J. Aquat. Plant Manage. 51: 39–44

Impact of aquatic herbicide combinations on nontarget submersed plants

CHRISTOPHER R. MUDGE*

ABSTRACT

The tolerances of four nontarget submersed aquatic plant species to low-dose herbicide and herbicide combinations were evaluated in 2 growth chamber experiments. The first experiment assessed the response of narrow-leaf and wide-leaf biotypes of vallisneria (*Vallisneria americana* Michaux), whereas the second experiment tested American pondweed (*Potamogeton nodosus* Poir.), Illinois pondweed (*Potamogeton illinoensis* Morong), and coontail (*Ceratophyllum demersum* L.). The federally registered aquatic herbicides bispyribac-sodium (2,6-bis[(4,6-dimethoxy-2-pyrimidinyl)oxy]benzoic acid-sodium), diquat (6,7-dihydrodipyrido[1,2-a:29,19-c]pyrazinediium ion), endothall (7-oxabicyclo[2.2.1]heptane-2,3-dicarboxylic acid), flumioxazin (2-[7-fluoro-3,4-dihydro-3-oxo-4-(2-propynyl)-2H-1,4-benzoxazin-6-yl]-4,5,6,7-tetrahydro-1H-isoindole-1,3(2H)-dione), imazamox (2-[4,5-dihydro-4-methyl-4-(1-methylethyl)-5-oxo-1H-imidazol-2-yl]-5-(methoxymethyl)-3-pyridinecarboxylic acid), and penoxsulam (2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c]pyrimidin-2-yl)-6-(trifluoromethyl)benzenesulfonamide) were applied alone and in combination to determine species selectivity. All herbicides applied alone to narrow-leaf and wide-leaf vallisneria, except the dipotassium salt of endothall 500 μg ai L^{-1} (500 ppb), failed to reduce shoot biomass compared to the nontreated control 11 wk after treatment (WAT). Combination treatments resulted in a range of selectivity and injury to both vallisneria biotypes; however, none of the single or combination treatments completely eliminated biomass. Conversely, increased injury was noted when these treatments were applied to the pondweed species and coontail. Only bispyribac-sodium at 10 μg ai L^{-1} (B10), flumioxazin at 50 μg ai L^{-1} (F50), penoxsulam at 50 μg ai L^{-1} (P5), and F50 + P5 failed to reduce shoot biomass of American pondweed compared to the nontreated control 8 WAT. All herbicides and herbicide combinations—except flumioxazin applied alone—decreased Illinois pondweed shoot biomass after 8 wk. Similar to Illinois pondweed, coontail was very susceptible to the single and combination herbicide treatments evaluated in this study, and all treatments except B10, P5, and B10 + P5 reduced shoot biomass 8 WAT. These data indicate that nontarget aquatic plants vary in their tolerance to low-dose herbicide and herbicide combination treatments.

Key words: bispyribac-sodium, *Ceratophyllum demersum*, diquat, endothall, flumioxazin, herbicide selectivity, imazamox, penoxsulam, *Potamogeton illinoensis*, *Potamogeton nodosus, Vallisneria americana*.

*Research Biologist, U.S. Army Engineer Research and Development Center, Vicksburg, MS 39180. Corresponding author's E-mail: Christopher.R.Mudge@usace.army.mil. Received for publication December 12, 2011 and in revised form November 5, 2012.

INTRODUCTION

Mixed populations of invasive species such as hydrilla [*Hydrilla verticillata* (L. f.) Royle] and nontarget native plants can often be found coexisting until the invasive plant ultimately takes over the aquatic ecosystem. It is important to manage the weedy species before monotypic stands form. Several new herbicidal modes of action have recently received federal registration or experimental use permits (EUP) in the United States for aquatic weed management. These products control hydrilla in a number of ways; for example, quinclorac (3,7-dichloro-8-quinolinecarboxylic acid) is a synthetic auxin. Bispyribac-sodium, bensulfuron methyl (2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)amino]carbonyl]amino]sulfonyl]methyl]benzoic acid), imazamox, and penoxsulam inhibit acetolactate synthase (ALS), whereas flumioxazin and topramezone ([3-(4,5-dihydro-3-isoxazolyl)-2-methyl-4-(methylsulfonyl)phenyl](5-hydroxy-1-methyl-1H-pyrazol-4-yl)methanone) inhibit protoporphyrinogen IX oxidase (PPO) and *p*-hydroxyphenylpyruvate dioxygenase (HPPD), respectively (Mossler et al. 2006, Koschnick et al. 2007, Mossler and Fishel 2007, Senseman 2007). Only bispyribac-sodium, flumioxazin, imazamox, and penoxsulam are currently registered by the U.S. Environmental Protection Agency under Section 3 of the Federal Insecticide, Fungicide, and Rodenticide Act for aquatic use.

