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Invasive Plant Science and Management

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Indaziflam controls nonnative *Alyssum* spp. but negatively affects native forbs in sagebrush steppe

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Research Article

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Abstract

Nonnative plant invasions can have devastating effects on native plant communities; conversely, management efforts can have nontarget and deleterious impacts on desirable plants. In the arid sagebrush steppe rangelands of the western United States, nonnative winter annual species affect forage production and biodiversity. One method proposed to control these species is to suppress the soil seedbank using the preemergent herbicide indaziflam. Our goal was to evaluate the efficacy of indaziflam to control nonnative annual mustards (*Alyssum* spp.) and to understand potential nontarget effects of management on the diverse mountain sagebrush steppe plant communities within Yellowstone National Park. Six sites were established along an elevation gradient (1,615 to 2,437 m), each with high and low *Alyssum* spp. infestations. We applied 63g ai ha^{-1} of indaziflam in late summer of 2018 and evaluated plant community cover in situ for 2 yr after treatment and emergence of forb species from the soil seedbank ex situ. Indaziflam was highly effective at controlling emergence of *Alyssum* spp. for 2 yr. Richness and Shannon's diversity of the nontarget plant community were significantly lower in sprayed plots than in the control, and both decreased along the elevation gradient. These reductions were due to a decrease in perennial forbs and native annual forbs in the sprayed plots; perennial graminoids were not affected. Overall, the aboveground and seedbank community composition was negatively impacted by indaziflam, and these effects were strongest for the native annual forbs that rely on annual regeneration from the seedbank. The effects of this herbicide to the nontarget community should be evaluated beyond the length of our study time; however, we conclude that indaziflam should likely be reserved for use in areas that are severely invaded and have seedbanks that are composed of nondesirable species rather than diverse, native mountain sagebrush communities.

Introduction

Nonnative plant invasions can have devastating impacts on native plant communities (Elton 1958; Mack and D'Antonio 1998; Tilman 1999; Vilà et al. 2011). However, chemical efforts to control nonnative plants can also damage co-occurring native species and communities (Crone et al. 2009; Kettenring and Adams 2011; Rinella et al. 2009; Wagner and Nelson 2014), sometimes causing more harm than the invader itself (Ortega and Pearson 2011; Skurski et al. 2013). Understanding the efficacy of herbicides on target species and potential adverse impacts on nontarget desirable species is key to developing effective management strategies.

Controlling invasive plants in semiarid grasslands is critical to maintaining the essential functions and services this ecosystem provides to humans around the world (Lund 2007; O'Mara 2012). The sagebrush steppe in the western United States is a diverse ecological community that provides forage for livestock operations and habitat for wildlife species (Beck et al. 2012). This semiarid ecosystem is threatened by land use change, climate change, and nonnative plant invasions (DiTomaso 2000; Knapp 1996; Vasquez et al. 2010), particularly winter annual grass species that can alter fire regimes and disrupt ecosystem functions (Balch et al. 2013; Billings 1994; Young and Fay 1997), creating novel plant communities far less diverse than the native communities they replaced (Allen and Knight 1984).

The preemergent herbicide indaziflam is promoted as an option to control invasive annual grasses in sagebrush steppe (Clark et al. 2020; Sebastian et al. 2016). Indaziflam is a nonselective herbicide that inhibits cellulose biosynthesis (Brabham et al. 2014) and provides residual control in the top few centimeters of soil (0 to 2.5 cm). Originally developed for turf and orchard use, indaziflam was recently approved for use in natural areas and grazed rangeland (Bayer 2020) and may be used to provide long-term control of invasive annual grasses by depleting the soil

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Management Implications

The sagebrush steppe rangelands of the American West are threatened by infestations of annual nonnative plant species, and an effective approach to controlling these species is through seedbank depletion. Indaziflam is a preemergent herbicide that can effectively deplete the soil seedbank of invasive annual grasses. This herbicide shows great promise for control of these annual invaders due to its residual soil activity of up to 3 yr. There have been no documented negative effects to established perennial vegetation; however, nontarget impacts to the native seedbank are expected. A field study was conducted to examine the impacts of indaziflam on nontarget species aboveground, and a greenhouse study addressed impacts to the soil seedbank. The mountain big sagebrush (*Artemisia tridentata* ssp. *vaseyana*) sites had localized infestations of nonnative annual mustards (*Alyssum* spp.) and high levels of species richness and diversity. Overall indaziflam controlled *Alyssum* spp. for 2 yr but also reduced richness and diversity of the nontarget forb community. Annual forbs were the most impacted, both aboveground—with reductions of up to 50% cover—and in the soil. Perennial forbs were also prevented from regenerating from the soil seedbank. Native annual forb species are of particular concern, as they rely on annual regeneration from the seedbank, whereas established perennial species can repopulate after the herbicide has degraded. The soil seedbank is often relied upon for passive restoration efforts, and broadscale use of indaziflam may not be appropriate in areas with minimal infestations. Our findings highlight the importance of considering nontarget impacts when using indaziflam in areas with diverse native plant communities and low infestations of annual nonnative species.

seedbank (Sebastian et al. 2017a, 2017b). Indaziflam shows great promise to reduce target species, as this herbicide remains active in the soil for up to 3 yr (Sebastian et al. 2016) and would potentially require fewer applications than other less persistent herbicides (Sebastian et al. 2017a).

The efficacy of indaziflam in sagebrush rangeland has been evaluated primarily in highly disturbed areas dominated by invasive annual grasses with little remaining native vegetation (Sebastian et al. 2017a). The control of invasive annual grasses results in an increase in growth of existing perennial plants due to reduced competition (Sebastian et al. 2017a, 2020). Established perennial vegetation is largely unaffected by indaziflam (Clark et al. 2019), likely because roots of perennial plants often extend below the zone of herbicide activity. However, new recruitment of a nonnative perennial species was inhibited by indaziflam (Sebastian et al. 2017c), and it is likely that any germinating seeds in the zone of herbicide activity will not emerge, as indaziflam inhibits cellulose biosynthesis in the radicle and is nonselective (Brabham et al. 2014). Species with annual life cycles and short-lived seedbanks will be more impacted by indaziflam than either perennial species that do not rely on annual germination or species with long-term seedbanks. Only a couple of indaziflam studies have examined impacts to annual forbs (Clark et al. 2019; Sebastian et al. 2020), and one of those found a slight increase in native annual cover after indaziflam treatment (Sebastian et al. 2020).

