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Imazapic Activity in a Semiarid Climate in Downy Brome (*Bromus tectorum*)–Infested Rangeland and CRP Sites

Krista A. Ehlert, Richard E. Engel, and Jane M. Mangold*

Chemical control of downy brome has focused on imazapic; however, imazapic efficacy in semiarid climates is unpredictable, possibly because of variable residual soil activity. Our objective was to characterize imazapic activity over 9 mo in rangeland and a Conservation Reserve Program (CRP) site following its application in the fall as affected by rate (0, 80, 160, 240 g ai ha^{-1}) and quantity of plant residue (reduced, ambient). Greenhouse bioassays were conducted over two seasons (2010 to 2011 and 2011 to 2012) using soil collected at multiple dates after imazapic application. Quantity of plant residue did not affect downy brome biomass or response to imazapic. Imazapic reduced downy brome biomass ($P < 0.05$) across all sampling dates in both seasons, and the response to rates was consistent up to 200 d post application. Imazapic activity over time conformed to a biphasic model with activity being consistent, or slightly improving, up to about 160 and 150 d post application, and then dropping rapidly to the final sampling event 287 and 272 d post application in rangeland and at CRP sites, respectively. These results indicate that fall imazapic applications in semiarid climates persist into the spring, thus providing control of both fall-emerging downy brome seedlings and seeds that overwinter and emerge the following spring.

Nomenclature: Imazapic; downy brome, *Bromus tectorum* L. BROTE.

Key words: Bioassay, chemical weed control, plant residue, Plateau®, soil residual activity.

El control químico de *Bromus tectorum* se ha enfocado en imazapic. Sin embargo, la eficacia de imazapic en climas semiáridos es impredecible, posiblemente por su variable actividad residual en el suelo. Nuestro objetivo fue caracterizar la actividad de imazapic a lo largo de 9 meses después de su aplicación en el otoño, la influencia de sitios bajo el Programa de Reservas para Conservación (CRP), el efecto de la dosis (0, 80, 160, 240 g ai ha^{-1}) y la cantidad de residuos vegetales (ambiental, reducida). Se realizaron bioensayos de invernadero durante dos temporadas (2010 a 2011 y 2011 a 2012) usando suelo colectado en múltiples fechas después de la aplicación de imazapic. La cantidad de residuo vegetal no afectó la biomasa de *B. tectorum* o la respuesta a imazapic. Imazapic redujo la biomasa de *B. tectorum* ($P < 0.05$) en todas las fechas de muestreo en ambas temporadas, y la respuesta a las dosis fue consistente hasta 200 d después de la aplicación. La actividad de imazapic a lo largo del tiempo se ajustó a un modelo bifásico teniendo una actividad consistente o ligeramente mejorando, hasta cerca de 160 y 150 d después de la aplicación, y luego cayendo rápidamente en el evento final de muestreo a 287 y 272 d después de la aplicación en pastizales y en sitios CRP, respectivamente. Estos resultados indican que las aplicaciones de imazapic en el otoño en climas semiáridos persisten hasta la primavera, bridando así control de plántulas que emergen en el otoño de *B. tectorum* y semillas que sobreviven el invierno y emergen durante la siguiente primavera.

The invasion of downy brome is considered one of the most significant plant invasions in North America, with 22.3 million ha infested in the western United States (Rice 2005). Downy brome has been problematic in the Great Basin and more recently in Montana, where its expansion may be enhanced in the future by global climate change (Bradley 2009). Chemical control of downy brome on rangelands in the West has focused primarily on imazapic, an acetolacetate synthase inhibitor and member of the imidazolinone herbicide family (Davison and Smith 2007; Elseroad and Rudd 2011; Morris et al. 2009). Efficacy of imazapic applications in Montana has been inconsistent (Mangold et al. 2013), which encouraged us to investigate imazapic activity in Montana's semiarid climate. Imazapic is typically applied pre- or post-emergent in the fall to control fall-emerging downy brome seedlings. However, land managers often question whether or not imazapic persists into the following spring, because it could potentially

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control downy brome seeds that do not germinate in the fall, but instead emerge the spring after application.

