Effects of Alligatorweed Control in Seasonal Wetlands Managed for Waterfowl

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Abstract: Wildlife managers commonly use herbicides to control invasive plant species and maintain early-successional vegetation communities in seasonally flooded moist-soil wetlands. However, there is limited information on how herbicides influence plant and animal communities following application. Thus, we investigated the response of vegetation, food density, and the abundance and activities of dabbling ducks (Anatini) to application of imazapyr herbicide in moist-soil wetlands in Tennessee to control invasive alligatorweed (*Alternanthera philoxeroides*). Imazapyr was applied topically at Tennessee National Wildlife Refuge during July 2011 following an early water drawdown and dry weather conditions. Food density and use and activities of dabbling ducks were similar between treatment and control plots during the year of application and in the subsequent year. Dabbling duck use was negatively related to increasing water depth during winter, independent of treatment type. Imazapyr treatments reduced canopy cover of alligatorweed in the year of and one-year post-treatment in most plots, but repeated herbicide applications and a more integrated approach are likely needed for long-term control in moist-soil wetlands where alligatorweed is established. Using an integrated pest management approach to control invasive species in moist-soil wetlands is important for National Wildlife Refuges to meet their waterfowl energy objectives in support of the North American Waterfowl Management Plan.

Key words: dabbling duck, herbicide, imazapyr, mallard, moist-soil management

Journal of the Southeastern Association of Fish and Wildlife Agencies 9: 105-113

Management of seasonally flooded, herbaceous wetlands (i.e., moist-soil management) is commonly used to promote early-succession vegetation communities that provide food and other habitat resources for waterfowl and other wetland-dependent wildlife (Low and Bellrose 1944, Fredrickson and Taylor 1982). Moist-soil management involves dewatering wetlands during late spring and summer to encourage grasses, sedges, and forbs that produce food for waterfowl (Gray et al. 2013). Seeds, tubers, and aquatic invertebrates within moist-soil wetlands provide a diverse suite of nutrients that complement high-energy agricultural crops used in state wildlife management areas and federal National Wildlife Refuges (NWR) to meet energy and habitat objectives for waterfowl (Hagy et al. 2020).

Moist-soil management often includes physical practices (i.e.,

mowing, disking) or use of herbicides to control invasive plants that displace desirable species and reduce food production for waterfowl (Madsen 1997, Holmes 2002, Strader and Stinson 2005). Unfortunately, some mechanical practices can spread invasive species and make the problem worse in the future, such as in the case of alligatorweed (*Alternanthera philoxeroides*). Alligatorweed is an exotic perennial macrophyte that has no known food value for waterfowl and can outcompete native aquatic vegetation in the southeastern United States (Quimby and Kay 1977, Buckingham 1996, Holm et al. 1997). Alligatorweed spreads vegetatively, forming dense mats that shade out desirable vegetation and obstruct water control structures, which reduces management capabilities (Vogt et al. 1992; U.S. Fish and Wildlife Service 2007, 2010). Alligatorweed poses a bit of a paradox in that traditional moist-soil

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management practices, such as disking and mowing, can invigorate alligatorweed by facilitating vegetative reproduction that hinders competition from desirable species (Holm et al. 1997, Strader and Stinson 2005). Thus, land managers currently use herbicides and, to a lesser degree, biological agents (e.g., *Agasicles hygrophila*) to control alligatorweed in managed wetlands in the Southeast (Tucker et al. 1994, Buckingham 1996, Allen et al. 2007). However, persistence and perpetual reoccurrence of alligatorweed in some wetlands is severe enough to limit vegetation management options leaving only active management (e.g., agriculture rotation and herbicides) as the viable option for control and early-succession vegetation management (U.S. Fish and Wildlife Service 2007, 2010, 2014).

