THESIS

SOIL SEED BANK COMPOSITION AND IMPLICATIONS FOR ECOLOGICAL RESTORATION IN DEGRADED COLORADO SHRUBLANDS

Submitted by

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ABSTRACT

SOIL SEED BANK COMPOSITION AND IMPLICATIONS FOR ECOLOGICAL RESTORATION IN DEGRADED COLORADO SHRUBLANDS

Soil seed banks of shrub-dominated ecosystems in western North America are poorly understood. The potential of the soil seed bank – the species composition and abundance of seeds – to impact ecological restoration has rarely been considered in ecological restoration of shrublands and could influence management decisions. I analyzed the germinable soil seed bank composition and distribution in two high-conservation priority ecosystems in Colorado. Studies were carried out to characterize seed bank composition and relationship to aboveground vegetation in “undesirable” and “desirable” plant communities; determine if “shrub islands” influence seed bank distribution; and assess the landscape and vertical distribution of the seed bank in a *Bromus tectorum* L. (cheatgrass) invaded rangeland. For all seed bank studies, soil seed bank samples were collected to a depth of 5 cm and grown in greenhouse conditions to determine the species composition and abundance of germinable seeds. I found that seed bank species richness and Shannon-Wiener diversity (H) did not differ in either shrubland between undesirable sites dominated by non-native vegetation and desirable sites dominated by native vegetation. Total seed abundance in a montane sagebrush shrubland was significantly greater in desirable sites (1401 ± 165 seeds m^{-2}) compared to undesirable sites (588 ± 190 seed m^{-2}). In a salt desert shrubland of the Colorado Plateau, total seed abundance did not differ, but on average non-native species seeds made up more than 60% of the total seed bank in undesirable sites, compared to 40% in desirable sites. In a separate study, shrub islands across Colorado were not associated with increased seed

bank species richness or seed abundance compared to adjacent shrub-less interspaces. Differences in seed bank Shannon Wiener diversity (H) varied between shrublands, with salt desert shrublands having significantly greater ($p\text{-value} < 0.05$) seed bank diversity inside of shrub islands compared to shrub-less interspaces. Another study was conducted in a *Bromus tectorum* L. (cheatgrass) invaded rangeland of the Colorado Plateau to determine the seed bank horizontal and vertical distribution. The germinable soil seed bank had a greater abundance and lower spatial variability of native species seeds (3390 seeds m^{-2}, CV = 75%) than non-native species seeds (1880 seeds m^{-2}, CV: 124%) across the sampled landscape. Non-native species (primarily *Bromus tectorum* L.) seed were concentrated in the upper 2 cm soil (1294 ± 155, $p\text{-value} <0.0001$), but were found in substantive abundance in the 2 – 5 cm seed bank layer (585 ± 91).

In addition to seed bank studies, in the fall of 2018, I established a study in a montane shrubland to test the effectiveness of seeding a high diversity native seed mix (39 species, 1496 PLS m^{-2}) and treatments to increase site heterogeneity to increase native plant species diversity. One growing season following plot establishment, I found that plots that received a high diversity seed mix and those that received heterogeneity treatments had greater seeded species diversity (H) and richness than control plots.

ACKNOWLEDGEMENTS

This work would not have been possible without the help, guidance, patience, and support of many people. To the many unlisted faces and names, thank you.

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Second, I am incredibly grateful to the USDA Forest Service, the DOI Bureau of Land Management, and Shell for funding and to have had the opportunity to work in some of the most

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Finally, I can’t say how blessed I am to have the love and support of my family. There’s no way that I can express my gratitude to my parents, Carol and Aubrey, and sister, Lauren, for their encouragement to succeed and to do my best. Thank you, and I love you.

I am forever grateful to have had the opportunity to work in some of the most beautiful landscapes in Colorado – from the montane sagebrush parkland of California Park, to the shale hills of the salt desert – these public lands and the people who manage them are truly special. What follows is my small contribution to the knowledge to help manage and restore these amazing landscapes.

DEDICATION

This thesis is dedicated to the public land managers across The United States of America who work to manage lands for multiple uses and have the impossible task of making everyone happy all of the time.

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CHAPTER I INTRODUCTORY AND BACKGROUND INFORMATION

Introduction

Sagebrush and salt desert shrub-dominated ecosystems span millions of hectares in the western United States (West 1983, Blaisdell and Holmgren 1984, Chambers et al. 2017, Jonas et al. 2018, Winkler et al. 2018). These landscapes, often viewed as “barren” by the common observer, provide countless ecosystem goods and services such as forage for livestock, wildlife habitat, carbon sequestration capacity, outdoor recreation, and others (Hoover et al. 2019). Sagebrush (*Artemisia* spp. L.) and saltbush (*Atriplex* spp. L.) dominated systems are habitat for more than 350 vertebrate species, several are of high conservation priority, including sage-grouse (*Centrocercus* spp.) (Chambers et al. 2017). These ecosystems have experienced over-utilization of their resources through a variety of means (Kachergis et al. 2012, Chambers et al. 2017, Jonas et al. 2018, Hoover et al. 2019). Such over-utilization has caused plant community and ecosystem degradation and reduced or outright eliminated the capacity of large areas of these landscapes to perform their ecological functions (Coates et al. 2016, Chambers et al. 2017, Crist et al. 2019). In that vein, ecological restoration of sagebrush and salt desert shrublands has received increased attention in the past decade by researchers, federal agencies, and conservation organizations alike (Schlaepfer et al. 2014, Chambers et al. 2017, Grant-Hoffman et al. 2018, Jonas et al. 2018, Winkler et al. 2018).

A common hurdle to overcome during ecological restoration is dealing with plant communities and their propagules – the soil seed bank – that are, or are not, already there. The soil seed bank is defined as, “All living seeds in a soil profile, including those on the soil surface” (Gallagher 2014). Soil seed banks can have negative or positive impacts on ecological restoration sites and projects (Bossuyt and Honnay 2008, Faist et al. 2013, Rayburn et al. 2016). Many degraded shrublands have been historically planted to, or invaded by, various non-native plant species that form large soil seed banks such as *Agropyron cristatum* (L.) Gaertn. (crested wheatgrass), *Phleum pratense* L. (Timothy), *Bromus inermis* Leyss. (smooth brome), *Bromus tectorum* L. (cheatgrass), *Salsola tragus* L. (prickly Russian thistle), and *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass) (Marlette and Anderson 1986, Humphrey and Schupp 2001, Kachergis et al. 2012, 2013, Pekas and Schupp 2013, Sayre 2017, Grant-Hoffman et al. 2018, Jonas et al. 2018). These species' soil seed banks can emerge quickly and out-compete native plant seedlings for resources like water, light, and nutrients when they become available – as is often the case of restoration treatments (Humphrey and Schupp 2001, Bossuyt and Honnay 2008, Gioria and Pyšek 2015, Rayburn et al. 2016). On the other-hand, there may be a seed bank of desirable native plant species present at a degraded site that is not being expressed in the above-ground plant community (Faist et al. 2013, Gioria and Pyšek 2015, Vandvik et al. 2016). Such lack of expression could be due to competition, lack of resources, or the seed bank being buried too deeply (Chambers 2000, Benvenuti 2003, Burmeier et al. 2010).

Knowing the composition of the soil seed bank present – undesirable or desirable – can be a critical factor in the planning and success of an ecological restoration project. If an undesirable seed bank is present, with an abundance of highly competitive non-native species, then treatments could be used to deplete that seed bank. Numerous treatment methods for depleting the soil seed bank have been developed including solarization with plastic tarps for localized areas or broad-scale use of pre-emergent herbicides such as indaziflam, imazapic, chlorsfuron, sulfometuron methyl, etc. (Shinn and Thill 2004, Dear et al. 2006, Sebastian et al. 2016, 2017a). Such treatments could allow follow-up seeding of more desirable plant species to be more able to compete, establish, and persist (Clark et al. 2019). Whereas if a desirable seed bank is present, with an abundance of native plant species, then passive restoration may be a suitable option (Von Blanckenhagen and Poschlod 2005, Valkó et al. 2011, Cowan and Anderson 2014, Haussmann et al. 2019). If a desirable seed bank is not expressing itself in the aboveground vegetation, management actions could be used to express the seed bank and reduce costs by utilizing seed *in situ* (Von Blanckenhagen and Poschlod 2005, Bossuyt and Honnay 2008, Vandvik et al. 2016, Rayburn et al. 2016).

Both the U.S. Forest Service (USFS) and the Bureau of Land Management (BLM) working in Colorado shrublands seek to understand how to maximize the use of their limited resources for ecological restoration (Chambers et al. 2014c, 2017, Grant-Hoffman et al. 2018). These agencies have a shared goal of increasing native biodiversity, landscape heterogeneity, and ecological functionality (Fuhlendorf and Engle 2001, USDA Forest Service 2003, Grant-Hoffman et al. 2012, Ware et al. 2014, Chambers et al. 2017). The soil seed bank is a variable that can either add to, or detract from ecosystem restoration efforts, by being a source plant propagules – whether they be native species, non-native species, or some of both (Von Blanckenhagen and Poschlod 2005, Bossuyt and Honnay 2008, Rayburn et al. 2016). Soil seed banks and their implications for ecological restoration in arid and semi-arid shrub-dominated ecosystems have received little attention in peer-reviewed literature, relative to other ecosystems (Hopfensperger 2007, Bossuyt and Honnay 2008, Martin et al. 2012, Jonas et al. 2018). This research seeks to quantify and understand the composition of soil seed banks and their potential to influence plant community dynamics in degraded versus intact shrublands in Colorado. The purpose of this chapter is to provide pertinent background information on the soil seed bank and the ecosystems studied in this thesis, including their ecological characteristics and management history. Nomenclature for all plant species follows the USDA NRCS PLANTS Database (2019).

Soil Seed Bank Background

The soil seed bank – it's composition and similarity to the present vegetation – of shrublands in western North America is largely unknown (Hopfensperger 2007, Pekas and Schupp 2013, Gioria and Pyšek 2015, Barga and Leger 2018, Jonas et al. 2018, Haight et al. 2019). The soil seed bank can represent a memory of past vegetation that no longer exists on a site and represents the regenerative potential of species that reproduce via seed at a given site (Faist et al. 2013, Gioria and Pyšek 2015, Vandvik et al. 2016).

The two main classifications of seed banks are transient and persistent. Transient seed banks encompass seeds that rarely survive for more than one year. Persistent seed banks are those seeds that remain viable for more than one year and have the potential to have viable seed in the soil environment for decades, even centuries if conditions are right (Pakeman et al. 2012, Gallagher 2014, Gremer and Venable 2014, Schwartz-Lazaro and Copes 2019). Some plants produce both transient and persistent seed banks in order to respond to variable environmental conditions and respond to disturbance, known as bet hedging (Gremer & Venable 2014; Gallagher, 2014).

Seed bank success and persistence is often related to soil nitrate (Pakeman et al. 2012), ground cover (Barga and Leger 2018), and disturbance that promotes the expression of those seeds in the seed bank (Chambers and MacMahon 1994, Benech-Arnold et al. 2000). Soil pathogens, such as harmful fungi, can have negative impacts and accelerate the rate at which seeds become non-viable (Chambers and MacMahon 1994, Pakeman et al. 2012, Gallagher 2014). However, some species require particular mycorrhizae in order to germinate (Gallagher 2014). If such mycorrhizae are present, the survivability of seeds and seedlings is improved (Gallagher 2014).

Vegetation dynamics are crucial to the development and distribution of seed banks (Bossuyt and Honnay 2008, Pekas and Schupp 2013, Gioria and Pyšek 2015, Vandvik et al. 2016). Seed rain and the traits of plants to shed and disperse seeds drive the spatial distribution of seed banks (Chambers and MacMahon 1994, Gallagher 2014). Dense vegetation and litter layers can act as natural seed traps (Chambers 2000, Barga and Leger 2018, Filazzola et al. 2019). However, dense vegetation and litter cover can also prevent germination of some seeds that need mineral soil contact in order to germinate (Chambers and MacMahon 1994, Baskin and Baskin 2014, Gallagher 2014). "Soil seed banks show very high spatial heterogeneity" (Gallagher, 2014, pg. 272) that can cause sampling error and misrepresentation of the presence or absence of species in a landscape, depending upon the sampling scheme used (Coffin and Lauenroth 1989, Hopfensperger 2007, Bossuyt and Honnay 2008, Vandvik et al. 2016). Structural heterogeneity of aboveground vegetation and "nurse" plants (i.e. shrubs) seem to play a role in accumulating greater numbers of seeds relative to interspaces with little vegetation cover, especially in arid environments (Koniak and Everett 1982, Chambers and MacMahon 1994, Guo et al. 1998, Martyn et al. 2016, Barga and Leger 2018, Castillo-Escrivà et al. 2019).

Soil Seed Bank Dynamics and Similarity to Standing Vegetation

The species composition of the soil seed bank and its similarity to aboveground vegetation are important factors to understand the successional trajectory of a site following disturbance (Hopfensperger 2007, Bossuyt and Honnay 2008, Pekas and Schupp 2013, Rayburn et al. 2016, Haight et al. 2019). The stability of a plant community and frequency of disturbance have been found to influence the seed bank composition and similarity to standing vegetation (Coffin and Lauenroth 1989, Hopfensperger 2007, Bossuyt and Honnay 2008, Pekas and Schupp 2013, Barga and Leger 2018). Hopfensperger (2007) found that similarity between the seed bank and aboveground vegetation decreased with time since disturbance in forest and wetland ecosystems and increased with time since disturbance in grasslands. Relatively few studies have been conducted in shrublands to analyze the influence of disturbance and aboveground vegetation composition on the soil seed bank (Bossuyt and Honnay 2008, Vandvik et al. 2016).

Bossuyt & Honnay (2008) reviewed 102 seed bank studies of European plant communities conducted between 1990 and 2006. The review comprised four distinct vegetation types (grasslands, forests, marshes, and heathlands) and analyzed factors such as seed density, species richness, and similarity between seed bank and standing vegetation. Of these 102 studies, only eight were conducted in "heathlands" – a type of shrubland – in temperate oceanic and polar climate zones. No studies in this review were located in arid or semi-arid climates. This being said, they found that heathland seed banks have relatively low species richness (5 – 20 species), low Shannon-Wiener diversity (0.75 – 1.75), intermediate seed density with a mean seed abundance of ~6000 seeds m^{-2} and Jaccard similarity to aboveground vegetation ranging from 0.25 – 0.45. They found that, consistent with Hopfensperger (2007), species richness and diversity of the soil seed bank and similarity in seed bank and aboveground vegetation is associated with the stability of the ecosystem in question. Compared to grasslands and wetlands, heathlands (shrublands) and forests are generally more stable ecosystems and thus have greater dissimilarity.

Soil Seed Bank Implications for Ecological Restoration

The similarity of soil seed banks to standing vegetation and historical vegetation in shrubland environments may play an important role during restoration of disturbed areas. However, this role has primarily been found to be a negative one. Bossuyt & Honnay (2008) and Hopfensperger (2007) surmise that relying only on seed germination from the seed bank for restoration of a target community is not feasible. This is due to the seed bank, in most cases, being dominated by early successional species. Seed banks also tend to be dominated by a relatively low number of species and only few rare species tend to be found in the seed bank (Hopfensperger 2007, Gioria and Pyšek 2015, Vandvik et al. 2016). Complicating factors for restoration of shrub-dominated ecosystems is that late-successional woody species, rely mainly on spatial seed dispersal and not *in situ* germination, as their seeds are often transient (Bossuyt and Honnay 2008, Gallagher 2014). The review found that most authors agree that restoration based on the seed bank is likely only possible at sites that were degraded less than five years ago (Bossuyt and Honnay 2008).

Bekker et al. (1997) had a similar conclusion as Bossuyt & Honnay (2008) regarding the possible use of the soil seed bank in areas that had been intensively managed for agriculture and then had restoration implemented. They found that agricultural practices that improve sites from natural “nutrient poor conditions” may cause native species adapted to those nutrient poor conditions to disappear rapidly from both the standing vegetation and the seed bank (Bekker et al. 1997).

These studies suggest that the soil seed bank present in arid and semi-arid shrubland sites will likely only be able to play a small positive role in future restoration efforts. Degradation of most of the sites included in this study first occurred more than 50 years ago and the legacy impacts of these disturbances are still evident (West 1983, USDA Forest Service 2003, Stier 2012, Kachergis et al. 2014, Duniway et al. 2018, Jonas et al. 2018). Additionally, subsequent planting or invasion of exotic species after those historic disturbances will likely hinder restoration projects. Such exotic species are often early-successional and produce large amounts of seed that contribute significant propagule pressure (Marlette and Anderson 1986, Humphrey and Schupp 2001, Smith et al. 2008, Isselin-Nondedeu and Bédécarrats 2009, Jonas et al. 2018). The challenge will be to deplete the seed bank of the exotic species while at the same time incorporating early-, mid-, and late-successional native species back into the seed bank and aboveground vegetation (Barr et al. 2017, Sebastian et al. 2017a). My study will be able to address whether or not the seed bank may have a net positive or net negative influence on restoration management in these shrub-dominated plant communities.

Study Sites

My study sites present restoration challenges in terms of edaphic factors and land management history. One area is a Forest Service Special Interest Area (SIA) known as California Park. California Park is managed by the Medicine Bow-Routt National Forest in north-central Colorado. This high elevation sagebrush park is a biodiversity hotspot for both vegetation and wildlife (USDA Forest Service, 2003). My second study area is a region of salt desert shrublands in western Colorado, along the eastern edge of the Colorado Plateau. These areas of salt desert vegetation are managed by the Bureau of Land Management in and around the McInnis Canyon, Gunnison Gorge, and Dominguez-Escalante National Conservation Areas. Though in two very different climates (**Appendix Table 1**) and managed by different agencies, this research seeks to address the knowledge gap regarding the soil seed bank composition and potential to influence ecological restoration of these shrublands.

California Park Sagebrush Parkland

California Park is located in Routt County, Colorado on the Medicine Bow-Routt National Forest and is managed by the USDA Forest Service (USFS) Hahns Peak-Bears Ears Ranger District. California Park is an 11,000 hectare silver sagebrush (*Artemisia cana* Pursh) park tucked between the Elkhead Mountains and the Park Range about 60 km northwest of Steamboat Springs (**Figure 1.1**). The Forest Service classified this area as a Special Interest Area due to its rich diversity of plant and animal life, cultural heritage, and archeological and geologic significance

(USDA Forest Service 2003). Birds such as Greater Sandhill cranes (*Grus canadensis tabida*), Gunnison sage-grouse (*Centrocercus minimus*), Greater sage-grouse (*Centrocercus urophasianus*), and Columbian sharp-tailed grouse (*Tympanuchus phasianellus columbianus*) use the park's uplands to breed; Boreal toads (*Bufo boreas boreas*) and Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*) can be found along the riparian areas that feed into the Yampa River; and large herds of elk (*Cervus canadensis*), mule deer (*Odocoileus hemionus*), and pronghorn antelope (*Antilocapra americana*) forage throughout the park (USDA Forest Service, 2003). These same factors and resources have attracted people to California Park for millennia.

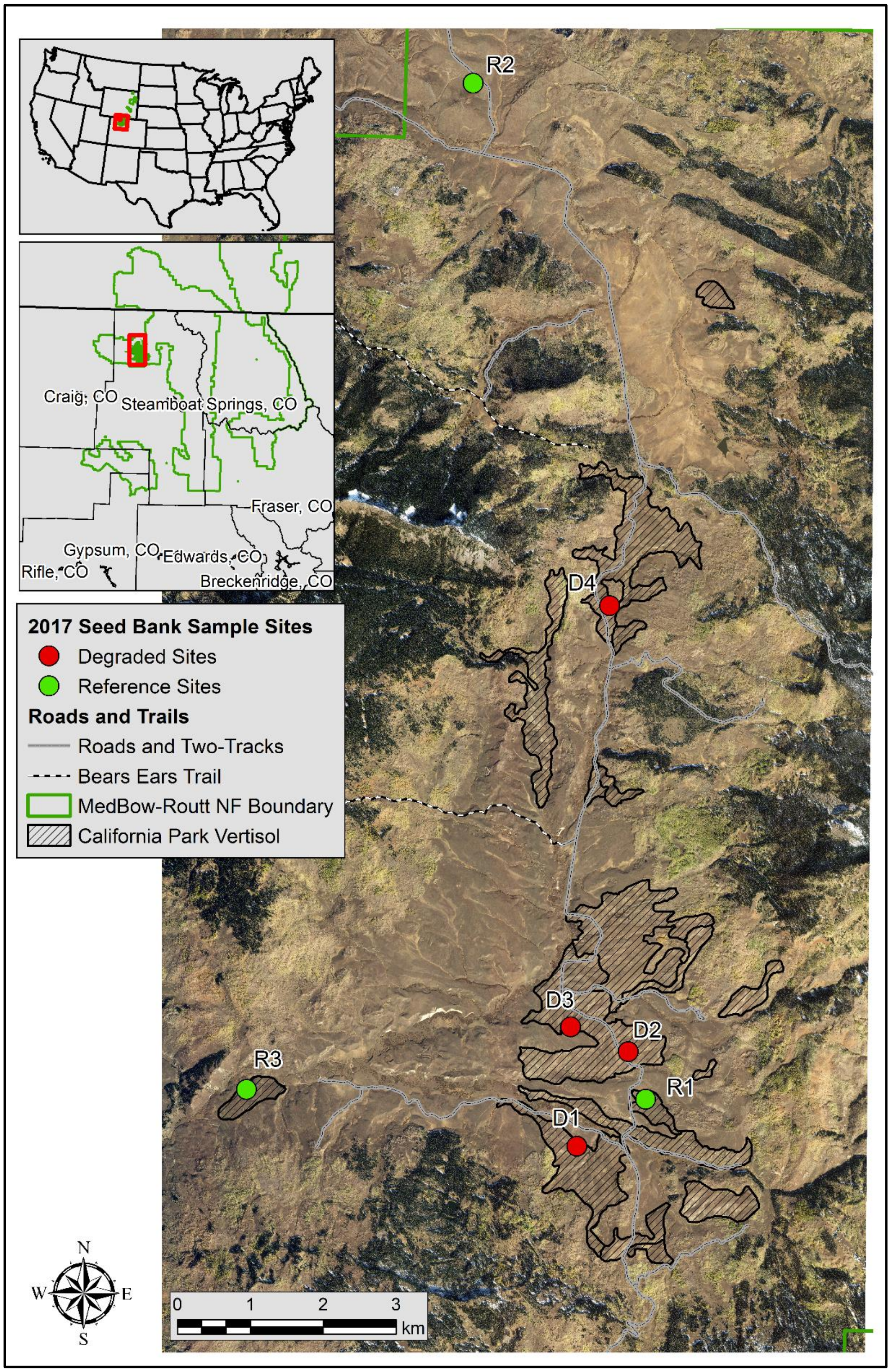


Figure 1.1: Map of California Park study area in northern Colorado. Desirable sites (R1 – R3) are plant communities that Forest Service resource managers consider to be in desirable states, with 15-35% sagebrush cover, diverse herbaceous understories, and little bareground. Undesirable sites (D1 – D4) are plant communities that resource managers consider to be in degraded, undesirable states with little native plant diversity and/or low plant cover. Aerial imagery from fall 2017 National Agricultural Imagery Program (USDA-FSA, 2017).

The land use history of California Park is extensive and includes Paleo-Indian hunter-gatherers, Ute peoples, cattleman, sheepherders, homesteaders, Forest Service personnel, and recreationists. To understand the interest in ecological restoration and soil seed bank research being done in California Park, some context is required as the land has a rich history of use and management.

The arrival of large cattle and sheep stock drives and the myth of superabundance in the late 1800's likely resulted in the over-use of California Park and the legacy impacts seen today (USDA Forest Service 2003, Kachergis et al. 2014). This high elevation park's lush summer range and abundant water from the surrounding Elkhead Mountains attracted cattlemen and sheepherders. California Park was also part of a stock drive route known as the "Beef Trail" which was used to move large herds of livestock to railyards in northern Colorado and southern Wyoming. Beginning around 1871 and continuing until the establishment of the Taylor Grazing Act of 1934, heavy grazing and trampling by tens of thousands of livestock occurred (USDA Forest Service 2003; Kachergis et al. 2011, 2014; John Sundberg, personal communication). This overuse denuded the vegetation, compacted the heavy clay soils, and altered the ecology of California Park (USDA Forest Service 2003, Kachergis et al. 2012).

The geomorphology, landforms, and associated soils of this SIA are highly variable throughout the relatively small area of the park (~11,000 hectares) and present unique challenges. The dominant geology throughout California Park includes landslide deposits and residuum from sedimentary rocks including the Lance Formation and Lewis shale. The soils derived from Lewis shale (a marine shale) tend to pose the greatest management concerns as the fine-grained material is highly erodible and susceptible to landslides and earth flows (USDA Forest Service 2003). The mineralogy of soils derived from Lewis shale is dominated by smectitic clays that can exhibit shrink-swell cracking and are classified as Vertisols (Soil Survey Staff, 2006). Historical overuse and denudation of vegetation caused soil compaction of these clay soils and exacerbated shrink-swell cracking due to the lack of ground cover (USDA Forest Service 2003).

To address the resource concerns that resulted from the large stock drives and heavy grazing, natural resource managers have worked to revegetate and improve range conditions throughout the park (USDA Forest Service 2003). Objectives and approaches of range improvement have changed over time, but in the mid-20th century had generally focused on attempting to revegetate bare areas, reduce soil erosion, and increase forage production (USDA Forest Service 2003). One method used in the early- to mid-20th century was the planting of non-native pasture grass species such as *Bromus inermis* Leyss. (smooth brome), *Phleum pratense* L. (Timothy), *Poa pratensis* L. (Kentucky bluegrass), and *Dactylis gomerata* L. (orchardgrass) (Kachergis et al. 2014, Sayre 2017, Grant-Hoffman et al. 2018). From the 1950's through the 1980's, the broadleaf herbicide 2-4-D was sprayed over large areas of California Park (USDA Forest Service 2003). The herbicide 2-4-5-T (Agent Orange) was also used in the 1950's (USDA Forest Service 2003). These herbicides were used to reduce the cover of the increasing *Artemisia tridentata* Nutt. (big sagebrush) and *Artemisia cana* Pursh (silver sagebrush) and patches of *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) a native forb that became dominant after heavy grazing (USDA Forest Service 2003, Kachergis et al. 2012, 2014). These management activities were largely effective in what they set out to do, but resource objectives have since changed (USDA Forest Service 2003, Chambers et al. 2017).

Resource objectives in California Park have shifted towards increasing wildlife habitat quality and reducing sedimentation into the Elkhead Creek drainage. Large areas of the uplands were (and still are) dominated by non-native pasture grasses like *Phleum pratense* L. (Timothy) and *Bromus inermis* Leyss. (smooth brome), with few forbs and shrubs to support upland birds (Crawford et al. 2004, Kachergis et al. 2012, 2014, Bates et al. 2017). Other sloping uplands are dominated by bare ground with sparse vegetation that frequently slumps or dominated by a native forb – *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) – that has created large "monocultures" (USDA Forest Service 2003, Kachergis et al. 2014). These high sloping areas with relatively little vegetation or vegetation that senesces rapidly (like *Wyethia*) can cause sedimentation into the Elkhead Creek that flows into the Yampa River (USDA Forest Service 2003, USDA Forest Service personnel, personal communication).

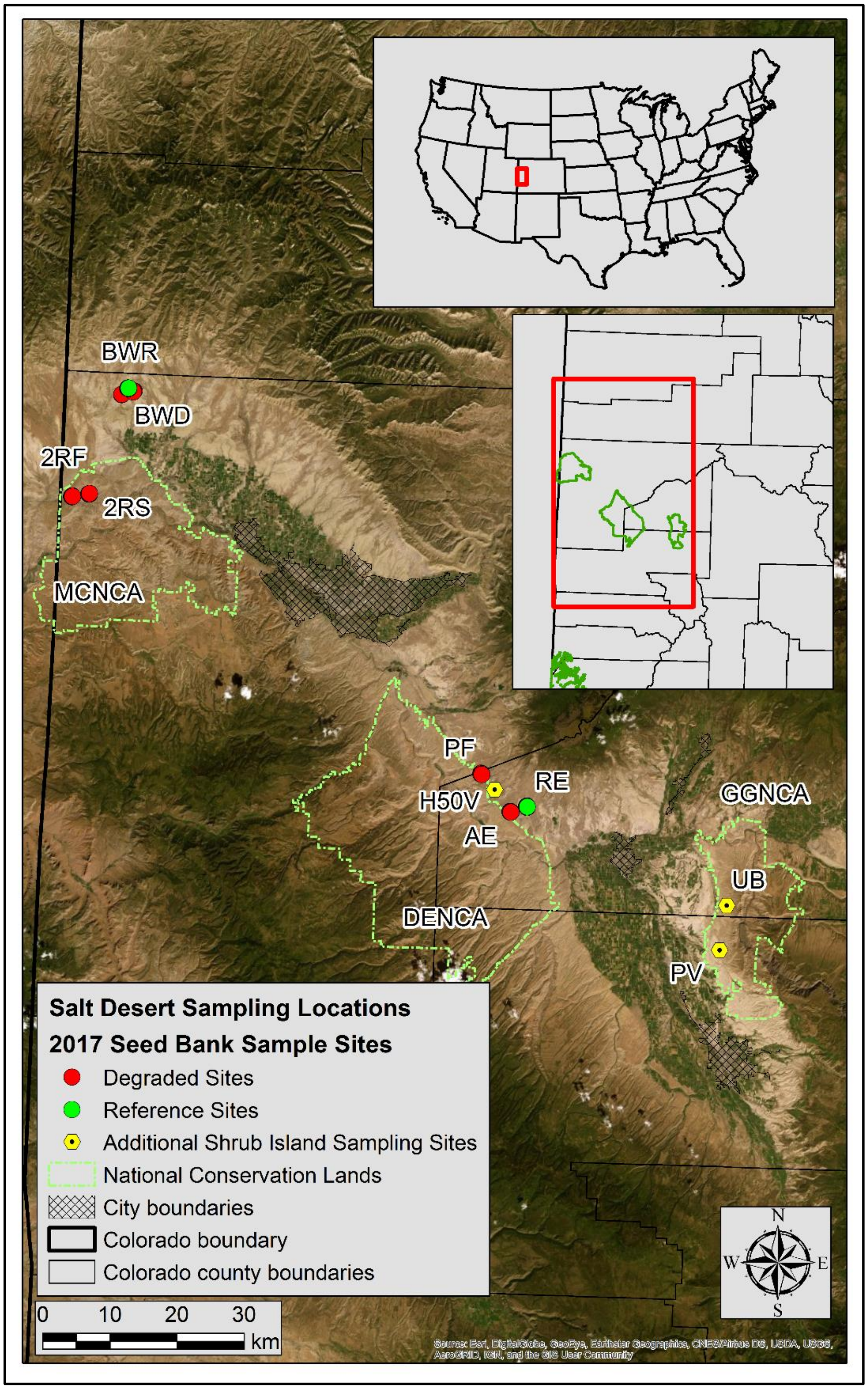
Kachergis et al. (2014) developed a state-and-transition model (STM) for the silver sagebrush shrublands found in California Park. The model describes three potential states and the associated transitions between states. Two of these states are "degraded" states the Forest Service is concerned with and are working to manage and restore. The state considered desirable by USFS resource managers (the reference state) includes two potential communities – one dominated by silver sagebrush with native herbaceous understory and the other described as a native grassland. The other two states are considered undesirable by USFS and include one with an overstory of *Artemisia cana* Pursh (silver sagebrush) and *Phleum pratense* L. (Timothy) in the understory, and another dominated by *Wyethia amplexicaulis* (Nutt). Nutt. (mule-ears). Kachergis found that the state transitions from the desirable state to the undesirable *Phleum*- and *Wyethia*- dominated states have been influenced by gradual increases in *Phleum pratense* L.(Timothy) and overgrazing, respectively. One state transition from the *Wyethia*-dominated state back to the native grassland community was identified and resulted from spraying broadleaf-specific herbicides that killed *Wyethia*.