Imidazolinone persistence has not been studied extensively in rangeland and Conservation Reserve Program (CRP) ecosystems of the semiarid West. Most studies on imidazolinone persistence have been conducted in cropland field sites. Irrigated cropping system studies in southern Alberta, Canada, found that imidazolinone (imazethapyr and imazamethabenz) persistence reduced yield of rotational crops seeded 1 yr post application (Moyer and Esau 1996). Dryland cropping system studies in the Pacific Northwest found that imazamox carryover was affected by rainfall (Ball et al. 2003). Little herbicide injury or carryover to a succeeding crop was observed at field sites in high-rainfall locations, whereas herbicide carryover was of particular concern at sites with low rainfall and low soil pH (pH 5.6 to 6.2). Soil pH is known to affect sorption and bioavailability of polar, ionizable chemicals, such as imidazolinones. Sorption of imidazolinones is greater at low pH, resulting in decreased availability for microbial degradation (Aichele and Penner 2005; Bresnahan et al. 2000). Similarly, imidazolinone persistence has been reported to be greater in soil with high clay and organic matter content (Colquhoun 2006; Ulbrich et al. 2005) because of increased sorption.

Soil bioassays are an alternative and affordable method to assess herbicide persistence and degradation relative to costly analytical methods (Cobucci et al. 1998; Ranft et al. 2010; Streibig 1988). Bioassays have been conducted in cropping systems to determine imazapic persistence as it relates to carryover effects and cropping interval recommendations (Onofri 1996; Ulbrich et al. 2005); however, few studies have investigated imazapic persistence as it relates to weed control. We present results from a bioassay conducted with soil samples collected from a field experiment that investigated the effect of imazapic rate and plant residue on downy brome control. Experiments were conducted for two seasons in rangeland and at a CRP site, which were characterized by a dense layer of dried plant residue at the soil surface. Our objective was to characterize imazapic activity over a 6-mo period (late September to early April) in season 1 and a 9-mo period (late September to late June/early July) in season 2 as affected by application rate and plant residue presence.

Materials and Methods

Site and Field Experiment Description. Soil for the bioassay was collected from a field experiment conducted at two locations during 2010 to 2011 and 2011 to 2012, hereafter referred to as 2011 and 2012, respectively. The field sites included a rangeland site 35 km south of Big Timber, MT (45.60°N, 110.17°W) and a CRP site 23 km south of Havre, MT (48.45°N, 109.88°W) (hereafter referred to as the rangeland and CRP sites, respectively). Soil at the rangeland site is a Winspect cobbly loam (Typic Calciustoll) with pH 6.6 (0 to 10 cm depth) and 36.8 g kg^{-1} organic carbon (C). Mean annual precipitation and air temperature are 387 mm and 7.2 C, respectively. The plant community consisted of perennial grasses (e.g., *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass) and *Pseudoroegneria spicata* (Pursh) Á. Löve (bluebunch wheatgrass)), exotic perennial forbs (e.g., *Taraxacum officinale* F.H. Wigg. (common dandelion) and *Tragopogon dubius* Scop. (yellow salsify)), and native perennial forbs (e.g., *Artemesia fridgida* Willd. (fringed sage)). Soil at the CRP site is an Evanston loam (Aridic Agriustolls) with pH 7.1 (0 to 10 cm depth) and 17.9 g kg^{-1} organic C. Mean annual precipitation and air temperature are 295 mm and 5 C, respectively. The plant community was dominated by seeded native and perennial grasses and to a lesser extent exotic perennial forbs, most of which were also found at the rangeland site. Both sites were infested with downy brome.