Previous studies have suggested imazapyr, a broad-spectrum post-emergent herbicide, is effective at controlling alligatorweed (Tucker et al. 1994, Allen et al. 2007, Hofstra and Champion 2010). Other herbicides, with active ingredients such as glyphosate, triclopyr, picloram, and 2,4-D amine are either ineffective or have application restrictions that make them suboptimal for alligatorweed control on NWRs (Bowmer et al. 1993, Tucker et al. 1994, Hofstra and Champion 2010). Adding to the challenge, anecdotal evidence from NWRs in the Southeast suggests that one-time applications of imazapyr have not always controlled alligatorweed across multiple years and may have short-term negative effects on desirable plant species. Moreover, as a broad-spectrum herbicide with residual soil activity, imazapyr can affect non-target plants at the time of application and has the potential to affect subsequent plant growth later in the growing season or possibly the next (Dugdale et al. 2020). NWR staff previously has observed that an integrated approach — timing water drawdown and application of herbicide with dry conditions during mid-summer - results in maximum alligatorweed control (Allen et al. 2007; U.S. Fish and Wildlife Service 2007, 2010). However, effects of imazapyr on desirable moist-soil vegetation and related food production for waterfowl, especially in the year following treatment, could negate benefits of alligatorweed control. Thus, an approach that investigates shortand long-term effects of alligatorweed control on vegetation composition, food production, and use by waterfowl in moist-soil wetlands would substantially improve managers' ability to incorporate imazapyr into an integrated pest management approach for this invasive species.

Herein, we investigated the effects of imazapyr herbicide on moist-soil vegetation, food density, and wetland use and activities of dabbling ducks (Anatini) within treated and non-treated plots in Tennessee. We hypothesized that imazapyr application would 1) reduce cover of alligatorweed in the year of (year 1) and one year following application (year 2), 2) decrease food density and cover of desirable vegetation in year 1, 3) increase cover of desirable plant species and decrease cover of undesirable plant species in year 2, and 4) result in decreased use by and foraging activity of dabbling ducks in year 1 relating to potential decreases in emergent vegetation and food abundance. Our results are needed to inform management of moist-soil wetlands to ensure contributions to conservation goals supporting the North American Waterfowl Management Plan (NAWMP) (NAWMP 2012, Hagy et al. 2020).

Study Area

We studied moist-soil wetlands within the Duck River Unit of Tennessee NWR (35°57'30N, 87°57'00W) in the Tennessee River Valley of west-central Tennessee. The Duck River Unit of Tennessee NWR (10,820 ha) is located at the confluence of the Duck and Tennessee rivers in Benton and Humphreys counties, Tennessee. Public access is restricted on Tennessee NWR from 15 November to 15 March to provide sanctuary conditions for non-breeding waterfowl. The NWR is surrounded by private and public hunting areas, making it an important waterfowl sanctuary in this region (U.S. Fish and Wildlife Service 2010, 2014). The Duck River Unit consists of seasonal wetlands, forested wetlands, and upland agricultural fields all within a leveed bottomland subdivided by seasonally flooded impoundments for waterfowl. Wintering waterfowl are a priority resource of concern for the Tennessee NWR, which supports up to 150,000 ducks and geese annually. Habitat resource management focuses on moist-soil wetlands, agricultural crops, forested and scrub-shrub wetlands, and riverine resources (i.e., mudflats, open water, and submerged aquatic vegetation). Moistsoil vegetation and agricultural crops were primarily provided in managed impoundments where herbicide applications and water level manipulation were the primary practices to encourage desirable waterfowl forage. Alligatorweed is the foremost invasive species within managed impoundments at Tennessee NWR and thus its control was an emphasized research and adaptive management objective (U.S. Fish and Wildlife Service 2010, 2014).

Methods

Experimental Design

In June–July 2011, we delineated four, non-adjacent, 1-ha blocks of moist-soil vegetation, each of which was divided into two 0.5-ha plots that received either a single application of imazapyr herbicide (hereinafter, herbicide plot) or no manipulation (hereinafter, control). Herbicide plots were broadcast treated with Habitat (BASF, Research Triangle Park, North Carolina) and Sun Energy surfactants at a rate of 4.677 L/ha and 1.169 L/ha, respectively, via tractor-mounted sprayer. We established blocks specifically in areas where alligatorweed was present and likely to become present

during the growing season based on occurrence in past years. We selected areas with visually homogenous vegetation coverage and species composition. We separated blocks by ≥ 200 m to reduce the likelihood of double-counting waterfowl, ensure spatial independence of vegetation and food response, and guard against chemical drift. We delineated plot boundaries with white polyvinyl chloride markers prior to treatment and randomly assigned plots within each block.

