# Response of Eurasian and Hybrid Watermilfoil to Low Use Rates and Extended Exposures of 2,4-D and Triclopyr

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# **ABSTRACT**

Eurasian watermilfoil (Myriophyllum spicatum L.) and hybrid watermilfoil (Myriophyllum spicatum  $\times$  M. sibiricum) are invasive submersed plants that coexist in the Great Lakes and Pacific Northwest regions. The auxin-mimic herbicides triclopyr (3,5,6-trichloro-2-pyridinyloxyacetic acid) and 2,4-D (2,4-dichlorophenoxy acetic acid) are commonly used to control these species at recommended use rates of 1.5 to 2.5 mg L<sup>-1</sup> and 2.0 to 4.0 mg L<sup>-1</sup>, respectively. Recent field data suggest that following some early season applications, control of watermilfoil may be related to extended exposures to low concentrations of these herbicides. Two greenhouse studies were conducted to determine the efficacy of lower concentrations and extended exposures of 2,4-D and triclopyr on both Eurasian and hybrid watermilfoil. Concentrations evaluated included 0, 25, 70, 100 and 250 µg L<sup>1</sup> 2,4-D amine or triclopyr. At 7 weeks after treatment, Eurasian watermilfoil biomass was significantly reduced with all rates of 2,4-D and triclopyr in both studies. Triclopyr at rates of 70 to 250 µg L<sup>-1</sup> controlled hybrid watermilfoil by 88 to 100% in study 1, while all rates of triclopyr resulted in 100% control of hybrid watermilfoil in study 2. The 2,4-D treatments of 25 to 100 µg L<sup>-1</sup> were not different from the untreated control, whereas the 250 µg L<sup>-1</sup> treatment resulted in a 95% biomass reduction for hybrid milfoil in study 1. In study 2, 2,4-D at 70 ug L<sup>-1</sup> and higher controlled hybrid watermilfoil by 93 to 100%. Results from these studies indicate that low rates and extended exposures of both triclopyr and 2,4-D can be effective at controlling both Eurasian and hybrid watermilfoil; however, different hybrid watermilfoil accessions may respond differently to low concentrations of the auxin-mimic herbicides. The ability to utilize low concentrations of these compounds in areas with limited water exchange may represent a cost-effective, selective, and large-scale treatment strategy not fully utilized today.

Key words: aquatic herbicides, chemical control, Myriophyllum spicatum, Myriophyllum spicatum × Myriophyllum sibiricum.

### INTRODUCTION

Triclopyr (3,5,6-trichloro-2-pyridinyloxyacetic acid) and 2.4-D (2.4-dichlorophenoxy acetic acid) are commonly used to provide selective milfoil control in the field, with recommended rates ranging from 2 to 4 mg L<sup>-1</sup> 2,4-D or 1.5 to 2.5 mg L<sup>1</sup> triclopyr (Carpenter et al. 1988, Getsinger et al. 1997, Parsons et al. 2001, Poovey et al. 2004). Past research has focused on operational use rates and short exposures (up to 72) h) for both 2,4-D and triclopyr (Elliston and Steward 1972, Green and Westerdahl 1990, Netherland and Getsinger 1992). Prolonged exposure to lower rates of both herbicides could potentially provide good watermilfoil control; however, extended exposure time scenarios have not been established for these use patterns (Green and Westerdahl 1990). Recent field residue data suggest that following some early season large-scale applications, widespread control of watermilfoil may be related to extended exposures to low concentrations of these herbicides (Asplund 2009).

The hybridization of Eurasian watermilfoil (Myriophyllum spicatum L.) with the native northern watermilfoil (M. sibiricum; Moody and Les 2002, 2006) has resulted in anecdotal reports of differences in response between hybrid (M. spicatum × M. sibiricum) and Eurasian watermilfoil to herbicide applications. Several studies have been conducted to address this concern, and to date, there has been no evidence that Eurasian and hybrid watermilfoil differ in their response to operational use rates of fluridone, 2,4-D or triclopyr (Poovey et al. 2007, Slade et al. 2007). In studies conducted by Poovey et al. 2007 and Slade et al. 2007, use rates >1 mg L<sup>-1</sup> and exposures ranging from 6 to 72 h were evaluated. Poovey et al. (2007) also found that rates ≥0.27 mg L<sup>1</sup> triclopyr or 2,4-D with a 24-h exposure reduced shoot biomass by 95 to 100% for both Eurasian and hybrid watermilfoil. Differences, however, were noted between both species at sublethal rates of both 2,4-D and triclopyr.

