

UNIVERSITY OF WISCONSIN-LA CROSSE

Graduate Studies

BREAKING THE CYCLE: CAN GLYPHOSATE AND WILLOW CUTTINGS
DISRUPT POSITIVE FEEDBACKS THAT SUSTAIN REED CANARYGRASS
(*PHALARIS ARUNDINACEA*) MONOCULTURES?

A Chapter Style Thesis Submitted in Partial Fulfillment of the Requirements for the
Degree of Master of Science

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Biology

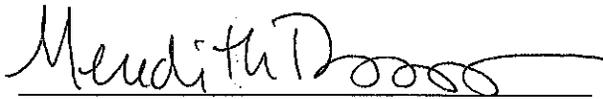
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By Michael Paul Merriman

We recommend acceptance of this thesis in partial fulfillment of the candidate's requirements for the degree of Master of Science in Biology

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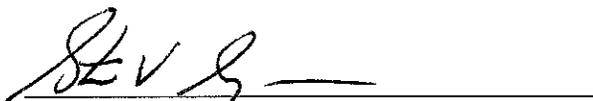


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ABSTRACT

Merriman, M.P. Breaking the cycle: Can glyphosate and willow cuttings disrupt positive feedbacks that sustain reed canarygrass (*Phalaris arundinacea*) monocultures? MS in Biology, May 2014, 51pp. (M. Thomsen and D.T. Gerber)

Restoring invaded ecosystems requires the reduction of any positive feedbacks created by the invader. Feedbacks created by reed canarygrass (*Phalaris arundinacea*) in the Upper Mississippi River floodplain include early season growth, a thick canopy and an extensive belowground rhizome system. We tested a novel restoration strategy designed to reduce these feedbacks by applying broad spectrum herbicide (glyphosate) in late fall 2012 and 2013, and by planting willows as a native competitor. We conducted the experiment in a site dominated by reed canarygrass in the Upper Mississippi River (UMR) floodplain south of La Crosse, WI over two growing seasons. In the first year of the experiment, glyphosate reduced reed canarygrass percent cover early in the growing season. At the end of the second year, glyphosate reduced reed canarygrass belowground biomass. Willow stakes that were planted in fall resulted in poor establishment. Stakes that were planted in spring exhibited higher height and canopy spread in glyphosate-treated plots. Ground-level light availability was higher in plots that had surviving willows than in plots without willows. Thus, results demonstrate the potential for willow stakes to establish in dense reed canarygrass, but additional information on how to maximize the success of the approach is still needed.

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CHAPTER I

REED CANARYGRASS (*PHALARIS ARUNDINACEA* L.), THE UPPER MISSISSIPPI RIVER FLOODPLAIN, AND FEEDBACKS

Reed Canarygrass (*Phalaris arundinacea* L.): A Noxious Invader of North American Wetlands

Invasive plants are problematic because they displace native species and decrease biodiversity, leading to degraded ecosystems with altered functions (Vitousek et al. 1997, Galatowitsch et al. 1999, Zedler and Kercher 2004, Kim et al. 2006, Zedler 2009). Estimates of the annual cost of efforts to restore invaded ecosystems range from millions to billions of dollars (Pimentel et al. 2000). Although invasive plants affect all ecosystems, wetlands are especially vulnerable to invasions (Zedler and Kercher 2004). Wetlands are landscape sinks that accumulate materials that can facilitate invasion, such as sediments and nutrients (Zedler and Kercher 2004). Wetlands are also ecosystems of critical importance, due to their high productivity, high levels of diversity, and contribution to crucial ecosystem functions (Lavergne and Molofsky 2004, Zedler and Kercher 2005). Because invasion of wetlands comes at such a high ecological cost, it is imperative that we develop efficient restoration techniques for invasive plants in wetlands (Zak et al 2011).

One of the most noxious invaders of North American wetlands is reed canarygrass (*Phalaris arundinacea* L., hereafter RCG; Galatowitsch et al. 1999, Zedler and Kercher 2004). In North America, RCG invasion is problematic in the Pacific Northwest (Kim et

al. 2006), the upper Midwest (Knutson et al. 2005) and southeastern United States (Foster and Wetzel 2005). For example, in Wisconsin wetlands, RCG dominates (> 80% cover) more than 40,000 ha (98,800 acre; Bernthal and Willis 2004). Although it was long believed that RCG was introduced to North America as a single European strain in the early 19th C, recent research has found that RCG in the U.S. today is a mix of native North American and multiple European genotypes (Lavergne and Molovsky 2007). Genetic recombination of the multiple RCG populations in North America is believed to have resulted in selection for high colonization rates, phenotypic plasticity, and a high degree of regional specialization (Lavergne and Molovsky 2007). Despite its damaging effects on wetland ecosystems, RCG is still widely cultivated today, bred into cultivars for use as forage crops or as persistent perennial cover for pastures (Lavergne and Molovsky 2004), erosion and sedimentation control (Lefor 1987, Kim et al. 2006), and for bioenergy and paper production (Lavergne and Molovsky 2004).

Reed canarygrass is a cool-season perennial grass that possesses a key set of characteristics that many wetland plant invaders share: prolific seed production and long-range dispersal of propagules, broad environmental tolerance, and competitive dominance relative to native species (Zedler and Kercher 2004, Lavergne and Molovsky 2004). Multiple modes of reproduction – from seeds, rhizomes, and plant fragments (Lefor 1987, Kercher and Zedler 2004, Lavergne and Molovsky 2004) – allow established stands of RCG to increase in density and expand. The large numbers of seeds that RCG produces each year are capable of remaining dormant for many years (Vose 1962), creating long-lived seed-banks that only require moist soils and light for germination (Lindig-Ciseros and Zedler 2002). Disturbance events, such as tree-falls, extended

flooding, or irregularly high flood flow velocity, create opportunities for seed germination and RCG establishment, potentially delivering RCG propagules at the same time. Following canopy disturbances in forests, RCG is able to out-compete tree seedlings and replace forested areas with near-monocultures of grass (Knutson and Klaas 1998, Urich et al. 2002, Knutson et al. 2005, Romano 2010, Reinhardt Adams et al. 2011). Once established, RCG monocultures leave behind thick layers of RCG litter at the end of every growing season (Annen 2011). This litter covers the ground well into spring, shading out any competing species (Annen 2011), especially tree seedlings – further increasing RCG’s already strong early-season growth. Additionally, established stands of RCG in riverine wetlands clog waterways, choking water circulation and preventing native plant propagules from traveling downstream (Lavergne and Molofsky 2004).

RCG monocultures have substantial impacts on community trophic interactions, further increasing the harm done to the marshes it invades (Galatowitsch et al. 1999, Zedler and Kercher 2004, Lavergne and Molovsky 2004). Formation of RCG monocultures decreases quantity, diversity, and quality of available host forage plants and may cause declines in consumers and their predators including arthropods, herbivorous small mammals, insectivorous small mammals, and the larger predators of these organisms (Spyreas et al. 2010). These authors also found that increasing RCG dominance leads to a decrease in plant diversity and overall floristic quality, with a corresponding decrease in the diversity and abundance of Homopterans. In addition, they found that voles and shrews (grassland specialists) became more abundant, while mice (often found in weedier areas with less dense vegetation and more exposed ground)

became less abundant, suggesting a change in mammalian community structure with increasing RCG dominance (Spyreas et al. 2010). Various strains of RCG have been bred to have either low or high concentrations of a diversity of alkaloid defense chemicals depending on how it is being used (Lavergne and Molofsky 2004). Because some invasive hybrids of RCG have high concentrations of alkaloids, herbivores preferentially graze on native plant species over RCG (Marten et al. 1976, Gauthier and Bedard 1991, Lavergne and Molofsky 2004, Bush et al. 2007). Preferential herbivory reduces both native plant cover and native plant propagules, deterring native plant reproduction. Furthermore, as a result of a variety of human activities, deer populations are above levels seen historically (Kellogg and Bridgham 2004), increasing pressures on native plant species. The cumulative effects of herbivory on native plants may further contribute to the ability of RCG to become established in sites previously dominated by native plant species.

Management Options for Controlling Reed Canarygrass

Many studies have assessed control methods for RCG in different regions and habitats, with none having emerged as clearly superior to the others (Lavergne and Molofsky 2006). Control efforts have found that RCG management requires multiple treatments in order for native plant re-establishment to succeed (Kellogg and Bridgham 2004, Foster and Wetzel 2005, Kim et al. 2006, Reinhardt Adams and Galatowitsch 2006, Hovick and Reinartz 2007, Annen 2011, Thomsen et al. 2012). Suppressing RCG via herbicide (pre-planting treatment) and then introducing native plant species is one of the most effective ways to replace RCG monocultures with native vegetation (Kellogg and Bridgham 2004, Hovick and Reinartz 2007, Kim et al. 2006, Thomsen et al. 2012). In

particular, late-season glyphosate and planting of willow stakes is a promising combination to achieve these two goals.