We assessed vegetation composition in treatment and control plots in June 2011 (pre-treatment period; treatment plot only), September 2011 (post-treatment period 1), June 2012 (post-treatment period 2), and September 2012 (post-treatment period 3). We sampled vegetation at 10 subplots systematically along a transect spanning each plot using a 1-m² frame quadrat. The direction and starting point of each transect was randomized and vegetation sampling locations were then placed systematically to cover the entire plot (Hagy and Kaminski 2012b). Within each subplot, we estimated percent horizontal cover of alligatorweed, plant genera known to produce food for waterfowl (hereinafter, desirable; Osborn 2015), plant genera that generally did not produce food for waterfowl (hereinafter, undesirable), and ground litter cover. We also visually assessed mean height of vegetation present (cm). We averaged cover and height estimates among subplots for each transect to obtain estimates for each plot.

We enumerated and estimated proportional activities of dabbling ducks by species once weekly in all plots during late December through late February each winter (2011–2012, 2012–2013). We initiated observations when each block was \geq 60% flooded. We surveyed ducks from camouflaged tree stands between sunrise and 5 h thereafter (~0700–1200 hours) using pre-determined routes that were rotated weekly among observers (Osborn et al. 2017). We assumed diurnal use adequately represented habitat resource use and selection within waterfowl sanctuaries and was useful to guide wetland management activities (Hagy and Kaminski 2012*b*, Osborn et al. 2017). When blocks were >60% inundated, we measured average water depth among 20–30 locations along one or more randomly placed transects within each plot and erected a fixed gauge to indicate average depth during surveys without disturbing waterfowl (Hagy and Kaminski 2012*b*, Osborn et al. 2017).

We conducted a single scan of each plot using binoculars and a digital voice recorder to simultaneously enumerate, identify, and record distances and instantaneous activities of dabbling ducks within plot boundaries (Altmann 1974, Hagy and Kaminski 2012*b*, Osborn et al. 2017). We visually estimated distances of dabbling ducks to the nearest 10 m to aid in density estimation (Buckland et al. 2001). Markers were placed at 100 and 200 m from the blind using laser range finders to assist observers in visually estimating distances. If waterfowl flushed or were disturbed from a plot immediately prior to or during a survey, we censored the survey and returned another morning. If a minor disturbance occurred and birds did not abandon or redistribute within the plot, we waited at least 5 min for birds to resume normal behavior before conducting a survey. We classified dabbling duck activities as foraging (surface feeding, tipping up), resting (sleeping, loafing, inactivity), locomotion (walking or swimming), aggression (chasing, biting, fighting), courtship (displaying, copulation), alert (inactive with head erect), and maintenance (preening, bathing, stretching) (Morton et al. 1989, Osborn et al. 2017). We did not include birds in flight during surveys (Buckland et al. 2001) and we did not survey waterfowl in dense fog or if winds exceeded 30 kph (Hagy and Kaminski 2012*b*). We recorded average water depth and an ocular estimate of horizontal emergent vegetation cover during each survey.

We estimated density of plant (i.e., seeds and tubers) and animal (i.e., aquatic macroinvertebrates) foods of waterfowl in early winter (November-December) of 2011 and 2012 using a standard core sampler (10 cm diameter and depth; Osborn et al. 2017). We collected five core samples systematically along a randomly positioned transect spanning each plot lengthwise, washed them in the field using a 500-µm sieve bucket (Wildco, Buffalo, New York), and placed them in polyethylene bags for storage and transport. We preserved all samples in 70% ethyl alcohol and stored them at -10 °C (Salonen and Sarvala 1985). In the lab, we thawed food samples, stained them with 1% rose bengal solution, washed them through a series of graduated sieves [#4 (4.75 mm), #14 (1.40 mm), #50 (300 µm)], and removed with forceps all aquatic macroinvertebrates commonly consumed by dabbling ducks (Hagy and Kaminski 2012a). We separated macroinvertebrates by order, dried them for 24 h at 60 °C, and weighed them to the nearest 0.1 mg (Murkin et al. 1994). Following removal of macroinvertebrates, we air-dried the remaining material for 24-48 h and extracted from #4 and #14 sieves all seeds and tubers known to be commonly consumed by dabbling ducks (Checkett et al. 2002, Hagy and Kaminski 2012a). We separated seeds and tubers by genus or species, dried them for 24 h at 60 °C, and weighed them to the nearest 0.1 mg (Hagy and Kaminski 2012a, Osborn et al. 2017).

