Management in Practice

Aquatic plant community restoration following the long-term management of invasive *Egeria densa* with fluridone treatments

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Abstract

The Sacramento-San Joaquin Delta is one of the largest estuaries in North America, providing water for more than 700,000 acres of agriculture, recreation and fisheries habitat. For decades, the exotic invasive plant *Egeria densa* has negatively impacted native habitat and navigation of vessels in the Sacramento-San Joaquin Delta. In 2007 the largest waterbody in the Delta, Franks Tract, began to be managed at operational scale utilizing the aquatic herbicide fluridone. A fluridone pellet formulation was applied to achieve in-water concentrations of fluridone between 2.5 and 3.5 ppb for 8 to 16 weeks in areas with dense *Egeria densa*. Fluridone applications were started as early as March and continued throughout the treatment period to sustain the target concentrations which were verified by an enzyme-linked immunoassay (ELISA) analytical test. Relative frequency of occurrence for native plants significantly increased from 2006 to 2017 ($P < 0.001$). Frequency of occurrence of most native species remained variable across years except for *Potamogeton richardsonii* where frequency of occurrence increased greatly from 3.6% in 2013 to 80% in 2017 ($P < 0.001$), and significantly increased each year sequentially except between 2015 to 2016 to become the most widespread species. The increase of native plants over the past five years, following management with fluridone, is likely to improve fisheries, native species habitat, and waterway traffic.

Key words: invasive aquatic plant, herbicide, native plant restoration, aquatic herbicides, macrophyte, aquatic weeds

Introduction

Egeria densa is an aquatic weed endemic to South America that has become invasive throughout many places in the world, often as a result of unintentional introductions (Matheson et al. 2005). *Egeria densa*'s ability to grow quickly and proliferate in low light conditions has made it an ideal aquarium plant and resulted in the transportation of this plant to many places of the world (June-Wells et al. 2012). Many introductions of aquatic invasive species are likely due to the unintentional release of aquarium plants, hitchhiking on water craft, or through propagule dispersal (Strecker et al. 2011; Havel et al. 2015). The Sacramento-San Joaquin Delta (hereafter referred to as the "Delta") has been infested with *Egeria densa* since the 1980's and fluridone herbicide treatment for the invasive plant has been going on since the late 1990s (Anderson 1999; Pennington and Sytsma 2009). Fluridone interferes with photosynthesis by blocking critical pigment production (carotenoids) that in turn results in the degradation and reduction of chlorophyll, leading to a "bleaching" effect in the leaves (Zou et al. 2018). Fluridone's mode of action avoids toxicity on non-target species such as mammals and fish and reduces impact to non-target plants through a combination of pellet placement in areas with highest *Egeria densa* densities and low target herbicide concentrations designed to impact the more sensitive invasive macrophyte (Arnold 1979; Hill et al. 2017). Managing invasive aquatic plants in the Delta is challenging due to the tidal system, where there are two tides daily. With tides and flowing water, maintaining an effective concentration of fluridone is significantly more challenging compared to static waterbodies. Additionally, fluridone must be kept under 5 ppb around any areas where the water could be used for irrigating plants within the Solanaceae family (SePRO 2017).

Invasive aquatic plants have negative impacts not only on habitat for native species, but also the economy due to the costs to manage them as well as the loss of recreation and other economic impacts. The dense monocultural stands of *Egeria densa* can damage native ecosystems by altering aquatic community habitat and restricting access to boaters (Hestir et al. 2008; Underwood et al. 2006). Invasive species control is a global issue due to intercontinental trade and the ability for invasive plants and animals to move across continents in a matter of days or hours. The European Union has taken a number of steps to ban certain invasive species and prevent new infestations from forming, as control of these species is much more costly than prevention (Official Journal of the European Union 2014). The state of Florida alone spends around $14.5 million annually to control *Hydrilla verticillata*, an invasive plant similar to *Egeria densa* (Center et al. 1997). *Hydrilla* infestations in just two Florida lakes have prevented recreation use, causing an annual loss of 10 million dollars (Center et al. 1997). A total of $100 million is invested annually in invasive aquatic weed control in the United States (OTA 1993).

