ or ▼ = Change Statistically Valid (Chi-square; α = 0.05)
▲ or ▼ = Change Not Statistically Valid (Chi-square; α = 0.05)

As previously indicated, 8 sub-sample locations were contained within Site E-11 and were sampled during the spring previous to the 2011 treatment and summer following this treatment. Two of these locations contained EWM before the treatment and 3 contained EWM following the treatment. While this sample size is too small for statistical analysis, these data clearly indicate that EWM continues to exist within this area following the herbicide treatment.

Renovate Max G is a relatively new herbicide that has not been widely used in Wisconsin. In efforts to learn more about this product, the herbicide manufacturer (SePRO) conducted residual monitoring at sites within 2011 treatment sites on North Twin Lake up to 40 hours following the application. Their preliminary results indicate that a pulse of Renovate Max G herbicide occurred between 4 and 8 hours following application and herbicide residuals were below 0.100 ppm a.e. by 40 hours after treatment. The herbicide levels were the highest for the three largest treatment sites (A-11, B-11, & G-11) and extremely low (< 0.200 ppm a.e.) for the smallest site (I-11). The full results of SePRO's study may be available at a later date.

2012 Treatment Strategy

Overall, the 2011 EWM treatment on North Twin Lake met with mixed results. Neither the quantitative nor qualitative criteria were met for the 2011 treatment. Also, 13.5 acres are proposed for treatment in 2012 (Map 9), just slightly less than what was treated in 2011 (Map 8).

The re-treatment of previously treated areas is not uncommon in EWM management as dense areas often require multiple years of treatment to significantly reduce a site's density and/or size. As discussed in past treatment reports, in order to reduce the treatable acreage of EWM on a lake, it is believed that multiple years of treatment is the key to success. Great strides in EWM control have been made in recent years on North and South Twin Lakes, but it must be noted that the remaining areas of EWM on North Twin Lake have been treated for multiple years.

During the 2011 summer surveys, a few areas where EWM has rebounded on South Twin Lake were located (Map 9). These areas are small, but it is recommended that these areas are aggressively targeted in 2012 to prevent these colonies from getting larger and spreading to new areas. Approximately 4.8 acres are proposed to be treated on South Twin Lake in 2012 (Map 9).

Please note that areas less than 0.5 acres were not proposed for treatment due to their extremely small size and unlikely nature of being treated successfully. As indicated above, treatment areas less than 5 acres fall into the micro-treatment category of spot treatments and are difficult to predict if they will be effective because of the rapid dilution of the herbicide. All of the 2012 proposed treatments on North and South Twin fall into this category. Therefore the proposed treatment strategy for North and South Twin Lakes includes an expanded buffer (40-foot) around the EWM colonies as well as a higher proposed application rate of herbicide (Map 9).

As previously indicated, this report marks the end of the current AIS Established Population Control (AIS EPC) Grant-funded projects (ACEI-029-08 & ACEI-079-10). Because herbicide application costs for some of the treatment years was likely less than budgeted for within these grants, the association could pursue extending the timeframe and scope of the project to include cost-coverage for the 2012 EWM treatment and associated monitoring. But if this is not an option, a prioritized treatment and continued monitoring may be appropriate for 2012 until additional funding becomes available.

NORTH & SOUTH TWIN LAKES 2008-2011 TREATMENT REVIEW

The goal of this four-year project was to control EWM on North and South Twin Lakes and reduce it to more manageable levels in an attempt to maintain the lake's ecological integrity. In this section, the overall effectiveness of this project is analyzed for its effectiveness in reducing EWM while at the same time minimizing impacts to the native aquatic plant community.

North Twin Lake

Figure 12 displays the acreage of EWM colonies located and mapped each year from 2007 to 2011 on North Twin Lake. As illustrated, the acreage of EWM colonies in North Twin Lake has increased over this time period. However, the density of EWM has been greatly reduced over this same time period. The majority of the colonies mapped in 2007-2009 were classified as having dominant or highly dominant EWM, while in 2010 and 2011, the majority of colonies had been reduced to scattered and highly scattered EWM. In addition, the acreage of highly scattered EWM in 2010 and 2011 is wholly comprised by a single colony of EWM that was discovered inside a dense bulrush colony 2010. This area has not been targeted for treatment due to its low density of EWM.

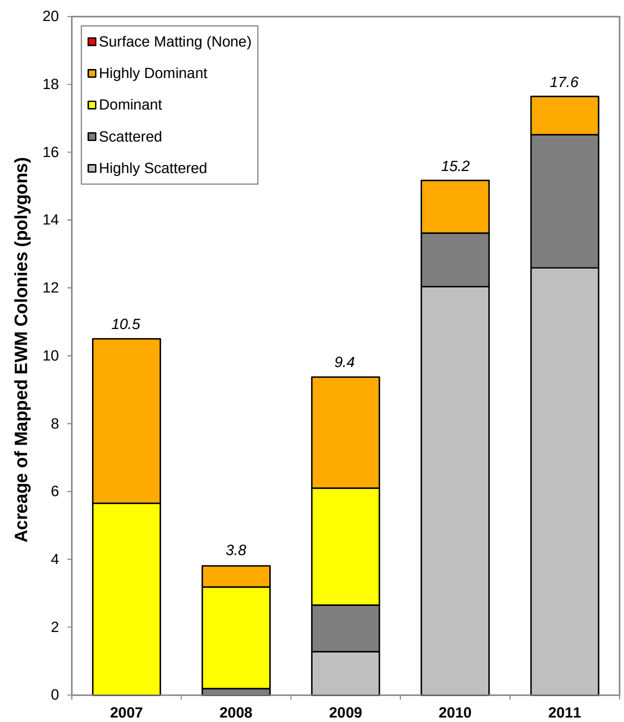

Figure 12. Acreage of mapped EWM colonies on North Twin Lake.

Two main areas in North Twin Lake, Sites A-11 and B-11 (Map 8) have been the target of herbicide treatments during this project. Actually, these areas have been treated for 5 consecutive years. Quantitative data collected from slightly less than 30 point-intercept locations in 2007-2011 from these sites provide valuable insight to how the herbicide treatments have affected the target species (EWM) as well as the non-target native plants.

Figure 13 shows that over the course of this project, EWM was reduced from an occurrence of 79% in 2007 to 7.1% in 2011, a statistically valid reduction of 91%. Within these treatment sites, no native aquatic plant species displayed statistically valid reductions over the course of this project (Figure 14). Two native aquatic plant species, coontail and common waterweed, increased in occurrence by 125% and 450%, respectively, from 2007 to 2011. The large increase in occurrence of these species may indicate that they are colonizing areas once occupied by EWM.

Overall, this four-year project has been very successful at reducing the occurrence of EWM while minimizing impacts to the native aquatic plant community on North Twin Lake. Areas that have been targeted for multiple years have seen large declines in EWM. As discussed in the previous section, areas of EWM still exist that require retreatment in 2012 in addition to two new areas discovered in 2011.

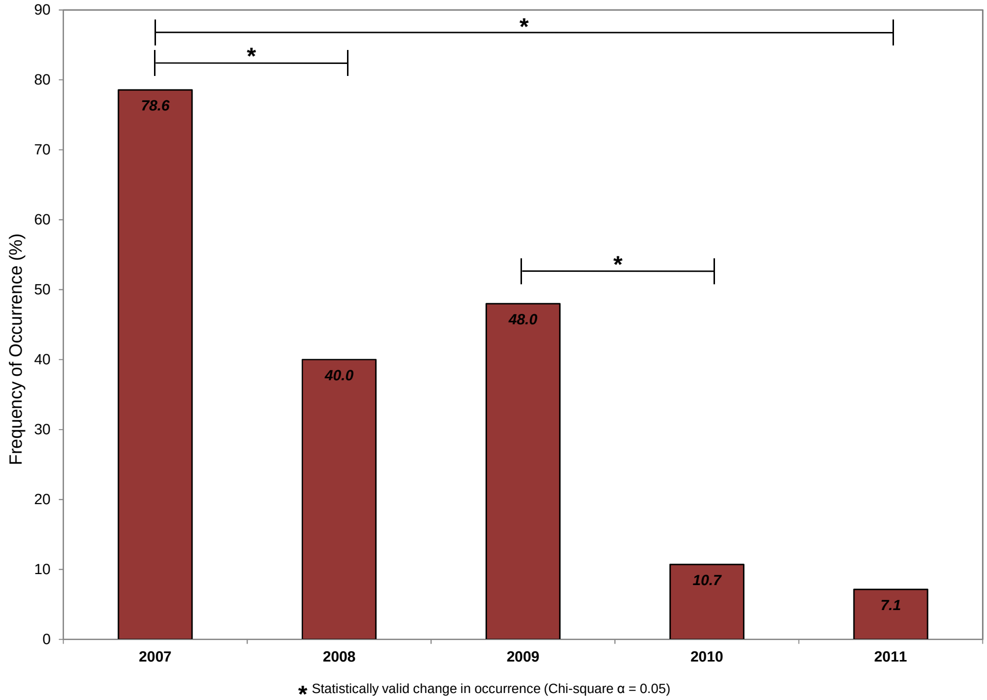

Figure 13. EWM frequency of occurrence within treatment sites A-11 and B-11 treated annually from 2008-2011. Created using sub-sample point-intercept data from 2007-2011.

