# Note

# Control of submersed flowering rush with contact and systemic aquatic herbicides under experimental conditions

ANGELA G. POOVEY, C. R. MUDGE, K. D. GETSINGER, AND H. SEDIVY\*

# INTRODUCTION

Flowering rush (Butomus umbellatus L.) grows both as an emergent plant along shorelines and as a submersed plant in deeper water of northern lakes and rivers (Countryman 1970). Native to Eurasia, it is an aggressive colonizer that can form dense monospecific stands, interfering with intended water uses and crowding out native plants (Countryman 1970). Ploidy has been strongly correlated with reproductive capacity of flowering rush (Lui et al. 2005). Diploid plants reproduce sexually through seed production, while triploid plants reproduce clonally through rhizome lateral branching. The production of bulbils, a vegetative reproductive structure, has been associated with diploid rather than triploid plants (Lui et al. 2005). According to Eckert et al. (2003), diploid populations are common in the eastern Great Lakes region and triploid populations are sparsely, but widely distributed across North America. For example, plants collected from Minnesota and Idaho lakes have been documented to be triploid, with no genetic differentiation among populations (Lui et al. 2005, Poovey et al. 2012).

Although the management of diploid flowering rush using aquatic herbicides has yet to be investigated, control of triploid flowering rush has been the focus of recent research efforts. Submersed applications of contact herbicides using short exposure times were found to be effective in controlling submersed plants (Poovey et al. 2012). Foliar applications of systemic herbicides have been tested in field sites for controlling emergent plants (Rice et al. 2009). To prevent new growth sprouting from rhizomes, bareground applications on dewatered sediment have been investigated in a mesocosm system (Woolf et al. 2011).

Submersed applications with systemic herbicides alone and in combination with contact herbicides have yet to be evaluated. Systemic herbicides and combinations of systemic and contact herbicides could augment control of flowering rush, with the potential to reduce roots and rhizomes as well as lateral root buds (Poovey et al. 2012). Chemical control strategies must also take into account potential herbicide damage to valuable non-target native plants that may be established adjacent to, or within, stands of flowering rush. Species-selective control of flowering rush, using any management approach, will be required to protect aquatic habitats (particularly with respect to threatened and endangered species) and community biodiversity.

Systemic herbicides can be either broad spectrum or highly specific in activity. Selectivity is achieved through the choice of herbicide, use rate, and timing of application (Poovey and Getsinger 2005). Two systemic herbicides commonly used for aquatic weed management are triclopyr ([(3,5,6-trichloro-2-pyridinyl)oxy]acetic acid) and 2,4-D ([2,4-dichlorophenoxy]acetic acid), which are synthetic auxins. Synthetic auxin herbicides are chemicals that act similarly to the plant hormone indole-3-acetic acid (IAA), and uptake of these herbicides leads to uncontrolled cell division and growth, which results in vascular tissue destruction (WSSA 2007). The maximum rate for submersed applications of triclopyr is 2.5 mg acid equivalent (ae)  $L^{-1}$ and 4.0 mg ae  $L^{-1}$  for 2,4-D (WSSA 2007). In addition, there are two formulations of 2,4-D available for aquatic weed control: the amine formulation, which is a liquid, and the low-volatile butoxyethyl ester formulation, which is a granular. Only the amine formulation of triclopyr is registered for aquatic sites. Triclopyr and 2,4-D require 1 to 3 d for effective control of susceptible submersed species (Green and Westerdahl 1990, Netherland and Getsinger 1992), and regrowth can occur in 4 to 8 wk following initial application. Broad leaf plants (dicots) are more susceptible to synthetic auxins than narrow leaf plants (monocots), such as flowering rush; however, herbicide activity on monocot aquatic plants has been reported (Belgers et al. 2007).

Other systemic herbicides interrupt biosynthetic pathways by blocking the production of specific plant enzymes. Two examples are herbicides with modes of actions that inhibit production of phytoene desaturase (PDS) and acetolactate synthase (ALS). The PDS enzyme is needed for carotene biosynthesis, and carotene pigments are essential for plants to photosynthesize (Bartels and Watson

<sup>\*</sup>First, second and third authors: Research Biologist, U.S. Army Engineer Research and Development Center, 3909 Halls Ferry Rd, Vicksburg, MS 39180. Fourth author: Weed Coordinator, Sanders County, 1111 Main St, Thompson Falls, MT 59873. Corresponding author: Angela.G.Poovey@usace.army.mil. Received for publication July 19, 2012 and in revised form November 2, 2012.

TABLE 1. CONCENTRATIONS AND EXPOSURE TIMES OF CONTACT AND SYSTEMIC AQUATIC HERBICIDES EVALUATED AGAINST FIELD-COLLECTED MINNESOTA AND IDAHO FLOWERING RUSH IN EXPERIMENT 1.

Herbicide treatment	Type of herbicide	Concentration	Exposure time (h)
Endothall	Contact	$1.5 \text{ mg ai } \text{L}^{-1}$	24
Flumioxazin	Contact	$0.4 \text{ mg} \text{ ai } \text{L}^{-1}$	24
2,4-D amine	Systemic	4.0 mg ae $L^{-1}$	24
2,4-D ester	Systemic	4.0 mg ae $L^{-1}$	24
Triclopyr	Systemic	1.25, 2.5 mg ae $L^{-1}$	24, 48
Triclopyr + endothall	Systemic + contact	1.25 mg ae $L^{-1}$ triclopyr	24
		$1.5 \text{ mg ai } \text{L}^{-1}$ endothall	
Triclopyr + flumioxazin	Systemic + contact	1.25 mg ae $L^{-1}$ triclopyr	24
1 /	,	$0.4 \text{ mg ai } \text{L}^{-1} \text{ flumioxazin}$	
Triclopyr $+$ 2,4-D amine	Systemic + systemic	1.25 mg ae $L^{-1}$ triclopyr	24
.,		4.0 mg ae $L^{-1}$ 2,4-D	
Reference		0	24

1978). The ALS enzyme is needed for the biosynthesis of branched-chain amino acids (isoleucine, valine, and leucine), which are protein building blocks and integral to plant growth (Tranel and Wright 2002).