Optimal use patterns for efficacy and selectivity of the newly registered and EUP herbicides are still under investigation. The ALS inhibitors have resulted in varying levels of hydrilla control in operational field applications. These products are typically applied at low concentrations (5 to 100 μg ai L^{-1} (5 to 10 ppb)) and generally require long continuous exposures (60 to 120+ d). Consequently, repeat applications are often necessary to maintain adequate exposure periods. In contrast to the long-term exposure requirements of ALS herbicides, the contact herbicide flumioxazin is degraded by hydrolysis in approximately 4 d, 16 h, and 17 min at pH 5.0, 7.0, and 9.0, respectively (Katagi 2003). Consequently, field trials with flumioxazin have resulted in variable control of hydrilla, with regrowth at or near the water surface less than 8 WAT of water with a pH > 8.0 (Mudge 2007). The long exposure and higher concentration requirements of the ALS herbicides, in addition to the rapid degradation of the PPO inhibitor flumioxazin, present disparate scenarios that have forced researchers and managers to find alternative ways to use these products.

Initial laboratory and small-scale field trials suggest that hydrilla control may be improved by applying low concentrations of ALS herbicides in combination with each other or in combination with contact herbicides. For example, combinations of penoxsulam and endothall decreased exposure requirements for penoxsulam, reduced endothall use rates for initial reduction of hydrilla biomass, and increased duration of control versus endothall alone (Heilman et al. 2009). These results suggest that the use of contact-plus-systemic herbicide combinations may expand. Preliminary growth chamber research has demonstrated that combinations of many of these products can effectively reduce hydrilla dry weight up to 97% by 9 WAT (C. R. Mudge, unpub. data).

No research has been published and little is known about the impact of these combinations on nontarget aquatic plants. Native submersed aquatic vegetation (SAV) is ecologically important, and could be exposed to these combination treatments following large-scale hydrilla management if both hydrilla and native SAVs coexist. Therefore, 2 experiments were undertaken to evaluate combinations of recently registered herbicides (bispyribac-sodium, flumioxazin, imazamox, and penoxsulam) and commercial standards diquat and endothall at low use rates to determine selectivity against desirable species that are native to Florida as well as other regions of the United States. The first experiment used 2 biotypes of vallisneria as there have been anecdotal claims of differential herbicide susceptibility between the 2 biotypes (narrow-leaf and wide-leaf) (M. D. Netherland, pers. comm.). The second experiment used American pondweed, Illinois pondweed, and coontail.

MATERIALS AND METHODS

Experiment 1

Experiment 1 was initiated in February 2010 at the U.S. Army Engineer Research and Development Center (USAERDC) in Vicksburg, MS, in a large controlled-environment chamber equipped with 60 55-L glass aquaria specifically designed and operated for growing submersed plants. Conditions conducive to maintaining healthy submersed plant growth were maintained: temperature of 24.0 ± 1 C (75.4°F) with a light intensity of 189.74 ± 74 μmol m^{-2} s^{-1} and a 14h : 10 h (light : dark) photoperiod.

Ramets of narrow-leaf vallisneria were collected from Lake Tohopekaliga, FL, and ramets of wide-leaf vallisneria were purchased from a commercial aquatic plant nursery[1], and shipped overnight to USAERDC. Narrow-leaf plants averaged 17.58 ± 1.02 g (0.60 ± 0.0359 oz) fresh weight (mean ± SE) and 15 leaves per plant, whereas wide-leaf plants averaged 3.36 ± 0.58 g fresh weight and 6 leaves per plant. Individual ramets were planted into 750-ml (25.4 oz) plastic beakers filled with sediment composed of 3 : 1 topsoil[2] : sand. Sediments were amended with Osmocote® 19–6–12 fertilizer[3] (2 g L^{-1} sediment). The sediment surface was top-dressed with a 1-cm (0.39 inches) layer of silica sand to reduce sediment and nutrient suspension in the water column. One beaker of narrow-leaf vallisneria was placed in each aquarium filled with growth culture solution (Smart and Barko 1985). Due to limited plant material, 1 beaker of wide-leaf vallisneria was placed in 33 of the 60 tanks. Plants were allowed to grow for 5 wk before herbicide treatment.

Federally registered herbicides including bispyribac-sodium[4], diquat[5], endothall (dipotassium salt)[6], flumioxazin[7], imazamox[8], and penoxsulam[9] were applied alone and in combination at low use rates (Table 1). Herbicide combinations and concentrations were chosen to determine efficacy among different herbicide modes of action. Previous growth chamber research revealed that many of these combinations were efficacious against hydrilla and warranted further investigation to determine their effects on nontarget species (C. R. Mudge, unpub. data). Each individual herbicide, with regard to the combination treatments, was applied individually to the aquariums and not mixed together in a stock solution prior to treatment. Herbicide exposure times were as follows: bispyribac-sodium, static; diquat, 8 h; endothall, 3 d; flumioxazin, 24 h; imazamox, 14 d; and penoxsulam, static. At the termination of assigned exposure times, aquaria were drained and filled to remove herbicide residue, and retreated with the herbicide that required additional exposure. The exposure times were chosen based on previous growth chamber research, product half-life under field conditions, and slower degradation in the growth chamber. The low-dose concentrations were selected from previous hydrilla field research (M. D. Netherland, unpub. data) as concentrations that would provide limited hydrilla control when applied alone, but that might, in combination with other products products, result in greater efficacy. Nontreated reference aquaria were also used to compare plant growth in the absence of herbicide. Water quality

