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An operational study of repeated diquat treatments to control submersed flowering rush

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INTRODUCTION

Flowering rush (*Butomus umbellatus* L.) was first introduced to eastern North America from its native Eurasia over 100 yr ago (Core 1941). Additional introductions of unique genetic strains, likely as garden ornamentals that escaped to natural areas, led to its establishment in the western United States (J. Gaskin, unpub. data). The first flowering rush record in western North America was from the Snake River, Idaho in 1949 (Anderson et al. 1974).

Flowering rush is able to exploit a wide range of habitats, making it an impressive invasive plant. In the northwestern United States, it grows as an emergent along shorelines, graduating out to water depths of more than 6 m where it can grow completely submersed. It can grow in all conditions, ranging from still water with muddy substrate to flowing water with rocky substrate. Flowering rush thrives in areas with fluctuating water levels, but also persists and spreads in stable water conditions (Hroudová 1989, Hroudová et al. 1996). It invades and dominates native plant beds (Madsen et al. 2012) and can also colonize habitats previously barren of plant growth (Parkinson et al. 2010). Flowering rush has been termed an ecosystem engineer for its ability to alter habitat by sediment accretion (Gunderson et al. 2016). These characteristics, along with the rapidly expanding population, have raised concern about potential impacts on habitat and irrigation water delivery if flowering rush becomes well-established in northwestern North America.

Flowering rush has both diploid and triploid cytotypes that vary in their reproductive strategies (Hroudová et al. 1996). Most of the flowering rush in Washington State is triploid, including at the study site, Silver Lake (Poovey et al. 2012, J. Gaskin, unpub. data). As such, it is expected to rarely, if ever, produce viable seed (Hroudová et al. 1996, Lui et al. 2005). Instead, it spreads locally by rhizomes and disperses long distances by natural and anthropogenic movement of rhizome fragments and buds, and less commonly from bulbils in the inflorescence (Hroudová et al. 1996).

Flowering rush was first recorded from Silver Lake, Washington in 1997 (Parsons 1998). In 2009 flowering rush achieved a Class A weed designation on the Washington Noxious Weed List, making it a high priority for control and eradication. At that time the Silver Lake population was thought to be one of two isolated flowering rush locations in the state, leading to a desire for control. In 2008 to 2009 small-plot herbicide trials evaluated imazapyr, triclopyr, and glyphosate for management of emergent flowering rush, finding that imazapyr was most effective at reducing plant cover. All products required emergent leaves to extend at least 0.6 m above the water to be effective, and none provided 100% control (T. W. Miller, Faculty, Washington State University Extension, pers. comm.). Therefore, repeated treatments would be required. Emergent growth was never treated on a large scale in Silver Lake because most of the flowering rush grew in deep water.

In 2011, submersed plants were targeted with a trial of three herbicides: granular triclopyr, and a granular triclopyr/2,4-D amine combination in 4-ha plots each; and liquid imazamox in a 3-ha plot. None of the treatments resulted in a reduction of flowering rush biomass or frequency of occurrence in the year of treatment. Herbicide concentration and exposure time data indicated herbicide contact with the plants was insufficient to provide control (J. K. Parsons, unpub. data).

Meanwhile, results had shown the most promising short-term submersed flowering rush control was achieved with contact herbicides (Madsen et al. 2012, Poovey et al. 2012). These herbicides typically are fast-acting but only kill that part of the plant they come into contact with. However, some plant species are more sensitive than others, and repeated use can reduce long term viability of sensitive species. Based off the research of Madsen et al. (2012), diquat was applied in a 4-ha treatment area in Silver Lake. The success of that treatment in controlling aboveground growth led to subsequent treatments in the same area over the course of an additional 4 yr. This is a report on monitoring data collected as part of this operational project.

MATERIALS AND METHODS

Silver Lake is a 73-ha lake, with a maximum depth of 9.1 m and a mean depth of 5.2 m (Bortleson et al. 1976). The water is generally clear, with secchi depth readings averaging 6 m during the study. It is located in the Cascade foothills about 30 mi (48.3 km) northeast of the town of Bellingham, Washington and just south of the Canadian border. It is long and narrow in a north-to-south direction, with small inflows to the north half and a slowly-flowing ungauged outlet through a wetland at the south. The shoreline is moderately developed with parks, organization camps, and single family homes. It is stocked with rainbow trout to provide seasonal fishing opportunities.