Reinhardt Adams and Galatowitsch (2006) found that fall glyphosate treatments (late August and late September) were twice as effective as those applied in spring (mid-May) at controlling RCG, due to lack of rhizome-based shoots and increased storage of carbohydrates in rhizomes in the fall. Hovick and Reinartz (2007) followed up on these findings by evaluating the effectiveness of fall glyphosate applications followed by planting treatments that included willow stakes. Willow stakes were both purchased from a wholesale nursery and locally harvested; species used included slender willow (*Salix petiolaris* Sm.), Bebb's willow (*Salix bebbiana* Sarg.), and pussy willow (*Salix discolor* Muhl.). They found that herbicide treatment dramatically decreased RCG cover relative to controls. In general, willow stake survival was low, but willow survival was also species-dependent; there was significantly higher survival in locally-harvested slender willow over all others tested. While this study had poor willow survival, they speculated that willows could not establish due to lower than optimal soil moisture levels for the unrooted stakes. Wetter conditions, combined with effective RCG control, may allow for native species to establish from planted stakes.

Thomsen et al. (2012) conducted a similar restoration study with the goal of managing RCG and re-establishing native species in a floodplain forest site. The treatment plots were cleared of RCG using a forestry mower, then disked and then treated with pre-emergent herbicides, rather than glyphosate. In addition to hand broadcasting plots with green ash (*Fraxinus pennsylvanica* Marshall), black ash (*Fraxinus nigra* Marshall), river birch (*Betula nigra* L.), and Dutch elm disease-resistant American elm

(*Ulmus americana* L.) seeds, they planted live stakes. Live stake plantings consisted of 1000 cottonwood (*Populus sp.*L), an undetermined number of red osier dogwood (*Cornus stolonifera* Michx.), and willow cuttings; an undetermined number of black willow (*Salix nigra* Marshall) and sandbar willow (*Salix interior* Rowlee) cuttings, in addition to 2,500 more willow cuttings four weeks later. Within the first year the herbicide treatments effectively decreased the cover of RCG and tree seedling density in the treatment plots was approximately ten times the amount of untreated controls. Willow stake cuttings only showed 12% survival after two years. However, the investigators did speculate low willow survival might have been due to herbicide remnants in the willow root zone that inhibited willow stake root production.

A study by Kim et al. (2006) found better rates of survival for live willow stakes planted in RCG monocultures in riparian wetlands. Their pre-planting treatment consisted of multiple applications of glyphosate during summer followed by planting of live willow stakes in different density treatments. They further suppressed RCG regeneration using mowing followed by wood chip mulch. After evaluating the correlation between willow planting density and RCG % cover their results showed that RCG responded significantly to willow planting density; willow shading reduced total RCG biomass by 44.9 to 68.0% (depending on willow age) in their high-density (0.60 m (1.97 ft) spacing) treatment. Although Kim et al. (2006) demonstrated a method for controlling RCG in a wetland site using affordable native plants, they did not quantify survival of introduced native willows.

Several authors have speculated that if willows could be successfully established in sites dominated by RCG, they could ultimately control RCG (Kim et al. 2006, Hovick

and Reinartz 2007, Thomsen et al. 2012). Most species of willow produce shrubby canopies, which could shade out RCG and suppress its recolonization (Hovick and Reinartz 2007). In addition, willows are water-loving plants that are tolerant to waterlogging, flood flow stress, and sediment deposition, making them ideal wetland competitors for RCG (Amlin and Rood 2001). Although there is evidence that increased willow-planting density decreases RCG % cover, there is little research that explores relationships between survivorship of live stakes and site conditions in riparian areas (Pezeshki et al. 2007, Caplan et al. 2012). There is almost no information on what height to use for live stakes; taller stakes may more rapidly shade out RCG if they sprout from their upper regions, and they may be better able to tolerate flooding if their height keeps them above water. We also lack information about willow growth responses to fall versus spring stake plantings. In one study spring cuttings survived better than fall live stakes (Densmore and Zasada 1978), while others found no survival variability with planting timing (Hoag et al. 1991, Hoag 1993). Willow stakes are typically harvested in early spring while plants are still dormant and then planted as soon as possible (Hoag 1993). When spring floodwaters delay planting, stakes must be stored, a process that likely decreases willow stake survival. However, if native species plantings are made in the fall, the plants may have a greater chance to establish in spring while RCG is still suppressed by the glyphosate applied the previous fall.

The Upper Mississippi River Floodplain Forest: What is it Supposed to Be, What Has it Become?

One ecosystem under particular threat to invasion by RCG is the Upper Mississippi River (UMR) floodplain forest. The UMR floodplain forest is located on the primary and secondary floodplain terraces of the Mississippi River, Illinois River, and their major tributaries (Romano 2010); it is a biome of crucial importance both economically and ecologically. Historically, the UMR was utilized for commercial navigation, flood control, as a timber source, for water supply, and for water quality improvements; most of these features are still heavily utilized (Romano 2010, De Jager et al. 2012). Additionally, the UMR floodplain forest provides a large wildlife habitat, has tremendous aesthetic value, and is a popular recreational area. For all of these reasons, maintaining the biological integrity of the UMR floodplain forest is critical. The first step towards this goal is to develop an understanding of the UMR biological community.

Forests are essential in the UMR and other riparian ecosystems for maintaining nutrient levels and water quality (USACE 2006). Forests prevent stream bank erosion through root establishment (Hoag and Landis 2001) and provide habitat for a wide range of vertebrates (Knutson and Klaas 1998). Furthermore, maintaining existing UMR floodplain forests is critical because in some areas up to 40% of the UMR's forested habitats have been destroyed since the late 1890s (Knutson and Klaas 1998). For example, only 22 to 25% of the UMR floodplain stretch from St. Paul, Minnesota to Alton, Illinois is presently occupied by forest, when 50 to 70% of this area was occupied by forest prior to European settlement (Knutson and Klaas 1998).

Humans have contributed to loss of forested sites in the UMR floodplain through construction of the lock and dam system in the late 1930s, in addition to other anthropogenic impacts over the last 150 years (USACE 2007, Theiling 1995). The UMR south of Minneapolis has been converted to a series of navigational pools between dams, where the main channel was deepened by approximately 1.4 to 2.7 m (4.59 to 8.86 ft; DeJager et al. 2012). As a result, northern reaches of each pool now have higher-than-historic water levels, and low-lying forests have been lost at the southern end of each pool.

The remaining UMR floodplain hosts a diverse array of plant species; a study by Knutson and Klaas (1998) identified 138 plant taxa from UMR floodplain forests sites between Winona, Minnesota and Guttenburg, Iowa, spanning 177 km (110 mi) of river length. Out of all tree species, these authors found that silver maple (*Acer saccharinum* L.) had extreme dominance (especially at size classes of 20 cm (7.87 in) and larger), followed by elm (*Ulmus sp.* L.), green ash (*Fraxinus Pennsylvania* Marshall), and swamp white oak (*Quercus bicolor* Willd.). Additionally, they found that elm and ash trees were represented primarily by young trees and were not well represented as mature canopy. Other canopy tree species associated with the UMR floodplain forest include hackberry (*Celtis occidentalis* L.), pin oak (*Quercus palustris* Muench.), eastern cottonwood (*Populus deltoids* Marsh.), sycamore (*Platanus occidentalis* L.), black willow (*Salix nigra* Marsh), sweetgum (*Liquidambar styraciflua* L.), and others depending on location within the UMR region (Phares and Larsson 1980, Romano 2006). Under former natural conditions, primary forest succession on mud flats and sandbars led to establishment of willows (*Salix sp.* L.), eastern cottonwood, and silver maple (Galatowitsch and McAdams

1994). These early-successional species were then replaced by swamp white oak, American elm, and bitternut hickory (*Carya cordiformis* Wangenh. K.Koch; Galatowitsch and McAdams 1994). The reasons for current successional trends leading to extreme dominance by silver maple are unknown, but may include a higher water table, increased flood frequency and duration, reduced channel migration, replacement of diseased American elms, and cutting of forests in 1930's prior to lock and dam construction (Knutson and Klaas 1998).

Further loss and decreased diversity of forested areas in the UMR could pose devastating problems for wildlife, especially birds that depend on forest canopies for resources, nests, and roosting (Knutson and Klaas 1998). The upper canopy of UMR forests are frequently occupied by herons and egrets (family Ardeidae), bald eagles (*Haliaeetus leucocephalus* L.), red-shouldered hawks (*Buteo lineatus* Gmelin), great horned owls (*Bubo virginianus* Gmelin), flycatchers (family Tyrannidae), blue-gray gnatcatchers (*Poliophtila caerulea* L.), yellow-throated vireos (*Vireo flavifrons* Vieillot), warbling vireos (*Vireo gilvus* Vieillot), red-eyed vireos (*Vireo olivaceus* L.), cerulean warblers (*Dendroica dominica* L.), and Baltimore orioles (*Icterus galbula* L.; Knutson 1995, Knutson and Klaas 1998). Within the last decade the cerulean warbler became a species of management concern due to steep population declines. One hypothesis for the decline of the cerulean warbler in the UMR is loss of mature American elm trees that were taller and with stronger and larger limbs at high canopy levels than other tree species; Knutson and Klass (1998) observed that cerulean warblers tended to perch in the highest tree species, such as elm.