Both PDS and ALS chemistries require long exposure times to be efficacious on susceptible plants. The PDSinhibitor fluridone (1-methyl-3-phenyl-5-[3-(trifluoromethyl)phenyl]-4(1H)-pyridinone) is effective with exposures of 45 to 90 days (Netherland and Getsinger 1993). Two ALSinhibitors, imazamox (2-[4,5-dihydro-4-methyl-4-(1-methylethyl)-5-oxo-1H-imidazol-2-yl]-5-(methoxymethyl)-3-pyridinecarboxylic acid) and bispyribac-sodium (sodium 2,6bis[(4,6-dimethoxy-2-pyrimidinyl)oxy]benzoate), are effective with exposure times ranging from 30 to 100 d (Koschnick et al. 2007). Fluridone is a broad-spectrum herbicide, but can be used to selectively control target weeds with low application rates (Netherland et al. 1997, Getsinger et al. 2002). Conversely, imazamox and bispyribac-sodium (hereafter bispyribac) are highly specific for certain emergent and submersed aquatic macrophyte species (Getsinger et al. 1994, Chiconela et al. 2004, Koschnick et al. 2007, Glomski and Netherland 2008).

In order to develop field guidance for species-selective chemical control of flowering rush, we evaluated several aquatic herbicides. In one experiment, we used triploid plant populations from Minnesota and Idaho to evaluate the effects of synthetic auxin herbicides and combinations of these herbicides with contact herbicides on submersed flowering rush. In a second experiment, we determined susceptibility of Minnesota and Idaho populations of flowering rush to PDS and ALS chemistries in a static exposure.

#### MATERIALS AND METHODS

# **Experiment 1**

This experiment was conducted in a large controlled environment growth chamber (48 m<sup>2</sup>) at the US Army Engineer Research and Development Center (ERDC) in Vicksburg, MS. Ambient conditions were set to provide optimum growth for submersed plants air temperature of  $21 \pm 2$  C (75  $\pm$  5 F), light intensity of 700 µmol m<sup>-2</sup> sec<sup>-1</sup>, and photoperiod of 14 h : 10 h light : dark cycle. Flowering rush rhizomes were field-collected from Detroit Lake, Pelican River Watershed, MN, and Lake Pend Oreille, ID, and shipped overnight to ERDC. Site descriptions of these lakes are provided in Poovey et al. (2012). Rhizomes were surrounded by sediment and subjected to a cold (4 C) dark treatment for at least 3 wk before sprouting. Rhizomes 4 to 5 cm (1.6 to 2 in) in length, 4 to 5 g (0.14 to 0.18 oz FW) were then washed to remove sediment, placed in culture solution (Smart and Barko 1985) that was aerated, and allowed to sprout in the environmental growth chamber for 3 wk.

Two different sediments were used for propagating sprouted rhizomes from each plant population based on observations from the field. Minnesota plants were planted in silty topsoil<sup>1</sup>. The pH of the potting soil (5.3) was raised to 6.8 by adding 150 mg L<sup>-1</sup> of calcium carbonate and 1.5 g L<sup>-1</sup> of sodium bicarbonate. The Idaho plants were planted in topsoil<sup>2</sup> that had remnants of bark, which was more acidic (pH = 5.2). Both sediments were fertilized with 150 mg L<sup>-1</sup> ammonium chloride.

Each sprouted Minnesota rhizome containing one shoot  $(7.7 \pm 1.0 \text{ g FW}, \text{shoot length} = 33 \pm 2.4 \text{ cm}, \text{root length} = 20 \pm 1.3 \text{ cm}; n = 20)$  was planted to a depth of 4 cm in a 1-L high-density polyethylene (HDPE) beaker filled with unfertilized topsoil. Each sprouted Idaho rhizome containing one shoot  $(11 \pm 1.0 \text{ g FW}, \text{ shoot length} = 34 \pm 2.9 \text{ cm}, \text{ root length} = 11 \pm 0.9 \text{ cm}; n = 20)$  was planted to a depth of 4 cm in a 1-L HDPE beaker filled with topsoil. A 2-cm layer of masonry sand was added to the sediment surface in each beaker to prevent dispersion of nutrients and sediment into the water column. Two beakers of each plant population were placed in each 48-L aquarium filled with culture solution (Smart and Barko 1985) amended with 0.1 mg L<sup>-1</sup> chelated iron. Plants grew for 3 wk prior to herbicide application.

Herbicide concentrations evaluated were selected based on several factors. Since aqueous exposure times in the Detroit Lake and Lake Pend Oreille can be short (< 24 hr), high concentrations of each product, except endothall, were used. A low rate of endothall was chosen based on a previous experiment in which 1.5 mg ai L<sup>-1</sup> for a 24 h exposure provided > 75% control (Poovey et al. 2012). An exposure period of 24 h was used for all treatments (Table 1). In addition, an exposure period of 48 h was used for triclopyr. Using high concentrations with exposure periods of 24 to 48 h can determine efficacy of synthetic auxin herbicides against submersed plants in small-scale experiments (Green and Westerdahl 1990, Netherland and Getsinger 1992).

Stock solutions of endothall<sup>3</sup>, flumioxazin<sup>4</sup> (Clipper<sup>®</sup>, Valent USA Corp.), 2,4-D amine<sup>5</sup>, and triclopyr<sup>6</sup> were prepared by diluting formulation concentrates in distilled water. A special liquid 2,4-D ester formulation was provided by NuFarm Americas Inc.<sup>7</sup> for this experiment; it also was diluted in distilled water for stock preparation. From the stock, each herbicide was applied subsurface using a pipette to provide nominal concentrations in the treatment aquaria for the appropriate exposure time (Table 1). Untreated reference aquaria were included to assess plant growth in the absence of herbicide exposure. Immediately following herbicide exposure times, all aquaria, including references, were drained and filled with fresh culture solution two times to remove all aqueous herbicide residues. The experiment was concluded 6 wk after treatment (WAT) to allow for plants to potentially recover from treatments.