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When flowering rush was first noted in Silver Lake in 1997, it was already well-established at the north end, occupying a wide zone from shallow water with mostly emergent plants to deep fully submersed plants. It is not known when flowering rush was introduced to Silver Lake; however, Hatzic Lake, British Columbia, located just north of Silver Lake, had flowering rush established since at least 1978 (Consortium of Pacific Northwest Herbaria 2017). It is possible the two populations were introduced in the respective lakes about the same time because triploid flowering rush was available in the horticulture trade (Lui et al. 2005), and travel between British Columbia and this part of Washington State is common.

This project began in 2012 with treatment of one 4-ha plot with the contact herbicide diquat[1] applied at the maximum label rate (370 parts per billion [ppb]) on 4 June and again on 14 August. Weighted trailing hoses were used to apply the herbicide about 1 m below the water surface. Due to success of the 2012 diquat treatment, this treatment regime was repeated in 2013 on 2 July and 26 September. In 2014, 2015, and 2016, diquat treatments occurred once per summer (3 September, 10 August, and 26 July, respectively). All treatments were applied to the same 4-ha plot at the same herbicide rate. Variations in treatment timing were due to scheduling with the herbicide applicator and county personnel.

Following the June 2012 treatment, water samples were collected for diquat concentration analysis from three points in the 4-ha treatment plot and one point at the south end of the lake (approximately 700 m from the treatment plot). Samples were collected from within the plant bed approximately 1 m off the lake bottom using a Kemmerer sampler. Samples were collected approximately 1, 6, 9, and 24 h after treatment (HAT) in the treatment plot, and 6, 9, and 24 HAT at the southern point. Following the second herbicide application in August 2012, the location and frequency of water samples were reduced to one point in the treatment plot and one point at the southern end. Samples were collected 1, 6, 24, and 48 HAT in the treatment plot, and 3, 6, 24, and 48 HAT at the southern point. Sample analysis was performed by a contracted lab using EPA method 549.2. The practical quantitation limit was 2 ppb, and the method detection limit was 0.3 ppb.

In 2012, aquatic plant frequency of occurrence and biomass samples were collected in May, prior to the initial diquat treatment, and again at the end of August following the two diquat treatments. In 2013 through 2017, plant sampling was repeated in May or June, prior to herbicide treatment for that year. The end-of-season plant sampling was not conducted in those years.

Plant frequency-of-occurrence data were collected following Madsen and Wersal (2012) at points less than 8 m deep, which was the maximum depth of plant growth. Points were located in the field using a Trimble Geo XT[2] Global Positioning System (GPS) unit. In the treatment plot the point density was a 25-m grid; for the rest of the littoral zone a 50-m grid was used. The number of samples collected varied slightly year-to-year due to fluctuating water levels. At each point a sampling-rake (two metal bow rakes bolted back to back with handles replaced by a marked rope) was dropped to the sediment from a boat, and all plant and macroalgae species collected were recorded, along with the depth. In shallow water areas where plants could be clearly seen, plant species were recorded by visual observation in an approximately 1 m^2 area.

Scuba divers collected plant biomass following Madsen and Wersal (2012) from a random subset of 20 plant frequency points at depths less than 4 m in the treatment plot in all years except 2015. In 2015 the divers were unavailable. Sample points were located by GPS and marked by the boat's crew with a weighted float. The divers then placed a 0.1 m^2 sample frame on a predetermined side of the weight. All plant material, including roots and rhizomes, within the sample frame was collected and placed in a mesh bag. The bag and plants were taken to the boat where the support crew transferred the plants to labeled plastic bags. Plant samples were later cleaned of mud and debris by washing with pressurized water. The clean plants were sorted by species, placed into preweighed and -labeled paper bags, dried to a constant weight at 70 C, and weighed. Flowering rush root/rhizome (below ground) biomass was dried separately from leaf (above ground) biomass.

Plant frequency data were analyzed using chi square analysis with a significance level of $P < 0.05$. Plant biomass data were log-transformed and analyzed using ANOVA with a post hoc Tukey's Honestly Significant Difference Test on significantly different comparisons ($P < 0.05$). SYSTAT®[3] was used for all analyses.

RESULTS AND DISCUSSION

Herbicide concentration

Diquat concentrations for June 2012 in the treatment plot averaged 94.8 ppb 1 HAT, 99.5 ppb 6 HAT, 36.3 ppb 9 HAT, and 24.9 ppb 24 HAT. The concentration at the southern sample point was 0.6 ppb 6 HAT and not detected 9 or 24 HAT. In August 2012, diquat concentration in the treatment plot was 72.1 ppb 1 HAT, then dropped to 0.5 ppb 6 HAT, 3.9 ppb 24 HAT, and 2.2 ppb 48 HAT. At the southern sample point diquat was not detected until 48 HAT at 2 ppb.