A third type of degraded state are areas termed “scabs” or “mobile real-estate” (USDA Forest Service, 2003; USDA Forest Service personnel, personal communication). These areas are primarily on the Jokodowski soil series (Fine, smectitic Typic Humicryerts) and are dominated by bare ground or undesirable short-lived plants like *Madia glomerata* Hook. (mountain tarweed – native annual forb), *Collomia linearis* Nutt. (tiny trumpet – native annual forb), and *Lomatium leptocarpum* (Torr. & A. Gray) J.M. Coult. & Rose (gumbo-lomatium – native perennial forb). These sloping areas of shrink-swell clay soils (Vertisols) turn to “gumbo” when wet and frequently slump (Soil Survey Staff, 2006; John Sundberg, personal communication). Despite revegetation attempts, these areas do not easily support perennial vegetation (USDA Forest Service 2003; USDA Forest Service personnel, personal communication).

Starting in 2000, Forest Service resource managers have used numerous restoration methods to try to increase cover of native grasses, forbs, and sagebrush on degraded areas in California Park. Some techniques have included plowing, sub-soil ripping, direct seeding using a seed drill, fertilization, herbicide application, and limited grazing exclusion (USDA Forest Service 2003). Despite these efforts, relatively little success of increasing plant species richness has been seen in the uplands over the past two decades (USDA Forest Service personnel, personal communication).

Salt Desert Shrublands

The second shrub-dominated ecosystem that is part of my studies is the salt desert shrubland of the Colorado Plateau in western Colorado. Large areas of this ecosystem are owned and managed by the U.S. Department of the Interior Bureau of Land Management (BLM) (Blaisdell and Holmgren 1984, Sleeter et al. 2012, Stier 2012, Jonas et al. 2018). The sites being studied are located throughout the Grand Valley in Delta, Mesa, and Montrose counties and are in and around several National Conservations Areas (NCAs) (**Figure 1.2**). These NCAs include Dominguez Escalante NCA (DENCA), Gunnison Gorge NCA (GGNCA), and McInnis Canyon NCA (MCNCA).



Source: Esri, DigitalGlobe, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AeroGRID, IGN, and the GIS User Community

Figure 1.2: Map of Salt Desert study area in the Grand Valley of western Colorado. Undesirable sites include Two Road Fire (2RF), Two Road Seeded (2RS), Alkali Exclosure (AE), Badger Wash Degraded (BWD), and Peeples Fire (PF). Desirable sites include Badger Wash Reference (BWR) and Relic Exclosure (RE). Additional sites sampled in 2019 include the Highway 50 Valley (H50V), Peach Valley (PV), and Ute Bedground (UB).

These National Conservation Lands are classified as such due to their rich diversity in natural and cultural resources. Numerous migratory birds, including golden eagles (*Aquila chrysaetos*), western snowy plover (*Charadrius alexandrines nivosus*), gray vireo (*Vireo vicinoir*) utilize these habitats and a portion of the McInnis NCA is a designated Audubon Important Bird Area (Grant-Hoffman et al. 2012). Large herds of pronghorn antelope (*Antilocapra americana*) use salt desert shrublands in the Grand Valley as winter range and winter concentration areas. Other animal species of interest include white-tailed prairie dogs (*Cynomys leucurus*), burrowing owls (*Athene cunicularia*), kit fox (*Vulpes macrotis*), and several bat species (Grant-Hoffman et al. 2012). The Colorado Plateau is regarded as a "center for endemic plants" (Welsh 1978) and more than 340 individual taxa have been found throughout the region (Welsh 1978, Winkler et al. 2018). Such ecological diversity has attracted people to this harsh landscape for millennia. Cultural heritage and archaeological sites, and paleontological resources abound throughout these NCAs and are important legacies that the BLM works to preserve and protect (Grant-Hoffman et al. 2012).

Jonas et al. (2018) provides a thorough review of the biotic, abiotic, and management conditions of salt desert shrublands and the history of ecological restoration attempts. To summarize, salt deserts in the Intermountain West occur in arid climates, with highly variable annual precipitation, ranging from 100 to 350 mm. Soils tend to be underlain by Mancos shale, a marine shale that has a high concentration of mineral salts, resulting in saline or alkaline soil conditions (Lusby et al. 1963, Duniway et al. 2018, Jonas et al. 2018). Plant productivity is low and biological soil crusts play an important role in soil stabilization (West 1983, Blaisdell and Holmgren 1984, Faist et al. 2017, Duniway et al. 2018, Jonas et al. 2018). Desirable salt desert shrubland plant communities are typically characterized by slow-growing *Atriplex* spp. L.

(saltbush) shrubs, perennial grasses, and biological soil crusts (BSC) in interspaces, with ≤ 20 % plant canopy cover (Duniway et al. 2018, Jonas et al. 2018). Perennial forbs and native annuals tend to make up lesser component of the plant communities, but can be locally abundant when soil moisture is high, especially some *Eriogonum* spp. Michx. (buckwheat) and *Calochortus* spp. Pursh (mariposa lily) (West 1983, Blaisdell and Holmgren 1984, Jonas et al. 2018).

Numerous land-uses in the salt desert have caused significant perturbations and changes to ecosystem function (Lusby et al. 1963, Lusby 1979, Blaisdell and Holmgren 1984, Sleeter et al. 2012, Jonas et al. 2018). Widespread and heavy grazing by cattle and sheep began in the 1880's and continued through the Taylor Grazing Act of 1934 (Blaisdell and Holmgren 1984, Duniway et al. 2018). Heavy grazing broke-up BSCs and caused shifts in plant community composition and production (Blaisdell and Holmgren 1984, Jonas et al. 2018). More recently, human development in the form of energy and transportation corridors, surface mining, military operations, recreation, and off-road vehicle use has caused severe soil disturbance, further destroying fragile BSCs and transported non-native species propagules (Blaisdell and Holmgren 1984, Stier 2012, Jonas et al. 2018).

These land uses and soil disturbances have caused vast areas of salt desert shrublands to be susceptible to invasion by non-native annual species, especially *Bromus tectorum* L. (cheatgrass), *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass), and *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover) (West 1983; Schwinning et al. 2005; Grant-Hoffman et al. 2018; Jonas et al. 2018; Duniway et al. 2018). Annual grass invasion has led to significant alterations of the normal fire return interval, from >80 years to <10 years (West 1983, Blaisdell and Holmgren 1984, Jonas et al. 2018). The change in fire-return interval and other factors has caused state-transitions where weedy non-native annual species are dominant and native shrubs are unable to re-establish (Chambers et al. 2014b, Grant-Hoffman et al. 2018, Jonas et al. 2018).

Attempts to restore salt desert shrubland communities following wildfire, overgrazing, and soil disturbance have occurred since the 1950's, but little success has been observed (Grant-Hoffman et al. 2018, Jonas et al. 2018). Active ecological restoration of these communities is thought to be limited by the inherently stressful climatic and edaphic conditions, aggressive non-native annual species, and failure of seeded or transplanted species to establish (Jonas et al. 2018). A few soil seed bank studies have taken place in ecosystems in the Colorado Plateau (Koniak and Everett 1982, Haight et al. 2019), but no studies were found that have taken place in salt desert shrublands.

Research Questions

Soil seed banks represent a memory of past and present vegetation, and regulate the regenerative potential of species reproducing by seed (Gioria and Pyšek 2015). A seed bank dominated by native species may allow for passive restoration and reduced restoration costs, while a seed bank dominated by non-native invasive species may pose a significant limitation to ecological restoration activities (Von Blanckenhagen and Poschlod 2005, Bossuyt and Honnay 2008, Chambers et al. 2014c, Sebastian et al. 2017a, Jonas et al. 2018). My main research question on soil seed banks of shrub-dominated ecosystems is: **What are the compositional differences of soil seed banks in intact versus degraded areas, and what is the potential of these seed banks to hinder ecological restoration or contribute to passive ecological restoration?**

There is a lack of literature on the soil seed bank composition in shrub-dominated ecosystems throughout the world, especially in semi-arid and arid climates (Hopfensperger 2007, Bossuyt and Honnay 2008, Martin et al. 2012, Jonas et al. 2018). Only recently has this gap started to be addressed (Pekas and Schupp 2013, Martyn et al. 2016, Barga and Leger 2018, Haight et al.

2019, Hu et al. 2019). My research in the silver sagebrush parkland of California Park and the salt desert shrublands of western Colorado will help to further address this knowledge gap in soil seed bank composition of rangelands. Additionally, I hope that by shedding light on the seed bank composition, I will be able to help inform federal land management agencies how to go about restoration management while taking the soil seed bank into consideration.

REFERENCES

Barga, S., and E. A. Leger. 2018. Shrub cover and fire history predict seed bank composition in Great Basin shrublands. Journal of Arid Environments 154:40–50.

Barr, S., J. L. Jonas, and M. W. Paschke. 2017. Optimizing seed mixture diversity and seeding rates for grassland restoration. Restoration Ecology 25:396–404.

Baskin, C. C., and J. M. Baskin. 2014. Germination ecology of seeds with nondeep physiological dormancy. Pages 79–117 Seeds.

Bates, J. D., K. W. Davies, A. Hulet, R. F. Miller, and B. Roundy. 2017. Sage grouse groceries: Forb response to piñon-juniper treatments. Rangeland Ecology and Management 70:106–115.

Bekker, R. M. R., G. L. G. Verweij, R. E. N. Smith, R. Reine, J. P. Bakker, and S. Schneider. 1997. Soil seed banks in European grasslands: does land use affect regeneration perspectives? Journal of Applied Ecology 34:1293–1310.

Benech-Arnold, R. L., R. A. Sanchez, F. Forcella, B. C. Kruk, and C. M. Ghersa. 2000. Environmental controls of dormancy in weed seed bank soils. Field Crops Research 67:105–122.

Benvenuti, S. 2003. Soil texture involvement in germination and emergence of buried weed seeds. Agronomy Journal 95:191–198.

Blaisdell, J. P., and R. C. Holmgren. 1984. Managing intermountain rangelands - Salt-desert shrub ranges. General Technical Report - US Department of Agriculture, Forest Service.

Von Blanckenhagen, B., and P. Poschlod. 2005. Restoration of calcareous grasslands: the role of the soil seed bank and seed dispersal for recolonisation processes. Biotechnol. Agron. Soc. Environ 9:143–149.

Bossuyt, B., and O. Honnay. 2008. Can the seed bank be used for ecological restoration? An overview of seed bank characteristics in European communities. Journal of Vegetation Science 19:875–884.

Burmeier, S., R. L. Eckstein, A. Otte, and T. W. Donath. 2010. Desiccation cracks act as natural seed traps in flood-meadow systems. Plant and Soil 333:351–364.

Castillo-Escrivà, A., G. M. López-Iborra, J. Cortina, and J. Tormo. 2019. The use of branch piles to assist in the restoration of degraded semiarid steppes. Restoration Ecology 27:102–108.

Chambers, J. C. 2000. Seed movements and seedling fates in disturbed sagebrush steppe ecosystems: Implications for restoration. Ecological Applications 10:1400–1413.

Chambers, J. C., J. L. Beck, J. B. Bradford, J. Bybee, S. Campbell, J. Carlson, T. J. Christiansen, K. J. Clause, G. Collins, M. R. Crist, J. B. Dinkins, K. E. Doherty, F. Edwards, S. Espinosa, K. A. Griffin, P. Griffin, J. R. Haas, S. E. Hanser, D. W. Havlina, K. F. Henke, J. D. Hennig, L. A. Joyce, F. F. Kilkenny, S. M. Kulpa, L. L. Kurth, J. D. Maestas, M. Manning, K. E.

Mayer, B. A. Mealor, C. McCarthy, M. Pellant, M. A. Perea, K. L. Prentice, D. A. Pyke, L. A. Wiechman, and A. Wuenschel. 2017. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 1. Science basis and applications. USDA Forest Service - General Technical Report RMRS-GTR.

Chambers, J. C., and J. A. MacMahon. 1994. A day in the life of a seed: Movements and fates of seeds and their implications for natural and managed systems. Annual Review of Ecology and Systematics 25:263–292.

Chambers, J. C., R. F. Miller, D. I. Board, D. A. Pyke, B. A. Roundy, J. B. Grace, E. W. Schupp, and R. J. Tausch. 2014a. Resilience and resistance of sagebrush ecosystems: Implications for state and transition models and management treatments. Rangeland Ecology and Management 67:440–454.

Chambers, J. C., D. A. Pyke, J. D. Maestas, M. Pellant, C. S. Boyd, S. B. Campbell, S. Espinosa, D. W. Havlina, and K. E. Mayer. 2014b. Using resistance and resilience concepts to reduce impacts of invasive annual grasses and altered fire regimes on the sagebrush ecosystem and greater sage-grouse: a strategic multi-scale approach. Gen. Tech. Rep. RMRS-GTR-326. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.

Clark, S. L., D. J. Sebastian, S. J. Nissen, and J. R. Sebastian. 2019. Effect of indaziflam on native species in natural areas and rangeland. Invasive Plant Science and Management 12:60–67.

Coates, P. S., K. E. Doherty, T. Kroger, E. J. Blomberg, C. A. Hagen, and M. L. Casazza. 2016. Wildfire, climate, and invasive grass interactions negatively impact an indicator species by reshaping sagebrush ecosystems. Proceedings of the National Academy of Sciences of the United States of America 113.

Coffin, D. P., and W. K. Lauenroth. 1989. Spatial and temporal variation in the seed bank of a semiarid grassland. American Journal of Botany 76:53–58.

Cowan, O. S., and P. M. L. Anderson. 2014. The peninsula shale renosterveld of devil's peak, western cape: A study into the vegetation and seedbank with a view toward potential restoration. South African Journal of Botany 95:135–145.

Crawford, J. A., R. A. Olson, N. E. West, J. C. Mosley, M. A. Schroeder, T. O. M. D. Whitson, R. F. Miller, M. A. Gregg, C. S. Boyd, J. A. 2004. Ecology and management of sage-grouse and sage-grouse habitat. Journal of Range Management 57:2–19.

Crist, M. R., J. C. Chambers, S. L. Phillips, K. L. Prentice, and L. A. Wiechman. 2019. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 2 . Management applications. General Technical Report RMRS-GTR-389. Fort Collins, CO.

Dear, B. S., G. A. Sandral, and B. C. D. Wilson. 2006. Tolerance of perennial pasture grass seedlings to pre- and post-emergent grass herbicides. Australian Journal of Experimental Agriculture 46:637–644.

Duniway, M. C., E. L. Geiger, T. J. Minnick, S. L. Phillips, and J. Belnap. 2018. Insights from long-term ungrazed and grazed watersheds in a salt desert Colorado Plateau ecosystem. Rangeland Ecology & Management 71:492–505.

Faist, A. M., S. Ferrenberg, and S. K. Collinge. 2013. Banking on the past: Seed banks as a reservoir for rare and native species in restored vernal pools. AoB PLANTS 5:1–11.

Faist, A. M., J. E. Herrick, J. Belnap, J. W. V. Zee, and N. N. Barger. 2017. Biological soil crust and disturbance controls on surface hydrology in a semi-arid ecosystem. Ecosphere 8.

Filazzola, A., A. R. Liczner, M. Westphal, and C. J. Lortie. 2019. Shrubs indirectly increase desert seedbanks through facilitation of the plant community. PLoS ONE 14:1–15.

Fuhlendorf, S. D., and D. M. Engle. 2001. Restoring heterogeneity on rangelands: Ecosystem management based on evolutionary grazing patterns. BioScience 51:625.

Gallagher, R. S. 2014. Seeds: The ecology of regeneration in plant communities. (R. S. Gallagher, Ed.). CABI, Wallingford.

Gioria, M., and P. Pyšek. 2015. The legacy of plant invasions: changes in the soil seed bank of invaded plant communities. BioScience 66:40–53.

Grant-Hoffman, M. N., A. Lincoln, and J. Dollerschell. 2018. Post-fire native seed use in Western Colorado: A look at burned and unburned vegetation communities. Natural Areas Journal 38:286–297.

Grant-Hoffman, M. N., K. A. Stevens, M. H. DeChandenedes, and M. Preseton. 2012. Science plan for McInnis Canyon National Conservation Area. Science Plan for the McInnis Canyon National Conservation Area. Washington, D.C.

Gremer, J. R., and D. L. Venable. 2014. Bet hedging in desert winter annual plants: Optimal germination strategies in a variable environment. Ecology Letters 17:380–387.

Guo, Q., P. W. Rundel, and D. W. Goodall. 1998. Horizontal and vertical distribution of desert seed banks: Patterns, causes, and implications. Journal of Arid Environments 38:465–478.

Haight, J. D., S. C. Reed, and A. M. Faist. 2019. Seed bank community and soil texture relationships in a cold desert. Journal of Arid Environments 164:46–52.

Haussmann, N. S., C. Delport, V. Kakembo, K. K. Mashiane, and P. C. le Roux. 2019. Restoration potential of invaded abandoned agricultural fields: What does the seed bank tell us? Restoration Ecology 27:813–820.

Hoover, D. L., B. Bestelmeyer, N. B. Grimm, T. E. Huxman, S. C. Reed, O. Sala, T. R. Seastedt, H. Wilmer, and S. Ferrenberg. 2019. Traversing the wasteland: A framework for assessing ecological threats to drylands. BioScience XX:1–13.

Hopfensperger, K. N. 2007. A review of similarity between seed bank and standing vegetation across ecosystems. Oikos 116:1438–1448.

Hu, A., J. Zhang, X. Chen, S. Chang, and F. Hou. 2019. Winter grazing and rainfall synergistically affect soil seed bank in semiarid area. Rangeland Ecology and Management 72:160–167.

Humphrey, L. D., and E. W. Schupp. 2001. Seed banks of Bromus tectorum-dominated communities in the Great Basin. Western North American Naturalist 61:85–92.

Isselin-Nondedeu, F., and A. Bédécarrats. 2009. Assessing the dominance of Phleum pvatense cv. climax, a species commonly used for ski trail restoration. Applied Vegetation Science 12:155–165.

Jonas, J. L., M. N. Grant-Hoffman, and M. W. Paschke. 2018. Restoration of North American salt deserts: A look at the past and suggestions for the future. Ecological Restoration 36:177–194.

Kachergis, E., M. E. Fernandez-Gimenez, and M. E. Rocca. 2012. Differences in plant species composition as evidence of alternate states in the sagebrush steppe. Rangeland Ecology & Management 65:486–497.

Kachergis, E. J., C. N. Knapp, M. E. Fernandez-Gimenez, J. P. Ritten, J. G. Pritchett, J. Parsons, W. Hibbs, and R. Roath. 2013. Tools for resilience management: Multidisciplinary development of state-and-transition models for northwest Colorado. Ecology and Society 18.

Kachergis, E., M. E. Rocca, and M. E. Fernandez-Gimenez. 2011. Indicators of ecosystem function identify alternate states in the sagebrush steppe. Ecological Applications 21:2781–2792.

Kachergis, E., M. E. Rocca, and M. E. Fernández-Giménez. 2014. Long-term vegetation change provides evidence for alternate states in silver sagebrush. Rangeland Ecology & Management 67:183–194.

Koniak, S., and R. L. Everett. 1982. Seed reserves in soils of successional stages of pinyon woodlands. The American Midland Naturalist 108:295–303.

Lusby, G. C. 1979. Effects of grazing on runoff and sediment yield from desert rangeland at Badger Wash in western Colorado, 1953-73. Geological Survey Water-Supply Paper 1532-1. Was.

Lusby, G. C., G. T. Turner, J. R. Thompson, and V. H. Reid. 1963. Hydrologic and biotic characteristics of grazed and ungrazed and watersheds of the Badger Wash basin in western Colorado, 1953-58. Geological Survey Water-Supply Paper 1532-B.

Marlette, G. M., and J. E. Anderson. 1986. Seed banks and propagule dispersal in crested-wheatgrass stands. Journal of Applied Ecology 23:161–175.

Martin, L. J., B. Blossey, and E. Ellis. 2012. Mapping where ecologists work: Biases in the global distribution of terrestrial ecological observations. Frontiers in Ecology and the Environment 10:195–201.

Martyn, T. E., J. B. Bradford, D. R. Schlaepfer, I. C. Burke, and W. K. Lauenroth. 2016. Seed bank and big sagebrush plant community composition in a range margin for big sagebrush. Ecosphere 7:1–11.

Pakeman, R. J., J. L. Small, and L. Torvell. 2012. Edaphic factors influence the longevity of seeds in the soil. Plant Ecology 213:57–65.

Pekas, K. M., and E. W. Schupp. 2013. Influence of aboveground vegetation on seed bank composition and distribution in a Great Basin Desert sagebrush community. Journal of Arid Environments 88:113–120.

Rayburn, A. P., C. Schriefer, A. Zamora, and E. A. Laca. 2016. Seedbank-vegetation relationships in restored and degraded annual California grasslands: Implications for restoration. Ecological Restoration 34:277–284.

Sayre, N. F. 2017. The Politics of Scale: A History of Rangeland Science. The University of Chicago Press, Chicago.

Schlaepfer, D. R., W. K. Lauenroth, and J. B. Bradford. 2014. Natural regeneration processes in big sagebrush (Artemisia tridentata). Rangeland Ecology and Management 67.

Schwartz-Lazaro, L. M., and J. T. Copes. 2019. A review of the soil seedbank from a weed scientists perspective. Agronomy 9:1–13.

Schwinning, S., B. I. Starr, and J. R. Ehleringer. 2005. Summer and winter drought in a cold desert ecosystem (Colorado Plateau) part II: Effects on plant carbon assimilation and growth. Journal of Arid Environments 61:61–78.

Sebastian, D. J., S. J. Nissen, J. R. Sebastian, and K. G. Beck. 2017. Seed bank depletion: The key to long-term downy brome (Bromus tectorum L.) management. Rangeland Ecology and Management 70:477–483.

Sebastian, D. J., J. R. Sebastian, S. J. Nissen, and K. G. Beck. 2016. A potential new herbicide for invasive annual grass control on rangeland. Rangeland Ecology and Management 69:195–198.

Shinn, S. L., and D. C. Thill. 2004. Tolerance of several perennial grasses to imazapic 1. Weed Technology 18:60–65.

Sleeter, B. M., T. S. Wilson, and W. Acevedo, editors. 2012. Status and trends of land change in the Western United States—1973 to 2000. U.S. Geological Survey Professional Paper 1794-A. U.S. Geological Survey.

Smith, D. C., S. E. Meyer, and V. J. Anderson. 2008. Factors affecting Bromus tectorum seed bank carryover in Western Utah. Rangeland Ecology & Management 61:430–436.

Soil Survey Staff, Natural Resources Conservation Service, United States Department of Agriculture. 2006. Official Soil Series Descriptions. Jokodowski Series. Available online at https://soilseries.sc.egov.usda.gov/OSD_Docs/J/JOKODOWSKI.html. Accessed [8/27/2019].

Stier, M. P. 2012. Colorado Plateaus Ecoregion. Pages 219–227 *in* and W. A. Benjamin M. Sleeter, Tamara S. Wilson, editor. Status and Trends of Land Change in the Western United States—1973 to 2000. U.S. Geological Survey.

USDA Farm Service Agency (FSA) Aerial Photography Field Office. 2017 Release. Available at https://www.usgs.gov/centers/eros/science/usgs-eros-archive-products-overview?qt-science_center_objects=0#qt-science_center_objects

USDA Forest Service. 2003. Integrated management plan for the California Park Special Interest Area. Steamboat Springs, Colorado.

USDA, NRCS. 2019. The PLANTS Database (http://plants.usda.gov, 28 September 2019). National Plant Data Team, Greensboro, NC 27401-4901 USA.

Valkó, O., P. Török, B. Tóthmérész, and G. Matus. 2011. Restoration potential in seed banks of acidic fen and dry-mesophilous meadows: Can restoration be based on local seed banks? Restoration Ecology 19:9–15.

Vandvik, V., K. Klanderud, E. Meineri, I. E. Måren, and J. Töpper. 2016. Seed banks are biodiversity reservoirs: Species-area relationships above versus below ground. Oikos 125:218–228.

Ware, I. M., P. Terletzky, and P. B. Adler. 2014. Conflicting management objectives on the Colorado Plateau: Understanding the effects of bison and cattle grazing on plant community composition. Journal for Nature Conservation 22:293–301.

Welsh, S. L. 1978. Problems in plant endemism on the Colorado Plateau. Great Basin Naturalist Memoirs:191–195.

West, N. E. 1983. Great Basin-Colorado Plateau sagebrush semi-desert. Pages 331–349 *in* N. E. West, editor. Temperate Deserts and Semi-Deserts. Elsevier, Amsterdam.

Winkler, D. E., D. M. Backer, J. Belnap, J. B. Bradford, B. J. Butterfield, S. M. Copeland, M. C. Duniway, A. M. Faist, S. E. Fick, S. L. Jensen, A. T. Kramer, R. Mann, R. T. Massatti, M. L. McCormick, S. M. Munson, P. Olwell, S. D. Parr, A. A. Pfennigwerth, A. M. Pilmanis, B. A. Richardson, E. Samuel, K. See, K. E. Young, and S. C. Reed. 2018. Beyond traditional ecological restoration on the Colorado Plateau. Restoration Ecology 26:1055–1060.

CHAPTER II SOIL SEED BANK COMPOSITION AND RELATIONSHIP TO ABOVEGROUND VEGETATION OF TWO COLORADO SHRUBLANDS

Introduction

The soil seed bank – it's composition and similarity to the present aboveground vegetation – of intact and disturbed shrublands in western North America is poorly understood (Humphrey and Schupp 2001, Hopfensperger 2007, Pekas and Schupp 2013, Gioria and Pyšek 2015, Jonas et al. 2018). The soil seed bank is defined by Gallagher (2014) as, "all living seeds in the soil and on the soil surface" and is a reservoir of plant propagules that can confer resilience to a functioning and stable ecosystem (Hopfensperger 2007, Maestas et al. 2016). Such resilience is crucial to the ability of a given site to recover following disturbance, e.g. fire, drought, or grazing (Hopfensperger 2007). The seed bank can be viewed as both a "memory" of vegetation that is no longer present at a site, and the regenerative potential of species that reproduce primarily by seed (Gioria and Pyšek 2015, Vandvik et al. 2016, Barga and Leger 2018). In a degraded or invaded ecosystem, however, the soil seed bank could play a negative role in ecosystem function and maintain a site in an undesirable alternate state (Humphrey and Schupp 2001, Von Blanckenhagen and Poschlod 2005, Chambers et al. 2014a, Haussmann et al. 2019).

Plant species primarily produce one of two different types of seed banks, termed transient or persistent. Transient seed banks are those that remain viable for less than one year (Baskin and Baskin 1989, Gallagher 2014, Gremer and Venable 2014). Woody shrub species like *Artemisia* spp. L. (sagebrush) and many perennial species are known to produce a transient seed bank (Humphrey and Schupp 2001, Jurado and Flores 2005). Persistent seed banks are those that can remain viable for more than one year, and have the potential to remain dormant for decades if conditions are favorable – some species seed can even remain dormant for centuries (Jurado and

Flores 2005, Pakeman et al. 2012, Gallagher 2014, Gremer and Venable 2014, Gioria and Pyšek 2015). Shrub species such as *Atriplex confertifolia* (Torr. & Frém.) S. Watson (shadscale saltbush) and *Atriplex canescens* (Pursh) Nutt. (fourwing saltbush) form persistent seed banks, in addition to many annual plant species (Meyer et al. 1998, Humphrey and Schupp 2001, Jurado and Flores 2005). Some plants produce both transient and persistent seed banks in order to respond to variable environmental conditions and respond to disturbance, known as bet hedging (Gallagher 2014, Gremer and Venable 2014, Shefferson et al. 2018).

