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Effect of indaziflam on native species in natural areas and rangeland

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Abstract

Minimizing the negative ecological impacts of exotic plant invasions is one goal of land management. Using selective herbicides is one strategy to achieve this goal; however, the unintended consequences of this strategy are not always fully understood. The recently introduced herbicide indaziflam has a mode of action not previously used in non-crop weed management. Thus, there is limited information about the impacts of this active ingredient when applied alone or in combination with other non-crop herbicides. The objective of this research was to evaluate native species tolerance to indaziflam and imazapic applied alone and with other broadleaf herbicides. Replicated field plots were established at two locations in Colorado with a diverse mix of native forbs and grasses. Species richness and abundance were compared between the nontreated control plots and plots where indaziflam and imazapic were applied alone and in combination with picloram and aminocyclopyrachlor. Species richness and abundance did not decrease when indaziflam or imazapic were applied alone; however, species abundance was reduced by treatments containing picloram and aminocyclopyrachlor. Species richness was only impacted at one site 1 yr after treatment (YAT) by these broadleaf herbicides. Decreases in abundance were mainly due to reductions in forbs that resulted in a corresponding increase in grass cover. Our data suggest that indaziflam will control downy brome (*Bromus tectorum* L.) for multiple years without reduction in perennial species richness or abundance. If *B. tectorum* is present with perennial broadleaf weeds requiring the addition of herbicides like picloram or aminocyclopyrachlor, forb abundance could be reduced, and in some cases there could be a temporary reduction in perennial species richness.

Introduction

Downy brome (*Bromus tectorum* L.), an exotic winter annual grass, has emerged as one of the most invasive and problematic weeds in western rangeland and natural areas, with an estimated 14% annual spread rate (DiTomaso et al. 2010; Duncan et al. 2004). Although *B. tectorum* typically germinates in the fall after cool, wet weather, plants are opportunistic and can germinate anytime the growing conditions are favorable (Beck 2009). This variable germination cycle has allowed *B. tectorum* to thrive in arid and semiarid western climates by rapidly growing and depleting available soil moisture and nutrients before most native species break dormancy in the spring (Knapp 1996; Mack and Pyke 1983). Invasions in natural ecosystems can cause severe negative impacts by reducing native plant diversity and lowering community productivity, increasing fire frequency, and displacing native vegetation that is critical wildlife and pollinator habitat (Abatzoglou and Kolden 2011; Beck 2009; Billings 1994; DiTomaso et al. 2010; Knapp 1996; Monaco et al. 2017; Whisenant 1990).

By the 1930s researchers had begun to recognize *B. tectorum* invasions as a serious issue in rangeland (Mack 1981; Price et al. 1948; Warg 1938). Since then, extensive research has been conducted on mechanical, cultural, biological, and chemical control of this exotic grass. Thus far, herbicides have been the most effective and widely used weed management strategy for *B. tectorum* on rangeland and natural areas (Diamond et al. 2012; Mangold et al. 2013; Monaco et al. 2017). Since its release in 1996, imazapic has been the primary herbicide used to control *B. tectorum* on rangeland, because it has both PRE and POST activity and is selective at relatively low use rates (Anonymous 1996; Kyser et al. 2013; Mangold et al. 2013). Several other herbicides, including glyphosate, sulfometuron, and rimsulfuron, have traditionally been used for *B. tectorum* control in non-crop sites (Kyser et al. 2013; Sebastian et al. 2016). Although adequate control is often achieved with these herbicide options the first year of application, control can be inconsistent or short term, and injury to desirable species can occur

Management Implications

Rangeland weeds cause severe ecological impacts, including depleting soil moisture and nutrients, reducing plant diversity and community productivity, altering fire frequency, and reducing recreational land values. Several herbicides approved for use in natural areas and rangeland can negatively affect native species, while the duration of weed control can be highly variable. Long-term weed control is critical in allowing sufficient time for native species recovery; therefore, herbicide options are needed that provide multiyear control without impacting the native plant community. Indaziflam, a newer herbicide option for preemergent invasive winter annual grass management, can provide control for 3 or more years, although there has been limited research on its effects on native species. A field study was conducted to evaluate changes in the native plant community composition from two annual grass herbicides, imazapic and indaziflam, as well as changes from two broadleaf herbicides, picloram and aminocyclopyrachlor, in diverse native perennial grass and forb communities. The study evaluated species richness and species abundance in the plant community for 2 yr. Picloram decreased native species abundance throughout the duration of the study across both sites, while aminocyclopyrachlor decreased species abundance at one site. Imazapic and indaziflam did not decrease species abundance or richness at either site over 2 yr. The results presented here suggest that indaziflam is an option for land managers to control winter annual grasses without negatively impacting existing native perennial species. In sites with a remnant native plant community, the multiyear winter annual grass control provided by indaziflam may allow enough time to achieve native species recovery.

(Kelley et al. 2013; Kyser et al. 2013; Mangold et al. 2013; Morris et al. 2009; Thacker 2009; Whitson and Koch 1998; Whitson et al. 1997). In many invasive situations, short-term control (<2 yr) does not allow the remnant native plant community enough time to reestablish/recover and become competitive (Chambers et al. 2014; Elseroad and Rudd 2011). As *B. tectorum* infestations continue to spread, shifting native perennial grass systems to ecosystems dominated by winter annual grasses, land managers need strategies that provide long-term control of this weed.

Indaziflam is a broad-spectrum, PRE herbicide first released in 2011 for use in several perennial cropping systems and later used for weed control in turfgrass, ornamentals, forestry, and non-crop industrial sites (Anonymous 2011a, 2011b; Brabham et al. 2014). In 2016 a supplemental label for indaziflam was approved for the release or restoration of desirable vegetation in natural areas, open spaces, wildlife management areas, and fire rehabilitation areas, specifically targeting invasive winter annual grass control in these sites (Anonymous 2016).

