

# Prospects for Disrupting Rhizome Apical Dominance Prior to Chemical Treatment of *Phalaris arundinacea*

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## ABSTRACT

Reed canarygrass (*Phalaris arundinacea*) is a widely distributed invasive species that dominates many natural areas and restoration sites. Cost-effective suppression and restoration strategies need to be developed for plant communities affected by this species. Pretreatments designed to disrupt rhizome apical dominance may augment herbicide performance by making reed canarygrass rhizomes more susceptible to herbicide applications. I tested whether coupling pretreatment disking or kinetin application to herbicide application would enhance chemical control relative to only solitary herbicide application. I also evaluated the relative performance of two grass-selective herbicides, sethoxydim and fluzifop. All treatments suppressed reed canarygrass and indirectly led to improvements in existing native species abundance compared to the untreated control. In terms of reed canarygrass suppression, non-reed canarygrass aboveground biomass, and species diversity (Shannon's diversity), fluzifop performed as well as sethoxydim. Reed canarygrass biomass was consistently lower in plots where either disking or kinetin pretreatments were coupled with herbicide application than in plots receiving only herbicide treatment, though the degree of additional suppression varied with choice of herbicide. When sethoxydim was used for follow-up herbicide applications, disking reduced reed canarygrass biomass more than the kinetin pretreatment, but when fluzifop was used, kinetin pretreatments and disking were similar in their suppressive effect. Results of this study suggest that coupling these pretreatments with herbicide application can improve grass-selective herbicide performance on reed canarygrass.

**Keywords:** apical dominance, fluzifop, kinetin, reed canarygrass (*Phalaris arundinacea*), sethoxydim

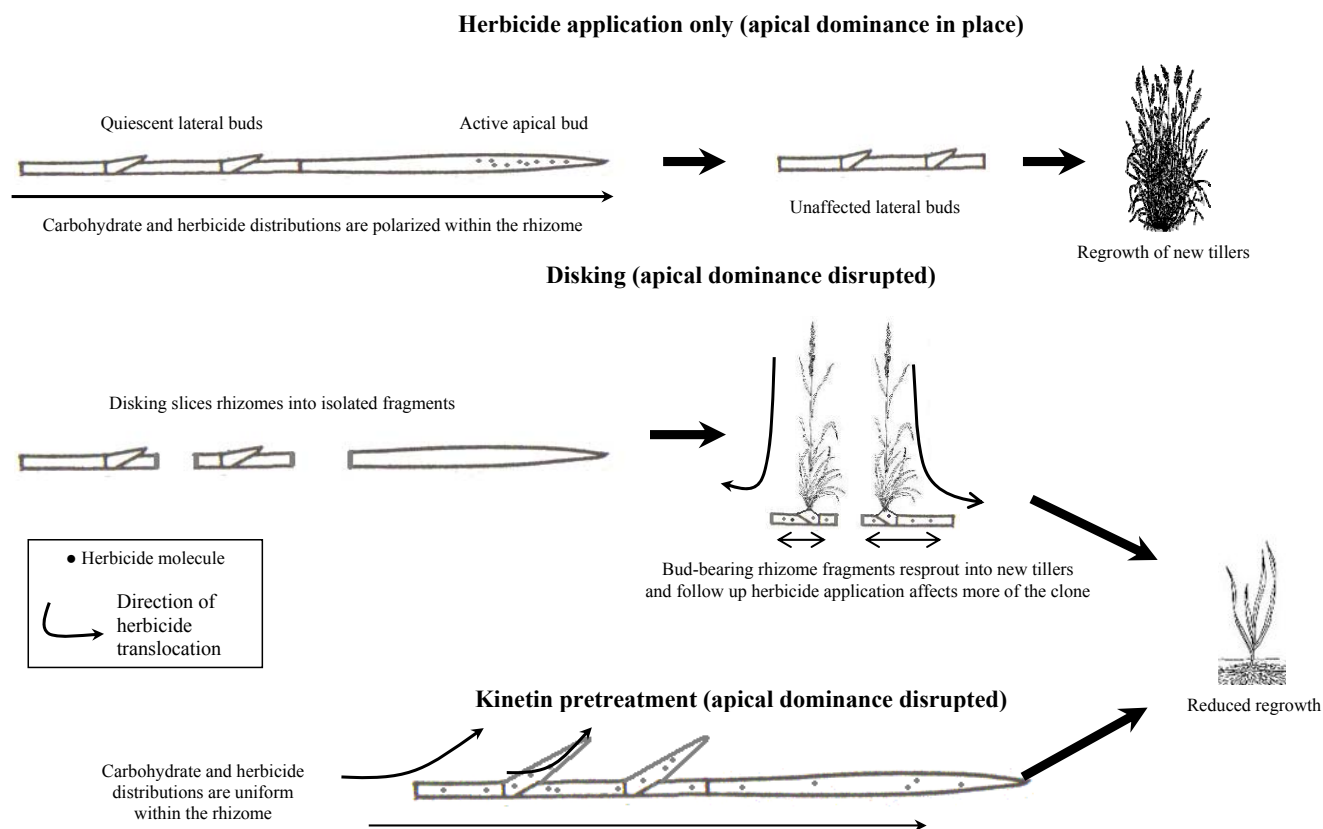
Reed canarygrass (*Phalaris arundinacea*) is a widely distributed perennial grass that invades and dominates natural areas and restoration sites that have been disturbed by sedimentation (Werner and Zedler 2002), nutrient enrichment (Wetzel and van der Valk 1998, Green and Galatowitsch 2001), hydrological instability or modification (Galatowitsch et al. 2000, Bonilla-Warford and Zedler 2002, Miller and Zedler 2003), or any combination of these factors (Kercher and Zedler 2004). This invasive species covers more than 202,000 ha of Wisconsin, USA (Bernthal and Hatch 2008), where it has fidelity for