The potential of the soil seed bank to influence ecological restoration – the make-up of native and non-native species seeds – in shrublands is largely unknown (Hopfensperger 2007, Bossuyt and Honnay 2008, Vandvik et al. 2016, Jonas et al. 2018). It is also a potential constraint that has largely been ignored in ecological restoration planning, but can have significant consequences on the outcome of restoration activities (Von Blanckenhagen and Poschlod 2005, Bossuyt and Honnay 2008, Pekas 2010, Sebastian et al. 2017a). The soil seed bank can have either positive or negative implications for ecological restoration (Bossuyt and Honnay 2008). Sites with abundant native species in the seed bank could help to reduce treatment costs and allow for passive restoration (Von Blanckenhagen and Poschlod 2005, Hopfensperger 2007, Cowan and Anderson 2014, Rayburn et al. 2016). Whereas sites with low abundance of native seeds would likely require supplemental seeding; and sites with high abundance of non-native species seeds would likely require seed bank depletion treatments prior to seeding (Hopfensperger 2007, Rayburn et al. 2016, Sebastian et al. 2017a, Grant-Hoffman et al. 2018).

Some seed bank research has been conducted in Great Basin sagebrush shrublands and shortgrass-steppe, but little research has been done in sagebrush or salt desert shrub dominated systems in Colorado (Coffin and Lauenroth 1989, Humphrey and Schupp 2001, Pekas 2010, Pekas and Schupp 2013, Haight et al. 2019). These gaps in literature make it difficult for land managers to make well-informed ecological restoration decisions at the local level (Knapp and Fernandez-Gimenez 2009, Jonas et al. 2018). My research aims to address these gaps to help public land managers in Colorado in high conservation-priority shrublands make restoration decisions that take the potential of the soil seed bank into account.

In this study, I ask: how do seed bank species richness, diversity, and seed abundance compare between undesirable sites that are in need of restoration and desirable sites? I hypothesize that non-diverse plant communities will contain non-diverse seed banks relative to more diverse plant communities (Hypothesis 2.1).

Materials and Methods

Study Site Description

Plant communities in this study include a high-elevation (2400-2500m) silver sagebrush parkland in Northern Colorado (California Park) and a region of salt desert shrubland vegetation in the Colorado Plateau of western Colorado. California Park is a montane silver sagebrush parkland in the Southern Rockies ecoregion and is managed by the USDA Forest Service as a Special Interest Area (SIA) on the Medicine Bow-Routt National Forest (USDA Forest Service 2003). This SIA designation is due to its ecological, recreational, cultural, and geological significance (USDA Forest Service 2003, Kachergis et al. 2012, 2014). Between 1981 and 2010, mean annual temperature (MAT) and precipitation (MAP) were 4.2°C and 718 mm, respectively (PRISM climate group, 1981-2010 climate normals). Salt desert shrubland communities sampled consist of several low-elevation (1400-1600m) sites in western Colorado. These sites are in Mesa and Delta counties and are located in the Colorado Plateau ecoregion. Between 1981 and 2010, mean annual temperature (MAT) and precipitation (MAP) for Grand Junction, Colorado were

11.7°C and 230 mm, respectively (PRISM climate group, 1981-2010 climate normals). These areas are managed by the Bureau of Land Management (BLM) and are in, or on the periphery of, the McInnis Canyon and Dominguez-Escalante National Conservation Areas (NCA). See "Study Sites" section in Chapter I for additional background about the ecology and management history of both sites.

Sites in California Park were chosen in coordination with Forest Service personnel. In total, seven sites were sampled throughout the California Park SIA that represent areas of "desirable" and "undesirable" vegetation and ecological characteristics, according to Forest Service resource managers (**Figure 2.1**). Three desirable sites were sampled, and four undesirable sites were sampled. All sample sites were located in mapped areas of the Jokodowski soil series, taxonomically classified as Fine, smectitic Typic Humicryerts (Soil Survey Staff, 2006). Desirable sites (Reference sites #1 - #3 [R1 – R3]) were characterized as having 15 - 35% sagebrush (*Artemisia* spp. L.) cover, low amounts of exposed bare ground, and a diverse community of native herbaceous vegetation (**Figure 2.2**). These desirable sites were considered to be of high habitat and range "quality" by Forest Service resource managers from a vegetation perspective. Undesirable sites were characterized as being of low habitat and range "quality". These include three degraded states described throughout the park by Kachergis et al. (2014) and Forest Service personnel (USDA Forest Service 2003). The undesirable states include sites dominated by *Phleum pratense* L. (Timothy – non-native pasture grass) (D1; **Figure 2.3**); sites termed "scabs" with low plant cover most years, large shrink-swell cracking, and are prone to mass-movement (D2 and D3; **Figure 2.4**); and sites dominated by "monocultures" of a native forb *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) (D4; **Figure 2.5**) (USDA Forest Service 2003, Kachergis et al. 2014).

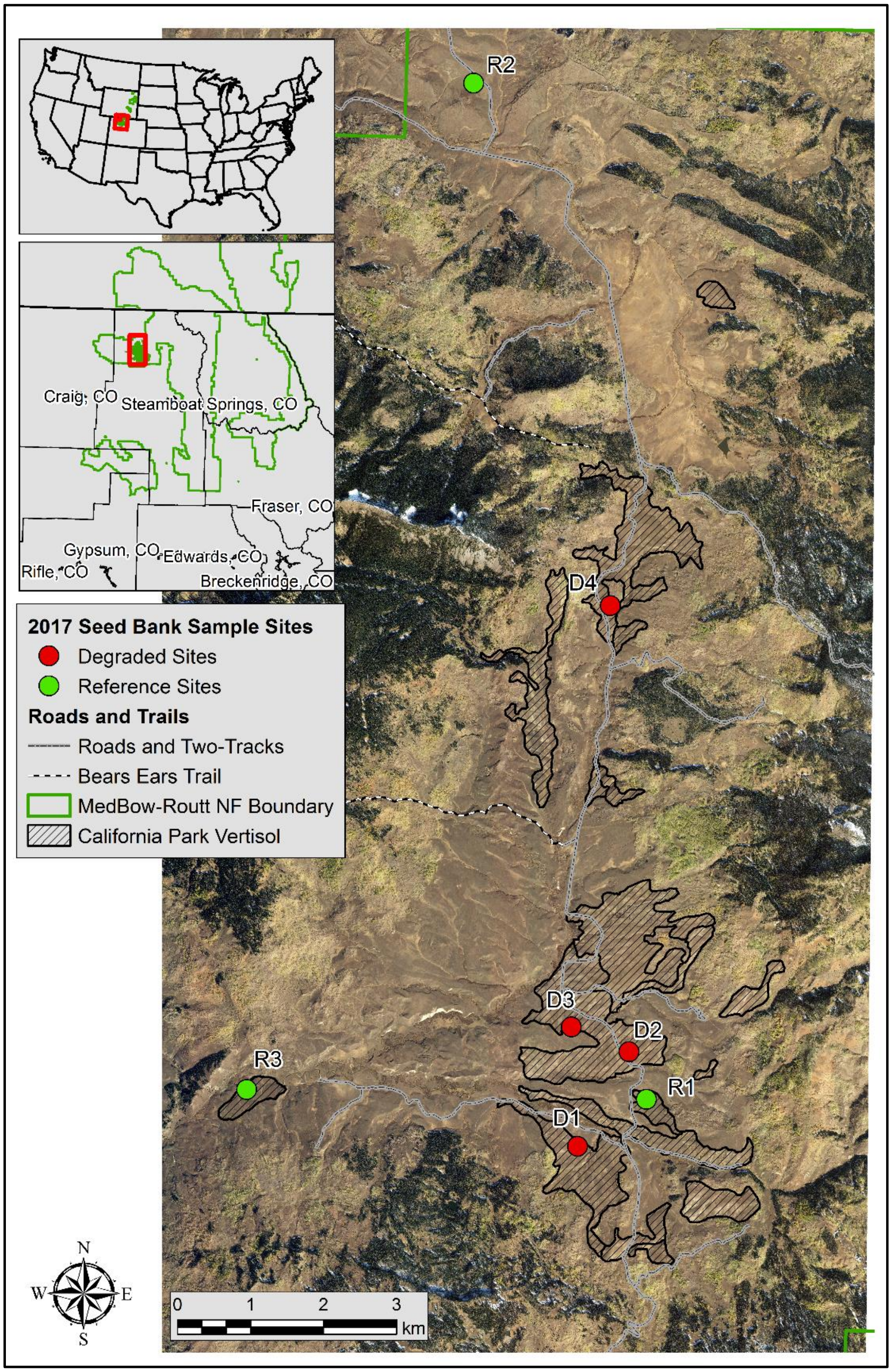


Figure 2.1: Map of California Park in northern Colorado on the Medicine Bow-Routt N.F. Undesirable (red) and Desirable (green) sites had soil seed bank sampling conducted in August 2017 and aboveground vegetation sampling conducted in June and July 2019. Map units of the Jokodowski soil series (Fine, smectitic Typic Humicryerts) displayed. Aerial imagery from fall of 2017 (USDA-FSA, 2017).

Figure 2.2: Photo of site R3 in the southwestern portion of California Park, looking southwest above Elkhead Creek. This site exhibits ecological characteristics desired by resource managers, including 15-35% *Artemisia cana* Pursh (silver sagebrush) and *Artemisia tridentata* Nutt. ssp. *vaseyana* (Rydb.) Beetle (mountain big sagebrush) cover and a diverse understory. Photo taken 8/02/2017 by Ryan Schroeder.

Figure 2.3: Photo of site D1 in the southern portion of California Park, looking south. This site is dominated by the non-native pasture grass *Phleum pratense* L. (Timothy) and represents as degraded state that resource managers desire to restore to a condition with sagebrush and higher native forb diversity. Photo taken 8/01/2017 by Ryan Schroeder.

Figure 2.4: Photo of site D2 in California Park, looking northwest towards Bears Ears Peak (left of tape) and Sugarloaf Mountain (right of tape). Most years, this “scab” site has very little plant cover but following the above-average winter and spring precipitation of 2019, the site was dominated by *Lomatium leptocarpum* (Torr. & A. Gray) J.M. Coult. & Rose (gumbo-lomatium), a native short-lived perennial forb. This site represents as degraded state that resource managers desire to restore to a condition with greater year-round plant cover and species diversity. Photo taken 6/10/2019 by Ryan Schroeder.

Figure 2.5: Photo of site D4 in the northern portion of California Park, looking west towards Saddle Mountain. This site is dominated by the native forb *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) and represents a degraded state that resource managers wish to restore to a condition with greater year-round plant cover and species diversity. Photo taken 7/17/2018 by Ryan Schroeder.

Salt desert sites were selected in coordination with BLM staff from those sampled by Jonas et al. (2018). Seven sites were chosen that represent areas of "desirable" and "undesirable" salt desert ecological and vegetation characteristics in this physiographic region (**Figure 2.6**). Two desirable sites were sampled – Badger Wash Reference (BWR; **Figure 2.7**) and Relic Exclosure (RE) – and five undesirable sites were sampled – Alkali Exclosure (AE), Badger Wash Degraded (BWD), Peeples Fire (PF), Two Road Fire (2RF; **Figure 2.8**), and Two Road Seeded (2RS). These sample sites span a wide topo-edaphic gradient but are characterized by Jonas et al. (2018) as having salt desert soil characteristics with the successional potential to reach a salt desert shrub-dominated state similar to that of the reference sites sampled. Desirable sites are considered to be of high habitat and range “quality” by BLM resource managers. Undesirable sites are characterized as being of low habitat and range “quality”.

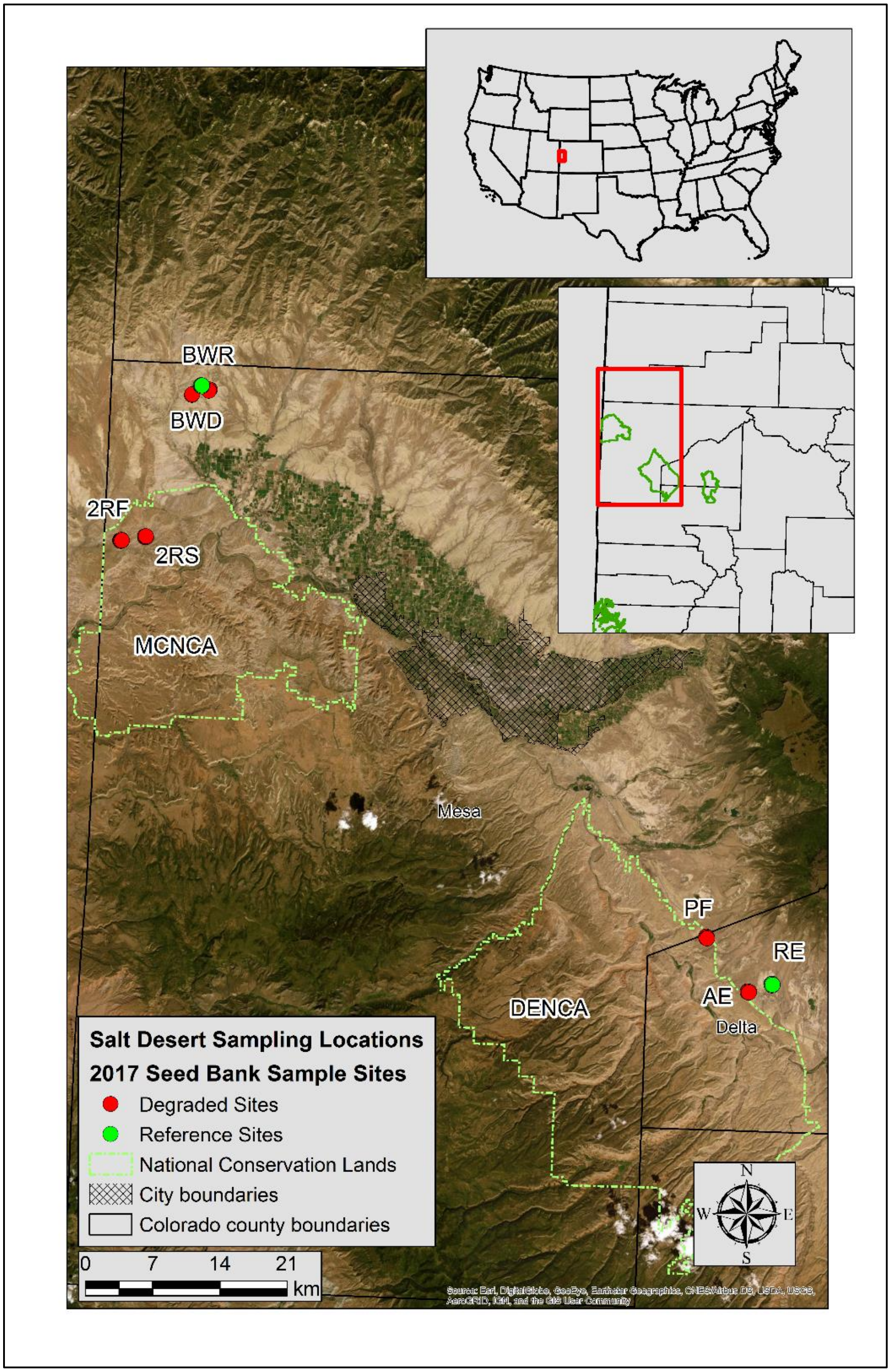


Figure 2.6: Map of Salt Desert study sites in western Colorado within and in the vicinity of the McInnis Canyon National Conservation Area (MCNCA) and Dominguez-Escalante National Conservation Area (DENCA). Undesirable (red) and Desirable (green) sites had soil seed bank sampling conducted in October 2017 and aboveground vegetation sampling conducted in May 2019.

Figure 2.7: Photo of the Badger Wash Reference (BWR) site in western Colorado, looking south. The site is inside a long-term grazing exclosure and has been the subject of several long-term studies of salt desert shrublands (Lusby et al. 1963, Lusby 1970, 1979, Duniway et al. 2018, Jonas et al. 2018). This site exhibits ecological characteristics desired by resource managers for salt desert shrublands, including a diverse community of native perennial halotolerant shrubs, grasses, forbs, and intact biological soil crusts. Photo taken 5/20/2019 by Ryan Schroeder.

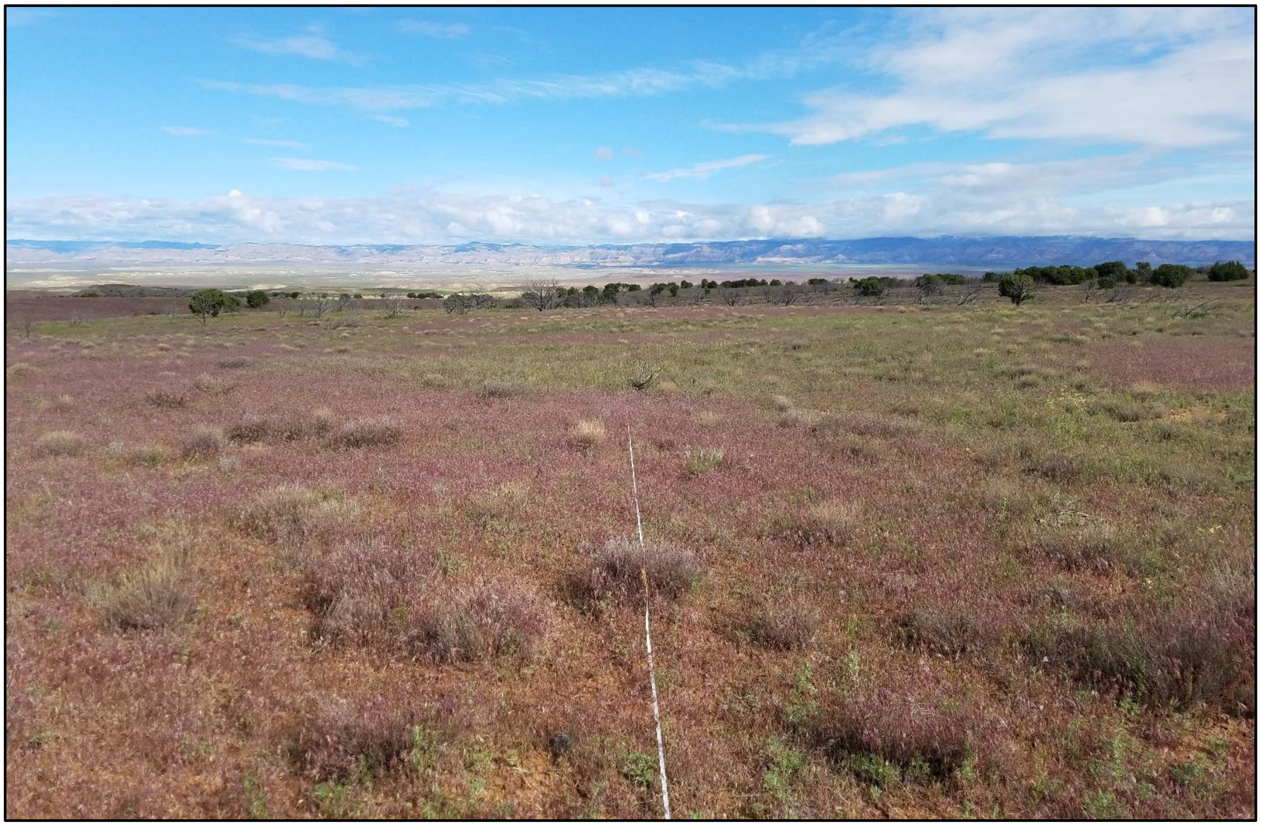


Figure 2.8: Photo of the Two Road Fire (2RF) site in western Colorado, looking north towards the Book Cliffs. The site is dominated by the non-native annual grass *Bromus tectorum* L. (cheatgrass) and is in a degraded state that resource managers desire to restore to greater native species diversity. Photo taken 5/21/2019 by Ryan Schroeder.

Desirable sites are inside of long-term grazing exclosures and have relatively high cover of halotolerant shrub species such as *Atriplex confertifolia* (Torr. & Frém.) S. Watson (shadscale saltbush), *Atriplex gardnerii* (Moq.) D. Dietr. (Gardner's saltbush), *Sarcobatus vermiculatus* (Hook.) Torr. (greasewood), and *Artemisia tridentata* Nutt. ssp. *tridentata* (basin big sagebrush). These desirable sites also have a diversity of patchily distributed native grasses and forbs, low cover of non-native annual grasses and forbs, and intact biological soil crusts in the interspaces (Duniway et al. 2018, Jonas et al. 2018). The undesirable sites were described by Jonas et al. (2018) as being in an "invaded non-native annual" alternate state (**Figure 2.8**). These sites are characterized as having little to no shrub cover, few intact biological soil crusts, and vegetation communities dominated by non-native annual grasses and forbs such as *Bromus tectorum* L. (cheatgrass), *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass), *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill), and *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover).

Soil Seed Bank Sampling and Propagation

Soil seed bank sampling was conducted during early August (Aug. 1-3) 2017 in California Park and late October (Oct. 23-25) 2017 in the salt desert. To attempt to capture the intra-site variability of the soil seed bank, three 100-m transects were established at each site. The start and end points of each transect were recorded with a GPS unit. A soil core (7.32 cm diameter x 5 cm deep) was collected every 5-m along each transect using a bulb-corer. Each core was placed in a clean plastic bucket, resulting in 20 cores per transect and a total of 60 cores (0.2526 m^2 sample area) that were then composited for each site (sample unit) in a plastic tub. The dominant ground cover at the point of each core was recorded. In California Park, the cover types recorded included: bare ground, grass, forb, and shrub (Appendix Table 2d). For salt desert sites, the cover types recorded included: bare ground, biological soil crust (BSC), annual grass, perennial grass, forb, and shrub (Appendix Table 2e). Samples were kept cool and dry before being transported back to the lab where they were weighed and stored at 5° C before being prepared for propagation.

To prepare the soil seed bank samples for propagation, the samples were allowed to air-dry for several days on a lab bench. The samples were then sieved through a 12.5 mm screen to remove rocks (coarse fragments), green vegetation, large root fragments, and tubers from species like *Orogenia linearifolia* S. Watson (Great Basin Indian potato) and *Claytonia lanceolata* Pail. ex Pursh (lanceleaf springbeauty). These removed materials were weighed, stored in Ziploc bags, and retained. This sieving also served to break up large soil aggregates. The remaining soil was returned to the plastic tub, mixed to homogenize the sample, and weighed to the nearest gram. After coarse fragment and vegetation removal, soil seed bank samples from California Park weighed between 9,600 g and 11,600 g, and Salt Desert sample weighed between 13,800 g and 15,200g. The soil samples were subsampled for soil texture, macro-nutrients, metals (for California Park), and sodium-absorption ratio (for salt desert) analyses. Lastly, each sample had ~25% of the original sample weight separated and held in reserve in case of mishaps in the greenhouse or if additional analyses became warranted. This resulted in an effective soil area of 0.1895 m^2 (by 5 cm deep) or 0.009745 m^3 of soil for each site (sample unit) to be propagated in the greenhouse. This value of 0.1895 m^2 was used to convert the number of seedlings that emerged to seed abundance (seeds m^{-2}).

Soil subsamples for all sites were sent to the Soil, Water and Plant Testing Laboratory at Colorado State University for routine nutrient analysis and soil texture by hydrometer. California Park's soils, derived from marine shales that are known to contain selenium, warranted further trace element analysis (Hettinger and Roberts 2005, Statwick and Sher 2017). A Mehlich III extraction was conducted at the Rocky Mountain Research Station's biogeochemistry laboratory (Mehlich 1984). The extractant was analyzed at the Soil, Water and Plant Testing Lab by Inductively Coupled Plasma-Atomic Emission Spectroscopy (ICP-AES) for a suite of 33 elements (**Appendix Table 2b**). In addition to the routine soil nutrient analysis, soils from the Salt Desert were analyzed for Sodium-Absorption Ratio (**Appendix Table 2c**).

The sieved seed bank samples were propagated in the Plant Growth Facility (PGF) on Colorado State University's campus. Samples from California Park were started on October 3, 2017 and Salt Desert samples were started on November 11, 2017 and were grown through August 2018. Over the course of the study, supplemental lighting in the greenhouse bay provided a photoperiod of 16 hours and an average light output of 300 µmol m^{-2} sec^{-1}. Seed bank samples were propagated in 25.4 x 52 cm plastic flats. The flats had a 3 cm base of ProMix BX potting soil spread across the bottom of the flats, and the field soil was spread over the potting soil to a depth of 2 cm or less (Benvenuti 2003, 2007). The overall volume of soil from each site meant that more than one flat was needed per sample unit to maintain a depth of 2 cm. Each California Park sample required three flats and Salt Desert samples required four flats. Flats were set on top of Redi-Heat Propagation Mats (Model #: RHM2015, Phytotronics Inc) set to maintain a constant minimum soil temperature of 21°C based on estimated optimum germination temperature for most species in these ecosystems (Rawlins et al. 2012).

Flats were kept moist to promote germination and were rotated on a bi-weekly basis to account for heterogeneity of greenhouse conditions. There were seven control trays (with only ProMix BX potting soil) randomly distributed among the sample trays to capture any contaminant seeds. Several species *Oxalis corniculata* L. (creeping woodsorrel), *Ericameria nauseosa* (Pall. ex Pursh) G.L. Nesom & Baird (rubber rabbitbrush), *Salix* spp. L. (willow), *Populus deltoides* W. Bartram ex Marshall ssp. *monilifera* (Aiton) Eckenwalder (plains cottonwood), and *Miscanthus* spp. Andersson (silvergrass) were detected in control and study flats and were thus presumed to be contaminants. To help promote seed germination and plant growth, applications of 21-18-18 fertilizer at a concentration of 200 ppm nitrogen were applied as a solution once monthly as a soil soaking (Walck et al. 2011).

Seedlings were identified after emergence, counted, and removed. Seedlings that could not be identified quickly were noted and allowed to either remain in the flats or were transferred into smaller pots to grow-out. Plants were identified to the finest taxonomical level possible –

mostly to the species level. Nomenclature for all plant species follows the USDA NRCS PLANTS Database (2019).

Specimens of each species identified were pressed as vouchers. As flats had all seedlings identified and removed in late spring 2018, the soil was allowed to dry-down for a minimum of two weeks. Seed bank samples were then mixed by hand (with care taken to retain all field soil), soil aggregates crushed, and then fertilized as mentioned above to attempt to induce germination of remaining seeds (Chambers and MacMahon 1994, Walck et al. 2011, Faist et al. 2013, Haight et al. 2019). The seed bank study was terminated after all plants in the trays could be identified and no new emergence was detected for two weeks.

Aboveground Vegetation Sampling

To compare the germinable soil seed bank composition to the aboveground plant community, vegetation sampling was conducted in the summer of 2019. Sampling of salt desert sites occurred on May 20 & 21, following a spring with above-average precipitation that resulted in a "super-bloom" (**Appendix Table 1**). Sampling in California Park occurred on June 10 & 11 for five of the seven sites – with two desirable sites being inaccessible due to high water from above-average spring precipitation (**Appendix Table 1**). The remaining two desirable sites were sampled on July 24. Transects were laid out using the GPS coordinates of the start- and end-points of the original soil seed bank sampling transects as a guide, and the start- and end-points of the vegetation transects recorded. Aboveground vegetation was measured as rooted frequency (presence-absence) of species occurrence using 0.67- x 0.28-m (0.1876 m^2) quadrats. Each transect had ten quadrats sampled, totaling 30 quadrats per site. This allows for the calculation of the relative frequency of species across each site and reflects the probability of encountering the species across the site (Despain et al. 1997). Nomenclature for all plant species followed the USDA NRCS PLANTS Database (2019).

Statistical Methods

Prior to analysis, contaminant species that germinated in the control trays were removed to reduce inflation of species richness values (Faist et al. 2013, Rayburn et al. 2016). To facilitate seed bank richness, diversity, and abundance analyses, total seedling abundance in greenhouse trays was converted to seed abundance (seeds m^{-2}). To facilitate similarity analyses, seedling counts and aboveground plant frequency measures were converted to presence-absence using the decostand function in the Vegan R-package (Oksanen et al. 2019).

I tested my hypothesis by examining soil seed bank composition (species richness, Shannon-Wiener diversity (H), total seed abundance, native species seed abundance, and non-native seed abundance), aboveground vegetation species richness, and site seed bank similarity to aboveground vegetation. Analyses were conducted using the statistical program R v3.6.1 (R Core Team, 2019) and functions in the Vegan R-package (Oksanen et al. 2019). Sample means were compared using Welch two-sample t-tests to account for unequal variance between samples, and an $\alpha = 0.05$ was used for significance testing. Similarity comparisons were calculated using a presence-absence transformed Jaccard dissimilarity indices using the vegdist function of the Vegan package. Jaccard dissimilarity index returns values between 0 and 1, with a value of zero signifying the species composition of the seed bank and aboveground vegetation are identical, and a value of one signifying the composition of the seed bank and aboveground vegetation share no species.

Nonmetric multi-dimensional scaling (NMDS; Kruskal 1964) was employed to describe the dominant soil seed bank species composition and their relationship to site environmental variables. This method of indirect ordination arranges sites in an ordination space that displays ecological dissimilarity. Environmental variables were correlated with the ordination axes after the NMDS ordination was performed in order to see any relationships between seed bank species composition and environmental conditions, those correlated at a p-value <0.10 were displayed on the ordination (McCune and Grace, 2002). NMDS also does not require any assumptions about underlying environmental gradients, and therefore is well suited to these data, as it is not known if soil seed bank composition in shrublands are driven by environmental conditions, above-ground vegetation communities, site history, etc. (Pekas and Schupp 2013, Kachergis et al. 2014, Barga and Leger 2018). NMDS was performed in R v3.6.1 (R Core Team 2019) using the package vegan (Oksanen et al., 2019). The Bray-Curtis distance measure, shared abundance divided by total abundance, was used because it performs well in community ecology analyses (McCune and Grace, 2002).

In order to conduct multivariate analyses of the seed bank composition, species seed abundance values were transformed to relative abundance. This was done in order to characterize and display the dominant species in each site's seed bank. Species that occurred in only one site's seed bank were omitted to reduce the noise in the data (McCune and Grace, 2002) and to ensure that the common species in the seed bank drove the analyses. This resulted in 26 species in California Park and 19 species in the salt desert seed banks. Environmental variables were then correlated to the NMDS axes using the envfit function of the Vegan R package (Oksanen et al. 2019). These environmental variables include several soil nutrient and physical property (sand, silt, clay) measurements, along with aggregate cover variables calculated from the dominant cover where each core was collected from each site. Environmental variables highly colinear with one another were removed from the analysis (McCune and Grace 2002). This ordination method was conducted to see if there were dominant relationships between site classification (desirable or undesiable), seed bank composition, and environmental conditions.