Just as UMR floodplain forests are important to canopy-dependent avian species, seasonally inundated wetlands in the UMR support 485 species of mussels, fishes, amphibians, reptiles, birds, and mammals – including 50 that five states of the UMR list rare, threatened, or endangered (Sparks et al. 1998). Aquatic organisms are affected by floodplain forest dynamics, such as nutrient fluxes due to woody debris and water shading by canopy trees (Opperman et al. 2010), leading to further effects on fish macroinvertebrate prey and vascular plants. Terrestrial herbivores, such as white-tailed deer, muskrats, and arthropod herbivores, are dependent on the resources provided by UMR plants; for example food sources and cover from predators (Knutson and Klaas 1998, USACE 2006).

Taking a Feedback Approach

If we wish to keep the UMR floodplain forested, a restoration plan needs to be developed using our knowledge of RCG as an invasive plant, our knowledge of the UMR floodplain forest, and an alternative stable state ecological model so that we can project long-term results. Ecological restoration is the repair of the species diversity and dynamics of a degraded ecosystem back to a “similar-to-remnant” (native) condition, consisting of native species and historic ecosystem functions. Restoring an ecosystem that has been heavily invaded to the state of a monoculture is a complex and lengthy process (Reinhardt Adams and Galatowitsch 2006, Annen 2011, Thomsen et al. 2012), and the results may not completely resemble the original state. Models have been used to predict the path that an ecosystem takes from degraded to restored, in order to improve predictability of management outcomes (Suding and Hobbs 2009). These models include a classic successional model and the newer alternative stable state model (Zedler 2009).

The successional model predicts that as an ecosystem degrades it follows a linear path through time until it reaches a monoculture state – restoration taking the same path in the opposite direction (Suding and Hobbs 2009). While the linear model has the appeal of simplicity, the alternative stable state model is more realistic in many cases (Zedler 2009); especially where you are looking at two drastically different states, such as RCG monoculture and floodplain forest. The alternative state model predicts that the two states, remnant and degraded, are rather stable until they pass a threshold point, after which they collapse or recover quickly (Suding and Hobbs 2009).

Passing the threshold point to the degraded condition is typically the result of a disturbance that makes the landscape vulnerable to heavy invasion (Annen 2011). The ease with which an ecosystem enters a degraded state following a disturbance is known as the ecosystem's resilience: which may be defined as the capacity of an ecosystem to withstand disturbance and then reorganize to retain its normal regime (Gunderson 2000). Stability is the persistence of an ecosystem state with or without the presence of a disturbance (Walker and Del Moral 2009). Stable ecosystems with large resilience are able to withstand a large range of disturbances, while those with low resilience can be pushed into a degraded state with a small disturbance (Gunderson 2000). The resiliency of a stable state to disturbance is determined by the number, strength, and interactivity of internal system feedbacks.

Internal feedbacks can maintain an invaded (degraded) state in an ecosystem following disturbance (Figure 1). In the case of our study system, disturbance events cause canopy gaps in forested sites. Common disturbance events include windstorms, extended inundation, and high flood velocities. Once canopy gaps are established, RCG

propagules are typically already present, or delivered by floodwaters. In the first growing season, RCG quickly develops a thick canopy, out-shading any germinating tree seedlings. Established RCG individuals increase the amount of the RCG propagule sources for the next growing season, and leave a thick layer of litter behind that inhibits native plant growth during the following growing season. Sites with high nitrogen (N) levels also favor the suppression of native plant species while facilitating RCG invasion (Green and Galatowitsch 2002, Herr-Turoff and Zedler 2005). All of these feedbacks further increase RCG dominance, reinforcing RCG monoculture. Thus, invasive plants engineer their ecosystems to promote their own growth, to the detriment of native vegetation. Our goal is to push this alternate stable state back towards a state dominated by native woody plants by disrupting the feedbacks that maintain an established RCG monoculture.

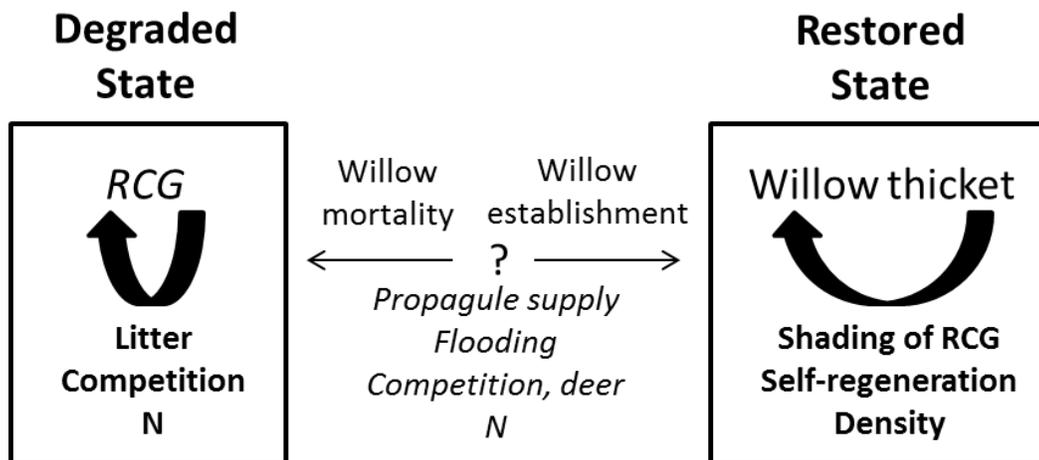


Figure 1. Summary of mechanisms from our proposed model. Several feedbacks keep RCG in a stable state including litter accumulation, competition and elevated nitrogen (N) levels. Manipulation of propagule supply and competition could promote the establishment of native plants such as willows. Willow thickets will be short-lived, however, unless the feedbacks that maintain a willow thicket are facilitated: canopy development, self-regeneration, and high density of willow individuals to discourage the re-invasion of RCG.

CHAPTER II

**WILLOW STAKE PLANTINGS AND GLYPHOSATE MAY DISRUPT
FEEDBACKS THAT MAINTAIN REED CANARYGRASS (*PHALARIS
ARUNDIACEA* L.) MONOCULTURES IN THE UPPER MISSISSIPPI RIVER
FLOODPLAIN FOREST**

Introduction

Invasive species are one of the primary causes for the loss of native species biodiversity in many ecosystems (Vitousek et al. 1997). Invasive plant species are capable of increasing their spatial distribution by expanding into existing plant communities – typically producing offspring in very large numbers and at large distances from parent plants (Richardson et al. 2000). Because of this, controlling many invasive plant species proves to be a tremendous challenge to land management agencies. The management strategies implemented are continuously evolving based on ecosystem dependent factors, with novel approaches in high demand. Management of invasive plant species in wetlands is particularly challenging (Galatowitsch et al 1999, Zedler and Kercher 2004).

One of the most aggressive wetland invaders in the temperate U.S. is reed canarygrass (*Phalaris arundinacea* L., hereafter RCG), a cool-season grass that is problematic in the Pacific Northwest (Kim et al. 2006), the upper Midwest (Knutson et al. 2005) and Southeastern United States (Foster and Wetzel 2005). For example, in Wisconsin wetlands, RCG dominates (> 80 % cover) more than 40,000 ha (98,842 acre;

Bernthal and Willis 2004). RCG is capable of quickly invading forested areas following canopy disturbances, and once established, quickly forms monocultures (Urich et al. 2002). Once RCG monocultures are present, methods of restoring have not yet proved successful to return the ecosystem to its original condition, forming an alternate stable state (Zedler 2009).

Feedbacks that maintain RCG monocultures must be disrupted to push the ecosystem back from the degraded (heavily invaded) state into an unstable state, which can then be restored (Suding and Hobbs 2009, Zedler 2009, Annen 2011); this process is lengthy, complex, and expensive (Annen 2011, Lavergne and Molovsky 2006). At the propagule level, RCG creates a long-lived seedbank with seeds capable of remaining dormant for three years (Zedler and Kercher 2004). Furthermore, once established, RCG forms a dense system of rhizomes from which the plant re-sprouts each year (Coops et al. 1996). Elevated nitrogen (N) levels also accelerate RCG spread while decreasing native wetland vegetation (Green and Galatowitsch 2002, Herr-Turoff and Zedler 2005). Monocultures of RCG grow quite tall and thick, with a dense litter layer – shading native plants that could potentially compete with RCG after they develop past their seedling states (Annen 2011). This effect is bolstered by the fact that RCG is a C3, or cool-season, grass that is productive at the beginning of the growing season, creating a canopy before other plants, particularly seedlings, are capable of beginning growth.