Water temperature was measured continuously with an Optic Stowaway<sup>®</sup> Temperature Probe<sup>8</sup> in reference aquaria, which were  $21 \pm 0.02$  C during the experiment. The pH was measured in each aquaria at time of herbicide application with a handheld multi-parameter probe<sup>9</sup>. The pH was 8.6.

Herbicide efficacy was assessed by measuring shoot, root, and rhizome biomass. A pretreatment biomass assessment was conducted by randomly selecting four aquaria and harvesting all biomass for each plant population. Biomass was dried and weighed for a dry weight measurement (g DW). At 6 WAT, biomass from one beaker in each aquarium was harvested, dried, and weighed. A growth recovery assessment was also included; shoots were clipped at the sediment surface from one beaker, which was placed back in the aquarium for an additional 2 wk to monitor re-growth. Afterwards, biomass was harvested, dried, and weighed.

Treatments were randomly assigned to individual aquaria and replicated four times, including the reference. All shoot, root, and rhizome data were analyzed using one-way analysis of variance (ANOVA) to determine herbicide effects. If effects were significant ( $P \le 0.05$ ), means were compared using Fisher's Least Significant Difference test (LSD).

# **Experiment 2**

This experiment was conducted in the controlled environment growth chamber under conditions described above. Light intensity was 594  $\mu$ mol m<sup>-2</sup> sec<sup>-1</sup> in Experiment 2. Water temperatures and pH were similar to Experiment 1.

Flowering rush rhizomes were field-collected from Detroit Lake, Pelican River Watershed, MN, and Lake Pend Oreille, ID, and shipped overnight to ERDC. They were subjected to a cold dark treatment and sprouted for 5 wk as described in the previous experiment.

Sediment preparation and plant propagation followed the same procedures as Experiment 1. One sprouted Minnesota rhizome containing one shoot  $(5.1 \pm 0.8 \text{ g FW}, \text{shoot length} = 24 \pm 2 \text{ cm}, \text{ root length} = 5.3 \pm 1.5 \text{ cm}; n = 15)$ 

TABLE 2. CONCENTRATIONS OF PLANT-ENZYME SPECIFIC AQUATIC HERBICIDES
evaluated against field-collected Minnesota and Idaho flowering rush in
Experiment 2. Plants were exposed to herbicides for 5 wk.

Herbicide	Concentration ( $\mu$ g ai L <sup>-1</sup> )	
Fluridone	10, 20	
Bispyribac	20, 40	
Imazamox	50, 100	
Reference	0	

and one sprouted Idaho rhizome containing one shoot (5.9  $\pm$  0.6 g FW, shoot length = 23  $\pm$  1.5 cm, root length = 9.3  $\pm$  0.6 cm; n = 15) were planted in 1 L HDPE beakers filled with topsoil. One beaker of each plant population was placed in each aquarium (volume = 48 L) filled with culture solution (Smart and Barko 1985) amended with chelated iron (0.1 mg L<sup>-1</sup>). Plants were allowed to established for 5 wk.

Herbicide concentrations were selected based on current operational use patterns for ALS and PDS herbicides against submersed plants: low use rates (< 100  $\mu$ g ai L<sup>-1</sup>) with long-term aqueous exposures of 4 to 12 wk. Stock solutions of fluridone<sup>10</sup>, bispyribac<sup>11</sup>, and imazamox<sup>12</sup> were prepared by diluting formulation concentrates in distilled water. Each herbicide was applied subsurface using a pipette to provide nominal concentrations in the treatment aquaria for a static exposure of 5 wk (Table 2). Untreated reference aquaria were included to assess plant growth in the absence of herbicide exposure. After 5 wk, the experiment was concluded.

Herbicide efficacy was assessed by measuring shoot, root, and rhizome biomass. A pretreatment biomass assessment was conducted by randomly selecting three aquaria and harvesting all biomass for each plant population. Biomass was dried and weighed for a dry weight measurement. Biomass in each aquarium was harvested, dried, and weighed 5 WAT.

Treatments were randomly assigned to individual aquaria and replicated three times, including the reference. All shoot, root, and rhizome data were analyzed using one-way ANOVA to determine herbicide effects. If effects were significant ( $P \le 0.05$ ), means were compared using LSD.

#### **RESULTS AND DISCUSSION**

#### Experiment 1

Reduction of Minnesota shoot biomass occurred with endothall (76%) and endothall combined with triclopyr (85%), flumioxazin (63%) and flumioxazin combined with triclopyr (82%; Figure 1A). The addition of triclopyr to endothall or flumioxazin did not significantly improve control compared to these products alone. Plants exposed to these herbicides showed symptoms of herbicide injury by 1 WAT and were necrotic by 4 WAT. There was little shoot recovery from endothall treatments compared to the reference (Figure 2A) due to significant herbicide effects on root biomass (Figure 1B). Although Minnesota root biomass reduction occurred in all treatments compared to the reference (Figure 1B), only the endothall treatment reduced root biomass below pretreatment levels.