11 different plant community types (Curtis 1959). Reed canarygrass invasions displace native plant species and alter restoration trajectories and vegetation succession patterns (Apfelbaum and Sams 1987, Maurer et al. 2003, Annen et al. 2008). Long-term, sustained control of this species continues to be a difficult management objective to achieve, particularly at sites where the disturbances that contribute to reed canarygrass invasions cannot be remedied prior to implementing control programs.

Herbicide application, alone or in combination with other treatments, is the method most commonly employed for reed canarygrass control. Herbicide applications are effective in the short term, but repeated treatments are usually required because this species can resprout from its seed

bank and/or rhizome bud bank (Reyes 2004, Annen 2008). For this reason, I propose adopting the term "suppression" rather than "control" when discussing herbicide effects on reed canarygrass to take into account the fact that these effects often do not persist beyond one or two growing seasons (and also to prevent confusion when the term "control" is used in experimental design terminology).

Post-application regrowth in reed canarygrass has been documented in several herbicide studies (Preuninger and Umbanhowar 1994, Kilbride and Paveglio 1999, Rachich and Reader 1999, Reinhardt and Galatowitsch 2004, Lesica and Martin 2004, Wilcox 2004, Annen et al. 2005, Hovick and Reinartz 2007, Wilcox et al. 2007, Annen 2008), and a mechanism to explain resprouting from rhizomes was



**Figure 1.** The effects of apical dominance and pretreatments on the distribution of herbicide within a perennial grass rhizome. When apical dominance is active, lateral buds are unaffected by herbicide and will develop into new tillers that will perpetuate the clone unless retreated with additional herbicide applications. When apical dominance is disrupted prior to herbicide application, herbicides are more uniformly distributed throughout the rhizome/tiller system, resulting in enhanced suppression and reduced regrowth.

postulated by Annen (2008). Herbicide performance depends on several factors, including choice of herbicide, proper use of additives, and application timing, and further depends on how an herbicide is used in the context of the life history traits of the target species. Life history traits such as perenniality and clonal expansion, both of which are conferred by rhizome systems, complicate control efforts. In order to completely kill a perennial species such as reed canarygrass, foliar-applied herbicides need to be translocated to rhizomes in toxic quantities and must also be distributed throughout the entire rhizome system. Regrettably, systemic herbicides tend to concentrate only in the distal, actively growing portions of perennial grass rhizomes because of apical dominance (Harker and Dekker 1988). Accessory treatments that disrupt rhizome apical dominance and stimulate lateral rhizome bud outgrowth may make

reed canarygrass rhizomes more susceptible to the effects of herbicides (Hillman 1985, Harker and Vanden Born 1997, Annen 2004). Coupling accessory treatments (such as tillage or growth regulator pretreatment) with herbicide application has been shown to improve reed canarygrass suppression and reduce the magnitude of regrowth in several recent studies (Kilbride and Paveglio 1999, Paveglio and Kilbride 2000, Hovick and Reinartz 2007, Annen 2008).