One ecosystem that is under heavy pressure from RCG invasion is the Upper Mississippi River (UMR) floodplain forest. Prior to the establishment of the lock and dam system in the 1930s, the UMR was fairly continuous with a defined floodplain. Aerial photographs from 1929 and 2010 show how lower pool regions were altered from

a continuous forested area, becoming a series of backwater islands as a result of rise in water level (Figure 2). Even in mid- and upper-pool sites, forest senescence appears to be followed by RCG invasion, rather than the recruitment of new trees to the canopy. Land managers in the UMR are therefore seeking reforestation methods for areas invaded by RCG (Urich et al. 2002, Romano 2010).

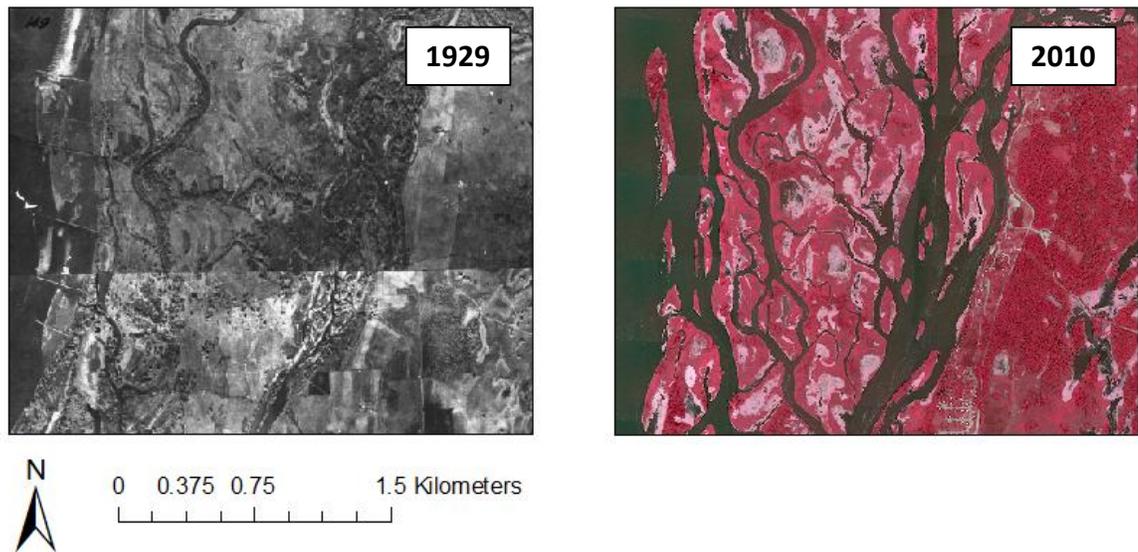


Figure 2. Upper Mississippi floodplain south of La Crosse, WI in 1929 and 2010. Construction of the lock and dam downstream created a radically altered environment from the wooded and connected area seen in 1929. By 2010, the area is a series of marshy islands connected by backwater sloughs. The 1929 image is a historic aerial photo mosaic (courtesy of Army Corps of Engineers and the 2010 image is an orthoreferenced color infrared image (courtesy of USGS).

Several studies have found that it is possible to restore RCG monocultures by treating the area with herbicide and then introducing native plant species (Kellogg and Bridgham 2004, Kim et al. 2006, Hovick and Reinartz 2007, Thomsen et al. 2012). However, research is needed to determine the best timing for herbicide application, and to assess when and how to plant native wetland plant competitors to effectively reduce RCG % cover. Systemic herbicides have been found to be twice as effective against RCG when applied in fall rather than in spring (Reinhardt Adams and Galatowitsch

2006). Because carbohydrates are being transported into rhizomes for storage in fall, systemic herbicides applied at this time also translocate more efficiently into belowground tissues, causing greater damage and limiting the early-season growth of RCG; carbohydrate flux and herbicide effectiveness have been documented with purple loosestrife (*Lythrum salicaria* L.; Katovich et al. 1998). This gives native plant species a chance to establish early in the growing season, a time that RCG usually dominates. Furthermore, one study found that herbicide application to RCG following the first killing frost is very effective (Hovick and Reinartz 2007).

When selecting wetland native plant competitors for RCG, water-loving plants that are tolerant of waterlogging, flood flow stress, and sediment deposition are ideal. Willows fit these requirements, and are easily planted as stakes in remote areas. The technique of staking willows, also known as “live staking” (Gray and Sotir 1996), has been used to stabilize streambanks and in other bioengineering techniques for hundreds of years (Ying et al. 2011). Live staking is possible because willows regenerate from root and shoot fragments; when a lateral stem is cut from a willow, and staked into the ground, it is capable of growing into a new adult plant. In spite of the ease of willow staking, ideal methods for willow staking are poorly understood (Pezeshki et al. 2007, Caplan et al. 2012). Willow sprouting and long term survival varies depending on site conditions such as competition with exotics and herbivory (Pezeshki et al. 2007). A number of studies are beginning to look at how vegetative stake height affects sprouting and survival (Zahawi 2008), however, little data exist on the effect of willow stake height on short and long-term survival.

The first objective of our study is to evaluate the effect of fall applications of glyphosate in a RCG monoculture. A broad-spectrum herbicide should effectively disrupt the feedbacks created by the early season growth of RCG, allowing other species to compete early in the growing season. This should be evident by decreased RCG canopy percent cover and belowground biomass, improving conditions for native plant competition. Our second objective is to analyze the effect of herbicide application, planting date (fall vs. spring) and stake height (1.52 vs. 0.91 m, 5 vs. 3 ft) on the sprouting and survival of planted willow stakes, to determine optimum planting strategies for willow stakes in RCG. Finally, we will determine whether or not established willow shrubs are capable of out-competing RCG. The willow plantings could disrupt the shading effect created by the RCG canopy, potentially aiding in the recruitment of other native species.

Methods

Site

The study site is on an island in a backwater channel of the Upper Mississippi River (UMR) south of La Crosse, Wisconsin, U.S.A. (43°44'19"N, 91°13'53" W; Figure 3). The site elevation ranges from approximately 632.5 to 634.0 m (2,075 to 2,080 ft) and is subject to frequent flooding. Over the last 30 years, the average annual flood duration at the site was 41 days out of a 180 day growing season, meaning the site was flooded 23% of the time from April through September (De Jager et al. 2012). Over-story growth diversity in the UMR tends to decline when flood duration increases from 0 to 40% of the growing season, with a single species, silver maple (*Acer saccharinum* L.), dominating at sites inundated for greater than 40% of the time (De Jager et al. 2012).

Areas inundated less than 40% of the time typically have a comparatively more diverse forest community with a wider range of species compositions. Thus, based on our site's 23% annual flood duration, it should be capable of supporting a diverse floodplain forest.