The objectives of these studies were to (1) establish new concentration exposure time parameters for low use rates of 2,4-D and triclopyr and (2) determine if differences exist for these use patterns between 2,4-D and triclopyr for control of Eurasian and hybrid watermilfoil.

## **MATERIALS AND METHODS**

Two studies were conducted in a greenhouse at the U.S. Army Engineer Research and Development Center, Lewisville Aquatic Ecosystem Research Facility (LAERF) in Lewisville, Texas. Studies were conducted with Eurasian

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watermilfoil obtained from an LAERF pond and hybrid watermilfoil obtained from White Bear Lake (study 1) and Otter Lake (study 2) in Minnesota. For both studies, two apical tips of watermilfoil (15 cm) were planted in plastic pots (750 ml), filled with LAERF pond sediment amended with 3 g L¹ Osmocote (16-8-12). Pots were topped with a 1-cm layer of sand, and four pots were placed in each aquarium (66 L). Aquariums were filled with alum-treated Lake Lewisville water and were situated in 1000-L fiberglass tanks filled with water. Water temperatures in the aquariums were maintained at 24 C by either aquarium heaters or by circulating water through a Pacific Coast Imports C-1000 chiller.

In both studies, aquariums were treated with herbicide when watermilfoil had grown to the water's surface. Treatments included static exposures to 0, 25, 70, 100, and 250 µg L <sup>1</sup> acid equivalent (ae) 2,4-D amine (DMA 4 IVM, Dow Agro-Sciences, Indianapolis, IN) or triclopyr (Renovate 3, SePRO Corporation, Carmel, IN). Treatments were replicated three times. Water samples were collected 1, 7, 21, 35, and 49 days after treatment (DAT) on 25 and 100 µg L<sup>-1</sup> treated tanks, and residues were analyzed via enzyme-linked immunosorbent assay (ELISA) technique. At 7 weeks after treatment (WAT), all viable shoot biomass was harvested and dried at 65 C. All data were subjected to a one-way analysis of variance (ANOVA). Due to statistical differences, data from study 1 and 2 were not pooled. Where treatment differences were detected, a posthoc test was conducted using the Student-Newman-Keuls method (SNK;  $\alpha = 0.05$ ). When necessary, data were square root transformed to meet the assumptions of normality and equal variance. Nontransformed data are presented.

# **RESULTS AND DISCUSSION**

Results of the residue analysis indicate that triclopyr, which degrades via photolysis (WSSA 2007), did not degrade over the 7-week exposure period (data not shown). However 2,4-D, which degrades microbially (WSSA 2007), did degrade by about 50% through 21 DAT, and residues continued to decline for the remainder of the exposure period.

All rates of 2,4-D amine and triclopyr reduced Eurasian watermilfoil shoot biomass compared to the controls in both studies (Figure 1A and B). Triclopyr reduced biomass 90 to 100% in both studies, whereas 2,4-D amine reduced biomass 42 to 100% in study 1 (Figure 1A) and 50 to 100% in study 2 (Figure 1B). Growth of the untreated Eurasian watermilfoil control plants was much greater in study 1; however, the trends in plant control remained similar for both studies. Other than the lowest rate of triclopyr resulting in greater efficacy than the lowest rate of 2,4-D, there were no differences noted between the two herbicides.