### Rhizome Apical Dominance and Reed Canarygrass Control

Apical dominance is the inhibitory influence of the rhizome apex on lateral rhizome bud outgrowth and rhizome branching. Holt (1954), Marquis and others (1979), and Reyes (2004) presented evidence that an apical dominance system operates in

reed canarygrass clones. Apical dominance gives rise to rhizome buds that are physiologically, anatomically, and morphologically heterogeneous (Hillman 1985, Sachs 2002). As a result of apical dominance, perennial grass rhizomes possess two types of buds: actively growing apical buds that give rise to new tillers, and metabolically inactive (dormant) lateral buds that initiate renewed growth whenever a disturbance affects top growth or the rhizome apex. These dormant lateral rhizome buds may contribute substantially to reed canarygrass resurgence capacity: Reyes (2004) measured an average of 820–900 dormant rhizome buds per square meter (47%–76% of the total buds) in a well-established reed canarygrass stand.

Apical dominance causes carbohydrates and inorganic nutrients to concentrate at the rhizome apex rather than being uniformly distributed throughout the entire rhizome (Figure 1).

Herbicides applied to aboveground leaves and culms likewise accumulate in rhizome apices because they are transported along with the carbohydrate assimilate stream. Harker and Dekker (1988) measured the distal/basal accumulation ratio (a measure of the degree of uniformity of herbicide distribution) for a series of postemergence herbicides in quackgrass (*Elytrigia repens*) rhizomes. Their results indicated that postemergence herbicides translocated toward the tips of rhizomes and that lateral rhizome buds were largely unaffected by herbicide applications to top growth. Similarly, Marquis and others (1979) reported that glyphosate applied to reed canarygrass leaves accumulated in distal meristematic tissues. As a consequence of apical dominance, systemic herbicides kill only a portion of the rhizome. Unaffected lateral buds can resprout into new tillers when the herbicide degrades. From a management perspective, apical dominance necessitates multiple-year herbicide applications to deplete the pool of dormant lateral rhizome buds within a reed canarygrass stand.

Two accessory treatments that disrupt rhizome apical dominance are tillage and plant growth regulator application (Figure 1). Tillage enhances herbicide performance by decapitating rhizome apices and slicing rhizomes into isolated multinodal fragments, which initiates active growth in dormant lateral buds (Leakey et al. 1975). Resprouting makes the rhizome more susceptible to complete herbicide translocation. Pretreatment with a growth regulator biochemically promotes lateral bud outgrowth and increases herbicide performance.

In recent field experiments (Kilbride and Paveglio 1999, Paveglio and Kilbride 2000, Hovick and Reinartz 2007, Annen 2008), the combined use of tillage and herbicide application suppressed reed canarygrass abundance more effectively than only solitary herbicide application. For sites where tillage is troublesome or impractical (such as wet sites or

sites with remnant sod and no prior history of tillage), no-till methods that can elicit similar effects on rhizome suppression (such as plant growth regulator applications) are potential pretreatment options, but further research is required before protocols can be established for their use. Harker and Taylor (1994) tested a mixture of the growth regulators chlormequat chloride and ethephon for enhancing quackgrass control with sethoxydim, and Annen (2008) evaluated this same mixture for reed canarygrass control. Although Harker and Taylor (1994) reported a 60% increase in suppression of aboveground biomass when this growth regulator system was applied as a pretreatment, Annen (2008) found that when this mixture was applied at the same rate it lessened reed canarygrass regrowth by only 26% and was cost prohibitive as a treatment option. Sachs and Thimann (1967) and McIntyre (1971) tested the synthetic cytokinin growth regulator kinetin (6*N*-furfurylaminopurine) for releasing quackgrass lateral buds from apical dominance. A 20 ppm kinetin solution applied directly to dormant lateral rhizome buds resulted in activation of growth and elongation, though the effects diminished within two weeks. Cline (1991) reviewed the literature and reported that inactive lateral buds began to synthesize proteins similar to those produced by actively elongating apical buds after treatment with kinetin. Since herbicide transport follows carbohydrate source-sink patterns, herbicide translocation to dormant lateral buds might be enhanced by pretreatment with the growth regulator kinetin. X-Cyte growth regulator (Stoller Enterprises, Houston TX) is a water-soluble postemergence formulation of kinetin (a synthetic cytokinin) that is used in agriculture to promote tillering, enhance translocation of substances from leaves, increase leaf surface area, and increase root initiation in cereal grasses. This growth regulator may have potential for enhancing herbicide performance on reed canarygrass.