Indaziflam is a cellulose biosynthesis inhibitor, representing a unique site of action with no reported cases of resistance in the field (Brabham et al. 2014; Tateno et al. 2016). Indaziflam has a longer soil residual than other herbicides commonly used for *B. tectorum* management, providing 3 or more years of control (Sebastian et al. 2016, 2017a). In most rangeland situations, this length of control allows enough time for release of the remnant native perennial community (Chambers et al. 2014; Sebastian et al. 2016, 2017a). Sebastian et al. (2016, 2017a) found that indaziflam will selectively control *B. tectorum* without impacting perennial grass and forb biomass, even leading to significant increases in biomass due to reductions in *B. tectorum* (Sebastian et al. 2016, 2017a). This research suggests native perennial species are tolerant to indaziflam, although studies assessing impacts to community composition and native species abundance following indaziflam applications have not been conducted.

The objective of this study was to evaluate tolerance of several native species to indaziflam applications and compare tolerance with other commonly used grass and broadleaf rangeland herbicides. We hypothesized that indaziflam would significantly reduce *B. tectorum* cover without decreasing native species abundance compared with the other herbicides evaluated.

Materials and Methods

Site Description

The experiments were established in 2015 at two sites in Jefferson County, CO, containing *B. tectorum* with a co-occurring native grass, forb, and shrub community. Site 1 (39.760°N, 105.239°W) was located on Mount Galbraith Open Space, and Site 2 (39.894°N, 105.271°W) was located on El Dorado Mountain Open Space. Sites were approximately 15 km apart in the Western High Plains region of the Great Plains ecoregion. In June 2015, before herbicide application, we conducted an initial inventory of the plant species by recording all plant species present at each site within the boundaries of the plots. A visual estimate of *B. tectorum* canopy cover (%) was also done at both sites. Site 1 was categorized as ~30% to 40% *B. tectorum* cover with 33 native species and 5 co-occurring nonnative species (Table 1). By the following year (2016), *B. tectorum* cover at Site 1 decreased to ~5% and maintained a similar cover level throughout the course of the study. Site 2 had ~60% to 70% *B. tectorum* cover with 35 native species and 6 co-occurring nonnative species (Table 1).

The soil at Site 1 was Ratake rocky loam (loamy-skeletal, micaceous, frigid, shallow Typic Haplustolls), with 2.3% organic matter and 6.0 pH in the top 20 cm (USDA-NRCS 2014). Site 1 was located on a 30° rocky slope, and the average elevation was 1,839 m (6,035 ft). The soil at Site 2 was Flatirons stony sandy loam (clayey-skeletal, smectitic, mesic Aridic Paleustolls), with 4.9% organic matter and 6.6 pH in the top 20 cm (USDA-NRCS 2014). Site 2 was located on a 25° rocky slope, and the average elevation was 1,995 m (6,544 ft). Mean annual precipitation based on the 30-yr average (1981 to 2010) was 468 mm at Site 1 and Site 2 based on the closest weather station (approximately 8 km from each site) (Western Regional Climate Center 2018).

Both sites received an additional 252 mm of precipitation above their 30-yr average in 2015. A statewide drought occurred in 2016, and total precipitation for the sites decreased to 148 mm below the 30-yr average (Western Regional Climate Center 2018). In 2017 the sites received precipitation similar to the 30-yr average. The 30-yr mean annual temperature for both sites was 10.2 C, and the average temperature for 2015 was close to the 30-yr average. The average temperatures for 2016 and 2017 were 1.8 C and 2.2 C warmer, respectively (Western Regional Climate Center 2018).

Experimental Design

Herbicide applications were made June 2, 2015 and timed in accordance with label recommendations for Dalmatian toadflax [*Linaria dalmatica* (L.) Mill.] control, one of the co-occurring nonnative species (Anonymous 2018). The herbicides targeting *B. tectorum* were applied at an early PRE application timing. *Bromus tectorum* was in the ripening stage and actively setting seed. Native forb growth stage ranged from postflowering, early flowering to preflowering. Ten

Table 1. List of species occurring at Site 1 and Site 2 with their nativity status.

Scientific name	Common name	Site 1	Site 2	Nativity
Allium textile	Prairie onion	X	X	Native
Alyssum simplex	Annual alyssum	X	X	Nonnative
Ambrosia psilostachya var. *coronopifolia*	Western ragweed	X	X	Native
Andropogon gerardii	Big bluestem	X	X	Native
Aristida purpurea	Purple threeawn	X	X	Native
Artemisia campestris	Field sagewort	X	X	Native
Artemisia frigida	Fringed sagebrush	X		Native
Artemisia ludoviciana	White sagebrush	X	X	Native
Astragalus shortianus	Short’s milkvetch	X	X	Native
Bouteloua gracilis	Blue grama	X	X	Native
Bromus tectorum	Downy brome	X	X	Nonnative
Castilleja integra	Wholeleaf Indian paintbrush		X	Native
Cerastium arvense	Field chickweed		X	Native
Cirsium undulatum	Wavyleaf thistle	X		Native
Cryptantha virgata	Miner’s candle	X		Native
Dalea purpurea	Purple prairie clover	X		Native
Delphinium carolinianum ssp. *virescens*	Carolina larkspur	X		Native
Descurainia pinnata	Western tansymustard		X	Native
Erigeron flagellaris	Trailing fleabane	X	X	Native
Eriogonum alatum	Winged buckwheat	X		Native
Eriogonum umbellatum	Sulfur-flower buckwheat	X	X	Native
Erodium cicutarium	Redstem filaree	X		Nonnative
Euphorbia brachycera	Horned spurge		X	Native
Gaillardia aristata	Blanketflower	X		Native
Helianthus pumilus	Little sunflower	X	X	Native
Hesperostipa comata	Needle-and-thread	X	X	Native
Heterotheca villosa	Hairy false goldenaster	X	X	Native
Iris missouriensis	Rocky Mountain iris	X	X	Native
Koeleria macrantha	Praire Junegrass		X	Native
Lappula occidentalis	Flatspine stickseed	X		Native
Lesquerella ludoviciana	Foothill bladderpod	X		Native
Leucocrinum montanum	Common starlily	X	X	Native
Liatris punctata	Dotted blazing star	X	X	Native
Linaria dalmatica	Dalmatian toadflax	X	X	Nonnative
Lomatium orientale	Northern Idaho biscuitroot	X	X	Native
Noccaea fendleri	Fendler’s pennycress		X	Native
Oenothera suffrutescens	Scarlet beeblossom		X	Native
Opuntia polyacantha	Plains pricklypear	X	X	Native
Oxytropis sericea	White locoweed	X		Native
Pascopyrum smithii	Western wheatgrass		X	Native
Penstemon secundiflorus	Sidebells penstemon	X	X	Native
Penstemon virens	Front Range beardtongue		X	Native
Poa compressa	Canada bluegrass		X	Nonnative
Pseudocymopterus montanus	Alpine false springparsley		X	Native
Psoralidium tenuiflorum	Slimflower scurfpea	X	X	Native
Ratibida columnifera	Upright prairie coneflower		X	Native
Rosa woodsii	Woods’ rose		X	Native
Schizachyrium scoparium	Little bluestem	X		Native
Senecio spartioides	Broom-like ragwort	X	X	Native
Toxicodendron rydbergii	Western poison-ivy		X	Native
Tragopogon dubius	Yellow salsify	X	X	Nonnative
Verbascum thapsus	Common mullein		X	Nonnative
Viola nuttallii	Nuttall’s violet	X	X	Native