There appears to be a need for additional research that relates to the restoration of native plant communities in response to the control and management of invasive plant species in different waterbodies. Studies on fluridone and invasive aquatic plant control tend to focus on efficacy on target invasive plants without necessarily reviewing the potential for restoration and improving native plant communities. A study in Loomis Lake, Washington showed species richness recovering starting two years after fluridone treatment (Parsons et al. 2009). Native plant recovery has also been observed in several Minnesota lakes after fluridone treatment (Crowell et al. 2006; Valley et al. 2006). Fluridone has been a useful tool for treatment of aquatic invasive plants in locations such as Loomis Lake, Washington, Schutz lake, Minnesota and Withlacoochee River, Florida (Fox et al. 1994; Valley et al. 2006; Parsons et al. 2009).

The ability of *Egeria densa* to rapidly produce apical shoots under low light conditions allows it to outcompete native macrophyte species and form dense surface mats (Mazzeo et al. 2003). By creating a dense surface canopy and homogenizing the water column, vegetation can greatly alter fisheries habitat, oftentimes reducing species diversity by altering feeding habits of fishes and providing refuge for zooplankton (Pelicice et al. 2005; Pelicice and Agostinho 2006). The Delta is home to robust fisheries, waterfowl hunting, and provides habitat for the endangered delta smelt (*Hypomesus transpacificus*). The delta smelt is endemic to the estuary, and was historically abundant, but it's rapid decline prompted it to be listed as federally endangered (Moyle et al. 2016). Changing fisheries trophic status to thick, dense, monotypic vegetated mats may negatively affect the endangered delta smelt by supporting increased predation, through support of intermediate size predators (Sweetnam 1999; Kimmerer 2011; Miller et al. 2012; Nobriga et al. 2013). Supporting fisheries trophic status that favors larger predators that are not as likely to prey on delta smelt may be a strategy to increase smelt populations, while also increasing large game fishes (Carpenter and Lodge 1986; Weaver et al. 1997; Petr 2000). In addition to trophic level influences, thick stands of invasive macrophytes reduce oxygen exchange with the surface, which has shown to have a negative impact on aquatic life (Schultz and Dibble 2012). Furthermore, reduced light penetration due to surface matting of dense submersed vegetation, as commonly occurs with *Egeria densa* in the Delta, reduces phytoplankton and zooplankton presence (Cattaneo et al. 1998), the latter being the primary food source for the delta smelt.

The judicious use of herbicides as part of Integrated Pest Management (IPM) is an effective and environmentally sensitive approach to pest management. This approach relies on strategically employed practices that result in achieving measurable management goals. Successful IPM programs deploy proven methods that are tailored to achieve the most efficient effect on target plant life cycles, developmental and reproductive stages and also take into account the environmental drivers that affect these phenological events. This information, in combination with available pest control methods is used to manage pest damage by the most economical means and with the least possible hazard to people, property, and the environment. Currently the Aquatic Invasive Plant Control Program (AIPCP) is utilizing herbicide control in the Delta, until further tools can be approved for use such as benthic mats and bladders. These methods are contingent upon approval of two Biological Opinions from the United States Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS).

Materials and methods

Study Site

Covering 3,500 acres, Franks Tract is the largest single body of water within the San Joaquin Delta. Originally used for agriculture, Franks Tract was flooded in 1936 and is a popular spot for fishing and waterfowl hunting (California Department Parks and Recreation 2018) In 2006 Franks Tract began to be managed at operational scale utilizing fluridone.