The forb desert alyssum or desert madwort (*Alyssum desertorum* Stapf) is a nonnative winter annual species common to the sagebrush steppe (Noack 2020) and is purported to displace native vegetation (Mosley 2014), though quantitative evidence of its impacts is lacking. Two other nonnative annual mustards [pale madwort (*Alyssum alyssoides* (L.) L.) and alyssum (*Alyssum simplex* Rudolphi)], can co-occur but receive less attention. In Yellowstone National Park (YNP), *A. desertorum* has recently been found above its documented elevational range, primarily in areas disturbed by wildlife, tourists walking off designated routes, or recent construction (H Anderson, personal communication). This spread has prompted YNP land managers to consider controlling localized infestations of *A. desertorum* to maintain the native diversity of these areas. Managers are controlling infestations of *A. desertorum* with indaziflam in one area of YNP that has experienced over a century of intense human land use. Using this herbicide to control *Alyssum* spp. in less disturbed and more species-diverse areas of YNP needs to be evaluated. Previous indaziflam studies in other semiarid grasslands have primarily focused on the response of perennial forbs and perennial grasses, as they are desirable components of rangeland plant communities; however, annual ephemeral forbs are also key components of rangelands (Pokorny et al. 2004). They provide critical spring forage for many wildlife species (Drut et al. 1994; Luna et al. 2018) and occupy an early successional niche. We sought to assess the impacts of indaziflam on the native plant community with a primary focus on annual forbs.

Our study examined the efficacy of preemergent indaziflam to control nonnative *Alyssum* spp. and evaluated impacts to the diversity of the nontarget plant community. The objectives were: (1) to evaluate the efficacy of indaziflam to control the target species; (2) to assess the effect of indaziflam on the richness and diversity of the whole plant community, distinguishing between perennial and annual species; and (3) to evaluate the effect of indaziflam on perennial and annual forb germination from the soil seedbank.

Materials and Methods

Study Area and Experimental Design

Six field sites were established along an elevational gradient (1,615 m to 2,347 m) in the northern range of YNP, USA (Table 1). Sites were randomly selected from a roadside survey of *A. desertorum* populations conducted with aid of the park botanist (H Anderson). However, further evaluation found a mix of *A. desertorum* with *A. alyssoides* and *A. simplex* at these sites (*Alyssum* spp. hereafter). The plant communities at all sites were characterized as mountain big sagebrush steppe (dominated by *Artemisia tridentata* Nutt. ssp. *vaseyana*).

We defined high and low invasion levels of *Alyssum* spp. at each site, with high levels equal to or above 10 individuals m^{-2}, and adjacent low levels with fewer than 10 individuals m^{-2}. Before spray application we assessed *Alyssum* spp. in each plot, using density classes (1: 0; 2: <10; 3: 10 to 100; and 4: >100 individuals m^{-2}). High-invasion plots were mainly in the high (4) category (75%, 78 plots) with the rest in the medium (3) level (25%, 26 plots [104 plots total]). Most of the low-invasion plots had no *Alyssum* spp. (94%, 96 of 102 plots [2 plots lost to the study]); only 6 had between 1 and 10 *Alyssum* spp. At five of the sites, eighteen 1-m^2 plots were established in each of the high- and low-invasion areas. In each of the invasion treatments, 12 plots were sprayed with indaziflam (Esplanade®, Bayer CropScience, Cary, NC, USA, 27513) and six plots were left unsprayed to serve as controls. At the sixth and highest site, fourteen 1-m^2 plots were established in each high- and low-invasion area, with seven

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Table 1. Site environmental characteristics for the six sites along an elevation gradient within Yellowstone National Park.

Site name	Species richness	Elevation	Coordinates		Soil texture	Invasion[a]	Aspect	Percent slope
		m	Latitude °N	Longitude °W				
Mammoth	62	1,615	44.98449	110.70107	Loam	High	West	11
						Low	West	7
Blacktail	72	1,980	44.95592	110.59104	Loam	High	South	2
						Low	South	2
Lamar	61	1,981	44.91088	110.32497	Loam	High	North	4
						Low	North	9
Phantom	76	2,042	44.95556	110.50489	Clay-loam	High	South	22
					Loam	Low	South	6
Swan	70	2,225	44.91345	110.72971	Loam	High	Flat	0.2
						Low	Flat	0.4
Hayden	38	2,347	44.65238	110.46549	Loam-sand	High	South	27
					Loam	Low	South-East	17

[a]High: areas with >10 *Alyssum* spp. individuals m^{-2}; Low: areas with <10 *Alyssum* spp. individuals m^{-2}.

of the plots in each invasion treatment sprayed with indaziflam and seven plots left unsprayed as controls. We had a high level of replication and chose our plot size to ensure we had a high detection rate for small ephemeral annual species, allowing adequate representation of the plant community. Plots were randomly assigned to the spray or control treatment. All spray plots were treated in August 2018 with 63 g ai ha^{-1} indaziflam using a backpack sprayer fitted with a XR11002 flat spray nozzle (TeeJet® Spraying Systems, P.O. Box 7900, Wheaton, IL 60187) at 195 L ha^{-1} at 138 kPa. Ocular estimates of foliar cover (to the nearest 1%) for each species and ground cover were conducted 1 and 2 yr after treatment during peak vegetative season, for the central 0.75 m^2 of each plot to account for potential edge effects of the herbicide. Species with "trace" (<0.5%) cover were recorded and analyzed as 0.1% cover. Estimates were allowed to exceed 100% to account for understory canopy structure. The sites were sampled from low to high elevation with the aim of sampling each site at comparable growth phases. Half of the spray plots were sampled 1 mo after the other plots at each site; this enabled us to us to determine whether we had missed any species and provided a better understanding of the plant community (Pokorny et al. 2004).