## Evaluating Fluazifop as an Alternative to Sethoxydim

Sethoxydim and fluazifop are two postemergence systemic graminicides with the same mode of action (non-competitive inhibition of lipid synthesis) but from different chemical families. Annen and others (2005), Wilcox and others (2007), Annen (2008), and Healy and Zedler (2010) evaluated the efficacy of sethoxydim on reed canarygrass, but to date no empirical studies have documented the effects of fluazifop on reed canarygrass or compared its performance to sethoxydim in the same experiment. Herbicide activity follows a dose-response model that can be quantified by an  $IC_{50}$  value—the concentration of herbicide molecules required to inhibit 50% of target enzyme activity; a more effective herbicide will have a lower  $IC_{50}$  value. Devine (1997) assayed the  $IC_{50}$  of several graminicides applied to yardgrass (*Eleusine indica*) and reported a value of 1.4  $\mu\text{mol/L}$  for fluazifop and 1.3  $\mu\text{mol/L}$  for sethoxydim. He concluded that sethoxydim and fluazifop had a comparable level of activity on grasses. However, individual species can vary in their responses to herbicide treatments (cf. Marquis et al. 1979). Harker and Dekker (1988) reported that sethoxydim had a higher distal/basal accumulation ratio than fluazifop when applied to quackgrass, and based on their results we could predict that sethoxydim performance will be more affected by apical dominance than fluazifop, the latter being a more effective chemical treatment. Since it is not clear which graminicide will perform better against reed canarygrass, I wanted to test if fluazifop was more effective than sethoxydim at suppressing reed canarygrass. Fluazifop offers two additional advantages over sethoxydim: it is less expensive on a per hectare basis (\$12 vs. \$20, 2008 prices, not including additives) and more resistant to ultraviolet degradation (see Matysiak and Nalewaja 1999 or Annen 2006) owing to double bond resonance in its molecular structure.

The objectives of this study were to determine if disking or kinetin pretreatment followed by graminicide application enhances chemical control of reed canarygrass relative to solitary graminicide application and to compare relative reed canarygrass suppression with the graminicides sethoxydim and fluazifop.

## Methods

### Study Site and Design

This study was conducted within a 23 ha restored wet prairie at the Swamplovers Foundation Nature Preserve, a 186 ha land trust located in the Driftless Area of southwestern Wisconsin, USA (89°40'N, 43°7½'W). Hydrological input for the wet prairie is primarily surface flow from the adjacent landscape, and the site has a 227 ha drainage basin. During the course of this investigation, surface water was present at the site for only one week following early spring rainfall in 2006, and for three weeks following autumn rainfall in 2007. The research site was used for row-crop agriculture for several decades prior to being restored to wet prairie in 1987. Although 79 species were originally planted at the site, at the beginning of the experiment reed canarygrass dominated the planting at ca. 85% cover. The next most abundant species were, in decreasing order of abundance, Canada goldenrod (*Solidago canadensis*), tall white beard-tongue (*Penstemon digitalis*), New England aster (*Aster novae-angliae*), common ragweed (*Ambrosia artemisiifolia*), and Virginia mountain-mint (*Pycnanthemum virginianum*) (nomenclature follows Gleason and Cronquist 1991). Twenty-five additional species (22 planted native and 3 weedy non-natives) were also present in low density at the beginning of the experiment. As part of ongoing management at the preserve, the entire experimental area was burned on 22 April 2006 and 8 May 2007.

The effects of tillage and kinetin pretreatments on reed canarygrass control

with graminicides were tested in a randomized complete block split-plot experiment in 2006 and 2007. Four main effect treatments were tested in 100 m<sup>2</sup> whole-plot experimental units: 1) tillage (light disking to a depth of 10–15 cm) followed by herbicide application (at the 3- to 4-leaf growth stage, approx. 21 d treatment interval); 2) two sets of kinetin applications to reed canarygrass during the 2- to 3-leaf growth stage (approx. 12 d interval) followed by herbicide application at the 3- to 4-leaf growth stage (5 d after the second kinetin application); 3) herbicide application only (at the 3- to 4-leaf growth stage); and 4) untreated control. In addition, two herbicide split-plot effects were tested in 50 m<sup>2</sup> experimental subunits: 1) sethoxydim (Sethoxydim E Pro) and 2) fluazifop butyl ester (Fusilade DX). Treatments were randomly assigned to subplots and split plots and replicated three times.

