

Boulder County Parks & Open Space
2021 Small Grant Program Report

Incorporation of indaziflam (Rejuvra®) into Boulder County Parks and Open Space
Weed Management: A Post-Fire Assessment

Final Report
Submitted by Denver Botanic Gardens

Prepared by Christina Alba and Michelle DePrenger-Levin

Contact: Christina Alba, PhD
Christina.alba@botanicgardens.org
720-865-3561

CONTENTS

CONTENTS	2
REPORT OVERVIEW	3
METHODS	4
Study design	4
Vegetation sampling	4
Functional Groups and Coefficients of Conservatism	5
Statistical analysis	6
RESULTS	7
Spray efficacy	7
Total species richness	9
Species richness of short-lived forbs	11
Proportional richness by functional groups	13
Coefficients of Conservatism	14
Total percent cover	15
Percent cover of short-lived forbs	16
Relativized percent cover by functional groups	17
DISCUSSION	18
Spray efficacy	18
Species richness	19
Species cover	20
Long-lived forbs and grasses	20
CONCLUSIONS	21
REFERENCES CITED	22

REPORT OVERVIEW

Land managers are increasingly tasked with understanding interacting global change phenomena. Biological invasions and altered fire regimes are two aspects of global change that together shape vegetation dynamics and ecological function in the western US (Alba et al. 2014). Non-native winter annual grasses such as *Bromus tectorum* L. (cheatgrass), which respond well to fire and change fuel characteristics, can create a plant-fire cycle that leads to loss of native biodiversity and attendant ecosystem function (D'Antonio and Vitousek 1992; Germino et al. 2016). Recent advances with pre-emergent herbicides have provided managers with an additional tool for controlling non-native annuals and biennials (see US EPA Pesticide Product Label, Rejuvra Plus, 2020 for a list of target species), while potentially reducing unwanted impact on native perennials (Sebastian et al. 2017; Clark et al. 2019; Clark 2020). Indaziflam (Rejuvra®) has recently been approved for use in natural areas, where its effect on propagule availability could mediate plant community response to pulsed disturbances such as fire. For example, given the targeted effect of Rejuvra® on short-lived species, it is of interest to assess whether its use could thwart establishment by short-lived native species that recruit post-fire.

Here we assessed plant community composition in areas of Heil Valley Ranch and nearby locations in Boulder County, Colorado, that were treated pre-fire (or not) with spraying regimens that include Rejuvra®, and that were burned (or not) during the 2020 Calwood Fire (setting up a naturally occurring factorial combination of treatments). Our overall objective was to determine whether sprayed areas are better primed for post-fire recruitment of desirable native species than untreated areas. Within the umbrella of this objective we:

- Assessed whether burning affected the efficacy of herbicide treatment in controlling the target non-native grasses *Bromus tectorum* and *Bromus japonicus*.
- Determined how the richness and relative abundances of native and introduced species responded to burning, spraying, and burning plus spraying; we include a focus on short-lived functional groups, given the targeted effect of Rejuvra® on short-lived species.

- Assessed whether burning or spraying affect the presence of species with different levels of tolerance to habitat degradation (using the Coefficient of Conservatism metric)

METHODS

Study design

The study area is located on the far eastern extent of the Calwood Fire burn perimeter, including Heil Valley Ranch and locations to the north that run largely adjacent to Highway 36. Burn severity was low across the entire extent of the predominantly grassland study area. We used several kmz layers provided by Boulder County Parks & Open Space (BCPOS) to haphazardly place transect locations across the study area (Table 1). The layers included the Calwood Fire burn perimeter, vegetation mapping polygons, herbicide spray polygons (note in Table 1 the full complement of herbicides used to spray each transect), and the location of prairie dog colonies, which were avoided. Our original design was a balanced factorial design with N = 9 transects located in each of the following treatment types: unburned, unsprayed = UU; unburned, sprayed= US; burned, unsprayed = BU; burned, sprayed = BS. A late-season excursion to ground-truth the spray polygons with the contractor resulted in an uneven design as follows: UU, N = 7; US, N = 10; BU, N = 5; BS, N = 11.

Vegetation sampling

We visited the study area throughout the growing season (late April through early August) to increase the veracity of our species identifications. Plants were observed as they matured from early-season vegetative to late-season flowering and fruiting states, and we made targeted specimen collections (e.g., of graminoids and other plants requiring microscopy for certain identification) to be housed in the Kathryn Kalmbach Herbarium at Denver Botanic Gardens. In a small number of cases, seedlings or other plants without the proper identifying structures were left at the genus or family level.

We estimated plant species richness and percent cover across the four treatment types during peak growing season (June 21 to July 8). We placed Daubenmire frames (50 cm x 20 cm)

at 5-m intervals along 33 50-m-long transects (Table 1) for a total of 10 quadrats per transect and 330 quadrats total. Percent cover of bare ground, litter, rocks, and plant species were estimated to the nearest percent. Trace amounts of vegetation were assigned a cover estimate of 0.5%. After estimating percent cover, we walked the entire length of the transect and counted all additional species present within a 1-m strip on each side of the tape (i.e., species counts were made in a 50 m x 2 m belt transect).

Functional Groups and Coefficients of Conservatism

We used the USDA Plants Database and Ackerfield 2015 to assign plant longevity as follows: short-lived = strict annuals and biennials, plus species that can range from annual to biennial; long-lived = perennial; “variable” includes a small subset of plants that can range from annual to perennial (e.g., *Argemone polyanthemos*). For habit, we grouped plants as forbs, graminoids (including *Carex* and *Juncus*), sub-shrubs to shrubs (e.g., *Artemisia frigida* and *Gutierrezia sarothrae*), and shrubs/trees (e.g., *Rhus trilobata* and *Celtis reticulata*). Table 2 includes the functional groups assignments for all species.

The Coefficient of Conservatism is an indicator that uses plant species composition to describe an area’s ecological condition (Rocchio 2007), which can range from degraded to fully intact. Two concepts underlie development of the conservatism indicator: 1) plant species differ in their tolerance to various types and magnitudes of disturbance and 2) plant species differ in the degree to which they depend on fully intact habitat. In this context, “fully intact” habitat reflects conditions prior to European settlement, which ushered in rapid changes in land use and disturbance regimes, as well as the introduction of non-native species. Species are assigned a ranking from zero to one. Those on the low end of the continuum show little fidelity to natural areas, while those on the high end occur only in relatively pristine sites. The indicator rankings are defined as follows:

0-3: Introduced species (always zero), or native species that occur in moderately to highly degraded sites

4-6: Native species that show some affinity to natural areas and are often dominant or are present across a wide range of habitats and environments

7-8: Native species associated mostly with natural areas, but that can sometimes persist in degraded habitat

9-10: Native species that tolerate very little or no habitat degradation

Statistical analysis

All statistical analyses were conducted in R version 3.6.2. We interpret the best-fit model for each response variable according to AICc, which measures the information lost between two statistical models and applies a bias correction for small sample size (Burnham et al. 2011). We used generalized linear models with a Poisson distribution to model species richness and *C* values. For total species richness and species richness of short-lived forbs, we compared a global model including the fixed effects of Burn status, Spray status, Origin (native or introduced) and their interactions to models without the interactions. For the proportion of species belonging to each functional group across treatment types, we separately analyzed native and introduced species. The global models included Burn status, Spray status, Functional group type, and their interactions. We analyzed *C* values only for native species.

The approach with the percent cover data of native and introduced species was similar to that used for richness models, except that we used general linear models with a Gaussian distribution. The global models for percent cover of *Bromus tectorum* and *Bromus japonicus* included Burn status, Spray status, and their interaction. For native species relativized cover, we visualize all functional groups (Figure 6), but do not include short-lived graminoids, shrubs/trees, and variable forbs in the models due rank deficiency (i.e., not occurring in all treatment types). Appendix 1 contains all model outputs showing significant fixed effects and their effect sizes (“Incidence Rate Ratios” for Poisson models and “Estimates” for Gaussian models). Post-hoc pairwise comparisons for all models were conducted with the “emmeans” package using the Tukey method to account for multiple comparisons. Post-hoc pairwise comparisons are presented in the text and figures.

Please note that we do not herein present richness broken out by introduced and native short-lived grasses because 1) the two native annual grasses, *Vulpia octoflora* and *Hordeum murinum*, were too uncommon to statistically assess and 2) percent cover of introduced short-

lived grasses is more informative than richness given that only two short-lived grasses, *Bromus tectorum* and *Bromus japonicus*, were broadly represented at the study site (with *Aegilops cylindrica* and *Bromus briziformis* too uncommon to statistically assess).

Finally, the figures make use of dotplots to show that we chose not to remove outliers, as these are part of the biological story and should not be clipped from the data set. They also illustrate how the unbalanced design contributes to variability around the means for each treatment type.

RESULTS

Spray efficacy

Bromus tectorum and *Bromus japonicus* were successfully controlled by the BCPOS spraying regimen that included Rejuvra®, and this was true in both burned and unburned areas (Figures 1 and 2; Appendix 1, Table A1, see effect sizes and significance of Spray status term). For *Bromus tectorum*, the magnitude of the spray effect was similar in burned (96% lower cover in sprayed areas) and unburned (93% lower cover) areas (Figure 1). For *Bromus japonicus*, the effect of spraying was more pronounced in burned areas (96% lower cover) than unburned areas (83% lower cover; Figure 2; see Appendix 1, Table A1 for significant Burn status x Spray status interaction). Comparing *B. japonicus* cover in unsprayed areas (Figure 2, treatments BU and UU) indicates there was a significant (post-hoc comparison, t ratio = 4.16, P -value = 0.0001) post-fire flush in its cover (76% higher) when not controlled. There was no evidence of a post-fire flush for *B. tectorum* (compare treatments BU and UU in Figure 1).