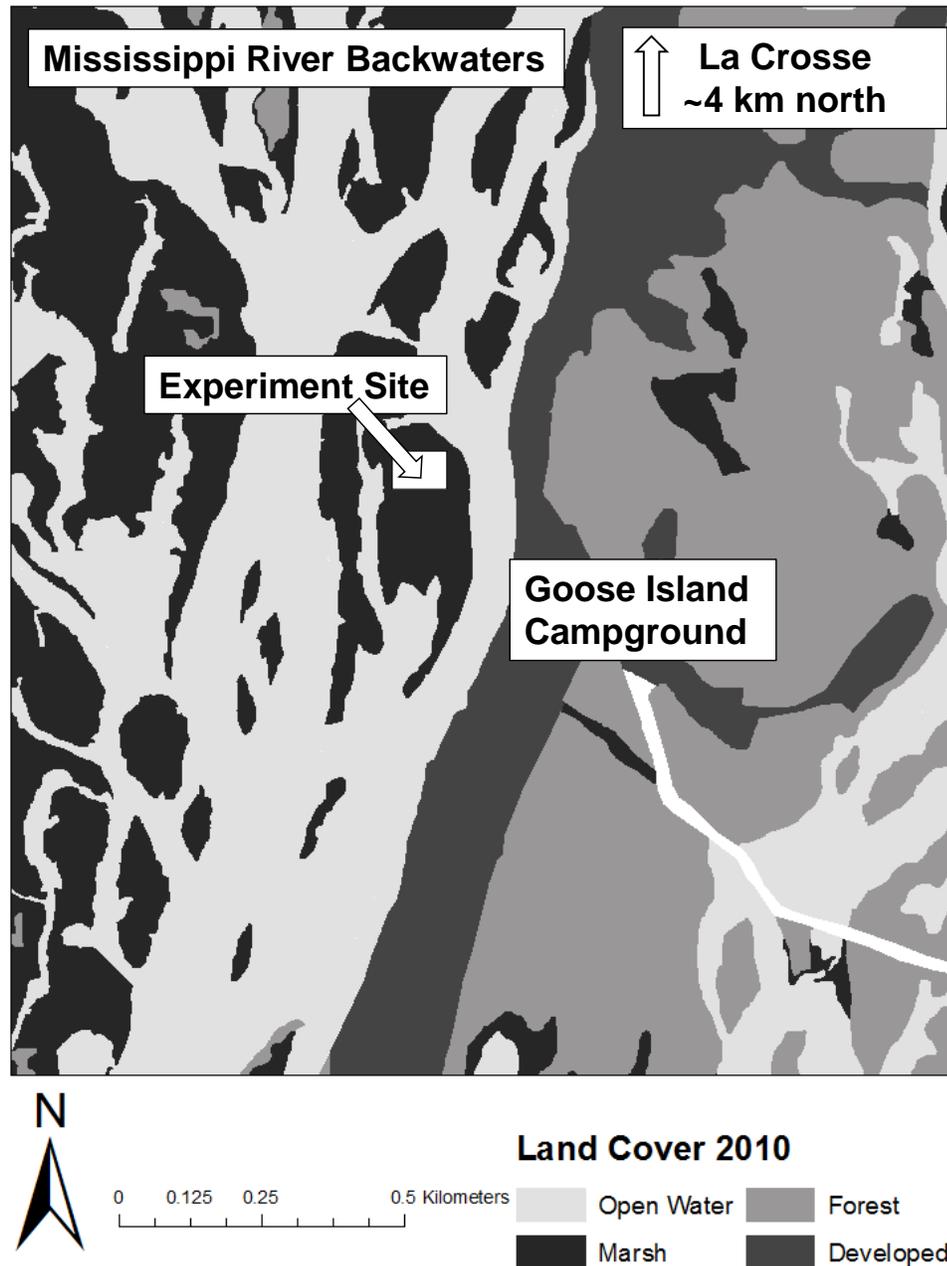


Figure 3. Map of experimental site in the Upper Mississippi River floodplain. The site is located on an island across from the Goose Island Landing and Campground, south of La Crosse, WI (GIS landcover data for map courtesy of USGS).

Plots were established in the northern portion of the island. At the time of plot establishment the experimental area was dominated by RCG (voucher specimen deposited in the University of Wisconsin, La Crosse herbarium). Thatch depth of the grass at the site, outside of experimental plots, was 21 cm and average biomass increased from 18 g m⁻² (160 lb acre⁻¹) at the beginning of the growing season (April 2012) to 394 g m⁻² (3,509 lb acre⁻¹) at peak growth (July 2012; *unpublished data*). Other plant species primarily inhabit the perimeter of the island including arrowhead (*Sagittaria sp. L.*), bulrush (*Shoenoplectus sp. Torr.*), duckweed (*Lemna sp. L.*), willow (*Salix sp. L.*), and a single silver maple (*Acer saccharinum L.*). Terrestrial vertebrates observed at the site include meadow voles (*Microtus sp. Ord.*), muskrats (*Ondatra zibethicus L.*), frogs (order Anura), and turtles (order Chelonii). Avian species observed in the vicinity include red-winged blackbird (*Agelaius phoeniceus L.*), red-tailed hawk (*Buteo jamaicensis Gmelin*), great blue heron (*Ardea Herodias L.*), great white egret (*Ardea alba L.*), and bald eagle (*Haliaeetus leucocephalus L.*).

Experimental Design

Eighty 1.5- by 1.5-m (4.92- by 4.92-ft) plots with a 1.5 m (4.92 ft) buffer zone between plots, were randomly assigned to 8 replicates of 10 treatments (Table 1). Half of all plots were sprayed with glyphosate in October 2011 and November 2012. Sixteen plots were left unplanted while the remaining 64 plots have willow stake plantings. Willows were planted either in the fall (32 plots) or spring (32 plots). The unplanted plots consist of half unsprayed plots and half glyphosate-treated plots, allowing us to monitor RCG growth in the presence and absence of glyphosate. Planted plots have five willows: one in the center and four equidistant from the center willow and the plot corners (Figure

4). Of the 64 planted plots, 32 plots have tall willow stake plantings cut to lengths of 1.52 m (5 ft), while the other 32 plots have short willow stakes cut to lengths of 0.91 m (3 ft).

Table 1. Description of plots by treatment at the site. All plots are 2.25 m² (24.2 ft²) in size. Treatment combinations can be read from left to right, with the number of plots after each treatment type in parentheses. For example, there are 32 plots that were planted in fall, and out of those 32 plots, 16 were sprayed with glyphosate. Out of those 16 that were sprayed with glyphosate, 8 were planted with short and 8 with tall willow stakes.

Planting date	Herbicide	Stake height
Fall (32)	Spray (16)	Short (8)
		Tall (8)
	No spray (16)	Short (8)
		Tall (8)
Spring (32)	Spray (16)	Short (8)
		Tall (8)
	No spray (16)	Short (8)
		Tall (8)
Unplanted (16)	Spray (8)	
		No spray (8)

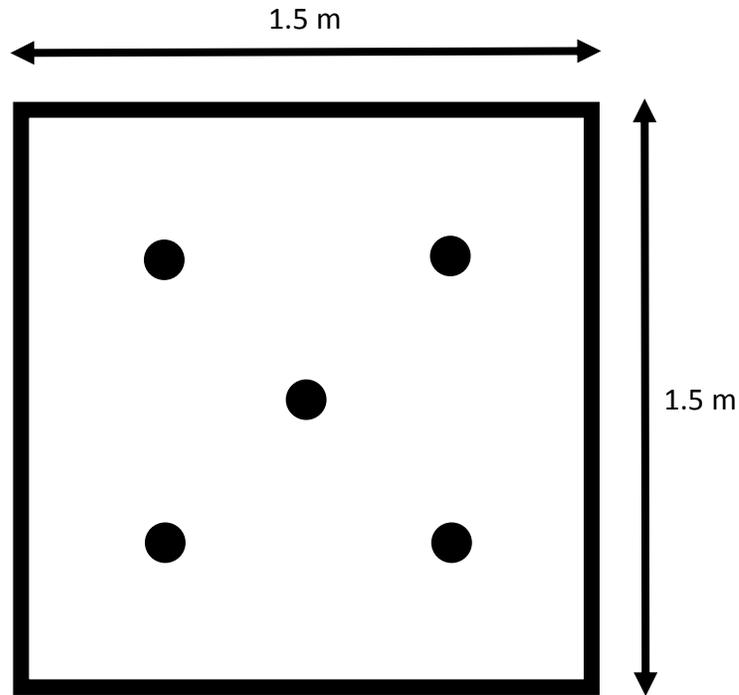


Figure 4. Plot design. Plots were squares 1.5 m (4.92 ft) on a side. Planted plots had five willow stakes. The willow stakes were arranged as seen by the black circles, with one in the center of the plot and the other four between the center of the plot and each of the four corners.

For glyphosate herbicide treatments, a calibrated backpack Solo® Economy 3 gallon backpack sprayer (Newport News, VA) was used in good spraying conditions in 2011 and 2012. These conditions included lack of wind, sunny skies, and relatively warm temperatures to maximize treatment effectiveness and prevent herbicide drift. Glyphosate (Roundup Concentrate®, Monsanto, St Louis, MO) was mixed as 2.5 fluid ounces of 18% glyphosate per gallon of water and applied to the 40 plots included in the herbicide treatment. Live willow cuttings were obtained from a sandbar willow (*Salix exigua* Nutt.) stand near the experiment site. This species was used because it provides a fast-growing shrubby canopy and exhibits vigorous growth at low elevations close to the water (Amlin and Rood 2001). Willow stakes were cut using reference tall and short

stakes of 1.52 m (5 ft) and 0.91 m (3 ft) respectively, with diameters of approximately 15.5 mm (0.6 in). Once all 160 short and 160 tall willow stakes were cut, they were immediately transported to the site and planted by pushing them 30.5 cm (1 ft) into the ground – making the final aboveground height of the tall and short willow stakes 1.22 m (4 ft) and 0.61 m (2 ft), respectively. Once planted, the base of each stake was wrapped with 15.2 cm (6 in) tall 1.3 cm (0.5 in) mesh Spectra® gutter screen to provide protection from rodent herbivory. Fall willow stakes were planted five days following herbicide application, in November, 2011; spring willow stakes were planted in April, 2012. Because fall planted willow stakes had poor sprouting during the 2012 growing season, we removed all fall-planted individuals and repeated the fall planting in November 2012 using the same methods as for the original planting.