herbicide treatments and one nontreated control were established in 3 by 6 m plots arranged in a randomized complete block design with six replications (Table 2). All treatments were applied with a CO_2-pressurized custom-built backpack sprayer using 11002LP flat-fan nozzles (TeeJet® Spraying Systems, P.O. Box 7900, Wheaton, IL 60187) delivering 187 L ha^{-1} at 207 kPa.

Treatment Evaluations and Data Analysis

To account for variability across the study area, native forb and shrub species were counted individually throughout the entire area of each plot from May to August (2016 and 2017) to determine species richness (total number of species) and abundance (number of individuals per species). Counts were conducted biweekly, targeting different species each time to account for varying life cycles, and individual species were counted only once per growing season. For rhizomatous or clonal plants, each clumping patch or grouping of stems was counted as one individual. To determine *B. tectorum* and native grass canopy cover, percent cover estimates of all grass species were determined by conducting visual evaluations across each entire plot (18-m^2 plot area) in August 2016 and 2017. Species richness was defined as the total number of different species occurring by plot (18-m^2 plot area), while species abundance was defined as total number of individuals per species per plot (18-m^2 plot area). Native grass cover was collected as percent cover per species; however, due to variability across the sites, species were combined

Table 2. Herbicides and rates applied in evaluating *Bromus tectorum* control and native species tolerance.

Common name	Rates applied[a] g ai ha^{-1}	Manufacturer
Picloram	561	Dow AgroSciences, Indianapolis, IN
Aminocyclopyrachlor	57	Bayer CropScience, Research Triangle Park, NC
Imazapic	105	BASF Specialty Products, Research Triangle Park, NC
Indaziflam	44	Bayer CropScience, Research Triangle Park, NC
Indaziflam	73	Bayer CropScience, Research Triangle Park, NC
Indaziflam	102	Bayer CropScience, Research Triangle Park, NC
Aminocyclopyrachlor + indaziflam	57 + 102	Bayer CropScience, Research Triangle Park, NC Bayer CropScience, Research Triangle Park, NC
Aminocyclopyrachlor + imazapic	57 + 105	Bayer CropScience, Research Triangle Park, NC BASF Specialty Products, Research Triangle Park, NC
Picloram + indaziflam	561 + 102	Dow AgroSciences, Indianapolis, IN Bayer CropScience, Research Triangle Park, NC
Picloram + imazapic	561 + 105	Dow AgroSciences, Indianapolis, IN BASF Specialty Products, Research Triangle Park, NC

[a] All treatments included 0.25% v/v nonionic surfactant.

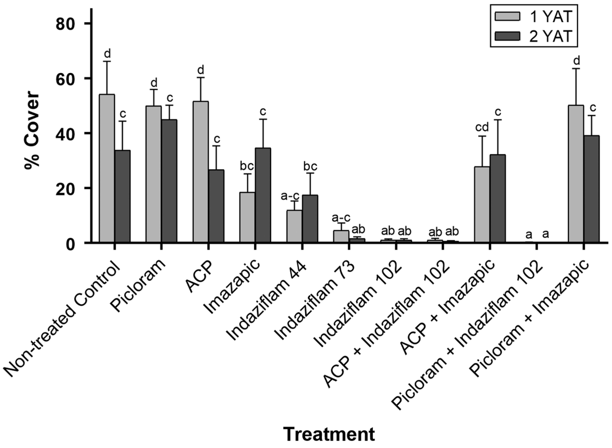

Figure 1. Percentage *Bromus tectorum* cover at Site 2 at 1 yr after treatment (YAT) (2016) and 2 YAT (2017). Letters indicate significant differences among herbicide treatments across years, using least-squares means ($P < 0.05$). Herbicide treatment rates are as follows: picloram (561 g ai ha^{-1}), aminocyclopyrachlor (ACP; 57 g ai ha^{-1}), imazapic (105 g ai ha^{-1}), indaziflam (44, 73, and 102 g ai ha^{-1}), and nontreated control.

into cool-season (C_3) and warm-season (C_4) cover categories. Cover data for *B. tectorum* and perennial grasses were arcsine square-root transformed to meet the assumptions of normality.

To test treatment effects on *B. tectorum* cover, a repeated-measures linear mixed-effects model was created using the 'lme4' package in R v. 3.4.3, testing for treatment effects at $\alpha = 0.05$ (R Core Team 2017). The fixed factors included in the model were treatment, year, and interactions, with year as the repeated measure; block was included as a random factor. Further analysis of the treatment and year effect was performed using the 'lsmeans' package in R (R Core Team 2017) to obtain comparisons between all pairs of least-squares means by year with a Tukey adjustment ($P < 0.05$). For grass cover data, the same analysis was performed for C_3 grass cover and C_4 grass cover. After rejection of the null hypothesis of equal variance for Sites 1 and 2, grass cover was analyzed separately by site.

Species richness was calculated by determining the number of native species in each plot. After failure to reject the null hypothesis of equal variance ($P = 0.3401$), sites were combined. A generalized linear mixed model was used to analyze count data, with treatment and year as fixed factors and block as the random factor. Count data for species richness were assumed to follow a Poisson distribution after failure to reject the null hypothesis that sample frequencies differed significantly from the expected frequencies under a Poisson distribution ($P = 0.1113$). Significant pairwise differences between richness were determined post hoc using a least-squares means test with a Tukey adjustment ('stats' and 'lsmeans' packages; R Core Team 2017).