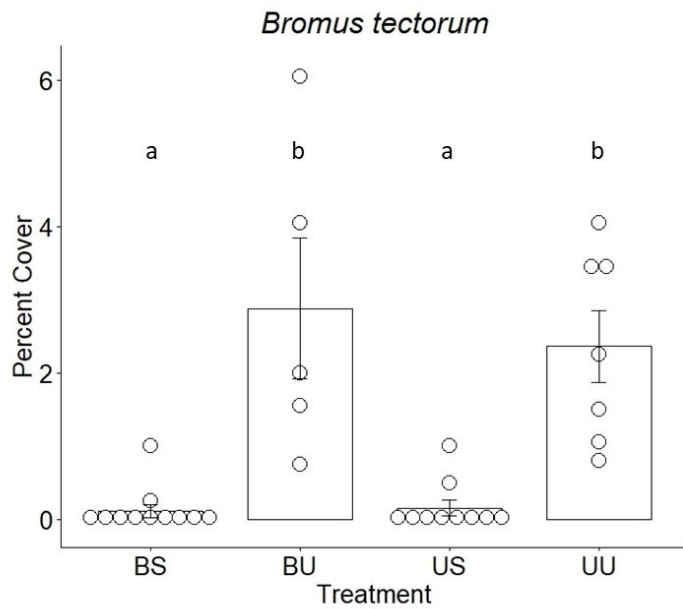


Figure 1. Mean (\pm SE) species richness of *Bromus tectorum* across four treatment types: BS = burned and sprayed; BU = burned and unsprayed; US = unburned and sprayed; UU = unburned and unsprayed. Dots are estimates for each transect per treatment type. Different letters on bars indicate significant differences at $P = 0.05$ based on post-hoc pairwise comparisons. See Appendix 1, Table A1 for effect sizes of main effects.

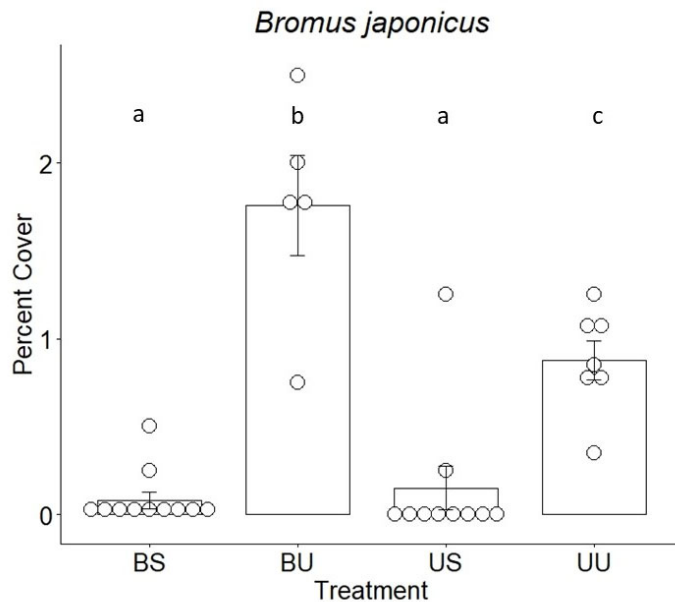


Figure 2. Mean (\pm SE) species richness of *Bromus japonicus* across four treatment types: BS = burned and sprayed; BU = burned and unsprayed; US = unburned and sprayed; UU = unburned and unsprayed. Dots are estimates for each transect per treatment type. Different letters on bars indicate significant differences at $P = 0.05$ based on post-hoc pairwise comparisons. See Appendix 1, Table A1 for effect sizes of main effects and interaction(s).

Total species richness

We identified 157 species present across the study area (Table 2), of which 33% are introduced (39 of 118). The richness of introduced species was slightly higher in burned than unburned areas (mean \pm SE burned = 11.3 ± 1.5 ; mean \pm SE unburned = 9.29 ± 1.0), while the richness of native species was slightly lower in burned than unburned areas (mean \pm SE burned = 25 ± 2.0 , mean \pm SE unburned = 29.8 ± 1.2). This different direction in the response of introduced versus native species to fire is evidenced by a significant Burn status x Origin interaction (Appendix 1, Table A2). The richness of introduced species was significantly lower in sprayed areas regardless of burn status (37% lower in burned areas; 34% lower in unburned areas; Figure 3). For native species, while spraying tended to reduce richness in burned areas

(21% lower), the effect was not significant (Figure 3; post-hoc comparison, z ratio = -2.26, $P = 0.11$; also see Appendix 1, Table A2 for significant Spray status x Origin interaction). In unburned areas, spraying reduced native richness even less so, and again non-significantly (8.6% lower; Figure 3; post-hoc comparison, z ratio = -2.26, $P = 0.11$).

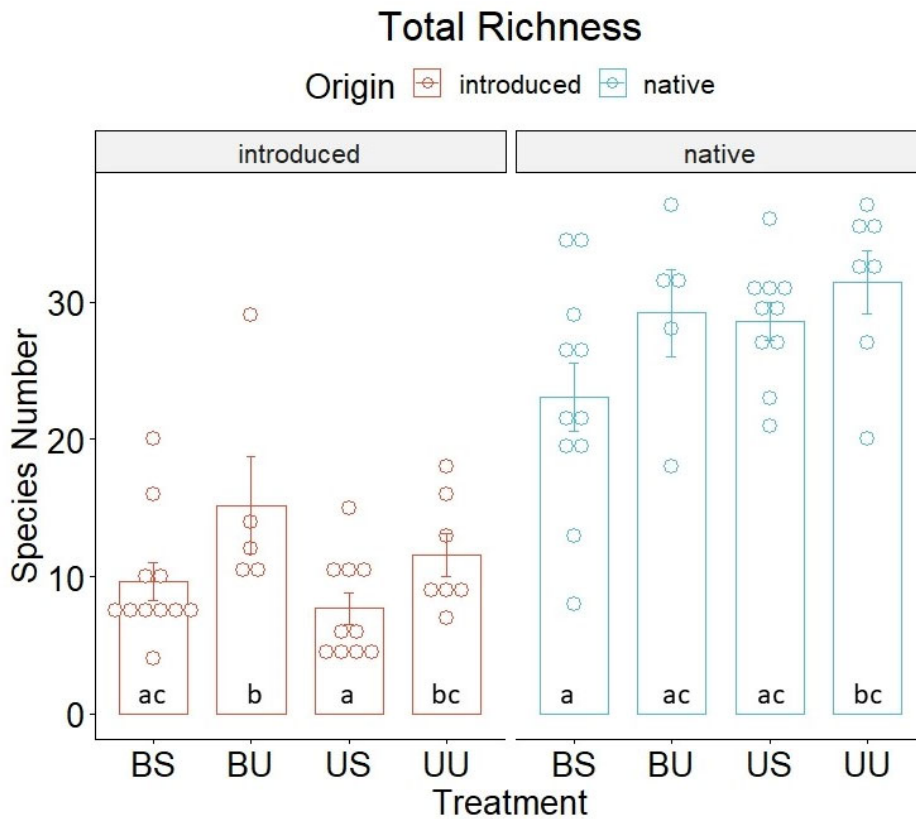


Figure 3. Mean (\pm SE) species richness of introduced and native species across four treatment types: BS = burned and sprayed; BU = burned and unsprayed; US = unburned and sprayed; UU = unburned and unsprayed. Dots are estimates for each transect per treatment type. Different letters on bars indicate significant differences at $P = 0.05$ based on post-hoc pairwise comparisons. See Appendix 1, Table A2 for effect sizes of main effects and interaction(s).

Species richness of short-lived forbs

Given the targeted effect of Rejuvra® on short-lived species, we examined the species richness of introduced and native short-lived forbs separately from the other functional groups (see Table 2 for how we defined functional groups and which species are short-lived). The richness of introduced short-lived forbs was 55% lower in burned areas that had been sprayed than burned areas that had not been sprayed (Figure 4; post-hoc comparison, z ratio = -3.39, P = 0.004). In unburned areas, introduced forb richness was 43% lower in sprayed areas, a marginally significant effect (post-hoc comparison, z ratio = -2.32, P = 0.09; Figure 4). There were 15 species of introduced short-lived forbs (Table 2), with the most abundant being *Erodium cicutarium*, *Alyssum simplex*, and *Tragopogon dubius* (further detailed in the section *Percent cover of short-lived forbs*).

For native species, spraying had no effect on richness in unburned areas (Figure 4; see Appendix 1, Table A3 for marginally significant Burn status x Spray status x Origin interaction). However, in burned areas, richness was 75% lower in sprayed areas than unsprayed areas, and this difference was highly significant (Figure 4; post-hoc comparison, z ratio = -4.7, P = 0.0001; See Appendix 1, Table A3 for effect size of Treatment x Short-lived forb interaction). There were 15 species of native-short lived forbs (comprising only 13% of the native species pool; Table 2), including three species of *Euphorbia* (*E. dentata*, *E. marginata*, and *E. spathulata*) and two species of *Triodanis* (*T. leptocarpa* and *T. perfoliata*). Most species were exceedingly sparsely distributed, occurring on only a handful of transects (Table 2). The most widespread short-lived native species was *Erigeron flagellaris* (Table 2), a biennial that occurred on 73% of the transects. Fourteen of the 15 species occurred on burned and unsprayed transects; 7 occurred on burned and sprayed, and 7 on unburned and sprayed, transects; and 9 occurred on unburned and unsprayed transects (Table 2).