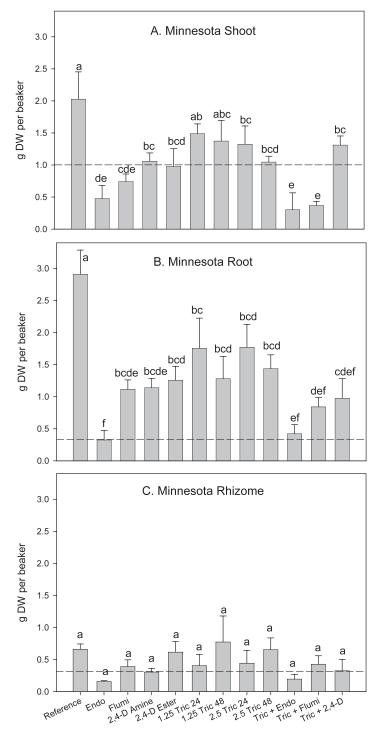


Figure 1. Experiment 1: Minnesota flowering rush (A) shoot, (B) root, and (C) rhizome biomass (mean  $\pm$  1 SE g DW, n = 4) 6 wk after exposure to endothall (Endo, 1.5 mg ai L<sup>-1</sup>), flumioxazin (Flumi, 0.4 mg ai L<sup>-1</sup>), 2,4-D amine (4.0 mg ae L<sup>-1</sup>), 2,4-D ester (4.0 mg ae L<sup>-1</sup>), triclopyr (Tric, 1.25 or 2.5 mg ae L<sup>-1</sup>) and combinations of triclopyr (1.25 mg ae L<sup>-1</sup>) with endothall (1.5 mg ai L<sup>-1</sup>), flumioxazin (0.4 mg ai L<sup>-1</sup>), and 2,4-D amine (4.0 mg ae L<sup>-1</sup>). Numbers in front of Tric represent concentrations (mg ae L<sup>-1</sup>) followed by exposure time (h). Treatments with the same letter are not significantly different (ANOVA P  $\leq$  0.05 shoot LSD = 0.663; root ANOVA P  $\leq$  0.05, LSD = 0.786; rhizome ANOVA P = 0.32). Dashed line represents pretreatment biomass.

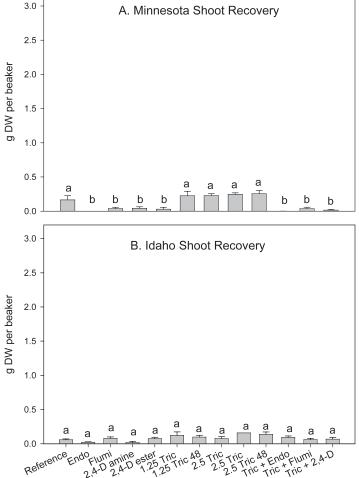


Figure 2. Experiment 1 Regrowth: (A) Minnesota and (B) Idaho flowering rush shoot biomass (mean  $\pm$  1 SE g DW, n = 4) collected as an assessment of recovery 8 wk after treatment with endothall (Endo, 1.5 mg ai L<sup>-1</sup>), flumioxazin (Flumi, 0.4 mg ai L<sup>-1</sup>), 2,4-D amine (4.0 mg ae L<sup>-1</sup>), 2,4-D ester (4.0 mg ae L<sup>-1</sup>), triclopyr (Tric, 1.25 or 2.5 mg ae L<sup>-1</sup>) and combinations of triclopyr (1.25 mg ae L<sup>-1</sup>) with endothall (1.5 mg ai L<sup>-1</sup>), flumioxazin (0.4 mg ai L<sup>-1</sup>), and 2,4-D amine (4.0 mg ae L<sup>-1</sup>). Numbers in front of Tric represent concentrations (mg ae L<sup>-1</sup>) followed by exposure time (h). Treatments with the same letter are not significantly different (Minnesota ANOVA P  $\leq$  0.05, LSD = (0.095; Idaho ANOVA P = 0.058). Dashed line represents pretreatment biomass.

All triclopyr treatments applied alone reduced Minnesota root biomass compared to the untreated reference (Figure 1B); however, the 1.25 mg ae  $L^{-1}$  treatments (24 and 48 h exposures) were not effective in reducing shoot biomass (Figure 1A). No triclopyr treatment applied alone eliminated shoot re-growth (Figure 2A). Conversely, both 2,4-D formulations were effective in reducing shoot and root biomass (Figures 1A and 1B). Flowering rush treated with triclopyr and 2,4-D exhibited initial herbicide symptoms of chlorosis and epinasty along the stems, but by 3 WAT, plants were growing vigorously with many green healthy shoots, although some browning was evident on a few decayed stems.

Reduction of Idaho shoot biomass ranged from 73 to 82% with endothall combined with triclopyr and endothall alone, respectively (Figure 3A). Likewise, flumioxazin alone (49%) and flumioxazin combined with triclopyr (65%)

reduced Idaho shoot biomass compared to the reference. Two other treatments that reduced shoot biomass > 50% included 2,4-D combined with triclopyr and 2.5 mg ae L<sup>-1</sup> triclopyr for the 48 h exposure time. Treatments that reduced biomass by 50% or more (or below pretreatment levels) were statistically similar (LSD = 0.488), including the flumioxazin alone treatment.

Herbicide effects on Idaho root biomass followed the same general trend as shoot biomass (Figure 3B). Treatments that significantly reduced root biomass compared to the reference were endothall, endothall combined with triclopyr, 2.5 mg ae  $L^{-1}$  triclopyr for 48 h exposure period, and 2,4-D amine combined with triclopyr. No herbicide treatments reduced root biomass below pretreatment levels. Shoot regrowth occurred in all Idaho plants (Figure 2B) as substantial root and rhizome biomass remained beneath the sediment to sustain plant recovery (Figures 3B and 3C). Most shoots sprouted within 1 wk after initial shoot removal at the 6 WAT harvest.

Rhizome biomass was not impacted by herbicide treatments in either the Minnesota or Idaho populations compared to the references (Figures 1C and 3C). Nonetheless, rhizome biomass was below pretreatment levels in plants exposed to endothall, including endothall combinations with triclopyr, for both plant populations. In addition, Idaho rhizome biomass was below pretreatment levels for all combinations of triclopyr with flumioxazin and 2,4-D as well as the maximum label rate of triclopyr for 48 h exposure time (Figure 3C).

Results from this experiment compare favorably to the results of a previous small-scale contact herbicide experiment on flowering rush where 1.5 mg ai  $L^{-1}$  endothall (24 h exposure) provided good control of Minnesota shoot biomass (> 80% at 4 WAT), and a treatment of 3 mg ai  $L^{-1}$  significantly reduced both shoot and root biomass in the Idaho population (Poovey et al. 2012). These data further confirm endothall efficacy in reducing flowering rush root biomass in both the Minnesota and Idaho triploid plant populations.