To test treatment effects on overall native forb and shrub community abundance, dissimilarity matrices were generated on the collected abundance data using the Bray-Curtis method in Primer v. 7 (Bray and Curtis 1957; Clarke and Gorley 2015). Due to varying species amounts and types occurring at each site, sites and years were analyzed separately. Count data for each species were square-root transformed before creation of a resemblance matrix for each site and year by using Bray-Curtis dissimilarity measures. Homogeneity of variance (or dispersion) at each site by year was tested using permutational analysis of multivariate dispersions and was significant for Site 2 in 2017 (Site 1, 2016 $P = 0.109$; Site 2, 2016 $P = 0.257$; Site 1, 2017 $P = 0.055$; Site 2, 2017 $P = 0.002$). There was a dispersion effect at Site 2 in 2017. Permutational multivariate analysis of variance (PERMANOVA) is largely unaffected by heterogeneity for balanced designs and is more powerful than other tests in detecting actual changes in community structure (Anderson and Walsh 2013). Therefore, the resulting resemblance matrices were used to generate principal coordinate analyses (PCoA) to visualize differences among treatments. PERMANOVA was conducted to test treatment effects on native forb and shrub community composition (Anderson 2001; Anderson et al. 2008). PERMANOVA can be used as a nonparametric alternative to MANOVA and allows analysis of multiple variables (i.e., species counts) when data do not meet the assumptions of MANOVA. PERMANOVA were conducted using partial sums of squares on 999 permutations of residuals under a reduced model. Factors considered in the model were treatment as a fixed factor and block as a random factor. All multivariate analyses were conducted using PRIMER v. 7 and PERMANOVA+ (Primer-E, Plymouth, UK). Pairwise tests were performed by treatment levels using PERMANOVA+ (Primer-E, Plymouth, UK). Similarity percentage analysis (SIMPER) (Primer v. 7) for significant treatments was then used to identify specific species accounting for greater than 60% of the dissimilarity in community composition compared with the nontreated control. The analysis revealed whether the dissimilarity was primarily due to increases or decreases in species abundance.

Results and Discussion

Bromus Tectorum Control

Bromus tectorum cover decreased significantly at Site 1 during 2016 and 2017; therefore, only Site 2 was analyzed for treatment impacts to *B. tectorum* cover. Treatment was the only significant factor impacting *B. tectorum* cover ($P < 0.001$) (Supplementary Table S1). Compared with the nontreated control, all treatments containing indaziflam had less *B. tectorum* cover at 1 yr after

Table 3. Mean percentage cover of perennial cool season (C_3) grasses at both sites 1 and 2 yr after treatment (YAT).

	Perennial C_3 grass cover[a]			
	Site 1		Site 2	
	1 YAT	2 YAT	1 YAT	2 YAT
	%		%	
Nontreated control	27 a	43a	37 a	34 a
Picloram	55 bcd	65 ab	41 a	34 a
Aminocyclopyrachlor	68 cd	65 ab	32 a	49 a
Imazapic	39 ab	45 ab	44 a	34 a
Indaziflam 44	46 abc	50 ab	32 a	62 a
Indaziflam 73	42 abc	53 ab	28 a	51 a
Indaziflam 102	37 ab	54 ab	38 a	65 a
Aminocyclopyrachlor + indaziflam	52 a-d	56 ab	42 a	52 a
Aminocyclopyrachlor + imazapic	56 bcd	63 ab	39 a	53 a
Picloram + indaziflam	68 cd	70 b	45 a	50 a
Picloram + imazapic	74 d	69 ab	33 a	42 a

[a] Means followed by the same letter do not differ significantly within year at $P < 0.05$.

treatment (YAT) (0% to 22% cover). Indaziflam at the highest rate (102 g ai ha^{-1}) alone or tank mixed with aminocyclopyrachlor or picloram resulted in only 0.7 ± 0.3% (mean ± SE) *B. tectorum* cover at 1 YAT. The only other treatment to reduce *B. tectorum* cover at 1 YAT was imazapic applied alone (21.6 ± 0.6%) (Figure 1). Indaziflam at the highest rates (73 and 102 g ai ha^{-1}) alone or tank mixed with aminocyclopyrachlor or picloram continued to reduce *B. tectorum* cover at 2 YAT (0.8 ± 0.3%) (Figure 1). Although our data only represent one site, they were consistent with past studies showing multiyear *B. tectorum* control with indaziflam treatments compared with short-term (<1 yr) control with imazapic treatments (Kyser et al. 2007; Mangold et al. 2013; Morris et al. 2009; Sebastian et al. 2016, 2017a).

Impacts to Native Grasses

All native grasses occurring across the two sites were perennial grasses. Site 1 had significant treatment ($P < 0.001$) and year ($P = 0.02$) effects for C_3 grasses; however, the interaction of year by treatment was not significant (Supplementary Table S1). Comparisons made to the nontreated control showed increases in C_3 grass cover at 1 YAT for treatments containing picloram and aminocyclopyrachlor (average of 27% C_3 grass cover in nontreated control plots compared with 52% to 74% cover in the picloram- and aminocyclopyrachlor-treated plots). By 2 YAT, the only significant difference in C_3 grass cover at Site 1 was between the nontreated control and picloram plus indaziflam treatment (Table 3). There was no difference in warm-season grass cover at 1 or 2 YAT at Site 1 (Supplementary Tables S1 and S2). With little competition from *B. tectorum* at Site 1, differences in perennial grass cover were likely due to the forb reduction from the broadleaf herbicides. At Site 2 there was no treatment effect on C_3 grass cover ($P = 0.6324$) (Supplementary Table S1). For C_4 grass cover, a post hoc Tukey test revealed no significant pairwise differences between individual means separated by treatment (Supplementary Tables S1 and S2).

Impacts to Species Richness

The treatment by year interaction was not significant ($P = 0.4609$) for species richness although there was a treatment effect ($P < 0.001$). The only treatment to impact species richness was picloram combined with indaziflam 1 YAT, which reduced species richness compared with the nontreated control. The picloram plus indaziflam treatment had an average richness of 7.4 ± 0.66 compared with the nontreated control with an average species richness of 12.8 ± 0.59. By 2 YAT no impacts to species richness were observed (Supplementary Figure S1).