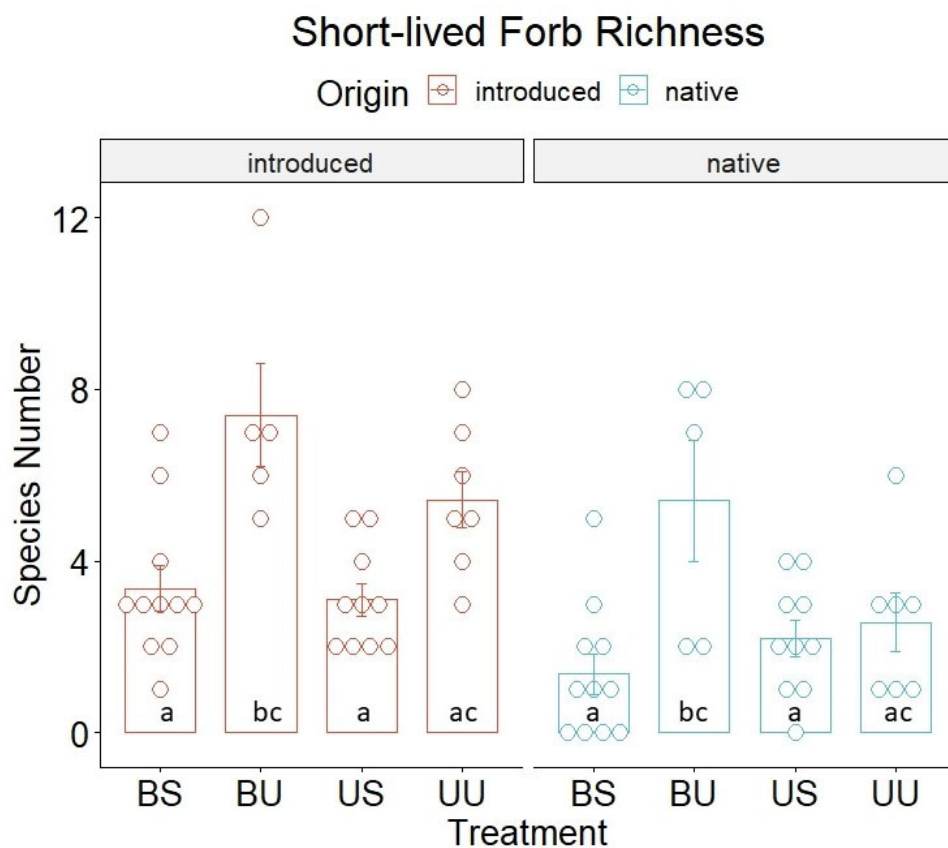


Figure 4. Mean (\pm SE) species richness of introduced and native short-lived forbs across four treatment types: BS = burned and sprayed; BU = burned and unsprayed; US = unburned and sprayed; UU = unburned and unsprayed. Dots are estimates for each transect per treatment type. Different letters on bars indicate significant differences at $P = 0.05$ based on post-hoc pairwise comparisons. See Appendix 1, Table A3 for effect sizes of main effects and interaction(s).

Proportional richness by functional groups

The proportional contribution of different functional groups to total species richness illustrates a few points. First, not surprisingly, introduced species are disproportionately represented by short-lived forbs and grasses, and are otherwise functionally depauperate relative to natives (lacking any sub-shrubs, shrubs, or trees). Second, the higher richness of native short-lived forbs in burned and unsprayed areas (Figure 4) also leads to their higher proportional representation relative to other functional groups in this treatment type (Figure 5, yellow bars; Appendix 1, Table A5, significant Treatment x Short-lived Forb interaction; pairwise comparison, z ratio = -4.27, $P = 0.0045$). Third, there were no other shifts in the proportional representation of native functional groups among different treatments (Appendix 1, Table A5; note the significant interactions of Treatment x Variable Forbs did not remain significant under the Tukey family-wise error rate).

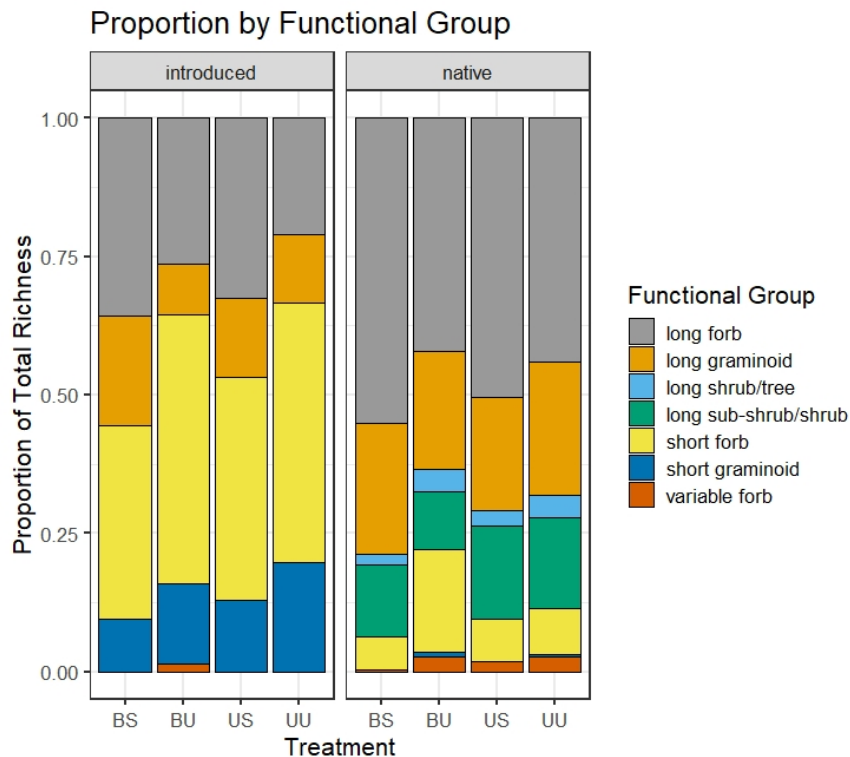


Figure 5. Proportion of total species richness by functional group. See Appendix 1, Tables A4 (introduced species) and A5 (native species) for effect sizes of main effects and interactions.

Coefficients of Conservatism

Neither burning nor spraying strongly shaped plant communities in terms of how natives species' Coefficients of Conservatism (C values) were distributed across treatment types (Figure 6; BS mean \pm SE = 4.7 ± 0.09 ; BU mean \pm SE = 4.48 ± 0.15 ; US mean \pm SE = 4.65 ± 0.09 ; UU mean \pm SE = 4.57 ± 0.11 ; no effects [burning, spraying, burning x spraying] were significant using a generalized linear model with Poisson distribution; data not shown). The species with the highest C value of 9, *Andropogon gerardii*, was the fourth most-common species in the study (Table 3), highlighting the importance of this area for high-quality, remnant populations of this tallgrass. Several species had C values of 7, including *Calochortus gunnisonii*, *Carex inops* subsp. *heliophila* (also one of the most common species; Table 3), *Carex occidentalis*, *Coryphantha missouriensis*, *Hybanthus verticillatus*, *Oenothera serrulata* var. *serrulata*, and *Pseudoroegneria spicata*.

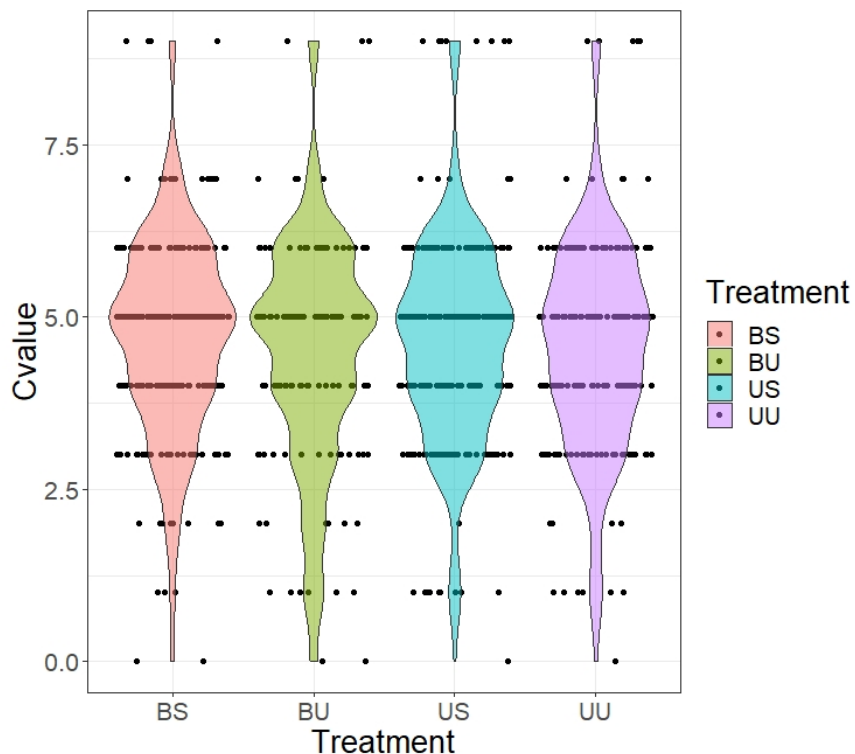


Figure 6. Violin plots showing the distribution of native species' Coefficients of Conservatism across the four treatment types.

Total percent cover

Only burning significantly affected the cover of introduced species (Figure 6; t ratio = 1.96, $P = 0.055$), with 58% higher cover in burned (mean \pm SE = 21.2%) than unburned (mean \pm SE = 13.4%) areas. There was, however, a trend toward lower cover (by 38%) of introduced species in unburned areas that had been sprayed (Figure 7; compare treatment US to UU). Total cover of native species did not vary significantly in relationship to burning or spraying (Figure 7; Appendix 1, Table A6, see significant Burn status x Origin interaction). Twenty-three species had an average cover of at least 0.5% (see Table 3 for all species and how their cover varies across treatment types). Of these common species, 8 (34%) were introduced. The top 10 most abundant species include *Convolvulus arvensis*, *Pascopyrum smithii*, *Psoralidium tenuiflorum*, *Andropogon gerardii*, *Poa compressa*, *Bouteloua curtipendula*, *Ambrosia psilostachya*, *Rhus trilobata*, *Symphytotrichum ericoides*, and *Erodium cicutarium*.

