

## NOTES

# Response of waterlily, spatterdock, and hardstem bulrush to liquid and granular triclopyr treatments

LEEANN M. GLOMSKI AND MICHAEL D. NETHERLAND\*

### INTRODUCTION

The use of herbicides for control of Eurasian watermilfoil (*Myriophyllum spicatum* L.) can result in a wide range of concentrations and exposure times for both target and nontarget vegetation (Getsinger et al. 2000, 2002, Netherland et al. 2002, Poovey et al. 2004, Wersal et al. 2010). Recent monitoring in lakes of the upper Midwest suggest that large-scale treatments with the auxin mimic herbicides 2,4-D (2,4-dichlorophenoxy acetic acid) and triclopyr (3,5,6-trichloro-2-pyridinyloxyacetic acid) can result in extended exposures that may last from 1 to several weeks (Asplund 2009). Moreover, recent mesocosm trials have demonstrated that extended exposures to low concentrations of auxin mimics can provide an effective strategy for controlling Eurasian watermilfoil (Glomski and Netherland 2010). Treatment strategies that provide extended exposures to lower concentrations or granular applications that are purported to improve efficacy against target plants also raise questions about the response of native plants to these treatments. In particular, resource managers in the northern states have expressed specific concerns regarding the response of native waterlily (*Nymphaea odorata* Aiton), spatterdock (*Nuphar lutea* (L.) Sm), and hardstem bulrush (*Schoenoplectus acutus* Muhl. ex Bigelow) to large-scale treatments with auxin-mimic herbicides. These species grow in proximity to or intermixed with Eurasian watermilfoil and they are typically considered valuable plants that provide food and cover for macroinvertebrates and fish (Borman et al. 1997). Moreover, waterlily, spatterdock, and hardstem bulrush are visually prominent emergent species and extended or short-term observations of significant visual injury to these emerged plants can result in negative feedback regarding the selective nature of Eurasian watermilfoil treatments by both resource agency and private stakeholder groups. As new large-scale strategies are

developed for applying auxin-mimic herbicides, it is important to determine the likelihood of both visual injury and potential for subsequent control of key emergent plant species.

Prior research conducted by Glomski and Nelson (2008) evaluating short-term exposures (24 h) to submersed applications of triclopyr and 2,4-D ester on spatterdock and waterlily demonstrated that impacts were largely growth regulating at 6 wk after treatment (WAT) at concentrations ranging from 0.25 to 2.5 mg L<sup>-1</sup>. Injury symptoms included leaf curling and petiole elongation. A subsequent trial by Glomski et al. (2009) found limited injury after a 24-h exposure to triclopyr and 2,4-D ester on American bulrush (*Schoenoplectus americanus* (Pers.) Volkart ex Schinz & R. Keller), whereas soft-stem bulrush (*Schoenoplectus tabernaemontani* (C.C. Gmel.) Palla) was more susceptible by 6 WAT to both herbicides at concentrations ranging from 0.25 to 2.5 mg L<sup>-1</sup>. Glomski et al. (2009) also exposed spatterdock to triclopyr, 2,4-D amine, and 2,4-ester concentrations ranging from 0.25 to 0.75 mg L<sup>-1</sup> for a 4-wk exposure period. Although epinasty was observed early in the study, the authors found no reduction in shoot or root biomass at 4 WAT. These prior studies provided evidence of the recovery potential of waterlily and two species of bulrush after short-term exposures to triclopyr and 2,4-D. Given the observations of longer-term exposures to auxin mimics associated with large-scale treatments (Asplund 2009), there are additional exposure scenarios that require more detailed evaluation. As noted above, many of these large block, whole-bay, or whole-lake treatments can result in exposure to herbicides for several days to several weeks. The objective of this study was to determine the effect of granular and liquid triclopyr formulations on waterlily, spatterdock, and hardstem bulrush under a broad range of concentrations and exposure times.

### MATERIALS AND METHODS

This study was conducted at the U.S. Army Engineer Research and Development Center's Lewisville Aquatic Ecosystem Research Facility (LAERF) located in Lewisville, TX. One rhizome section of waterlily, spatterdock, or hardstem bulrush was planted into 3.78-L pots filled with

\*First author: Research Biologist, U.S. Army Engineer Research and Development Center, Lewisville Aquatic Ecosystem Research Facility, 201 E Jones St, Lewisville, TX 75057. Second author: Research Biologist, U.S. Army Engineer Research and Development Center, Center for Aquatic and Invasive Plants, Gainesville, FL 32653. Corresponding author's E-mail: LeeAnn.M.Glomski@usace.army.mil. Received for publication July 11, 2013 and in revised form April 23, 2014.