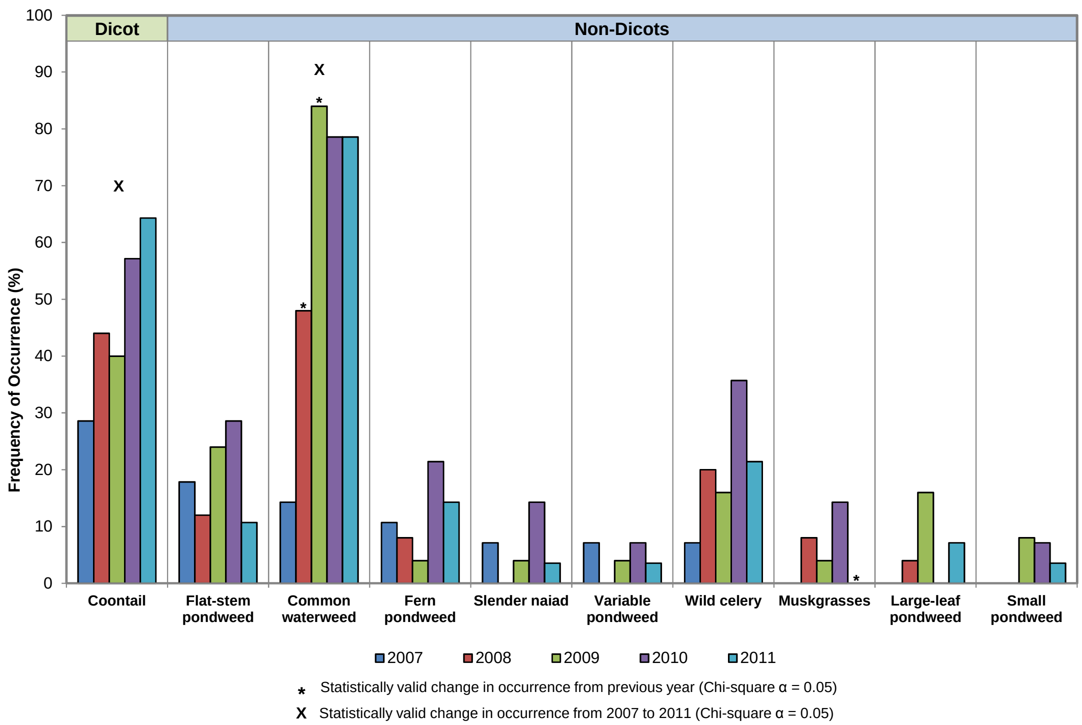

Figure 14. Frequency of occurrence of ten frequently encountered native aquatic plant species within treatment sites A-11 and B-11 treated annually from 2008-2011. Created using sub-sample point-intercept data from 2007-2011.

South Twin Lake

While EWM has only been located in isolated colonies in the southeastern portion of North Twin Lake over the course of this project, EWM was widespread throughout littoral areas in South Twin Lake at the beginning of this project. Spot treatments over areas of EWM conducted in 2007 and 2008 met with little or no success. In 2008, approximately 135 acres of colonized EWM was mapped on South Twin Lake (Figure 15).

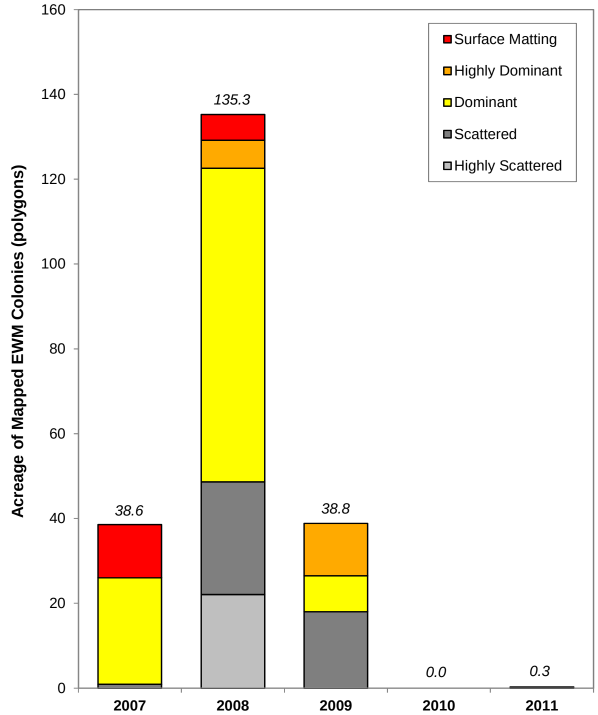

Figure 15. Acreage of mapped EWM colonies on South Twin Lake.

A more aggressive strategy was undertaken in 2009 where a whole-lake liquid 2,4-D treatment was implemented. The intent of that treatment was to apply liquid 2,4-D at the rate of 1.75 ppm a.e. over the treatment areas and as a result, reach a lake-wide concentration of 0.167 ppm a.e. Based upon the herbicide residual monitoring of the treatment, South Twin Lake's measured herbicide levels were between 0.100 and 0.150 ppm a.e. for at least 7 days. While the target concentration of 0.167 ppm a.e. was not met, the levels did remain at or above 0.100 ppm a.e. for an extended time. Colonized EWM reduced drastically after the 2009 treatment (Figure 15) and lake-wide EWM frequency of occurrence reduced from 20.7% to 10.2% (Figure 16). The treatment proved to be effective and was proposed for use again in 2010, but at a slightly higher herbicide concentration.

The target concentration of the 2010 herbicide treatment was 0.240 ppm a.e. However, the mean lake-wide herbicide concentration on South Twin Lake from application to 7 days after treatment was measured to be 0.575 ppm a.e. – almost 2.5 times higher than the target concentration. While emerging results appear clear that liquid 2,4-D mixes horizontally within the lake, little information existed at that time regarding if 2,4-D mixes vertically into deep areas of the lake during stratification. After discussions between Onterra and John Skogerboe from the USACE, it was hypothesized that if the lake was thermally stratified, the 2,4-D would dissipate throughout the warmer, upper zone of the lake (epilimnion), but not into the cooler, deeper water zones of the lake (metalimnion and hypolimnion). This hypothesis was tested on a few lakes and the results indicate that the herbicide residuals did not vertically mix throughout the entire water column. While this was not specifically tested on South Twin Lake, it is likely that the lake was thermally stratified at the time of the treatment and the 2,4-D only mixed within the upper zone of the lake contributing to the higher than expected 2,4-D concentrations.

No EWM was located in South Twin Lake following the 2010 treatment, and therefore no treatment was conducted in 2011. Figure 15 illustrates the extreme decline in colonial acreage of EWM following the large-scale treatments in 2009 and 2010.

Because the goal of the large-scale treatments conducted in 2009 and 2010 was to disperse herbicide throughout the entire lake, annual treatment monitoring was also conducted on a lake-wide scale. The whole-lake point-intercept survey, as discussed in the Aquatic Plant Section, comprised of 621 point-intercept sampling locations was completed by the WDNR in the summer of 2008 and by Onterra in the summers of 2009, 2010, and 2011. Although no herbicide treatment occurred on South Twin Lake in 2011, a whole-lake point-intercept survey was conducted to continue an ongoing study in cooperation with the WDNR and USACE to assess the 2009 and 2010 large-scale treatment in terms of EWM control and non-target effects on the native aquatic plant community one year later.

Figure 16 shows that the large-scale treatments conducted in 2009 and 2010 were successful in reducing the lake-wide occurrence of EWM. Over the course of this project, the littoral frequency of occurrence of EWM in South Twin Lake was reduced from 20.7% in 2008 to 0.3% in 2011 (Figure 16). This reduction represents a statistically valid reduction in occurrence of over 98%.