Non-target emergent and floating-leaf plants that may be growing in mixed communities with flowering rush in the field would probably be unharmed by the endothall CET used in this small-scale experiment. In an outdoor mesocosm study, spatterdock (Nuphar advena (Aiton) W.T. Aiton), pickerelweed (Pontederia cordata L.), and cattail (Typha latifolia L.) were not injured by endothall concentrations of 1.5 mg ai  $L^{-1}$  with a 24 h exposure (Skogerboe and Getsinger 2002). In another study, water lily (Nymphaea odorata Aiton.) and arrowhead (Sagittaria latifolia Willd.) were significantly impacted by 2.0 mg ai  $L^{-1}$  endothall in a static exposure of 120 h; however, these species are not likely to be impacted at lower concentrations and shorter exposure times (Skogerboe and Getsinger 2001). It is unknown how the addition of triclopyr will impact selectivity of endothall when used in combination; therefore, the selectivity of this treatment combination needs further investigation.

Flumioxazin has the potential to control submersed flowering rush at the maximum label rate (0.4 mg ai  $L^{-1}$ ), even with the water column pH  $\geq$  9. In this experiment, flumioxazin was effective against Minnesota and Idaho

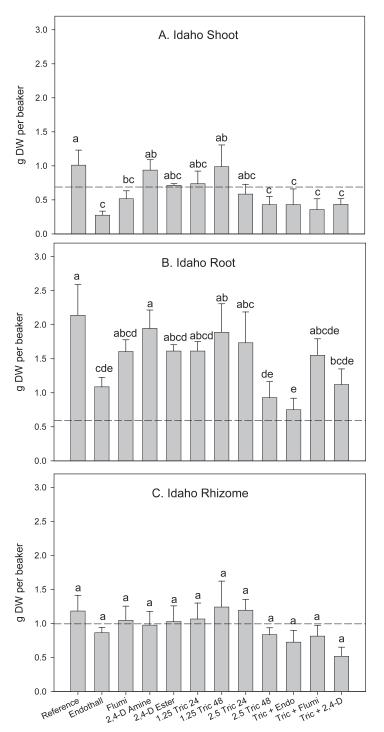


Figure 3. Experiment 1: Idaho flowering rush ((B) root, and (C) rhizome biomass (mean  $\pm 1$  SE g DW, n = 4) 6 wk after treatment with endothall (Endo, 1.5 mg ai L<sup>-1</sup>), flumioxazin (Flumi, 0.4 mg ai L<sup>-1</sup>), 2,4-D amine (4.0 mg ae L<sup>-1</sup>), 2,4-D ester (4.0 mg ae L<sup>-1</sup>), triclopyr (Tric, 1.25 or 2.5 mg ae L<sup>-1</sup>) and combinations of triclopyr (1.25 mg ae L<sup>-1</sup>) with endothall (1.5 mg ai L<sup>-1</sup>), flumioxazin (0.4 mg ai L<sup>-1</sup>), and 2,4-D amine (4.0 mg ae L<sup>-1</sup>). Numbers in front of Tric represent concentrations (mg ae L<sup>-1</sup>) followed by exposure time (h). Treatments with the same letter are not significantly different (shoot ANOVA P  $\leq$  0.05, LSD = 0.448; root ANOVA P  $\leq$  0.05, LSD = 0.800; rhizome ANOVA P = 0.39). Dashed line represents pretreatment biomass.

flowering rush, verifying its efficacy against Idaho flowering rush in a previous experiment (Poovey et al. 2012). Preliminary experimental data indicate that 0.4 mg ai  $L^{-1}$ may negatively impact water lily and spatterdock; however, re-growth of those plants may occur within the growing season of herbicide application (authors' unpublished data). Differences in plant susceptibility to flumioxazin can depend on application technique. For example, submersed applications of 0.4 mg ai  $L^{-1}$  injured emergent vegetation, but foliar applications using 841 g ha<sup>-1</sup> did not (Mudge and Haller 2012). Other research indicates that flumioxazin selectivity is variable, with little pattern of plant susceptibility among families or genera (Mudge and Haller 2010, Glomski and Netherland 2012a). Increased efficacy of flowering rush would likely be achieved in waterways with neutral pH ranges (pH = 7), since hydrolysis of flumioxazin occurs in 16 to 18 h at lower pH levels (Katagi 2003, Mudge and Haller 2010).

Use of synthetic auxin herbicides for control of triploid flowering rush had mixed results. Reduction of shoot biomass ranged from 27 to 48% for triclopyr (all CETs) and 48 to 52% for 2,4-D (both formulations) in Minnesota plants. Root biomass reduction occurred in all but the 1.25 mg ae  $L^{-1}$  triclopyr treatments. In Idaho plants, there was little shoot or root reduction compared to the references. The combination of 2,4-D and triclopyr did not substantially enhance efficacy over each product alone. Selectivity differs between 2,4-D and triclopyr, and may differ between liquid and granular formulations. American bulrush (Schoenoplectus americanus Pers.) has been reported as tolerant to high concentrations of triclopyr (2 mg ae  $L^{-1}$ ) and moderate concentrations of 2,4-D ester (2.5 mg ae  $L^{-1}$ ) using a 24 h exposure time; however, soft-stem bulrush (S. taberbaemontani (C.C. Gmel.) Palla) biomass was significantly reduced with these CETs in the same experiment (Glomski et al. 2009). Hard-stem bulrush (S. acutus (pers.)Volkart ex Schinz & R. Keller) biomass was not affected when exposed to triclopyr concentrations of 1 mg ae  $L^{-1}$  using a liquid formulation, but was reduced using the granular formulation (Glomski and Netherland 2012b). In another experiment, Glomski and Nelson (2008) found that water lily was initially injured by triclopyr (2 mg ae  $L^{-1}$ ) and 2,4-D ester (2.5 mg ae  $L^{-1}$ ) using a 24 h exposure time. Plants recovered from the triclopyr treatment, but not the 2,4-D ester treatment. These same herbicide treatments were used against spatterdock, where shoot biomass was reduced by 48% following herbicide application, although root biomass was not affected by herbicide treatment, and re-growth from roots and rhizomes was observed. Since submersed applications of the synthetic auxins in this experiment provided only fair control (50%) of submersed flowering rush at CETs that compromise selectivity, additional research with these products should focus on foliar applications on the emergent form of flowering rush.