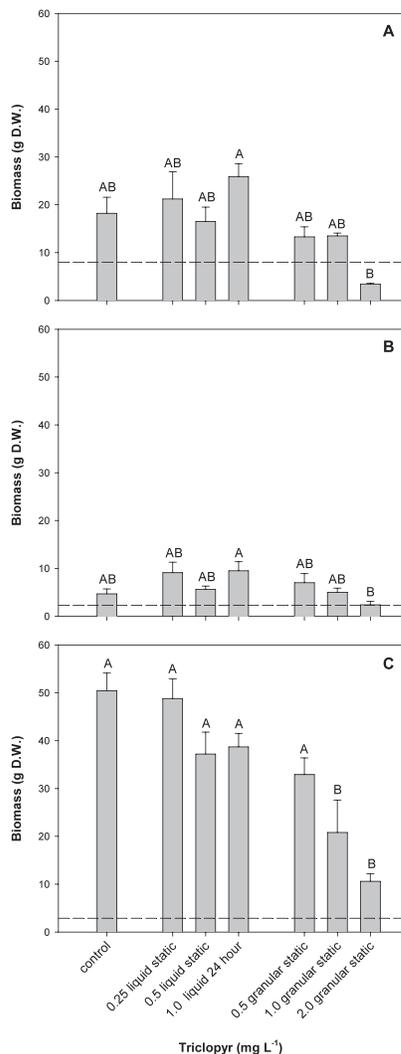


Figure 1. Mean ( $\pm$  SE) dry weight of (A) waterlily, (B) spatterdock, and (C) hardstem bulrush 42 d after the June triclopyr treatments. Bars sharing the same letter do not significantly differ from each other (Student–Newman–Keuls method;  $\alpha = 0.05$ ). The dashed line represents pretreatment biomass.

LAERF pond sediment amended with  $3 \text{ g L}^{-1}$  osmocote<sup>1</sup> (16–8–12). Three pots of waterlily<sup>2</sup> and spatterdock<sup>2</sup> and two pots of hardstem bulrush<sup>3</sup> were placed into each 760-L Rubbermaid tank. Tanks were filled with Lake Lewisville water to a depth of 50 cm and plants were allowed to grow for 4 wk before the first treatment and 6 wk before the second. The tanks were under a structure covered with 30% shade fabric.

The first treatment took place on 20 June 2011 (June treatment) and included 6-wk static exposures of 0.25 and  $0.50 \text{ mg L}^{-1}$  liquid triclopyr<sup>4</sup>, and 0.50, 1.00 and  $2.00 \text{ mg L}^{-1}$  granular triclopyr<sup>5</sup>. A 24-h exposure of  $1.00 \text{ mg L}^{-1}$  liquid and an untreated control were also included. A second set of tanks was treated on 5 July 2011 (July treatment) and included a  $1.50 \text{ mg L}^{-1}$  granular for 24-h, 48-h, and 4-wk static exposures. After each exposure time, tanks were drained and refilled with untreated water to remove aqueous herbicide residues.

Treatments were randomly assigned to tanks and replicated three times. Water samples were collected from selected treatment tanks at 1, 2, 3, 7, 10, 15, and 21 d after the static treatments to provide estimated half-lives of triclopyr applications. Triclopyr was analyzed via use of an enzyme-linked immunoassay<sup>6</sup> (Fox et al. 2002). Triclopyr concentration data were subjected to regression analysis and half-lives were estimated for the static treatments using an exponential decay model (Fox et al. 1993). On 1 August 2011 all viable shoot biomass from both the June (42 d after treatment [DAT]) and July (28 DAT) treatments was harvested and dried at  $65 \text{ C}$  to a constant weight. Although the June application provided a longer period of recovery compared with the July treatment, the decision to harvest at a single date allowed us to quantify if biomass is being affected earlier in the progression of the treatment. When necessary, data were square root transformed to meet the assumptions of normality and equal variance. All biomass data were subjected to a one-way ANOVA. Where treatment differences were detected, a post hoc test was conducted using the Student–Newman–Keuls method ( $\alpha = 0.05$ ). Nontransformed data are presented.