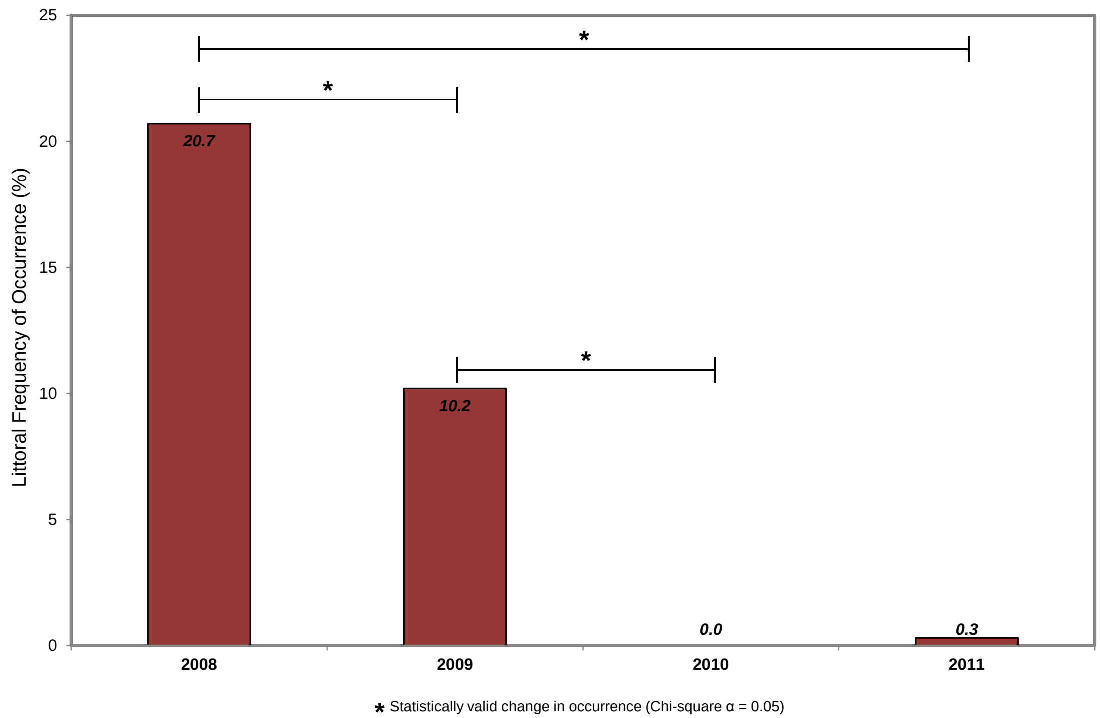

Figure 16. EWM littoral frequency of occurrence within South Twin Lake from 2008-2011. Created using sub-sample point-intercept data from 2007-2011.

In the Aquatic Plant Section, the native aquatic plant community of South Twin Lake was shown to have maintained its high quality throughout this four-year project. The most notable changes to the aquatic plant community brought forth within the floristic quality analysis was that northern water milfoil, dwarf water milfoil, and white-water crow foot were impacted by the herbicide treatment program to the degree that they were not located during subsequent point-intercept surveys. It is likely that these three dicots continue to exist within South Twin Lake, but at population levels undetectable by this survey methodology. North Twin Lake contains healthy populations of northern water milfoil (~17% littoral frequency) and white-water crowfoot (~11% littoral frequency of occurrence) and it is suspected that over time, these species will increase in occurrence in South Twin Lake as result of expansion of current populations or colonization from North Twin Lake. Please note that dwarf water milfoil is not known to exist in North Twin Lake.

Looking at this data more closely, some native species were not lost completely as a result of the treatment, but their populations were significantly reduced (Figure 17). Another dicot species, water marigold reduced in occurrence following the large-scale treatments. One native dicot species, coontail, did not exhibit a statistically valid change in occurrence over the course of the project.

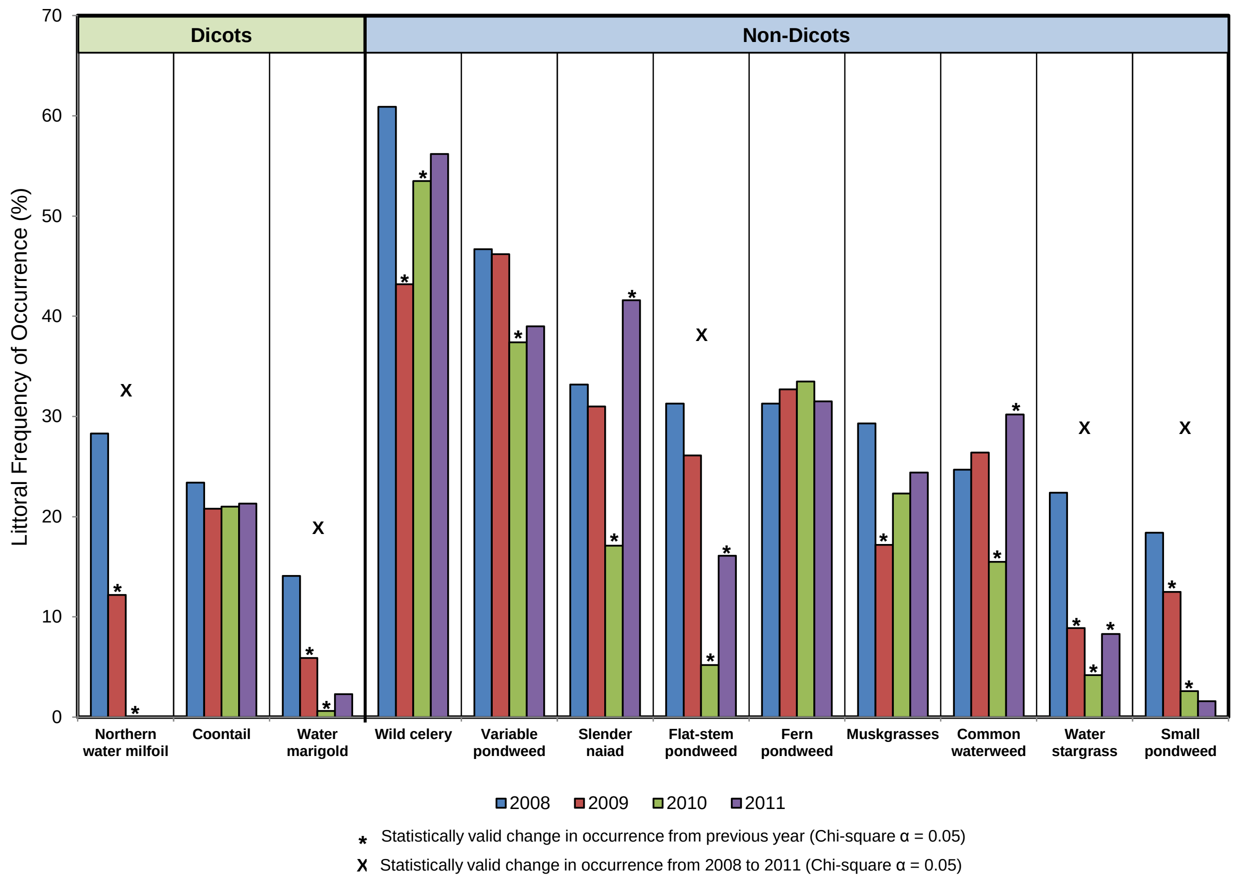

Figure 17. Littoral frequency of occurrence of 12 frequently encountered native aquatic plant species in South Twin Lake from 2008-2011. Created using sub-sample point-intercept data from 2007-2011.

Flat-stem pondweed, water stargrass, and small pondweed also demonstrated statistically valid reductions in occurrence between 2008 and 2011. These species are monocots which were historically not thought to be particularly sensitive to dicot-selective herbicides like 2,4-D. However, data gathered from South Twin Lake and other Wisconsin lakes with similar large-scale liquid treatments, suggests that some of these plants may be prone to decline after treatment.

The 2011 survey on South Twin Lake revealed that some native aquatic plant species had rebounded after declining following the 2009 and 2010 treatments. The occurrences of wild celery, variable pondweed, muskgrasses, and common waterweed declined following the 2009 and/or the 2010 treatment. However, in 2011 these plant species increased in occurrence to levels that were determined not to be statistically different from before whole-lake treatments occurred (2008) (Figure 17). While flat-stem pondweed and water stargrass continue to be statistically less frequent than in 2008, these species have also rebounded (statistically valid) in occurrence in 2011.

Overall, this four-year EWM control project on North and South Twin Lakes has decreased the targeted EWM population. Treatment effectiveness on these lakes is largely a result of treating a particular area multiple times to reduce the population to levels that no longer warrant treatment. The data show that some native aquatic plant species in South Twin Lake have been impacted by the treatments and this list of species includes both dicot and non-dicot species. However, the aquatic plant community as a whole has maintained a high diversity and is of higher quality than most lakes in the region and the state. The resurgence of many species was also observed in 2011, an indicator of their recovery potential.