Herbicide treatments

A fluridone pellet formulation was applied annually to achieve in-water concentrations of fluridone between 2.5 and 3.5 ppb for 8 to 16 weeks per-year in areas with dense *Egeria densa*. Fluridone pellets were purchased from SePro Corporation under trade names Sonar One, Sonar PR and Sonar Q. Fluridone applications were started as early as March and continued throughout the treatment period in variable intervals to sustain the target concentrations which were verified by an ELISA analytical test. Treatment areas were determined and maintained for each season based on *Egeria densa* and Curlyleaf pondweed *(Potamogeton crispus)* distribution. Vegetation distribution and density were determined by macrophyte monitoring prior to upcoming cycle of management. Pellets were applied via boat-mounted air blowers to achieve homogenous distribution over *Egeria densa* beds, per the product label.

Macrophyte Monitoring for Species Presence

To assess and document changes in macrophyte species presence and relative abundance, we conducted point intercept surveys (Madsen 1999; Madsen and Wersal 2017) in the fall of each year, with dates ranging from October 7 to December 6, however; most surveys were conducted in the second week of October. Sampling points were chosen by generating a grid of points with a layover of the study area using various GIS programs. Each grid represented a full coverage of all of Frank's Tract at evenly spaced intervals, although the number of points on the grid was variable each year due to logistical ability and time needed to accurately monitor a representative area. All points were used in the analysis regardless of water depth, as this metric was inconsistent due to tide schedule or water abundance.

To sample each point we used a weighted, double-headed, 0.33 m wide rake, which was dragged for ~ 3 m along the bottom and then pulled up to the boat for analysis. We recorded each species of submerged macrophyte that was present on the rake. The frequency of occurrence for each species was calculated as the number of sites with that species present divided by the total number of sample sites for each survey. Frequency of occurence was performed independently for all native and invasive species detected. In addition, the relative frequency of occurrence was calculated for native and non-native plants as 2 separate groups.

To calculate a value for each point representing the number of native or non-native species per point, the number of different species for each respective group was totaled. This tabulation allowed determination of a mean and standard deviation for each survey and provided a metric to complete statistical analysis on species diversity.

Statistical analysis

Statistical analyses were carried out using IBM SPSS Statistics v. 25 and all tests were considered significant if $P < 0.05$. To test between-year changes in plants per point for both native and non-native groups, we compared each year to the previous year using an ANOVA with Tukey HSD post hoc test, we also used the same test to compare beginning year of treatment to the last year of treatment for each group respectively. To assess change in frequency of occurrence for *Egeria densa* and for native and non-native species between years, we used a chi-squared analysis.

Results

Relative frequency of non-native plants decreased over the timeframe from 2006 to 2017 ($P = 0.18$) with variability between years (Figure 1), likely due to differences in area covered with fluridone treatments between years and non-treatment years in 2013 and 2015 (Table 1). The greatest reduction in relative frequency of non-native plants occurred between 2013 and 2014. It should be noted that 2013 was a non-treatment year and that in 2014, 1872 acres were treated, during which time non-native plants decreased from 60.4% to 27.7% ($P < 0.01$; Figure 1). The largest decrease in non-native species frequency of occurrence was that of *Potamogeton crispus* from 56.0% in 2016 to only 4% in in 2017, *Cabomba caroliniana* and *Myriophyllum spicatum* were found in 2006 at 1.5% and 3.1% respectively, but not in any surveys after 2006 (Table 2). The primary non-native management target species, *Egeria densa,* had variable frequency of occurrence throughout 2006 to 2017, despite the overall relative decrease of non-native plants compared to that of native species (Table 2). Relative frequency of occurrence for native plants significantly increased from 2006 to 2017 ($P < 0.001$) and showed significant increases between 2007 thru 2013 ($P < 0.001$), 2013 to 2014 ($P < 0.001$), and 2014 to 2015 ($P < 0.001$) (Figure 1). Compared to non-native species, a general increase in relative frequency of native species was observed, whereas non-natives show the inverse. However, frequency of occurrence of most native species remains variable across years except for