## RESULTS AND DISCUSSION

After the liquid triclopyr applications in June, half-lives averaged  $5.9 \pm 0.8 \text{ d}$  ( $r^2 = 0.91$ ) and the granular treatments averaged  $7.6 \pm 1.1 \text{ d}$  ( $r^2 = 0.83$ ). Half-lives in the static treatment after the granular applications in July averaged  $7.1 \pm 0.6 \text{ d}$  ( $r^2 = 0.87$ ). Concentrations at 1 DAT after the granular applications were within 15% of nominal target concentrations, suggesting an initial rapid release of triclopyr from the granules. Herbicide degradation in these studies was within the range of half-lives observed after numerous large-scale liquid and granular treatments conducted in the upper Midwest.

Initial visual symptoms on waterlily were similar for both liquid and granular triclopyr formulations. Within 2 DAT, all treated waterlilies had elongated petioles and notable leaf curling. Although some epinasty was noted after the  $0.25 \text{ mg L}^{-1}$  liquid application in June, no further injury symptoms were observed. In contrast, waterlilies treated at concentrations  $> 0.5 \text{ mg L}^{-1}$  (liquid and granular) resulted in yellowing of the majority of the surface leaves by 7 DAT. The surface leaves of waterlilies treated at the higher granular rates (1 to  $2 \text{ mg L}^{-1}$ ) with static exposures became necrotic within 2 wk of application. This type of injury can be quite visual after an operational application and can elicit concern by numerous stakeholders. Despite the early severe injury symptoms after the June treatments, there were no biomass differences detected when comparing 24 h versus static exposures and liquid versus granular applications for triclopyr concentrations ranging from 0.25 to  $2.0 \text{ mg L}^{-1}$  (Figure 1A). The  $2.0 \text{ mg L}^{-1}$  application was the only treatment that resulted in biomass that did not recover above the initial weight. The initial severity of injury and the subsequent lack of biomass reduction at 42 DAT demonstrates a strong potential for regrowth or recovery in shallow water after exposure to triclopyr. Given the

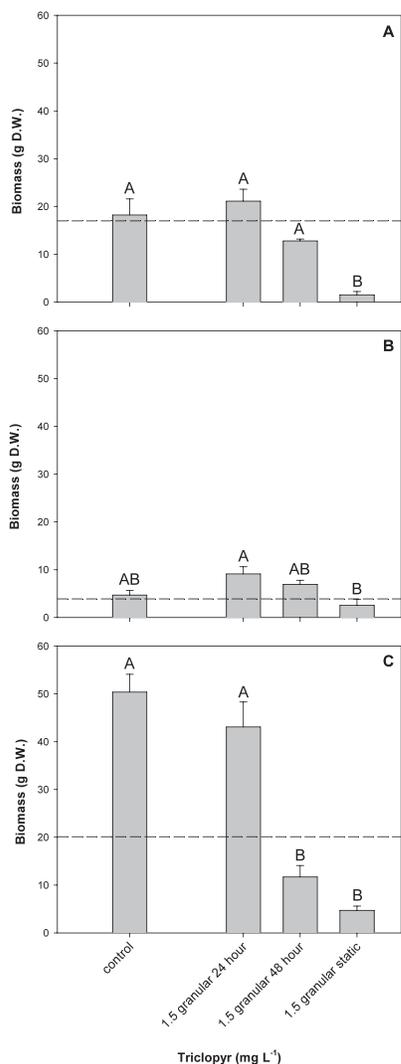


Figure 2. Mean ( $\pm$  SE) dry weight of (A) waterlily, (B) spatterdock, and (C) hardstem bulrush 28 d after the July 1.5 mg L<sup>-1</sup> triclopyr treatments. Bars sharing the same letter do not significantly differ from each other (Student–Newman–Keuls method;  $\alpha = 0.05$ ). The dashed line represents pretreatment biomass.

product half-lives noted above, triclopyr remained above 0.25 mg L<sup>-1</sup> for ~15 to 22 d after the 1.0 and 2.0 mg L<sup>-1</sup> treatments.

After the July applications at 1.5 mg L<sup>-1</sup>, waterlilies showed similar initial symptoms to the 24-h, 48-h, and static treatments. Although biomass recovered to untreated control levels after the 24- and 48-h exposures, a 92% reduction in biomass was noted after the static exposure (Figure 2A). Although all treatments resulted in severe initial injury, the recovery of waterlily biomass after the shorter-term exposures demonstrates that extended exposure to low concentrations can have impacts on the rate of recovery and initial loss of biomass. The 28-DAT harvest interval does not allow us to predict the potential for the static treatment to recover given a longer period of time, but it does suggest that extended exposure periods can result in greater initial biomass reduction.