The field experiment at each site consisted of a factorial combination of four imazapic application rates (Plateau® WSC, BASF; 0 (control), 80, 160, and 240 g ai ha^{-1}) and two plant residue treatments (reduced, ambient). The experiment was arranged in a randomized split block design with imazapic rate as the whole plot (18.3 by 3.0 m) and residue treatment as the subplot (9.1 by 3.0 m), with four replications. The reduced residue treatment was achieved by hand raking away from the shared edge of the subplot with a lawn rake immediately before imazapic application. The ambient residue treatment was undisturbed. We collected and weighed the raked residue for each trial at both sites, then

Table 1. Application date and corresponding weather conditions for 2011 and 2012 at the rangeland and Conservation Reserve Program (CRP) sites.

Date	Weather conditions	Date	Weather conditions
2011 Rangeland		2012 Rangeland	
September 29, 2010	1.0 km h^{-1} wind 10 C 63% relative humidity	September 20, 2011	8.0 km h^{-1} wind 8 C 62% relative humidity
2011 CRP		2012 CRP	
September 23, 2010	1.0 km h^{-1} wind 17 C 51% relative humidity	September 28, 2011	6.0 km h^{-1} wind 18 C 23% relative humidity

dried it at 50 C for 72 h. Residue removed at the CRP site was equivalent to 3.8 ± 0.5 and 9.8 ± 0.7 g m^{-2} for the 2011 and 2012 trials, respectively. Residue removed at the rangeland site for the 2011 trial was not measured; however, 3.8 ± 0.3 g m^{-2} was removed for the 2012 trial. After implementing plant residue treatments, imazapic was mixed with water plus a non-ionic surfactant (0.10% v/v, Penetrator®, Helena Chemical Company) and applied using a CO_2 backpack sprayer delivering 157 L ha^{-1} water at 294 kPa across a boom width of 3 m. Date of application and corresponding weather conditions are summarized in Table 1.

Bioassay Experiment Description. Soil for the bioassay was collected from all experimental units, including the nontreated control at the rangeland and CRP sites. Samples were collected on five and six dates at the CRP site in 2011 and 2012, and four and six dates at the rangeland site in 2011 and 2012, respectively (Table 2). At each sampling date, three soil cores were collected at three random locations within each plot (nine cores per plot) using a 7-cm-diam tulip bulb planter. Soil core depth was 10 cm at the CRP site but only 8 cm at the rangeland site because of rock fragments. All plant residue at the soil surface was retained with the soil samples. The nine soil cores collected from each plot were composited and placed in 2-L Ziploc© freezer bags and frozen (0 C) within 4 h of sampling. Soil samples remained frozen until all sampling dates were completed. Soils were then processed for the bioassay by drying in an oven at 50 C for 72 h followed by sieving to remove coarse rock fragments (> 2 mm). Soils were oven-dried at a modest temperature instead of air-dried, because imazapic volatility was not a concern (Tu et al. 2001).

Table 2. Soil sampling date[a] and corresponding days after treatment (DAT) at the rangeland and Conservation Reserve Program (CRP) sites for 2011 and 2012.

	Rangeland		CRP	
Trial	Date	DAT	Date	DAT
2011	Sept 29, 2010	0	Sept 24, 2010	0
	Oct 13, 2010	14	Oct 8, 2010	14
	Oct 29, 2010	30	Oct 22, 2010	28
	N/A	—	Nov 19, 2011	56
	Apr 10, 2011	193	Apr 14, 2011	202
	N/A	—	N/A	—
2012	Sept 20, 2011	0	Sept 29, 2011	0
	Oct 5, 2011	15	Oct 12, 2011	13
	Oct 18, 2011	28	Oct 25, 2011	26
	Nov 17, 2011	58	Nov 28, 2011	60
	Mar 23,2012	185	Mar 29, 2012	182
	July 3, 2012	287	June 27, 2012	272

[a] Abbreviations: Sept, September; Oct, October; N/A, no soil sampling occurred; Nov, November; Apr, April; Mar, March.