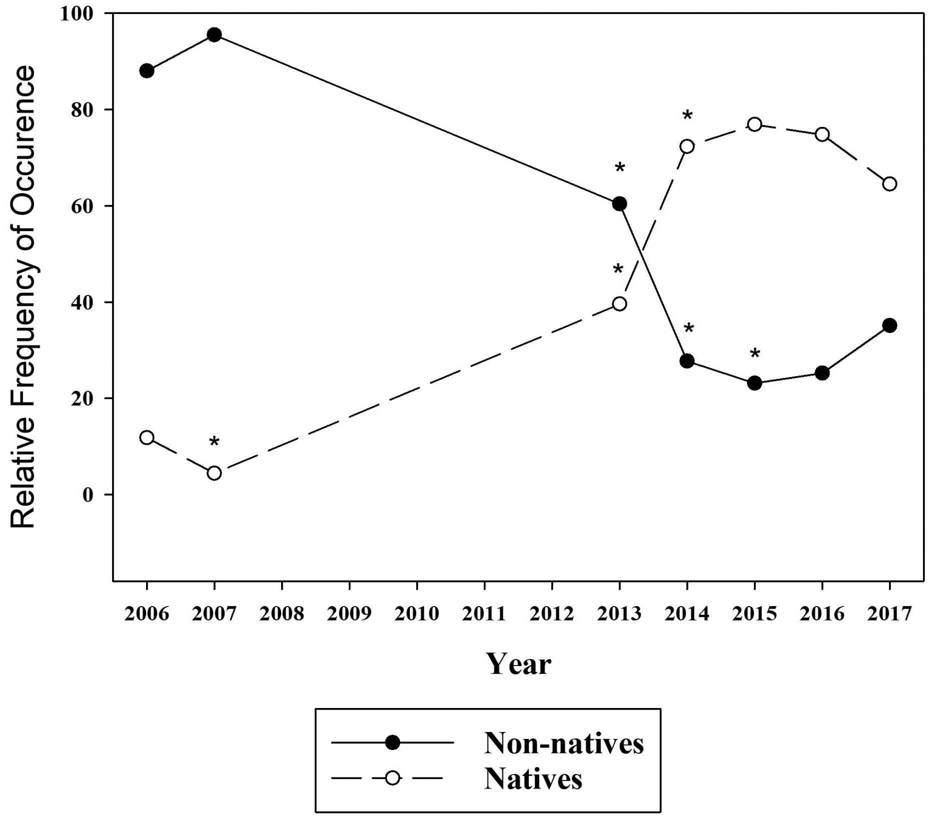

Figure 1. Relative frequency of occurrence of all native and non-native macrophyte species in Frank's Tract for each survey year. (*) indicates significant difference between prior year using Chi-squared analysis. Closed circles, relative frequency of non-native plants; open circles, relative frequency of native plants.

Table 1. Year, month surveyed, survey points per-survey and acres treated with fluridone in Frank's Tract: 2006 to 2017.

Year	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017
Survey Month	December	October	N/A	N/A	N/A	N/A	N/A	October	October	October	October	October
Survey Points	452	702	N/A	N/A	N/A	N/A	N/A	195	100	200	45	100
Area Treated	140	3247	3247	0	500	2413	700	0	1872	0	1040	1097

Table 2. Frequency of occurrence of non-native and native macrophyte species in Frank's Tract by year of survey.

Species	2006	2007	2013	2014	2015	2016	2017
Non-Natives							
Egeria densa	49.6	47.0	77.9	55.0	55.0	49.0	70.0
Potamogeton crispus	2.0	1.4	34.4	25.0	14.5	56.0	4.0
Myriophyllum spicatum	3.1	–	–	–	–	–	–
Cabomba caroliniana	1.5	–	–	–	–	–	–
Natives							
Ceratophyllum demersum	18.1	6.0	37.9	72.0	48.0	42.0	46.0
Potamogeton richardsonii	–	–	3.6	40.0	66.0	53.0	80.0
Najas guadalupensis	–	–	50.8	30.0	55.5	53.0	14.0
Elodea canadensis	–	–	9.2	39.0	21.5	38.0	8.0
Stuckenia filiformis	1.8	8.1	14.9	13.0	18.0	29.0	16.0
Stuckenia pectinata	–	0.7	20.5	12.0	15.0	62.0	5.0
Potamogeton foliosus	–	–	–	–	6.5	51.0	–
Potamogeton nodosus	–	0.3	–	1.0	0.5	4.0	–
Nitella sp.	–	–	–	–	0.5	–	8.0
Potamogeton illinoensis	–	–	–	–	0.5	2.0	–
Native Richness	2	4	6	7	10	9	7