In contrast to the initial injury symptoms in waterlilies, spatterdock leaves were only slightly curled in treated tanks through 7 DAT. All treated plants looked similar to the controls at the end of the experiment and there were no differences in final biomass (Figures 1B and 2B). Results obtained by Glomski et al. (2009) evaluating static exposures to triclopyr rates of 0.25 to 0.75 mg L<sup>-1</sup> yielded similar outcomes to our current studies and suggest a consistent response by spatterdock to an extended exposure period at even higher concentrations. The combined results from the June and July treatments suggest that spatterdock is generally more tolerant than waterlily to submersed applications of triclopyr across a broad range of concentrations, exposure times, and formulations.

Hardstem bulrush did not show strong initial injury symptoms or changes in biomass after static liquid or granular triclopyr treatments up to 0.5 mg L<sup>-1</sup> or to the 1.0 mg L<sup>-1</sup> 24-h exposure treatment (Figure 1C). In contrast, treated bulrush was beginning to turn yellow at 2 DAT in tanks treated with 1.0 and 2.0 mg L<sup>-1</sup> static granular rates. Soon after stems turned yellow, they broke off at the sediment surface. By 42 DAT, new shoots were observed in both the 1 and 2 mg L<sup>-1</sup> static granular tanks, but final biomass was reduced by 59 and 79% respectively compared with the untreated control (Figure 1C).

The July treatments of 1.5 mg L<sup>-1</sup> granular triclopyr resulted in a difference in hardstem bulrush biomass when the 24-h exposures were compared with the 48-h and static exposures (Figure 2C). Biomass of plants exposed for 48 h and a static exposure was reduced by 77 and 88% respectively. The response to the 48-h exposure was similar to the static exposure.

Results of these trials suggest that hardstem bulrush response to triclopyr is likely concentration and exposure dependent. The lack of visual injury after static treatments of 0.25 to 0.5 mg L<sup>-1</sup> would suggest some level of inherent tolerance; however, the significant visual injury to existing bulrush stems at concentrations above 1.0 mg L<sup>-1</sup> was consistent. Biomass reduction at concentrations above 1.0 mg L<sup>-1</sup> ranged from 76 to 91% (Figures 1 and 2). The shallow nature of the study tanks (0.5 m) likely favors the ability of the bulrush to recover from underground storage tissue. The influence of water depth on potential recovery by plants such as bulrush and waterlily after herbicide application was not addressed in this trial, but it deserves additional research attention.

In summary, waterlily showed strong initial visual injury symptoms to triclopyr across a range of concentrations tested. This injury did not necessarily translate to biomass reductions by 42 DAT; however, these visual symptoms can result in negative sentiment toward treatment selectivity if resource managers are conducting surveys during the height of this visual injury. Aquatic managers should be aware that visual injury symptoms on waterlily may be significant and sustained over a period of weeks after application of triclopyr. These studies suggest that significant underground storage organs can result in rapid recovery once the triclopyr exposures are removed. Spatterdock showed a much greater tolerance to triclopyr and the limited visual injury symptoms were short-lived at concentrations ranging

from 0.25 to 2.0 mg L<sup>-1</sup>. Hardstem bulrush showed strong and rapid visual injury symptoms at triclopyr concentrations of 1.0 mg L<sup>-1</sup> and greater with 48-h and static exposures. The loss of entire shoots after initial treatment indicates that recovery was occurring from rhizomes. This strong response to increased triclopyr exposure suggests that caution should be exercised when using triclopyr at higher concentrations in areas where hardstem bulrush is an important resource. Future research to determine the impact of water depth on the ability of waterlily and bulrush to withstand herbicide exposure is recommended.

### SOURCES OF MATERIALS

- <sup>1</sup>Osmocote®, The Scotts Company, PO Box 606, Marysville, OH 43040.  
<sup>2</sup>Joe Snow's Aquatic Plants, Argyle, TX 76226.  
<sup>3</sup>Kester's Wild Game Food Nurseries, Inc, Omro, WI 54963.  
<sup>4</sup>Renovate® 3, SePRO Corporation, 11550 North Meridian Street, Suite 600, Carmel, IN 46032.  
<sup>5</sup>Renovate® OTF, SePRO Corporation, 11550 North Meridian Street, Suite 600, Carmel, IN 46032.  
<sup>6</sup>Strategic Diagnostics Inc, 111 Pencader Drive, Newark, DE 19702-3322.

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