Evaluating the Effect of Herbicide on the Soil Seedbank

To assess the effect of herbicide on the seedbank, soil samples were collected at the two lowest sites in April 2019. At each site, soil cores were taken in each of six spray plots and six control plots in both areas of invasion treatments ($n = 48$). Six soil cores (10-cm diameter by 6-cm deep) were taken per plot in the area outside the vegetation quadrat but within the 1-m^2 plot and combined into one soil sample. Soil samples (2,832 cm^3) were stored in a cold storeroom at 5 C for 2 wk, then spread out in trays (28 by 13 cm) on top of 2.5 cm of sterilized soil (1:1:1 ratio of mineral soil, sphagnum peat moss, and washed concrete sand) in the Montana State University Plant Growth Center (Bozeman, MT, 59717). Trays were placed in a greenhouse (22 to 18 C, 16-h photoperiod) and watered twice each day for 5 min using a drip irrigation system. Seedling emergence above the soil surface was recorded at least once per week. Seedlings that emerged were identified, counted, and then removed from the tray. Seedlings that were unidentifiable at the 1- to 2-leaf stage were repotted and grown until they were identifiable. Emerging graminoids were removed and not counted. There was no germination for the first 45 d; therefore, trays were moved to a cold, wet, and dark stratification chamber (4 C) for 30 d to break dormancy, after which the trays were returned to the greenhouse. After germination diminished again (6 mo) the trays were returned to the stratification chamber for 56 d, and then returned to the greenhouse. Any additional germinants were identified and removed. The experiment was terminated after 17 mo.

Statistical Analysis

All analyses were completed using R v. 3.6.1 (R Core Team 2019) and the packages BIODIVERSITYR (Kindt and Coe 2005), LME4 (Bates et al. 2015), LMERTEST (Kuznetsova et al. 2017), MUMIN (Bartoń 2019), and VEGAN (Oksanen et al. 2019).

To evaluate the effects of indaziflam on the abundance of *Alyssum* spp. (objective 1), the high invasion data were subset, and a linear mixed model following a Gaussian distribution was fit. The saturated model included herbicide treatment (spray, control), elevation, and year (2019, 2020) as main effects. To account for repeated measures of plots between years, the unique tag number for each plot was included as a random effect. For objective 2, indaziflam effects on the plant community, the full data set was used, including both levels of *Alyssum* spp. invasion. Species richness and Shannon's diversity index were assessed for the whole community, perennial graminoids, perennial forbs, and annual native forbs. Percent cover estimates were used to calculate Shannon's diversity index for each plot. Shannon's diversity index was calculated as

$$H = -\sum_{i=1}^{S} p_i * \log b * p_i \quad [1]$$

where p_i is the proportion of species i, S is the number of species, and log$b$ is the logarithm base 10. Species richness was evaluated with generalized linear mixed-effects models following a Poisson distribution, and Shannon's diversity was evaluated following a Gaussian distribution. A quasi-Poisson distribution was used when overdispersion was present. Saturated models with all possible interactions were fit with main and random effects as noted earlier and including invasion (high, low) as a main effect. Any significant interactions were maintained in the final models. Normality and heteroscedasticity assumptions were assessed, and no transformations were necessary. The model intercept was the lowest elevation

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Table 2. Cover (%) of *Alyssum* spp. (ALSPP) in high-invasion plots in response to indaziflam treatment (control, sprayed), elevation, and year (2019, 2020).[a]

		Fixed effects					Random effect Variance	
Response	Predictor	Est.	SE	df	*t*-value	P(>)	Tag	Residual
% cover ALSPP	Intercept	4.11	1.06	105	2.06	**<0.01**	0.62 ± 0.79	1.42 ± 1.19
$R^2 = 0.63$	Indaziflam	−3.29	0.29	185	−11.19	**<0.01**		
	Elevation	3.7×10^4	5.1×10^4	101	−0.72	0.47		
	Year	−0.88	0.28	102	−3.19	**<0.01**		
	Indaziflam* year	1.01	0.35	102	2.93	**0.004**		

[a]Intercept is unsprayed control, 2019. Results are from a linear mixed model regression. P-values in bold indicate statistically significant difference ($P < 0.05$).

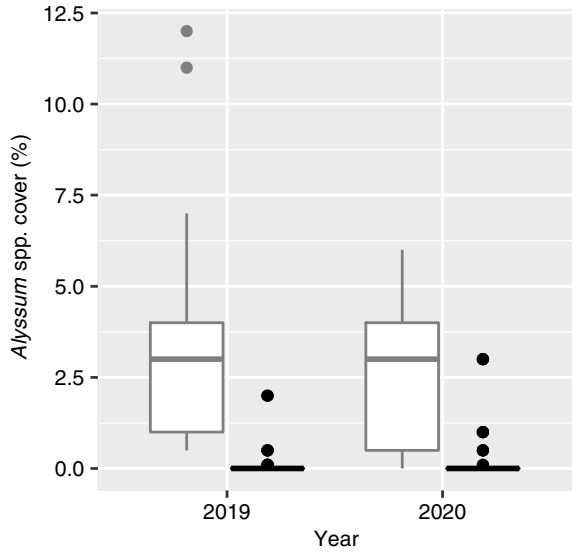


Figure 1. *Alyssum* spp. cover after indaziflam treatment (gray, control; black, sprayed) at sites in Yellowstone National Park along an elevational gradient, in 2019 and 2020. Data shown are from plots with high levels of *Alyssum* spp. invasion.

site, control, and low invasion. A type II ANOVA was used to evaluate treatment effects.

For objective 3, to evaluate impacts to perennial and annual forbs in the soil seedbank, the abundance, richness, and Shannon's diversity were analyzed with linear models similar to those previously described. Abundance and Shannon's diversity responses were fit following a Gaussian distribution, and the richness responses were fit with a quasi-Poisson distribution to account for overdispersion; treatment effects were evaluated with a type II ANOVA.

Results and Discussion

Indaziflam Efficacy on Alyssum spp.

Indaziflam provided excellent control of *Alyssum* spp. 1 and 2 yr after treatment ($P < 0.01$). There was no difference by elevation ($P = 0.47$); however, there was a difference by year ($P < 0.01$) and a significant interaction between year and indaziflam ($P < 0.01$) (Table 2). This is likely explained by natural variation of *Alyssum* spp. abundance in the control plots (Figure 1). The mean (±SE) cover of *Alyssum* spp. in spray plots was 0.1% (±0.04%) in 2019 and 0.2% (±0.1%) in 2020; in control plots, mean cover was 3.7% (±0.43%) in 2019 and 2.5% (±0.32%) in 2020. This reduction in sprayed plots equates to mean *Alyssum* spp. cover of 97% less in 2019 and 91% less in 2020 compared with control plots.