Results

California Park Results

California Park Aboveground Vegetation Communities

Aboveground vegetation sampling in the summer of 2019 identified 74 unique species across the seven sites in California Park. Species richness across all sites ranged from 24 to 43 species. Aboveground species richness averaged 30.3 species on undesirable sites, 33.3 species on desirable sites, and did not differ significantly (**Table 2.1**). Undesirable sites tended to be dominated by few species that have short lifespans, including *Lomatium leptocarpum (*Torr. & A. Gray) J.M. Coult. & Rose (gumbo-lomatium – native perennial forb), *Cerastium arvense* L. (field chickweed – native perennial forb), and *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears – native perennial forb). Desirable site plant communities were more variable but tended to be dominated by perennial species. Dominant species across desirable sites include grasses such as *Festuca idahoensis* Elmer (Idaho fescue – native perennial grass), *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass – native perennial grass), and *Phleum pratense* L. (Timothy – non-native pasture grass), native shrubs such as *Artemisia cana* Pursh (silver sagebrush), *Artemisia arbuscula* Nutt. (low sagebrush), and *Artemisia tridentata* Nutt. (big sagebrush), and numerous annual and perennial forbs.

Table 2.1: Aboveground vegetation species richness of undesirable (n=4) and desirable (n=3) sites in California Park, Colorado. Means reported ± standard error. Statistical significance of comparison between means: $^{*}p \leq 0.05$; $^{**}p \leq 0.01$; $^{\dagger}p \leq 0.10$.

Site Classification	Aboveground Species Richness	Aboveground Native Species Richness	Aboveground Non-native Species Richness
Undesirable	30.3 ± 3.6	27.0 ± 3.2	3.3 ± 0.5
Desirable	33.3 ± 4.8	30.0 ± 4.0	3.3 ± 0.9
	t = -0.51	t = -0.58	t = -0.083
	$p = 0.637$	$p = 0.590$	$p = 0.939$

California Park Seed Bank Composition

A total of 49 species were identified across seven sample sites. Soil seed bank species richness values did not differ between undesirable and desirable sites in California Park. Mean seed bank species richness was 16.8 and 16 for undesirable and desirable sites, respectively (**Table 2.2**). Seed bank Shannon-Wiener diversity (H) did not differ between site classifications. Diversity of desirable sites was more variable than that of undesirable sites. Overall seed abundance is statistically different between undesirable and desirable sites, with undesirable sites averaging less than half of the overall seeds m^{-2} of desirable sites, 588 seeds m^{-2} versus 1401 seeds m^{-2}, p-value = 0.0232 (**Table 2.2**).

Table 2.2: Mean species richness, species Shannon-Wiener diversity (H), and seed abundance (seeds m^{-2}) of seed banks across undesirable (n=4) and desirable (n=3) sites in California Park, Colorado. Means reported ± standard error. Statistical significance of comparison between means: $^{*}p \leq 0.05$; $^{**}p \leq 0.01$; $^{\dagger}p \leq 0.1$.

Site Classification	Seed Bank Species Richness	Seed Bank Diversity	Seed Abundance (seeds m^{-2})	Native Seed Abundance (seeds m^{-2})	Non-native Seed Abundance (seeds m^{-2})
Undesirable	16.8 ± 1.4	2.13 ± 0.097	588 ± 190	394 ± 113	194 ± 104
Desirable	16.0 ± 2.5	1.92 ± 0.416	1401 ± 165	830 ± 202	572 ± 354
	t = 0.26	t = 0.49	t = -3.24	t = -1.88	t = -1.02
	$p = 0.811$	$p = 0.666$	$p = 0.0232^{*}$	$p = 0.150$	$p = 0.400$

When broken down between native and non-native species seeds, the statistical difference did not persist. The seed banks of sites throughout California Park were largely made up of native species seeds (except for R1 – **Appendix Table 3**). Non-native species seeds, primarily of the non-native pasture grasses *Phleum pratense* L. (Timothy) and *Poa pratensis* L. (Kentucky bluegrass), were found throughout both desirable and undesirable sites with variable abundance. These non-native grasses made up between 3% and 71% of the total seed bank abundance of sites. This illustrates the extent to which these non-native pasture grasses have come to persist throughout California Park.

Although species richness and diversity values do not differ between desirable and undesirable sites, the dominant species composition of the seed bank varied from site-to-site. This is illustrated by the non-metric multidimensional scaling (NMDS) ordination in **Figure 2.9**. NMDS found two convergent solutions with a stress approaching zero (stress = 0.0036), indicating excellent model fit (McCune and Grace 2002), with two axes being optimal. This NMDS displays the Bray-Curtis dissimilarity of the dominant species composition of the seed bank of sites in

California Park, weighted by species' seed relative abundance. Sites that are closer together are relatively similar in dominant seed bank composition, while those farther apart are more different. Species codes are overlaid on the ordination and indicate the dominant species present in the seed bank of each site – the closer a species code is to the site, the more abundant that species seed was at the site. Species close to the center of the ordination, including *Festuca idahoensis* Nutt. (Idaho fescue [FEID] – native perennial grass), *Melica spectabilis* Scribn. (purple oniongrass [MESP] – native perennial grass), *Achillea millefolium* L. (common yarrow [ACMI2] – native perennial forb), *Potentilla* spp. L. (cinquefoil [POTEN] – native perennial forb), etc. were found across several sites in intermediate abundances. Only one environmental variable – soil electrical conductivity (EC) – was correlated with the NMDS axes following a permutation procedure (p-value = 0.08).

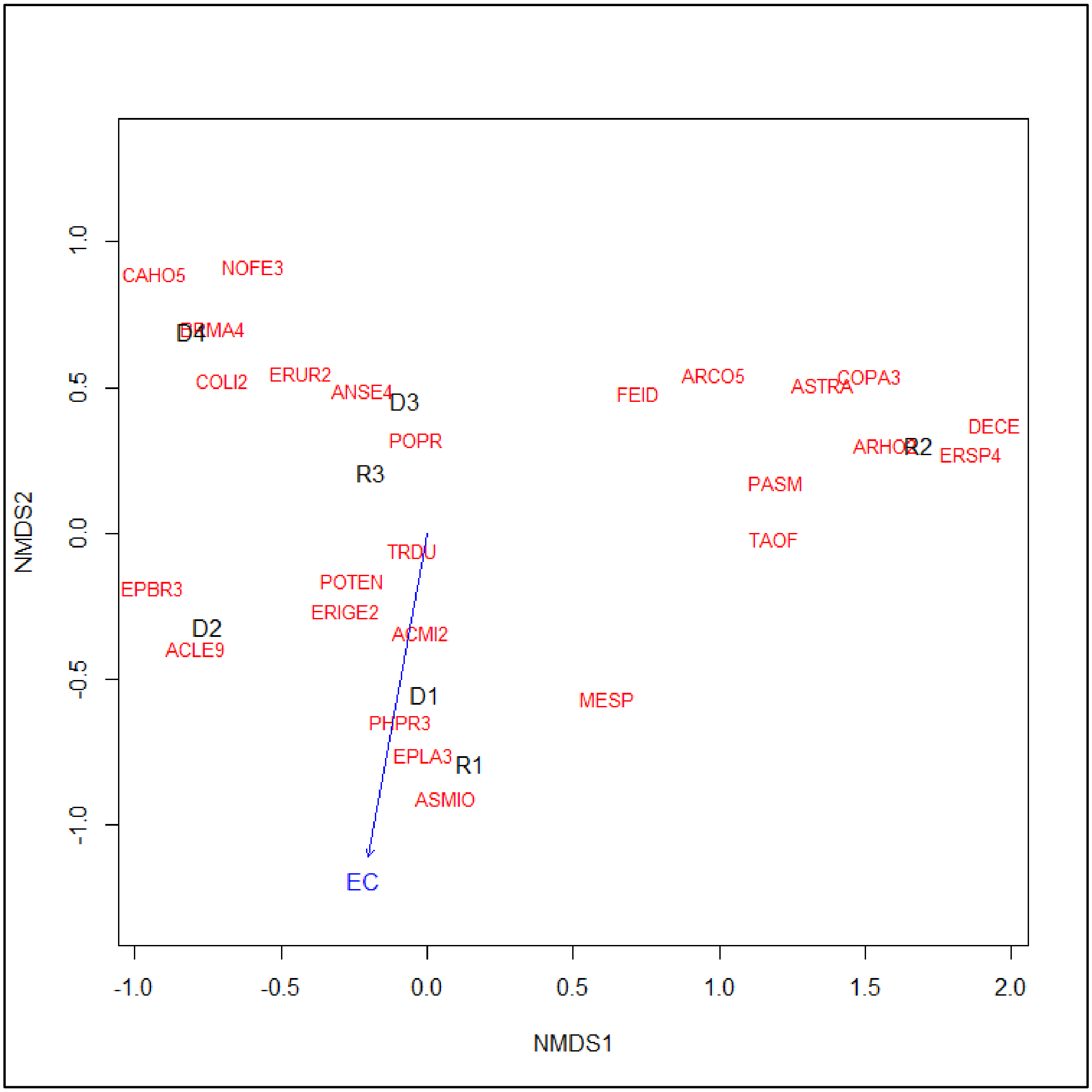


Figure 2.9: This NMDS ordination displays the Bray-Curtis dissimilarity of the dominant seed bank species composition in California Park, weighted by species' seed relative abundance. Final stress = 0.0036. Environmental variables were correlated to the NMDS axes *post hoc* to determine if any environmental variables were correlated with site dominant seed bank composition. Black site names represent the site (plot) scores (R# = desirable sites, D# = undesirable sites). Sites that are closer together are relatively similar in composition, while those farther apart are more different. Seed bank species scores are represented by red species symbols (USDA, NRCS 2019). Solid blue lines represent environmental variables fitted with the NMDS axes with a p-value < 0.10 include soil electrical conductivity (EC; p-val = 0.08). The NMDS was driven by the seed bank composition of sites and the environmental variable overlay indicates that increased EC is correlated with sites such as D1 and R1 that have relatively high abundance of *Phleum pratense* L. (Timothy [PHPR3]) in the seed bank.

Sites R1 and D1 seed banks are similar to each other, being dominated by the non-native pasture grass *Phleum pratense* L. (Timothy [PHPR3]). Site R3, D3, and D4 seed banks had large portions of native forbs including *Erigeron ursinus* D.C. Eaton (Bear River fleabane [ERUR2] – native perennial forb), *Androsace septentrionalis* L. (pygmyflower rockjasmine [ANSE4] – native annual forb), and *Collomia linearis* Nutt.(tiny trumpet [COLI2] – native annual forb). Site D2 had a seed bank dominated by *Achnatherum lettermannii* (Vasey) Barkworth (Letterman's needlegrass [ACLE9] – native perennial grass) and *Epilobium brachycarpum* C. Presl (tall annual willowherb [EPBR3] – native annual forb). The seed bank of site R2 was quite dissimilar from the other sites in California Park and had a diversity of native forbs and grasses, including *Arabis holboellii* Hornem. (Holboell's rockcress [ARHO2] – native perennial forb), *Erigeron speciosus* (Lindl.) DC. (aspen fleabane [ERSP4] – native perennial forb), and *Deschampsia cespitosa* (L.) P. Beauv. (tufted hairgrass [DECE] – native perennial grass).

California Park Seed Bank Similarity to Aboveground Vegetation

Across California Park's seven sites, dissimilarity values between the seed bank and aboveground vegetation of the same site ranged from 0.736 to 0.814. Mean dissimilarity of undesirable sites was 0.77 (± 0.017) and desirable sites was 0.77 (± 0.015) and did not differ statistically ($t = 0.226$, p-value = 0.83). Interestingly, no *Lomatium leptocarpum* (Torr. & A. Gray) J.M. Coult. & Rose (gumbo-lomatium) or *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) seeds were germinated from the seed bank, despite their prevalence in the aboveground vegetation in undesirable and some desirable sites in 2019. Results indicate that seed bank-vegetation dissimilarity is quite high, sites sharing less than 25% of the species present in both the seed bank and aboveground vegetation. Common species found both in the seed bank and aboveground vegetation of a majority of sites included *Achnatherum lettermannii* (Letterman's needlegrass), *Achillea millefolium* L. (yarrow), *Collomia linearis* Nutt. (tiny trumpet), *Festuca idahoensis* Elmer (Idaho fescue), *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass), *Phleum pratense* L. (Timothy), *Poa pratensis* L. (Kentucky bluegrass), and *Potentilla* spp. L. (cinquefoil).

Salt Desert Results

Salt Desert Aboveground Vegetation Communities

Aboveground vegetation sampling of salt desert sites in the spring of 2019 identified 60 unique species across the seven sites. Species richness values ranged from 4 to 37 across all sites. Mean species richness of undesirable sites was 16.4 and mean species richness of desirable sites was 31, and not statistically different (**Table 2.3**). Undesirable sites tended to be dominated by numerous non-native annual species such as *Bromus tectorum* L. (cheatgrass), *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass), *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover), and *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill). Desirable site plant communities were variable but tended to be dominated by native shrubs, numerous native annual and perennial forbs and grass species, and interspaces with biological soil crusts. Dominant species across desirable sites include shrub species such as *Atriplex gardneri* (Moq.) D. Dietr. (Gardner's saltbush) and *Atriplex confertifolia* (Torr. & Frém.) S. Watson (shadscale saltbush), with *Artemisia tridentata* Nutt. ssp. *tridentata* (basin big sagebrush) a component of the uplands of Badger Wash Reference. Native annual species prevalent in these reference sites included *Cryptantha minima* Rydb. (little cryptantha) and *Descurainia pinnata* (Walter) Britton (western tansymustard). The forb species *Calochortus nuttallii* Torr. & A. Gray (sego lily) was prevalent across Badger Wash Reference during the super bloom of 2019 (**Figure 2.7**). *Bromus tectorum* L. (cheatgrass) and other non-native annuals more common in undesirable sites were found patchily throughout both reference sites.

Table 2.3: Aboveground vegetation species richness of undesirable (n=5) and desirable (n=2) sites in the salt desert shrublands of western Colorado. Means reported ± standard error. Statistical significance of comparison between means: * $p \leq 0.05$; ** $p \leq 0.01$; † $p \leq 0.10$.

Site Classification	Aboveground Species Richness	Aboveground Native Species Richness	Aboveground Non-native Species Richness
Undesirable	16.4 ± 5.2	12.0 ± 4.7	4.4 ± 1.1
Desirable	31.0 ± 6.0	25.0 ± 6	6.0 ± 0
	t = -1.83	t = -1.71	t = -1.43
	p = 0.174	*p* = 0.210	*p* = 0.227

Salt Desert Seed Bank Composition

A total of 46 species were identified across the seven sites sampled. Soil seed bank species richness values did not differ between undesirable and desirable sites of the salt desert. Mean species richness was 11.6 and 17.5 for undesirable and desirable sites, respectively, p-value = 0.0543 (**Table 2.4**). Seed bank Shannon-Wiener diversity (H) did not differ between site classifications. Seed bank diversity of undesirable sites was more variable than that of desirable sites. Overall seed abundance was not different between undesirable and desirable sites, with undesirable sites averaging 774 seeds m^{-2} and reference sites 504 seeds m^{-2} (**Table 2.4**).

Table 2.4: Mean species richness, species diversity (Shannon-Wiener), and seed abundance (seeds m^{-2}) of seed banks across undesirable (n=5) and desirable (n=2) sites in the salt desert shrublands of western Colorado. Means reported ± standard error. Statistical significance of comparison between means: $^{*}p \leq 0.05$; $^{**}p \leq 0.01$; $^{\dagger}p \leq 0.1$.

Site Classification	Seed Bank Species Richness	Seed Bank Diversity	Seed Abundance (seeds m^{-2})	Native Seed Abundance (seeds m^{-2})	Non-native Seed Abundance (seeds m^{-2})
Undesirable	11.6 ± 2.2	1.56 ± 0.243	774 ± 158	304 ± 119	470 ± 112
Desirable	17.5 ± 0.5	2.22 ± 0.066	504 ± 50	293 ± 34	211 ± 15
	t = -2.61	t = -2.64	t = 1.63	t = 0.089	t = 2.29
	$p = 0.0543^{\dagger}$	$p = 0.0511^{\dagger}$	*p* = 0.168	*p* = 0.933	$p = 0.0810^{\dagger}$

Seed banks throughout the salt desert had non-native species seed present (**Table 2.4**). Undesirable sites have soil seed banks composed of more than half non-native seeds. In contrast, reference sites have approximately 40% non-native seeds. This illustrates the extent to which non-native annual plant species have invaded and come to persist across a wide topo-edaphic and management gradients of salt desert ecosystems.

Although species richness and diversity values do not statistically differ between desirable and undesirable sites, the dominant species composition of the seed bank varied from site-to-site. This is illustrated by the non-metric multidimensional scaling (NMDS) ordination in **Figure 2.10**. NMDS found two convergent solutions with a low stress (stress = 0.0448) indicating good model fit (McCune and Grace 2002), with two axes being optimal. Species codes are overlaid on the ordination and indicate the dominant species present in each site – the closer a species code is to the site, the more abundant that species seed was at the site. Species close to the center of the ordination, including *Erysimum capitatum* (Douglas ex Hook.) Greene (sanddune wallflower [ERCA14] – native biennial/perennial forb), *Gutierrezia sarothrae* (Pursh) Britton & Rusby (broom snakeweed [GUSA2] – native sub-shrub), *Cyrptantha ambigua* (A. Gray) Greene (basin cryptantha [CRAM3] – native annual forb), and *Descurainia pinnata* (Walter) Britton (western tansymustard [DEPI] – native annual forb) were found across several sites in intermediate abundances. Six environmental variables were significantly correlated (p-value < 0.10) with the NMDS axes following a permutation procedure. These included biological soil crust (BSC) cover (p-val = 0.062), Forb cover (p-val = 0.059), soil organic matter (OM) content (p-val = 0.056), total soil phosphorus (P; p-val = 0.024), total soil copper (Cu; p-val = 0.002), and soil Sodium-Absorption Ratio (SAR; p-val = 0.016).

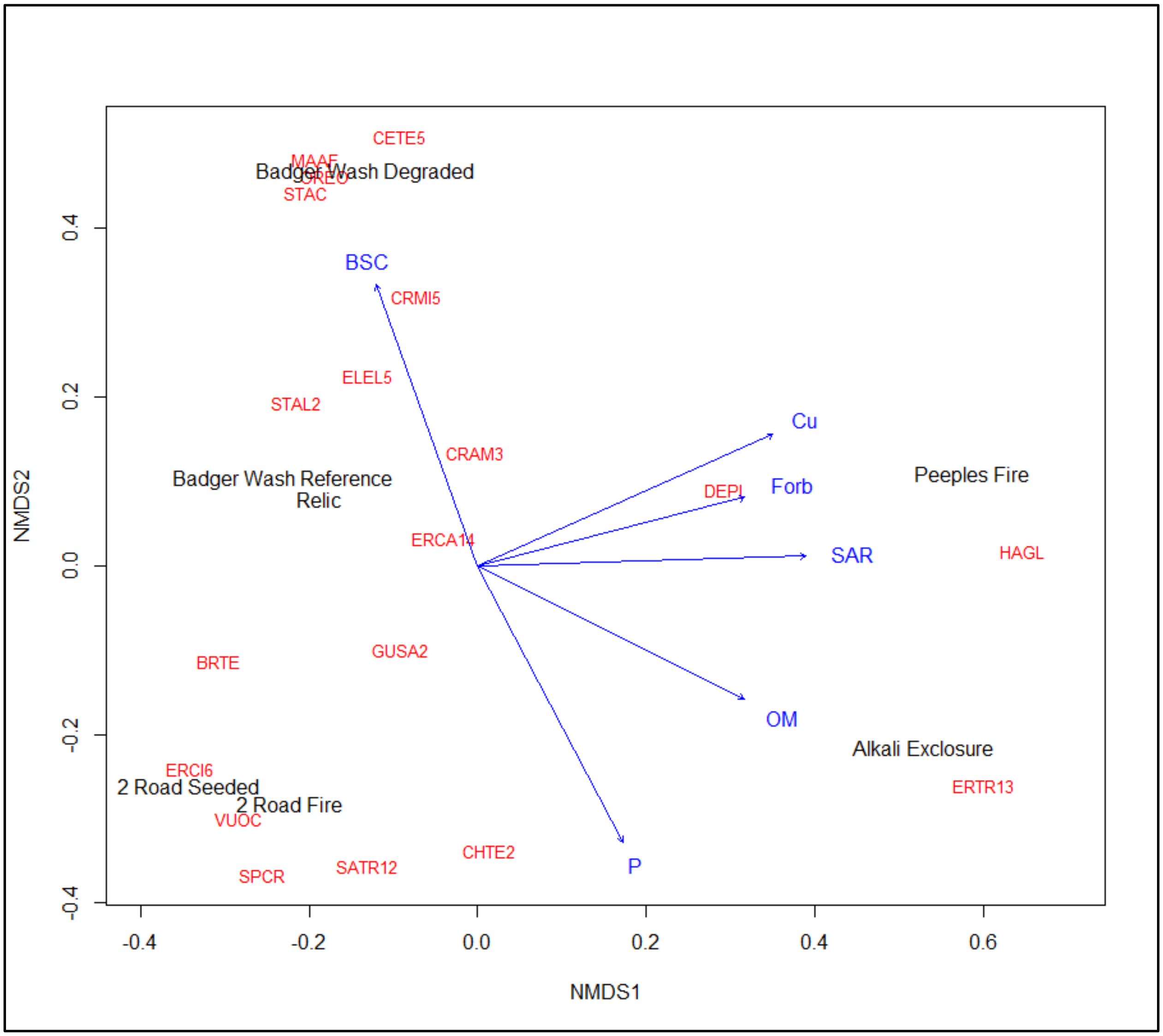


Figure 2.10: This NMDS ordination displays the Bray-Curtis dissimilarity of the dominant species composition of the seed bank of sites in the salt desert, weighted by species' seed relative abundance. Final stress = 0.0448. Environmental variables were correlated to the NMDS axes *post hoc* to determine if any environmental variables were correlated with site dominant seed bank composition. Black site names represent the site (plot) scores. Sites that are closer together are relatively similar in composition, while those farther apart are more different. Desirable sites include Badger Wash Reference and Relic. Undesirable sites include 2 Road Fire, 2 Road Seeded, Alkali Exclosure, Badger Wash Degraded, and Peeples Fire. Seed bank species scores are represented by red species symbols (USDA, NRCS 2019). Solid blue lines represent environmental variables fitted with the NMDS axes with a p-value < 0.10 include biological soil crust (BSC) cover (p-val = 0.062), Forb cover (p-val = 0.059), soil organic matter (OM) content (p-val = 0.056), total soil phosphorus (P; p-val = 0.024), total soil copper (Cu; p-val = 0.002), and soil Sodium-Absorption Ratio (SAR; p-val = 0.016).

The two desirable sites in the salt desert (Badger Wash Reference and Relic) cluster close together with a diversity of native species, but also high relative abundance of cheatgrass [BRTE] seed making up the seed bank. Native species common in the seed banks of these desirable sites include *Descurainia pinnata* (Walter) Britton (western tansymustard [DEPI] – native annual forb), *Cryptantha ambigua* (A. Gray) Greene (basin cryptantha [CRAM3] – native annual forb), and *Elymus elymoides* (Raf.) Swezey (squirreltail [ELEL5] – native perennial grass). These desirable sites shared similar environmental characteristics as well, including moderate BSC cover and relatively low soil organic matter (OM), Sodium-Absorption Ratio (SAR), total soil phosphorus (P), and total soil copper (Cu).

The undesirable sites are on the edges of the ordination space, their seed banks being dominated by a few species (mostly non-natives). Alkali Exclosure and Peeples Fire were dominated by non-native annuals *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass [ERTR13]) and *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover [HAGL]), respectively. Increased soil organic material (OM) and Sodium-Absorption Ratio (SAR) are highly correlated with the seed bank communities of these undesirable sites. The 2 Road Fire and 2 Road Seeded sites cluster together, both being dominated by *Bromus tectorum* L. (cheatgrass [BRTE] – non-native annual grass) and other non-native forbs such as *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill [ERCI6] – non-native annual forb), *Salsola tragus* L. (prickly Russian thistle [SATR12] – non-native annual forb), and *Choripsora tenalla* (Pall.) DC. (crossflower [CHTE2] – non-native annual forb). The Badger Wash Degraded site is interesting in that it has several native species seed in relatively low abundances in the seed bank, as well as several non-native species (that dominate the seed bank), while also having relatively high BSC cover. Non-native species that predominate in this site's seed bank are *Malcolmia africana* (L.) W.T. Aiton (African mustard

[MAAF] – non-native annual forb) and *Ceratocephala testiculata* (Crantz) Roth (curveseed butterwort [CETE5] – non-native annual forb). Native species seed present in lower abundances at Badger Wash Degraded included *Stenotus acaulis* (Nutt.) Nutt. (stemless mock goldenweed [STAC] – native perennial forb), *Oreocarya* spp. Greene (cryptantha [OREO] – native perennial forb), *Cryptantha minima* Rydb. (little cryptantha [CRMI5] -native annual forb), and *Stanleya albescens* M.E. Jones (white princesplume [STAL2] – native perennial forb).

Salt Desert Seed Bank Similarity to Aboveground Vegetation

Across the seven salt desert sites, Jaccard dissimilarity values between the seed bank and aboveground vegetation ranged from 0.45 to 1.0. Mean Jaccard dissimilarity of undesirable sites was 0.72 (± 0.096) and desirable sites was 0.78 (± 0.029) and did not differ statistically ($t = -0.543$, p-value = 0.61). Dominant native and non-native annual species were the most similar species found between the soil seed bank and aboveground vegetation. On average, sites shared slightly more than 25% of the species present in both the seed bank and aboveground vegetation. Common species found in both the seed bank and aboveground vegetation across a majority of sites include *Bromus tectorum* L. (cheatgrass – non-native annual grass), *Cryptantha minima* Rydb. (little cryptantha – native annual forb), *Descurainia pinnata* (Walter) Britton (western tansymustard – native annual forb), *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill – non-native annual forb), and *Gutierrezia sarothrae (Pursh)* Britton & Rusby (broom snakeweed – native sub-shrub).

Discussion

In this study, I hypothesized that non-diverse plant communities (undesirable sites) will contain non-diverse seed banks relative to more diverse plant communities (desirable sites). These results from aboveground vegetation species richness and germinable soil seed bank composition do not support this hypothesis in either shrubland ecosystem studied. Neither aboveground species richness nor soil seed bank species richness and Shannon-Wiener diversity (H) statistically differed from one another between desirable and undesirable sites.

Mean soil seed bank species richness values were similar to values reported in literature of other intact and invaded shrubland ecosystems (Bossuyt and Honnay 2008, Pekas 2010, Pekas and Schupp 2013, Gioria and Pyšek 2015, Martyn et al. 2016, Vandvik et al. 2016, Haight et al. 2019). The high Jaccard dissimilarity of species composition between seen bank and aboveground vegetation were also similar to the limited values available in literature (Bossuyt and Honnay 2008, Martyn et al. 2016). Soil seed bank Shannon-Weiner diversity (H) values were greater in both of these shrublands than values reported by Bossuyt and Honnay (2008) for European heathlands. Soil seed bank abundances (seeds m^{-2}) were similar to values found in other studies (Bossuyt and Honnay 2008, Martyn et al. 2016).

Few shrub seedlings were germinated from sites in either California Park or the salt desert (**Appendix Table 3 and Appendix Table 4**). This is contrary to what Bossuyt and Honnay (2008) found in European heathlands, but similar to seed bank studies conducted in more arid shrublands (Pekas and Schupp 2013, Martyn et al. 2016, Barga and Leger 2018, Haussmann et al. 2019). Shrub seedlings that did germinate were found in both desirable and undesirable sites, but in relatively low abundance 5 – 26 seeds m^{-2}. Non-native species seed were present across desirable and undesirable sites in both shrublands, indicating the extent to which these non-native species have become established in these ecosystems. Interestingly, desirable sites tended to have an equal or greater number of non-native species relative to degraded sites, but with lower overall abundance in the seed bank. This may be an indication of “the rich getting richer”, where desirable sites are able to support a greater number of total species – both native and non-native (Stohlgren et al. 2002, 2003). Importantly, the presence of such non-native species seed in reference site seed banks may be an indicator of vulnerability to non-native species increases if disturbances were to occur (Stohlgren et al. 1999, 2002, Chambers et al. 2014b, Kachergis et al. 2014).

The seed bank composition between desirable and undesirable sites in these shrublands was more diverse and nuanced than expected. Similar numbers of species were found in the seed banks of desirable and undesirable sites for the corresponding shrubland, but there were differences in the dominant species and the functional groups of species present. There are important management and restoration implications that the seed bank poses in these different ecosystems. The specifics of the soil seed bank composition, the relationship to the aboveground plant community, and their implications for ecological restoration differ by the shrubland ecosystem being considered.

California Park

In the high elevation silver sagebrush parkland of California Park, undesirable sites had a statistically lower abundance (seeds m^{-2}) of seeds compared to desirable sites. This suggests that a lack of propagules in the undesirable sites could pose a limitation for passive ecological restoration (Bossuyt and Honnay 2008, Rayburn et al. 2016, Haussmann et al. 2019). Although relatively few non-native species were present in both the aboveground vegetation and seed bank, the two that were most prevalent – *Phleum pratensis* L. (Timothy) and *Poa pratensis* L. (Kentucky bluegrass) – made up between 8% and 46% of the total number of seeds m^{-2} in undesirable sites. The prevalence of these non-native pasture grass seeds could pose an additional constraint in the form of propagule pressure by these highly competitive species (Lockwood et al. 2005, Isselin-Nondedeu and Bédécarrats 2009, Kachergis et al. 2014, DeKeyser et al. 2015). These two factors indicate that the soil seed bank has a low potential to be used for passive restoration in undesirable areas in California Park (Von Blanckenhagen and Poschlod 2005, Bossuyt and Honnay 2008, Rayburn et al. 2016). Additionally, in sites that are dominated by non-native pasture grasses, the depletion of the non-native seed bank is warranted prior to restoration seeding (DeKeyser et al. 2015, Sebastian et al. 2017a, Clark et al. 2019).