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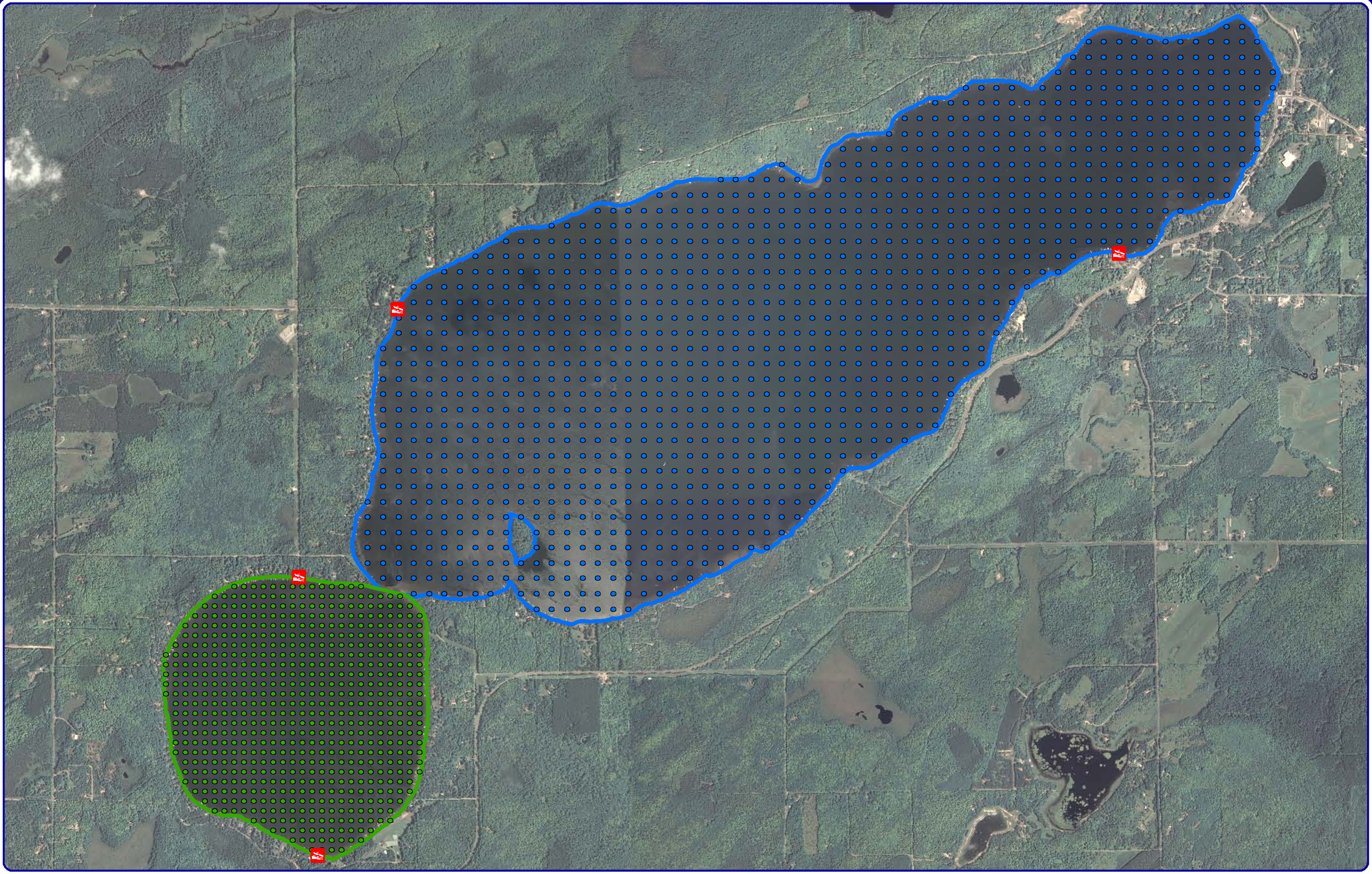

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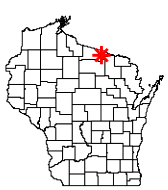
www.onterra-eco.com

Sources:
Hydro: WDNR
Orthophotography: NAIP, 2010
Map Date: January 10, 2012
Filename: *Map1_NSTwin_Location.mxd*



Project Location in Wisconsin

Legend

North Twin Lake ~2,788 acres

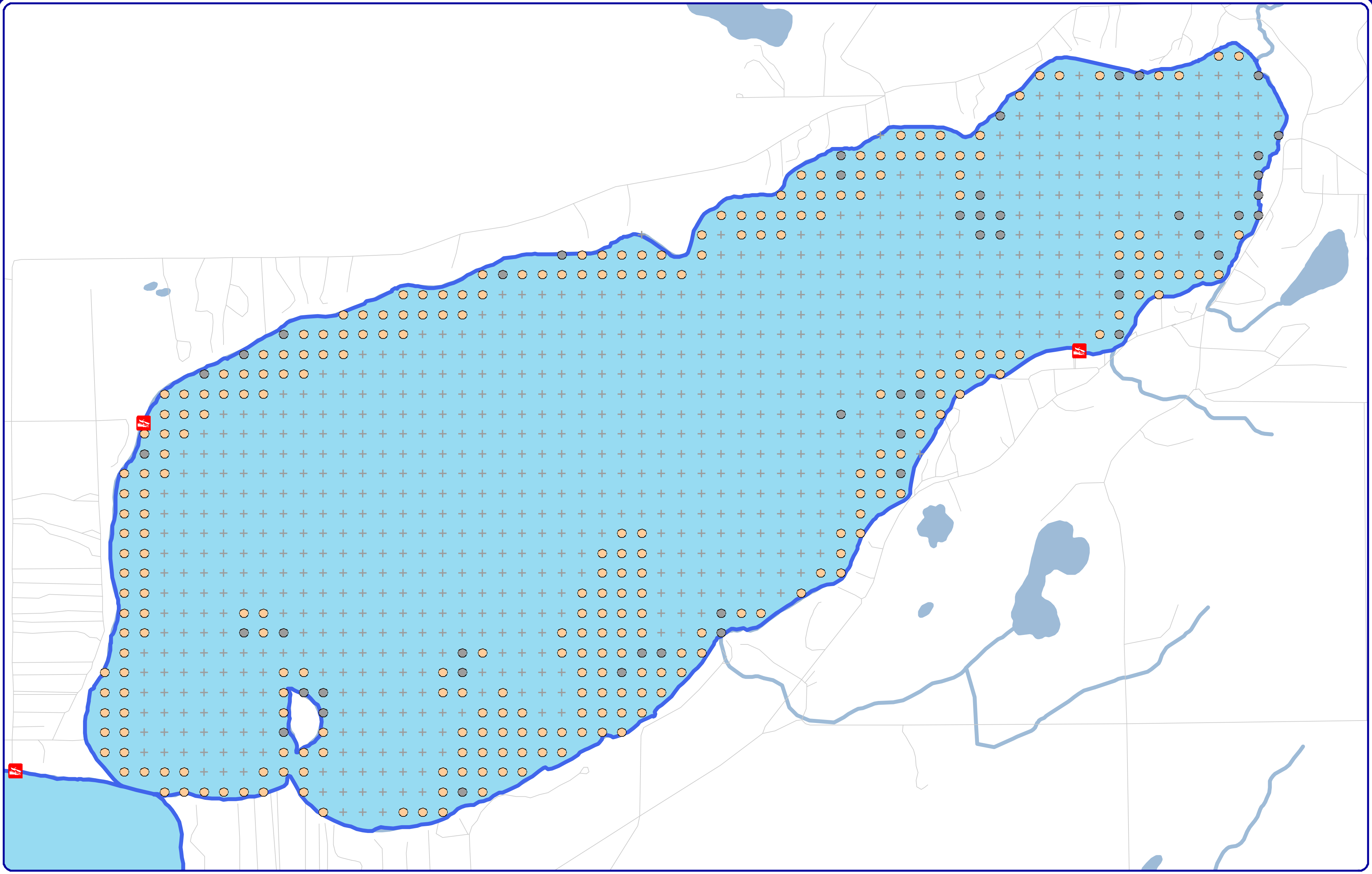
South Twin Lake ~642 acres

Point-intercept Survey Location
100-meter spacing - 1,163 total points

Point-intercept Survey Location
63-meter spacing - 621 total points

Public Access

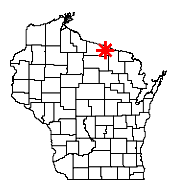
Map 1
North & South Twin Lakes
Vilas County, Wisconsin
Project Location
& Lake Boundaries