Table 1. Aquatic herbicides applied alone and in combination to nontarget submersed aquatic plants.[1]

Herbicide treatments	Concentration (μg ai L^{-1})	Plants treated
Alone treatments		
Bispyribac-sodium	10	NL, AP, IL, C
Diquat	100	NL, AP, IL, C
Endothall	500	NL, WL, AP, IL, C
Flumioxazin	50	NL, WL, AP, IL, C
Imazamox	50	NL, WL, AP, IL, C
Penoxsulam	5	NL, WL, AP, IL, C
Combination treatments		
Bispyribac-sodium + diquat	10 + 100	NL, AP, IL, C
Bispyribac-sodium + endothall	10 + 500	NL, AP, IL, C
Bispyribac-sodium + flumioxazin	10 + 50	NL, AP, IL, C
Bispyribac-sodium + imazamox	10 + 50	NL, AP, IL, C
Bispyribac-sodium + penoxsulam	10 + 5	NL, AP, IL, C
Diquat + flumioxazin	100 + 50	NL, AP, IL, C
Diquat + penoxsulam	100 + 5	NL, AP, IL, C
Endothall + flumioxazin	500 + 50	NL, WL, AP, IL, C
Endothall + imazamox	500 + 50	NL, WL, AP, IL, C
Endothall + penoxsulam	500 + 5	NL, WL, AP, IL, C
Flumioxazin + imazamox	50 + 50	NL, WL, AP, IL, C
Flumioxazin + penoxsulam	50 + 5	NL, WL, AP, IL, C
Imazamox + penoxsulam	50 + 5	NL, WL, AP, IL, C
Nontreated control	0	NL, WL, AP, IL, C

[1]Abbreviations: NL, narrow-leaf vallisneria; WL, wide-leaf vallisneria; AP, American pondweed; IL, Illinois pondweed; C, coontail.

variables were measured with a hand-held multi-parameter probe.[10] At treatment, water temperature was 22.6 ± 0.2 ° C and pH ranged from 7.6 to 8.6.

Three beakers of each plant species were used to measure pretreatment shoot biomass. Narrow-leaf vallisneria pretreatment shoot and root dry weights were 1.7 ± 0.2 and 0.5 ± 0.2 g, respectively, and wide-leaf vallisneria shoot and root dry weights were 0.6 ± 0.1 g and 0.5 ± 0.2 g, respectively. All viable plant material was harvested 11 WAT, placed in a forced-air drying oven at 70 C for 1 wk, weighed, and analyzed for shoot and root dry weight biomass. The study was randomized with 3 replications per treatment. All data were analyzed using analysis of variance and means were separated using Fisher's Protected LSD ($P \leq 0.05$). Normality assumptions were assessed for all response variables. Wide-leaf vallisneria root biomass did not meet normality assumptions and thus was transformed using a base-10 log transformation. Respective means were back-transformed for tabular depictions.

Experiment 2

Experiment 2 was conducted under similar conditions at USAERDC in the controlled-environment growth chamber. Coontail, American pondweed, and Illinois pondweed were collected from Orange Lake (Florida), the Lewisville Aquatic Ecosystem Research Facility (Lewisville, TX), and the Center for Aquatic and Invasive Plants (Gainesville, FL), respectively, and shipped overnight to USAERDC. One healthy stem clipping of coontail (30 cm), 2 American pondweed stem clippings (30 cm, average 30 leaves per plant), and 2 Illinois pondweed clippings (30 cm, average 35 leaves per plant) were planted separately into 750-ml plastic beakers filled with sediment composed of 3 : 1 topsoil : sand. Sediments were amended with Osmocote® 19–6–12 fertilizer (2 g L^{-1} sediment). The sediment surface was top-dressed with a 1-cm layer of silica sand to reduce sediment and nutrient suspension in the water column. All plants were placed in each aquarium filled with growth culture solution (Smart and Barko 1985). Two beakers of Illinois pondweed were placed in each aquarium, whereas single beakers were utilized for each of the other 2 species. Plants were allowed to grow for 6 wk before herbicide treatment.

As in Experiment 1, herbicides were applied alone and in combination at low use rates (Table 1). All herbicide combinations were applied individually and exposure times were the same as those described in Experiment 1. At the termination of assigned exposure times, aquaria were drained and filled to remove herbicide residue, and retreated with the herbicide that required additional exposure. Nontreated reference aquaria were also used to compare plant growth in the absence of herbicide. At treatment, water temperature was 22.8 ± 0.1 C and pH ranged from 6.9 to 8.1.