Impacts to Community Composition

Visualization of the PCoA plots suggested changes in community composition due to herbicide treatments at both sites, so a PERMANOVA analysis was performed to determine any treatment differences (Figure 2). For Site 1, PERMANOVA analysis showed impacts to the community composition of native species from herbicide treatments at 1 and 2 YAT ($P < 0.001$). All treatments containing picloram and aminocyclopyrachlor impacted species abundance compared with nontreated control plots at both 1 and 2 YAT (Table 4). Further analysis with SIMPER revealed that both broadleaf herbicide treatments decreased the abundance of most native forbs and shrubs included in the analysis (Supplementary Data File S1). Hairy false goldenaster [*Heterotheca villosa* (Pursh) Shinners var. *villosa*] and western ragweed [*Ambrosia psilostachya* DC. var. *coronopifolia* (Torr. & A. Gray) Farw.] were most impacted, contributing to more than 20% of the dissimilarity to the nontreated control at both 1 and 2 YAT (Supplementary Data File S1). No treatments resulted in increased species abundance at Site 1 (Table 4; Figure 2). At Site 2, the PERMANOVA also showed treatment effects to community composition of native species in both years ($P < 0.001$). At 1 YAT, treatments containing picloram reduced the abundance of most species, while treatments of imazapic alone and indaziflam at 44 g ai ha^{-1} increased species abundance compared with the nontreated control (Table 4). In year 2, treatments containing picloram still reduced species abundance; however, no treatments increased species abundance compared with the nontreated control (Table 4; Figure 2). Picloram had the greatest impacts to *H. villosa* and trailing fleabane (*Erigeron flagellaris* A. Gray), accounting for more than 30% and 40% of the dissimilarity to the nontreated control in 1 and 2 YAT, respectively (Supplementary Data File S1). In the imazapic-alone treatment, the greatest increases to abundance were to *A. psilostachya*, western poison-ivy [*Toxicodendron rydbergii* (Small ex Rydb.) Greene], and *H. villosa*, accounting for almost 40% of the dissimilarity to the nontreated control (Supplementary Data File S1). Indaziflam (44 g ai ha^{-1}) had the greatest increases to *A. psilostachya*, horned spurge (*Euphorbia brachycera* Engelm.), Nuttall's violet (*Viola nuttallii* Pursh), and sidebells penstemon (*Penstemon secundiflorus* Benth.), accounting for almost 45% of the dissimilarity (Supplementary Data File S1). Additional species contributing to the dissimilarity to the nontreated control can be viewed in Supplementary Data File S1. Reducing *B. tectorum* abundance can lead to increases in perennial grass and forb abundance as the competition for resources is removed (Monaco et al. 2017; Sebastian et al. 2016, 2017a; Thill et al. 1984; Whitson and Koch 1998); therefore, the increases in species abundance in Site 2 in the indaziflam and imazapic treatments are likely due to the reduction in *B. tectorum* cover.

The impacts to the native plant community differed between the two sites, although some responses to treatments were the same. At Site 1, no treatments resulted in increased native forb or shrub abundance compared with the nontreated control, while broadleaf herbicides increased C_3 grass cover. At the same site, both broadleaf herbicides (picloram and aminocyclopyrachlor) reduced