2,100
Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Roads and Hydro: WDNR
Orthophotography: NAIP, 2010
Aquatic Plant Survey: Onterra, 2011
Map Date: January 10, 2012
Filename: Map2_NTwin_SubstratePI.mxd



Project Location in Wisconsin

Legend

Point-intercept Sampling Locations

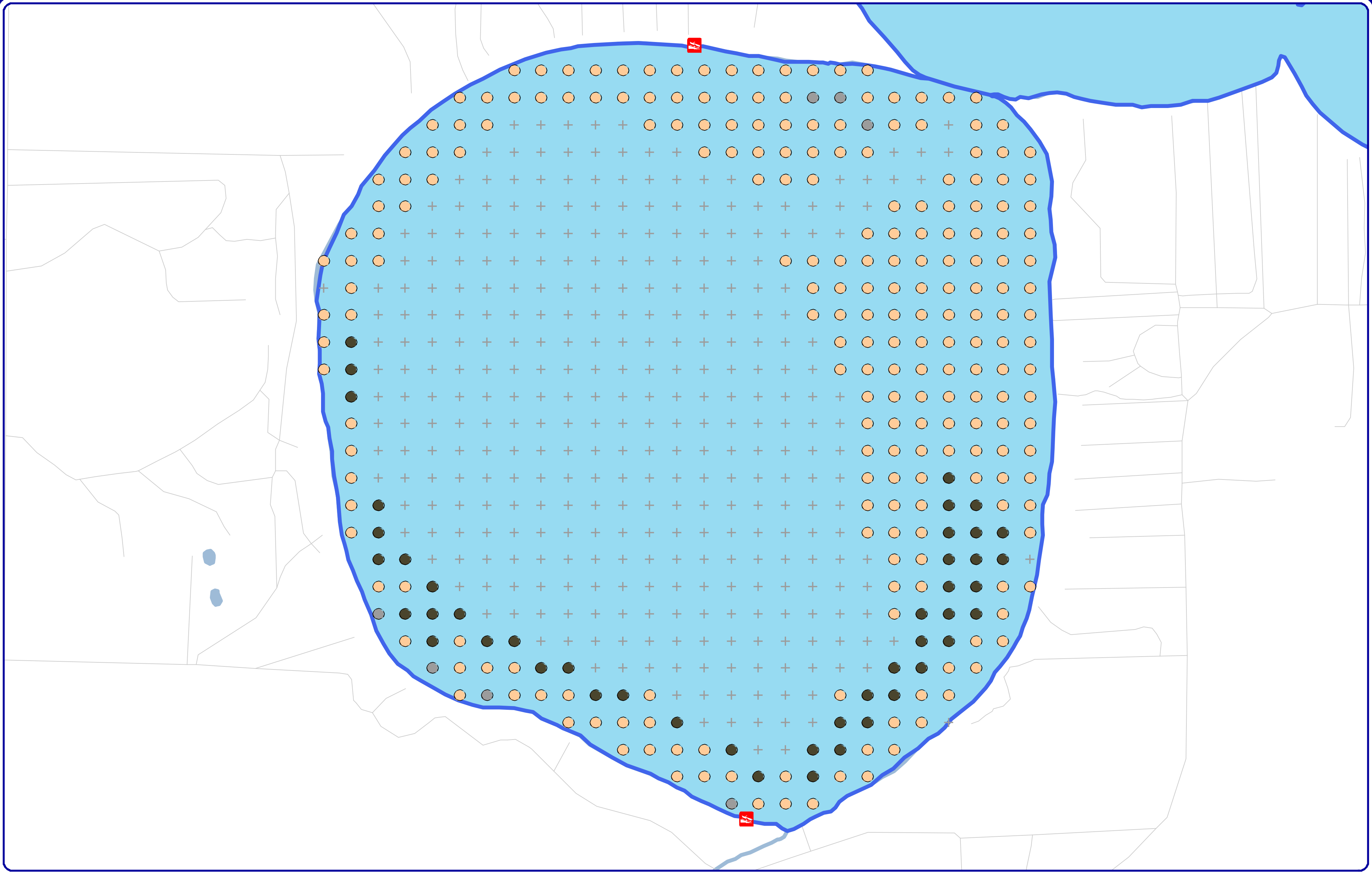
- No Data *(Non-navigable or Too Deep)*
- Muck *(None)*
- Sand
- Rock

Map 2

North Twin Lake

Vilas County, Wisconsin

PI Survey: Substrate Types

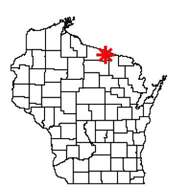


1,000

Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Roads and Hydro: WDNR
Orthophotography: NAIP, 2010
Aquatic Plant Survey: Onterra, 2011
Map Date: January 10, 2012
Filename: *Map3_STwin_SubstratePI.mxd*



Project Location in Wisconsin

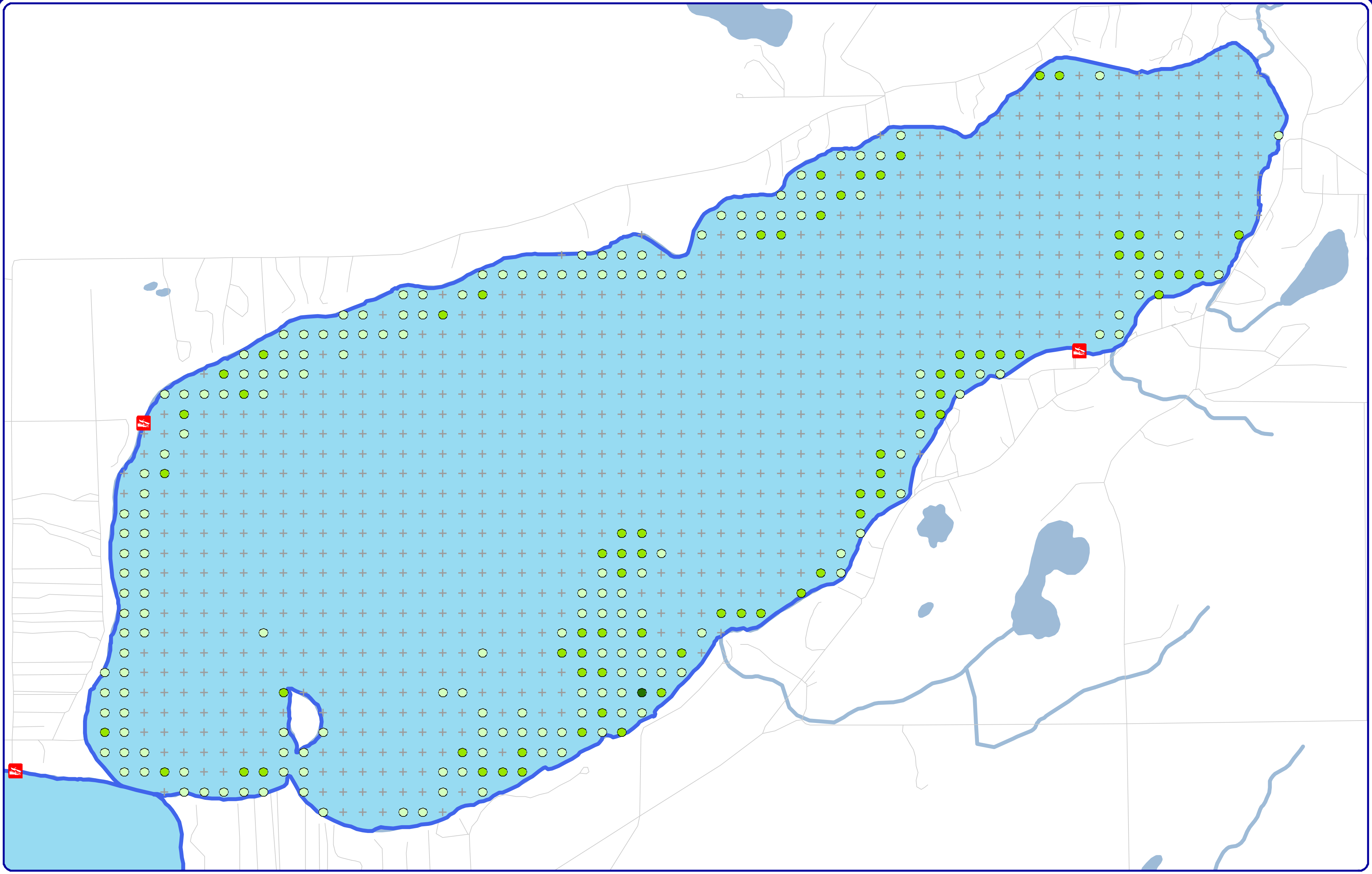
Legend

Point-intercept Sampling Locations

- + No Data *(Non-navigable or Too Deep)*
- Muck
- Sand
- Rock

Map 3
South Twin Lake
Vilas County, Wisconsin

PI Survey: Substrate Types

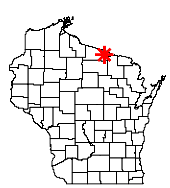


2,100

Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Roads and Hydro: WDNR
Orthophotography: NAIP, 2010
Aquatic Plant Survey: Onterra, 2011
Map Date: January 10, 2012
Filename: *Map4_NTwin_TRFPI.mxd*