American pondweed pretreatment shoot and root dry weights were 1.2 ± 0.1 and 0.1 ± 0.02 g, respectively; Illinois pondweed shoot and root dry weights were 0.5 ± 0.2 and 0.1 ± 0.03 g, respectively; and coontail shoot dry weight was 2.9 ± 0.9 g. Two beakers of Illinois pondweed and 1 beaker each of American pondweed and coontail per aquarium were harvested 8 WAT, placed in a forced-air drying oven at 70 C for 1 wk, weighed, and analyzed for shoot and root dry weight biomass. The study was randomized with 3 replications per treatment. All data were analyzed using analysis of variance and means were separated using Fisher's Protected LSD ($P \leq 0.05$). Normality assumptions were assessed for all response variables. Coontail shoot biomass data did not meet normality assumptions and thus were transformed using a plus-1 square transformation. Respective means were back-transformed for tabular depictions.

RESULTS AND DISCUSSION

Experiment 1

None of the herbicides applied alone to narrow-leaf vallisneria, except the dipotassium salt of endothall (500 µg ai L^{-1} [E500]), reduced shoot biomass compared to the nontreated control 11 WAT (Table 2). Only 5 of the 13 combination treatments were injurious to narrow-leaf vallisneria: specifically, bispyribac-sodium at 10 µg ai L^{-1} (B10) + diquat at 100 µg ai L^{-1} (D100), B10 + E500, E500 + flumioxazin at 50 µg ai L^{-1} (F50), E500 + imazamox at 50 µg ai L^{-1} (I50), and E500 + penoxsulam at 5 µg ai L^{-1} (P5) reduced shoot biomass by 47 to 83% compared to non-treated control plants. Four of these combinations included the fast-acting contact herbicide endothall. Combinations that included endothall were not significantly different from the treatment with endothall alone; therefore, plant injury is likely due to endothall rather than to the herbicide combination. This is supported by Skogerboe and Getsinger's 2002 report, which stated that 500 µg ai L^{-1} of endothall significantly reduced vallisneria dry weight 6 WAT. In Experiment 1, narrow-leaf vallisneria shoots were not impacted by combinations of the slow-acting systemic ALS herbicides bispyribac-sodium, imazamox, or penoxsulam as well as combinations with the PPO inhibitor flumioxazin (Table 2).

Treatments with a single herbicide had a similar effect on root dry weight, with only E500 reducing narrow-leaf vallisneria root dry weight by 53% 11 WAT (Table 2). In addition, the combination treatments of B10 + D100, B10 + E500, E500 + F50, E500 + I50, and E500 + P5 decreased root biomass 43 to 74%. All other herbicides alone or in combination reduced narrow-leaf vallisneria root dry weight by ≤ 41%.

Wide-leaf vallisneria shoots were affected by the herbicide treatments E500, E500 + F50, and E500 + I50 (Table 2). As stated above, this injury was likely due to endothall alone, with little or no phytotoxicity attributable to flumioxazin or imazamox. For example, Mudge and Haller (2010) showed that when flumioxazin was applied at pH 7.0, a concentration of 1,244 µg ai L^{-1} (more than 24 orders of magnitude greater than the concentration tested in these experiments) was required to reduce dry weight biomass by 50%. Wide-leaf vallisneria roots were affected more by single and combination herbicide treatments than any other plant response variable evaluated in Experiment 1 (Table 2). Flumioxazin, penoxsulam, and the combination of these 2 herbicides were the only treatments evaluated in this study

Table 2. Effect of the aquatic herbicides bispyribac-sodium at 10 μg ai L^{-1} (B10), diquat at 100 μg ai L^{-1} (D100), endothall at 500 μg ai L^{-1} (E500), flumioxazin at 50 μg ai L^{-1} (F50), imazamox at 50 μg ai L^{-1} (I50), and penoxsulam at 5 μg ai L^{-1} (P5) alone and in combination on narrow-leaf and wide-leaf vallisneria dry weight (g, mean ± 1 SE) 11 wk after treatment.