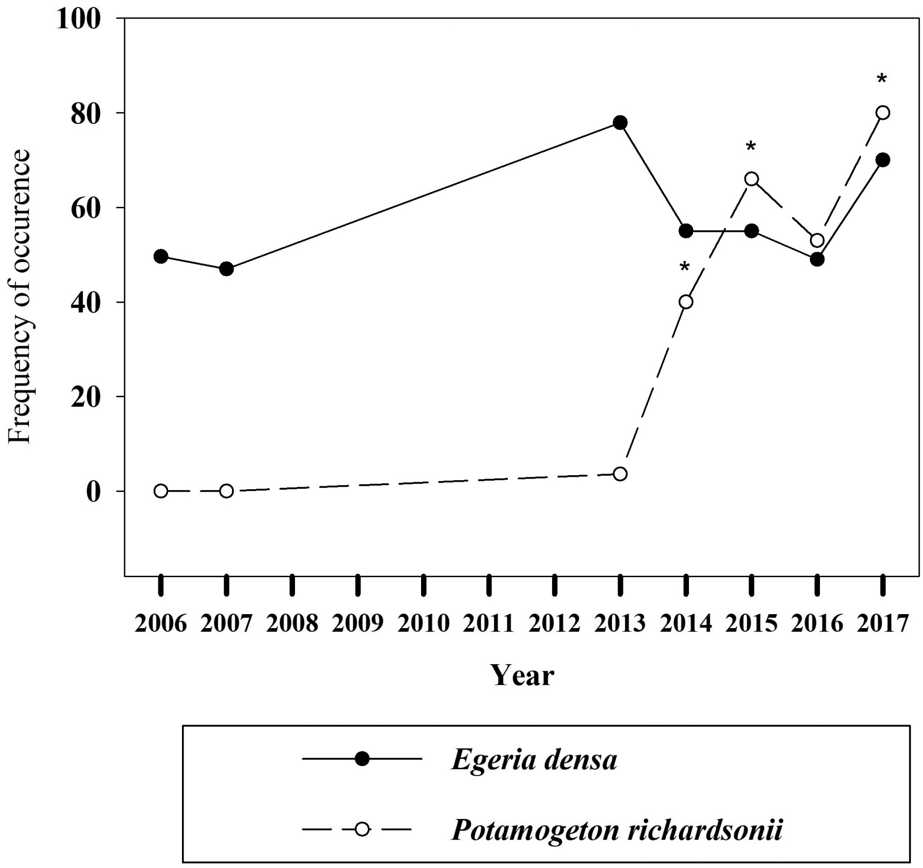

Figure 2. Frequency of occurrence of *Egeria densa* and *Potamogeton richardsonii* in Frank's Tract for each survey year. (*) indicates significant difference between prior year using Chi-squared analysis. Closed circles, frequency of invasive *Egeria densa*; open circles, frequency of the native *Potamogeton richardsonii*.

Potamogeton richardsonii where frequency of occurrence increased greatly from 3.6% in 2013 to 80% in 2017 ($P < 0.001$), and significantly increased each year sequentially except between 2015 to 2016 (Figure 2). The reduction of non-native plant relative frequency initially led to an overall increase in species richness (Table 2) and increase in overall plants per sample point. Prior to 2013, data on plants per sample point were limited and thus per-point data was restricted to 2013 thru 2017. A Tukey HSD showed significant differences in plants per point between years at 0.99 α. Non-native plant species per point significantly decreased between 2013 and 2017 ($P < 0.001$) and varied between sequential years ($P < 0.001$ to 0.127; Figure 3). Conversely, native plant species per point increased every year ($P < 0.001$ to 0.34) except for 2017 where a significant decrease compared to 2016 was observed (Figure 3). This decrease may potentially be due to the increase and dominance of one native species, *Potamogeton richardsonii* which significantly increased every year sequentially from 2013 to 2017 (Figure 2), even during a year of no treatment (2014 *vs*. 2015).