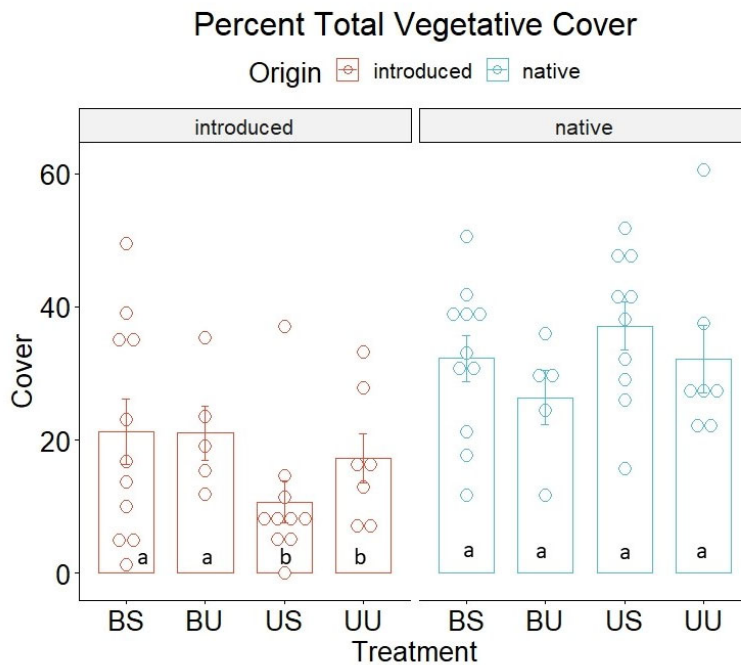


Figure 7. Mean (\pm SE) percent cover of all introduced and native species across four treatment types: BS = burned and sprayed; BU = burned and unsprayed; US = unburned and sprayed; UU = unburned and unsprayed. Dots are estimates for each transect per treatment type. Different

letters on bars indicate significant differences at $P = 0.05$ based on post-hoc pairwise comparisons. See Appendix 1, Table A6 for effect sizes of main effects and interactions.

Percent cover of short-lived forbs

Percent cover of introduced short-lived forbs was lower in sprayed than unsprayed areas, both in the presence (Figure 8; 63% lower cover; post-hoc comparison, t ratio = -4.1, $P = 0.0008$) and absence (58% lower cover; post-hoc comparison, t ratio = -4.1, $P = 0.0008$) of fire. The most common introduced short-lived forbs were *Erodium cicutarium*, *Alyssum simplex*, and *Tragopogon dubius* (see Table 3 for means \pm SEs across treatment types). Conversely, while there was a slight trend toward lower cover of natives in sprayed than unsprayed areas (Figure 8), it was not significant (see Appendix 1, Table A7 for Spray status x Origin interaction).

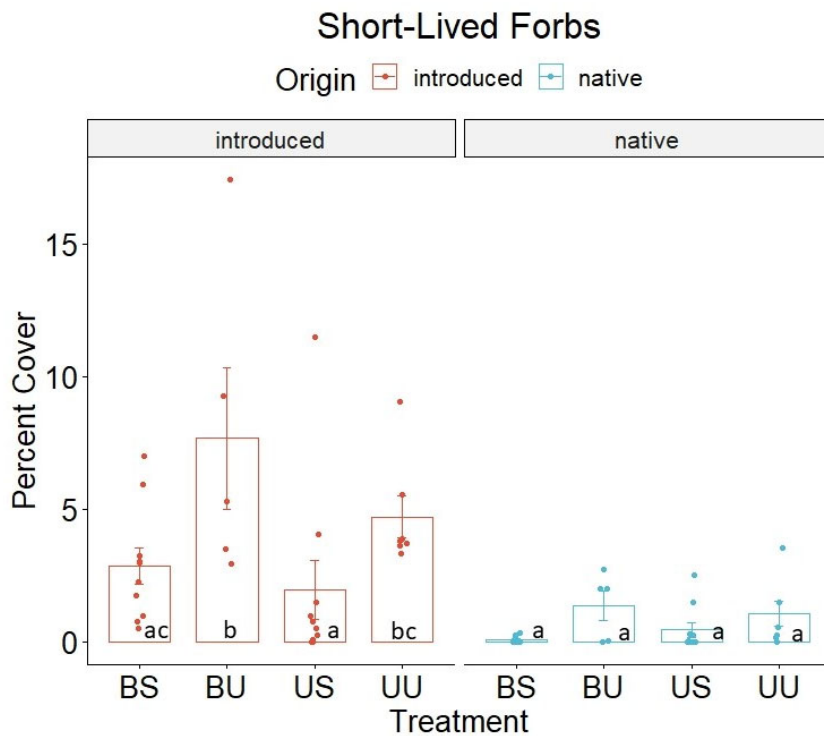


Figure 8. Mean (\pm SE) percent cover of introduced and native short-lived forbs across four treatment types: BS = burned and sprayed; BU = burned and unsprayed; US = unburned and sprayed; UU = unburned and unsprayed. Dots are estimates for each transect per treatment

type. Different letters on bars indicate significant differences at $P = 0.05$ based on post-hoc pairwise comparisons. See Appendix 1, Table A7 for effect sizes of main effects and interactions.

Relativized percent cover by functional groups

For introduced species, the proportion of cover made up of short-lived forbs was lower in sprayed areas that were burned (Figure 9; Appendix 1, Table A8, Treatment x Short-lived forb interactions), however this difference did not remain significant when performing post-hoc comparisons using the Tukey Method (t ratio = -0.61, $P = 0.92$). The same pattern held for short-lived graminoids (Appendix 1, Table A8, see significant Treatment x Short-lived graminoid interactions; compare to post-hoc analysis, t ratio = -1.2, $P = 0.6$). For native species, there were few significant interactions between the relativized cover of functional groups and burning and spraying, and we retained the better fit model without the interaction terms (Appendix 1, Table A9).

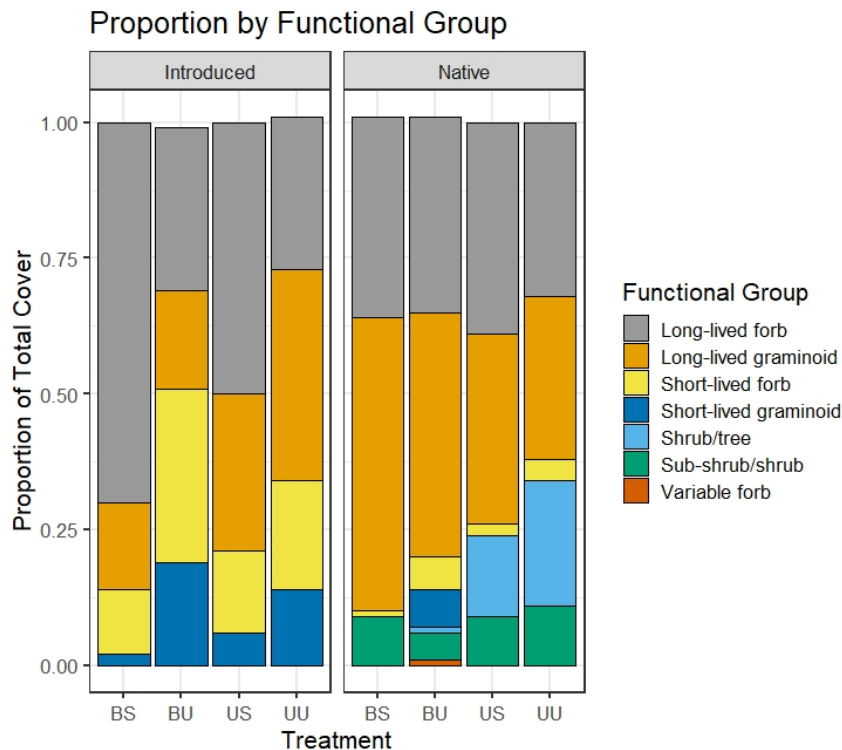


Figure 9. Proportion of relativized percent cover by functional group. See Appendix 1, Tables A9 (introduced species) and A10 (native species) for effect sizes of main effects and interactions.

DISCUSSION

Spray efficacy

Our results show that spraying regimens that include Rejuvra® successfully control *B. tectorum* and *B. japonicus* on Heil Valley Ranch and nearby Open Space land. We detected a signal of its efficacy across variability associated with several vegetation types (Table 1) and show that it remains effective in burned areas. Despite *B. tectorum* displaying post-fire increases in performance in other systems (Davies and Nafus 2013), we did not observe that phenomenon here (at least in the first year post-fire). There is a growing acknowledgment of geographic variability in how cheatgrass responds to fire (Taylor et al. 2014), with several other factors, including grazing, topography, and precipitation, also affecting its prevalence (Williamson et al. 2020). Conversely, *B. japonicus* did show some indication of a post-fire flush in cover, a pattern that contradicts other studies showing that fire reduces *B. japonicus* performance, for example through removal of litter that creates good microsites for seedling establishment (Whisenant 1990).

The overall cover estimates of the two grasses appear low. It may be that while our study design effectively captured relative differences in *Bromus tectorum* and *Bromus japonicus* cover across the treatment types, it did not optimally estimate the absolute abundance of the grasses. Indeed, we sometimes saw dense patches of cheatgrass near, but not within, our transects. However, this scenario is in some ways a testament to the fact that treatment differences arose without any *a priori* information about cheatgrass distributions used when haphazardly placing the transects across treatment types. It is also the case that the average percent cover (not only for these grasses, but for all species in the study) can seem low once the range of variability gets averaged over. For example, many of the quadrats along our transects contained 10-25% cover of *B. tectorum*.

Species richness

When considering all species, we found that introduced species tended to have higher richness in burned areas and natives tended to have lower richness, a pattern that emerges across many ecosystems worldwide (Alba et al. 2014). However, while the direction of the response to fire was different for these two groups, the magnitude of the responses was not particularly pronounced. Therefore, it can be hypothesized that low-severity fire in foothills grassland ecosystems may not strongly exclude native species from the species pool, nor widely facilitate establishment of introduced species, although the observed trend suggests caution (see Fornwalt et al. for a similar example in ponderosa pine–Douglas-fir forests). Spraying had a larger effect on introduced species richness than did burning, with the successful outcome of lower richness in both burned and unburned areas. At the same time, spraying did not appear to strongly affect overall native species richness.

A separate analysis of short-lived introduced forbs showed that spraying successfully reduced their richness in both burned and unburned areas. However, the results also suggest that spraying had non-target effects on native species, with burned and unsprayed areas having high richness indicative of a post-fire flush that did not occur in burned and sprayed areas. Conversely, spraying did not affect native richness in unburned areas. As such, experimental research should be undertaken to explore this pattern further (e.g., using a before-after-control-impact (BACI) study design). Such work can help determine whether it is necessary to mitigate non-target effects of spraying during post-fire community assembly, for example by seeding with native forbs known to recruit during the high-resource window created by fire.