Project Location in Wisconsin

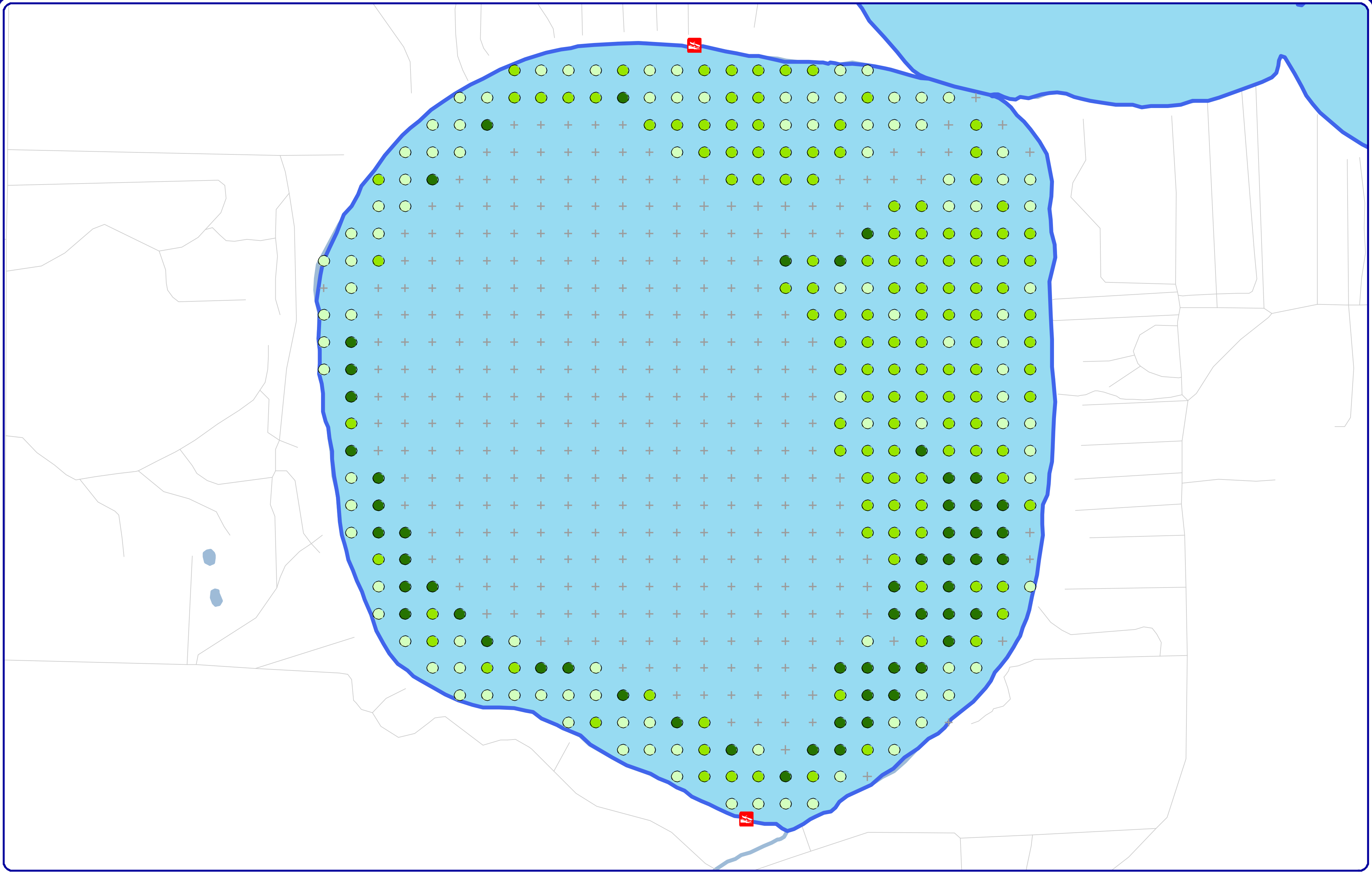
Legend

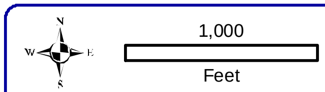
Point-intercept Sampling Locations

- No Vegetation or Non-navigable
- Rake-fullness = 1
- Rake-fullness = 2
- Rake-fullness = 3

Map 4
North Twin Lake
Vilas County, Wisconsin

PI Survey:
Aquatic Plant Distribution

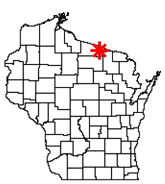




Onterra LLC
Lake Management Planning

815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Roads and Hydro: WDNR
Orthophotography: NAIP, 2010
Aquatic Plant Survey: Onterra, 2011
Map Date: January 10, 2012
Filename: *Map5_STwin_TRFPI.mxd*



Project Location in Wisconsin

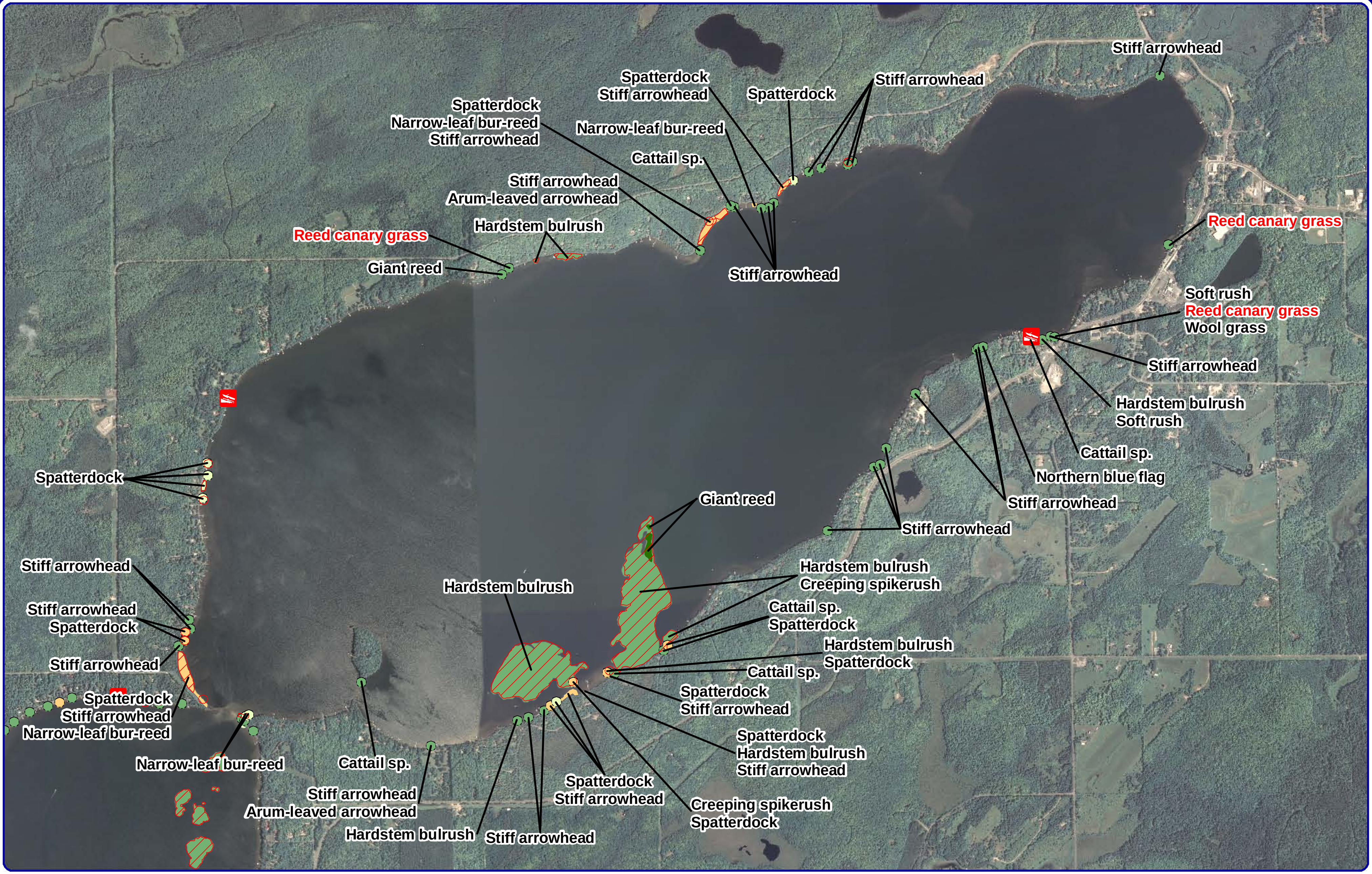
Legend

Point-intercept Sampling Locations

- + No Vegetation or Non-navigable
- Rake-fullness = 1
- Rake-fullness = 2
- Rake-fullness = 3