	Narrow-leaf vallisneria		Wide-leaf vallisneria	
Herbicide treatment	Shoots[1]	Roots	Shoots	Roots
B10	2.76 ± 0.25 a–c	2.21 ± 0.52 a–c	—	—
D100	2.17 ± 0.09 b–e	2.09 ± 0.41 a–d	—	—
E500	1.42 ± 0.26 d–f	1.38 ± 0.26 c–f	0.92 ± 0.52 c–e	0.73 ± 0.30 ef
F50	3.03 ± 0.23 ab	2.88 ± 0.55 a	2.14 ± 0.46 ab	2.17 ± 0.48 ab
I50	3.04 ± 0.84 ab	2.23 ± 0.67 a–c	1.69 ± 0.36 a–c	1.00 ± 0.23 c–e
P5	3.61 ± 1.13 a	2.50 ± 0.54 a–c	2.50 ± 0.25 a	1.67 ± 0.24 a–c
B10 + D100	1.76 ± 0.47 c–f	1.33 ± 0.25 c–f	—	—
B10 + E500	0.98 ± 0.20 ef	0.87 ± 0.07 ef	—	—
B10 + F50	2.96 ± 0.55 a–c	1.88 ± 0.43 a–f	—	—
B10 + I50	2.85 ± 0.10 a–c	2.30 ± 0.14 a–c	—	—
B10 + P5	2.94 ± 0.23 a–c	1.98 ± 0.28 a–e	—	—
D100 + F50	2.17 ± 0.17 b–e	2.04 ± 0.21 a–e	—	—
D100 + P5	2.58 ± 0.42 a–d	1.95 ± 0.10 a–e	—	—
E500 + F50	0.99 ± 0.09 ef	1.00 ± 0.14 d–f	0.56 ± 0.04 de	0.71 ± 0.05 d–f
E500 + I50	1.29 ± 0.50 ef	1.66 ± 0.63 b–f	0.37 ± 0.12 e	0.36 ± 0.04 f
E500 + P5	0.55 ± 0.29 f	0.75 ± 0.30 f	1.18 ± 0.36 b–e	1.06 ± 0.29 b–e
F50 + I50	2.85 ± 0.14 a–c	2.01 ± 0.29 a–e	1.48 ± 0.48 a–d	1.04 ± 0.16 b–e
F50 + P5	2.69 ± 0.66 a–c	2.64 ± 0.51 ab	2.03 ± 0.47 ab	1.54 ± 0.19 a–d
I50 + P5	3.01 ± 0.18 a–c	2.07 ± 0.53 a–e	1.21 ± 0.18 b–e	1.08 ± 0.13 b–e
Control	3.34 ± 0.20 ab	2.93 ± 0.63 a	2.21 ± 0.49 ab	3.05 ± 0.87 a

[1]Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD ($P \leq 0.05$); $n = 3$.

that failed to reduce root biomass 11 WAT. All other treatments decreased wide-leaf vallisneria root biomass by 65 to 88% compared to the nontreated control, with minimal differences among these treatments.

None of the individual or combination treatments completely eliminated biomass of these 2 nontarget submersed plants, and only few treatments reduced shoot or root biomass to below pretreatment levels. Several of the treated plants were beginning to produce new leaves from older stems and rhizomes as well as new stolons (daughter plants) at 5 to 11 WAT. Many of the contact-plus-systemic combinations evaluated in this research also reduced hydrilla biomass 75 to 100% compared to the nontreated control in previous growth chamber experiments (C. R. Mudge, unpub. data).

Although no statistical comparison was made across plants, both narrow- and wide-leaf vallisneria responded similarly to most herbicide treatments. The biotypes evaluated in this study are just 2 of many vallisneria biotypes found throughout the United States, and DNA marker research is currently being conducted at the University of Florida to determine the genetic relationship among vallisneria biotypes throughout Florida (L. A. Gettys, pers. comm.). It is possible that other accessions of vallisneria from various lakes may respond differently to the herbicide combinations tested in these experiments. For example, the submersed aquatic plant cabomba (*Cabomba caroliniana* Gray) has 3 distinct phenotypes. The 3 cabomba phenotypes exhibited distinctly different photosynthetic responses to a wide range of contact and systemic herbicides in growth chamber studies (Bultemeier et al. 2009), and 1 of these phenotypes has recently become problematic in the northern regions of the United States. In addition, 2 biotypes of alligatorweed (*Alternanthera philoxeroides* [Mart.] Griseb.) (slender-stem and broad-stem) responded differently to quinclorac (3,7-dichloro-8-quinolinecarboxylic acid), with the slender-stem biotype more susceptible than the broad-stem biotype (Kay 1992). The aforementioned research and the vallisneria research demonstrate the need for further evaluation of other plant species with different biotypes, ecotypes, or phenotypes that could have differential responses to herbicide treatments.

Experiment 2

Bispyribac-sodium (B10), flumioxazin (F50), and penoxsulam (P5) applied alone to American pondweed did not reduce shoot biomass compared to the nontreated control 8 WAT (Table 3); however, individual applications of diquat (D100), imazamox (I50), and endothall (E500) reduced shoot biomass by 59, 70, and 96%, respectively (Table 3). Combinations that included diquat, endothall, or imazamox were not significantly different from these products alone; therefore, the injury is likely attributed to their activity rather than the herbicide combination, despite the relatively low concentrations used in this experiment. Concentrations of diquat and endothall in these studies were almost half those used in previous experiments in which American pondweed shoot biomass significantly declined (Skogerboe and Getsinger 2001, Skogerboe et al. 2006). All 13 combination treatments except F50 + P5 were injurious to American pondweed shoots. Combinations of bispyribac-sodium with flumioxazin (B10 + F50) and penoxsulam (B10 + P5) reduced American pondweed shoot biomass by approximately 50%, whereas combinations of D100 + P5 and F50 + I50 decreased shoot biomass by 60%. All other combinations reduced shoot biomass > 85%.