We conducted the soil bioassays in the Montana State University Plant Growth Center using 1.3-L pots filled with approximately 500 g of soil. Five downy brome seeds were planted in each pot, and a toothpick was placed immediately adjacent to the seed. Greenhouse light and temperature conditions were maintained at 21.1/12.8 C day/night. Supplemental light was applied as needed to achieve 12 h d^{-1}. Pots were moved by block every 5 to 7 d to minimize the effect of location in the greenhouse. Pots were watered daily throughout both trials to avoid drought stress (i.e., visual evidence of wilting). In 2011, downy brome seedlings that emerged from the seedbank (volunteer seedlings) were not removed; however, in 2012, volunteer seedlings were removed approximately 14 d after planting to avoid density-dependent growth, which would confound biomass measurements. Volunteer seedling density removed per pot ranged from 5 to 175. At 28 d for both trials, total biomass (roots and shoots) was harvested and weighed. Roots were separated from the soil and gently triple-washed with water. Total biomass was determined by whole plant dry weight (50 C for 72 h) expressed as milligrams per plant (mg $plant^{-1}$).

Statistical Analysis. Two separate analyses were performed to determine whether downy brome biomass was affected by imazapic rates and residue and to describe the trend of imazapic activity over time. To determine the effects of imazapic rates and residue, an analysis of variance for individual sampling events (two sites by two seasons) was conducted using the mixed model procedure, or Proc Mixed, in SAS 9.3 (SAS Institute). A preliminary analysis of the data using Levene's test for homogeneity of variances found the variances between imazapic rates were not equal. Thus, a model with unequal variances was specified in Proc Mixed by using the REPEATED statement with the GROUP = option; GROUP = rate was used to specify a different residual variance for each imazapic rate (Littell et al. 2006). Imazapic application rate (control, 80, 160, and 240 g ha^{-1}), plant residue (reduced, ambient), block, and the interaction of rate and residue were included as independent effects. The PDIFF option in the LSMEANS statement was used to separate means when independent effects were found to be significant ($\alpha = 0.05$).

In preparation for the analysis of imazapic activity over time, downy brome biomass was normalized at all sampling events (two sites by two seasons) relative to the nonsprayed control (BM_0). Imazapic-reduced biomass (iBM) was calculated by expressing biomass of the 80, 160, and 240 g ha^{-1} rates as a percentage of BM_0:

$$iBM(\%) = \left[1 - (BM_{80,160,240}/BM_0)\right] \times 100$$

where $BM_{80,160,240}$ refers to the biomass of sprayed plots. To describe imazapic activity over time, scatter diagrams of iBM vs. days after treatment (DAT) were developed, and data were fitted to a piecewise linear regression model in SAS 9.3 using the protocol described by Mendenhall and Sincich (2011).

Results and Discussion

Imazapic Rate and Residue Effects. At the rangeland site, downy brome biomass was reduced by imazapic at all sampling dates in 2011 and 2012 ($P < 0.05$). Residue and the interaction of residue and rate did not affect downy brome biomass at any sampling date. Mean absolute downy brome biomass for each sampling date, averaged across the two residue treatments, is provided in Table 3. Differences among imazapic rates (80, 160, and 240 g ha^{-1}) were small or insignificant the first three and four sampling dates in 2011 and 2012, respectively. These periods correspond to sampling dates that occurred during the fall immediately after application. Soil moisture conditions were dry through much of the fall (September to November) when precipitation was 111 and 41 mm in 2011 and 2012, respectively. In contrast, precipitation totaled 336 and 101 mm in the spring (March to May) of 2011 and 2012, respectively. The drier conditions experienced in 2012 likely resulted in the lower downy brome cover we observed in the field at all imazapic rates, except the control, compared with 2011 (rate by trial interaction, $P = 0.0001$). Significant differences among imazapic rates appeared at the last sampling date in 2011 and 2012 (Table 3). Differences in downy brome biomass in the field in response to imazapic rates in the field were only apparent in 2012, when 80, 160, and 240 g ai ha^{-1} similarly reduced downy brome cover (3 $\pm$ 1, 2 $\pm$ 1, and 0%, respectively) relative to the control (18 $\pm$ 3%).

Table 3. Mean absolute downy brome biomass as affected by imazapic rate for each sampling date (days after treatment) in 2011 and 2012 at the rangeland and Conservation Reserve Program (CRP) sites.