Impacts of Indaziflam on the Whole Plant Community

There were 160 species across all sites: 100 perennial forbs, 32 perennial graminoids, 1 annual graminoid, 16 annual forbs, and 11 shrubs (Supplementary Table S1). There were some co-occurring nonnative species: 4 perennial graminoids, 1 annual graminoid, 7 perennial forbs, and 2 annual forbs (Supplementary Table S1). Mean (±SE) cover of nonnative species across all elevations was 3.9% (±0.86), which included the targeted *Alyssum* spp. populations. Total species richness at each site ranged from 38 to 76 species (Table 1). The mean species richness of the whole plant community was lower in sprayed plots ($P < 0.01$) and high *Alyssum* spp. invasion plots ($P = 0.05$), decreased as elevation increased ($P < 0.01$) but did not differ by year ($P = 0.26$) (Table 3; Figure 2). Sprayed plots also had lower Shannon's diversity than control plots ($P < 0.01$), and diversity decreased as elevation increased ($P < 0.01$) but did not differ between levels of invasion ($P = 0.39$) or by year ($P = 0.18$) (Table 4). Species richness has shown a decrease or hump-shaped response along elevation gradients in mountain habitats globally (Haider et al. 2018; Pauchard et al. 2009).

The significant difference in total species richness between high- and low-invasion treatments was generally fewer than two species and mainly related to the differences in richness between invasion treatments at the two highest elevations (Figure 2). To evaluate the impacts of an invader on richness and diversity, the values for the invader should be removed to avoid artificially inflating the response (Thomsen et al. 2016). This additional analysis was performed and found similar patterns (data not shown). However, when annual forb richness and diversity were evaluated, neither differed between the invasion treatments (Meyer-Morey 2021). These results suggest that *Alyssum* spp. is a weak invader in the mountain sagebrush plant community. However, further evaluation of *Alyssum* spp. impact is necessary to determine whether and which populations to prioritize for management (Rew et al. 2007).

Our findings that richness and diversity decreased with herbicide application is in contrast with other rangeland studies that found increased richness and abundance 1 yr after treatment (Sebastian et al. 2020) and no effects on species richness after 2 yr (Clark et al. 2019) for the whole community. These studies were conducted in areas with dense infestations of downy brome (*Bromus tectorum* L.) (between 30% and 70%) and another four to five nonnative species, and the increased abundance and richness was attributed to release from competition. Our study sites were in relatively undisturbed mountain big sagebrush plant communities that are more resilient to disturbance and resistant to annual species invasion than lower-elevation sagebrush communities (Chambers et al. 2014).

Richness was also higher at our sites than in the study sites of Clark et al. (2019; 33 to 35 native species), and we only observed *B.*

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Table 3. Effects of indaziflam herbicide on species richness of the whole plant community, perennial graminoids, perennial forbs, and annual forbs, controlling for level of *Alyssum* spp. invasion (high, low), elevation, and year (2019, 2020).[a]

Fixed effects						Random effect
Response						Variance
richness	Predictor	Est.	SE	*z*-value	P(>)	tag
Whole community	Intercept	4.55	0.16	28.92	<**0.01**	0.03 ± 0.18
$R^2 = 0.52$	Indaziflam	−0.24	0.04	−6.27	<**0.01**	
	Invasion	-7×10^3	0.04	−1.93	**0.05**	
	Elevation	-9×10^4	7.6×10^5	−11.76	<**0.01**	
	Year2020	-3×10^3	0.03	−1.14	0.26	
Perennial graminoids	Intercept	1.99	0.31	6.51	<**0.01**	0.0 ± 0.0
$R^2 = 0.08$	Indaziflam	2×10^3	0.05	0.05	0.96	
	Invasion	0.96	0.43	2.25	**0.02**	
	Elevation	-3×10^4	1×10^4	−1.76	0.08	
	Year2020	1×10^3	0.05	0.02	0.98	
	Elevation*invasion	-5×10^4	2×10^4	−2.55	**0.01**	
Perennial forbs	Intercept	4.90	0.22	22.17	<**0.01**	0.09 ± 0.29
$R^2 = 0.56$	Indaziflam	−0.15	0.06	−2.59	**0.01**	
	Invasion	−0.13	0.06	−2.39	**0.02**	
	Elevation	-1×10^3	1×10^4	−12.98	<**0.01**	
	Year2020	−0.10	0.04	−2.59	**0.01**	
Annual forbs	Intercept	−3.08	0.70	−4.39	<**0.01**	0.32 ± 0.56
$R^2 = 0.52$	Indaziflam	−1.85	0.15	−12.40	<**0.01**	
	Invasion	0.42	0.14	3.03	<**0.01**	
	Elevation	1×10^3	2×10^4	5.15	<**0.01**	
	Year2020	0.14	0.93	1.46	0.14	

[a]Intercept is unsprayed control, low invasion, 2019. Results are from mixed-effects models with a Poisson distribution. Values in bold indicate statistically significant differences ($P < 0.05$).