The only stand-alone herbicide treatment that decreased root biomass compared to the nontreated control was E500, which reduced American pondweed root dry weight by

Table 3. Effect of the aquatic herbicides bispyribac-sodium at 10 μg ai L^{-1} (B10), diquat at 100 μg ai L^{-1} (D100), endothall at 500 μg ai L^{-1} (E500), flumioxazin at 50 μg ai L^{-1} (F50), imazamox at 50 μg ai L^{-1} (I50), and penoxsulam at 5 μg ai L^{-1} (P5) alone and in combination on American pondweed, Illinois pondweed, and coontail shoot and root dry weight (g, mean ±1 SE) 8 wk after treatment.

	American pondweed		Illinois pondweed		Coontail
Herbicide treatment	Shoots[1]	Roots	Shoots	Roots	shoots
B10	4.15 ± 1.97 ab	1.00 ± 0.39 a–c	0.48 ± 0.22 cd	0.24 ± 0.10 a–d	3.00 ± 0.37 bc
D100	2.28 ± 1.57 b–d	0.66 ± 0.45 a–e	0.40 ± 0.40 cd	0.21 ± 0.18 a–d	0.00 ± 0.00 d
E500	0.21 ± 0.21 cd	0.14 ± 0.08 de	0.07 ± 0.03 d	0.05 ± 0.00 d	1.13 ± 0.97 d
F50	4.01 ± 1.31 ab	1.36 ± 0.54 a	1.35 ± 0.39 ab	0.43 ± 0.11 a	0.89 ± 0.46 d
I50	1.66 ± 0.42 b–d	0.59 ± 0.10 b–e	0.80 ± 0.19 bc	0.35 ± 0.11 ab	2.61 ± 0.54 c
P5	4.00 ± 1.89 ab	1.26 ± 0.44 ab	0.43 ± 0.14 cd	0.14 ± 0.04 b–d	3.94 ± 1.50 a–c
B10 + D100	0.91 ± 0.59 cd	0.35 ± 0.20 c–e	0.07 ± 0.07 d	0.05 ± 0.01 d	0.06 ± 0.02 d
B10 + E500	0.00 ± 0.00 d	0.06 ± 0.02 e	0.01 ± 0.01 d	0.05 ± 0.00 d	0.07 ± 0.07 d
B10 + F50	2.83 ± 0.50 bc	0.84 ± 0.19 a–d	0.84 ± 0.60 bc	0.32 ± 0.24 a–c	0.62 ± 0.32 d
B10 + I50	0.63 ± 0.41 cd	0.30 ± 0.14 c–e	0.34 ± 0.17 cd	0.13 ± 0.05 b–d	2.84 ± 0.34 c
B10 + P5	2.55 ± 0.90 b–d	0.65 ± 0.26 a–e	0.52 ± 0.23 cd	0.23 ± 0.12 a–d	5.22 ± 0.42 a
D100 + F50	0.52 ± 0.13 cd	0.18 ± 0.08 de	0.06 ± 0.02 d	0.07 ± 0.01 cd	0.00 ± 0.00 d
D100 + P5	2.11 ± 0.35 b–d	0.84 ± 0.06 a–d	0.22 ± 0.12 cd	0.12 ± 0.04 b–d	0.09 ± 0.02 d
E500 + F50	0.07 ± 0.06 d	0.09 ± 0.04 e	0.13 ± 0.08 d	0.06 ± 0.01 d	0.00 ± 0.00 d
E500 + I50	0.00 ± 0.00 d	0.05 ± 0.02 e	0.01 ± 0.01 d	0.06 ± 0.02 d	0.05 ± 0.05 d
E500 + P5	0.00 ± 0.00 d	0.04 ± 0.02 e	0.02 ± 0.01 d	0.04 ± 0.01 d	0.09 ± 0.05 d
F50 + I50	2.12 ± 0.80 b–d	0.60 ± 0.13 b–e	0.51 ± 0.22 cd	0.20 ± 0.03 a–d	0.55 ± 0.25 d
F50 + P5	3.86 ± 1.63 ab	1.25 ± 0.44 ab	0.23 ± 0.03 cd	0.11 ± 0.03 b–d	0.66 ± 0.29 d
I50 + P5	0.73 ± 0.18 cd	0.13 ± 0.06 de	0.24 ± 0.14 cd	0.08 ± 0.03 cd	2.52 ± 0.83 c
Control	5.59 ± 1.27 a	1.24 ± 0.07 ab	1.63 ± 0.19 a	0.42 ± 0.12 a	4.82 ± 1.18 ab

[1]Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD ($P \leq 0.05$); $n = 3$.

> 90% by the conclusion of the study (Table 3). Herbicide combinations of B10 + F50, B10 + P5, D100 + P5, F50 + I50, and F50 + P5 were similar to the control, whereas all other herbicide combinations significantly reduced root biomass by ≥ 75%.