Data Collection

During the 2012 growing season we quantified RCG performance, willow stake establishment and performance, and site conditions. We visually estimated RCG percent cover during the months of April, May, June, and August 2012 (high water prevented our access to the site in July 2012). Belowground biomass was measured in the months of May, June, and August by collecting a single root core in each unplanted plot; to observe the growth of RCG in the presence and absence of glyphosate. Cores were collected using a bulb planter (10 cm height by 28 cm² area; 3.94 in by 4.35 in²). Root cores were refrigerated until being washed in deionized water over a 2.00 mm (0.08 in) sieve to isolate RCG roots and rhizomes. Once cleaned, belowground biomass was dried for a minimum of 48 hours at 60°C (140°F) and weighed to determine belowground biomass per core. Willow establishment was quantified by measuring the number of willows that

had sprouted by May 2012. Willow performance was quantified by measuring the heights and canopy spread (lengths of two longest stems added together) of surviving willows in August. Soil nitrogen was measured in July 2012 using ion exchange resin probes (PRS probes, Western Ag Innovations, Saskatoon, Saskatchewan). Three anion and three cation probes were placed in each unplanted plot for three weeks. They were then collected, washed, and sent in for analysis.

In 2013 we were not able to start data collection at the site until July due to lengthy site inundation. RCG percent cover and belowground biomass were collected in July and August using the same methods as 2012. In July, we also determined the number of surviving willows from the spring 2012 planting, and the establishment of the stakes planted in November 2012. In August 2013 we quantified willow performance using the same methods as in 2012. Finally, the availability of photosynthetically active radiation (PAR) was measured outside of the plot, under the willow canopy (if present) and at the soil surface at the center of each plot using a hand-held quantum sensor (LI-COR Environmental, Lincoln, NE) in August 2013. Light measurements were made in all plots with surviving willows (11 plots) and in 11 randomly-selected plots without willows.

Analysis

Data analysis was performed with JMP® 10 (SAS Institute Inc., Cary North Carolina). RCG percent cover and belowground biomass data were square root transformed to normalize distributions and equalize variances. Percent cover and belowground biomass were analyzed in the unplanted plots using a repeated measures ANOVA to determine the effect of glyphosate over time on RCG growth. When the

time*treatment effect was significant, the effect of treatment was examined for each month using t-tests and a Bonferroni adjustment. Data for the number of established willows in May 2012 were not normally distributed, so non-parametric Wilcoxon tests were used to evaluate the effect of herbicide, planting date, and stake height treatments separately. A Bonferoni adjustment was then applied to account for multiple comparisons. Due to poor sprouting of fall plantings, we did not include them in the willow performance analyses. ANOVA was used to test the effect of glyphosate on individual willow height and canopy spread in 2012 and 2013. Plot was included as a random effect in these analyses to avoid pseudoreplicating observations of individual willows within plots. ANOVA was used to compare total nitrogen between plots where herbicide was applied verses unsprayed plots. Light data were non-normal so we performed a non-parametric Wilcoxon test on percentage of ambient light reaching the soil surface in plots where willows survived vs. plots without willows.

Results

Reed Canarygrass Performance

Surveys of RCG % cover in unplanted plots indicated that application of glyphosate significantly affected RCG % cover during 2012, but that effect varied over time (Figure 5, Table 2). RCG % cover generally increased over the growing season. During the months of April, May and June there was significantly lower RCG % cover in the plots where glyphosate was applied. By August the effect of the fall glyphosate application had disappeared and all plots all had close to 100% cover of RCG. During the 2013 season, herbicide treatment had no effect on RCG % cover at the time points sampled.

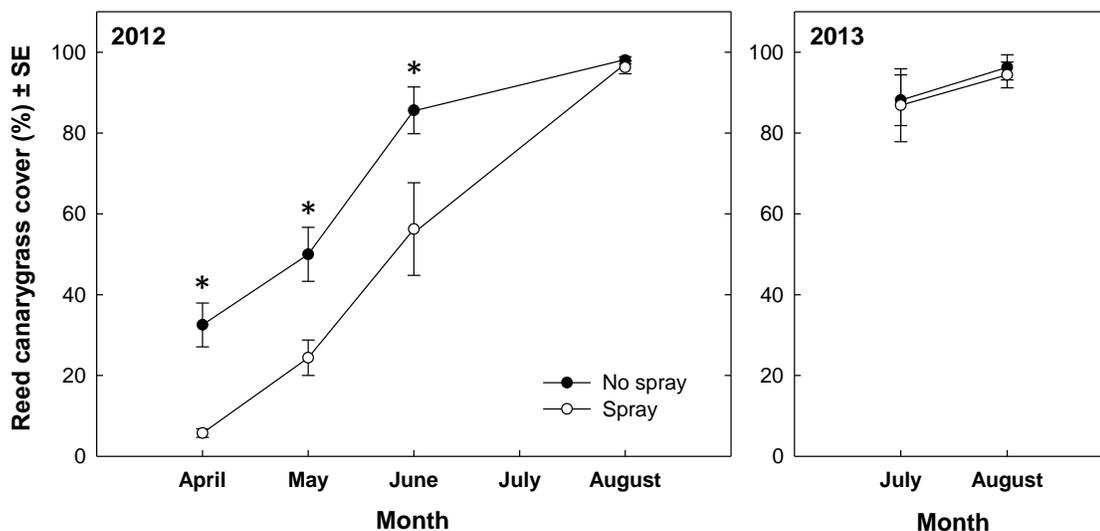


Figure 5. RCG % cover in plots without willow stakes, April – August 2012 and July – August 2013. Glyphosate application the previous fall significantly decreased RCG % cover in the months of April, May and June 2012 (indicated by *). There was also a significant increase in RCG % cover through time during the 2012 season. No effect of glyphosate was apparent at the 2013 sample time points.

Table 2. Results of a repeated measures ANOVA for the relationship between herbicide, mean RCG % cover and time in 2012 and 2013.

Year	Treatment	F	P
2012	Herbicide	$F_{1,14} = 18.93$	0.0007
	Time	$F_{3,12} = 152.95$	< 0.0001
	Time*Herbicide	$F_{3,12} = 10.74$	0.0010
2013	Herbicide	$F_{1,14} = 0.08$	0.7878
	Time	$F_{1,14} = 3.70$	0.0751
	Time*Herbicide	$F_{1,14} = 0.01$	0.9100

In 2012, belowground biomass generally decreased throughout the growing season (Figure 6, Table 3). However, there was no significant effect of the herbicide treatment on biomass. In 2013, glyphosate significantly decreased belowground biomass, but the biomass densities were similar at both time points sampled.

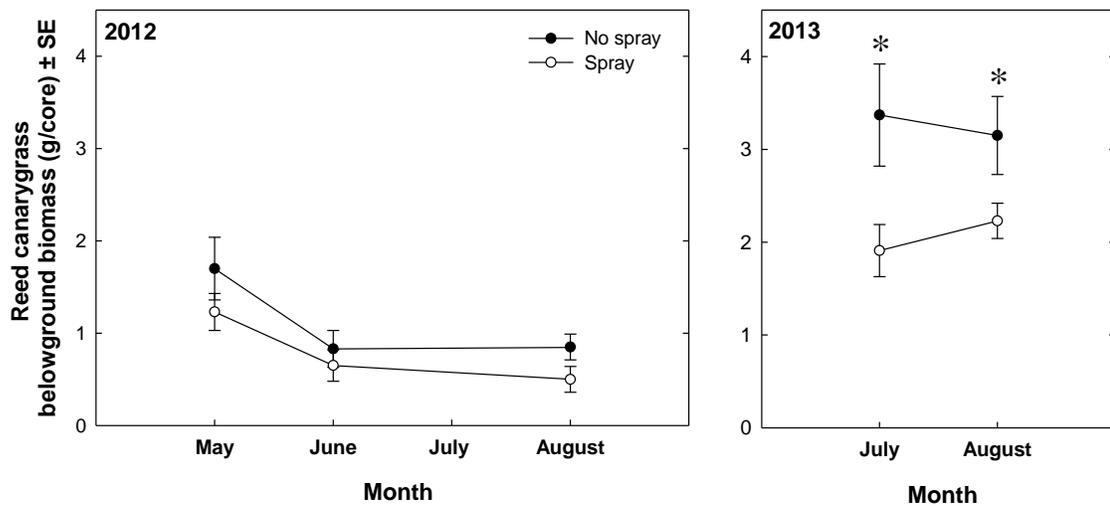


Figure 6. RCG belowground biomass. Belowground biomass (g/core) as sampled with soil cores 10 cm height by 28 cm² in size. Soil cores were collected in May – August 2012 and July – August 2013, in plots where no willows were planted. In 2012, belowground biomass decreased significantly over time (Table 2). In July and August 2013 belowground biomass was significantly lower in plots in which glyphosate was applied the previous fall (Table 3).

Table 3. Results from repeated measures ANOVAs for the relationship between herbicide, RCG belowground biomass and time in 2012 and 2013.

Year	Treatment	F	P
2012	Herbicide	F _{1,14} = 2.90	0.1105
	Time	F _{2,13} = 0.42	0.0042
	Time*Herbicide	F _{2,13} = 8.60	0.6685
2013	Herbicide	F _{1,14} = 14.01	0.0025
	Time	F _{1,14} = 0.20	0.6582
	Time*Herbicide	F _{1,14} = 0.38	0.5383

Willow Performance

Willow stakes that were planted in spring 2012 established at a significantly higher rate than those planted in fall 2011 ($\chi^2_1 = 30.94$, $P < 0.0001$; Figure 7, Table 4). For spring-planted stakes, 56% established, compared to 8% of the fall-planted stakes. Neither stake height nor herbicide had a significant effect on willow establishment.