We did not find a strong response of plant communities to burning or spraying based on the distribution of *C* values of native species among treatment types. We had hypothesized that more ruderal, early successional native species (lower *C* value) might be over-represented following fire, or that spraying might disrupt plant communities such that sensitive species with high affinity to “pristine” conditions might non-randomly drop out. While we did not uncover a strong shift in *C* values, it is the case that many of the native short-lived forbs have low *C* values, indicating a ruderal nature that may favor them in a post-fire landscape (e.g., *Euphorbia dentata*, *Euphorbia marginata*, and *Helianthus annuus* = 1; *Lappula occidentalis*, *Lepidium*

virginicum, and *Plantago patagonica* = 2; *Ellisia nyctelea*, *Erigeron flagellaris*, and *Triodanis leptocarpa* = 3).

Species cover

Burning significantly increased the cover of introduced species but did not affect the cover of native species. Spraying did not have a significant effect on the total cover of introduced species, although there was a trend toward lower cover in sprayed versus unsprayed areas that were not burned. In this case, the outlier should be noted, as it is well removed from all the other transect means (due to very high cover of *Convolvulus arvensis* on transect US2020_2 in Heil Valley Ranch; see Table 1 for location information). Thus, while not significant, there is support for spray efficacy in unburned areas, with this efficacy negated by burning.

There was no evidence that spraying affected the cover of native species, and in fact some of the highest estimates of native cover occurred on sprayed transects. Separately considering short-lived forbs illustrated that spraying was effective in lowering introduced cover in both burned and unburned areas, while native species were not significantly affected (again, despite some non-significant trends toward a reduction in sprayed areas). Finally, visualization of the relative cover of different functional groups across treatment types showed lower proportional representation of introduced short-lived grasses and forbs in sprayed than unsprayed areas; however, these patterns were not significant when held up to the Tukey's family-wise errors rates during post-hoc analysis. Native proportional cover of different functional groups did not significantly vary across treatment types.

Long-lived forbs and grasses

While we framed our report to focus on hypotheses surrounding short-lived species, we want to highlight that the current BCPOS spray regimens appear either neutral (in terms of richness) or beneficial (in terms of percent cover) to long-lived native forbs and grasses (Table 4). Specifically, we note that long-lived native forb cover was 19% higher in sprayed than unsprayed areas that burned and 40.5% higher in sprayed areas that did not burn. Long-lived

native grass cover was 40% higher in sprayed than unsprayed areas that burned and 39% higher in sprayed areas that did not burn. These patterns corroborate previous findings that the targeted effect of Rejuvra® facilitates long-lived species (Clark et al. 2019), which in the case of forbs, may have attendant positive effects on pollinators (Seshardri and Hardin 2018). We note that final interpretation of these patterns relies on statistical analyses that we were unable to complete by the report deadline. The formalized results will be made public upon completion.

CONCLUSIONS

Overall, our findings suggest that the incorporation of targeted-action Rejuvra® into BCPOS weed management regimens achieves the goal of suppressing annual grasses while lessening the impact on long-lived native species. However, there is also evidence of potential non-target effects on the richness of short-lived native species under post-fire conditions. It may thus be of interest to design future experimental studies (e.g., using a before-after-control-impact [BACI] study design) that explicitly test how disturbances like fire interact with Rejuvra® to affect the performance of both introduced and native short-lived species.

REFERENCES CITED

- Alba C, Skálová H, McGregor KF, D'Antonio CM and P Pyšek. 2014. Native and exotic plant species respond differently to wildfire and prescribed fire as revealed by meta-analysis. *Journal of Vegetation Science* 26:102-113.
- Burnham KP, Anderson DR and KP Huyvaert. 2011. AIC model selection and multimodel inference in behavioral ecology: Some background, observations, and comparisons. *Behavioral Ecology and Sociobiology* 65:23-35.
- Clark SL. 2020. Using herbicides to restore native species and improve habitat on rangelands and wildlands. *Outlooks on Pest Management* 31:57-60.
- Clark SL, Sebastian DJ, Nissen SJ, and JR Sebastian. 2019. Effects of indaziflam on native species in natural areas and rangeland. *Invasive Plant Science and Management* 12:60-67.
- D'Antonio CM and PM Vitousek. 1992. Biological invasions by exotic grasses, the grass/fire cycle, and global change. *Annual Review of Ecology and Systematics* 23:63-87.
- Davies KW and AM Nafus. 2013. Exotic annual grass invasion alters fuel amounts, continuity and moisture content. *International Journal of Wildland Fire* 22:353-358.
- Fornwalt PJ, Kaufmann MR and TJ Stohlgren. 2010. Impacts of mixed severity wildfire on exotic plants in a Colorado ponderosa pine–Douglas-fire forest. *Biological Invasions* 12:2683-2695.
- Germino MJ, Belnap J, Stark JM, Allen EB, and BM Rau. Ecosystem impacts of exotic annual invaders in the genus *Bromus*. *In Exotic brome-grasses in arid and semiarid ecosystems of the Western US 2016* (pp. 61-95). Springer, Cham.
- Rocchio J. 2007. Colorado Natural Heritage Program. Floristic Quality Assessment Indices for Colorado Plant Communities.
- Sebastian DJ, Fleming MB, Patterson EL, Sebastian JR, and SJ Nissen. 2017. Indaziflam: A new cellulose-biosynthesis-inhibiting herbicide provides long-term control of invasive winter annual grasses. *Pest Management Science* 73:2149-2162.
- Seshadri A and J Hardin. 2018. Bringing back flowering plants and pollinators through effective control of invasive winter annual grasses with Esplanade herbicide. Draft Small Grant report to Boulder County Open Space.
- Taylor K, Brummer T, Rew LJ, Lavin M, and BD Maxwell. 2014. *Bromus tectorum* response to fire varies with climate conditions. *Ecosystems* 17:960-973.
- Whisenant SG. 1990. Postfire populations dynamics of *Bromus japonicus*. *The American Midland Naturalist*. 123:301-308.

Williamson MA, Fleishman E, MacNally RC, Chambers JC, Bradley BA, Dobkin DS, Board DI, Fogarty FA, Norning N, Leu M, and MW Zillig. 2020. Fire, livestock grazing, topography, and precipitation affect occurrence and prevalence of cheatgrass (*Bromus tectorum*) in the central Great Basin, USA. *Biological Invasions* 22:663-680.

Table 1. Transect identifiers and associated sample date, location, and vegetation alliance. BS = burned, sprayed; BU = burned, unsprayed; US = unburned, sprayed; UU = unburned, unsprayed. Four transects in bold had their treatment type amended following ground-truthing of sprayed areas with BCPOS spray contractor (those ending in S changed to sprayed. Those ending in U changed to unsprayed). Three transects in bold and italics (ending in D) were dropped from the data set when their spray status could not be confidently assigned during ground-truthing. Spray regimen details provided by BCPOS.

Transect ID	Sample Date	Origin Latitude	Origin Longitude	End Latitude	End Longitude	Elevation (m)	Bearing from Origin	BCPOS Vegetation Mapping Layer Alliance Description	Spray Regimen (Esplanade now Rejuvra®)
BS2018_1	7/8/2021	40.15343	105.2795	40.15306	105.27979	1730	206	Annual-dominated Upland Disturbance	Esplanade 7 + Glyphosate 12 + NIS; Quinstar to control field bindweed
BS2018_2	6/21/2021	40.16819	105.2669	40.16808	105.26747	1718	250	Blue Grama - Buffalograss Shortgrass Prairie	Esplanade 7 + Glypho 12 + NIS; Quinstar to control field bindweed
BS2018_3	6/24/2021	40.17223	105.26691	40.17266	105.26671	1734	18	Needle-and-Thread Northwestern Great Plains Herbaceous	Esplanade 7 + Glypho 12 + NIS
BS2019_1	7/8/2021	40.15498	105.27807	40.15452	105.27822	1748	190	Annual-dominated Upland Disturbance	Esplanade + Glyphosate; Quinstar to control field bindweed
BS2019_2	6/25/2021	40.16431	105.27136	40.16384	105.27129	1744	180	Western Wheatgrass Herbaceous Alliance	Esplanade + Glyphosate; Quinstar to control field bindweed
BS2019_3	6/21/2021	40.16833	105.2691	40.16851	105.26857	1723	60	Big Bluestem - (Yellow Indiangrass) Herbaceous	Esplanade 7 + Glyphosate 12 + NIS
BS2020_1	6/28/2021	40.13689	105.30101	40.13734	105.30087	1787	9	Ponderosa Pine Tallgrass Savannah Herbaceous	Esplanade 7 + Escort 1 + Glyphosate 10
BS2020_2	7/7/2021	40.1361	105.3013	40.1359	105.3008	1704	108	Ponderosa Pine Tallgrass Savannah Herbaceous	Esplanade 7 + Escort 1 + Glyphosate 10

Table 1. Transect identifiers and associated sample date, location, and vegetation alliance. BS = burned, sprayed; BU = burned, unsprayed; US = unburned, sprayed; UU = unburned, unsprayed. Four transects in bold had their treatment type amended following ground-truthing of sprayed areas with BCPOS spray contractor (those ending in S changed to sprayed. Those ending in U changed to unsprayed). Three transects in bold and italics (ending in D) were dropped from the data set when their spray status could not be confidently assigned during ground-truthing. Spray regimen details provided by BCPOS.