All herbicides and herbicide combinations decreased Illinois pondweed shoot biomass by 8 wk, except flumioxazin applied alone (F50); however, combinations of flumioxazin with other herbicides reduced shoot biomass by ≥ 50% (Table 3). Illinois pondweed root biomass was not affected by many of the herbicides applied alone, including B10, D100, F50, and I50, and herbicide combinations that were similar to the nontreated control were B10 + F50, B10 + P5, and F50 + I50. Herbicides and herbicide combinations that reduced Illinois pondweed root biomass by 60 to 85% were B10 + I50, D100 + P5, and F50 + P5 (Table 3). All other treatments reduced root dry weight by ≥ 85%.

Similar to Illinois pondweed, coontail shoots were very susceptible to stand-alone and combination herbicide treatments evaluated in this study (Table 3). Bispyribac-sodium (B10), penoxsulam (P5), and the combination of these 2 herbicides (B10 + P5) were the only treatments that failed to reduce shoot biomass 8 WAT. Treatments of I50, B10 + I50, and I50 + P5 decreased coontail shoot biomass by 41 to 48 %. All other treatments decreased coontail shoot biomass by ≥ 75%, with no differences between these treatments.

Unlike the previous experiment with narrow- and wide-leaf vallisneria, many individual or combination treatments severely injured American pondweed, Illinois pondweed, and coontail. Endothall, diquat, and combinations with these herbicides reduced shoot or root biomass by 90% or greater. In general, American and Illinois pondweed recovery was observed as early as 4 WAT when treated with B10, F50, P5, or F50 + P5. Pondweeds exposed to the other individual or combination treatments were declining in health or severely injured at the conclusion of the experiment. Conversely, coontail was either highly tolerant or highly susceptible to the herbicide or herbicide combination treatments.

Based on these data, American and Illinois pondweeds are likely to be impacted by these newly registered herbicides or combinations of these products with older chemistries. Other native pondweed species could be negatively impacted by these herbicide treatments and further research should be conducted to determine pondweed selectivity. In southern water bodies where hydrilla is managed, these native species usually germinate at the same time as hydrilla and would therefore be exposed to herbicide treatments employed for hydrilla control. On the other hand, native pondweeds in northern U.S. lakes typically germinate after noxious plant species such as curlyleaf pondweed (*Potamogeton crispus* L.) and Eurasian watermilfoil (*Myriophyllum spicatum* L.) have completed most of their life cycle; as a result, the native species are typically not exposed to herbicide treatments (Poovey et al. 2002).

Reducing use concentrations to approximately one-half or one-fourth the current recommended rate, when used in combination, will reduce herbicide exposure for nontarget plants, which will likely result in less injury to some nontarget species. Although the benefit of combining fast- and slow-acting herbicides, such as endothall and penoxsulam, has been a reduction in the exposure period required to control hydrilla, combinations of contact-plus-systemic herbicides were more injurious to the nontarget plant species tested in these experiments than combinations of systemic-plus-systemic herbicides.

Systemic-plus-systemic combinations (bispyribac-sodium, imazamox, or penoxsulam) provided no additional injury to vallisneria biotypes compared to these products applied alone; however, negative effects of these combinations were observed on pondweeds and coontail. Although the ALS herbicides have shown promise for hydrilla control, the prolonged exposure times needed to manage weeds may injure some nontarget SAV and emergent plant species. Additionally, there are concerns regarding the development of ALS herbicide resistance in aquatic weed control (Richardson 2008). Given these factors, other combinations of systemic-plus-systemic herbicides should be evaluated to determine their effects on hydrilla and non-target SAV.

In conclusion, these data indicate that the narrow-leaf and wide-leaf biotypes of vallisneria were minimally impacted by herbicide combinations. In addition, both taxa responded similarly to herbicide treatments. Other vallisneria biotypes in Florida or other regions of the United States may respond differently to these combinations, but vallisneria present in mixed SAV stands with hydrilla should survive herbicide treatments. Conversely, American pondweed, Illinois pondweed, and coontail were adversely affected by many of the herbicide combinations evaluated in these experiments, and the different species of pondweed responded similarly to the herbicide treatments. It is uncertain whether these plants would survive exposure to herbicides used to control hydrilla or other invasive weeds, but recovery in the following season may occur because pondweeds reproduce by seed (Godfrey and Wooten 1979).

Herbicides and their combinations studied in these experiments should be tested in outdoor mesocosms to allow for herbicide degradation in a seminatural system, which could verify the results of these growth chamber studies. Future research goals include evaluation of the effects of these combinations on other nontarget submersed and emergent plant species. Results of these studies will be useful for aquatic plant management in water bodies where hydrilla and nontarget plants coexist and can provide information regarding which herbicide combinations should be used to control hydrilla while maintaining a healthy native plant community.

SOURCES OF MATERIALS

[1]Wide-leaf vallisneria, Suwannee Labs, 1205 SW King St., Lake City, FL 32024.