To account for seeds that potentially passed through the 300-µm sieve, we subsampled sieve contents of five cores samples from one herbicide plot and the adjacent control plot within a single block from the first month of sampling (November–December) to create correction factors (Osborn et al. 2017). We sorted all seeds from a 25% mass portion randomly selected from the combined set of five subsamples and dried and weighed them using the same protocols as for the #4 and #14 sieves (Hagy et al. 2011, Osborn et al. 2017). We multiplied seed biomasses from subsamples x4 and

compared taxon-specific estimates to the corresponding mass of the #4 and #14 sieve contents, by which we established a constant correction factor among plots by treatment type and year. We applied taxon-specific correction factors to seed and tuber biomass estimates in all sieves to account for recovery bias (Hagy et al. 2011) and we expressed biomass as density (kg[dry]/ha).

Statistical Analyses

We used paired *t*-tests to compare vegetation metrics (i.e., alligatorweed cover, desirable plant cover, undesirable plant cover, litter cover, and mean plant height) among blocks and between treatments for each vegetation assessment period (post-treatment periods 1-3 in 2011-2012). Additionally, we used paired *t*-tests to compare vegetation metrics before (pre-treatment period) and after (post-treatment period 1) herbicide application in treatment plots in 2011. Due to an abundance of zeros in subplot quadrats, we also used logistic regression to interpret the likelihood of alligatorweed presence in plots and used the Hosmer-Lemeshow test to assess model fit (Hosmer and Lemeshow 1989). We used one-way analysis of variance to test for treatment effects separately on seed and tuber density and invertebrate density during early winter of the year of treatment and early winter of the year after treatment. We designated dry biomass (kg/ha) as the response variable and treatment as a fixed effect.

We used multiple covariates distance sampling (Buckland et al. 2001) in Distance 6.0 release 2 (Thomas et al. 2010) to estimate dabbling duck densities in herbicide and control plots and account for visibility bias (Smith et al. 1995, Osborn et al. 2017). Because of low abundances of species other than mallards (Anas platyrhynchos), we pooled weekly abundances among species and analyzed all dabbling ducks collectively (i.e., treated them as a dabbling duck guild). Because of small sample sizes for most individual waterfowl species within individual plots, we estimated global detection functions and average detection probabilities pooled across plots and moist-soil sites from a concurrent study among years (Osborn et al. 2017). We used percent emergent vegetation cover as a categorical covariate with four levels (0-25%, 30%-50%, 55%-75%, 80%-100%) to account for potential visibility bias. We applied detection functions and detection probabilities from global models to estimate weekly densities of dabbling ducks (Thomas et al. 2009). We adjusted weekly density estimates using monthly flooded areas (ha) obtained from light detection and ranging imagery, water gauge data, and aerial imagery in ArcGIS 10.1 (Environmental Systems Research Institute Inc., Redlands, California) (Thomas et al. 2009).

Blocks flooded asynchronously among locations and years, so we designated the first week post-flooding for each block as the

first sampling period regardless of Julian date to standardize dabbling duck observations among blocks and reduce potential bias. We used a general linear mixed model to test for treatment effects on dabbling duck densities. We designated dabbling duck density as a response variable, treatment as a fixed effect, block nested within year as a random effect, and week as a repeated measure. We used Spearman rank correlation (ρ) to measure the association of weekly dabbling duck densities with weekly water depth estimates (McKinney et al. 2006, Osborn et al. 2017). To analyze instantaneous activities of dabbling ducks, we summed counts across all weekly surveys by treatment type and activity, and we performed separate chi-square tests of homogeneity to test for a difference in the percent occurrence of activities among control and treatment plots (Zar 1999). We excluded courtship, aggression, alertness, and maintenance from total counts and final analyses because of low occurrence (<10%).

Prior to all analyses, we examined histograms, box plots of group means and variances, residual distributions, and variances of response variables to assess variable distributions (Quinn and Keough 2002, Littell et al. 2006). We transformed dabbling duck density using natural logarithm to equalize variances (Quinn and Keough 2002, Zuur et al. 2009). When using repeated measures, we estimated degrees of freedom via Kenward-Rogers and compared Akaike Information Criterion scores to select an appropriate covariance structure (Littell et al. 2006). We designated $\alpha = 0.05$ and performed Tukey's pairwise multiple comparison test of means when a main effect was significant. We calculated means and standard errors from untransformed or back-transformed data as appropriate. All analyses were performed in SAS 9.4 (SAS Institute Inc, Cary, North Carolina).