Shrink-swell cracking on undesirable sites like "scabs" (D-2 and D-3) or *Wyethia*-dominated communities (D-4) may pose an additional limitation. Large shrink-swell (desiccation) cracks on Vertisols have been found to act as "seed traps" that can translocate seeds below successful germination depths (Chambers and MacMahon 1994, Benvenuti 2003, Burmeier et al. 2010). These shrink-swell cracks could act to further reduce the available seed bank in this ecosystem (Burmeier et al. 2010). A potential benefit is that these cracks may hold a high density of seeds with high species richness that, if germinated, could quickly return a large number and diversity of species to a site, but this is speculative (Burmeier et al. 2010).

The prevalence of non-native pasture grass seeds in sites classified as desirable is also of concern. The dominance of *Phleum pratense* L. (Timothy) seed in desirable site R-1 (71% of the overall seeds m^{-2}) had a large effect on reducing seed bank diversity (H) values of desirable sites ($H_{R1} = 1.08$ compared to $H > 2.0$ for the other two desirable sites). The presence of these non-native pasture grasses in both the aboveground vegetation and the soil seed bank in desirable sites (R-1 and R-3, see **Appendix Table 3**) may indicate that these sites are either vulnerable to a state-transition following disturbance or were in the *Phleum*-dominated understory alternate state described by Kachergis et al. (2014). This also brings up concern with hind-sight assumptions about broadly classifying areas as "desirable" and "undesirable". When sampling took place in 2017, these desirable sites were assumed to be in the "desirable" sagebrush with native herbaceous understory state. Follow-up aboveground vegetation sampling in 2019 indicated that this may have been an incorrect assumption. This can partially be attributed to the extent to which plant communities on Jokodowski soils throughout California Park were degraded and continue to exhibit the legacy impacts of historical overutilization (USDA Forest Service 2003, Kachergis et al. 2014).

This being said, in both the soil seed bank and aboveground vegetation, desirable sites tended to have more long-duration perennial grasses and shrubs present, compared to undesirable sites – except for site R1. Undesirable sites' aboveground vegetation tended to be dominated by species that senesce early in the year and have been considered undesirable by resource managers (USDA Forest Service 2003). These species included *Lomatium leptocarpum* (Torr. & A. Gray) J.M. Coult. & Rose (gumbo-lomatium – native perennial forb), *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears – native perennial forb), *Claytonia lanceolate* Pail. ex Pursh (lanceleaf springbeauty – native perennial forb), *Allium* spp. L. (onion – native perennial forb), and *Delphinium nuttallianum* Prtiz. ex Walp. (twolobe larkspur – perennial native forb) (USDA Forest Service personnel, personal communication). However, these species may provide important food sources for granivores and other non-game species (Crawford et al. 2004, Dumroese et al. 2016, Bates et al. 2017). Dominance of species such as these on undesirable sites could be attributed to the stressful edaphic conditions – high clay content and low litter cover limiting moisture availability and shrink-swell cracking that can cause root-shear and death of some perennial plants like shrubs (Kachergis et al. 2014, Malongweni et al. 2019). Interestingly, few of the species that were dominant in aboveground vegetation were found in the seed bank of degraded sites. This may have been due to a lack of viable seeds of these species in the seed bank or that their specific germination conditions were not met in the greenhouse propagation portion of the study. The samples were not assessed following termination of the germination trial to determine if viable seeds remained. Another possibility is high rates of granivory of the seeds of these plant species (Kemp 1989, Chambers and MacMahon 1994, Sartor and Marone 2010, Dumroese et al. 2016). Large numbers of birds were observed in the large *Wyethia* stands and eating their seeds in the summer of 2019.

Further study is needed to understand what factors continue to limit native species recovery across California Park. Few trends were evident when comparing soils data to seed bank composition. Soils (upper 5 cm analyzed) were found to be seleniferous ($\geq$ 2 mg kg^{-1} total Se) [**Appendix Table 2b**] across all sites in California Park, with values ranging from 2.2 – 4.6 mg/kg elemental Se. However, we were not able to assess which species of selenium were present and whether or not the selenium is bioavailable, which is an important factor in determining if selenium can be inhibiting plant growth (Schmidt 1982, Statwick and Sher 2017).

When it comes to management and ecological restoration implications, the soil seed bank of undesirable sites in California Park poses limitations by having low overall seed abundance relative to desirable sites and propagule pressure by the non-native pasture grasses *Phleum pratense* L. (Timothy) and *Poa pratensis* L. (Kentucky bluegrass). Therefore, similar to the findings of others, the soil seed bank likely cannot be relied on solely to passively restore undesirable areas (Bossuyt and Honnay 2008, Cowan and Anderson 2014, Rayburn et al. 2016). In areas like scabs or *Wyethia*-dominated sites, the relatively low abundance of seed in the seed bank poses a limitation that could potentially be addressed by supplemental seeding of native species. Whereas in sites dominated by non-native pasture grasses, a combination of a lack of native species seed and propagule pressure from the pasture grasses pose additional limitations (Lockwood et al. 2005). Such pasture grass-dominated sites would likely require treatments to both reduce the cover of such non-native perennial species and seed bank depletion to reduce the competitive effects and propagule pressure of these grasses (Shinn and Thill 2004, Dear et al. 2006, Sebastian et al. 2016, 2017a). Such a seed bank could potentially be depleted by utilizing pre-emergent herbicides such as indaziflam, imazapic, chlorsfuron, sulfometuron methyl, etc. Indaziflam is a particularly effective pre-emergent herbicide for use on annual grasses while not negatively impacting perennial forbs present in the aboveground vegetation, but there is no literature on its effectiveness on perennial grass seedlings (Sebastian et al. 2017a, 2017b, Clark et al. 2019). Such treatments could then be followed by supplemental site seeding with a high diversity seed mix of native grasses and forbs (Barr et al. 2017).

Incorporating shrubs seeds into a seed mix should be taken into careful consideration, especially on sites that exhibit shrink-swell cracking. Kachergis et al (2014) suggests that heavy clay sites – claypan ecological sites - that exhibit shrink-swell cracking are unsuitable for *Artemisia* spp. due to resulting root shear (Malongweni et al. 2019). Interestingly, the surface soil textures across the sites sampled – all in mapped Jokodowski soil units – had lower clay content than typical Jokodowski soils (**Appendix Table 2a**) and those reported by Kachergis et al. (2012) and it is unclear why these soil textures differ (Soil Survey Staff 2006). Information on the native seed abundance in these degraded areas could be important to help resource managers determine which species to include in a seed mix – those species already *in situ* could aid in planning the seed mix and seeding rate and reduce seeding costs.

A power analysis was conducted on the species richness of the soil seed bank to determine if our sample size was able to sufficiently capture a difference between the two site classifications. The small sample size, resultant means, and relatively large standard deviations within groups yielded little statistical power (Power = 0.05).

Salt Desert

Throughout the salt desert shrubland sites sampled, total seed abundance (seeds m^{-2}) was similar to or greater than other desert shrublands or Colorado Plateau ecosystems (Guo et al. 1998, Humphrey and Schupp 2001, Haight et al. 2019). Many non-native species were found across all site's seed banks and aboveground vegetation. In undesirable sites – those dominated by bare ground and non-native annuals – non-native species were prevalent and made up a majority of the soil seed bank. Whereas in less-disturbed desirable sites, native species made up a majority of the soil seed bank. Soil seed bank species richness and diversity did not differ between degraded and reference sites (**Table 2.4**). However, sample size was low (Desirable n = 2 and Undesirable n = 5) and resulted in limited statistical power. A power analysis was conducted on the species richness of the soil seed bank to determine if the sample size was able to sufficiently capture a difference between the two site classifications. The sample size, resultant means and relatively large standard deviation in the degraded sites group yielded less than ideal statistical power (Power = 0.369).

Relatively low overall abundance of native species' seed could indicate a limited regenerative potential of these species (Von Blanckenhagen and Poschlod 2005, Gioria and Pyšek 2015). Additionally, the presence of non-native annual seeds in desirable sites could indicate that these areas are vulnerable to non-native invasions following disturbance (Stohlgren et al. 2002, Chambers et al. 2014a, Grant-Hoffman et al. 2018, Jonas et al. 2018).

Interestingly, in undesirable sites, the abundance of native species seeds were similar to those in desirable sites (**Table 2.4**). However, the relative dominance of non-native annuals including *Bromus tectorum* L. (cheatgrass), *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass), *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill), and *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover) in the seed bank poses a constraint for ecological restoration of such sites (Humphrey and Schupp 2001, Pekas and Schupp 2013, Chambers et al. 2014c, 2014a, 2017, Sebastian et al. 2017a, Haight et al. 2019). Dominance of non-native species seed in undesirable sites also seemed to be influenced by or to influence certain edaphic conditions, displayed in the NMDS (**Figure 2.10**) (Blaisdell and Holmgren 1984, Wolkovich et al. 2010). These include high soil resource availability like relatively higher soil organic matter (OM) content, soil phosphorus (P), and soil nitrate-nitrogen (co-linear with OM) (**Figure 2.10**, and **Appendix Table 2c**) (Blaisdell and Holmgren 1984, Sperry et al. 2006, Wolkovich et al. 2010, Jonas et al. 2018). Soil conditions like high Sodium-Absorption Ratio (SAR) are also correlated with the dominance of *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover) in the seed bank at sites including Peeples Fire and Alkali Exclosure.

These factors indicate that the soil seed bank has a low potential to be used for passive restoration in salt desert shrublands of the Colorado Plateau, similar to the findings of others (Humphrey and Schupp 2001, Bossuyt and Honnay 2008, Pekas 2010). Additionally, in sites that are dominated by non-native annual species, the depletion of the non-native seed bank is warranted prior to restoration seeding to remove the propagule pressure of such non-native annuals (DeKeyser et al. 2015, Sebastian et al. 2017a, Clark et al. 2019).

Conclusions and Management Implications

Seed bank species richness and diversity values did not differ between sites classified as “Desirable” and “Undesirable” in either California Park or the salt desert. Thus, I reject Hypothesis 2.1. This study found that the soil seed bank is a potential constraint to ecological restoration of these high conservation-priority shrublands.

An important takeaway is that undesirable sites are not necessarily non-diverse (species-poor) relative to desirable sites when it comes to species richness. This was found in both the aboveground vegetation and soil seed bank composition in this study, and is similar to findings in other disturbed landscapes (Stohlgren et al. 1999, 2003, Rayburn et al. 2016). Although species richness numbers did not differ, the dominant species composition varied from site-to-site. Sites classified as undesirable in both ecosystems tended to have more short-lived plant species that are not typically "desirable" from a natural resource management standpoint (USDA Forest Service personnel, personal communication). In California Park, these included both short-lived perennials including *Lomatium leptocarpum* (Torr. & A. Gray) J.M. Coult. & Rose (gumbo-lomatium) and *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) and annuals including *Madia glomerata* Hook. (mountain tarweed) and *Collomia linearis* Nutt. (tiny trumpet). Although not typically considered "desirable", these species produce seeds that may be important for wildlife such as sage-grouse (Hermann 1966, Crawford et al. 2004, Bates et al. 2017). In the salt desert, these short-lived species were typically non-native weedy annuals including *Bromus tectorum* L. (cheatgrass), *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass), *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill), and *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover). Sites classified as desirable in both ecosystems tended to have more "desirable" long-lived perennial species, including shrubs, perennial grasses, and perennial forbs that provide groundcover year-round.

This study provides information to help resource managers make management decisions that incorporate the soil seed bank as a potential ecological constraint. The soil seed bank in degraded plant communities of California Park and the salt desert show limited potential for the seed bank to be relied upon for passive restoration to the desired shrub-dominated states. As few shrub seedlings were germinated from the seed banks and non-native species seed tended to make up large proportions, >30%, of the seed bank (Von Blanckenhagen and Poschlod 2005, Gioria and Pyšek 2015, Haussmann et al. 2019). It is unknown what an optimal or preferred abundance of native species in seed banks of shrublands are to achieve successful revegetation. Though it likely would need to be similar to high seeding rates suggested for the respective ecosystem (Blaisdell and Holmgren 1984, Barr et al. 2017, Baughman et al. 2017, Grant-Hoffman et al. 2018, Jonas et al. 2018). Sites where non-native species predominate in both the aboveground vegetation and the seed bank pose additional constraints on ecological restoration. Such highly competitive non-native species likely need to be addressed and treated – both the standing vegetation and the seed bank – to increase the likelihood of restoration success (Sebastian et al. 2016, 2017a, Clark et al. 2019, Schwartz-Lazaro and Copes 2019).

Seed bank depletion could be accomplished by utilizing pre-emergent herbicides including indaziflam, imazapic, chlorsfuron, sulfometuron methyl, rimsulfuron, etc. (Kyser et al. 2013, Sebastian et al. 2017a, 2017b, Clark et al. 2019). These herbicides have been found to be highly effective on annual grasses such as cheatgrass, however little research has been done on the impact of such herbicides on non-native pasture grass seedlings that are of concern in California Park such as Timothy, Kentucky bluegrass, and smooth brome (Shinn and Thill 2004, Dear et al. 2006, Kyser et al. 2013, Clark et al. 2019). Additionally, several native forb and grass species (both annuals and perennials) were found to occur in the soil seed bank across degraded sites that could potentially express themselves. Herbicides like this have been found to primarily target monocot seedlings and may not have a significant negative impact on native forb seedlings (Sebastian et al. 2017b, Clark et al. 2019). However, additional research is needed to determine the impacts of such pre-emergent herbicides on the seeds of native annual and perennial forbs and grasses. Such treatments, if effective, could then be followed by supplemental site seeding with a high diversity seed mix of native grasses and forbs (Barr et al. 2017). In California Park, such management actions could potentially reduce forage grass production and should be considered with the relevant stakeholders (Shinn and Thill 2004, Dear et al. 2006).

Due to the limited timeframe of this study, I was not able to assess the soil seed bank dynamics in desirable and undesirable sites of these shrublands. The temporal and spatial dynamics of the seed bank (and aboveground vegetation) are important factors when considering how the soil seed bank may impact ecological restoration and management activities in rangeland systems (Coffin and Lauenroth 1989, Bossuyt and Honnay 2008, Wright and Zuur 2014, Sebastian et al. 2017a).

The findings of this study provide resource managers useful information on the soil seed bank composition and potential to impact ecological restoration activities in these high conservation-priority shrublands. The seed bank is an important factor to consider when planning ecological restoration treatments in undesirable sites and if not addressed can likely reduce the likelihood of restoration success. Additional questions emerge from these findings. Are the native species seeds present in undesirable sites part of a persistent seed bank or are they part of a transient seed bank that is not able to express itself in the aboveground plant community? Can the seed abundance of native species *in situ* be incorporated into seed mix development and seeding rate calculations for follow-up treatments? Can the soil seed bank be an indicator of the current "state" or potential future "state" of these shrublands were disturbance to occur? Further study is warranted to address these knowledge gaps and increase the understanding of the impact of seed banks in ecological restoration of shrublands.

REFERENCES

Barga, S., and E. A. Leger. 2018. Shrub cover and fire history predict seed bank composition in Great Basin shrublands. Journal of Arid Environments 154:40–50.

Barr, S., J. L. Jonas, and M. W. Paschke. 2017. Optimizing seed mixture diversity and seeding rates for grassland restoration. Restoration Ecology 25:396–404.

Baskin, J. M., and C. C. Baskin. 1989. Physiology of dormancy and germination in relation to seed bank ecology. Ecology of Soil Seed Banks. Academci Press, Inc.

Bates, J. D., K. W. Davies, A. Hulet, R. F. Miller, and B. Roundy. 2017. Sage grouse groceries: Forb response to piñon-juniper treatments. Rangeland Ecology and Management 70:106–115.

Baughman, O. W., R. Burton, M. Williams, P. J. Weisberg, T. E. Dilts, and E. A. Leger. 2017. Cheatgrass die-offs: A unique restoration opportunity in northern Nevada. Rangelands 39:165–173.

Benvenuti, S. 2003. Soil texture involvement in germination and emergence of buried weed seeds. Agronomy Journal 95:191–198.

Benvenuti, S. 2007. Natural weed seed burial: effect of soil texture, rain and seed characteristics. Seed Science Research 17:211.

Blaisdell, J. P., and R. C. Holmgren. 1984. Managing intermountain rangelands - Salt-desert shrub ranges. General Technical Report - US Department of Agriculture, Forest Service.

Von Blanckenhagen, B., and P. Poschlod. 2005. Restoration of calcareous grasslands: the role of the soil seed bank and seed dispersal for recolonisation processes. Biotechnology, Agronomy, Society and Environment 9:143–149.

Bossuyt, B., and O. Honnay. 2008. Can the seed bank be used for ecological restoration? An overview of seed bank characteristics in European communities. Journal of Vegetation Science 19:875–884.

Burmeier, S., R. L. Eckstein, A. Otte, and T. W. Donath. 2010. Desiccation cracks act as natural seed traps in flood-meadow systems. Plant and Soil 333:351–364.

Chambers, J. C., J. L. Beck, J. B. Bradford, J. Bybee, S. Campbell, J. Carlson, T. J. Christiansen, K. J. Clause, G. Collins, M. R. Crist, J. B. Dinkins, K. E. Doherty, F. Edwards, S. Espinosa, K. A. Griffin, P. Griffin, J. R. Haas, S. E. Hanser, D. W. Havlina, K. F. Henke, J. D. Hennig, L. A. Joyce, F. F. Kilkenny, S. M. Kulpa, L. L. Kurth, J. D. Maestas, M. Manning, K. E. Mayer, B. A. Mealor, C. McCarthy, M. Pellant, M. A. Perea, K. L. Prentice, D. A. Pyke, L. A. Wiechman, and A. Wuenschel. 2017. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 1. Science basis and applications. USDA Forest Service - General Technical Report RMRS-GTR.

Chambers, J. C., B. A. Bradley, C. S. Brown, C. D'Antonio, M. J. Germino, J. B. Grace, S. P. Hardegree, R. F. Miller, and D. A. Pyke. 2014a. Resilience to stress and disturbance, and resistance to Bromus tectorum L. invasion in cold desert shrublands of western North America. Ecosystems 17:360–375.

Chambers, J. C., and J. A. MacMahon. 1994. A day in the life of a seed: Movements and fates of seeds and their implications for natural and managed systems. Annual Review of Ecology and Systematics 25:263–292.

Chambers, J. C., R. F. Miller, D. I. Board, D. A. Pyke, B. A. Roundy, J. B. Grace, E. W. Schupp, and R. J. Tausch. 2014b. Resilience and resistance of sagebrush ecosystems: Implications for state and transition models and management treatments. Rangeland Ecology and Management 67:440–454.

Chambers, J. C., D. A. Pyke, J. D. Maestas, M. Pellant, C. S. Boyd, S. B. Campbell, S. Espinosa, D. W. Havlina, and K. E. Mayer. 2014c. Using resistance and resilience concepts to reduce impacts of invasive annual grasses and altered fire regimes on the sagebrush ecosystem and greater sage-grouse: a strategic multi-scale approach. Gen. Tech. Rep. RMRS-GTR-326. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.

Clark, S. L., D. J. Sebastian, S. J. Nissen, and J. R. Sebastian. 2019. Effect of indaziflam on native species in natural areas and rangeland. Invasive Plant Science and Management 12:60–67.

Coffin, D. P., and W. K. Lauenroth. 1989. Spatial and temporal variation in the seed bank of a semiarid grassland. American Journal of Botany 76:53–58.

Cowan, O. S., and P. M. L. Anderson. 2014. The peninsula shale renosterveld of devil's peak, western cape: A study into the vegetation and seedbank with a view toward potential restoration. South African Journal of Botany 95:135–145.

Crawford, J. A., R. A. Olson, N. E. West, J. C. Mosley, M. A. Schroeder, T. O. M. D. Whitson, R. F. Miller, M. A. Gregg, C. S. Boyd. 2004. Ecology and management of sage-grouse and sage-grouse habitat. Journal of Range Management 57:2–19.

Dear, B. S., G. A. Sandral, and B. C. D. Wilson. 2006. Tolerance of perennial pasture grass seedlings to pre- and post-emergent grass herbicides. Australian Journal of Experimental Agriculture 46:637–644.

DeKeyser, E. S., L. A. Dennhardt, and J. Hendrickson. 2015. Kentucky bluegrass (Poa pratensis) invasion in the Northern Great Plains: A story of rapid dominance in an endangered ecosystem. Invasive Plant Science and Management 8:255–261.

Despain, D., P. Ogden, G. . Ruyle, and E. L. Smith. 1997. Some methods for monitoring rangelands and other natural area vegetation. (G. B. Ruyle, Ed.) Extention Report 9043. The University of Arizona, College of Agriculture, Tuscon, AZ.

Dumroese, R. K., T. Luna, J. R. Pinto, and T. D. Landis. 2016. Forbs: Foundation for restoration of monarch butterflies, other pollinators, and greater sage-grouse in the western United States. Natural Areas Journal 36:499–511.

Duniway, M. C., E. L. Geiger, T. J. Minnick, S. L. Phillips, and J. Belnap. 2018. Insights from long-term ungrazed and grazed watersheds in a salt desert Colorado Plateau ecosystem. Rangeland Ecology & Management 71:492–505.

Faist, A. M., S. Ferrenberg, and S. K. Collinge. 2013. Banking on the past: Seed banks as a reservoir for rare and native species in restored vernal pools. AoB PLANTS 5:1–11.

Gallagher, R. S. 2014. Seeds: The ecology of regeneration in plant communities. Page (R. S. Gallagher, Ed.). CABI, Wallingford.

Gioria, M., and P. Pyšek. 2015. The legacy of plant invasions: changes in the soil seed bank of invaded plant communities. BioScience 66:40–53.

Grant-Hoffman, M. N., A. Lincoln, and J. Dollerschell. 2018. Post-fire native seed use in Western Colorado: A look at burned and unburned vegetation communities. Natural Areas Journal 38:286–297.

Gremer, J. R., and D. L. Venable. 2014. Bet hedging in desert winter annual plants: Optimal germination strategies in a variable environment. Ecology Letters 17:380–387.

Guo, Q., P. W. Rundel, and D. W. Goodall. 1998. Horizontal and vertical distribution of desert seed banks: Patterns, causes, and implications. Journal of Arid Environments 38:465–478.

Haight, J. D., S. C. Reed, and A. M. Faist. 2019. Seed bank community and soil texture relationships in a cold desert. Journal of Arid Environments 164:46–52.

Haussmann, N. S., C. Delport, V. Kakembo, K. K. Mashiane, and P. C. le Roux. 2019. Restoration potential of invaded abandoned agricultural fields: What does the seed bank tell us? Restoration Ecology 27:813–820.

Hermann, F. J. 1966. Notes on Western range forbs. Agriculture Handbook No. 293. U.S. Department of Agriculture, Washington, D.C.

Hettinger, R. D., and L. N. R. Roberts. 2005. Lewis total petroleum system of the southwestern Wyoming province, Wyoming, Colorado, and Utah. Page USGS Digital Data Series.

Hopfensperger, K. N. 2007. A review of similarity between seed bank and standing vegetation across ecosystems. Oikos 116:1438–1448.

Humphrey, L. D., and E. W. Schupp. 2001. Seed banks of Bromus tectorum-dominated communities in the Great Basin. Western North American Naturalist 61:85–92.

Isselin-Nondedeu, F., and A. Bédécarrats. 2009. Assessing the dominance of Phleum pvatense cv. climax, a species commonly used for ski trail restoration. Applied Vegetation Science 12:155–165.

Jonas, J. L., M. N. Grant-Hoffman, and M. W. Paschke. 2018. Restoration of North American salt deserts: A look at the past and suggestions for the future. Ecological Restoration 36:177–194.

Jurado, E., and J. Flores. 2005. Is seed dormancy under environmental control or bound to plant traits? Journal of Vegetation Science 16:559–564.

Kachergis, E., M. E. Fernandez-Gimenez, and M. E. Rocca. 2012. Differences in plant species composition as evidence of alternate states in the sagebrush steppe. Rangeland Ecology & Management 65:486–497.

Kachergis, E., M. E. Rocca, and M. E. Fernández-Giménez. 2014. Long-term vegetation change provides evidence for alternate states in silver sagebrush. Rangeland Ecology & Management 67:183–194.

Kemp, P. R. 1989. Seed Banks and Vegetation Processes in Deserts. Pages 257–281 Ecology of Soil Seed Banks. Academci Press, Inc.

Knapp, C. N., and M. E. Fernandez-Gimenez. 2009. Knowledge in practice: Documenting rancher local knowledge in northwest Colorado. Rangeland Ecology & Management 62:500–509.

Kyser, G. B., R. G. Wilson, J. Zhang, and J. M. Ditomaso. 2013. Herbicide-assisted restoration of great basin sagebrush steppe infested with medusahead and downy brome. Rangeland Ecology and Management 66:588–596.

Lockwood, J. L., P. Cassey, and T. Blackburn. 2005. The role of propagule pressure in explaining species invasions. Trends in Ecology and Evolution 20:223–228.

Lusby, G. C. 1970. Hydrologic and biotic effects of grazing vs. non-grazing near Grand Junction, Colorado. Journal of Range Management 23:256.

Lusby, G. C. 1979. Effects of grazing on runoff and sediment yield from desert rangeland at Badger Wash in western Colorado, 1953-73. Geological Survey Water-Supply Paper 1532-1. Was.

Lusby, G. C., G. T. Turner, J. R. Thompson, and V. H. Reid. 1963. Hydrologic and biotic characteristics of grazed and ungrazed and watersheds of the Badger Wash basin in western Colorado, 1953-58. Geological Survey Water-Supply Paper 1532-B.

Maestas, J. D., S. B. Campbell, J. C. Chambers, M. Pellant, and R. F. Miller. 2016. Tapping soil survey information for rapid assessment of sagebrush ecosystem resilience and resistance. Rangelands 38:120–128.

Malongweni, S. O., Y. Kihara, K. Sato, T. Tokunari, T. Sobuda, K. Mrubata, and T. Masunaga. 2019. Impact of agricultural waste on the shrink–swell behavior and cracking dynamics of expansive soils. International Journal of Recycling of Organic Waste in Agriculture.

Martyn, T. E., J. B. Bradford, D. R. Schlaepfer, I. C. Burke, and W. K. Lauenroth. 2016. Seed bank and big sagebrush plant community composition in a range margin for big sagebrush. Ecosphere 7:1–11.

McCune, B., and J. Grace. 2002. Analysis of ecological communities. Page Analysis of ecological communities. MjM Software Design, Gleneden Beach, OR.

Mehlich, A. 1984. Mehlich 3 soil test extractant: A modification of Mehlich 2 extractant. Communications in Soil Science and Plant Analysis 15:1409–1416.

Meyer, S. E., S. L. Carlson, and S. C. Garvin. 1998. Seed germination regulation and field seed bank carryover in shadscale (Atriplex confertifolia: Chenopodiaceae). Journal of Arid

Environments 38:255–267.

Oksanen, J., Guillaume Blanchet, F., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P.R., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Szoecs, E., Wagner, H. (2019). vegan: Community Ecology Package. R package version 2.5-6. https://CRAN.R-project.org/package=vegan

Pakeman, R. J., J. L. Small, and L. Torvell. 2012. Edaphic factors influence the longevity of seeds in the soil. Plant Ecology 213:57–65.

Pekas, K. M. 2010. Seed pool dynamics of a Great Basin sagebrush community in the context of restoration. Utah State University.

Pekas, K. M., and E. W. Schupp. 2013. Influence of aboveground vegetation on seed bank composition and distribution in a Great Basin Desert sagebrush community. Journal of Arid Environments 88:113–120.

PRISM Climate Group, Oregon State University, http://prism.oregonstate.edu, created 4 Feb 2004.

R Core Team (2019). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Rawlins, J. K., B. A. Roundy, S. M. Davis, and D. Egget. 2012. Predicting germination in semi-arid wildland seedbeds. I. Thermal germination models. Environmental and Experimental Botany 76:60–67.

Rayburn, A. P., C. Schriefer, A. Zamora, and E. A. Laca. 2016. Seedbank-vegetation relationships in restored and degraded annual California grasslands: Implications for restoration. Ecological Restoration 34:277–284.

Sartor, C. E., and L. Marone. 2010. A plurality of causal mechanisms explains the persistence or transience of soil seed banks. Journal of Arid Environments 74:303–306.

Schmidt, V. A. 1982. Selenium in Agriculture:78–80.

Schwartz-Lazaro, L. M., and J. T. Copes. 2019. A review of the soil seedbank from a weed scientists perspective. Agronomy 9:1–13.

Sebastian, D. J., S. J. Nissen, J. R. Sebastian, and K. G. Beck. 2017a. Seed bank depletion: The key to long-term downy brome (Bromus tectorum L.) management. Rangeland Ecology and Management 70:477–483.

Sebastian, D. J., S. J. Nissen, J. R. Sebastian, P. J. Meiman, and K. G. Beck. 2017b. Preemergence control of nine invasive weeds with aminocyclopyrachlor, aminopyralid, and indaziflam. Invasive Plant Science and Management 10:99–109.

Sebastian, D. J., J. R. Sebastian, S. J. Nissen, and K. G. Beck. 2016. A potential new herbicide for invasive annual grass control on rangeland. Rangeland Ecology and Management 69:195–198.

Shefferson, R. P., T. Kull, M. J. Hutchings, M.-A. Selosse, H. Jacquemyn, K. M. Kellett, E. S. Menges, R. B. Primack, J. Tuomi, K. Alahuhta, S. Hurskainen, H. M. Alexander, D. S.

Anderson, R. Brys, E. Brzosko, S. Dostálik, K. Gregg, Z. Ipser, A. Jäkäläniemi, J. Jersáková, W. Dean Kettle, M. K. McCormick, A. Mendoza, M. T. Miller, A. Moen, D.-I. Øien, Ü. Püttsepp, M. Roy, N. Sather, N. Sletvold, Z. Štípková, K. Tali, R. J. Warren, and D. F. Whigham. 2018. Drivers of vegetative dormancy across herbaceous perennial plant species. Ecology Letters.