[2]Black Kow® topsoil, Black Gold Compost Co., PO Box 190, Oxford, FL 34484.

[3]Osmocote®, The Scotts Company, 14111 Scottslawn Rd., Marysville, OH 43041.

[4]Tradewind®, Valent Corporation, PO Box 8025, Walnut Creek, CA 94596.

[5]Reward®, Syngenta Professional Products, 3411 Silverside Rd., Wilmington, DE 19810.

[6]Aquathol® K, United Phosphorus Inc., 630 Freedom Business Center, Suite 402, King of Prussia, PA 19406.

[7]Clipper®, Valent USA Corporation, PO Box 8025, Walnut Creek, CA 94596.

[8]Clearcast®, SePRO Corporation, 11550 North Meridian St., Suite 600, Carmel, IN 46032.

[9]Galleon®, SePRO Corporation, 11550 North Meridian St., Suite 600, Carmel, IN 46032.

[10]YSI® Model 556, handheld multi-parameter probe, YSI Inc., 1700/1725 Brannum Lane, Yellow Springs, OH 45387.

ACKNOWLEDGEMENTS

Support for this project was provided by the U.S. Army Corps of Engineers Aquatic Plant Control Research Program (APCRP). Permission to publish this information was granted by the Chief of Engineers. Portions of this data were published as an APCRP Technical Note (APCPR-CC-17). The author would like to thank Angela Poovey, Morgan Sternberg, James Smith, Katharine DeRossette, and Heidi Sedivy for technical assistance as well as Mike Netherland, Lyn Gettys, and LeeAnn Glomski for plant collection. Manuscript reviews were provided by Angela Poovey and Lyn Gettys. 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 products.

LITERATURE CITED

Bultemeier BW, Netherland MD, Ferrell JA, Haller WT. 2009. Differential herbicide response among three phenotypes of *Cabomba caroliniana*. Invasive Plant Sci. Manag. 2:352–359.

Godfrey RK, Wooten JW. 1979. Aquatic and Wetland Plants of Southeastern United States: Monocotyledons. University of Georgia Press, Athens, GA. 712 pp.

Heilman MA, Koschnick TJ, Tarver DP, Hulon CM, Seacrist C. 2009. Penoxsulam and Endothall Combination for Control of *Hydrilla verticillata*. http://www.apms.org/programs/2009program.pdf. Accessed December 5, 2011.

Katagi T. 2003. Hydrolysis of n-phenylimide herbicide flumioxazin and its anilic acid derivative in aqueous solutions. J. Pestic. Sci. 28:44–50.

Kay SH. 1992. Response of two alligatorweed biotypes to quinclorac. J. Aquat. Plant Manag. 30:35–40.

Koschnick TJ, Netherland MD, Haller WT. 2007. Effects of three ALS-inhibitors on five emergent native plant species in Florida. J. Aquat. Plant Manag. 45:47–51.

Mossler M, Fishel F. 2007. Non-food Related Actions. Chemically Speaking. http://www.pested.ifas.ufl.edu/newsletters/march2007/Chemically__Speaking__March__2007__original.pdf. Accessed December 12, 2011.

Mossler M, Fishel F, Whidden N. 2006. Pesticide Registration and Actions. Chemically Speaking. http://pested.ifas.ufl.edu/newsletters/april2006/Chemically%20Speaking%20April%202006.pdf. Accessed December 12, 2011.

Mudge CR. 2007. Characterization of Flumioxazin as an Aquatic Herbicide. Ph.D dissertation. University of Florida, Gainesville, FL. 120 pp.

Mudge CR, Haller WT. 2010. Effect of pH on submersed aquatic plant response to flumioxazin. J. Aquat. Plant Manag. 48:30–34.

Poovey AG, Skogerboe JG, Owens CS. 2002. Spring treatment of diquat and endothall for curlyleaf pondweed control. J. Aquat. Plant Manag. 40:63–67.

Richardson RJ. 2008. Aquatic plant management and the impact of emerging herbicide resistance issues. Weed Technol. 22:8–15.

Senseman SA (ed.). 2007. Herbicide Handbook. 9th ed. Weed Science Society of America, Lawrence, KS. 458 pp.

Skogerboe JG, Getsinger KD. 2001. Endothall species selectivity evaluation: Southern latitude aquatic plant community. J. Aquat. Plant Manag. 39:129–135.

Skogerboe JG, Getsinger KD. 2002. Endothall species selectivity evaluation: Northern latitude aquatic plant community. J. Aquat. Plant Manag. 40:1–5.

Skogerboe JG, Getsinger KD, Glomski LM. 2006. Efficacy of diquat on submersed plants treated under simulated flowing water conditions. J. Aquat. Plant Manag. 44:122–125.

Smart RM, Barko JW. 1985. Laboratory culture of submersed freshwater macrophytes on natural sediments. Aquat. Bot. 21:251–263.