# **Experiment 2**

Compared to the reference, herbicide treatments in this experiment were not effective in significantly reducing shoot and rhizome biomass of either the Minnesota or

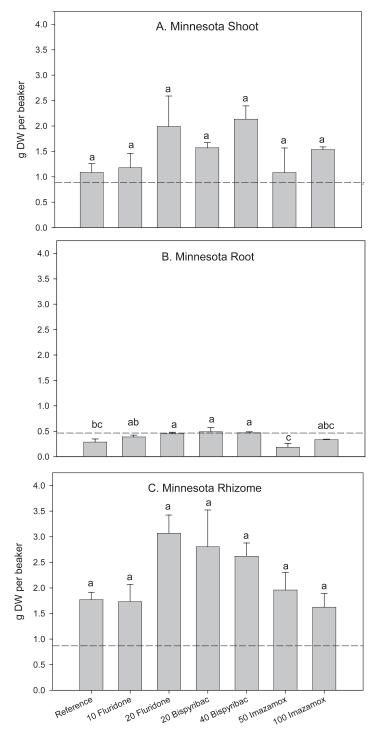


Figure 4. Experiment 2: Minnesota flowering rush (A) shoot, (B) root, and (C) rhizome biomass (mean  $\pm 1$  SE g DW, n = 3) 5 wk after treatment with fluridone, bispyribac, and imazamox. Numbers in front of herbicide active ingredient represent concentrations (µg ai L<sup>-1</sup>). Treatments with the same letter are not significantly different (shoot ANOVA P = 0.26; root ANOVA P  $\leq 0.05$ , LSD = 0.163; rhizome ANOVA P = 0.12). Dashed line represents pretreatment biomass.

Idaho flowering rush (Figures 4 and 5). The 100  $\mu$ g ai L<sup>-1</sup> imazamox and 40  $\mu$ g ai L<sup>-1</sup> bispyribac treatments reduced Idaho root biomass by 63 and 53%, respectively (Figure 5B). Rhizome biomass increased two-fold (MN) and three-fold

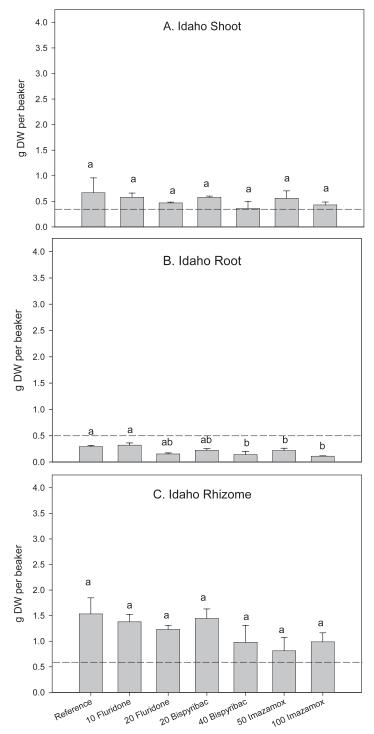


Figure 5. Experiment 2: Idaho flowering rush (A) shoot, (B) root, and (C) rhizome biomass (mean  $\pm 1$  SE g DW, n = 3) 5 wk after treatment with fluridone, bispyribac, and imazamox. Numbers in front of herbicide active ingredient represent concentrations (µg ai L<sup>-1</sup>). Treatments with the same letter are not significantly different (shoot ANOVA P = 0.75; root ANOVA P  $\leq 0.05$ , LSD = 0.112; rhizome ANOVA P = 0.28). Dashed line represents pretreatment biomass.

(ID) compared to pretreatment levels (Figures 4C and 5C). Hroudová and Zákravský (1993) found that triploid flowering rush allocated most of its biomass to rhizomes by the end of the growing season in preparation for overwintering.

J. Aquat. Plant Manage. 51: 2013

Visually, all herbicide treatments produced slight bleaching and some browning of shoots through 2 WAT. Shoots showed signs of herbicide-induced stress until the end of the experiment; however, biomass was not affected by herbicide treatment (Figures 4 and 5).

Lack of efficacy may be attributed to inherent tolerance of submersed flowering rush to subsurface applications of these plant enzyme-specific herbicides. Differential response of emergent and submersed aquatic plants to ALS-inhibiting herbicides has been reported (Getsinger et al. 1994, Chiconela et al. 2004, Koschnick et al. 2007, Glomski and Netherland 2008). Slight changes in the molecular structure of these herbicides greatly affect the potency and spectrum of plant susceptibility (Ladner 1991). For example, soft-stem bulrush was more susceptible to bispyribac than imazamox, but maidencane (Panicum hemitomon Schult.), pickerelweed (Pontederia cordata L.), and duck potato (Sagittaria lancifolia L.) were more susceptible to imazamox than bispyribac in an outdoor mesocosm experiment (Koschnick et al. 2007). In another experiment, differences were noted between Sagittaria species duck potato and arrowhead (S. latifolia Willd.), where arrowhead was significantly more susceptible to bispyribac than duck potato (Glomski and Mudge 2009).