	Rangeland downy brome biomass[a]										
	Days after treatment										
	2011					2012					
Rate g ai ha^{-1}	0	14	30	N/A[b]	193	0	15	28	58	185	287
	mg plant^{-1}										
0	30 c	33 b	43 c	—	92 c	35 b	21 b	135 c	155 c	188 b	33 c
80	6 b	8 a	9 b	—	25 b	3 a	4 a	8 b	11 b	46 a	28 b
160	5 ab	7 a	5 a	—	24 b	3 a	2 a	6 ab	5 a	16 a	27 a
240	4 a	4 a	4 a	—	11 a	3 a	3 a	3 a	4 a	11 a	25 a
	CRP downy brome biomass[a]										
	Days after treatment										
	2011					2012					
Rate g ai ha^{-1}	0	14	28	56	202	0	13	26	60	182	272
	mg plant^{-1}										
0	22 c	23 b	13 a	17 b	89 b	39 b	94 c	55 b	71 c	123 b	42 b
80	9 b	9 a	9 a	10 b	59 b	12 a	19 b	18 a	12 b	34 a	33 b
160	5 ab	7 a	9 a	6 ab	24 a	6 a	6 a	5 a	6 ab	8 a	17 a
240	3 a	5 a	6 a	5 a	12 a	4 a	5 a	4 a	4 a	8 a	17 a

[a] Within a column, means followed by the same letter are not significantly different at $\alpha = 0.05$.

[b] N/A, no soil sampling occurred.

At the CRP site, downy brome biomass was significantly ($P < 0.05$) affected by imazapic rate for all sampling dates in 2011 and 2012, with the exception of 28 DAT in 2011. Residue and the interaction of residue and rate generally did not affect downy brome biomass. Mean absolute downy brome biomass for each sampling date averaged across the two residue treatments is provided in Table 3. Significant differences among imazapic rates appeared at the last sampling date in 2011 and 2012. Specifically, 160 and 240 g ha^{-1} similarly reduced downy brome biomass. Similar to the rangeland site, these spring and early summer sampling dates provide evidence of decreased imazapic persistence. Moreover, they correspond with moist spring (March to May) soil conditions of 132 mm precipitation in 2011 and 159 mm precipitation in 2012, relative to dry soil moisture conditions the previous fall (September to November) when there was 79 and 24 mm of precipitation in 2011 and 2012, respectively. At the CRP site, there was no interaction in the field between imazapic rate and trial; however, there was a main effect of imazapic rate ($P < 0.0001$) on downy brome cover, with slight differences among 80, 160, and 240 g ai ha^{-1} (3 ± 1, 1 ± 0.2, and 0%, respectively) relative to the control (20 ± 4%).

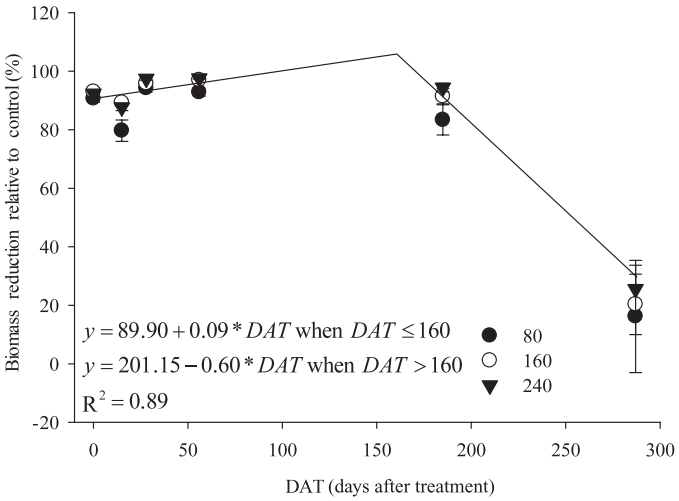

Figure 1. Imazapic-reduced downy brome biomass (percent reduced relative to the control) in imazapic-treated soil (80, 160, and 240 g ai ha^{-1}) vs. days after treatment (DAT) for the rangeland site in 2012. Error bars indicate standard errors of mean.