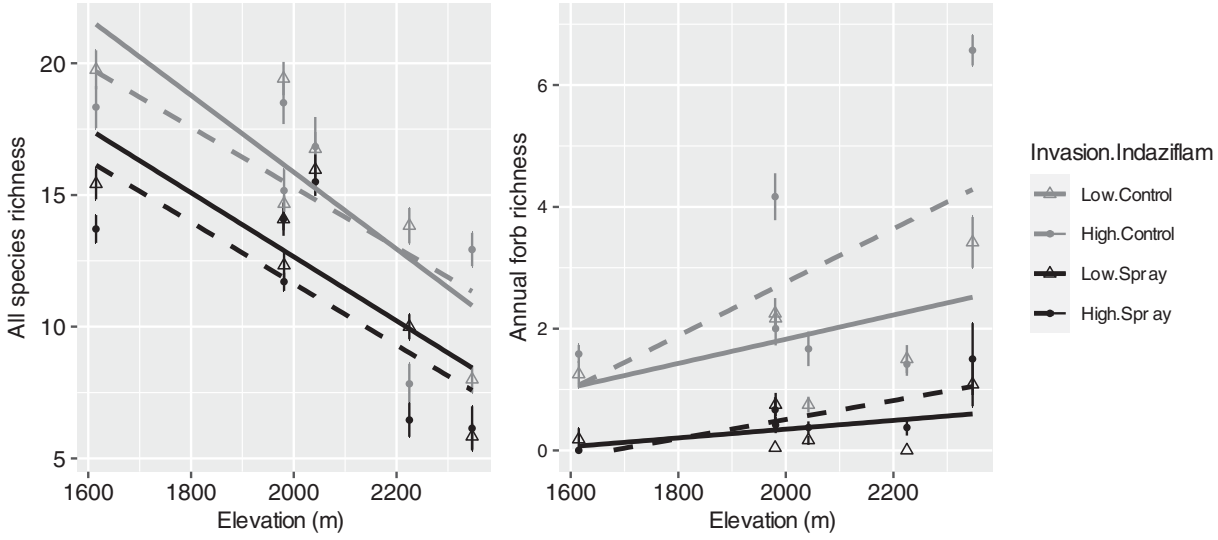


Figure 2. All species and annual forb mean (±SE) richness after indaziflam treatment (gray, control; black, sprayed) in two levels of *Alyssum* spp. invasion (Δ solid, low; • dashed, high) along an elevation gradient in Yellowstone National Park.

tectorum at one site, where its cover was very low (0.5%). To better understand the differences we observed, richness and diversity of perennial graminoids, perennial forbs, and annual forbs were analyzed separately.

Perennial Species Response

Established perennial graminoid richness and Shannon's diversity were largely unaffected by indaziflam ($P = 0.96$ and $P = 0.50$, respectively) (Tables 2 and 3, Supplementary Figure S1), as expected from previous studies (Clark et al. 2019, 2020). Perennial forb mean species richness was lower in sprayed plots ($P < 0.01$) and under high invasion ($P = 0.02$), decreased with elevation ($P < 0.01$), and differed between years ($P = 0.01$; Table 3, Supplementary Figure S1). Shannon's diversity of perennial forbs was lower in sprayed ($P = 0.04$) and high-invasion plots ($P = 0.05$), decreased with elevation ($P < 0.01$), and was lower in 2020 than 2019 ($P = 0.01$). Other studies have shown no injury to or reduction in abundance of existing perennial vegetation (Clark et al. 2019; Sebastian et al. 2017a); therefore, we theorize that reduced recruitment from the seedbank may have been the cause of perennial forb reduction, which our seedbank study results corroborate. Sebastian et al. (2017c) also concluded that indaziflam reduced recruitment of a perennial nonnative forb at their field study site.

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Table 4. Effects of indaziflam (spray, control) on Shannon's diversity of the whole plant community, perennial graminoids, perennial forbs, and annual forbs, controlling for level of *Alyssum* spp. invasion (high, low), elevation, and year (2019, 2020).[a]

Fixed effects							Random effect	
Response							Variance	
Shannon's diversity	Predictor	Est.	SE	df	*z*-value	P(>)	Tag	Residual
Whole community	Intercept	4.24	0.23	204	18.62	<**0.01**	0.11 ± 0.33	0.03 ± 0.16
$R^2 = 0.87$	Indaziflam	−0.18	0.05	204	−3.71	<**0.01**		
	Invasion	−0.04	0.05	203	0.86	0.39		
	Elevation	-1×10^3	1×10^4	203	−9.18	<**0.01**		
	Year	−0.02	0.02	204	−1.35	0.18		
Perennial graminoids	Intercept	3.00	0.24	204	12.67	<**0.01**	0.12 ± 0.34	0.03 ± 0.18
$R^2 = 0.83$	Indaziflam	0.04	0.05	204	0.68	0.50		
	Invasion	-6×10^2	0.05	203	−1.09	0.28		
	Elevation	-9×10^4	1×10^4	203	−8.31	<**0.01**		
	Year	3×10^3	0.02	204	−0.17	0.86		
Perennial forbs	Intercept	3.90	0.32	204	11.51	<**0.01**	0.21 ± 0.46	0.04 ± 0.19
$R^2 = 0.88$	Indaziflam	−0.13	0.07	203	−2.02	**0.04**		
	Invasion	−0.13	0.07	203	−1.94	0.05		
	Elevation	-1×10^3	1×10^4	203	−7.69	<**0.01**		
	Year	−0.07	0.02	204	−3.47	<**0.01**		
Annual forbs	Intercept	−0.43	0.20	204	−2.09	**0.04**	0.07 ± 0.27	0.05 ± 0.23
$R^2 = 0.71$	Indaziflam	−0.43	0.05	204	−9.55	<**0.01**		
	Invasion	0.06	0.04	203	1.37	0.17		
	Elevation	4×10^4	1×10^5	203	4.46	<**0.01**		
	Year	0.04	0.02	204	1.76	0.08		

[a]Intercept is control, low invasion, 2019. Results are from mixed-effects models. Values in bold indicate statistically significant differences ($P < 0.05$).

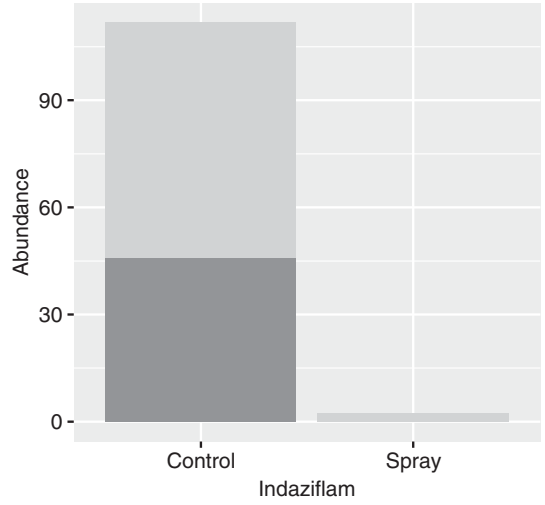


Figure 3. Soil seedbank total abundance by life form (light gray, annual forb; dark gray, perennial forb) after indaziflam treatment (sprayed, control) from all samples (2.26 m^2) collected at two sites (M, B) in Yellowstone National Park.