Transect ID	Sample Date	Origin Latitude	Origin Longitude	End Latitude	End Longitude	Elevation (m)	Bearing from Origin	BCPOS Vegetation Mapping Layer Alliance Description	Spray Regimen (Esplanade now Rejuvra®)
BS2020_3U	6/24/2021	40.16506	105.27263	40.16538	105.27222	1761	35	Big Bluestem - (Yellow Indiangrass) Herbaceous	N/A
BU2018_1	7/8/2021	40.15569	105.28132	40.15532	105.28149	1781	178	Ponderosa Pine Tallgrass Savannah Herbaceous	N/A
<i>BU2018_2D</i>	<i>6/21/2021</i>	<i>40.16984</i>	<i>105.26771</i>	<i>40.17024</i>	<i>105.26775</i>	<i>1724</i>	<i>0</i>	<i>Blue Grama - Buffalograss Shortgrass Prairie</i>	<i>N/A</i>
BU2018_3S	6/24/2021	40.17283	105.26716	40.1732	105.2675	1735	308	Needle-and-Thread Northwestern Great Plains Herbaceous Alliance	N/A
BU2019_1	7/8/2021	40.15548	105.27843	40.15507	105.27875	1798	194	Annual-dominated Upland Disturbance	N/A
BU2019_2S	6/25/2021	40.16374	105.27198	40.16372	105.27255	1747	269	Western Wheatgrass Herbaceous	Esplanade 7 + Glyphosate 10
BU2019_3	6/21/2021	40.1688	105.26995	40.16868	105.27051	1743	253	Big Bluestem - (Yellow Indiangrass) Herbaceous	N/A
BU2020_1	6/28/2021	40.13515	105.30154	40.13477	105.30183	1779	210	Ponderosa Pine Woodland	N/A

Table 1. Transect identifiers and associated sample date, location, and vegetation alliance. BS = burned, sprayed; BU = burned, unsprayed; US = unburned, sprayed; UU = unburned, unsprayed. Four transects in bold had their treatment type amended following ground-truthing of sprayed areas with BCPOS spray contractor (those ending in S changed to sprayed. Those ending in U changed to unsprayed). Three transects in bold and italics (ending in D) were dropped from the data set when their spray status could not be confidently assigned during ground-truthing. Spray regimen details provided by BCPOS.

Transect ID	Sample Date	Origin Latitude	Origin Longitude	End Latitude	End Longitude	Elevation (m)	Bearing from Origin	BCPOS Vegetation Mapping Layer Alliance Description	Spray Regimen (Esplanade now Rejuvra®)
BU2020_2S	7/7/2021	40.13483	105.30215	40.13488	105.30276	1794	288	Ponderosa Pine Woodland	Esplanade 7+ Milestone 5
<i>BU2020_3D</i>	<i>6/25/2021</i>	<i>40.16478</i>	<i>105.27247</i>	<i>40.16468</i>	<i>105.27188</i>	<i>1753</i>	<i>90</i>	<i>Big Bluestem - (Yellow Indiangrass) Herbaceous</i>	<i>N/A</i>
US2018_1	6/29/2021	40.17715	105.25732	40.17696	105.25784	1699	240	Big Bluestem - Indiangrass Mixed grass Western Plains Grassland	Esplanade 7 + Glyphosate 12 + NIS; Quinstar to control field bindweed.
US2018_2	6/22/2021	40.1787	105.25883	40.17862	105.25829	1735	92	Three-leaved Sumac Shrub Savannah Herbaceous	Esplanade 7 + Glyphosate 12 + NIS; Quinstar to control field bindweed.
US2018_3	6/23/2021	40.17918	105.2582	40.17892	105.25865	1719	220	Three-leaved Sumac Shrub Savannah Herbaceous	Esplanade 7 + Glyphosate 12 + NIS; Quinstar to control field bindweed.
US2019_1	7/2/2021	40.17697	105.26083	40.17659	105.26078	1697	150	Three-leaved Sumac Shrub Savannah Herbaceous	Esplanade + Glyphosate
US2019_2	7/2/2021	40.17829	105.26161	40.17784	105.26191	1676	192	Three-leaved Sumac Shrub Savannah Herbaceous	Esplanade + Glyphosate
US2019_3	6/22/2021	40.18106	105.2581	40.18108	105.2587	1749	273	Three-leaved Sumac Shrub Savannah Herbaceous	Esplanade + Glyphosate

Table 1. Transect identifiers and associated sample date, location, and vegetation alliance. BS = burned, sprayed; BU = burned, unsprayed; US = unburned, sprayed; UU = unburned, unsprayed. Four transects in bold had their treatment type amended following ground-truthing of sprayed areas with BCPOS spray contractor (those ending in S changed to sprayed. Those ending in U changed to unsprayed). Three transects in bold and italics (ending in D) were dropped from the data set when their spray status could not be confidently assigned during ground-truthing. Spray regimen details provided by BCPOS.

Transect ID	Sample Date	Origin Latitude	Origin Longitude	End Latitude	End Longitude	Elevation (m)	Bearing from Origin	BCPOS Vegetation Mapping Layer Alliance Description	Spray Regimen (Esplanade now Rejuvra®)
US2020_1	6/28/2021	40.13361	105.30104	40.13341	105.30051	1770	100	Ponderosa Pine Tallgrass Savannah Herbaceous	Esplanade 7 + Escort 1 + Glyphosate 10
US2020_2	7/7/2021	40.13383	105.3007	40.13358	105.30051	1773	142	Ponderosa Pine Tallgrass Savannah Herbaceous	Esplanade 7 + Escort 1 + Glyphosate 10
US2020_3	6/29/2021	40.18359	105.2542	40.18407	105.25401	1714	25	Needle-and-Thread Bunch Herbaceous	Esplanade 7 + Escort 1 + Glyphosate 10
UU2018_1S	7/8/2021	40.17847	105.25632	40.17873	105.25676	1688	298	Ponderosa Pine / Grass Understory Southern Rocky Mountain Open Woodland	Esplanade 7 + Glyphosate 12 + NIS; Quinstar to control field bindweed.
UU2018_2	6/23/2021	40.17829	105.25967	40.17873	105.25973	1718	349	Three-leaved Sumac Shrub Savannah Herbaceous	N/A
UU2018_3	6/23/2021	40.1791	105.25768	40.1791	105.25712	1720	90	Ponderosa Pine / Grass Understory Southern Rocky Mountain Open Woodland	N/A
UU2019_1	7/2/2021	40.17761	105.26059	40.17749	105.26007	1702	106	Three-leaved Sumac Shrub Savannah Herbaceous	N/A
UU2019_2	7/2/2021	40.17783	105.26259	40.1774	105.26273	1728	196	Three-leaved Sumac Shrub Savannah Herbaceous	N/A

Table 1. Transect identifiers and associated sample date, location, and vegetation alliance. BS = burned, sprayed; BU = burned, unsprayed; US = unburned, sprayed; UU = unburned, unsprayed. Four transects in bold had their treatment type amended following ground-truthing of sprayed areas with BCPOS spray contractor (those ending in S changed to sprayed. Those ending in U changed to unsprayed). Three transects in bold and italics (ending in D) were dropped from the data set when their spray status could not be confidently assigned during ground-truthing. Spray regimen details provided by BCPOS.

Transect ID	Sample Date	Origin Latitude	Origin Longitude	End Latitude	End Longitude	Elevation (m)	Bearing from Origin	BCPOS Vegetation Mapping Layer Alliance Description	Spray Regimen (Esplanade now Rejuvra®)
<i>UU2019_3D</i>	<i>6/22/2021</i>	<i>40.18154</i>	<i>105.25957</i>	<i>40.18201</i>	<i>105.25954</i>	<i>1770</i>	<i>355</i>	<i>Three-leaved Sumac Shrub Savannah Herbaceous</i>	<i>N/A</i>
UU2020_1	6/28/2021	40.13304	105.30037	40.13261	105.30064	1761	210	Ponderosa Pine Tallgrass Savannah Herbaceous	N/A
UU2020_2	7/7/2021	40.13398	105.30006	40.13442	105.29989	1764	344	Ponderosa Pine Tallgrass Savannah Herbaceous	N/A
UU2020_3	6/29/2021	40.18314	105.25541	40.18278	105.25559	1728	210	Needle-and-Thread Bunch Herbaceous	N/A

Table 2. Species listed by their presence (denoted by an "x") across treatments, or for native short-lived forbs, by the number of transects on which they occurred. UU = unburned, unsprayed; US = unburned, sprayed; BU = burned, unsprayed; BS = burned, sprayed. Transect replication per treatment follows treatment type. Species on Colorado's noxious weed list are denoted with superscripts (a = List A; b = List B; c = List C). Only taxa identified to the species level are included. We combined strictly annual, strictly biennial, and annual to biennial species into the grouping of "short-lived" for analysis. Species with variable longevity (can range from annual to biennial to perennial) were coded as "variable" for analysis. Other functional groups are self-explanatory. Nomenclature according to Ackerfield 2015.

Species	Native or Introduced	Longevity	Functional Group for Analysis	UU (N = 7)	US (N = 10)	BU (N = 5)	BS (N = 11)
<i>Aegilops cylindrica</i>^b	Introduced	Annual	Short-lived graminoid	x			
<i>Allium textile</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Alyssum simplex</i>	Introduced	Annual	Short-lived forb	x	x	x	x
<i>Ambrosia psilostachya</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Andropogon gerardii</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Argemone polyanthemus</i>	Native	Annual/Biennial/Perennial	Variable forb		x	x	x
<i>Aristida purpurea</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Artemisia campestris</i>	Native	Perennial	Long-lived forb				x
<i>Artemisia frigida</i>	Native	Perennial	Sub-shrub/shrub	x	x	x	x
<i>Artemisia ludoviciana</i>	Native	Perennial	Sub-shrub/shrub	x	x	x	x
<i>Asclepias engelmanniana</i>	Native	Perennial	Long-lived forb	x		x	
<i>Asclepias pumila</i>	Native	Perennial	Long-lived forb	x	x		
<i>Asclepias viridiflora</i>	Native	Perennial	Long-lived forb			x	
<i>Astragalus agrestis</i>	Native	Perennial	Long-lived forb		x		x
<i>Astragalus drummondii</i>	Native	Perennial	Long-lived forb			x	x
<i>Astragalus flexuosus</i> var. <i>flexuosus</i>	Native	Perennial	Long-lived forb				x
<i>Bouteloua curtipendula</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Bouteloua gracilis</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Brickellia eupatorioides</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Bromus briziformis</i>	Introduced	Annual	Short-lived graminoid			x	
<i>Bromus inermis</i>	Introduced	Perennial	Long-lived graminoid		x	x	x
<i>Bromus japonicus</i>	Introduced	Annual	Short-lived graminoid	x	x	x	x
<i>Bromus tectorum</i>^c	Introduced	Annual	Short-lived graminoid	x	x	x	x
<i>Buchloe dactyloides</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Calochortus gunnisonii</i>	Native	Perennial	Long-lived forb				x
<i>Camelina microcarpa</i>	Introduced	Annual/Biennial	Short-lived forb	x		x	x
<i>Carduus nutans</i>^b	Introduced	Annual/Biennial	Short-lived forb	x	x	x	
<i>Carex brevior</i>	Native	Perennial	Long-lived graminoid	x		x	
<i>Carex inops</i> subsp. <i>heliophila</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Carex occidentalis</i>	Native	Perennial	Long-lived graminoid	x			
<i>Castilleja integra</i>	Native	Perennial	Long-lived forb		x		
<i>Celtis reticulata</i>	Native	Perennial	Shrub/tree	x		x	
<i>Centaurea diffusa</i>^b	Introduced	Biennial/Perennial	Variable forb			x	
<i>Cerastium arvense</i> var. <i>strictum</i>	Native	Perennial	Long-lived forb			x	

Table 2. Species listed by their presence (denoted by an "x") across treatments, or for native short-lived forbs, by the number of transects on which they occurred. UU = unburned, unsprayed; US = unburned, sprayed; BU = burned, unsprayed; BS = burned, sprayed. Transect replication per treatment follows treatment type. Species on Colorado's noxious weed list are denoted with superscripts (a = List A; b = List B; c = List C). Only taxa identified to the species level are included. We combined strictly annual, strictly biennial, and annual to biennial species into the grouping of "short-lived" for analysis. Species with variable longevity (can range from annual to biennial to perennial) were coded as "variable" for analysis. Other functional groups are self-explanatory. Nomenclature according to Ackerfield 2015.