Map 5
South Twin Lake
Vilas County, Wisconsin

PI Survey:
Aquatic Plant Distribution

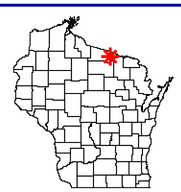


2,500

Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Aquatic Plants: Onterra, 2011
Orthophotography: NAIP, 2010
Map date: December 21, 2011
Filename: Map6_NTwin_Comm_2011.mxd



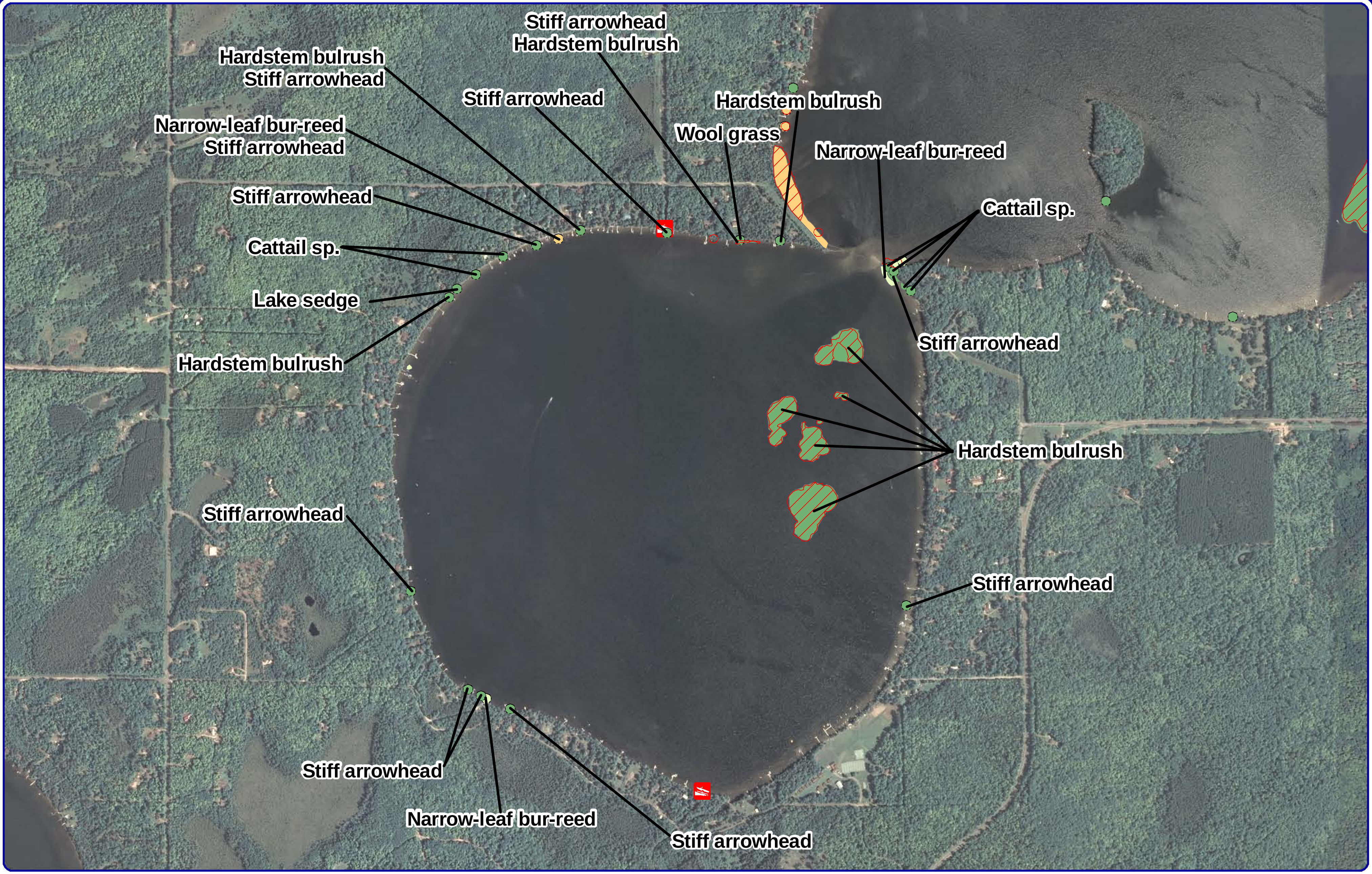
Project Location in Wisconsin

Legend

Small Plant Communities
- Emergent
- Floating-leaf
- Mixed Floating-leaf & Emergent
- 2004 Small Plant Community

Large Plant Communities
- Emergent
- Floating-leaf
- Mixed Floating-leaf & Emergent
- 2004 Large Plant Community

Map 6
North Twin Lake
Vilas County, Wisconsin
Floating-leaf & Emergent Aquatic Plant Communities

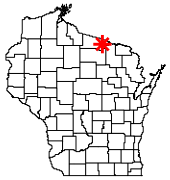


1,375

Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Aquatic Plants: Onterra, 2011
Orthophotography: NAIP, 2010
Map date: December 21, 2011
Filename: Map7_STwin_Comm_2011.mxd