Repeating the fall planting in 2012 resulted in a similar pattern of low establishment, with only 5% of fall 2012 willow stakes becoming established as sprouted plants the following spring (Table 4).

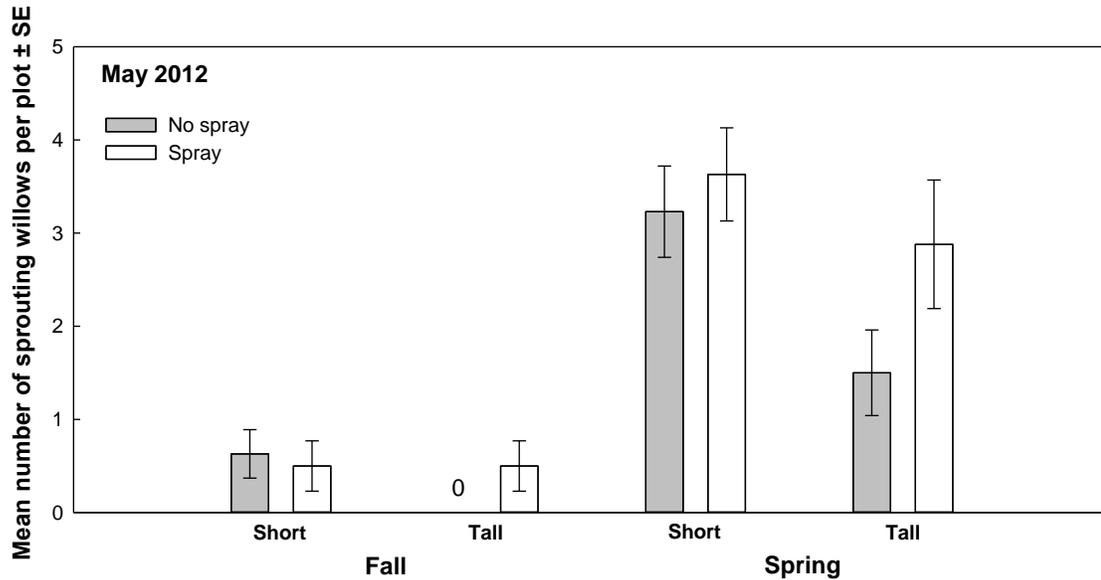


Figure 7. Willow stake establishment. Average number of established willow stakes per plot in May 2012 in all treatments. Five stakes were planted per plot; willow stakes planted in spring had significantly higher establishment than those planted in fall, but stake height had no effect on willow establishment in 2012. The “0” indicates no willow establishment in the plots that were planted in fall with tall willow stakes. Results for fall-planted stakes were similar for the second year of planting.

Table 4. Wilcoxon tests were performed on willow establishment data in 2012 and in 2013 due to its non-normal distribution. The effect of each experimental treatment on the number of willows establishing per plot was analyzed separately and significance of each factor determined using a Bonferroni adjustment for multiple comparisons (significant values in bold).

Date	Treatment	χ^2	P
2012	Planting date	30.94	< 0.0001
	Stake height	3.72	0.0538
	Herbicide	1.23	0.2665
2013 (second round of fall stakes only)	Stake height	4.41	0.0358
	Herbicide	0.0097	0.9216

Among the spring-planted willows surviving until August 2012, mean height and canopy spread were significantly greater in plots where glyphosate was applied (Figure 8, Table 5). Willow heights in the plots where glyphosate was applied vs. those in the unsprayed plots differed by an average of 18 cm (7.09 in). The surviving willow individuals in August 2013 had similar heights and canopy spreads in plots where glyphosate was applied and in the unsprayed plots. In the unsprayed plots, 38 willows survived to August 2012 (48% of those originally planted in spring plots), while 52 (65%) survived in plots where glyphosate was applied. As of August 2013, survivorship in unsprayed plots had dropped to 8 (10% of those originally planted) and 19 (24%) in glyphosate sprayed plots.

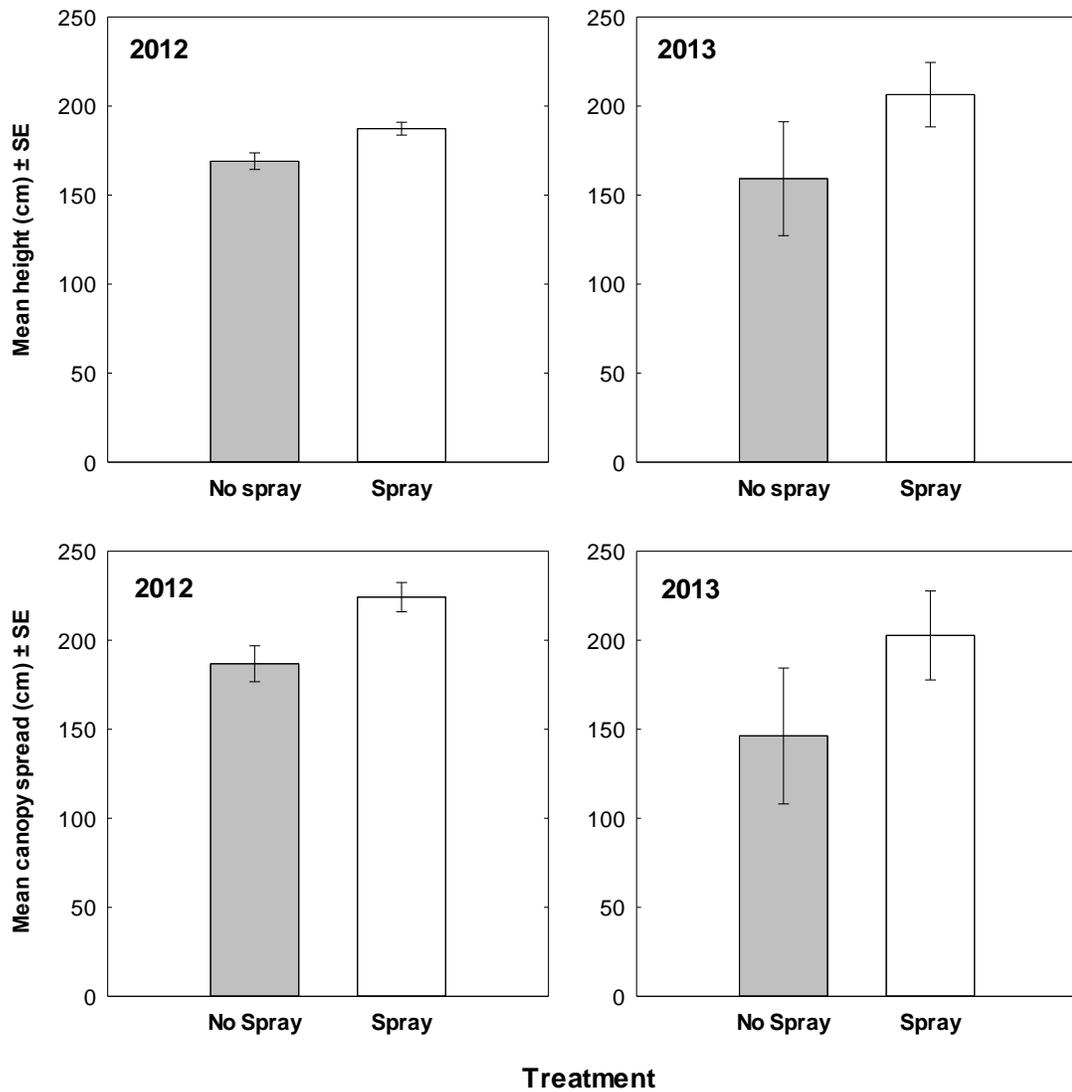


Figure 8. Performance of surviving willow. Mean willow height and canopy spread of all surviving spring-planted willow individuals in August 2012 and 2013, in glyphosate treated vs. unsprayed plots. In 2012, willows were significantly taller and had greater canopy spread in plots where glyphosate was applied (N=77, Table 4). However, in 2013 willow heights and canopy spreads were similar in plots where glyphosate was applied and in unsprayed plots (N=27).

Table 5. Effect of glyphosate on the height and canopy spread of surviving spring-planted willows in August 2012 and 2013. Plot was included as a random effect in the model.