Shinn, S. L., and D. C. Thill. 2004. Tolerance of several perennial grasses to imazapic 1. Weed Technology 18:60–65.

Sperry, L. J., J. Belnap, and R. D. Evans. 2006. Bromus tectorum invastion alters nitrogen dynamics in an undisturbed arid grassland ecosystem. Ecology 87:603–615.

Statwick, J., and A. A. Sher. 2017. Selenium in soils of western Colorado. Journal of Arid Environments 137:1–6.

Stohlgren, T. J., D. T. Barnett, and J. T. Kartesz. 2003. The rich get richer: patterns of plant invasions in the United States. Frontiers in Ecology and Environment 1:11–14.

Stohlgren, T. J., D. Binkley, G. W. Chong, M. A. Kalkhan, L. D. Schell, K. A. Bull, Y. Otsuki, G. Newman, M. Bashkin, and S. Yowhan. 1999. Exotic plant species invade hot spots of native plant diversity. Ecological Monographs.

Stohlgren, T. J., G. W. Chong, L. D. Schell, K. A. Rimar, Y. Otsuki, M. Lee, M. A. Kalkhan, and C. A. Villa. 2002. Assessing vulnerability to invasion by nonnative plant species at multiple spatial scales. Environmental Management 29:566–577.

USDA Farm Service Agency (FSA) Aerial Photography Field Office. 2017 Release. Available at https://www.usgs.gov/centers/eros/science/usgs-eros-archive-products-overview?qt-science_center_objects=0#qt-science_center_objects

USDA Forest Service. 2003. Integrated management plan for the California Park Special Interest Area. Steamboat Springs, Colorado.

USDA, NRCS. 2019. The PLANTS Database (http://plants.usda.gov, 28 September 2019). National Plant Data Team, Greensboro, NC 27401-4901 USA.

Vandvik, V., K. Klanderud, E. Meineri, I. E. Måren, and J. Töpper. 2016. Seed banks are biodiversity reservoirs: Species-area relationships above versus below ground. Oikos 125:218–228.

Walck, J. L., S. N. Hidayati, K. W. Dixon, K. Thompson, and P. Poschlod. 2011. Climate change and plant regeneration from seed. Global Change Biology 17:2145–2161.

Wolkovich, E. M., D. A. Lispon, R. A. Virginia, K. L. Cottingham, and D. T. Bolger. 2010. Grass invasion causes rapid increases in ecosystem carbon and nitrogen storage in a semiarid shrubland. Global Change Biology 16:1351–1365.

Wright, B. R., and A. F. Zuur. 2014. Seedbank dynamics after masting in mulga (Acacia aptaneura): Implications for post-fire regeneration. Journal of Arid Environments 107:10–17.

CHAPTER III VARIABLE INFLUENCE OF SHRUB ISLANDS ON SOIL SEED BANK COMPOSITION IN THREE COLORADO SHRUBLANDS

Introduction

The distribution of the soil seed bank and dispersal of seeds throughout an ecosystem is influenced by numerous biotic and abiotic factors (Chambers and MacMahon 1994, Benvenuti 2007, Gallagher 2014). Dispersal of germinable seeds plays a significant role in community assembly and the spatial patterning of plant communities (Chambers and MacMahon 1994, Funk et al. 2008, Hulvey and Aigner 2014, Vandvik et al. 2016). Chambers and MacMahon (1994) describe two "Phases" of seed dispersal, Phase I dispersal being the direct movement of seed from the parent plant to a surface and Phase II dispersal being secondary horizontal and vertical movements of seed. Microsites such as soil cracks and depressions, animal mounds, "islands" of taller vegetation, and accumulations of coarse litter are important features that capture seeds during Phase II dispersal (Koniak and Everett 1982, Chambers and MacMahon 1994, Chambers 2000, Schlaepfer et al. 2014, Chambers et al. 2017, Barga and Leger 2018, Ward et al. 2018). Understanding the factors and site characteristics that influence seed bank distribution in shrublands is important for their ecological restoration and management (Pekas 2010, Vandvik et al. 2016, Chambers et al. 2017, Crist et al. 2019).

In shrubland ecosystems in the western United States, dominant shrub genera like *Artemisia* spp. L. (sagebrush) and *Atriplex* spp. L (saltbush) provide a critical layer of structural heterogeneity (Blaisdell and Holmgren 1984, Crawford et al. 2004, Jonas et al. 2018). Shrubs are known to influence habitat selection by animals, increase snow and moisture accumulation, increase litter accumulation, and create "islands of fertility" (Charley and West 1975, Crawford et al. 2004, Schlesinger et al. 2006, Ward et al. 2018, Filazzola et al. 2019). These physical factors can also act as a physical barrier capturing wind-blow seeds and influence the distribution of the soil seed bank (Chambers and MacMahon 1994, Pekas and Schupp 2013, Hulvey et al. 2017). Shrubs also influence subcanopy microclimate – reducing temperatures and wind velocities – and fertility levels that can provide favorable microsites for germination and growth (Chambers and MacMahon 1994, Schlesinger et al. 2006, McAdoo et al. 2013, Pekas and Schupp 2013, Filazzola et al. 2019, Gonzalez and Ghermandi 2019). Studies in desert shrubland and arid woodland environments found that areas beneath shrubs accumulate a greater density and species richness of seeds, relative to shrub-less interspaces (Koniak and Everett 1982, Chambers and MacMahon 1994, Castillo-Escrivà et al. 2019, Filazzola et al. 2019). Pekas and Schupp (2013) found that, in a Great Basin semi-arid sagebrush steppe, microhabitats below *Artemisia tridentata* Nutt. (big sagebrush) shrubs accumulated more seed for some but not all species compared to interspace areas.

Loss of structural heterogeneity is a common feature of degraded shrublands (Longland and Bateman 2002, Chambers et al. 2017). Shrublands that have been degraded through over-utilization, wildfire, exotic species invasion, are often structurally homogenous (Fuhlendorf and Engle 2001, Chambers et al. 2017). A common goal of the ecological restoration of shrublands is to restore structural and functional heterogeneity (Chambers et al. 2017, Crist et al. 2019, Paschke et al. 2019). Several techniques have been utilized, including the installation of “shrub mimics”, branch piles, or vertical mulch to create “restoration islands” (Chambers 2000, Hulvey et al. 2017, Naeth et al. 2018, Castillo-Escrivà et al. 2019).

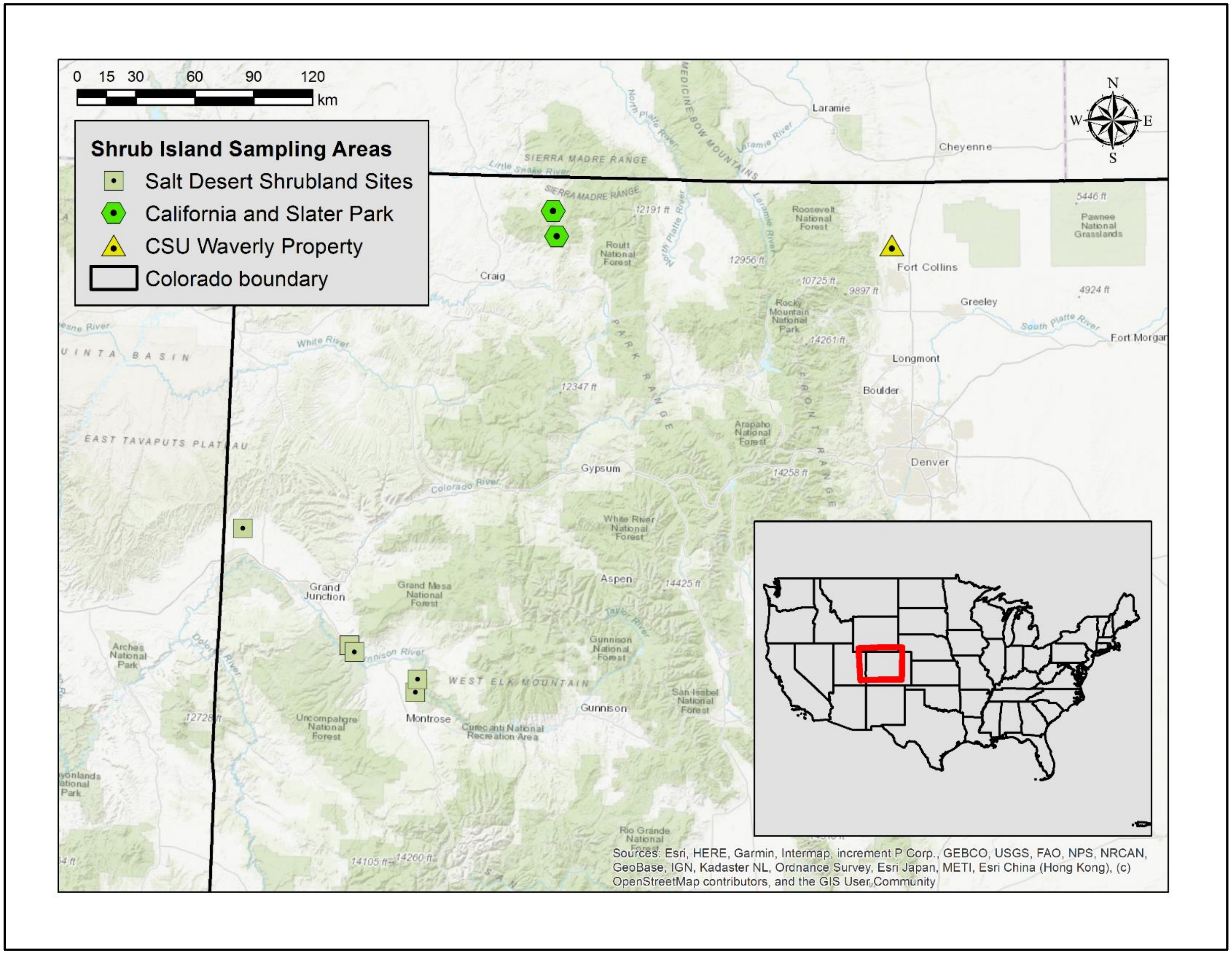
Few studies have evaluated the influence the presence of shrubs on seed banks in arid and semi-arid shrublands (Koniak and Everett 1982, Guo et al. 1998, Pekas and Schupp 2013, Barga and Leger 2018, Filazzola et al. 2019, Gonzalez and Ghermandi 2019). The objective of this study was to evaluate if "shrub islands" increase the species richness and abundance of seeds in the soil seed bank across several shrublands in Colorado, USA. I pose the questions, do shrub islands influence the distribution and composition of the soil seed bank and act as refugia for seeds? Do the areas within "shrub islands" have more species rich or diverse seed banks compared to shrub-less interspaces outside of shrub islands? If so, the use of artificial shrub islands could be a viable ecological restoration method in these shrubland ecosystems (Longland 1995, Chambers 2000, McAdoo et al. 2013, Boyd and Obradovich 2014, Hulvey et al. 2017, Castillo-Escrivà et al. 2019).

In this chapter, I hypothesize that the areas within shrub islands will have greater soil seed bank richness and diversity relative to areas between shrub islands where shrubs are lacking (Hypothesis 3.1). I also hypothesize that the areas within shrub islands will have greater soil seed bank abundance relative to areas between shrub islands where shrubs are lacking (Hypothesis 3.2).

Materials and Methods

Study Area Description

Shrub islands were identified and sampled in three shrublands throughout Colorado (**Figure 3.1**). These include the California Park Special Interest Area (SIA) and salt desert shrubland sites (see Chapter 1 "Study Sites"), along with the Colorado State University (CSU) Gabbard-Rutledge property near Waverly, Colorado (referred to as "Waverly"). California Park and the salt desert were selected to address soil seed bank knowledge gaps for the respective land management agencies and Waverly was added to facilitate an undergraduate research project and add a third shrubland to the study.



Sources: Esri, HERE, Garmin, Intermap, increment P Corp., GEBCO, USGS, FAO, NPS, NRCAN, GeoBase, IGN, Kadaster NL, Ordnance Survey, Esri Japan, METI, Esri China (Hong Kong), (c) OpenStreetMap contributors, and the GIS User Community

Figure 3.1: Map of shrub island soil seed bank sampling locations throughout Colorado

In California Park, sampling took place throughout California Park proper and the northern portion of the SIA known as Slater Park. This area is a high elevation silver sagebrush park in the Southern Rocky Mountains ecoregion, managed by the USDA Forest Service in Routt county Colorado (USDA Forest Service 2003, Kachergis et al. 2014). The dominant shrub species include *Artemisia cana* Pursh. (silver sagebrush) and *Artemisia tridentata* Nutt. spp. *vaseyana* (Rydb.) Beetle (mountain big sagebrush), with pockets of other shrub species including *Prunus virginiana* L. (chokecherry), *Symphoricarpos occidentalis* Hook. (western snowberry), and *Dasiphora fruticosa* (L.) Rydb. (shrubby cinquefoil). The shrubland areas sampled in California Park are currently grazed by domestic and wild ungulates, and the areas historically experienced heavy over-grazing and trampling (see Chapter 1 section "Study Sites"). The areas sampled were primarily located on upland locations in mapped units of the Jokodowski soil series (Fine, smectitic Typic Humicryerts) (Soil Survey Staff, 2006). The areas outside of the shrub islands (shrub island interspaces) were primarily vegetated with native grasses and forbs, and the non-native grass *Phleum pratense* L. (Timothy). Several shrub islands sampled in Slater Park exhibited features similar to "Mima mounds" that are thought to be created by pocket gophers (Cotter 1963, Gabet et al. 2014). The circular hillock features contained shrubs, along with native grasses and forbs in the understory. While the shrub-island interspace areas were dominated by both native and non-native grasses and *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears).

The salt desert shrubland sites (**Figure 3.1**) include areas studied in the preliminary soil seed bank study (see Chapter 2) including Alkali Exclosure and Badger Wash Reference, along with areas studied by Jonas et al. 2018 including the Peach Valley Exclosure, Ute, and Highway 50 Valley sites. The salt desert areas are located in the Colorado Plateau ecoregion in Mesa, Delta, and Montrose counties of western Colorado, managed by the Bureau of Land Management (BLM).

Dominant shrub species across these sites include several *Atriplex* L. species (saltbush), *Sarcobatus vermiculatus* (Hook.) Torr. (greasewood), and pockets of *Artemisia tridentata* Nutt. spp. *tridentata* (basin big sagebrush). The shrubland areas sampled in Peach Valley and Badger Wash Exclosure were in long-term (>50 year) grazing exclosures, and the remaining three sites were in areas that are able to be grazed by livestock. The shrub island interspaces across all sites were primarily bare ground.

The Waverly site is a 127 ha parcel of land located 20km north of Fort Collins, Colorado (**Figure 3.1**) in the Great Plains shortgrass steppe ecoregion in Larimer county (Barr et al. 2017). The sampling area is classified as a Loamy Plains ecological site and is dominated by the non-native pasture grass *Agropyrum cristatum* (L.) Gaertn. (crested wheatgrass) and *Chrysothamnus viscidiflorus* (Hook.) Nutt. (yellow rabbitbrush). The land was used for livestock grazing research from 1963 to approximately 2001. Following the cessation of grazing research, the land has been used for ecological restoration research. Since 2001, shrub species including *Chrysothamnus viscidiflorus* (Hook.) Nutt. (yellow rabbitbrush) and *Atriplex canescens* (Pursh) Nutt. (fourwing saltbush) were planted and established throughout the site. Shrub island interspace areas were primarily dominated by non-native pasture grasses.

Identifying Shrub Islands

For the purpose of the study, I aimed to develop a spatial definition to help guide sampling efforts. Here I define a “shrub island” as an aggregation of multiple individual shrubs, with an area between 5 m^2 and 100 m^2 (measured as an ellipse), and at least 3 meters distant to the nearest neighboring aggregation of shrubs. The spatial definition of a shrub island in this study was initially developed using GIS imagery analysis techniques. National Agriculture Imagery Program (NAIP) aerial imagery was analyzed using a supervised classification technique in ArcGIS v.10.6

(ESRI, release September 2017) and patch statistics generated using the program FRAGSTATS (McGarigal, Cushman, & Ene, 2012, USDA-FSA 2017). The output patch statistics from across two salt desert locations and California Park found that the mean shrub vegetation patch size was approximately 15.5 m^2 and mean Euclidean Nearest Neighbor distance to like patches was 4 meters. To attempt and account for the resolution of the imagery and create a flexible sampling criteria, the shrub island area range of 5 to 100 m^2 and minimum nearest neighbor distance of 3 meters was set. Aerial imagery was used to find areas where shrub islands that met the definition likely occurred. Potential shrub islands were identified and loaded into GPS units to guide us in the field. Then while on the ground we searched for shrub islands that met the definition, recorded their location, and sampled them.

Soil Seed Bank Sampling

Salt Desert sampling took place on June 12 and 14, 2018 at Badger Wash and Alkali Exclosure, and on May 23, 2019 at the Peach Valley exclosure, Ute, and Highway 50 Valley sites. Sampling in California and Slater Park took place on June 26 and 27, 2018. Sampling at Waverly took place on September 29 and 30, 2018. In the field, pre-identified shrub islands were verified and sampled or deemed to not meet the definition based upon their size and distance to nearest aggregation of shrubs. The area of each sampled shrub island was measured as an ellipse (**Equation 3.1**, **Figure 3.2**).

Equation 3.1: $\text{Area} = \pi * a * b$

a = major axis of the ellipse

b = minor axis of the ellipse

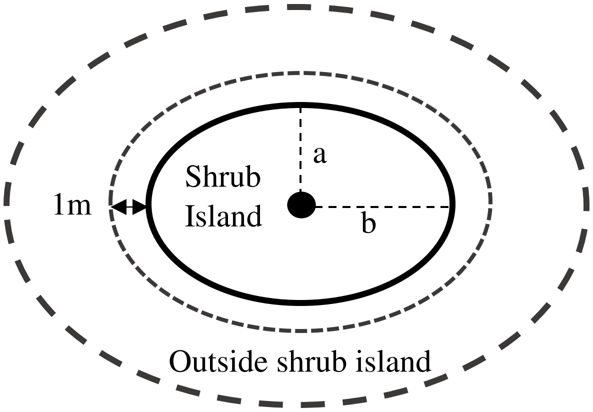


Figure 3.2: Diagram of a shrub island and the characteristics measured during sampling. “a” and “b” signify the major and minor axes, respectfully, used to calculate the area of the shrub island in Equation 3.1. The outside sample was sampled randomly at least 1 m outside of the perimeter of the shrub island.

The number of soil cores collected for each composite sample (both inside and outside shrub island samples) was proportional to the size of the shrub island, one core per 2 m^2 of shrub island area. To sample from inside the shrub island, a measuring tape was run through the canopy of the shrubs, either in a circular fashion for large islands, or straight through for smaller ones, staying at least 0.5 m inside from the edge of the perimeter of the shrub island. Points along the tape were chosen randomly using a random number table. A 1.5 cm diameter soil probe was used to collect soil cores to a 5 cm depth. The paired sample outside of the shrub island, or in the shrub island interspace, was sampled by randomly laying the measuring tape at least 1 m away from the shrub island. A new set of random numbers was selected as sampling points along the tape and the same number of cores taken inside the shrub island were sampled. These resulted in two composite sample units per shrub island – one “inside” sample unit and one “outside” sample unit that had an equivalent number of soil cores collected. Additional data collected for each shrub island include the dominant genera of the shrub island and the tallest living portion of shrub vegetation (measured to the nearest centimeter). In all, 40 shrub islands were sampled across California Park

(80 total sample units), 24 shrub islands across the five Salt Desert locations (48 total sample units), and 28 shrub islands at the Waverly property (56 total sample units). Following sampling, the soil samples were stored in a cooler before being transported back to the lab where they were stored at 5°C.

Seed Bank Propagation

After returning to the lab, the soil seed bank samples were weighed to the nearest gram and stored at 5°C prior to being processed for propagation. Prior to propagation, all samples were sieved through a 5.6 mm sieve to remove rocks (coarse fragments), living vegetation, large root fragments, and to break up soil aggregates. These samples were then propagated in a greenhouse with supplemental light 300 µmol m^{-2} sec^{-1} to extend the photoperiod to 16 hours. California Park samples were started on 11/02/2018 and terminated 8/26/2019, Waverly samples were started on 11/01/2018 and terminated 8/26/2019, and Salt Desert samples were started 5/29/2019 and terminated 9/19/2019.

Soil seed bank samples were propagated in round pots with a 15 cm diameter and 10 cm depth. Each pot had 7 cm of potting soil (ProMix BX) as a base and the field soil spread on top to a depth of 1 cm or less. If the field soil sample weighed greater than 150 g (larger shrub islands), the sample was evenly split into two pots (by weight) to achieve a depth less than 1 cm. To account for any contaminant seed that might blow into sample pots from the outside environment, control pots with 8 cm of potting soil were randomly distributed throughout the sample pots. The number of control pots used was equal to 10% of the number of sample units for each site (19 control pots total). Pots were set on top of Redi-Heat Propagation Mats (Model #: RHM2015, Phytotronics Inc) set to maintain a minimum soil temperature of 21°C based on estimated optimum germination temperature for most species in these ecosystems (Rawlins et al. 2012). Samples were randomly rotated on monthly basis to account for heterogeneity in greenhouse conditions. Samples were watered so that they remained moist throughout the germination and growth period.

Seedlings that germinated were recorded, identified, and removed from the pots. After all seedlings were removed and no new germination was recorded for two weeks, samples were allowed to dry-down for two weeks. The soil was then mixed by hand to break-up aggregates and fertilized with a 200ppm N solution of 21-18-18 to try and induce further germination (Chambers and MacMahon 1994, Walck et al. 2011). This method germinated additional seedlings from approximately 40% of the sample units for each study area. Samples were terminated after no new germination was detected for two weeks. Nomenclature for all plant species followed the USDA NRCS PLANTS Database (2019).

Statistical Methods

Prior to analysis, the mean number of contaminant plants of each species that germinated in control pots were subtracted from field soil values to reduce inflation of species richness values (Faist et al. 2013, Rayburn et al. 2016). These species included *Ericameria nauseosus* (Pall. ex Pursh) G.L. Nesom & Baird (rubber rabbitbrush), *Epilobium ciliatum* Raf. (fringed willowherb), *Populus deltoides* (Aiton) Eckenwalder (plains cottonwood), *Oxalis corniculata* L. (creeping woodsorrel), and *Miscanthus* spp. Andersson (silvergrass). To facilitate soil seed bank richness and diversity comparisons among sample units, seed bank density is expressed as seeds per m^2 of soil sampled.

My hypotheses were tested by examining soil seed bank composition – species richness, Shannon-Wiener diversity [H], and seed abundance – inside versus outside of shrub islands. Each site (i.e. California Park, salt desert, and Waverly) was tested independently from the others. Analyses were conducted using the statistical program R v3.6.1 (R Core Team, 2019) using the specnumber and diverse functions in the Vegan R-package (Oksanen et al. 2013). To test Hypothesis 3.1, a paired t-test approach was used to test the mean difference of the paired richness and diversity measures inside and outside each unique shrub island. To test Hypothesis 3.2, a paired t-test approach was used to test the mean difference of the paired total seed abundance measures inside and outside of each unique shrub island. Species seed abundance were also lumped by functional group – shrub, sub-shrub, native grass, native forb, and non-native – to compare seed bank abundance at a finer scale for each shrubland. This was done to account for the variability in seed bank species composition across shrub islands within one ecosystem and for comparison across shrublands.

Normality of the difference in means (inside versus outside sample) was checked using the Shapiro-Wilk normality test. If normality was violated, then a Wilcoxon signed-rank test – a non-parametric equivalent – was used and noted. The Wilcoxon signed-rank test compares whether the median of a sample population is different from zero and retains information about the magnitude of the difference. In this case, species richness, diversity, and seed abundance of the inside shrub island samples compared to the outside shrub island samples. Measurements where richness, diversity, or seed abundance inside a shrub island are greater than outside are assigned a positive value and where measurements outside a shrub island are greater than inside are assigned a negative value. An $\alpha = 0.05$ was used for significance testing.

Results

Shrub Island Seed Bank Species Richness

Samples from California Park had seedlings from 34 species germinate from both inside and outside shrub island samples, including 28 native and 6 non-native species. Samples from the salt desert had seedlings from 14 species germinate from both inside and outside shrub island samples, including 8 native and 6 non-native species. Samples from Waverly had seedlings from 18 species germinate from both inside and outside shrub island samples, including 11 native and 7 non-native species. Differences in species richness values between inside versus outside samples in California Park were not normally distributed and a Wilcoxon signed rank test was used. Differences in species richness between inside versus outside samples were normally distributed for the salt desert and Waverly samples and a paired t-test was used. Species richness did not statistically differ between inside and outside shrub island samples in the three shrublands at an $\alpha = 0.05$ (**Table 3.1**).

Table 3.1: Seed bank species richness and Shannon-Wiener diversity (H) inside and outside of shrub islands at three Colorado shrublands. Data are means ± standard error. Comparison between paired sample means evaluate using a Student's paired t-test. Where the difference in means inside vs. outside shrub islands was non-normal, the Wilcoxon signed-rank test was used . $^{*}p \leq 0.05$; $^{**}p \leq 0.01$; $^{\dagger}p \leq 0.10$.

Site	Sample Location	Seed Bank Richness	Seed Bank Diversity
California Park	Inside (n = 40)	2.4 ± 0.2	0.676 ± 0.085
	Outside (n = 40)	2.0 ± 0.3	0.490 ± 0.086
		Wilcoxon	Wilcoxon
		$p = 0.255$	$p = 0.0869^{\dagger}$
Salt Desert	Inside (n = 24)	1.2 ± 0.2	0.299 ± 0.084
	Outside (n = 24)	0.9 ± 0.2	0.123 ± 0.054
		t = 1.37	t = 2.12
		$p = 0.183$	$p = 0.0449^{*}$
Waverly	Inside (n = 28)	0.8 ± 0.2	0.162 ± 0.061
	Outside (n = 28)	0.5 ± 0.1	0.072 ± 0.040
		t = 1.43	t = 1.28
		$p = 0.164$	$p = 0.213$

Species commonly (>10% of sample units) found both inside and outside shrub islands in California Park included *Phleum pratense* L. (Timothy – non-native pasture grass) and *Androsace septentrionalis* L. (pygmyflower rockjasmine – native annual forb). Common species found primarily in inside shrub island seed banks included *Artemisia cana* Pursh (silver sagebrush –

native shrub) and *Potentilla* spp. L. (cinquefoil – native perennial forb). Common species found primarily in the interspaces included *Achnatherum lettermannii* (Vasey) Barkworth (Letterman's needlegrass – native perennial grass).

Species commonly found both inside and outside shrub islands in the salt desert included *Descurainia pinnata* (Walter) Britton (western tansymustard – native annual forb), *Amaranthus albus* L. (prostrate pigweed – non-native annual forb), and *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover – non-native annual forb). Common species found primarily in inside shrub island seed banks in the salt desert included *Gutierriezia sarothrae* (Pursh) Britton & Rusby (broom snakeweed – native sub-shrub). No common species were found only in interspace seed bank samples in the salt desert. At the Waverly site, species composition of the seed banks varied greatly from shrub island to shrub island and few species were found consistently between seed banks.

Shrub Island Seed Bank Diversity

Differences in seed bank Shannon-Wiener diversity (H) were not normally distributed for California Park and the Wilcoxon signed rank test was used. Differences in seed bank diversity were normally distributed for the salt desert and Waverly seed bank samples and a paired t-test was used. Seed bank diversity did not differ at California Park and Waverly at an $\alpha = 0.05$. However, seed bank diversity of California Park samples neared statistical difference (p-value = 0.0869) with inside seed bank samples having generally greater diversity. Seed bank diversity was significantly greater inside of shrub islands in the salt desert (**Table 3.1**; paired t = 2.12, p-val = 0.0449). Diversity (H) values for Waverly and the salt desert shrub islands were zero-inflated due to a large number of samples that had seedlings from only one species emerge (which resulted in an H = 0 for many samples). This zero-inflation issue thus yielded low statistical power.

Shrub Island Seed Bank Abundance

Total seed abundance did not statistically differ between inside and outside shrub islands in the three shrublands (**Table 3.2**). California Park seed banks inside of shrub islands averaged 2296 seeds m^{-2} and outside of shrub islands averaged 2705 seeds m^{-2}, with outside seed banks being much more variable (**Table 3.2**, **Figure 3.3**). Salt desert seed banks averaged 1180 seeds m^{-2} inside shrub islands and 861 seeds m^{-2} outside shrub islands (**Figure 3.4**). Waverly seed banks averaged 984 seeds m^{-2} inside shrub islands and 613 seeds m^{-2} outside shrub islands (**Figure 3.5**). From the salt desert (n = 24), five paired areas from the salt desert and six from Waverly (n = 28) had no seedlings emerge from either the inside or outside samples.

Table 3.2: Seed abundance (seeds m^{-2}) broken down by plant functional group inside and outside shrub islands across the three shrublands sampled in Colorado. Means reported ± standard error. N/A signifies that no seeds of the functional group were found in samples. Statistical significance of comparison between paired sample mean differences using a Student's paired t-test. If normality in the difference of means was violated, the Wilcoxon signed-rank test was used and noted: * $p \leq 0.05$; ** $p \leq 0.01$; † $p \leq 0.10$.