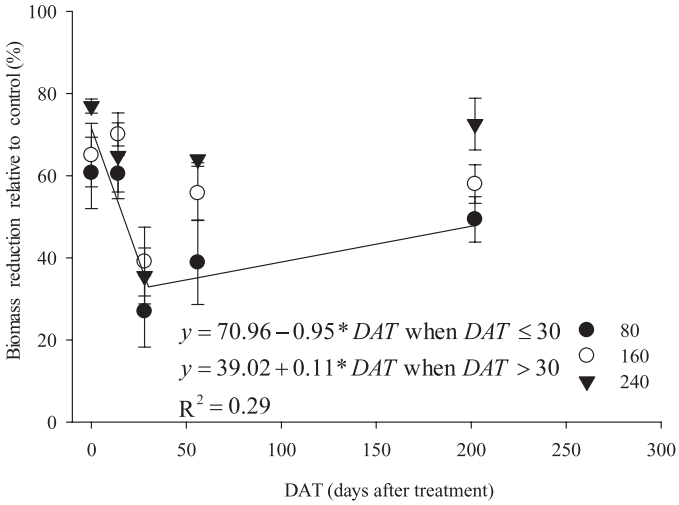

Figure 2. Imazapic-reduced downy brome biomass (percent reduced relative to the control) in imazapic-treated soil (80, 160, and 240 g ai ha^{-1}) vs. days after treatment (DAT) for the Conservation Reserve Program (CRP) site in 2011. Error bars indicate standard errors of mean.

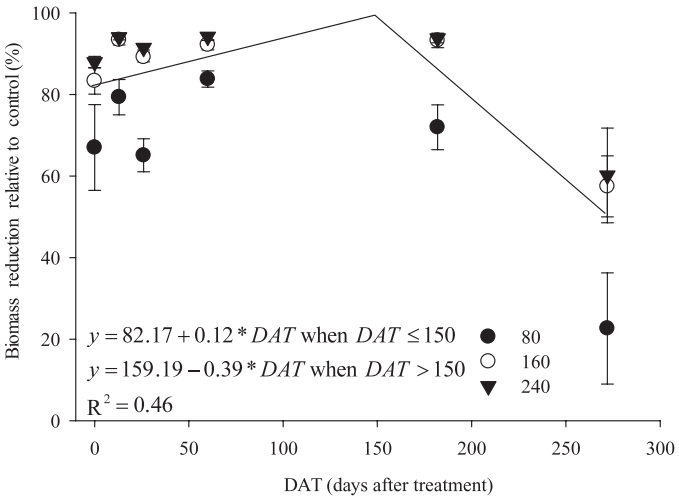

Figure 3. Imazapic-reduced downy brome biomass (percent reduced relative to the control) in imazapic-treated soil (80, 160, and 240 g ai ha^{-1}) vs. days after treatment (DAT) for the Conservation Reserve Program (CRP) site in 2012. Error bars indicate standard errors of mean.

Imazapic Activity over Time. At the rangeland site the relationship between downy brome biomass and DAT was well described by a biphasic model, but only for the 2012 trial. Persistence during the fall months into the following spring was evident in 2012 when the relationship between downy brome biomass and DAT was relatively linear from 0 to 160 DAT (Figure 1). Around 160 DAT, the relationship became biphasic as imazapic persistence decreased, and there was less of an effect on downy brome biomass through the last sampling date at 287 DAT. A biphasic model can also describe the relationship between downy brome biomass and DAT for the CRP site; however, the relationship varies for each trial. In 2011, downy brome biomass decreased rapidly as DAT increased until approximately 30 DAT, after which it slightly increased (Figure 2). In contrast, the relationship between downy brome biomass and DAT was relatively stable in 2012 during the fall and early spring sampling dates until approximately 150 DAT (Figure 3). Imazapic was less persistent after 150 DAT, as evidenced by the decreased effect of imazapic on downy brome biomass.