Species	Native or Introduced	Longevity	Functional Group for Analysis	UU (N = 7)	US (N = 10)	BU (N = 5)	BS (N = 11)
<i>Cercocarpus montanus</i>	Native	Perennial	Shrub/tree			x	
<i>Chamaesyce glyptosperma</i>	Native	Perennial	Long-lived forb			x	
<i>Chamerion angustifolium</i>	Native	Perennial	Long-lived forb		x		
<i>Chenopodium cf. album</i>	Introduced	Annual	Short-lived forb		x	x	x
<i>Chenopodium cf. leptophyllum</i>	Native	Annual	Short-lived forb	0	2	3	1
<i>Cirsium arvense</i>^b	Introduced	Perennial	Long-lived forb			x	
<i>Cirsium undulatum</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Collinsia parviflora</i>	Native	Annual	Short-lived forb	0	0	1	0
<i>Comandra umbellata</i>	Native	Perennial	Long-lived forb				x
<i>Convolvulus arvensis</i>^c	Introduced	Perennial	Long-lived forb	x	x	x	x
<i>Coryphantha missouriensis</i>	Native	Perennial	Sub-shrub/shrub			x	x
<i>Dactylis glomerata</i>	Introduced	Perennial	Long-lived graminoid	x		x	
<i>Dalea purpurea</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Delphinium carolinianum</i> subsp. <i>virescens</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Dianthus armeria</i>	Introduced	Annual/Biennial	Short-lived forb	x			
<i>Dichanthelium oligosanthes</i> subsp. <i>scribnerianum</i>	Native	Perennial	Long-lived graminoid	x	x	x	
<i>Echinocereus viridiflorus</i>	Native	Perennial	Sub-shrub/shrub				x
<i>Ellisia nyctelea</i>	Native	Annual	Short-lived forb	0	0	0	1
<i>Elymus elymoides</i>	Native	Perennial	Long-lived graminoid	x	x		
<i>Elymus repens</i>^c	Introduced	Perennial	Long-lived graminoid				x
<i>Ericameria nauseosa</i>	Native	Perennial	Sub-shrub/shrub				x
<i>Erigeron flagellaris</i>	Native	Biennial	Short-lived forb	7	8	3	6
<i>Eriogonum alatum</i>	Native	Perennial	Long-lived forb				x
<i>Erodium cicutarium</i>^c	Introduced	Annual/Biennial	Short-lived forb	x	x	x	x
<i>Euphorbia dentata</i>	Native	Annual	Short-lived forb	1	7	3	2
<i>Euphorbia marginata</i>	Native	Annual	Short-lived forb	0	1	1	1
<i>Euphorbia spathulata</i>	Native	Annual	Short-lived forb	3	2	3	0
<i>Evolvulus nuttallianus</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Fallopia convolvulus</i>	Introduced	Annual	Short-lived forb	x	x	x	x
<i>Gaillardia aristata</i>	Native	Perennial	Long-lived forb	x		x	
<i>Galium aparine</i>	Native	Annual	Short-lived forb	1	1	2	2
<i>Geranium caespitosum</i>	Native	Perennial	Long-lived forb				x
<i>Glandularia bipinnatifida</i>	Native	Perennial	Long-lived forb	x			
<i>Grindelia squarrosa</i>	Native	Annual/Biennial/Perennial	Variable forb	x	x	x	

Table 2. Species listed by their presence (denoted by an "x") across treatments, or for native short-lived forbs, by the number of transects on which they occurred. UU = unburned, unsprayed; US = unburned, sprayed; BU = burned, unsprayed; BS = burned, sprayed. Transect replication per treatment follows treatment type. Species on Colorado's noxious weed list are denoted with superscripts (a = List A; b = List B; c = List C). Only taxa identified to the species level are included. We combined strictly annual, strictly biennial, and annual to biennial species into the grouping of "short-lived" for analysis. Species with variable longevity (can range from annual to biennial to perennial) were coded as "variable" for analysis. Other functional groups are self-explanatory. Nomenclature according to Ackerfield 2015.

Species	Native or Introduced	Longevity	Functional Group for Analysis	UU (N = 7)	US (N = 10)	BU (N = 5)	BS (N = 11)
<i>Gutierrezia sarothrae</i>	Native	Perennial	Sub-shrub/shrub	x	x		x
<i>Helianthus annuus</i>	Native	Annual	Short-lived forb	0	0	1	0
<i>Helianthus pumilus</i>	Native	Perennial	Long-lived forb				x
<i>Hesperostipa comata</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Heterotheca foliosa</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Heterotheca villosa</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Hordeum jubatum</i>	Native	Perennial	Long-lived graminoid			x	
<i>Hordeum pusillum</i>	Native	Annual	Short-lived graminoid	x			
<i>Hybanthus verticillatus</i>	Native	Perennial	Long-lived forb			x	x
<i>Hypericum perforatum</i>^c	Introduced	Perennial	Long-lived forb	x	x	x	x
<i>Juncus interior</i>	Native	Perennial	Long-lived graminoid	x		x	x
<i>Koeleria macrantha</i>	Native	Perennial	Long-lived graminoid	x		x	x
<i>Lactuca serriola</i>	Introduced	Annual/Biennial	Short-lived forb	x	x	x	x
<i>Lappula occidentalis</i>	Native	Annual/Biennial	Short-lived forb	0	0	1	2
<i>Lathyrus lanszwertii</i> var. <i>leucanthus</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Lepidium campestre</i>	Introduced	Annual	Short-lived forb	x	x	x	x
<i>Lepidium virginicum</i>	Native	Annual	Short-lived forb	2	0	1	0
<i>Leucocrinum montanum</i>	Native	Perennial	Long-lived forb				x
<i>Liatris punctata</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Linaria dalmatica</i>^b	Introduced	Perennial	Long-lived forb	x	x	x	x
<i>Linum lewisii</i>	Native	Perennial	Long-lived forb			x	x
<i>Lithospermum incisum</i>	Native	Perennial	Long-lived forb				
<i>Lomatium orientale</i>	Native	Perennial	Long-lived forb		x		x
<i>Malva neglecta</i>	Introduced	Annual/Biennial	Short-lived forb				x
<i>Marrubium vulgare</i>	Introduced	Perennial	Long-lived forb			x	x
<i>Medicago sativa</i>	Introduced	Perennial	Long-lived forb			x	x
<i>Mertensia lanceolata</i>	Native	Perennial	Long-lived forb	x			
<i>Mirabilis hirsuta</i>	Native	Perennial	Long-lived forb		x		x
<i>Mirabilis lanceolata</i>	Native	Perennial	Long-lived forb	x			
<i>Mirabilis linearis</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Nassella viridula</i>	Native	Perennial	Long-lived graminoid				x
<i>Nothocalais cuspidata</i>	Native	Perennial	Long-lived forb				x
<i>Oenothera serrulata</i> var. <i>serrulata</i>	Native	Perennial	Long-lived forb		x		
<i>Oenothera suffrutescens</i>	Native	Perennial	Long-lived forb	x	x	x	x

Table 2. Species listed by their presence (denoted by an "x") across treatments, or for native short-lived forbs, by the number of transects on which they occurred. UU = unburned, unsprayed; US = unburned, sprayed; BU = burned, unsprayed; BS = burned, sprayed. Transect replication per treatment follows treatment type. Species on Colorado's noxious weed list are denoted with superscripts (a = List A; b = List B; c = List C). Only taxa identified to the species level are included. We combined strictly annual, strictly biennial, and annual to biennial species into the grouping of "short-lived" for analysis. Species with variable longevity (can range from annual to biennial to perennial) were coded as "variable" for analysis. Other functional groups are self-explanatory. Nomenclature according to Ackerfield 2015.