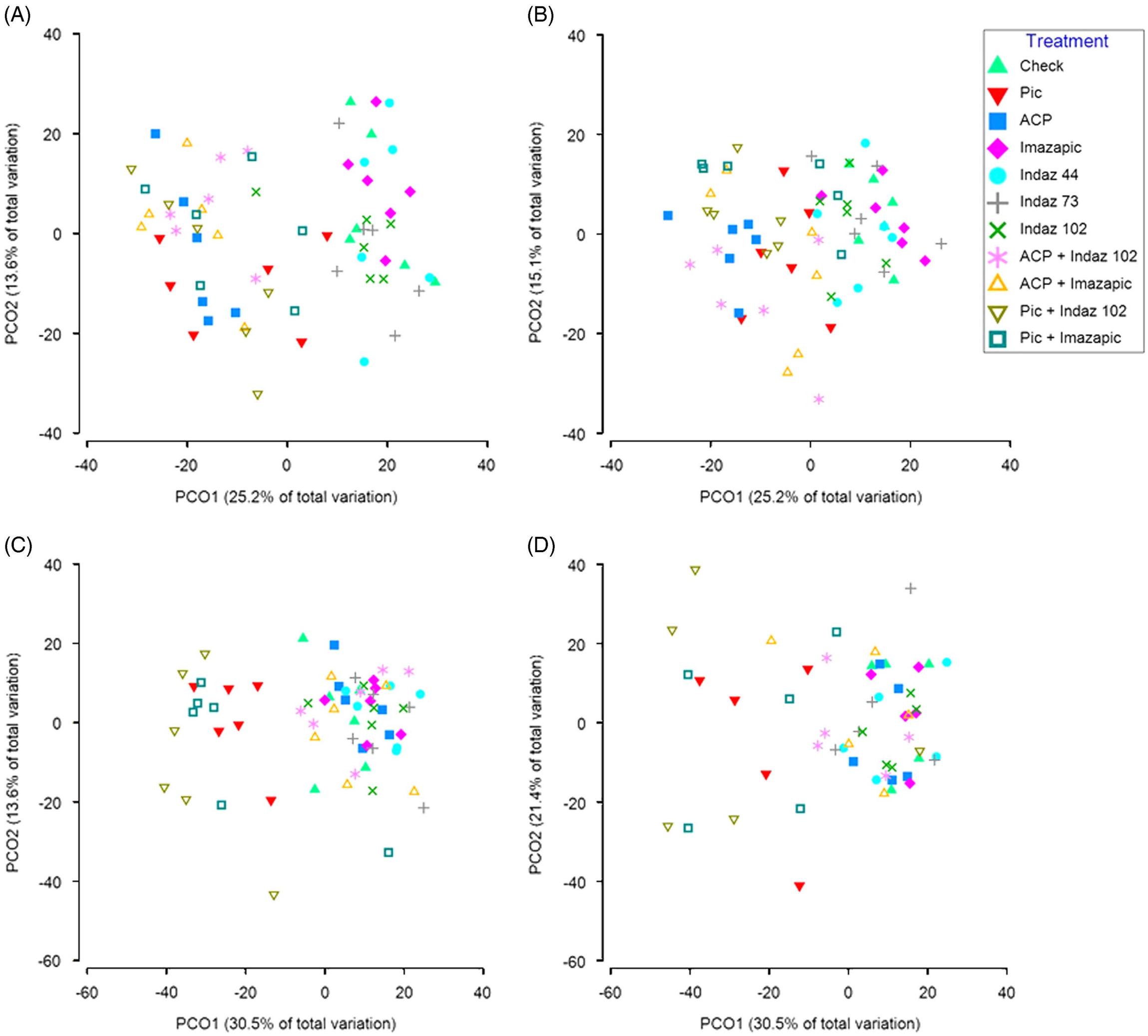

Figure 2. Principal coordinate analysis (PCoA) of native forb and shrub species abundance separated by treatment. Treatments farther away from the nontreated (check, represented by the green triangle) had more dissimilarities in community composition. The analysis was based on the Bray-Curtis dissimilarity matrix constructed using the square-root-transformed species counts from Site 1 in 2016 (A) and 2017 (B) and Site 2 in 2016 (C) and 2017 (D). The percent of variation explained is given in brackets on the x- and y-axes. ACP, aminocyclopyrachlor; Indaz, indaziflam; PIC, picloram.

native forb and shrub abundance, while the annual grass herbicides (indaziflam and imazapic) had no impact on the overall community composition. The shift to a more C_3 grass–dominated community in the plots treated with broadleaf herbicides is likely due to the reduction in forb and shrub abundance (Arnold and Santelmann 1966; Greet et al. 2016). At Site 2, which was characterized by 60% to 70% *B. tectorum* cover, only picloram decreased species abundance, while increases in species abundance at 1 YAT were observed among a few treatments that reduced *B. tectorum* cover. Decreases in species richness were also observed from one picloram treatment at this site as well. These findings support extensive research showing decreases in native forb abundance from picloram applications and more recent work showing transient forb decreases from aminocyclopyrachlor applications (Arnold and Santelmann 1966; Carter and Lym 2018; Greet et al. 2016; Ortega and Pearson 2011; Thilmony and Lym 2017; Wagner and Nelson 2014).

Much of the research examining herbicide impacts on native species abundance is compounded by noxious weed competition at the site (Arnold and Santelmann 1966; Beran et al. 1999; Carter and Lym 2018; Davies and Sheley 2011; Elseroad and Rudd 2011). This can make separating herbicide impacts from invasion impacts difficult. In sites dominated by invasive weeds, especially long-term invasions, the diversity of the native plant community has already been compromised, while in noninvaded, intact plant communities, there is a higher potential for loss as native species have not been impacted by nonnative invaders (Davies and Sheley 2011; Duncan et al. 2004). In a study conducted by Ortega and Pearson (2011), the authors presented an impact gradient for picloram that coincided with spotted knapweed (*Centaurea stoebe* L.) invasion levels. Their study found that native forb cover declined > 20% in treated plots versus control plots at noninvaded sites, while impacts to forb cover were minimal in sites with moderate to high *C. stoebe* invasion levels. The authors concluded that differences in picloram effects on native species was due to the strength of release effects from the invasive species, as the increased diversity in sites void of *C. stoebe* had more loss potential than sites already suffering the effects of invasion. This offers a possible explanation for why decreased native species abundances were observed from aminocyclopyrachlor treatments in the site with a more intact native plant community versus the site dominated by *B. tectorum*.

To date, the two published field studies showing indaziflam treatments resulting in long-term *B. tectorum* control reported no

Table 4. Pairwise comparisons of herbicide treatments versus nontreated control for native forb abundance from permutational multivariate analysis of variance analysis.

Treatment versus nontreated control	Site 1 2016	Site 1 2017	Site 2 2016	Site 2 2017
	P-value[a]		P-value[a]	
Picloram	0.017*	0.019*	0.01*	0.015*
Aminocyclopyrachlor	0.004*	0.006*	0.358	0.146
Imazapic	0.367	0.35	0.018^{+}	0.776
Indaziflam 44	0.581	0.557	0.045^{+}	0.177
Indaziflam 73	0.552	0.407	0.199	0.676
Indaziflam 102	0.453	0.162	0.324	0.2
Aminocyclopyrachlor + indaziflam	0.015*	0.032*	0.456	0.071
Aminocyclopyrachlor + imazapic	0.008*	0.012*	0.085	0.24
Picloram + indaziflam	0.026*	0.014*	< 0.001*	0.037*
Picloram + imazapic	0.012*	0.028*	0.033*	0.028*

[a] P-values marked with asterisks (*) are considered significant abundance reductions at the <0.05 level. P-values marked with plus signs (+) are considered significant abundance increases at the <0.05 level.

observable negative impacts to native species (Sebastian et al. 2016, 2017a). *Bromus tectorum* control with indaziflam at 58 g ai ha^{-1} lasted 3 yr with no injury to crested wheatgrass [*Agropyron cristatum* (L.) Gaertn.] and western wheatgrass [*Pascopyrum smithii* (Rydb.) Á. Löve] or impacts to forb species richness (Sebastian et al. 2016). Another study by Sebastian et al. (2017a) reported 2 yr of *B. tectorum* control from indaziflam (44, 73, and 102 g ai ha^{-1}), with increased perennial grass and forb biomass and no impact to forb species richness. Our data corroborate previous findings of native species tolerance to indaziflam applications, while also showing that the community composition and abundance of native species is not impacted. The literature on impacts to perennial species abundance with imazapic applications is more diverse, and past findings have been variable, showing no impact to species abundance or impacts to specific perennial species, especially in areas with low annual precipitation or during periods of drought (Beran et al. 1999; Kyser et al. 2007; Monaco et al. 2005; Morris et al. 2009; Shinn and Thill 2002). Our study found no evidence of decreases in species abundance with imazapic applications. Our findings from Site 2 are consistent with previous research showing multiyear *B. tectorum* control with indaziflam applications (Sebastian et al. 2016, 2017a) and variability in control with imazapic applications (Davies and Sheley 2011; Davison and Smith 2007; Elseroad and Rudd 2011).