Diquat did not reach the target concentration (370 ppb) in the treatment plot either time samples were collected. However, this was expected due to dilution and mixing because only a relatively small portion of the lake was treated. Weather conditions in August were calm at the time of treatment; however heavy southerly winds were present the day following treatment. Thus, wind mixing could have led to the lower treatment plot herbicide concentrations compared with June concentrations. During both treatments, diquat reached the southern sample point, although at very low concentrations.

Plant data

Plants grow to a depth of about 8 m in Silver Lake, with mainly a patchy growth of the macroalgae stonewort (*Nitella* Agardh spp.) at depths of 6 m or greater. Flowering rush abundance was greatest between about 2 to 3 m deep where

Table 1. Average biomass (g dry weight [DW] m^{-2}) in the treatment plot (n = 20), with standard deviation in parentheses. An * indicates the value is significantly different from pretreatment (May 2012) by ANOVA and post hoc test ($P < 0.05$).

	Average biomass (g DW m^{-2})					
	May 2012	August 2012	May 2013	May 2014	May 2016	May 2017
Flowering rush leaves	49.97 (68.1)	2.07 (3.3)*	2.15 (5.6)*	0.62 (2.6)*	0.11 (0.5)*	0.00 (0)*
Flowering rush roots, rhizomes	117.58 (162.8)	134.31 (239.0)	47.35 (106.0)	34.97 (87.7)	1.93 (8.1)*	0.31 (1.4)*
Macroalgae	11.35 (32.2)	31.02 (55.7)	72.34 (198.2)	25.21 (50.7)	4.28 (10.2)	2.98 (9.1)
Big-leaf pondweed	0.07 (0.3)	0.51 (2.3)	0	3.88 (10.5)	1.27 (3.9)	3.62 (9.3)
Thin-leaf pondweed	0	0.04 (0.2)	0	0	1.84 (5.6)	0.31 (0.5)
White-stem pondweed	0	3.38 (10.8)	0	2.50 (8.4)	0	0

it grew submersed or with a small amount of leaf emerging from the water. The maximum depth we recorded flowering rush was 6.5 m.

Flowering rush leaf biomass data from before treatment (May) and 8 d after the second diquat treatment (August) in 2012 showed a significant decrease in the treatment plot (Table 1). Other studies also have found that contact herbicides reduce flowering rush leaf biomass within a few weeks of treatment (Poovey et al. 2012, Madsen et al. 2016b). Flowering rush root/rhizome biomass did not change significantly between May and August 2012 (Table 1). In other studies, variable rhizome biomass results were obtained from contact herbicide in the year of treatment (Poovey et al. 2012, Madsen et al. 2016a,b). This could be because of high variability in the samples, sample timing, a difference in the rate of rhizome decomposition, or whether or not decomposing rhizomes were included in the sample. We included all rhizome material in our samples; thus, a change in mass would not be expected until dead rhizomes were decomposed. Flowering rush frequency of occurrence was not reduced between May and August 2012 in either the treatment plot or the rest of the lake (Tables 2 and 3).

Biomass of none of the other plant species changed significantly in the treatment plot between May and August 2012 (Table 1). Without management, one would expect plant biomass to increase between May and August because most submersed plants are at their peak biomass in August in western Washington. Thus, although plant biomass did not decrease significantly, the fact that it also did not increase significantly was likely due to the herbicide treatments. Frequency of occurrence of one species, American waterweed (*Elodea canadensis* Rich.), was reduced significantly in August 2012 when looking at the whole-lake data (Table 3).

In subsequent annual samples (May 2013 through May 2017), there was a significant reduction in treatment plot flowering rush leaf biomass for all years compared with the pretreatment sampling (May 2012). By 2016 and 2017 the reduction was seen in root/rhizome biomass as well (Table 1). We also found a significant reduction in flowering rush frequency in the treatment plot in 2014, 2016, and 2017 (Table 2). In 2015, data were collected in mid-June, so flowering rush growth was more advanced than in other years (the longer leaves being more easily caught by the sampling rake). The reduction in rhizome biomass would indicate that diquat's ability to reduce leaf biomass is weakening overall plant health, leading to rhizome biomass reductions that persist into the year following treatment. Treatment plot biomass and frequency continued to decline after treatments were reduced from twice to once per year (reflected in 2016 and 2017 data) (Tables 1 and 2). This corroborates results of a diquat trial on flowering rush in Minnesota lakes which similarly showed that diquat reduced flowering rush growth (Madsen et al. 2016a,b).