Species	Native or Introduced	Longevity	Functional Group for Analysis	UU (N = 7)	US (N = 10)	BU (N = 5)	BS (N = 11)
<i>Onosmodium bejariense</i>	Native	Perennial	Long-lived forb	x	x		x
<i>Opuntia macrorhiza</i>	Native	Perennial	Sub-shrub/shrub		x		x
<i>Opuntia phaeacantha</i>	Native	Perennial	Sub-shrub/shrub	x	x	x	x
<i>Opuntia polyacantha</i>	Native	Perennial	Sub-shrub/shrub	x	x	x	x
<i>Oxalis dillenii</i>	Native	Perennial	Long-lived forb	x	x	x	
<i>Packera plattensis</i>	Native	Biennial/Perennial	Variable forb	x			
<i>Panicum virgatum</i>	Native	Perennial	Long-lived graminoid	x		x	
<i>Pascopyrum smithii</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Phacelia hastata</i>	Native	Perennial	Long-lived forb				x
<i>Phemeranthus parviflorus</i>	Native	Perennial	Long-lived forb	x	x		
<i>Phyla cuneifolia</i>	Native	Perennial	Long-lived forb	x			
<i>Physalis hederifolia</i> var. <i>comata</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Physalis heterophylla</i>	Native	Perennial	Long-lived forb			x	x
<i>Physalis longifolia</i>	Native	Perennial	Long-lived forb	x			x
<i>Plantago patagonica</i>	Native	Annual	Short-lived forb	1	0	1	0
<i>Poa compressa</i>	Introduced	Perennial	Long-lived graminoid	x	x	x	x
<i>Poa pratensis</i>	Introduced	Perennial	Long-lived graminoid	x	x	x	x
<i>Polygonum aviculare</i>	Introduced	Perennial	Long-lived graminoid			x	x
<i>Potentilla fissa</i>	Native	Perennial	Long-lived forb	x			
<i>Potentilla recta</i>^b	Introduced	Perennial	Long-lived forb	x	x	x	x
<i>Prunus virginiana</i>	Native	Perennial	Shrub/tree			x	x
<i>Pseudoroegneria spicata</i>	Native	Perennial	Long-lived graminoid	x			
<i>Psoralidium tenuiflorum</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Ratibida columnifera</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Rhus trilobata</i>	Native	Perennial	Shrub/tree	x	x	x	x
<i>Rosa blanda</i>	Native	Perennial	Sub-shrub/shrub	x	x	x	
<i>Rumex acetosella</i>	Introduced	Perennial	Long-lived forb	x		x	
<i>Rumex crispus</i>	Introduced	Perennial	Long-lived forb			x	x
<i>Salsola tragus</i>	Introduced	Annual	Short-lived forb	x	x		x
<i>Schedonorus arundinaceus</i>	Introduced	Perennial	Long-lived graminoid			x	x
<i>Scorzonera laciniata</i>	Introduced	Perennial	Long-lived forb	x	x		x
<i>Scutellaria brittonii</i>	Native	Perennial	Long-lived forb	x	x		
<i>Silene antirrhina</i>	Native	Annual	Short-lived forb	1	0	3	0
<i>Sisymbrium altissimum</i>	Introduced	Annual	Short-lived forb			x	x

Table 2. Species listed by their presence (denoted by an "x") across treatments, or for native short-lived forbs, by the number of transects on which they occurred. UU = unburned, unsprayed; US = unburned, sprayed; BU = burned, unsprayed; BS = burned, sprayed. Transect replication per treatment follows treatment type. Species on Colorado's noxious weed list are denoted with superscripts (a = List A; b = List B; c = List C). Only taxa identified to the species level are included. We combined strictly annual, strictly biennial, and annual to biennial species into the grouping of "short-lived" for analysis. Species with variable longevity (can range from annual to biennial to perennial) were coded as "variable" for analysis. Other functional groups are self-explanatory. Nomenclature according to Ackerfield 2015.

Species	Native or Introduced	Longevity	Functional Group for Analysis	UU (N = 7)	US (N = 10)	BU (N = 5)	BS (N = 11)
<i>Solidago missouriensis</i>	Native	Perennial	Long-lived forb				x
<i>Sphaeralcea coccinea</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Sporobolus cryptandrus</i>	Native	Perennial	Long-lived graminoid	x	x	x	x
<i>Stephanomeria pauciflora</i>	Native	Perennial	Long-lived forb				x
<i>Symphotrichum ericoides</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Symphotrichum porteri</i>	Native	Perennial	Long-lived forb	x	x	x	
<i>Taraxacum officinale</i>	Introduced	Perennial	Long-lived forb	x	x	x	x
<i>Thelesperma megapotamicum</i>	Native	Perennial	Long-lived forb				x
<i>Thlaspi arvense</i>	Introduced	Annual	Short-lived forb			x	x
<i>Tradescantia occidentalis</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Tragia ramosa</i>	Native	Perennial	Long-lived forb	x	x	x	x
<i>Tragopogon dubius</i>	Introduced	Annual/Biennial	Short-lived forb	x	x	x	x
<i>Trifolium pratense</i>	Introduced	Perennial	Long-lived forb			x	
<i>Triodanis leptocarpa</i>	Native	Annual	Short-lived forb	1	1	1	0
<i>Triodanis perfoliata</i>	Native	Annual	Short-lived forb	1	0	3	0
<i>Verbascum thapsus</i>^c	Introduced	Biennial	Short-lived forb	x		x	
<i>Verbena bracteata</i>	Native	Annual/Biennial/Perennial	Variable forb		x	x	
<i>Viola nuttallii</i>	Native	Perennial	Long-lived forb	x	x		x
<i>Vitis riparia</i>	Native	Perennial	Long-lived forb		x		
<i>Vulpia octoflora</i>	Native	Annual	Short-lived graminoid			x	
<i>Yucca glauca</i>	Native	Perennial	Sub-shrub/shrub	x	x	x	x

Table 3. Percent cover of the most abundant species (those with a total mean percent cover of at least 0.5%) broken out by over in each treatment type. UU = unburned, unsprayed; US = unburned, sprayed; BU = burned, unsprayed; BS = burned, sprayed.

Species	Origin	Mean (SE)	Mean (SE)	Mean (SE)	Mean (SE)	Mean (SE)
		Total Percent Cover	Percent Cover UU	Percent Cover US	Percent Cover BU	Percent Cover BS
<i>Convolvulus arvensis</i>	Introduced	7.2 (1.7)	1.4 (0.7)	5.8 (1.7)	2.8 (1.9)	14.1 (4.2)
<i>Pascopyrum smithii</i>	Native	5.7 (1.1)	1.4 (0.6)	2.3 (0.9)	5.4 (1.8)	11.1 (2.0)
<i>Psoralidium tenuiflorum</i>	Native	3.2 (0.78)	1.5 (0.7)	3.0 (0.8)	1.4 (0.9)	5.3 (2.0)
<i>Andropogon gerardii</i>	Native	3.2 (0.77)	4.0 (1.9)	5.1 (1.8)	3.5 (1.5)	0.8 (0.5)
<i>Poa compressa</i>	Introduced	1.9 (0.43)	2.2 (0.9)	1.6 (0.9)	2.3 (0.9)	1.9 (0.7)
<i>Bouteloua curtipendula</i>	Native	1.9 (0.35)	2.3 (0.8)	2.7 (0.7)	1.6 (0.8)	1.1 (0.5)
<i>Ambrosia psilostachya</i>	Native	1.9 (0.39)	2.6 (0.66)	2.7 (0.7)	1.6 (1.1)	0.8 (0.68)
<i>Rhus trilobata</i>	Native	1.6 (0.80)	4.8 (3.3)	2 (1.1)	0.1 (0.1)	0
<i>Symphyotrichum ericoides</i>	Native	1.5 (0.42)	1.6 (0.8)	1.8 (0.8)	0.7 (0.4)	1.6 (0.9)
<i>Erodium cicutarium</i>	Introduced	1.4 (0.40)	0.20 (0.14)	1.4 (1.0)	2.5 (1.27)	1.6 (0.44)
<i>Tragia ramosa</i>	Native	1.3 (0.32)	1.3 (0.6)	1.5 (0.5)	3.3 (1.4)	0.2 (0.2)
<i>Sphaeralcea coccinea</i>	Native	1.3 (0.39)	0.07 (0.07)	2.2 (0.9)	0	1.8 (0.8)
<i>Poa pratensis</i>	Introduced	1.2 (0.53)	4.1 (2.1)	0.5 (0.5)	0.02 (0.02)	0.4 (0.3)
<i>Buchloe dactyloides</i>	Native	1.1 (0.51)	0.6 (0.4)	1.3 (1)	0.1 (0.1)	1.7 (1.2)
<i>Alyssum simplex</i>	Introduced	1.05 (0.31)	2.7 (0.5)	0	3.0 (1.2)	0.1 (0.1)
<i>Bromus tectorum</i>	Introduced	1.02 (0.27)	2.4 (0.5)	0.2 (0.1)	2.9 (0.9)	0.1 (0.1)
<i>Carex inops subsp. heliophila</i>	Native	0.9 (0.27)	0.9 (0.7)	0.9 (0.4)	0	1.3 (0.5)
<i>Artemisia ludoviciana</i>	Native	0.8 (0.33)	1.4 (0.8)	0.9 (0.8)	0.2 (0.2)	0.6 (0.5)
<i>Tragopogon dubius</i>	Introduced	0.7 (0.19)	1.0 (0.3)	0.3 (0.1)	1.1 (0.4)	0.8 (0.5)
<i>Opuntia phaeacantha</i>	Native	0.7 (0.20)	1.4 (0.5)	0.9 (0.5)	0.3 (0.2)	0.1 (0.1)
<i>Bouteloua gracilis</i>	Native	0.6 (0.27)	0.04 (0.04)	0.9 (0.4)	0.05 (0.05)	1.1 (0.7)
<i>Bromus japonicus</i>	Introduced	0.5 (0.12)	0.9 (0.1)	0.2 (0.1)	1.8 (0.3)	0.08 (0.05)
<i>Oenothera suffrutescens</i>	Native	0.5 (0.19)	0.3 (0.2)	0.6 (0.3)	0.5 (0.5)	0.6 (0.4)

Table 4. Means and standard errors of richness and percent cover of long-lived forbs and grasses across treatment types and by native and introduced origin.

Origin	Functional Group	Mean (SE) Richness				Mean (SE) Percent Cover			
		BS	BU	US	UU	BS	BU	US	UU
Introduced	Long-lived forbs	1.9 (0.37)	1.4 (0.68)	1.1 (0.31)	1.43 (0.30)	15.8 (4.1)	6.5 (2.2)	6.3 (1.8)	2.9 (1.4)
Native	Long-lived forbs	5.5 (0.72)	6.2 (1.3)	5.9 (0.46)	7.6 (0.48)	12.6 (2.8)	10.6 (3)	16.3 (2.5)	11.6 (1.9)
Introduced	Long-lived graminoids	1.9 (0.34)	1.4 (0.68)	1.1 (0.31)	1.4 (0.30)	2.6 (0.87)	2.9 (0.99)	2.2 (1.3)	6.4 (2.3)
Native	Long-lived graminoids	5.5 (0.72)	6.2 (1.3)	5.9 (0.46)	7.6 (0.48)	18.5 (2.4)	13.2 (3.1)	14.9 (1.9)	10.7 (1.8)