Site	**Sample Location**	**Total Abundance (seeds m^{-2})**	**Shrub (seeds m^{-2})**	**Subshrub (seeds m^{-2})**	**Native Grass (seeds m^{-2})**	**Native Forb (seeds m^{-2})**	**Non-native (seeds m^{-2})**
California Park	Inside	2296 ± 310	228 ± 83	N/A	239 ± 144	1057 ± 185	772 ± 168
(n = 40)	Outside	2705 ± 739	57 ± 48	N/A	301 ± 109	640 ± 152	1707 ± 738
		t = -0.497	t = 1.72		t = -0.333	Wilcoxon	t = -1.22
		$p = 0.622$	$p = 0.0927^{\dagger}$		$p = 0.741$	$p = 0.0317^{*}$	$p = 0.231$
Salt Desert	Inside	1180 ± 367	N/A	N/A	41 ± 29	482 ± 183	656 ± 344
(n = 24)	Outside	861 ± 238	N/A	N/A	0	389 ± 163	472 ± 193
		t = 0.741			t = 1.39	t = 0.362	t = 0.510
		$p = 0.464$			$p = 0.179$	$p = 0.721$	$p = 0.615$
Waverly	Inside	984 ± 252	97 ± 55	0	0	263 ± 116	625 ± 235
(n = 28)	Outside	613 ± 192	34 ± 34	155 ± 94	40 ± 40	108 ± 60	276 ± 137
		t = 1.20	t = 0.945	t = -1.66	t = -1.000	t = 1.29	t = 1.32
		$p = 0.241$	$p = 0.353$	$p = 0.109$	$p = 0.326$	$p = 0.209$	$p = 0.198$

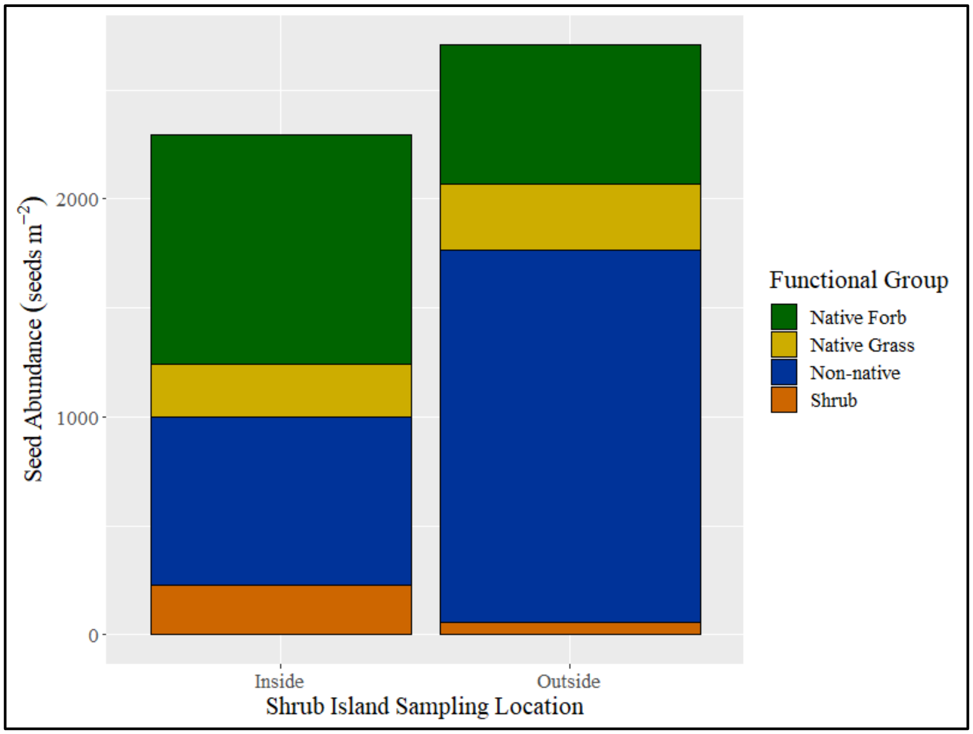


Figure 3.3: California Park mean seed abundance (seeds m^{-2}), broken down by functional group, comparing seed banks inside of shrub islands (Inside) and in the shrub-less interspaces (Outside). Total abundance ($t = -0.497$, $p = 0.622$).

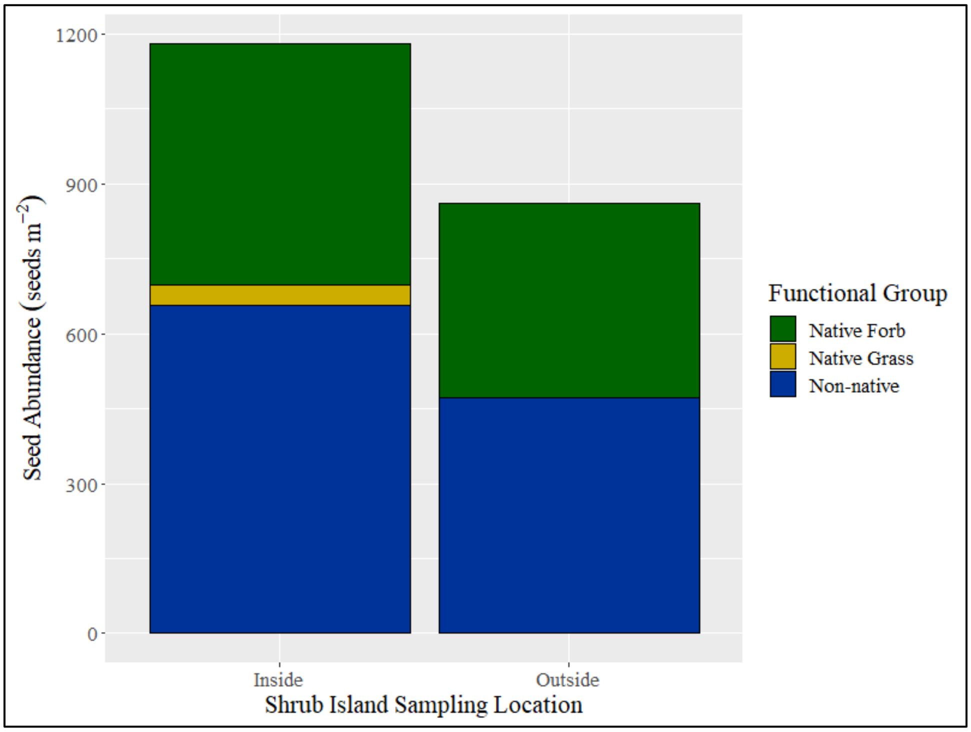


Figure 3.4: Salt desert mean seed abundance (seeds m^{-2}), broken down by functional group, comparing seed banks inside of shrub islands (Inside) and in the shrub-less interspaces (Outside). Total abundance ($t = 0.741$, $p = 0.464$).

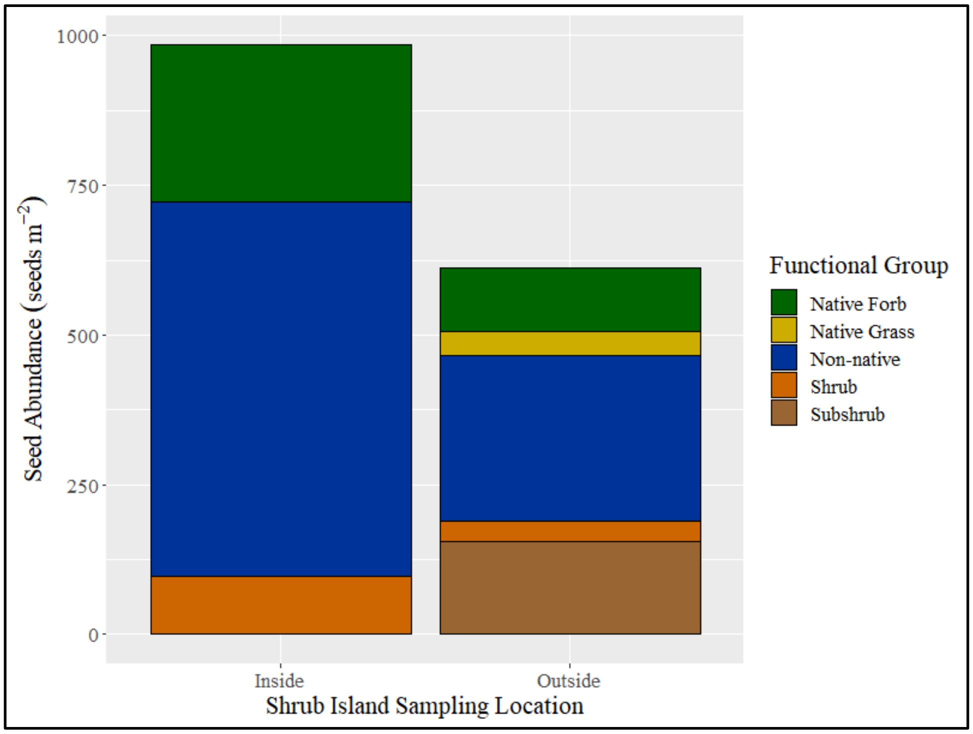


Figure 3.5: Waverly mean seed abundance (seeds m-2), broken down by functional group, comparing seed banks inside of shrub islands (Inside) and in the shrub-less interspaces (Outside). Total abundance (t = 1.20, p = 0.241).

Non-native species dominated the seed banks of the three shrublands (**Table 3.2**, **Figures 3.3 – 3.5**). Native forbs was the next largest functional group emerging from seed banks inside and outside of shrub islands. Shrub seeds germinated from both inside and outside samples in California Park (*Artemisia cana* Pursh– silver sagebrush) and Waverly (*Chyrsothamnus viscidiflorus* (Hook.) Nutt. – yellow rabbitbrush and *Ericameria nauseosa* (Pall. ex Pursh) G.L. Nesom & Baird – rubber rabbitbrush). No shrub seeds were found in the seed bank of the salt desert samples.

Significant differences in seed abundance by functional group were found in California Park seed banks (**Table 3.2**). Native forb seed abundance was greater (Wilcoxon signed rank test) inside of shrub islands than outside (p-value = 0.0317). Shrub seed abundance neared statistical significance (paired t-test) with shrub seeds tending to be more abundant inside of shrub islands (t

= 1.72, p-value = 0.0927). No statistical differences in seed abundance by functional group were found in either salt desert or Waverly seed bank samples. Although there were functional group seeds that only occurred in either inside or outside samples, their occurrence in relatively few samples resulted in zero-inflation and limited statistical inference (**Table 3.2**, **Figure 3.4**, **Figure 3.5**).

Discussion

Across three shrublands sampled in Colorado, no differences in germinable soil seed bank richness inside versus outside of shrub islands were found. Seed bank Shannon-Winer diversity (H) levels inside and outside shrub islands did not differ for the rabbitbrush-dominated Waverly shrubland. Seed bank diversity was significantly greater inside of shrub islands in the salt desert and neared statistical significance in California Park, but neither were paired with greater species richness. Thus, I reject the hypothesis that shrub islands have greater soil seed bank species richness and diversity, relative to shrub-less interspaces (Hypothesis 3.1) for these three shrublands. There also were no differences in overall seed abundance inside and outside shrub islands in the three shrublands. Thus, I reject the hypothesis that shrub islands have greater seed abundance, relative to shrub-less interspaces (Hypothesis 3.2). A caveat for these rejections is that the sample size for each shrubland was relatively low, with paired sample sizes between 24 and 40.

These data do not support the notion that “shrub islands” act as seed refugia or seed bank hotspots in the three shrublands studied. This is interesting, as these findings are contrary to other studies – primarily in desert shrublands and arid woodlands – that found seed banks were more species rich and had greater seed abundance beneath shrubs (Koniak and Everett 1982, Chambers and MacMahon 1994, Longland 1995, Chambers 2000, Longland and Bateman 2002, Wang et al.

2011, Barga and Leger 2018, Filazzola et al. 2019). However, the literature is mixed. Results from this study are similar to the findings of Pekas and Schupp (2013), who found no influence of shrubs or "microhabitat phase" on seed bank richness and overall density in a Great Basin sagebrush steppe (*Artemisia tridentata* Nutt. ssp. *wyomingensis* Beetle & Young [Wyoming big sagebrush]).

Although these results found that shrub islands did not increase both seed bank species richness *and* diversity, the finding that seed bank species diversity was greater inside of shrub islands in the salt desert and neared statistical significance in California Park is noteworthy. These results suggest that, in these two shrubland ecosystems, seed abundance of uncommon species was more even inside of shrub islands as opposed to the interspaces. This is similar to the results of Barga and Leger (2018) who found that *Artemisia tridentata* Nutt. ssp. *wyomingensis* Beetle & Young (Wyoming big sagebrush) shrubs had a large effect on increasing the presence of "rare" species in the seed bank.

Germinable seed bank diversity (H) both inside and outside of shrub islands (in all three shrublands) was quite low. This was due in part to there being few seedlings emerging from samples (especially in the salt desert and Waverly) and zero-inflation when a sample had only a single species emerge from the seed bank (resulting in an $H = 0$). The Shannon-Weiner index is also sensitive to changes in the proportions of rare species and decreases when few species make up a majority of the seed abundance. In both the salt desert and California Park, several "outside" samples had seed banks made up of few species' seeds, with one species seed making-up large proportions of the seed bank. These species that made up disproportionately high seed abundance tended to be seeds of non-native species. In California Park, these included species such as *Phleum pratense* L. (Timothy – non-native pasture grass) and *Spergularia rubra* (L.) J. Presl & C. Presl (red sandspurry – non-native annual forb) that were found in high (and highly variable) abundances. In the salt desert, this species was *Halogeton glomerata* (M. Bieb.) C.A. Mey. (saltlover – non-native annual forb).

Germinable seed bank compositional differences by plant species functional group were nuanced for California Park and are similar to the findings by Pekas and Schupp (2013). In the silver sagebrush parkland of California Park, overall seed abundance did not differ inside and outside shrub islands. However, native forb species seed were found to be statistically more abundant inside of shrub islands (**Table 3.2**). Shrub seed abundance also tended to be greater inside of shrub islands and neared statistical significance (p-value = 0.0927). Pekas and Schupp (2013) found that only shrub seed densities were significantly greater beneath shrubs in Great Basin sagebrush steppe.

Even though seeds of functional groups such as subshrubs (*Artemisia frigida* Willd. – prairie sagewort) and native grasses only occurred in shrub-less interspaces at the Waverly site, general lack of seeds in the seed bank resulted in low statistical power. The functional group breakdown of the seed bank also suggests that, in these three ecosystems, non-native species seed make up a large component of the seed bank and that their distribution is not influenced by the presence of shrub islands. This is similar to several other studies across shrub-dominated ecosystems (Chambers and MacMahon 1994, Humphrey and Schupp 2001, Pekas and Schupp 2013, Barga and Leger 2018, Filazzola et al. 2019).

Overall, few seedlings germinated from samples in both the salt desert and Waverly. Across the 24 shrub islands (48 total sample units) sampled in the salt desert, 97 seeds germinated. Across the 28 shrub islands (56 total sample units) sampled at Waverly, 42 seeds germinated. Whereas in California Park, across the 40 shrub islands sampled (80 total sample units), 355 seeds germinated. There are potential explanations for the lack of germinable seeds found in the seed bank, but these are speculative. Perhaps the most plausible is that there just were not many seeds present in the seed banks of the salt desert and Waverly. Another potential reason is the influence of sampling times. In the salt desert, 15 of the 24 shrub islands were sampled in the summer of 2018, and nine sampled in the spring of 2019 – during a wetter than average spring that resulted in a "super bloom" (**Appendix Table 1**). The differences in sampling dates likely introduced error, as seed banks can show large temporal fluctuations (Coffin and Lauenroth 1989, LaForgia et al. 2018). The above-average precipitation in spring of 2019 may have caused many of the seeds of plants in this shrubland system to germinate and the seed bank to be depleted. Four of the nine (44%) of the shrub islands sampled in 2019 had no seedlings emerge from either inside or outside seed bank samples. The remaining five shrub islands had only 12 seedlings germinated between them. Sampling at Waverly took place in the late summer and early fall of 2018, following seed set and rain of most plants. However, seed scarification of that years' seeds may not have occurred, and seeds present in the seed bank may still have been dormant (Jurado and Flores 2005, Sartor and Marone 2010). The soil samples were not assessed following the germination study to determine if additional seeds remained in the soil that did not germinate.

The Waverly site also has low aboveground plant diversity, due in part to historical management, and is dominated by non-native grasses such as *Agropyrum cristatum* (L.) Gaertn. (crested wheatgrass). The low diversity of the plant community may simply be reflected in the soil seed bank, as seed bank and aboveground plant community composition have been found to be more similar in grasslands such as shortgrass steppe (Coffin and Lauenroth 1989, Hopfensperger 2007) where the Waverly site is located, relative to later-successional shrublands (Bossuyt and Honnay 2008, Vandvik et al. 2016).

Comparison of seed bank abundances across these shrublands should be done with caution, as the age of shrubs and the successional stages of the sampling sites differ from one another. Although the age of shrub individuals was not directly assessed, generalizations can be made. In California Park, historical management throughout the park reduced shrub cover greatly through the 1950's and 1960's, but shrubs recovered in many areas in the 1980's (USDA Forest Service 2003, Kachergis et al. 2014). Additionally, many shrub islands were associated with "Mima mound" features that are thought to take decades or more to be created by burrowing mammals (Gabet et al. 2014). In the salt desert, the shrubs sampled were likely the oldest of the three shrublands sampled. Jonas et al. (2018) describes that the regeneration of mature salt desert shrubs can take 100 years or more. The dominant shrubs at Waverly – *Chrysothamnus viscidiflorus* (Hook.) Nutt. (yellow rabbitbrush) – are an early successional species and likely the youngest of the three shrublands sampled, having increased primarily after 2001 (Tilley and St. John 2012).

Intermediate and late-successional areas and associated shrub islands (i.e. California Park and the salt desert) may have had time to influence the understory communities and for more variable plant communities and seed banks to develop (Hopfensperger 2007, Bossuyt and Honnay 2008, Vandvik et al. 2016, Barga and Leger 2018). Whereas the early successional plant community and seed bank at Waverly is likely more homogenous and has had less time to influence seed bank development (Hopfensperger 2007, Bossuyt and Honnay 2008, Barga and Leger 2018).

Conclusions and Management Implications

The presence of shrub islands had variable effects on germinable soil seed bank composition between the three shrublands studied. Shrub islands had no significant effect on seed bank species richness. Seed bank Shannon-Wiener diversity (H) tended to be greater inside of shrub islands versus outside shrub islands in the salt desert and California Park, but was not different in the earlier-successional shrubland of Waverly. Total seed abundance was not increased by the presence of shrub islands in the three shrublands. Though contrary to studies of desert shrublands and arid pinon-juniper woodlands, these findings are consistent with those from the Great Basin sagebrush steppe (Koniak and Everett 1982, Chambers and MacMahon 1994, Pekas and Schupp 2013, Barga and Leger 2018, Castillo-Escrivà et al. 2019, Filazzola et al. 2019). Seed dispersal is highly variable and species-dependent, and multiple factors simultaneously influence seed dispersal and seed bank development (Chambers and MacMahon 1994, Bossuyt and Honnay 2008, Pekas and Schupp 2013, Gallagher 2014, Barga and Leger 2018). The general lack of germinable seed bank differences suggest that other factors are driving the dispersal and distribution of seed banks.

These results suggest that shrub islands are not primary drivers of seed bank development. The implications of these findings are that the establishment of "shrub mimics", branch piles, establishing pockets of shrubs may not help to focus the accumulation of germinable seeds to facilitate passive restoration. However, in California Park, it does appear that shrub islands have an effect on accumulating native forb seeds. As such, these methods could be used to achieve such a management objective which has been receiving greater attention (Dumroese et al. 2016). Forbs and forb seeds are an important consideration for sage-grouse management, as forb seeds are a primary food source (Dumroese et al. 2016, Bates et al. 2017). This study cannot elucidate the mechanisms behind the accumulation or the fate of those native forb seeds. However, this may be a factor of the relative age of the shrub islands and the length of time that shrub vegetation have been able to influence understory communities and their associated seed banks (Koniak and Everett 1982, Vandvik et al. 2016, Hulvey et al. 2017, Barga and Leger 2018).

Other studies have found that use of branch piles, “vertical mulch”, and “shrub simulates” can be used to benefit other aspects of ecological restoration. These include facilitating seedling survival and influencing the distribution of target species seeds that are dispersed by birds (Chambers 2000, Castillo-Escrivà et al. 2019, Filazzola et al. 2019). So although they may not accumulate greater species richness of seeds or total seed density, there are other benefits of the use of shrub islands in ecological restoration (Chambers 2000, Boyd and Obradovich 2014, Hulvey et al. 2017).

REFERENCES

Barga, S., and E. A. Leger. 2018. Shrub cover and fire history predict seed bank composition in Great Basin shrublands. Journal of Arid Environments 154:40–50.

Barr, S., J. L. Jonas, and M. W. Paschke. 2017. Optimizing seed mixture diversity and seeding rates for grassland restoration. Restoration Ecology 25:396–404.

Bates, J. D., K. W. Davies, A. Hulet, R. F. Miller, and B. Roundy. 2017. Sage grouse groceries: Forb response to piñon-juniper treatments. Rangeland Ecology and Management 70:106–115.

Benvenuti, S. 2007. Natural weed seed burial: effect of soil texture, rain and seed characteristics. Seed Science Research 17:211.

Blaisdell, J. P., and R. C. Holmgren. 1984. Managing intermountain rangelands - Salt-desert shrub ranges. General Technical Report - US Department of Agriculture, Forest Service.

Bossuyt, B., and O. Honnay. 2008. Can the seed bank be used for ecological restoration? An overview of seed bank characteristics in European communities. Journal of Vegetation Science 19:875–884.

Boyd, C. S., and M. Obradovich. 2014. Is pile seeding wyoming big sagebrush (Artemisia tridentata subsp. wyomingensis) an effective alternative to broadcast seeding? Rangeland Ecology and Management 67:292–297.

Castillo-Escrivà, A., G. M. López-Iborra, J. Cortina, and J. Tormo. 2019. The use of branch piles to assist in the restoration of degraded semiarid steppes. Restoration Ecology 27:102–108.

Chambers, J. C. 2000. Seed movements and seedling fates in disturbed sagebrush steppe ecosystems: Implications for restoration. Ecological Applications 10:1400–1413.

Chambers, J. C., J. L. Beck, J. B. Bradford, J. Bybee, S. Campbell, J. Carlson, T. J. Christiansen, K. J. Clause, G. Collins, M. R. Crist, J. B. Dinkins, K. E. Doherty, F. Edwards, S. Espinosa, K. A. Griffin, P. Griffin, J. R. Haas, S. E. Hanser, D. W. Havlina, K. F. Henke, J. D. Hennig, L. A. Joyce, F. F. Kilkenny, S. M. Kulpa, L. L. Kurth, J. D. Maestas, M. Manning, K. E. Mayer, B. A. Mealor, C. McCarthy, M. Pellant, M. A. Perea, K. L. Prentice, D. A. Pyke, L. A. Wiechman, and A. Wuenschel. 2017. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 1. Science basis and applications. USDA Forest Service - General Technical Report RMRS-GTR.

Chambers, J. C., and J. A. MacMahon. 1994. A day in the life of a seed: Movements and fates of seeds and their implications for natural and managed systems. Annual Review of Ecology and Systematics 25:263–292.

Charley, J. L., and N. E. West. 1975. Plant-induced soil chemical patterns in some shrub-dominated semi-desert ecosystems of Utah. Journal of Ecology 63:945–963.

Coffin, D. P., and W. K. Lauenroth. 1989. Spatial and temporal variation in the seed bank of a semiarid grassland. American Journal of Botany 76:53–58.

Cotter, J. F. 1963. Causation and plant succession in disturbed areas of southwestern Montana. Montana State University.

Crawford, J. A., R. A. Olson, N. E. West, J. C. Mosley, M. A. Schroeder, T. O. M. D. Whitson, R. F. Miller, M. A. Gregg, C. S. Boyd. 2004. Ecology and management of sage-grouse and sage-grouse habitat. Journal of Range Management 57:2–19.

Crist, M. R., J. C. Chambers, S. L. Phillips, K. L. Prentice, and L. A. Wiechman. 2019. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 2 . Management applications. General Technical Report RMRS-GTR-389. Fort Collins, CO.

Dumroese, R. K., T. Luna, J. R. Pinto, and T. D. Landis. 2016. Forbs: Foundation for restoration of monarch butterflies, other pollinators, and greater sage-grouse in the western United States. Natural Areas Journal 36:499–511.

Environmental Systems Research Institute (ESRI). (2017). ArcGIS Release 10.6. Redlands, CA.

Faist, A. M., S. Ferrenberg, and S. K. Collinge. 2013. Banking on the past: Seed banks as a reservoir for rare and native species in restored vernal pools. AoB PLANTS 5:1–11.

Filazzola, A., A. R. Liczner, M. Westphal, and C. J. Lortie. 2019. Shrubs indirectly increase desert seedbanks through facilitation of the plant community. PLoS ONE 14:1–15.

Fuhlendorf, S. D., and D. M. Engle. 2001. Restoring heterogeneity on rangelands: Ecosystem management based on evolutionary grazing patterns. BioScience 51:625.

Funk, J. L., E. E. Cleland, K. N. Suding, and E. S. Zavaleta. 2008. Restoration through reassembly: Plant traits and invasion resistance. Trends in Ecology and Evolution 23:695–703.

Gabet, E. J., J. T. Perron, and D. L. Johnson. 2014. Biotic origin for Mima mounds supported by numerical modeling. Geomorphology 206:58–66.

Gallagher, R. S. 2014. Seeds: The ecology of regeneration in plant communities. Page (R. S. Gallagher, Ed.). CABI, Wallingford.

Gonzalez, S. L., and L. Ghermandi. 2019. Dwarf shrub facilitates seedling recruitment and plant diversity in semiarid grasslands. PLoS ONE 14:1–17.

Guo, Q., P. W. Rundel, and D. W. Goodall. 1998. Horizontal and vertical distribution of desert seed banks: Patterns, causes, and implications. Journal of Arid Environments 38:465–478.

Hopfensperger, K. N. 2007. A review of similarity between seed bank and standing vegetation across ecosystems. Oikos 116:1438–1448.

Hulvey, K. B., and P. A. Aigner. 2014. Using filter-based community assembly models to improve restoration outcomes. Journal of Applied Ecology 51:997–1005.

Hulvey, K. B., E. A. Leger, L. M. Porensky, L. M. Roche, K. E. Veblen, A. Fund, J. Shaw, and E. S. Gornish. 2017. Restoration islands: a tool for efficiently restoring dryland ecosystems?

Humphrey, L. D., and E. W. Schupp. 2001. Seed banks of Bromus tectorum-dominated communities in the Great Basin. Western North American Naturalist 61:85–92.

Jonas, J. L., M. N. Grant-Hoffman, and M. W. Paschke. 2018. Restoration of North American salt deserts: A look at the past and suggestions for the future. Ecological Restoration 36:177–194.

Jurado, E., and J. Flores. 2005. Is seed dormancy under environmental control or bound to plant traits? Journal of Vegetation Science 16:559–564.

Kachergis, E., M. E. Rocca, and M. E. Fernández-Giménez. 2014. Long-term vegetation change provides evidence for alternate states in silver sagebrush. Rangeland Ecology & Management 67:183–194.

Koniak, S., and R. L. Everett. 1982. Seed reserves in soils of successional stages of pinyon woodlands. The American Midland Naturalist 108:295–303.

LaForgia, M. L., M. J. Spasojevic, E. J. Case, A. M. Latimer, and S. P. Harrison. 2018. Seed banks of native forbs, but not exotic grasses, increase during extreme drought. Ecology 99:896–903.

Longland, W. S. 1995. Desert rodents in disturbed shrub communities and their effects on plant recruitment. Pages 209–214 *in* B. Roundy, E. McArthur, J. Haley, and D. Mann, editors. Wildland Shrub and Arid Land Restoration Symposium. General Technical Report INT-GTR-315, Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT.

Longland, W. S., and S. L. Bateman. 2002. Viewpoint: The ecological value of shrub islands on disturbed sagebrush rangelands. Journal of Range Management 55:571–575.

McAdoo, K. J., C. S. Boyd, and R. L. Sheley. 2013. Site, competition, and plant stock influence transplant success of Wyoming big sagebrush. Rangeland Ecology and Management.

Naeth, M. A., A. C. Cohen Fernández, F. P. O. Mollard, L. Yao, S. R. Wilkinson, and Z. Jiao. 2018. Enriched topographic microsites for improved native grass and forb establishment in reclamation. Rangeland Ecology & Management 71:12–18.

Oksanen, J., Guillaume Blanchet, F., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P.R., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Szoecs, E., Wagner, H. (2019). vegan: Community Ecology Package. R package version 2.5-6. https://CRAN.R-project.org/package=vegan

Paschke, M. W., L. B. Perkins, and K. E. Veblen. 2019. Restoration for multiple use. Restoration Ecology 27:701–704.

Pekas, K. M. 2010. Seed pool dynamics of a Great Basin sagebrush community in the context of restoration. Utah State University.

Pekas, K. M., and E. W. Schupp. 2013. Influence of aboveground vegetation on seed bank composition and distribution in a Great Basin Desert sagebrush community. Journal of Arid Environments 88:113–120.

R Core Team (2019). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Rawlins, J. K., B. A. Roundy, S. M. Davis, and D. Egget. 2012. Predicting germination in semi-arid wildland seedbeds. I. Thermal germination models. Environmental and Experimental Botany 76:60–67.

Rayburn, A. P., C. Schriefer, A. Zamora, and E. A. Laca. 2016. Seedbank-vegetation relationships in restored and degraded annual California grasslands: Implications for restoration. Ecological Restoration 34:277–284.

Sartor, C. E., and L. Marone. 2010. A plurality of causal mechanisms explains the persistence or transience of soil seed banks. Journal of Arid Environments 74:303–306.

Schlaepfer, D. R., W. K. Lauenroth, and J. B. Bradford. 2014. Natural regeneration processes in big sagebrush (Artemisia tridentata). Rangeland Ecology and Management 67.

Schlesinger, W. H., S. L. Tartowski, and S. M. Schmidt. 2006. Nutrient cycling within an arid ecosystem. Page 24 *in* K. Havstad, L. F. Huenneke, and W. H. Schlesinger, editors. Structure and Function of a Chihuahuan Desert Ecosystem. The Jornada Basin Long-Term Ecological Research Site.

Tilley, D., and L. St. John. 2012. Plant guide for yellow rabbitbrush (Chrysothamnus viscidiflorus). Aberdeen, Idaho, Idaho 83210.