Higher doses and/or longer exposure times of the herbicides tested might result in better control of the submersed growth stage of flowering rush. Herbicide doses were chosen based on use patterns developed for ALS and PDS chemistries when controlling other submersed aquatic plants, such as hydrilla [Hydrilla verticillata (L.f.) Royle] and Eurasian watermilfoil (Myriophyllum spicatum L.), which currently focus on low use rates (< 100  $\mu g$  ai L<sup>-1</sup>) with long-term exposures of 4 to 12 wk (Netherland et al. 1993). Given that fluridone, bispyribac, and imazamox require long exposure times for submersed treatments, these herbicides would not be appropriate products to control submersed flowering rush in systems where adequate aqueous exposure times cannot be maintained. Exposure time may be extended through the use of barrier curtains, which reduce water movement in treated areas and potentially increase herbicide contact time. In reservoirs where water fluctuations are manipulated, greater efficacy could be achieved when herbicides are applied during periods of low water discharge, thereby increasing aqueous contact times around target plants (Getsinger et al. 2012).

Using aquatic herbicides in combination with another management practice, such as herbicide application after dewatering for bareground applications, may provide better control of flowering rush biomass. Bareground applications of fluridone were effective in reducing shoot and root biomass in mesocosm experiments (Woolf et al. 2011). Application of fluridone to newly sprouted vegetative propagules (tubers) after dewatering was successful in controlling monoecious hydrilla in Lake Gaston, a run of the river reservoir on the border of North Carolina and Virginia (Nawrocki 2011). An advantage of these integrated approaches is the reduction in the amount of herbicide used when applied to either the bareground or shallow water if application is immediately after dewatering; however, timing of application would be difficult if sprouting of flowering rush rhizomes is non-seasonal or random.

As with the synthetic auxin herbicides, foliar applications to emergent flowering rush may be the best management practice for the ALS herbicides. Both imazamox and imazapyr (2-[4,5-dihydro-4-methyl-4-(1-methylethyl)-5-oxo-1-H-imidazol-2-yl]-3-pyridinecarboxylic acid), another ALSinhibitor, have shown promise in controlling emergent flowering rush in field trials (Rice et al. 2009).

# ACKNOWLEDGEMENTS

Support and cooperation for this work were provided by the Pelican River Watershed District, the U.S. Army Engineer Seattle District, and the Aquatic Ecosystem Restoration Foundation. Mike Netherland, John Skogerboe, and Lee Ann Glomski kindly provided technical assistance and reviewed earlier versions of this manuscript. Special thanks are extended to Michelle Marko (Concordia College) and Betsy Hull (Albeni Falls Project, U.S. Army Engineer Seattle District) for rhizome collections. Additional support was provided by Craig Brengle (Albeni Falls Project, U.S. Army Engineer Seattle District) and Tera Guetter (Pelican River Watershed District). Permission was granted by the Chief of Engineers to publish this information. Citation of trade names does not constitute an official endorsement or approval of the use of such commercial products.

#### SOURCES OF MATERIALS

<sup>1</sup>Black Kow<sup>®</sup> Topsoil, Black Gold Compost Co., Oxford, FL.

<sup>2</sup>Scotts<sup>®</sup> Premium Topsoil, the Scotts Co., Marysville, OH.

<sup>3</sup>Aquathol<sup>®</sup> K, United Phosphorus Inc., King of Prussia, PA.

<sup>4</sup>Clipper<sup>®</sup>, Valent USA Corp., Walnut Creek, CA.

<sup>5</sup>DMA 4 IVM, Dow AgroSciences LLC, Indianapolis, IN.

<sup>6</sup>Renovate<sup>®</sup>, SePRO Corp., Carmel, IN.

<sup>7</sup>2,4-D ester liquid, NuFarm Americas Inc., Burr Ridge, IL.

<sup>8</sup>Optic Stowaway<sup>®</sup> Temperature Probe, Onset Computer Corp., Bourne, MA.

 $^9 \mathrm{YSI}$  Model 556, Handheld Multi-Parameter Probe, YSI, Yellow Springs, OH.

<sup>10</sup>Sonar<sup>®</sup> AS, SePRO Corp., Carmel, IN.

<sup>11</sup>Tradewind<sup>®</sup>, Valent USA Corp., Walnut Creek, CA.

<sup>12</sup>Clearcast<sup>®</sup>, SePRO Corp., Carmel, IN.

#### LITERATURE CITED

- Bartels PG, Watson CW. 1978. Inhibition of carotenoid synthesis by fluridone and norflurazon. Weed Sci. 26:198–203.
- Belgers JDM, Van Lieverloo RJ, Van der Pas LJT, Van den Brink PJ. 2007. Effects of the herbicide 2,4-D on the growth of nine aquatic macrophytes. Aquat. Bot. 86:260–268.
- Chiconela T, Koschnick TJ, Haller WT. 2004. Selectivity of metsulfuron methyl to six common littoral species in Florida. J. Aquat. Plant Manage. 42:115–116.
- Countryman WD. 1970. The history, spread and present distribution of some immigrant aquatic weeds in New England. Hyacinth Control J. 8:50–52.
- Eckert CG, Lui K, Bronson K, Corradini P, Bruneau A. 2003. Population genetic consequences of extreme variation in sexual and clonal reproduction in an aquatic plant. Mol. Ecol. 12:331-344
- Getsinger KD, Dick GO, Crouch RM, Nelson LS. 1994. Mesocosm evaluation of bensulfuron methyl activity on Eurasian watermilfoil, vallisneria, and American pondweed. J. Aquat. Plant Manage. 32:1–6.
- Getsinger KD, Madsen JD, Wersal RM, Skogerboe JG, Nawrocki JJ, Richardson RJ, Sternberg MR. 2012. Selective control of Eurasian

watermilfoil and curlyleaf pondweed in Noxon Rapids Reservoir, Montana: Aquatic herbicide evaluations 2009–2010. Technical Report ERDC/EL TR-12-00. U.S. Army Engineer Research and Development Center, Vicksburg (MS). 56 p.