### Application Protocol

Sethoxydim E Pro (13% a.i. sethoxydim) was applied at a rate of 3.75 pints/acre (4.45 L/ha). A water conditioning agent (ReQuest, Helena Chemical, Memphis TN) was added to sethoxydim spray mixtures at a rate of 2.5 mL/L (0.25% v/v) to sequester hardwater cations that can accelerate physical decomposition of sethoxydim (Shoaf and Carlson 1992, Annen 2006). A methylated seed oil/nonionic surfactant blend (MSO/NIS, Dyne-Amic, Helena Chemical, Memphis TN) was added to sethoxydim spray mixtures at a rate of 4 mL/L (0.375% v/v) to enhance foliar penetration and protect against UV degradation of sethoxydim, which can inactivate 50% of applied herbicide within ten minutes (Matysiak and Nalewaja 1999). Fusilade DX (24.5% a.i. fluazifop butyl ester) was applied at a rate of 24 fluid ounces/acre (1.75 L/ha). Both herbicides were applied as a 30 psi broadcast spray from a small capacity tank with a cone nozzle adjusted to provide a wide spray pattern. ReQuest water conditioning

agent and Dyne-Amic MSO/NIS were added to fluazifop spray mixtures at the same rate as for sethoxydim to standardize treatments and ensure that any treatment differences were not due to additives, which can substantially affect herbicide performance on reed canarygrass (Annen 2006). To prevent cross-contamination of herbicides, separate spray tanks were used to apply sethoxydim and fluazifop, and tanks were flushed out thoroughly between each subplot application to prevent buildup of herbicide concentrations.

X-Cyte (0.04% a.i. kinetin) was applied at a rate of 1.0 pint/acre (1.2 L/ha). Kinetin applications were made after 4:00 p.m. because this growth regulator formulation is light sensitive and requires an uptake period of several hours (Leo Brostowitz, professional crop consultant, pers. comm.). A nonionic surfactant and sticking agent (Induce pH, Helena Chemical, Memphis TN) was added to kinetin spray mixtures at a rate of 4 mL/L (0.375% v/v) to encourage penetration of the growth regulator into reed canarygrass leaves, prevent premature loss of applied growth regulator from leaf washing and rewetting, prevent evaporation of spray solutions from leaf surfaces, and stabilize tank mixture pH. McIntyre (1971) reported that the effects of kinetin application on lateral bud outgrowth wore off after 12 days. For that reason, kinetin was applied twice with a 12-day interval between applications. The 21-day interval between disking and subsequent herbicide application represented the time required for reed canarygrass regrowth to reach the 3- to 4-leaf growth stage after disking.

In 2006, kinetin was applied on 21 May and 2 June, plots were disked on 2 June, herbicide was applied to kinetin and herbicide-only plots on 7 June, and herbicide was applied to disked plots on 22 June. In 2007, kinetin was applied on 21 May and 1 June, plots were disked on 31 May, herbicide was applied to kinetin and herbicide-only plots on 8 June, and herbicide was applied to disked plots on 28 June.

**Table 1. Summary of mean ( $\pm$  SEM) treatment effects for an experiment to suppress reed canarygrass (RCG) biomass ( $\text{g}/0.125 \text{ m}^2$ ) and enhance biomass ( $\text{g}/0.125 \text{ m}^2$ ) and Shannon's Diversity ( $H'$ ) of native plant in a wet prairie in the Driftless Area of Wisconsin. Means with different letters were separate at  $\alpha = 0.05$ .**

Treatment	Response		
	RCG biomass (g)	non-RCG biomass (g)	Diversity ( $e^{H'}$ )
<i>Sethoxydim</i>			
Control	29.78 (2.0) a	6.93 (1.5) c	2.08 (0.05) a
Herbicide only	5.16 (1.6) b	46.43 (2.4) a	4.94 (0.08) a
Kinetin + herbicide	1.70 (0.6) c	41.87 (1.6) a	4.73 (0.03) a
Disk + herbicide	0.23 (0.3) d	33.47 (2.0) b	4.92 (0.11) a
<i>Fluazifop</i>			
Control	29.65 (1.6) a	15.37 (2.4) c	2.12 (0.05) a
Herbicide only	4.88 (1.5) b	53.60 (3.3) a	3.96 (0.08) a
Kinetin + herbicide	1.20 (0.8) c	39.38 (1.5) b	4.36 (0.06) a
Disk + herbicide	1.17 (0.7) c	42.08 (1.8) b	2.75 (0.12) a

Disked plots were treated with herbicide later than herbicide-only and kinetin-herbicide plots so that reed canarygrass phenology was standardized at the 3- to 4-leaf growth stage during herbicide application.