Hybrid watermilfoil showed some variation in response to treatment between the two studies. In study 1, biomass was reduced 95% at 250  $\mu g~L^{\rm 1}$  2,4-D amine and 88 to 100% for 70 to 250  $\mu g~L^{\rm 1}$  triclopyr (Figure 2A). Rates of 25 to 100  $\mu g~L^{\rm 1}$  2,4-D amine and 25  $\mu g~L^{\rm 1}$  triclopyr did not reduce biomass (Figure 2A). In study 2, however, 70 to 250  $\mu g~L^{\rm 1}$  2,4-D amine reduced biomass 93 to 100%, and all rates of triclopyr resulted in 100% control of hybrid watermilfoil (Figure 2B).

The hybrid watermilfoil used in these studies originated from two different lakes, and these results suggest that differ-

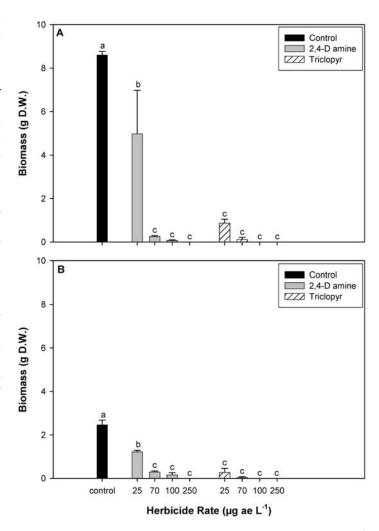


Figure 1. Mean ( $\pm$ SE) dry weight (g) of Eurasian watermilfoil biomass 7 weeks after treatment with 2,4-D amine and triclopyr in (A) study 1 and (B) study 2. Each bar represents the average of three replicate treatments. Bars sharing the same letter do not significantly differ from each other. Data were subjected to a one-way analysis of variance, and means were separated using the Student-Newman-Keuls Method (SNK;  $\alpha$  = 0.05).

ent accessions of hybrid watermilfoil may respond differently to low use rates of these herbicides. Because biomass of the untreated hybrid controls was similar for the two studies, this likely rules out differences in growth rates as an explanation for these results. These data also provide some support to the many anecdotal reports of differences between Eurasian and hybrid response to herbicides. Nonetheless, it should be noted that in these studies we were evaluating use rates that are 6 to 16 times lower than the recommended label rates. Further testing is being conducted to determine if differences in hybrid or milfoil accession is a factor in the effectiveness of low use rate applications of the auxin mimic herbicides.

Results of these studies indicate that long-term exposures of Eurasian and hybrid watermilfoil to low rates of 2,4-D or triclopyr can provide control. These data demonstrate technical feasibility of this approach; however, utilizing these products in this manner would require significant changes in use patterns and treatment timing. Current early season, low

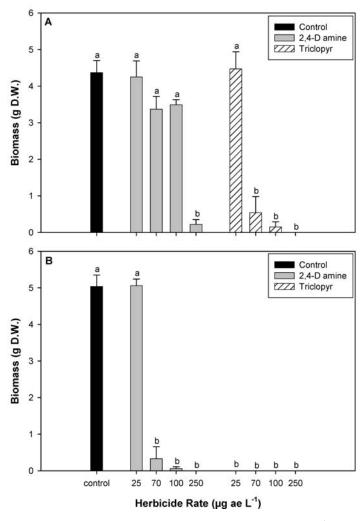


Figure 2. Mean ( $\pm$ SE) dry weight (g) of hybrid watermilfoil biomass 7 weeks after treatment with 2,4-D amine and triclopyr in (A) study 1 and (B) study 2. Each bar represents the average of three replicate treatments. Bars sharing the same letter do not significantly differ from each other. Data were subjected to a one-way analysis of variance, and means were separated using the Student-Newman-Keuls Method (SNK;  $\alpha$  = 0.05).

rate fluridone applications for milfoil control in northern tier states (Getsinger et al. 2001, 2002) may provide a useful template for designing low rate extended exposure treatments with either 2,4-D or triclopyr. Due to differences in degradation pathways between the two products, further studies need to be conducted to determine effective exposure periods between 72 hours and 4 weeks, which would aid in making product-specific recommendations. The impact of these types of applications on selectivity has not been evaluated, and while both products are recognized as being selective for numerous submersed monocots, changes in selectivity patterns (whether an increase or decrease) still need to be documented.

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