- Getsinger KD, Poovey AG, James WF, Stewart RM, Grodowitz MJ, Maceina MJ, Newman RM. 2002. Management of Eurasian watermilfoil in Houghton Lake, Michigan: Workshop Summary. Technical Report ERDC/EL TR-02-24. U.S. Army Engineer Research and Development Center, Vicksburg, MS. 88 p.
- Glomski LM, Mudge CR. 2009. Effect of submersed applications of bispyribac-sodium on non-target emergent vegetation. APCRP Technical Notes Collection (ERDC/TN APCRP-CC-12). U.S. Army Engineer Research and Development Center, Vicksburg, MS. 7 p.
- Glomski LM, Nelson LS. 2008. Evaluation of 2,4-D ester and triclopyr amine against waterlily and spatterdock. APCRP Technical Notes Collection (ERDC/TN APCRP-CC-07). U.S. Army Engineer Research and Development Center, Vicksburg, MS. 6 p.
- Glomski LM, Netherland MD. 2008. Efficacy of fluridone, penoxsulam, and bispyribac-sodium on variable-leaf milfoil. J. Aquat. Plant Manage. 46:193–196.
- Glomski LM, Netherland MD. 2012a. Utilization of a small-scale primary screening method to predict impacts of flumioxazin and carfentrazoneethyl on native and invasive submersed plants. J. Aquat. Plant Manage. In press.
- Glomski LM, Netherland MD. 2012b. Evaluation of liquid and granular triclopyr against water lily, spatterdock, and hardstem bulrush. Proceedings, 52<sup>nd</sup> Annual Meeting of the Aquatic Plant Management Society, July 22–25, Salt Lake City, UT.
- Glomski LM, Netherland MD, Nelson LS. 2009. Evaluation of 2,4-D and triclopyr against spatterdock and bulrush. APCRP Technical Notes Collection (ERDC/TN APCRP-CC-10). U.S. Army Engineer Research and Development Center, Vicksburg, MS. 8 p.
- Green WR, Westerdahl HE. 1990. Response of Eurasian watermilfoil to 2,4-D concentration and exposure times. J. Aquat. Plant Manage. 28:27–32.
- Hroudová Z, Zákravský p. 1993. Ecology of two cytotypes of *Butomus umbellatus* II. Reproduction, growth and biomass production. Folia Geobot. Phytot. 28:413-424.
- Katagi T. 2003. Hydrolysis of n-phenylimide herbicide flumioxazin and its anilic acid derivative in aqueous solutions. J. Pest. Sci. 28:44–50.
- Koschnick TJ, Netherland MD, Haller WT. 2007. Effects of three ALSinhibitors on five emergent native plant species in Florida. J. Aquat. Plant Manage. 45:47–51.
- Ladner DW. 1991. Structure-activity relationships among imidazolinones herbicides. In: D. L. Shaner and S. L. O'Connor (*eds.*). The Imidazolinone Herbicides. CRC Press, Boca Raton, FL. p. 31–51.
- Lui K, Thompson FL, Eckert CG. 2005. Causes and consequences of extreme variation in reproductive strategy and vegetative growth among invasive populations of a clonal aquatic plant, *Butomus umbellatus* L. (Butomaceae). Biol. Invasions 7:427–444.
- Mudge CR, Haller WT. 2010. Effect of pH on submersed aquatic plant response to flumioxazin. J. Aquat. Plant Manage. 48:30-34.
- Mudge CR, Haller WT. 2012. Response of target and non-target floating and emergent aquatic plants to flumioxazin. J. Aquat. Plant Manage. 50:111– 116.
- Nawrocki, J. 2011. Environmental and physiological factors affecting submersed aquatic weed management. MS Thesis. North Carolina State University, Raleigh, NC. 204 p.
- Netherland MD, Getsinger KD. 1992. Efficacy of triclopyr on Eurasian watermilfoil: Concentration and exposure time effects. J. Aquat. Plant Manage. 30:1–5.
- Netherland MD, Getsinger KD. 1993. Fluridone concentration and exposure time requirements for control of Eurasian watermilfoil and hydrilla. J. Aquat. Plant Manage. 31:189–194.
- Netherland MD, Getsinger KD, Skogerboe JG. 1997. Mesocosm evaluation of the species-selective potential of fluridone. J. Aquat. Plant Manage. 35:41-50.
- Netherland MD, Getsinger KD, Turner EG. 1993. Fluridone concentration and exposure time requirements for control of Eurasian watermilfoil and hydrilla. J. Aquat. Plant Manage. 31:189–194.
- Poovey AG, Getsinger KD. 2005. Use of herbicides to control the spread of aquatic invasive plants. J ASTM Intern. 2(10), Paper Id JAI13254.
- Poovey AG, Mudge CR, Thum R, James C, Getsinger KD. 2012. Evaluations of contact herbicides for controlling two populations of flowering rush. J. Aquat. Plant Manage. 50:48–54.

- Rice P, Dupuis V, Mitchell A. 2009. Results in the second summer after foliar application of herbicides to flowering rush. Report to the Montana Noxious Weed Trust Fund. Montana State University, Bozeman, MT. 7 p.
- Skogerboe JG, Getsinger KD. 2001. Endothall species selectivity evaluation: Southern latitude aquatic plant community. J. Aquat. Plant Manage. 39:129–135.
- Skogerboe JG, Getsinger KD. 2002. Endothall species selectivity evaluation: Northern latitude aquatic plant community. J. Aquat. Plant Manage. 40:1–5.
- Smart RM, Barko JW. 1985. Laboratory culture of submersed freshwater macrophytes on natural sediments. Aquat. Bot. 21, 251–263.
- Tranel PJ, Wright TR. 2002. Resistance of weeds to ALS-inhibiting herbicides: What have we learned. Weed Sci. 50:700-712.
- Weed Science Society of America (WSSA). 2007. Herbicide Handbook Ninth Edition. Weed Science Society of America, Lawrence, KS. 403 p.
- Woolf T, Madsen J, Wersal R. 2011. Flowering rush control project for Lake Pend Oreille, Idaho: Preliminary summary on mesocosm and field evaluations. GRI Report #5048. Geosystems Research Institute, Mississippi State University, Starkville, MS. 10 p.