Annual Forb Response

Annual forbs were greatly impacted by indaziflam. Mean species richness of annual native forbs was lower in sprayed plots ($P < 0.01$) by at least 50% (Figure 2), increased with elevation ($P < 0.01$), and was greater in high-invasion areas ($P < 0.01$) due to the presence of *Alyssum* spp., but did not differ between years ($P = 0.14$) (Table 3; Figure 2). Shannon's diversity of annual forbs was reduced by indaziflam ($P < 0.01$), increased significantly with elevation ($P < 0.01$), and did not differ by year ($P = 0.08$) or level of invasion ($P = 0.17$; Table 4). The increase with elevation was explained by blackfoot groundsmoke (*Gayophytum racemosum* Torr. & A. Gray) and northern linanthus [*Leptosiphon septentrionalis* (H. Mason) J.M. Porter & L.A. Johnson] at the highest elevation. The reduced richness and diversity of annual forbs after herbicide application is in contrast with another study that found no effect of indaziflam on annual forbs (Sebastian et al. 2020) and suggests that individual species of annual forbs may have differing sensitivities to indaziflam. There were 12 species of annual forbs plus *Alyssum* spp. at our sites, six were observed in both control and spray plots: *Alyssum* spp., tiny trumpet (*Collomia linearis* Nutt.), woodland draba (*Draba nemorosa* L.), *L. septentrionalis*, slender phlox [*Microsteris gracilis* (Hook.) Greene], and Douglas' knotweed (*Polygonum douglasii* Greene). The other six were only observed in the control plots: pygmyflower rockjasmine (*Androsace septentrionalis* L.), maiden blue-eyed Mary (*Collinsia parviflora* Lindl.), tall annual willowherb (*Epilobium brachycarpum* C. Presl.), pinyon groundsmoke (*Gayophytum ramosissimum* Torr. & A. Gray), *G. racemosum*, dwarf purple monkeyflower (*Mimulus nanus* Hook. & Arn.), and Suksdorf's monkeyflower (*Mimulus suksdorfii* A. Gray). Because annual species rely on annual regeneration from the seedbank, long-lived residual preemergent herbicides like indaziflam may have long-term impacts on all these species, depending on the species' seed decay rates, which are poorly quantified. We did not evaluate the relative fecundity of the species that emerged in control and sprayed plots, but they did produce seeds in both. Our results suggest the species only observed in the control plots are likely to be most affected, because for these species to remain in a community after herbicide application will require recolonization via seed movement from adjacent unsprayed areas.

Impacts of Indaziflam on Soil Seedbank Recruitment

Indaziflam greatly suppressed germination of both perennial and annual forbs from the soil seedbank (Figure 3). There were 29 forb species that emerged: 18 perennials and 11 annuals (Supplementary Table S1). Mean abundance of perennial forbs in the control soils was 23 (±15) individuals and there were no perennial forbs that emerged in the sprayed soils (Figure 3). This supports the results of our field study, which showed reduced richness and diversity

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Table 5. Effects of indaziflam on the abundance, species richness, and Shannon's diversity of annual forb seedlings emerged from the soil seedbank, controlling for site and level of *Alyssum* spp. invasion.[a]

Response	Predictor	Est	SE	*t*-value	P(>)
(G) Abundance	Intercept	49.29	10.79	4.57	<**0.01**
	Indaziflam	−25.96	10.79	−2.96	<**0.01**
	Site	−6.46	10.79	−0.60	0.55
	Invasion	−25.96	10.79	−2.41	**0.02**
(QP) Richness	Intercept	1.56	0.18	8.88	<**0.01**
	Indaziflam	−2.32	0.43	−5.35	<**0.01**
	Site	−1.00	0.28	−3.59	<**0.01**
	Invasion	−0.58	0.26	−2.26	**0.01**
(G) Shannon's diversity	Intercept	0.81	0.09	9.50	<**0.01**
	Indaziflam	−0.50	0.09	−5.89	<**0.01**
	Site	−0.41	0.09	−4.76	<**0.01**
	Invasion	−0.10	0.09	−1.20	<0.24

[a]Intercept is control (no indaziflam treatment), site M, and low invasion. Results are from linear models fit with Gaussian (G) and quasi-Poisson (QP) distributions. Values in bold indicate statistically significant difference ($P < 0.05$).

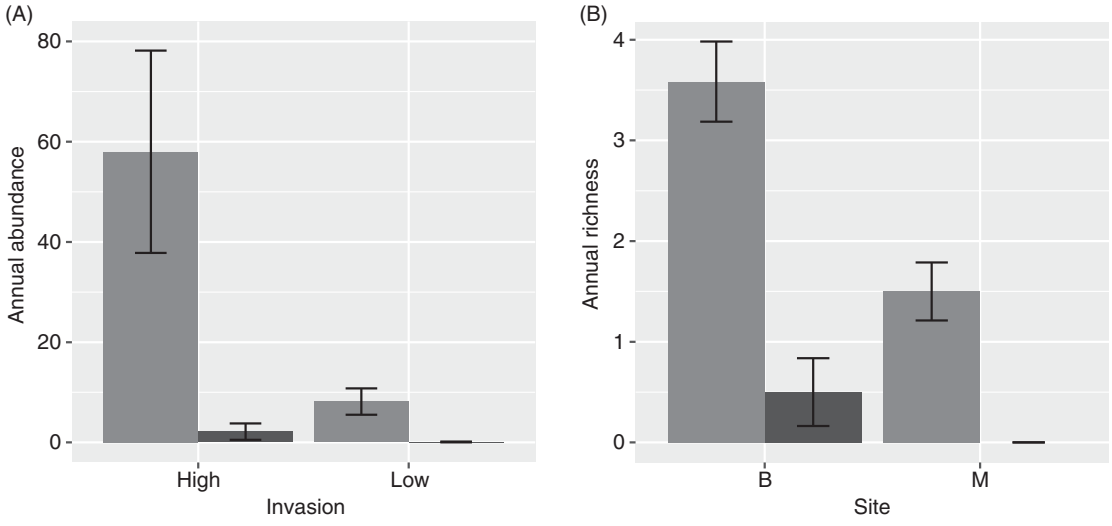


Figure 4. Mean annual forb soil seedbank (A) abundance and (B) richness after indaziflam treatment (gray, control; black, sprayed) from samples (471 cm^2) at two sites (M, B) in Yellowstone National Park and by level of *Alyssum* spp. invasion (low, high).

of perennial forbs in sprayed plots, and the results of Sebastian et al. (2017c), which documented reduced recruitment of seven perennial nonnative forbs in their seedbank study.