Biomass of the other plant species in the treatment plot did not change significantly in any year between 2013 and 2017 when compared with May 2012 (Table 1). Treatment plot frequency of occurrence changed significantly for macroalgae (a mix of muskwort [*Chara* Val. spp.] and stonewort) (Table 2). The macroalgae were collected more frequently in May 2013 and June 2015 than prior to treatments (May 2012). Macroalgae are unaffected by diquat, so would be expected to expand to colonize areas where flowering rush was declining (Clayton and Tanner 1988, Kelly et al. 2012). The reason macroalgae did not consistently increase could be due to competition from other plants colonizing those areas. Other native species were collected too infrequently to show a significant change.

Table 2. Treatment plot percent frequency of occurrence. An * indicates a significant difference from May 2012 at $P \leq 0.05$. Only species present at more than 5% of points on more than one date are presented.

	% Points where plant present						
	May 2012 (n = 62)	August 2012 (n = 64)	May 2013 (n = 64)	May 2014 (n = 64)	June 2015 (n = 64)	May 2016 (n = 63)	May 2017 (n = 64)
Flowering rush	16	20	9	5*	6	3*	2*
Macroalgae	24	22	42*	39	48*	35	38
Big-leaf pondweed	2	2	3	8	3	3	9
Thin-leaf pondweed	0	2	0	6	9	8	8
White-stem pondweed	8	3	5	0	6	0	0
No plants	60	59	52	61	45	60	61

Table 3. Whole lake (except treatment plot) percent frequency of occurrence for plants found in at least 1% of sample points for at least two sample dates, an * indicates a significant difference from May 2012 at $P \leq 0.05$.

	% Points where plant present						
	May 2012 (n = 190)	August 2012 (n = 188)	May 2013 (n = 189)	May 2014 (n = 193)	June 2015 (n = 190)	May 2016 (n = 188)	May 2017 (n = 188)
Flowering rush	15	16	9	9	12	10	5*
Macroalgae	44	49	62*	45	44	38	34*
American waterweed	7	2*	1*	1*	4	6	4
Water marigold	4	5	2	1	4	2	2
Big-leaf pondweed	3	1	4	6	8*	7*	12*
White-stem pondweed	1	3	1	1	2	1	1
Thin-leaf pondweed	0	0	0	0	3	17*	27*
No plants	38	35	31	45	44	46	47

Pondweed species (*Potamogeton* L. spp.) have been found to be fairly susceptible to diquat in other studies (Skogerboe et al. 2006, Mudge 2013). However, because they did not disappear from our treatment plot, the pondweed species present (white-stem pondweed (*Potamogeton praelongus* Wulf.), thin-leaf pondweed (*Potamogeton pusillus* L.) and big-leaf pondweed (*Potamogeton amplifolius* Tucker.) can tolerate low diquat concentrations.

In the rest of the lake (Table 3), flowering rush frequency was reduced by the last year of the study. This might have been caused by a combination of a cold winter and spring in 2017, causing later than normal flowering rush growth, and out-of-plot impacts from diquat drifting out of the treatment plot in low concentrations. Frequency of American waterweed was significantly reduced in 2013 and 2014, then it appears to have recovered after diquat treatments were reduced to once per year (reflected in 2015 through 2017 frequency data). American waterweed is susceptible to diquat at low concentrations (Glomski et al. 2005), but treating one relatively small area once per year allowed it to persist. Big-leaf pondweed and thin-leaf pondweed frequencies increased in the later years of the study. Direct observation showed that these plants were filling in areas that had been previously dominated by flowering rush. The frequency of points where no plants were collected did not change significantly over the treatment period. This would indicate that the repeated diquat treatments are not preventing native plants from recolonizing the flowering rush-dominated areas. Two species did not change significantly over the course of the study, water marigold (*Bidens beckii* Torr. ex Spreng.) and white-stem pondweed.

Timing of herbicide treatment varied in this study, with the late summer treatments occurring between late July and late September. Marko et al. (2015) found that flowering rush rhizome starch content was lower in late summer/early fall, indicating this would be the optimal time to apply herbicides due to reduced energy reserves to fuel recovery. Future treatments should focus on late summer treatments to take advantage of this.

In conclusion, this study showed that repeated diquat treatments over the course of 5 yr reduced flowering rush biomass and frequency in the treatment area, while allowing native species to recolonize that area.

SOURCES OF MATERIALS

[1]Diquat bromide (the following brand names were used, all with 37.3% active ingredient): Reward® and Tribune™, Syngenta Crop Protection, Inc., P.O. Box 18300, Greensboro, NC 27409; Littora®, SePRO Corp., 11550 No. Meridian St., Suite 600, Carmel, IN 46032.

[2]Trimble Geo XT 2005 Series, Trimble Inc., 345 SW Avery Ave., Corvallis, OR 97333

[3]SYSTAT Version 13.1 statistical software, Systat Software Inc., 2107 No. First St., Suite 360, San Jose, CA 95131.

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