Date	Measurement	F	P
2012	Height	$F_{1,22.8} = 9.84$	0.0047
	Canopy Spread	$F_{1,26.5} = 6.09$	0.0204
2013	Height	$F_{1,14.5} = 2.036$	0.1750
	Canopy Spread	$F_{1,14.5} = 1.58$	0.2281

Site Conditions

Our site was flooded for 33% of the first growing season (April 1 – September 30, 2012) and 61% of the second (2013). Soil nitrogen levels in unplanted plots in July 2012 were similar in unsprayed plots and plots where glyphosate was applied ($F_{1,14}=0.0003$, $P=0.9870$). In August 2013, plots where willows were planted had significantly greater light levels at the soil surface than in unplanted plots (Figure 9, $\chi^2_1 = 8.93$, $P = 0.0035$). RCG blocked almost all ambient light from reaching the ground level (percent of ambient light at soil surface = 0.13%) while willows allowed significantly more light to reach the ground level (1.13%). In comparison, willows allowed about half (49.45%) of the ambient light to penetrate their canopies, as measured at the top of the RCG canopy.

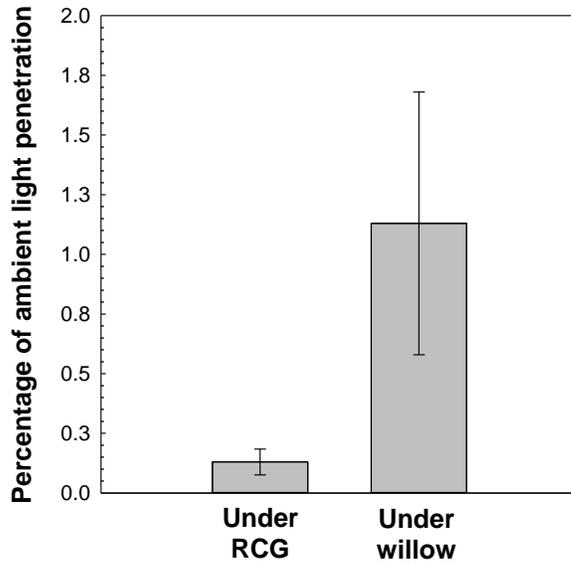


Figure 9. Percentage of ambient light \pm 1SE that reaches the ground in plots with willow survivors (under willow) and non-planted plots (under RCG). Willow canopies allowed a significantly larger % of ambient light to reach the ground than plots where willows were not planted.

Discussion

Late fall glyphosate application inhibited early season growth of RCG (Figure 5, Table 2), one of the primary feedbacks that maintain RCG monocultures (Zedler 2009). RCG % cover was lower in plots where glyphosate was applied during the first three months (April, May, and June) of the growing season in 2012. Although plots with and without the glyphosate treatment had similar % cover at the end of the growing season, inhibiting RCG at the beginning of the growing season is crucial. Increased light at the ground level early in the growing season would potentially aid germination of native wetland plant seeds and would encourage seedling establishment before the RCG canopy closes. However, increasing ground light level will only aid in native species establishment while RCG is suppressed in the presence of plentiful native propagules; research has found that increased ground light availability also leads to an increase in RCG invasion (Kercher et al. 2007). Studies are needed to see how ground light levels

affect native seedling establishment in RCG monocultures where RCG is being suppressed via herbicide. Recruitment of native plants and lowered RCG percent cover may lead to decreased lower amounts of RCG litter, a feedback that promotes RCG monoculture formation (Zedler 2009, Annen 2011), in the following growing season, further aiding native seedling recruitment. Unfortunately, we were not able to measure that level at the start of the following growing season due unanticipated site flooding through June 2013.

Repeated fall glyphosate application decreased the belowground biomass of RCG during the 2013 growing season (Figure 5, Table 3). Belowground biomass is another feedback that maintains RCG dominance in monoculture, not only because RCG resists removal due to its underground rhizomes (Zedler 2009), but also because RCG reproduces through its rhizomes, increasing RCG monoculture expansion (Coops et al. 1996, Kercher et al. 2007). RCG has been documented to have higher belowground competitive abilities than many native species, to the point where RCG suppresses the root growth of native communities (Lavergne and Molofsky 2004). Furthermore, if the herbicide application increases control each following year, it could be both biologically effective as well as cost effective. Although our data indicate that glyphosate limits the growth of RCG rhizomes and roots, belowground biomass was also higher in all treatments during our second season of sampling than the first. Our site was flooded for extended periods during the second sampling season, submerging all plants. Complete submergence in seasonal floodplains is a primary selective pressure due to lower oxygen availability in the root zone (Qin et al. 2013). Research found that RCG carbohydrate stores decrease with increasing submergence time (Qin et al. 2013), suggesting that

extended inundation should lead to RCG die-off (Stannard et al. 2001, Wisconsin RCG Management Working Group 2009, Qin et al. 2013). However, we observed the opposite pattern, with greater belowground biomass during the second growing season, which was flooded during the majority of the season (61%). It is likely that slight differences between sampling and processing techniques of the first and second year could account for the difference. It is also possible, however, that the assumption of rhizome die-off due to extended inundation may not apply to RCG growing in the UMR floodplain. There is a need for data quantifying the effect of extended inundation of RCG belowground systems across multiple sites.

Planting live willow stakes had varied results. Fall plantings had significantly lower sprouting than spring willow stakes, across two years of planting (Figure 5, Table 3). Thus, we conclude that sandbar willow stakes are more likely to sprout and develop into shrubs if they are planted in the spring. Willow stakes have been shown to have good survival in the years following their initial establishment (Pezeshki et al. 2007). However, there was a decline in survival of our sprouted willows from 2012 to 2013. We observed both caterpillars and vole damage on willow shrubs in the 2013 growing season, and the extended inundation of the site in 2013 may have also harmed the willows.

Plots where glyphosate was applied had significantly taller willows with greater canopy spread during the first growing season, perhaps as a result of early season RCG suppression. Ultimately, however, the differences in height observed in the first year were relatively minor, and the two-year survival rate for planted stakes relatively low, even in glyphosate-treated plots (approximately 25%). Survival and growth of willow cuttings is influenced by elevation relative to the water table and sediment texture

(Pezeshki et al. 2007), two components that we did not explicitly address in our study. Research is needed on how willow survival and growth in RCG monocultures is affected by sediment texture and water gradient across different sites. Furthermore, little literature exists on the survivorship of sandbar willow; most quantify survival and performance of black willow stakes. In order for planting live sandbar willow stakes to be a successful restoration technique, they should be planted in large numbers in spring, just as the growing season begins. Long-term survival of stakes is low as well, so it is recommended that dead stakes be replaced each following spring, until enough willow shrubs have been developed to provide a large enough propagule source to sustain a willow thicket. In alternative states theory, three natural states can exist (Zedler 2009), in this case the invaded state (RCG monoculture), the remnant state (floodplain forest), and the restored state (willow thicket). Zedler (2009) states that the invaded state is not always reversible, and that the restored state is not necessarily permanent in time. Thus, continued stake replacement may enforce feedbacks that maintain the restored, willow thicket, state.

RCG is particularly sensitive to shading and vulnerable to competition for light (Lindig-Cisneros and Zedler 2001). Willow shading reduced the RCG canopy, a result seen as an increase in the amount of ambient light reaching the ground in plots with willows as compared to those without them (Figure 7, Table 5). Increase in ground light level could improve growing conditions for newly recruited native seedlings. We did find silver maple seedlings in two willow plots, which could possibly be an outcome of willow plantings aiding in the recruitment of floodplain forest tree seedlings. If seedlings can establish, they could eventually become trees- which would not only further inhibit

RCG growth through shading and soil drying, but would increase native propagule recruitment as well.

Wetlands are highly subject to invasion due to flooding that causes accumulation of water, nutrients sediments, and invasive plant propagules that allow invasive plants to establish, displace native species, and expand (Zedler and Kercher 2004, Kercher et al. 2007). RCG, is the most noxious invader in Wisconsin (Bernthal and Willis 2004), and quickly moves into areas with available light, creating monocultures. Monocultures of RCG are maintained by feedbacks including early-season growth (Zedler 2009), a developed RCG canopy that out-shades native species (Kercher et al. 2007), a prolific propagule source in the form of an extensive belowground rhizome system (Coops et al. 1996), and a seedbank that lasts for years (Annen 2011), making it unlikely that any single restoration treatment will be able to eliminate a RCG monoculture. We succeeded in reducing RCG canopy and % cover leading to greater ground light penetration using willows and glyphosate. This has the potential to benefit the recruitment and development of native floodplain plant species. Continual use of these methods over more years may successfully return RCG monocultures in the UMR into willow thicket states, and possibly young forest ecosystems in the future. As willows proved to suppress the RCG canopy, sowing additional native seed mixes into plots where the RCG canopy is competing for light levels with developed willow shrubs could further restore the native community. Our site was on an island of RCG monoculture, while other studies large natural propagule sources because they were floodplain forest-adjacent (Hovick and Reinartz 2007, Thomsen et al. 2012). If a shrubby community continues to develop it may be beneficial to frequently apply spot systemic grass-specific herbicides to RCG.

While willows may open the door to the decline of a RCG monoculture, we need efficient methods that encourage the recruitment of large sources of propagules into the newly developed-thicket.

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