One important aspect land managers must take into account when considering the results from this study and developing large-scale weed management plans is interannual variability in plant community composition. Although *B. tectorum* was initially a target species for control in this study, the cover at one site decreased to a negligible level (<5% cover) the year after treatments were applied. *Bromus tectorum* invasion levels can decrease during periods of drought and return with increased fall and winter moisture (Mack and Pyke 1983). Climatic variation can also impact weed control and injury to native species from herbicide treatments (Evans et al. 1969; Sebastian et al. 2017c).

Indaziflam is an effective tool for multiyear *B. tectorum* control (Sebastian et al. 2016, 2017a), and our results suggest that this herbicide can be used in non-crop sites without impact to native perennial species. Land managers should consider impacts to the plant community when using broadleaf herbicides in these sites, as there is a potential to decrease forb and shrub abundance and shift to a more grass-dominated ecosystem. Integrating indaziflam into current management programs could provide the length of *B. tectorum* control needed to deplete the invasive annual grass seedbank and release the remnant plant community (Chambers et al. 2014; Elseroad and Rudd 2011; Sebastian et al. 2017b). Reestablishing the dominant native perennial plant community further increases the resistance and resilience of that ecosystem to future *B. tectorum* invasions and decreases fine fuels from invasive annual grass that are associated with wildfires (Chambers et al. 2014). Future studies should evaluate the length of *B. tectorum* control and native species tolerance across varying climates and soil types, specifically in more arid regions such as the Great Basin.

Supplementary material. To view supplementary material for this article, please visit https://doi.org/10.1017/inp.2019.4

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References

Abatzoglou JT, Kolden CA (2011) Climate change in western US deserts: potential for increased wildfire and invasive annual grasses. Rangeland Ecol Manag 64:471–478

Anderson MJ (2001) Permutation tests for univariate or multivariate analysis of variance and regression. Can J Fish Aquat Sci 58:626–639

Anderson MJ, Gorley RN, Clarke RK (2008) Permanova + for Primer: guide to software and statistical methods. Plymouth, UK: Primer-E Limited

Anderson MJ, Walsh DC (2013) PERMANOVA, ANOSIM, and the Mantel test in the face of heterogeneous dispersions: what null hypothesis are you testing? Ecol Monogr 83:557–574

Anonymous (1996) Plateau® herbicide, for weed control, native grass establishment and turf growth suppression on roadsides and other noncrop areas. PE47013. Parsippany, NJ: American Cyanamid Company. 17 p

Anonymous (2011a) Indaziflam 500 SC herbicide product label. Research Triangle Park, NC: Bayer CropScience. 11 p

Anonymous (2011b) Indaziflam 74SC herbicide product label. Research Triangle Park, NC: Bayer Environmental Science. 16 p

Anonymous (2016) Esplanade® 200SC supplemental label- for release or restoration of desirable vegetation. Research Triangle Park, NC: Bayer Environmental Science. 2 p

Anonymous (2018) Tordon® 22k herbicide product label. Indianapolis, IN: Dow AgroSciences. 11 p

Arnold WR, Santelmann PW (1966) The response of native grasses and forbs to picloram. Weeds 14:74–76

Beck KG (2009) Downy Brome (*Bromus tectorum*) and Japanese Brome (*Bromus japonicus*) Biology, Ecology, and Management. http://mining.state.co.us/SiteCollectionDocuments/DownybromeandJapanesebromeliteraturereviewColoradoDRMSDec09.pdf. Accessed: July 21, 2018

Beran DD, Masters RA, Gaussoin RE (1999) Grassland legume establishment with imazethapyr and imazapic. Agron J 91:592–596

Billings WD (1994) Ecological Impacts of Cheatgrass and Resultant Fire on Ecosystems in the Western Great Basin. General Technical Rep INT-GTR-313. Ogden, UT: U.S. Department of Agriculture, Forest Service, Intermountain Research Station. https://www.fs.fed.us/rm/pubs_int/int_gtr313/int_gtr313_022_030.pdf. Accessed: July 18, 2018

Brabham C, Lei L, Gu Y, Stork J, Barrett M, DeBolt S (2014) Indaziflam herbicidal action: a potent cellulose biosynthesis inhibitor. Plant Physiol 166:1177–1185

Bray JR, Curtis JT (1957) An ordination of the upland forest communities of southern Wisconsin. Ecol Monogr 27:325–349

Carter TR, Lym RG (2018) Native forb response to aminocyclopyrachlor. Invasive Plant Sci Manag 11:56–60

Chambers JC, Pyke DA, Maestas JD, Pellant M, Boyd CS, Campbell SB, Espinosa S, Havlina DW, Mayer KE, Wuenschel A (2014) Using Resistance and Resilience Concepts to Reduce Impacts of Invasive Annual Grasses and Altered Fire Regimes on the Sagebrush Ecosystem and Greater Sage-Grouse: A Strategic Multi-scale Approach. General Technical Rep RMRS-GTR-326. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. http://www.fs.fed.us/rm/pubs/rmrs_gtr326.pdf. Accessed: July 18, 2018

Clarke KR, Gorley RN, 2015. PRIMER v7: User Manual/Tutorial. Plymouth, UK: PRIMER-E. 296 p

Davies KW, Sheley RL (2011) Promoting native vegetation and diversity in exotic annual grass infestations. Ecol Restor 19:159–165

Davison JC, Smith EG (2007) Imazapic provides 2-year control of weedy annuals in a seeded Great Basin fuelbreak. Native Plants J 8:91–96

Diamond JM, Call CA, Devoe N (2012) Effects of targeted grazing and prescribed burning on community and seed dynamics of a downy brome (*Bromus tectorum*)–dominated landscape. Invasive Plant Sci Manag 5:259–269

DiTomaso JM, Masters RA, Peterson VF (2010) Rangeland invasive plant management. Rangelands 32:43–47

Duncan CA, Jachetta JJ, Brown ML, Carrithers VF, Clark JK, DiTomaso JM, Lym RG, McDaniel KC, Renz MJ, Rice PM (2004) Assessing the economic, environmental, and societal losses from invasive plants on rangeland and wildlands. Weed Technol 18:1411–1416

Elseroad AC, Rudd NT (2011) Can imazapic increase native species abundance in cheatgrass (*Bromus tectorum*) invaded native plant communities? Rangeland Ecol Manag 64:641–648