Results

Vegetation Response

Alligatorweed cover, desirable plant cover, undesirable plant cover, and litter cover were all less ($t_3 > 2.6$, P < 0.04) in herbicide plots than control plots in September 2011 (post-treatment period 1), approximately two months post herbicide treatment (Table 1). Mean vegetation height did not differ among control and treatment plots ($t_3=2.2$, P=0.06) in post-treatment period 1. Alligatorweed cover, desirable plant cover, and litter cover all declined between the pre-treatment period and the post-season period 3 (September 2012) in herbicide plots ($t_3>2.65$, P<0.04). Conversely, undesirable plant cover and mean vegetation height were greater in herbicide plots in post-treatment period 3 than in the pre-treatment period ($t_3<-4.1$, P<0.01).

In both sampling periods in 2012, alligatorweed cover, desirable plant cover, undesirable plant cover, litter cover, and mean

		Plot			Alligatorweed (%)		Desirable (%)		Undesirable (%)		Height (cm)		Litter (%)	
Туре	#	Year	Period ^a	<i>x</i>	SE	<i>X</i>	SE	<i>X</i>	SE	ñ	SE	<i>x</i>	SE	
Control	1	2011	Post 1	1.0	0.7	16.0	5.0	56.5	8.6	18.6	1.2	23.5	4.8	
		2012	Post 2	0.0	0.0	19.0	5.6	44.5	5.3	12.2	0.8	21.0	1.6	
			Post 3	4.0	1.8	35.5	6.9	52.0	6.4	16.9	0.9	2.5	2.5	
	2	2011	Post 1	6.0	1.9	44.5	5.3	25.0	6.2	21.1	1.8	21.5	3.7	
		2012	Post 2	0.0	0.0	50.5	5.8	26.5	4.0	8.2	1.2	3.0	0.8	
			Post 3	1.5	1.1	45.5	5.9	50.0	6.1	18.7	0.8	0.0	0.0	
	3	2011	Post 1	2.0	1.3	63.5	6.3	18.5	6.9	15.6	0.9	12.0	2.0	
		2012	Post 2	5.5	2.4	51.5	4.9	35.5	4.6	12.4	0.9	5.0	3.1	
			Post 3	3.0	1.3	33.5	2.6	38.5	3.1	26.0	2.3	0.0	0.0	
	4	2011	Post 1	4.5	2.4	55.0	6.2	33.0	6.4	25.2	1.8	7.5	3.7	
		2012	Post 2	1.5	1.1	24.5	5.5	54.0	6.3	13.5	1.4	9.5	2.5	
			Post 3	5.5	2.0	27.0	6.7	53.0	4.7	31.7	2.6	9.0	2.2	
	Mean	2011	Post 1	3.4	1.1	44.8	10.3	33.3	8.3	20.1	2.0	16.1	3.8	
		2012	Post 2	1.8	1.3	36.4	8.5	40.1	5.9	11.6	1.2	9.6	4.0	
			Post 3	3.5	0.8	35.4	3.8	48.4	3.4	23.3	3.4	2.9	2.1	
Treatment	1	2011	Pre	1.0	0.7	56.8	7.7	20.2	6.6	4.4	0.7	3.0	0.8	
			Post 1	0.5	0.5	1.0	0.7	9.0	3.6	10.4	1.0	83.5	3.8	
		2012	Post 2	0.0	0.0	22.5	3.0	62.0	6.8	16.7	2.0	6.0	2.6	
			Post 3	0.0	0.0	31.5	7.7	38.5	8.3	21.7	1.6	1.5	1.1	
	2	2011	Pre	10.0	4.8	36.5	9.6	23.5	6.0	5.7	1.2	4.5	3.0	
			Post 1	0.5	0.5	3.5	2.0	6.0	2.6	17.7	1.7	83.0	5.0	
		2012	Post 2	0.0	0.0	24.5	3.1	50.0	1.7	10.4	1.9	3.5	0.8	
			Post 3	0.0	0.0	35.0	5.2	48.5	4.1	26.9	2.4	0.0	0.0	
	3	2011	Pre	5.5	3.9	66.5	10.2	1.0	0.7	4.8	0.7	1.0	1.0	
			Post 1	0.0	0.0	0.0	0.0	2.2	0.9	16.3	1.5	78.3	5.5	
		2012	Post 2	0.0	0.0	23.0	4.4	39.5	3.0	10.8	0.6	8.5	2.8	
			Post 3	0.0	0.0	53.5	5.7	41.0	4.5	22.0	2.0	0.0	0.0	
	4	2011	Pre	9.0	3.1	59.0	7.4	17.5	5.3	7.2	1.0	0.0	0.0	
			Post 1	1.5	1.1	2.0	1.5	9.5	4.5	16.6	0.7	72.5	6.2	
		2012	Post 2	1.5	1.1	24.0	6.4	43.0	7.3	6.9	1.3	9.0	4.1	
			Post 3	6.0	3.1	44.5	5.2	31.0	5.0	22.0	3.2	0.0	0.0	
	Mean	2011	Pre	6.4	2.0	54.7	6.4	15.6	5.0	5.5	0.6	2.1	1.0	
			Post 1	0.6	0.3	1.6	0.7	6.7	1.7	15.3	1.6	79.3	2.6	
		2012	Post 2	0.4	0.4	23.5	0.5	48.6	5.0	11.2	2.0	6.8	1.3	
			Post 3	1.5	1.5	41.1	5.0	39.8	3.6	23.1	1.2	0.4	0.4	