Discussion

The recovery of native species abundance in response to vegetation management is a well-documented phenomenon and has been recorded in

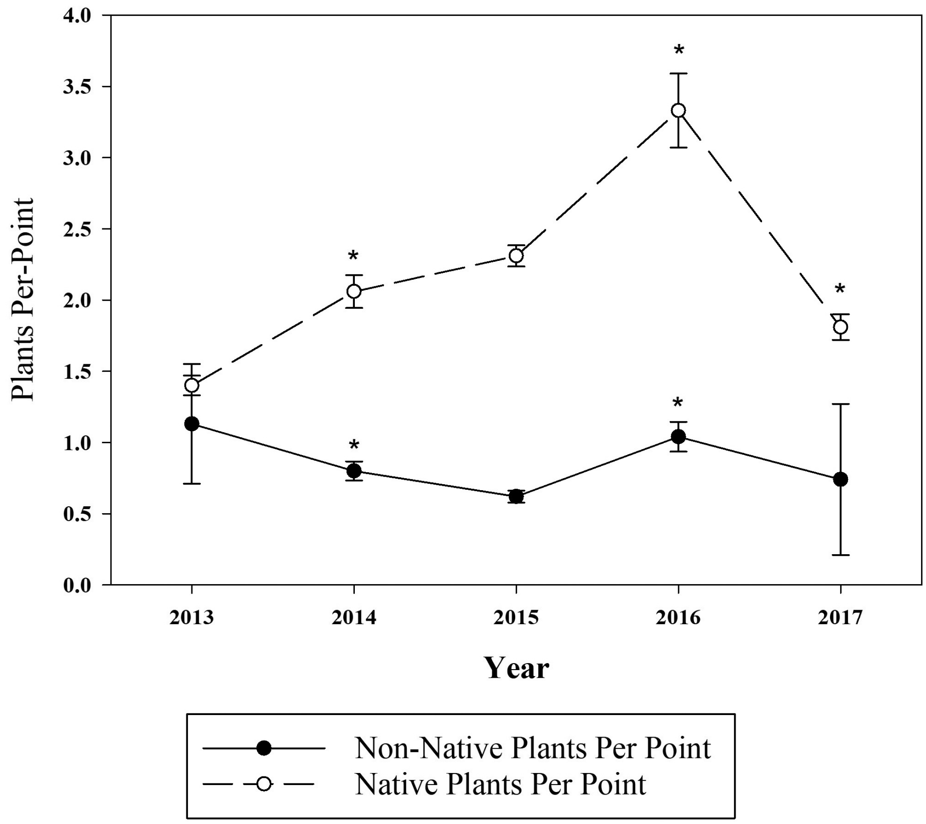

Figure 3. Non-native and native plants per point for each survey year for Frank's Tract where analysis could be conducted with available data. (*) indicates significant difference over the prior year using a ANOVA with Tukey HSD post hoc test. Closed circles, average number of non-native plant species per point; open circles, average number of native plant species per point. Error bars represent standard deviation of the mean.

many studies around the world (Jones et al. 2012; Michelan et al. 2010; Olson and Doherty 2014). Aquatic vegetation management in response to invasive non-native species is an integral part in the protection of native resources. In many cases maintaining the functionality of waterways through large scale vegetation management can also support the proliferation and diversity of native species, as documented in this study. Through the large-scale application of the aquatic herbicide fluridone, California Department of Parks and Recreation, Division of Boating and Waterways has managed *Egeria densa* in Franks Tract which likely allowed for an increase in species diversity since 2007 from mostly non-native to native species. Increasing the number of different native plant species can have several benefits on fisheries (Petr 2000; Valley and Bremigan 2002) compared to monotypic weed beds (Schultz and Dibble 2012) and help promote other native species populations throughout the trophic levels of the ecosystem such as waterfowl (Engelhardt and Ritchie 2001; Maltchik et al. 2010; Kuczyńska-Kippen and Joniak 2016).