Evans RA, Eckert RE, Burgess KL, Young JA (1969) Downy brome control by soil-active herbicides for revegetation of rangelands. Weed Sci 17:166–169

Greet BJ, Mealor BA, Kniss AR (2016) Response of *Delphinium occidentale* and associated vegetation to aminocyclopyrachlor. Rangeland Ecol Manag 69:474–480

Kelley WK, Fernandez-Gimenez ME, Brown CS (2013) Managing downy brome (*Bromus tectorum*) in the Central Rockies: land manager perspectives. Invasive Plant Sci Manag 6:521–535

Knapp PA (1996) Cheatgrass (*Bromus tectorum* L) dominance in the Great Basin Desert: history, persistence, and influences to human activities. Global Environ Chang 6:37–52

Kyser GB, DiTomaso JM, Doran MP, Orloff SB, Wilson RG, Lancaster DL, Lile DF, Porath ML (2007) Control of medusahead (*Taeniatherum caput-medusae*) and other annual grasses with imazapic. Weed Technol 21:66–75

Kyser GB, Wilson RG, Zhang J, DiTomaso JM (2013) Herbicide-assisted restoration of Great Basin sagebrush steppe infested with medusahead and downy brome. Rangeland Ecol Manag 66:588–596

Mack RN (1981) Invasion of *Bromus tectorum* L. into western North America: an ecological chronicle. Agro-Ecosystems 7:145–165

Mack RN, Pyke DA (1983) The demography of *Bromus tectorum*: variation in time and space. J Ecol 1:69–93

Mangold J, Parkinson H, Duncan C, Rice P, Davis E, Menalled F (2013) Downy brome (*Bromus tectorum*) control with imazapic on Montana grasslands. Invasive Plant Sci Manag 6:554–558

Monaco TA, Osmond TM, Dewey SA (2005) Medusahead control with fall and spring-applied herbicides on northern Utah foothills. Weed Technol 19:653–658

Monaco TA, Mangold JM, Mealor BA, Mealor RD, Brown CS (2017) Downy brome control and impacts on perennial grass abundance: a systematic review spanning 64 years. Rangeland Ecol Manag 70:396–404

Morris C, Monaco TA, Rigby CW (2009) Variable impacts of imazapic rate on downy brome (*Bromus tectorum*) and seeded species in two rangeland communities. Invasive Plant Sci Manag 2:110–119

Ortega YK, Pearson DE (2011) Long-term effects of weed control with picloram along a gradient of spotted knapweed invasion. Rangeland Ecol Manag 64:67–77

Price R, Parker KW, Hull AG Jr. (1948) Range practices and problems. Grass: The Yearbook of Agriculture 1948. U.S. Department of Agriculture, p 557–560

R Core Team (2017) R: A Language and Environment for Statistical Computing. Vienna, Austria: R Foundation for Statistical Computing. https://www.R-project.org Accessed: April 30, 2018

Sebastian DJ, Fleming MB, Patterson EL, Sebastian JR, Nissen SJ (2017a) Indaziflam: a new cellulose-biosynthesis-inhibiting herbicide provides long-term control of invasive winter annual grasses. Pest Manag Sci 73:2149–2162

Sebastian DJ, Nissen SJ, Sebastian JR, Beck KG (2017b) Seed bank depletion: the key to long-term downy brome (*Bromus tectorum* L.) management. Rangeland Ecol Manag 70:477–483

Sebastian DJ, Nissen SJ, Westra P, Shaner DL, Butters G (2017c) Influence of soil properties and soil moisture on the efficacy of indaziflam and flumioxazin on *Kochia scoparia* L. Pest Manag Sci 73:444–451

Sebastian DJ, Sebastian JR, Nissen SJ, Beck KG (2016) A potential new herbicide for invasive annual grass control on rangeland. Rangeland Ecol Manag 69:195–198

Shinn SL, Thill DC (2002) The response of yellow starthistle (*Centaurea solstitialis*), annual grasses, and smooth brome (*Bromus inermis*) to imazapic and picloram. Weed Technol 16:366–370

Tateno M, Brabham C, DeBolt S (2016) Cellulose biosynthesis inhibitors—a multifunctional toolbox. J Exp Bot 67:533–542

Thacker E, Ralphs MH, Monaco TA (2009) Seeding cool-season grasses to suppress broom snakeweed (*Gutierrezia sarothrae*), downy brome (*Bromus tectorum*), and weedy forbs. Invasive Plant Sci Manag 2:237–246

Thill DC, Beck KG, Callihan RH (1984) The biology of downy brome (*Bromus tectorum*). Weed Sci 32:7–12

Thilmony BM, Lym RG (2017) Effect of aminocyclopyrachlor on native prairie species in the Northern Great Plains. Invasive Plant Sci Manag 10:201–209

[USDA-NRCS] U.S. Department of Agriculture, Natural Resources Conservation Service (2014) Web Soil Survey. http://websoilsurvey.nrcs.usda.gov. Accessed: April 30, 2018

Wagner V, Nelson CR (2014) Herbicides can negatively affect seed performance in native plants. Ecol Restor 22:288–291

Warg SA (1938) Life History and Economic Studies on *Bromus tectorum*. Master's thesis. Missoula, MT: University of Montana. 38 p

Western Regional Climate Center (2018) Cooperative Climatological Data Summaries. http://www.wrcc.dri.edu/climatedata/climsum. Accessed: April 30, 2018

Whisenant SG (1990) Changing fire frequencies on Idaho's Snake River Plains: ecological and management implications. Pages 4–10 *in* McArthur ED, Romney EM, Smith SD, Tueller PT, eds. Proceedings of the Symposium on Cheatgrass Invasion, Shrub Die-Off, and Other Aspects of Shrub Biology and Management. Ogden, UT: USDA Forest Service, Intermountain Research Station General Technical Report INT–276. 351 p

Whitson TD, Koch DW (1998) Control of downy brome (*Bromus tectorum*) with herbicides and perennial grass competition. Weed Technol 1:391–396

Whitson TD, Majerus ME, Hall RD, Jenkins JD (1997) Effects of herbicides on grass seed production and downy brome (*Bromus tectorum*). Weed Technol 1:644–648