Table 1. Mean (SE) proportional alligatorweed cover, desirable plant cover, undesirable plant cover, litter cover, and maximum plant height in 0.5-ha plots (#) treated with imazapyr (treatment) and adjacent un-manipulated plots (control) during 2011 and 2012 at the Duck River Unit of Tennessee National Wildlife Refuge, Tennessee.

a. Pre: July, Post 1: September 2011, Post 2: June 2012, Post 3: September 2012

vegetation height were all similar among treatment plots ($t_3 < 2.0$, P > 0.07). Alligatorweed cover, desirable plant cover, litter cover, and mean vegetation height were all less ($t_3 > 3.0$, P < 0.03) in herbicide plots after treatment (post-treatment period 1) than before treatment (pre-treatment period). Undesirable plant cover did not differ in treatment plots ($t_3 = 2.3$, P = 0.05) before and after treatment (post-treatment period 1).

Prior to treatment, alligatorweed comprised 6.4% (SE = 1.8%,

n=4) of vegetation cover and occurred in 26.3% of vegetation surveys across treatment blocks. Two months post treatment, alligatorweed cover decreased 90.6% in treatment plots ($\bar{x}=0.6$, SE=0.3%, n=4) and 75.7% in control plots ($\bar{x}=3.4\%$ SE=0.9%, n=4; Table 1). One year after treatment, alligatorweed comprised 1.5% of treatment plots (SE=0.9%, n=4) and 3.5% of control plots (SE=0.8%, n=4). We included year and treatment in the final logistic regression model and found no significant evidence for lack of fit (Wald X^2 = 19.6, P < 0.001). Alligatorweed was 2.3 times more likely to occur in fall 2011 than fall 2012 surveys (Wald X^2 = 9.2, P = 0.003) and 2.5 times more likely to occur in control than treatment plots (Wald X^2 = 7.6, P = 0.006). When analyzed separately, alligatorweed was 4.2 times more likely to occur in control than treatment plots in 2011 (Wald X^2 = 5.3, P = 0.022) and 5.3 times more likely in control than treatment plots in 2012 (Wald X^2 = 10.1, P = 0.002).

Waterfowl Use and Activities

Dabbling duck density averaged 75.3 ducks/ha (SE=13.6) across treatments and years. Dabbling duck density did not differ between treatment and control plots ($F_{1,21.2}=0.71$, P=0.410) although we found it was negatively related to increasing water depth regardless of treatment type (herbicide: $\rho = -0.358$, P=0.09; control: $\rho = -0.349$, P=0.009: a 2.5-cm decrease in water depth resulted in an estimated increase of 167 dabbling ducks/ha). Percent occurrence of foraging, locomotion, and resting activities of dabbling ducks did not differ between herbicide and control plots during the year of treatment ($X^2 = 3.559$, P=0.169) or one year after treatment ($X^2 = 1.317$, P=0.599). Dabbling ducks spent most of their time foraging (48.7%), followed by resting (21.8%) and locomotion (21.4%).

Seed, Tuber, and Invertebrate Density

Seed and tuber biomass did not differ between herbicide and control plots at the time of initial flooding in the year of treatment ($F_{1,7}$ =0.92, P=0.375; Table 2) or one year after treatment ($F_{1,7}$ =1.08, P=0.340). Furthermore, we detected no difference in seed and tuber biomass between years in herbicide ($F_{1,7}$ =0.05, P=0.505) and control plots ($F_{1,7}$ =0.27, P=0.625). Seed and tuber density was 850.5 kg/ha (SE=273.7) in the year of treatment and 1238.6 kg/ha (SE=349.7) one year after treatment across control and herbicide plots.