The few species that emerged in the sprayed soils were all annual forbs: herb sophia [*Descurainia sophia* (L.) Webb ex Prantl], *C. linearis*, *D. nemorosa*, and *M. gracilis*. These were very low in abundance compared with the control soils; mean annual forb abundance was 1 (±0.83) individual in sprayed soils compared with 33 (±11.3) individuals in the controls (Figure 4A). The latter three species (*C. linearis*, *D. nemorosa*, and *M. gracilis*) were also found in the sprayed plots in the field experiment but also occurred at greater abundance in the control plots. Annual forb richness was lower in the sprayed soils ($P < 0.01$) and differed between the two sites ($P < 0.01$) and levels of invasion ($P = 0.03$). Mean annual richness in the controls was 1.5 (±0.3) species at the lower site (M) and 3.6 (±0.4) species at the higher site (B) and, in sprayed soils, was reduced to 0 (±0) and 0.5 (±0.34), respectively (Figure 4B). Shannon's diversity was also lower in sprayed soils ($P < 0.01$) and differed between sites ($P < 0.01$), though it did not differ between levels of invasion (Table 5). The soil seedbank at our sites had a high diversity of species (29 species) despite *Alyssum* spp. infestation, and these nontarget species, particularly the native annual forbs, were shown to be negatively impacted by indaziflam.

Previous indaziflam studies have primarily focused on the response of perennial forbs and perennial grasses, as they are desirable components of rangeland plant communities; however annual forbs are also key components of rangelands (Pokorny et al. 2004). They occupy an early successional niche and provide critical spring forage for many wildlife species (Drut et al. 1994; Luna et al. 2018). Additionally, native annual forbs are phenologically similar to nonnative winter annual grasses and likely use similar resource pools (Forbis 2010). The annual forbs in our study area are early-season and early seral species; early seral annual forbs can compete with nonnative winter annual grasses, specifically *B. tectorum* (Uselman et al. 2015). Therefore, knowing which annual species are tolerant to indaziflam is useful for creating restoration seed mixes for use after indaziflam application.

In the western United States, approximately 23 million ha of rangelands are infested with nonnative annual species (DiTomaso et al. 2017), presenting a considerable challenge to restore and preserve these working lands. Areas with extensive monocultures of nonnative plants require novel and innovative

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approaches, such as seedbank depletion, to achieve adequate long-term control; however, these same methods may not be appropriate at all levels of infestation or with all nonnative annual species. In areas where the seedbank is composed mostly of nonnative species that have formed persistent soil seedbanks, indaziflam is a valuable tool to deplete the seedbank of target species before active revegetation efforts (Clark et al. 2020). However, in areas with minimal infestations, an existing diverse plant community, and therefore likely a diverse seedbank, the use of indaziflam and its impacts on the seedbank of nontarget species should be carefully considered before broadscale use. Additionally, to determine whether active management is warranted, the actual impacts of an invader should be considered (Rew et al. 2007). Our results suggest *Alyssum* spp. is a weak invader, and using indaziflam to control it may do more harm than the target species itself in high-elevation sagebrush steppe.

Indaziflam significantly reduced species richness and diversity, particularly of nontarget annual forb species, resulting in a community more dominated by perennial species. Native annual forb species are often overlooked, though they occupy an important early successional niche and contribute to the biodiversity of rangelands. Maintaining biodiversity should be a priority for land managers, as communities that are higher in diversity are typically more resistant to ecosystem alteration and more resilient after disturbances (Chambers et al. 2014; Hobbs and Huenneke 1992; Standish et al. 2014). Understanding and minimizing the nontarget effects of invasive plant control on existing intact vegetation is critical when developing management strategies (Rew et al. 2007), especially in arid rangelands. Future studies should address the long-term impacts of indaziflam to plant community composition and explore the tolerances and seedbank longevities of native species for restoration seed mixes.

Supplementary material. To view supplementary material for this article, please visit https://doi.org/10.1017/inp.2021.31

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References

Allen EB, Knight DH (1984) The effects of introduced annuals on secondary succession in sagebrush-grassland, Wyoming. Southwest Nat 29:407–421

Balch JK, Bradley BA, D'Antonio CM, Gómez-Dans J (2013) Introduced annual grass increases regional fire activity across the arid western USA (1980–2009). Global Change Biol 19:173–183

Bartoń K (2019) MuMIn: Multi-Model Inference. R Package v. 1.43.6. https://CRAN.R-project.org/package=MuMIn. Accessed: December 6, 2020

Bates D, Mächler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using lme4. J Stat Softw 67:1–48

Bayer (2020) Rejuvra herbicide receives EPA approval. New herbicide is a long-term, economical solution for the control of invasive annual grasses. Cary, NC: Bayer Environmental Science. https://www.businesswire.com/news/home/20200630005526/en/Rejuvra%E2%84%A2-Herbicide-Receives-EPA-Approval. Accessed: November 3, 2021

Beck JL, Connelly JW, Wambolt CL (2012) Consequences of treating Wyoming big sagebrush to enhance wildlife habitats. Rangeland Ecol Manag 65: 444–455

Billings WD (1994) Ecological impacts of cheatgrass and resultant fire on ecosystems in the western Great Basin. Pages 22–30 *in* Monsen SB and Kitchen SG, eds. Proceedings—Ecology and Management of Annual Rangelands. Ogden, UT: USDA Forest Service, Intermountain Research Station, General Technical Report INT-GTR-313

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

Chambers JC, Bradley BA, Brown CS, D'Antonio C, Germino MJ, Grace JB, Hardegree SP, Miller RF, Pyke DA (2014) Resilience to stress and disturbance, and resistance to *Bromus tectorum* L. invasion in cold desert shrublands of western North America. Ecosystems 17:360–375

Clark SL, Sebastian DJ, Nissen SJ, Sebastian JR (2019) Effect of indaziflam on native species in natural areas and rangeland. Invasive Plant Sci Manag 12:60–67

Clark SL, Sebastian DJ, Nissen SJ, Sebastian JR (2020) Evaluating winter annual grass control and native species establishment following applications of indaziflam on rangeland. Invasive Plant Sci Manag 13:199–209

Crone EE, Marler M, Pearson DE (2009) Non-target effects of broadleaf herbicide on a native perennial forb: a demographic framework for assessing and minimizing impacts. J Appl Ecol 46:673–682