Results from this study support the idea that imazapic degradation occurs relatively slowly in Montana's semiarid climate after application in the fall, relative to its average half-life of 120 d (Tu et al. 2001). Consequently, fall applications of imazapic should be able to inhibit growth or kill downy brome into the following spring, particularly at rates $\geq$ 160 g ha^{-1}. This was generally supported by our field data (collected 9 mo after imazapic application), which demonstrated downy brome control with all imazapic rates relative to the control. Degradation of imidazolinone herbicide in soil occurs primarily because of microbial activity and as such is limited by low moisture and temperatures (Mangels 1991; Prostko et al. 2005). These conditions are consistent with Montana's climate, which is dry, cold, or both over much of the fall and winter months (October to March). For example, long-term weather records indicate that only 31% and 25% of the annual precipitation occurs over this 6-mo period at our rangeland and CRP sites, respectively. Furthermore, climatic data near our CRP and rangeland sites indicate that for both sites mean air temperatures fall to $\leq$ 0 C beginning early to mid-November and do not rise above freezing until mid-March.

Although most rangeland studies do not directly measure or follow imazapic concentrations in the soil over time, they do suggest that imazapic activity is sufficient to provide downy brome or annual grass control for greater than 1 yr in semiarid climates. Davison and Smith (2007) found that imazapic applied at 105 g ha^{-1} eliminated or significantly reduced growth of downy brome for two growing seasons in Nevada. Elseroad and Rudd (2011) stated

that imazapic (70 g ha^{-1}) could provide downy brome control for 3 or 4 yr after application in north-central Oregon. However, other studies from semiarid climates resulted in only 1 yr of control over downy brome and other invasive annual grasses. For example, Owen et al. (2011) reported that imazapic applied at 132 g ha^{-1} in the late fall reduced downy brome cover and biomass during the first season after application but had no effect during the second season. Similarly, Sheley et al. (2007) found that medusahead [*Taeniatherum caput-medusae* (L.) Nevski] in eastern Oregon was effectively controlled by imazapic ($\geq$ 140 g ha^{-1} rate) the first year after application, but control was reduced greatly during the second year (unburned treatments only). In contrast to studies conducted in semiarid regions, a cropland study in Georgia revealed that imazapic applied at 70 g ha^{-1} had degraded to such a degree that it no longer affected oat yields 4 mo post application (Prostko et al. 2005).

Imazapic efficacy in rangelands has been shown to be reduced in the presence of plant residue, or thatch, on the soil surface. This response has been attributed to adsorption of imazapic to the dried plant material, thereby making it unavailable for plant uptake (Kyser et al. 2007). Our investigation generally found no effect of the residue treatment on downy brome biomass. This may reflect the comparatively small effect that hand-raking had on residue quantity in the field compared with other management techniques like prescribed burning or tillage. In other studies, prescribed burning and tillage followed by imazapic application led to increased annual grass control compared with imazapic application alone (Kyser et al. 2007; Monaco et al. 2005; Sheley et al. 2007).

We recognize the limitations of a greenhouse soil bioassay and thus have recommendations for future research. Although bioassays act as a direct measure of plant available herbicide (Eberle and Gerber 1976), they are unable to provide a direct measure of soil herbicide concentration, unlike an analytical analysis. Moreover, in our study, the lack of equal variances between imazapic rates may have been associated with our perception that fertilization was unnecessary. Thus, it may be advisable to add soil nutrient amendments to future executions of greenhouse soil bioassays, in that doing so may alleviate density-induced reductions in plant biomass. These recommendations will build upon our research and contribute to the knowledge of imazapic activity in semiarid systems.

Downy brome control with imazapic is somewhat inconsistent (Mangold et al. 2013); based on our findings, we conclude that this inconsistency cannot be explained by its degradation rate in the soil. Both our field observations and results from the greenhouse bioassay indicate that when imazapic is applied PRE or POST in the fall, it persists to provide control of downy brome seeds that germinate and emerge the following spring. This is critical for control of downy brome, given its habit of prolonged germination and emergence period from the fall of one year through the spring of the next year (Young and Evans 1975).

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