Invertebrate biomass differed between herbicide and control

 Table 2. Mean (SE) biomass (kg[dry]/ha) of foods apparently consumed by waterfowl in 0.5-ha plots

 (n) treated with imazapyr and adjacent un-manipulated plots (control) during early winter 2011 and

 2012 at the Duck River Unit of Tennessee National Wildlife Refuge, Tennessee.

		Seeds	ubers	Aquatic invertebrates			
Year	Plot type	Ĩ	n	SE	Ĩ	n	SE
2011	Mean	850.5	8	273.7	2.3	8	0.8
	Control	586.6	4	254.4	4.0	4	1.0
	Herbicide	1114.4	4	488.3	0.6	4	0.4
2012	Mean	1238.6	8	349.7	6.0	8	1.9
	Control	877.8	4	504.4	5.4	4	2.6
	Herbicide	1599.5	4	478.9	6.6	4	3.2

plots at the time of initial flooding in the year of treatment ($F_{1,7} = 13.85$, P = 0.006) but not one year after treatment ($F_{1,7} = 4.26$, P = 0.07). Furthermore, we detected no difference in invertebrate biomass between years in herbicide ($F_{1,7} = 3.36$, P = 0.116) and control plots ($F_{1,7} = 0.26$, P = 0.630). Invertebrate biomass was 4.0 kg/ ha (SE = 1.0) in control and 0.6 kg/ha (SE = 0.4) in herbicide plots in the year of treatment. Invertebrate biomass across herbicide and control plots was 6.0 kg/ha (SE = 1.9) one year after treatment.

Discussion

Application of imazapyr herbicide substantially reduced coverage of alligatorweed, desirable plant species, and other undesirable plant species two months after application. Moreover, imazapyr treatments suppressed alligatorweed growth throughout the subsequent growing season compared to control plots (Allen et al. 2007). However, imazapyr treatment did not reduce seed and tuber density for waterfowl during the year of application nor did it, contrary to our expectations, result in reduced desirable plant species coverage relative to control plots in the year post treatment. Furthermore, seed and tuber densities were similar among treatment and control plots one year after application. In fact, mean seed and tuber densities in treatment plots one year after herbicide application were approximately double the mean density on managed lands in the Mississippi Alluvial Valley (Kross et al. 2008) and greater than several other large-scale studies in similar geographies (Stafford et al. 2011, Hagy and Kaminski 2012b). Herbicide application may have indirectly affected aquatic invertebrate densities in the year of application by increasing litter and reducing emergent vegetation cover after flooding. Regardless, overall invertebrate densities in both herbicide and control plots were low relative to other wetland types indicating limited significance for contributing to daily energy requirements of waterfowl (Osborn et al. 2017).

Imazapyr applications reduced the cover and frequency of occurrence of alligatorweed in the year of and after treatment, which is consistent with previous research (Allen et al. 2007). However, alligatorweed persisted at low coverages in both control and treatment plots 1 year after treatment, indicating the likely need for follow-up herbicide treatments or additional pest control strategies. Imazapyr requires active plant growth to translocate through plant tissues and inhibit amino acid synthesis, so short- and longterm control may vary with application timing, according to moisture conditions, or with other stressors (Parker and Boydston 2005). Indeed, in our study, alligatorweed coverage declined in both treated (90.6%) and control plots (75.7%) in the year of treatment, which we attribute to dry conditions during summer 2011 (Osborn 2015, Wei et al. 2014) and the natural growth period for alligatorweed (Shen et al. 2005). Thus, we speculate that opportunistically applying imazapyr during dry or drought periods could substantially reduce alligatorweed coverage and improve overall control (U.S. Fish and Wildlife Service 2007, 2014).

We recommend an integrated pest management approach where timing and amount of herbicides coincides with expected temperature and moisture regimes (i.e., rainfall, drawdown timing) to achieve optimal alligatorweed control. Although we were unable to test biological control agents (e.g., *Agasicles hygrophila*) in this study, we suspect that integrating biological control agents where feasible, such as in southern portions of the Southeast where overwintering is possible, could increase long-term control following or prior to initial herbicide treatment (Vogt et al. 1992). We recommend additional alligatorweed control studies along a latitudinal gradient that incorporates biological control agents into a multiyear integrated pest management strategy to better evaluate viability of these agents at more northerly portions of the Southeast such as Tennessee (Vogt et al. 1992; Harms and Shearer 2019, 2020).