### Response Variables and Data Analysis

Treatment responses were measured on 4–6 September 2007. Aboveground biomass (hereafter, biomass) of herbaceous species was sampled in four ( $0.5 \times 0.25 \text{ m}$ ) rectangular quadrats/subplot. Quadrat shape and size were appropriate for this type of vegetation (Brummer et al. 1994). All herbaceous species present within each quadrat were sampled. Biomass was determined on a dry-mass basis. Plants were clipped at the plant-soil interface, separated by species, trimmed to ca. 10-cm pieces, and then dried to constant mass. Plant biomass was measured to the nearest 0.1 g with an Acculab EC-211 analytical balance (Acculab Sartorius Group, Edgewood NY). Since biomass harvest is a destructive sampling technique that can alter treatment responses (Krebs 1989), and since Annen (2008) found that a two-year lag time existed to detect effects of similar treatments, plots were sampled only at the conclusion of the experimental treatments. Species richness (the number of

species per  $0.125 \text{ m}^2$ ) and Shannon's Diversity ( $H'$ ) were calculated for each main effect treatment and split plot treatment. Shannon's Diversity was calculated as  $H' = -\sum p_i (\ln p_i)$ , where  $p_i$  corresponds to the proportional abundance of the  $i$ th species. For clarity,  $H'$  estimates were converted into the same scale as species richness with MacArthur's  $N_1$  (where  $N_1 = e^{H'}$ ) (MacArthur 1965).

Data were tested for normality ( $\chi^2$  goodness-of-fit test) and homoscedasticity (Bartlett's Test) (TOXSTAT vers. 3.0, University of Wyoming, Laramie). Main effect (disking + herbicide, kinetin + herbicide, herbicide only, and control) and split effect (sethoxydim and fluazifop) treatment means were compared with a parametric analysis of variance (ANOVA) for a randomized block split-plot design (SPSS vers. 14.0, SPSS Inc., Chicago IL). Means were separated by the degree of overlap in the 95% confidence interval for each response (Day and Quinn 1989). When constructing confidence intervals for Shannon's Entropy, variance estimates were calculated following methods outlined by Magurran (1988). Statistical significance was tested at  $\alpha = 0.05$ . Treatment effects sizes were calculated as [(treatment mean – control)/treatment mean]  $\times$  100.

### Results

Compared to the untreated control, reed canarygrass aboveground biomass was lower and non-reed canarygrass abundance was higher in treated plots (Table 1). There was no difference in reed canarygrass suppression ( $F_{(1,2)} = 0.004$ ,  $p = 0.957$ ), non-reed canarygrass biomass ( $F_{(1,2)} = 1.334$ ,  $p = 0.367$ ), or herbaceous species diversity ( $F_{(1,2)} = 2.115$ ,  $p = 0.283$ ) due to choice of herbicide. Species richness was 55% greater in disked plots treated with sethoxydim than disked plots treated with fluazifop (9.0 vs. 5.8 species per sampling frame;  $F_{(1,2)} = 25.00$ ,  $p = 0.038$ ) (see Appendix available at [uwpress.wisc.edu/journals/journals/er\\_suppl.html](http://uwpress.wisc.edu/journals/journals/er_suppl.html)).

Compared to the untreated control, reed canarygrass biomass was nearly five times lower in plots treated with sethoxydim, equal to a suppressive effect of  $197 \text{ g}/\text{m}^2$  (Table 1). Reed canarygrass biomass was even lower when either disking or kinetin pretreatment was coupled to sethoxydim application. The combined suppressive effect on reed canarygrass of coupling treatments was equivalent to  $236 \text{ g}/\text{m}^2$  when disking was the pretreatment and  $225 \text{ g}/\text{m}^2$  when kinetin was the pretreatment (disk + sethoxydim > kinetin + sethoxydim > sethoxydim only > control). Non-reed canarygrass biomass was higher in treated plots

relative to the untreated control (Table 1). Although diversity was statistically similar among all treatments, it was twice as high in treated plots as in the untreated control and was ecologically significant (Table 1). Reed canarygrass comprised 81.2%, 10.0%, 3.9%, and 0.7% of the total aboveground biomass in the control, sethoxydim only, kinetin + sethoxydim, and disk + sethoxydim treatments, respectively. Mean species richness per sampling frame was highest in disked plots, similar in kinetin and sethoxydim-only plots, and lowest in control plots (online appendix).