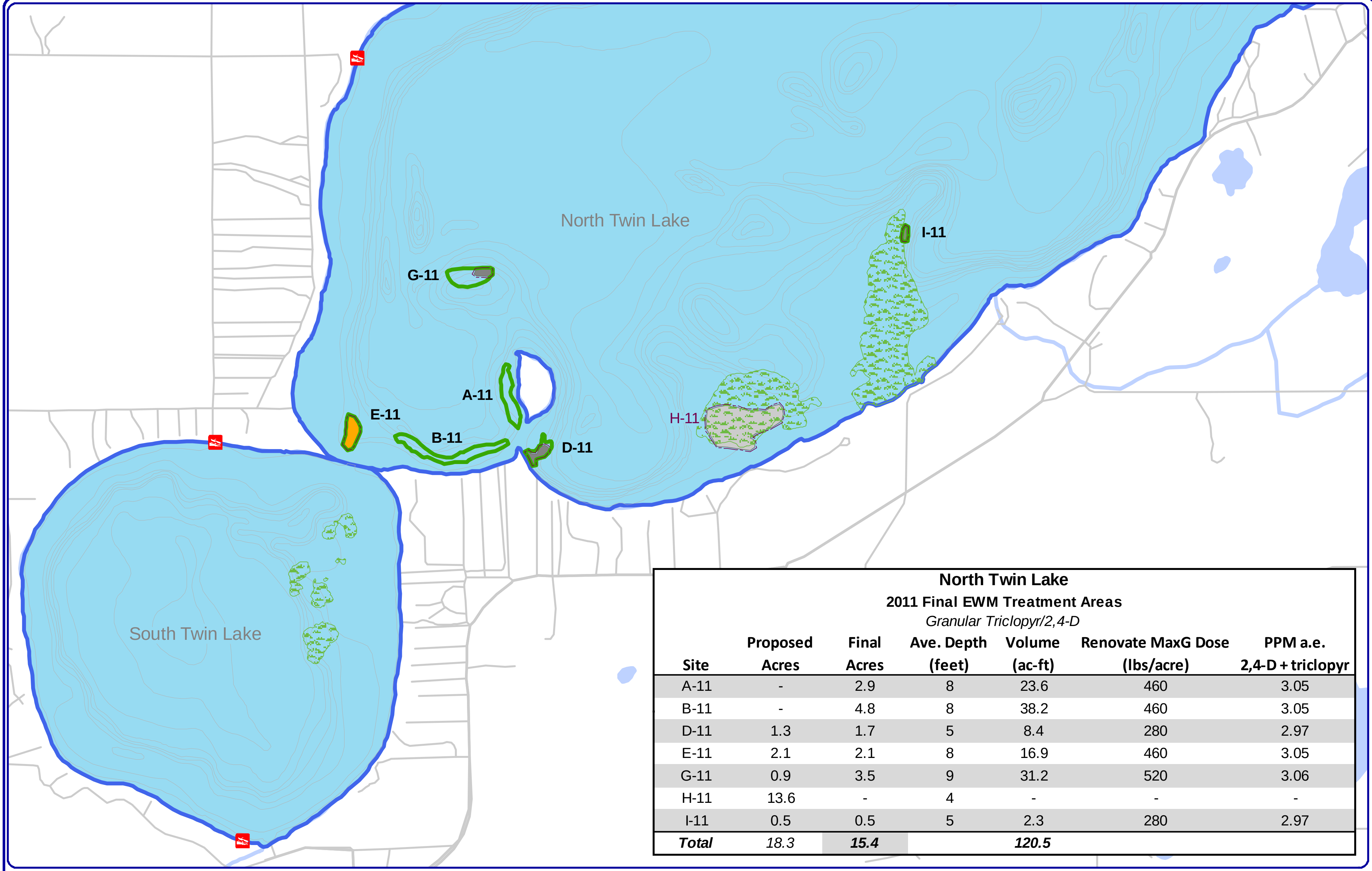
Project Location in Wisconsin

Legend

Small Plant Communities
- Emergent
- Floating-leaf
- Mixed Floating-leaf & Emergent
- 2004 Small Plant Community

Large Plant Communities
- Emergent
- Floating-leaf
- Mixed Floating-leaf & Emergent
- 2004 Large Plant Community

Map 7
South Twin Lake
Vilas County, Wisconsin
Floating-leaf & Emergent Aquatic Plant Communities



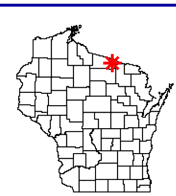
North Twin Lake
2011 Final EWM Treatment Areas
Granular Triclopyr/2,4-D

Site	Proposed Acres	Final Acres	Ave. Depth (feet)	Volume (ac-ft)	Renovate MaxG Dose (lbs/acre)	PPM a.e. 2,4-D + triclopyr
A-11	-	2.9	8	23.6	460	3.05
B-11	-	4.8	8	38.2	460	3.05
D-11	1.3	1.7	5	8.4	280	2.97
E-11	2.1	2.1	8	16.9	460	3.05
G-11	0.9	3.5	9	31.2	520	3.06
H-11	13.6	-	4	-	-	-
I-11	0.5	0.5	5	2.3	280	2.97
Total	*18.3*	***15.4***		***120.5***		

1,900
Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

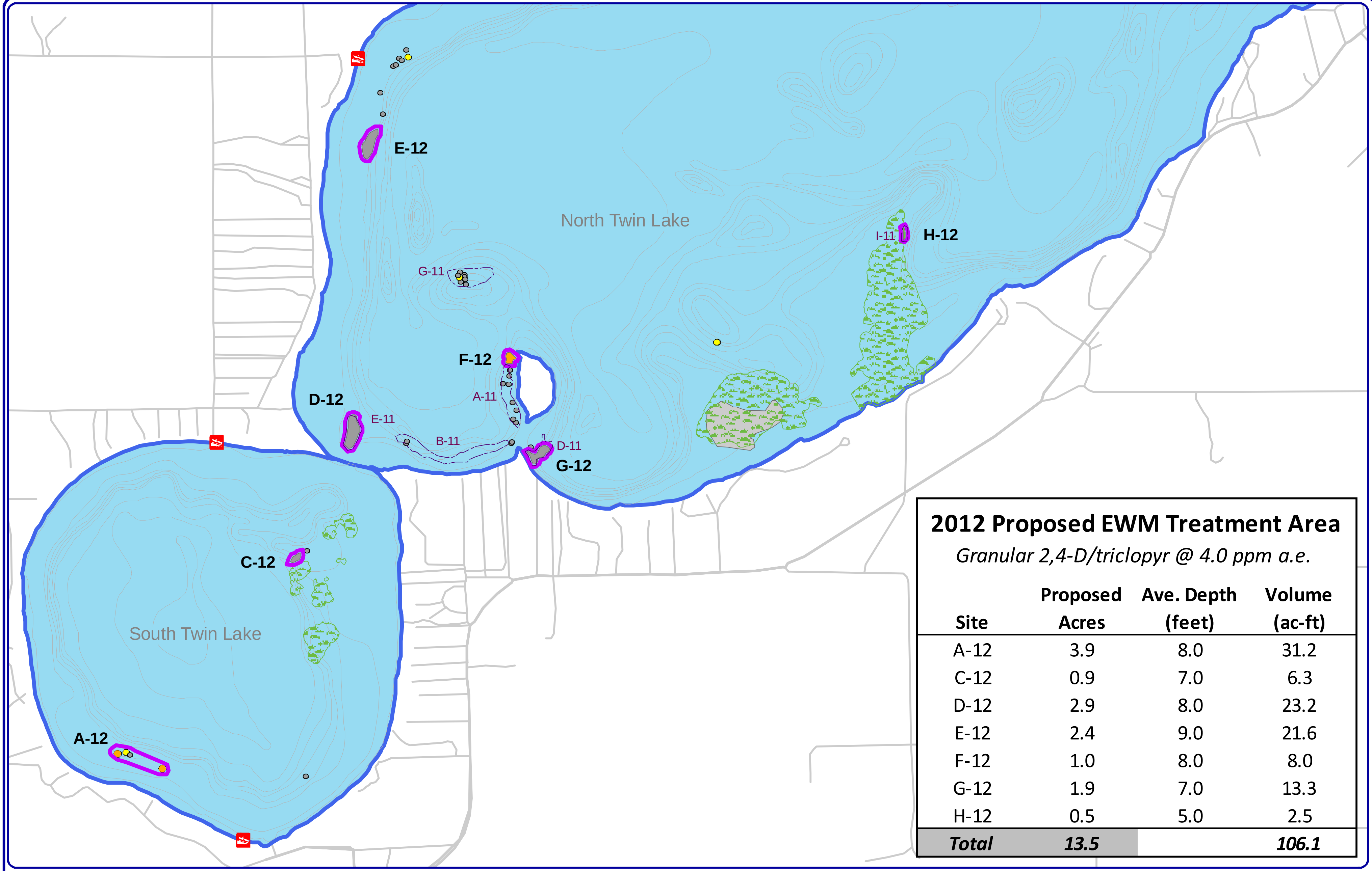
Sources:
Roads and Hydro: WDNR
Aquatic Plant Survey: Onterra 2010-11
Map Date: January 6, 2012
Filename: *Map8_NSTwin_EWM_2010PB_T2011.mxd*


Project Location in Wisconsin

Legend

Eurasian water milfoil
(September 2010)
- Highly Scattered
- Scattered
- Dominant *(None)*
- Highly Dominant
- Surface Matting *(None)*
- 2011 Conditional Treatment Area
- 2011 Final Treatment Area
- Hardstem Bulrush Communities
- Public Access

Map 8
North & SouthTwin Lakes
Vilas County, Wisconsin
2010 EWM Locations & 2011 Treatment Areas



2012 Proposed EWM Treatment Area

Granular 2,4-D/triclopyr @ 4.0 ppm a.e.

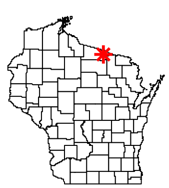
Site	Proposed Acres	Ave. Depth (feet)	Volume (ac-ft)
A-12	3.9	8.0	31.2
C-12	0.9	7.0	6.3
D-12	2.9	8.0	23.2
E-12	2.4	9.0	21.6
F-12	1.0	8.0	8.0
G-12	1.9	7.0	13.3
H-12	0.5	5.0	2.5
Total	***13.5***		***106.1***

1,900

Feet

Onterra LLC
Lake Management Planning
815 Prosper Rd
De Pere, WI 54115
920.338.8860
www.onterra-eco.com

Sources:
Roads and Hydro: WDNR
Aquatic Plant Survey: Onterra 2010-11
Map Date: January 6, 2012
Filename: *Map9_NSTwin_EWM_T2012_Cond1.mxd*



Project Location in Wisconsin

Legend

Eurasian water milfoil ***(September 2011)***

- Highly Scattered
- Scattered
- Dominant *(None)*
- Highly Dominant
- Surface Matting *(None)*
- Single or Few Plants
- Clumps of Plants
- Small Plant Colony
- 2011 Final Treatment Area
- 2012 Proposed Treatment Area
- Hardstem Bulrush Communities

Map 9

North & SouthTwin Lakes

Vilas County, Wisconsin

2011 EWM Locations & 2012 Proposed Treatment Areas v.1