When devising an aquatic macrophyte control program, many factors must be considered, such as: target plant physiology and phenology, economic feasibility and optimizing control methodologies. Numerous studies have concluded that aquatic herbicides can be very effective for aquatic plant control (Wersal et al. 2010; Johnson et al. 2012; Hussner et al. 2017). However, important factors govern how effective these treatments can be, including, but not limited to: time of treatment, water chemistry and exposure time (Netherland et al. 2000; Cedergreen et al. 2005). Franks Tract is part of the San Joaquin Delta, a tidal system which is fed by numerous rivers. The dynamic and complex water movement of the Delta can make herbicide exposure time extremely difficult to achieve, especially when consistent concentrations are needed over a lengthy time for efficacy (Netherland et al. 1993). Fortunately, *Egeria densa* is sensitive to low doses of fluridone (Cockreham and Netherland 2000; Tanaka et al. 2001). The application of fluridone in Franks Tract through a slow-release pellet provided persistent low doses of fluridone at sufficient levels to achieve management of *Egeria densa*. The management of *Egeria densa* has led to the prevalence of native species, even though *Egeria densa* fluctuated between 47–78% over the study period. By selectively targeting sensitive invasive species at the onset of vegetative growth, collateral damage to native or desirable species can be avoided while still gaining control over the target non-native species (Sethi et al. 2017). Although some native species like *Elodea canadensis* and *Stuckenia pectinata* are known to be sensitive to low rates of fluridone, the net benefit of *Egeria densa* removal is greater than the potential decline of these species (Poole et al. 2004).

The rationale for relying on fluridone in this project is based on both the mode of action and ability to deliver effective concentrations and exposure. The degree of fluridone efficacy can be species specific depending on concentration and exposure time. Whereas some genera such as *Hydrilla*, *Elodea*, and *Myriophyllum* show very good control at concentrations as low as 5 ppb of fluridone, other genera such as *Potamogeton* and *Vallisneria* require higher rates to produce the same level of control (Netherland et al. 1993, 1997; Netherland and Getsinger 1995). Differences in species specific rates required for macrophyte control can prove beneficial if the target species are sensitive to fluridone. Lower rates needed for macrophyte control can reduce program costs and ultimately reduce the impact on non-target organisms. In many cases, non-target organisms are plants that are native to the local ecosystem and play an important role in ecosystem function. Additionally, supporting native macrophyte growth in response to reducing invasive macrophyte monotypic growth may reduce opportunities for re-establishment of invasive macrophytes when a native species becomes the dominant plant in the ecosystem (Capers et al. 2007; Rodrigues and Thomaz 2010). Capers et al. (2007) also found that reduced invasibility only correlated with plant density of native species and not species richness. These findings may lead one to believe that the statistically significant reduction in number of native species in 2017 compared to 2016 may not indicate a decline in ecosystem health, but the potential aid of *Potamogeton richardsonii* in the competition of *Egeria densa*. *Potamogeton richardsonii* increased during a non-treatment year (2014 *vs*. 2015) as well as during treatment years such as (2013 *vs*. 2014). Additional benefits of *Potamogeton* and other native species include, providing food for waterfowl (Van Donk and Otte 1996; Lauridsen et al. 2003) and the potential for higher fish and other aquatic species diversity compared to a monotypic habitat dominated by non-native macrophytes (Keast 1984). Houston and Duivenvoorden (2002) found that increases of native plant species led to increases of native fishes and reductions of introduced fish species. Incorporating native species competition into an integrated pest management program in concert with a targeted herbicide program provides a more sustainable approach to aquatic resource management than other pest management tools alone. The control actions taken by the AIPCP has shown successful management of the invasive macrophyte *Egeria densa* to allow for the emergence of the native *Potamogeton richardsonii* as the dominant species in Franks Tract.

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