Compared to the untreated control, reed canarygrass biomass was five times lower in plots treated with fluazifop, a suppressive effect of 198 g/m<sup>2</sup> (Table 1). As with sethoxydim, reed canarygrass biomass was even lower when pretreatments were coupled to fluazifop application. The combined suppressive effect on reed canarygrass of coupling treatments was equivalent to 228 g/m<sup>2</sup> when either disking or kinetin pretreatment was used in conjunction with fluazifop (disk + fluazifop = kinetin + fluazifop > fluazifop only > control). Non-reed canarygrass biomass was higher in all treated plots compared to untreated controls (Table 1). As with sethoxydim, diversity was statistically similar among all treatments, but was 30% to 106% higher in treated plots compared to the untreated control (Table 1). Reed canarygrass comprised 65.9%, 8.3%, 2.9%, and 2.7% of the total aboveground biomass in the control, sethoxydim only, kinetin + sethoxydim, and disk + sethoxydim treatments, respectively. Species richness per unit area was similar among treated plots and lowest in the untreated control (see online appendix).

The online appendix summarizes the composition of post-treatment non-reed canarygrass vegetation. The total number of species sampled among all replications was lowest in the untreated control plots, intermediate in herbicide only and kinetin +

herbicide plots, and highest in tilled plots. Three species had a mean abundance of at least 1.0 g/quadrat in the untreated control plots, while treated plots had between six and nine species with mean abundance of at least 1.0 g/quadrat.

## Discussion

Reed canarygrass abundance was consistently lower and non-reed canarygrass abundance, species richness, and diversity consistently higher in all treated plots compared to the untreated controls (Table 1). In terms of reed canarygrass suppression and improvements in native species abundance and diversity, fluazifop performed as well as sethoxydim. Both herbicide formulations reduced reed canarygrass aboveground biomass by a factor of five (Table 1). Reed canarygrass abundance was even lower in plots where either disking or kinetin application was coupled to herbicide application, suggesting that these combinations of treatments were more conducive to desired endpoints than solitary herbicide use, at least in the short term.

### Use of Disking Prior to Herbicide Application

As reported in previous studies (Kilbride and Paveglio 1999, Paveglio and Kilbride 2000, Hovick and Reinartz 2007, Annen 2008), reed canarygrass was less abundant after sequencing pretreatment disking with herbicide application to augment herbicide performance. Non-reed canarygrass biomass in disked plots was intermediate between the control and the other treatment plots (Table 1), probably a result of the restart associated with disking. This restart affected both reed canarygrass and established prairie species. The nontarget planted prairie species, however, were allowed to recover from tillage by use of a selective graminicide on reed canarygrass regrowth. Nevertheless, repeated tillage could have negative consequences on native perennial species, such as

lowering seed production capability and fecundity.

Although species richness was greatest in disked plots, floristic quality was lower and several common agricultural weeds were present, for example, velvet-leaf (*Abutilon theophrasti*), black mustard (*Brassica nigra*), bull thistle (*Cirsium vulgare*), Canada thistle (*C. arvense*), yellow foxtail (*Setaria glauca*), green foxtail (*S. viridis*), and clover (*Trifolium* spp.), which was likely a result of the agricultural land-use history of the site and the fact that the wet prairie was a ten-year-old restored community. In contrast, Kilbride and Paveglio (1999), Paveglio and Kilbride (2000), and Annen (2008) reported increases in floristic quality after remnant sites were tilled. The size and composition of the native species propagule bank may affect treatment outcomes. Tillage can either bury (Combroux et al. 2002) or expose (Thompson and Luthin 2004) seed banks, and these agricultural weeds were nearly absent in other treatment plots. Therefore, another disadvantage of tillage is the potential for secondary weed outbreaks, particularly in restored sites. Disking adds \$8 (2008 USD) per hectare to control costs (although the cost of using this technique varies).