DiTomaso JM (2000) Invasive weeds in rangelands: species, impacts, and management. Weed Sci 48:255–265

DiTomaso JM, Monaco TA, James JJ, Firn J (2017) Invasive plant species and novel rangeland systems. Pages 429–465 *in* Briske D, ed. Rangeland Systems. Cham, Switzerland: Springer

Drut MS, Pyle WH, Crawford JA (1994) Technical note: diet and food selection of sage grouse chicks in Oregon. J Range Manag 47:90–93

Elton CS (1958) The Ecology of Invasions by Animals and Plants. Boston, MA: Springer. 196 p

Forbis TA (2010) Germination phenology of some Great Basin native annual forb species. Plant Spec Biol 25:221–230

Haider S, Kueffer C, Bruelheide H, Seipel T, Alexander JM, Rew LJ, Arévalo JR, Cavieres LA, McDougall KL, Milbau A, Naylor BJ, Speziale K, Pauchard A (2018) Mountain roads and non-native species modify elevational patterns of plant diversity. Global Ecol Biogeogr 27:667–678

Hobbs RJ, Huenneke LF (1992) Disturbance, diversity, and invasion: implications for conservation. Conserv Biol 6:324–337

Kettenring KM, Adams CR (2011) Lessons learned from invasive plant control experiments: a systematic review and meta-analysis. J Appl Ecol 48:970–979

Kindt R, Coe R (2005) Tree Diversity Analysis. A Manual and Software for Common Statistical Methods for Ecological and Biodiversity Studies. Nairobi: World Agroforestry Centre (ICRAF). 196 p

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

Kuznetsova A, Brockhoff PB, Christensen RHB (2017) Tests in linear mixed effects models. J Stat Softw 82(13):1–26

Luna T, Mousseaux MR, Dumroese RK (2018) Common Native Forbs of the Northern Great Basin Important for Greater Sage-Grouse. General Technical Report RMRS-GTR-387. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station; Portland, OR: U.S. Department of the Interior, Bureau of Land Management, Oregon–Washington Region. 76 p

Lund HG (2007) Accounting for the world's rangelands. Rangelands 29:3–10

Mack MC, D'Antonio CM (1998) Impacts of biological invasions on disturbance regimes. Trends Ecol Evol 13:195–198

Meyer-Morey J (2021) Impacts of a Non-native Forb, *Alyssum desertorum* Stapf., and Non-target Effects of Indaziflam in the Sagebrush Steppe of Yellowstone National Park. MS thesis. Bozeman, MT: Montana State University. 121 p

Mosley J (2014) Featured weed: yellow and desert alyssum. Big Sky Small Acres Spring/Summer:4–5

Noack R, Pokorny M, Jacobs J (2020) Plant guide for desert madwort (*Alyssum desertorum*). USDA Natural Resources, Bridger Plant Materials Center. https://www.blogs.nrcs.usda.gov/Internet/FSE_PLANTMATERIALS/publications/mtpmcpg13571.pdf. Accessed: December 6, 2020

Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, McGlinn D, Minchin PR, O'Hara RB, Simpson GL, Solymos P, Stevens MHH, Szoecs E, Wagner H (2019) Vegan: Community Ecology Package. R Package v. 2.5-6. https://CRAN.R-project.org/package=vegan. Accessed: December 6, 2020

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O'Mara FP (2012) The role of grasslands in food security and climate change. Ann Bot 110:1263–1270

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

Pauchard A, Kueffer C, Dietz H, Daehler CC, Alexander J, Edwards PJ, Arévalo JR, Cavieres LA, Guisan A, Haider S, Jakobs G, McDougall K, Millar CI, Naylor BJ, Parks CG, et al. (2009) Ain't no mountain high enough: plant invasions reaching new elevations. Front Ecol Environ 7:479–486

Pokorny ML, Sheley RL, Svejear TJ, Engel RE (2004) Plant species diversity in a grassland plant community: evidence for forbs as a critical management consideration. Western N Am Naturalist 64:219–230

Rew LJ, Lehnhoff EA, Maxwell BD (2007) Non-indigenous species management using a population prioritization framework. Can J Plant Sci 87:1029–1036

Rinella MJ, Maxwell BD, Fay PK, Weaver T, Sheley RL (2009) Control effort exacerbates invasive-species problem. Ecol Appl 19:155–162

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, Sebastian JR, Meiman PJ, Beck KG (2017c) Preemergence control of nine invasive weeds with aminocyclopyrachlor, aminopyralid, and indaziflam. Invasive Plant Sci Manag 10:99–109

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

Sebastian J, Swanson J, Sauer S, Clark S, Sebastian D (2020) Pollinator community and floral resource response to cheatgrass control with Esplanade 200 SC (indaziflam). *In* Proceedings of the Weed Science Society of America Western Society of Weed Science Joint Meeting. Maui, HI: Weed Science Society of America

Skurski TC, Maxwell BD, Rew LJ (2013) Ecological tradeoffs in non-native plant management. Bio Conserv 159:292–302

Standish RJ, Hobbs RJ, Mayfield MM, Bestelmeyer BT, Suding KN, Battaglia LL, Eviner V, Hawkes CV, Temperton VM, Cramer VA, Harris JA, Funk JL, Thomas PA (2014) Resilience in ecology: Abstraction, distraction, or where the action is? Biol Conserv 177:43–51

Thomsen MS, Wernberg T, South PM, Schiel DR (2016) To include or not to include (the invader in community analyses)? That is the question. Biol Invasions 18:1515–1521

Tilman D (1999) The ecological consequences of changes in biodiversity: a search for general principles. Ecology 80:1455–1474

Uselman SM, Snyder KA, Leger EA, Duke SE (2015) Emergence and early survival of early versus late seral species in Great Basin restoration in two different soil types. Appl Veg Sci 18:624–636

Vasquez EA, James JJ, Monaco TA, Cummings DC (2010) Invasive plants on rangelands: a global threat. Rangelands 32:3–5

Vilà M, Espinar JL, Hejda M, Hulme PE, Jarošík V, Maron JL, Pergl J, Schaffner U, Sun Y, Pyšek P (2011) Ecological impacts of invasive alien plants: a meta-analysis of their effects on species, communities and ecosystems. Ecol Lett 14:702–708

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

Young JA, Fay LA (1997) Cheatgrass and range science: 1930–1950. J Range Manag 50:530–535

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