Interestingly, mid-summer applications of imazapyr did not reduce seed and tuber biomass in the year of treatment during our study. Imazapyr can take several weeks to induce plant necrosis with relative slow absorption and translocation rates (Tucker et al. 1994), so annual plants may have had time to produce seeds in treatment plots similar to control plots. Additionally, early drawdowns (i.e., April-May) and delayed imazapyr application until mid-summer may have allowed some desirable plant species that are abundant seed producers (e.g., Echinochloa crus-galli) to mature before treatment. We observed increased tuber production of yellow nutsedge (Cyperus esculentus) in treated plots, which had densities that we estimated were nearly twice that in control plots (Osborn 2015). Imazapyr is a non-selective herbicide; foliar applications late in the growing season may kill only aboveground biomass of yellow nutsedge possibly leaving mature tubers unaffected and dormant until the next growing season (Wilen et al. 1999). The increased amount of yellow nutsedge could also be due to reduced competition from other plants, however, and not to stimulated tuber production, per se (Kelly 1990, Wilen et al. 1999). Given that NWR staff have anecdotally observed similar responses of yellow nutsedge following late-summer glyphosate applications, further research is needed on applications of broad-spectrum herbicides late in the growing season in moist-soil wetlands to better understand the effects on tuber production.

Although dabbling ducks commonly used moist-soil wetlands at Tennessee NWR (Osborn et al. 2017), contrary to our initial hypothesis we observed no difference in dabbling duck use between treated and control plots during our study. This suggests that vegetation and structural differences related to herbicide treatments did not influence waterfowl use. Dabbling ducks were more abundant in other western Tennessee moist-soil wetlands than either our treatment or control plots (Osborn et al. 2017), and these areas of greater use generally had less robust or undesirable emergent vegetation than our plots according to our anecdotal observations. We suspect that waterfowl may primarily select moist-soil wetlands for other reasons, such as sanctuary conditions, and that small differences in food density do not result in detectable and predictable functional or numerical responses of ducks as has been demonstrated previously (Hagy and Kaminski 2015, Hagy et al. 2017).

Dabbling duck abundance in our study was inversely related to water depth, which is consistent with findings from other studies (Colwell and Taft 2000, Hagy and Kaminski 2012b, Osborn et al. 2017). Flooding creates an interspersion of vegetation and open water desirable to waterfowl (Kaminski and Prince 1981, Smith et al. 2004, Moon and Haukos 2008), but birds may either abandon foraging areas when water depth prevents efficient foraging or use areas differently at night (Moon and Haukos 2008, Hagy and Kaminski 2012b, Monroe et al. 2021). Functional use and dabbling duck density did not differ between treated and control plots, which we speculate resulted from similar vegetation cover and food density among plots and possibly overall lower habitat quality among study plots relative to other nearby locations (Osborn et al. 2017). Our study suggests that treating invasive species with imazapyr herbicide during the growing season has minimal to no effects on waterfowl use and food availability during subsequent fall and winter periods on NWRs in Tennessee.

Management Implications

Mid- to late-summer imazapyr application may help control alligatorweed through the next growing season without negatively affecting seed and tuber density, coverage of desirable moistsoil vegetation in the year following application, or waterfowl use. Imazapyr applications can be effectively used as a component of an integrated pest management program to maintain alligatorweed control where it is established while providing waterfowl habitat resources to achieve habitat objectives on NWRs in support of the NAWMP. However, long-term alligatorweed control in wetlands managed for waterfowl will typically require repeated control techniques that are integrated in a formal approach, such as timing herbicide applications to coincide with dry periods during the summer, keeping impoundments dry for multiple years in sequence, and using rotational practices (e.g., agriculture) at short intervals with moist-soil management to maintain desirable vegetation communities.

Acknowledgments

We thank the staff at Tennessee NWR for providing access to lands, spatial and other data, and field housing for this study. We also thank the Black Duck Joint Venture, University of Tennessee Institute of Agriculture, Tennessee State Parks, Mississippi State University, Ducks Unlimited, U.S. Fish and Wildlife Service (USFWS) NWR System and Division of Migratory Birds, and the Illinois Natural History Survey for providing funding and in-kind support. We thank all the field and laboratory technicians for their assistance during this project. The findings and conclusions in this article are those of the authors and do not necessarily represent the views of the USFWS or other agencies and organizations. The use of trade, product, or firm names in this publication are for descriptive purposes only and does not imply endorsement by the U. S. government.

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