### Use of Kinetin Pretreatments

Coupling kinetin pretreatments to graminicide application also resulted in lower reed canarygrass biomass compared to solitary herbicide application. Kinetin pretreatments doubled reed canarygrass suppression when sethoxydim was used as the follow-up treatment and tripled it when fluazifop was used. Coupling kinetin pretreatments to graminicide application was as effective as preapplication disking when fluazifop was used but not sethoxydim (Table 1). Annen (2008) reported a 26% decrease in reed canarygrass resurgence capacity and Harker and Taylor (1994) described a 60% additional decrease in quackgrass biomass when 2:1 mixtures of the growth regulators chlormequat

chloride and ethephon were used as an herbicide pretreatment. In this study, use of kinetin as a growth-regulator pretreatment suppressed reed canarygrass up to two times more than growth regulators previously tested by Annen (2008) and up to four times more than those tested by Harker and Taylor (1994). Not only were kinetin applications more effective, they were considerably less expensive: paired kinetin treatments added only \$14 per hectare to suppression costs (as compared to \$175 per ha for CCC/ethephon pretreatments) (2008 prices in USD, not including labor or additives).

It is possible that additional growth-enhancing effects of kinetin may have been responsible for the results measured in this field experiment. Cytokinins are known to promote cell division and elongation, and increasing leaf surface area for herbicide contact could have resulted in absorption of greater concentrations of herbicide by reed canarygrass. Follow-up studies should consider incorporating a kinetin-only treatment with leaf characteristics as response variables to determine if kinetin application increases leaf surface area of reed canarygrass and associated species.

### **Posttreatment Species Composition**

Several species that subdominated the planting prior to initiating the experiment were among the most abundant species sampled at its conclusion (see Appendix available at [uwpress.wisc.edu/journals/journals/er\\_suppl.html](http://uwpress.wisc.edu/journals/journals/er_suppl.html)). With the exception of the weedy ruderal species that appeared in the tilled plots after disking, no species were sampled at the conclusion of the experiment that were not present when the experiment was initiated. Previous studies (Annen et al. 2005, Annen 2008, Healy and Zedler 2010) documented higher post-treatment non-reed canarygrass abundance concordant with graminicide applications in mixed vegetation stands. For Healy

and Zedler (2010), posttreatment vegetation composition was weedy or of low ecological quality when graminicides were used in restored sites with agricultural use histories. For Annen and colleagues (2005), it consisted of midsuccessional native species in low abundance when graminicides were used in degraded natural areas. For Annen (2008), posttreatment vegetation consisted of a diverse mix of semiconservative native species when graminicides were used in high-quality remnant sites with low initial reed canarygrass abundance. Posttreatment vegetation abundance depends on site factors such as land-use history and density and composition of remnant vegetation stands and propagule banks, and also on active revegetation efforts conducted in conjunction with herbicide applications. High-quality remnant sites typically respond more positively to graminicide treatments than restored sites with a long history of chronic disturbance and off-site impacts (pers. obs.).

In this experiment, increases in non-reed canarygrass abundance could have been the result of an indirect competitive release mechanism, although this experiment was not designed to specifically address this hypothesis. In this framework, the abundance of planted species initially present in the study area was suppressed by resource competition with reed canarygrass. Reed canarygrass thatch has a mulching effect on native species, which further contributed to reed canarygrass dominance. Combinations of prescribed burning (for litter removal) followed by selective reed canarygrass suppression treatments (and treatment combinations) may have opened up niche space in the vegetation matrix and enabled existing species to expand in response to new resource opportunities.

## **Conclusions and Management Implications**

1. Reed canarygrass abundance was lower and non-reed canarygrass abundance and diversity were higher in treated plots than in the untreated control.
2. Fluazifop was an effective substitute for sethoxydim in this experiment and may have some economic advantages. However, these herbicides may not be suitable for all abatements, and it is therefore important to read and understand the labeling of both herbicide products before deciding on a formulation to use.
3. Coupling pretreatment disking and kinetin applications with herbicide use augmented reed canarygrass suppression with sethoxydim and fluazifop relative to solitary herbicide use.
4. Secondary weed outbreaks and weed shifts can occur when tillage is used to augment reed canarygrass suppression in restored settings, and no-till methods of disrupting apical dominance may be more advisable in restored communities where reed canarygrass is problematic or in remnant sites with no prior history of tillage.
5. Kinetin pretreatments were an effective substitute for pretreatment disking when fluazifop was used as the follow-up herbicide.
6. Kinetin growth regulators were a less expensive, more efficacious alternative to chlormequat chloride/ethephon growth regulators reported by Harker and Taylor (1994) and Annen (2008).
7. Kinetin has potential for enhancing reed canarygrass suppression with graminicides and warrants further investigation.

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