USDA Farm Service Agency (FSA) Aerial Photography Field Office. 2017 Release. Available at https://www.usgs.gov/centers/eros/science/usgs-eros-archive-products-overview?qt-science_center_objects=0#qt-science_center_objects

USDA Forest Service. 2003. Integrated management plan for the California Park Special Interest Area. Steamboat Springs, Colorado.

Vandvik, V., K. Klanderud, E. Meineri, I. E. Måren, and J. Töpper. 2016. Seed banks are biodiversity reservoirs: Species-area relationships above versus below ground. Oikos 125:218–228.

Walck, J. L., S. N. Hidayati, K. W. Dixon, K. Thompson, and P. Poschlod. 2011. Climate change and plant regeneration from seed. Global Change Biology 17:2145–2161.

Wang, Y., W. Bao, and N. Wu. 2011. Shrub island effects on a high-altitude forest cutover in the eastern Tibetan Plateau. Annals of Forest Science 68:1127–1141.

Ward, D., J. Trinogga, K. Wiegand, J. du Toit, D. Okubamichael, S. Reinsch, and J. Schleicher. 2018. Large shrubs increase soil nutrients in a semi-arid savanna. Geoderma 310:153–162.

CHAPTER IV DISTRIBUTION AND VERTICAL STRATIFICATION OF A SOIL SEED BANK IN A CHEATGRASS (*Bromus tectorum* L.) INVADED RANGELAND

Introduction

Ecological restoration of salt desert shrublands and other rangelands in the Colorado Plateau ecoregion has received increased attention by scientists and land management agencies in recent years (Chambers et al. 2017, Grant-Hoffman et al. 2018, Jonas et al. 2018, Winkler et al. 2018, Crist et al. 2019). The Colorado Plateau is a center of ecological diversity and a hotspot of plant species endemism due in part to the diverse geomorphology, climate, and other abiotic conditions (Welsh 1978, Jonas et al. 2018, Winkler et al. 2018). The Colorado Plateau is also an area of great cultural (both historic and modern) value and significance (Grant-Hoffman et al. 2012, Stier 2012, Jonas et al. 2018, Winkler et al. 2018). Legacy impacts from historical land over-utilization, current impacts from increasing land use intensity, and a changing climate present a myriad of challenges to resource managers to sustain and restore biological diversity in these ecosystems (West 1983, Blaisdell and Holmgren 1984, Stier 2012, Duniway et al. 2018, Jonas et al. 2018, Winkler et al. 2018). One such challenge includes the prevalence of numerou non-native annual plant species such as *Bromus tectorum* L. (cheatgrass – non-native annual grass), *Eremopyrum triticeum* (Gaertn.) Nevski (annual wheatgrass – non-native annual grass), *Halogeton glomeratus* (M. Bieb.) C.A. Mey. (saltlover – non-native annual forb), and *Salsola tragus* L. (prickly Russian thistle – non-native annual forb) (Humphrey and Schupp 2001, Grant-Hoffman et al. 2012, Chambers et al. 2017, Jonas et al. 2018, Winkler et al. 2018).

These non-native species can quickly become dominant at sites affected by severe disturbances such as wildfire or intense soil disturbance resultant from pipeline or oil well-pad construction (West 1983, Blaisdell and Holmgren 1984, Jonas et al. 2018, Winkler et al. 2018).

Research has shown that these disturbances and subsequent plant invasion can shift ecosystems once characterized by diverse communities of native shrubs, forbs, grasses, and biological soil crusts to non-native annual-dominated alternate states (Knapp 1996, Sperry et al. 2006, Fuhlendorf et al. 2012, Jonas et al. 2018, Winkler et al. 2018). Such ecological state transitions have resulted in reduced forage productivity, reduced plant and animal community diversity, and loss of ecological resilience to disturbances (West 1983, Blaisdell and Holmgren 1984, Stier 2012, Jonas et al. 2018). Millions of hectares in the Intermountain West and the Colorado Plateau have transitioned to these alternate states (Sleeter et al. 2012, Chambers et al. 2017). Large amounts of time and money are expended attempting to manage them to reduce wildfire risk, and to restore biological diversity and ecosystem services and function (Knapp 1996, Stier 2012, Sebastian et al. 2017a, Jonas et al. 2018, Winkler et al. 2018). However, ecological restoration of rangelands throughout the Colorado Plateau has proven to be incredibly challenging and limited success has been realized (Bernstein et al. 2014, Grant-Hoffman et al. 2018, Jonas et al. 2018, Winkler et al. 2018). Thus, recent calls have been made for a better understanding of the ecology of ecosystems in the Colorado Plateau, (Jonas et al. 2018, Winkler et al. 2018) to make more informed restoration decisions.

One ecological aspect of these annual-dominated alternate states that has been understudied is the composition and spatial distribution of soil seed banks, both across a landscape and vertically in the soil profile (Guo et al. 1998, Vilà and Gimeno 2007, Pekas and Schupp 2013, Gioria and Pyšek 2015, Jonas et al. 2018). The soil seed bank is defined as, “All living seeds in a soil profile, including those on the soil surface” (Gallagher 2014). Soil seed banks represent a memory of past and present vegetation, and regulate the regenerative potential of species reproducing by seed at a given site (Hopfensperger 2007, Bossuyt and Honnay 2008, Gallagher 2014, Gioria and Pyšek

2015). A seed bank dominated by native species may confer resilience to invasion, while a seed bank dominated by non-native invasive species may pose a critical limitation to ecological restoration activities (Von Blanckenhagen and Poschlod 2005, Bossuyt and Honnay 2008, Chambers et al. 2014c, Sebastian et al. 2017a, Jonas et al. 2018).

Few studies have analyzed the impact of species invasion on the vertical distribution of the seed bank (Keddy et al. 1989, Guo et al. 1998, Gioria and Pyšek 2015, Gaggini et al. 2019). Knowledge of the absolute and relative abundance of non-native seeds across soil depths can be useful in assessing the degree of degradation of a site and the potential effort needed to restore native vegetation (Keddy et al. 1989, Pellant 1996, Smith et al. 2008, Faist et al. 2013, Rayburn et al. 2016). If an undesirable seed bank is present, with an abundance of highly competitive non-native species, then treatments could be used to deplete that seed bank. Numerous treatment methods for depleting the soil seed bank have been developed including solarization with plastic tarps for localized areas or broad-scale use of pre-emergent herbicides such as indaziflam, imazapic, chlorsfuron, sulfometuron methyl, etc. (Shinn and Thill 2004, Moyes et al. 2005, Dear et al. 2006, Sebastian et al. 2016, 2017a). Such treatments could allow follow-up seeding of more desirable plant species to be more able to compete, establish, and persist (Clark et al. 2019). Knowledge of the seed bank composition across soil depths is important, as pre-emergent herbicides tend to have greatest efficacy in the upper 2 cm of the soil (Alonso et al. 2015, Sebastian et al. 2016). If there is a significant seed bank below the region of greatest efficacy that has the potential to germinate from depth, knowing its composition would help to determine follow-up restoration treatments. If a desirable seed bank of native species is present at depth that could survive seed bank depletion treatments, then passive restoration or supplemental seeding may be suitable options (Von Blanckenhagen and Poschlod 2005, Valkó et al. 2011, Cowan and Anderson

2014, Haussmann et al. 2019). Whereas, if a non-native annual seed bank is present at depth that could survive and germinate, then additional treatments to reduce *in situ* propagule pressure and outside seed dispersal such as vegetation management with targeted grazing or herbicide application to would likely be needed (Hull 1964, Diamond et al. 2012, Kyser et al. 2013, Sebastian et al. 2017a, Perryman et al. 2018). This is especially important for *Bromus tectorum* L. (cheatgrass), as it is known to be able to successfully germinate from well below 2.5 cm, with reports of germination from a depth of more than 5 cm (Hull 1964).

Non-native annuals that are of major concern in the Colorado Plateau can produce large amounts of propagules (Pellant 1996, Lockwood et al. 2005, Jonas et al. 2018, Winkler et al. 2018). This propagule pressure and the high germination rates of these non-native annuals has been found to competitively exclude native species from recruiting into invaded plant communities (Hull 1964, Hassan and West 1986, Pellant 1996, Humphrey and Schupp 2001, Pekas and Schupp 2013, Gioria and Pyšek 2015, Sebastian et al. 2017a). Seed bank studies that have been conducted in cold deserts of the U.S., primarily the Great Basin, have found that non-native annuals, especially cheatgrass, can make up a majority of the soil seed bank (Knapp 1996, Pellant 1996, Humphrey and Schupp 2001, Diamond et al. 2012). However, the prevalence of cheatgrass seed can vary over time and be influenced by soil type and aboveground plant community composition (Hassan and West 1986, Humphrey and Schupp 2001, Smith et al. 2008, Pekas and Schupp 2013, Haight et al. 2019).

Soil seed banks in less-disturbed ecosystems are known to be highly heterogeneous in terms of species composition and abundance (Hopfensperger 2007, Bossuyt and Honnay 2008, Vandvik et al. 2016). But in invaded ecosystems, evidence suggests that non-native species' seed banks are more homogenously distributed and often represent >50% of the seed bank (Knapp 1996, Humphrey and Schupp 2001, Boudell et al. 2002, Gioria and Pyšek 2015, Rayburn et al. 2016).

Bromus tectorum L. (cheatgrass) can form a large seed bank, with reported seed abundances ranging from <5,000 to >12,000 seeds m^{-2} (Pellant 1996, Humphrey and Schupp 2001, Diamond et al. 2012). Cheatgrass primarily forms a transient seed bank (Smith et al. 2008, Sebastian et al. 2017a), but has been found to produce seeds that can persist for upwards of five years (Young et al. 1969, Pellant 1996, Diamond et al. 2012). It has been suggested that multi-year seed carryover is not the primary driving force of cheatgrass persistence on a given site, rather it is the annual seed bank replenishment that contributes to the persistence of these non-native annual alternate states (Hull 1964, Pellant 1996, Humphrey and Schupp 2001). However, multi-year seed persistence has been found to be an important characteristic of cheatgrass that has allowed it to recover from "die-offs" (Baughman et al. 2017) or stand failures that fail to replenish the seed bank. Long-term (4-5 year) control has been found to effectively deplete the seed bank of cheatgrass and increase native perennial species cover in Colorado (Sebastian et al. 2017b, 2017a).

Understanding and quantifying the composition and distribution of the seed bank can help resource managers target management actions. This is especially true as new tools become available for such long-term non-native annual control, e.g. the pre-emergent herbicide indaziflam (Sebastian et al. 2016, 2017b, Clark et al. 2019). However, it is important to understand what native species seeds might also be present in the seed bank, so that such management actions do not have adverse impacts on plant community assembly (Sebastian et al. 2017a). Such pre-emergent herbicides like indaziflam are moderately mobile in soil, but the herbicide has been found to have its highest efficacy in the upper 2 cm of the soil (Alonso et al. 2015, Sebastian et al. 2017b). Knowing the seed bank composition lower in the soil profile can provide information to give natural resource managers additional options. If native species seeds are present lower in the soil profile and relatively few non-native species seeds, then passive restoration using the seed bank (if native seeds are able to germinate from depth) or soil disturbance to bring up the seed bank could be viable options (Von Blanckenhagen and Poschlod 2005). However, if non-native species seed have moved downward in the soil profile, then a more nuanced treatment approach may be necessary.

In this study, I aimed to characterize the soil seed bank composition across a rangeland site that experienced a wildfire of (1696 ha) in 1994 and was subsequently invaded by *Bromus tectorum* L. (cheatgrass). At the time of sampling in 2018 and 2019, the site had >75% cheatgrass cover, with native grass and forb species making up minor components of the plant community. This study aims to, 1) describe the composition and spatial distribution of the surface (0-5 cm) soil seed bank; and 2) describe the vertical distribution of the soil seed bank between 0-2 cm and 2-5 cm in the soil profile. The site studied and associated disturbance followed by cheatgrass invasion is representative of large areas of publicly managed land in the Colorado Plateau (West 1983, Grant-Hoffman et al. 2018, Jonas et al. 2018, Winkler et al. 2018).

In this study, I posed the question, what is the spatial distribution of native and non-native species seeds in soil, both across the landscape (horizontally) and vertically in the soil profile? First, I hypothesized that the surface (0-5 cm) soil seed bank of non-native species is more homogenously distributed horizontally across the landscape than that of native species (Hypothesis 4.1). Second, I hypothesized that the soil seed bank of non-native species is concentrated in the 0-2 cm portion of the soil profile (Hypothesis 4.2).

Materials and Methods

Site Description

The study was conducted in western Colorado, U.S.A on the western edge of Mesa County near the Colorado-Utah border (UTM 12S 670061 E 4340606 N, **Figure 4.1**). The study area is in the McInnis Canyon National Conservation Area (MCNCA), which is an area of high conservation priority due to the significant biologic, geologic, and cultural resources present (Grant-Hoffman et al. 2012). The soils of the study area are derived from slope alluvium and eolian material derived from sandstone over residuum weathered from shale, soil texture of the soil surface (0-5 cm) is a sandy loam (55% sand, 22% silt, 19% clay), additional soil chemistry data in **Appendix Table 2c** (Soil Survey Staff, 1999). Topography of the site is relatively flat, with a small ephemeral drainage running along the southwestern quarter of the study area. The 30-year normal (1981-2010) mean annual temperature (MAT) for this area is 12.1°C and mean annual precipitation (MAP) is 272 mm (PRISM Climate Group, 39.1986°N 109.0316°W). The area experienced a wildfire in July of 1994, known as the "Two Road" fire, with a 1696 ha fire perimeter (**Figure 4.1**, MTBS 2019). The overall fire area primarily burned at low to moderate severity, 58% and 19% of the 1696 ha perimeter, respectively. The area sampled during this study primarily experienced low fire severity, with the southeast portion of the sampling area mapped as moderate fire severity (MTBS, 2019).

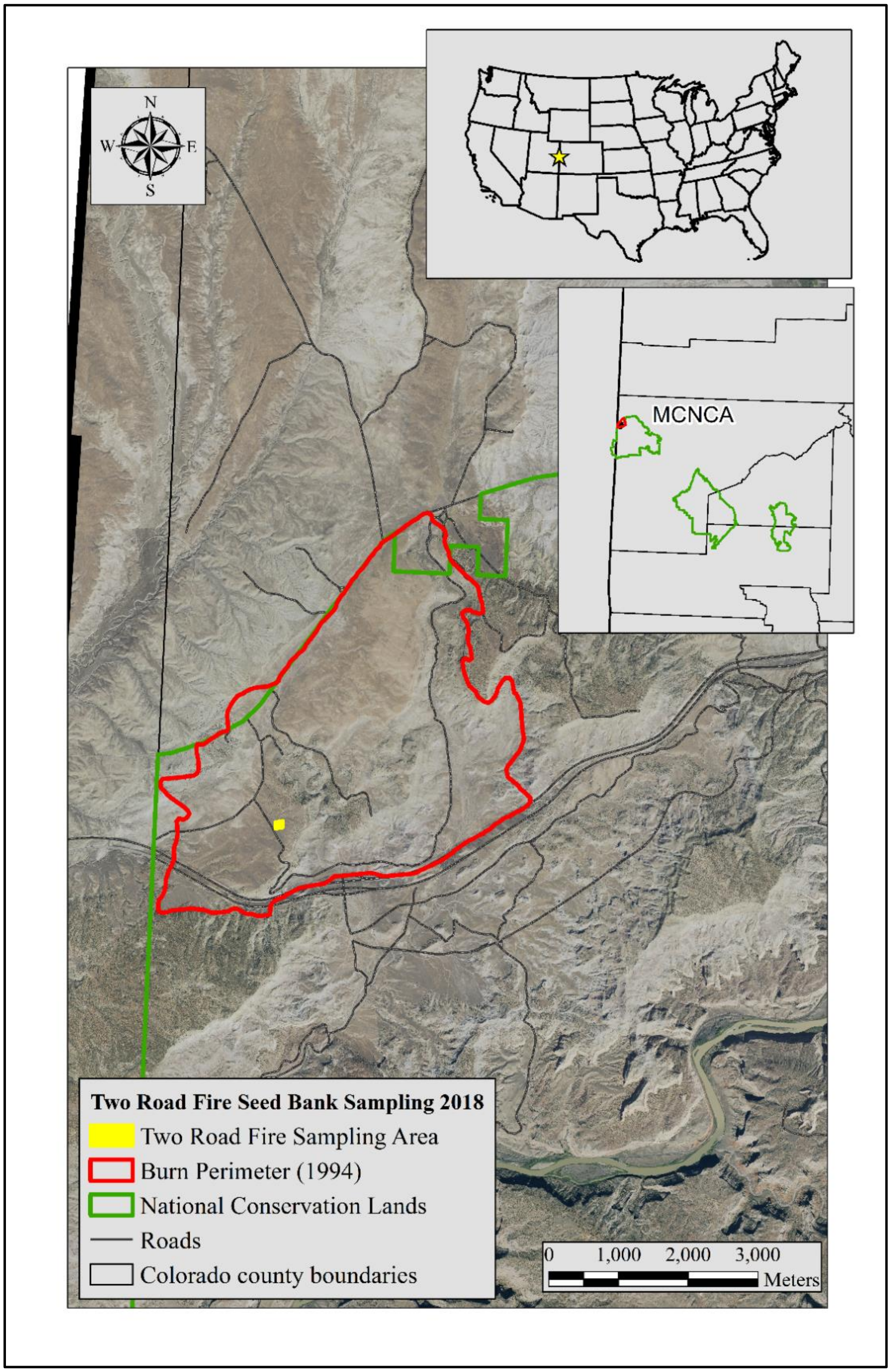


Figure 4.1: Map of the site location of the Two Road Fire sampling site, located in the McInnis Canyon National Conservation Area (MCNCA) and in the 1696 ha fire perimeter of the Two Road that burned in July 1994. Aerial imagery of Mesa County taken in 2017 by the National Agricultural Imagery Program (USDA-FSA, 2017), fire perimeter from the Monitoring Trends in Burn Severity group (MTBS, 2019).

In 2019, aboveground vegetation was measured as frequency (species presence-absence) of occurrence using 0.67- x 0.28-m (0.1876 m^2) quadrats. Three 100-m transects were laid out across the Two Road Fire site, and 10 quadrats measured per transect. The non-native species *Bromus tectorum* L. (cheatgrass) and *Sisymbrium altissimum* L. (tall tumblemustard) were the dominant species in the aboveground vegetation, both observationally in 2018 when seed bank sampling took place, and in 2019 when frequency sampling took place. The surrounding area also has scattered *Juniperus* spp. L. (juniper) trees and large individuals of *Echinocereus triglochidiatus* Engelm. (kingcup cactus).

Seed Bank Sampling and Propagation

Soil seed bank sampling was conducted June 12-13, 2018 across the Two Road Fire site. To capture the landscape-scale variability of the soil seed bank, a 110- x 100-m grid pattern was set-up (**Figure 4.2**). Along a baseline running west to east, 100-m transect tapes were run directly out to the north; one every ten meters along the baseline. This resulted in 12 north-south transects. To ensure ~10-m separation between north-south transect lines throughout the grid, the distance between transect tapes was checked randomly at three locations along each transect line and corrections made if necessary. A soil seed bank sample was then collected every ten meters along each transect, totaling 120 potential sample points. Due to large rocks present at the surface, two sample points were not used (n = 118 points overall).

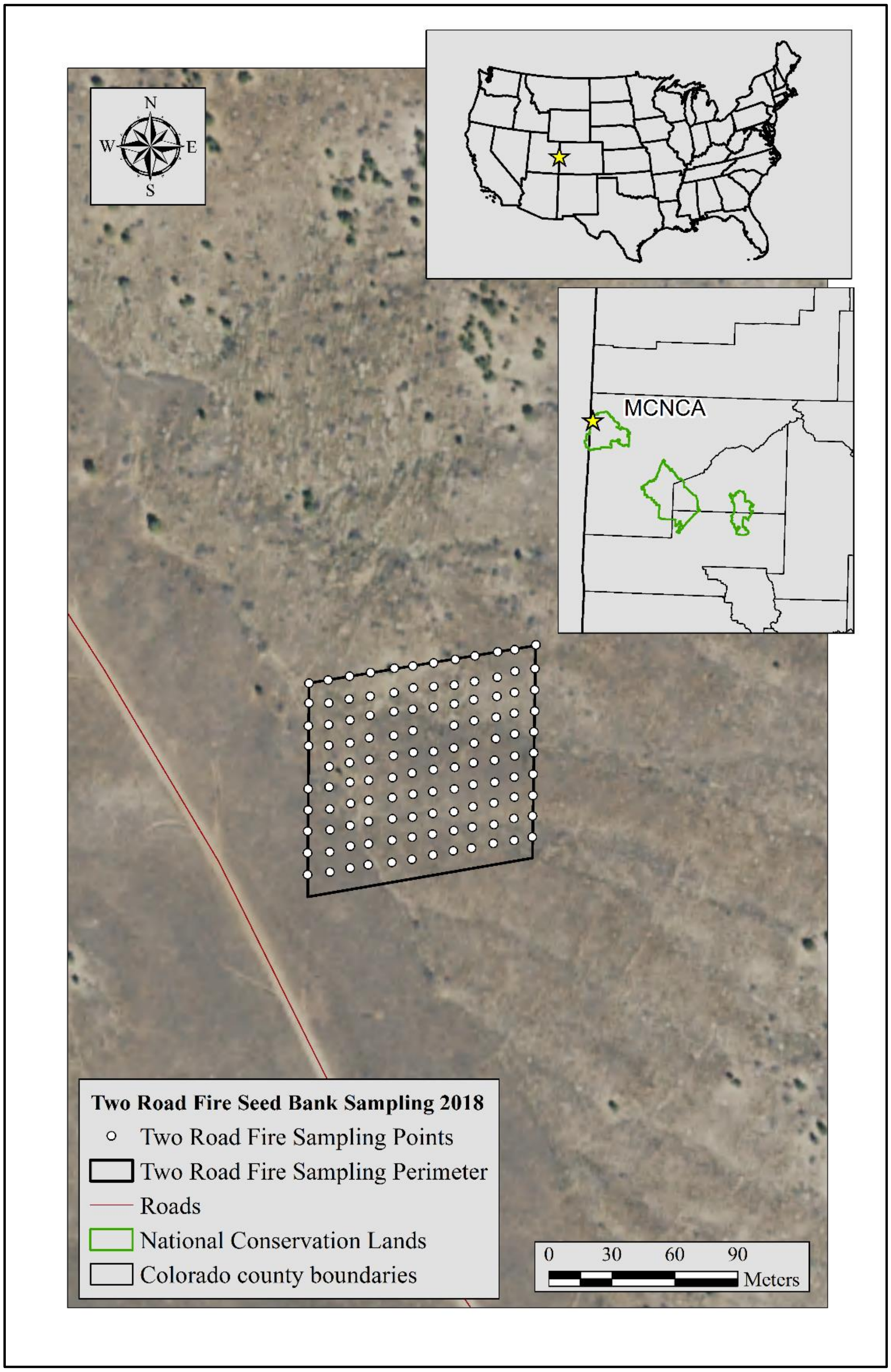


Figure 4.2: Site location of the Two Road Fire seed bank study area and layout of sampling points. The area was sampled in 2018 to analyze the distribution of the seed bank across the site and vertically in the soil profile. Aerial imagery of Mesa County taken in 2017 by the National Agricultural Imagery Program (USDA-FSA, 2017).

Soil seed bank samples were collected to a depth of 5 cm using a soil probe with a diameter of 1.5 cm. At each sample point, 10 cores were taken in a consistent, arbitrary sampling pattern. This resulted in a total sampled area of 0.2085 m^2, and a total volume of soil sampled equal to 0.0104 m^3 for the site. Each core was separated between 0-2 cm (top) and 2-5 cm (bottom) and each core section pooled for each sample point. This resulted in 236 unique soil seed bank samples for the 118 sample points. Seed bank samples were stored in a cool and dry place before being transported back to the lab where they were weighed to the nearest gram and stored at 5 °C.

To prepare the soil seed bank samples for propagation, the field soil was passed through a 5.6 mm sieve to remove rocks (coarse fragments), living vegetation, large root fragments, and to break up soil aggregates. Breaking up aggregates was especially important for the lower seed bank samples, as there was a calcic layer from 4 to 5 cm in several samples.

Soil seed bank samples were propagated at the Plant Growth Facility (PGF) on Colorado State University's campus. The PGF greenhouse bay had supplemental light 300 $\mu mol\ m^{-2}\ sec^{-1}$ to extend the photoperiod to 16 hours day^{-1}. The study was started on 9/14/2018 and terminated on 4/20/2019. Soil seed bank samples were germinated in small pots, 15 cm diameter by 10 cm depth. A base of 7 cm of ProMix BX growth media was spread in the pots and the field soils spread across the growth media to a depth of 1 cm or less. To account for any contaminant seed that might have blown into sample pots from the outside environment, control pots with 8 cm of ProMix BX growth media were randomly distributed among sample pots. The number of control pots used was equal to 10% of the number of sample units (24 control pots in total). Pots were set on top of Redi-Heat Propagation Mats (Model #: RHM2015, Phytotronics Inc) set to maintain a constant minimum soil temperature of 21°C based on estimated optimum germination temperature for most species in these ecosystems (Rawlins et al. 2012). Samples were watered regularly to ensure adequate moisture throughout the germination and growth period and randomly rotated on monthly basis to account for variability in greenhouse conditions.

Germinated seedlings were recorded, identified, and removed from the pots. After all seedlings were removed and no new germination was recorded for two weeks, samples were allowed to dry-down for two weeks. The soil was then mixed by hand to break-up aggregates and fertilized with a solution of 21-18-18 (200ppm N) to try and induce further germination (Chambers and MacMahon 1994, Walck et al. 2011). This method germinated additional seedlings from approximately 25% of the sample units. Samples were terminated after no new germination was detected for two weeks following the first dry-down and re-wetting period. Voucher specimens of each species that germinated were preserved for identity verification purposes. Nomenclature for all plant species followed the USDA NRCS PLANTS Database (2019).

Statistical Methods

To address Hypothesis 4.1, seedling counts from both the top (0-2 cm) and bottom (2-5 cm) seed bank layers of each sample point were added together to create a surface (0-5 cm) seed bank species matrix (n = 118). Seedling counts for each of the 118 sample points were then converted to seed abundance in seeds m^{-2} to facilitate further analysis. Seed abundance of all non-native species was summed to give a total non-native seed bank for each point. Seed abundance of all native species seed was summed to give a total native species seed bank for each point.

To test Hypothesis 4.1, analyses were conducted to determine if there was a difference in the mean native and non-native species seed abundance across the site, and how much variability there was in seed abundance of native and non-native species. A paired t-test approach was used to evaluate differences in native and non-native seed abundance at each point. This method determines if the difference in mean seed abundance differs from zero. The normality of differences between native and non-native seed abundance was verified. The coefficient of variation (CV) for native and non-native species seed abundance were used to describe spatial variability in seed abundance. Analyses were conducted in R v3.6.1 (R core team, 2019) and an $\alpha = 0.05$ was used for significance testing as a threshold for assigning statistical differences.

To visualize the data spatially, sample points were digitized in ArcGIS Pro v.2.4.1 (ESRI, 2019). Seed abundance data were joined by sample point and visualized by the abundance of native and non-native species seed in the overall (0-5 cm) seed bank.

To address Hypothesis 4.2, seedling counts for the top (0-2 cm) and bottom (2-5 cm) seed bank layers for each sample point were converted to seed abundance in seeds m^{-2}. Seed abundance of all non-native species was summed for each seed bank layer. Analyses were conducted to determine if there was a difference in the seed abundance of non-native species in the top (0-2 cm) versus bottom (2-5 cm) seed bank layer at each sample point and across the sampled site. A paired t-test was used to determine if the difference in seed abundance in the top versus bottom layers was different than zero. Normality of the difference between non-native seed abundance in the top and bottom seed bank layers was checked and found to be normal. Analyses were conducted in R v3.6.1 (R core team, 2019). An $\alpha = 0.05$ was used for significance testing as a threshold for assigning statistical differences. To determine if the seed bank of non-native species is concentrated to the upper portion of the soil profile, the mean of the differences must be greater than zero.

Results

Aboveground Vegetation Composition

Vegetation sampling in May of 2019 recorded 20 species (**Table 4.1**). Six non-native annual species dominated the vegetation of the site, with the most prevalent being *Bromus* *tectorum* L. (cheatgrass), *Sisymbrium altissimum* L. (tall tubmlemustard), and *Erodium cicutarium* (L.) L'Hér. ex Aiton (redstem stork's bill). The most common native species were the forbs *Cryptantha minima* Rydb. (little cryptantha – native annual forb), *Sphaeralcea coccinea* (Nutt.) Rydb. (scarlet globemallow – native perennial forb), and *Plantago patagonica* Jacq. (woolly plantain – native annual forb).

Table 4.1: Relative frequency of 20 species found in the aboveground vegetation of the Two Road Fire site in May 2019. Nomenclature follows USDA NRCS PLANTS database (2019). Cheatgrass had both a relative frequency of 100% and estimated cover >75% across the site.

Species name	**Common name**	**Native or Non-native**	**Relative Frequency**
Achnatherum hymenoides	Indian ricegrass	Native	7%
Bromus tectorum	cheatgrass	Non-native	100%
Calochorus nuttallii	sego lilly	Native	3%
Chaetopappa ericoides	rose heath	Native	23%
Chorispora tenella	blue mustard	Non-native	3%
Cryptantha minima	little cryptantha	Native	60%
Descurainia pinnata	western tansymustard	Native	40%
Descurainia sophia	herb sophia	Non-native	3%
Draba reptans	Carolina draba	Native	7%
Erodium cicutarium	redstem stork's bill	Non-native	57%
Gutierrezia sarothrae	broom snakeweed	Native	3%
Lepidium huberii	Huber's pepperweed	Native	23%
Lepidium perfoliatum	clasping pepperweed	Non-native	23%
Phlox longifolia	longleaf phlox	Native	3%
Pleuraphis jamesii	James' galleta	Native	20%
Plantago patagonica	woolly plantain	Native	47%
Sisymbrium altissimum	tall tumblemustard	Non-native	63%
Sphaeralcea coccinea	scarlet globemallow	Native	57%
Sporobolus cryptandrus	sand dropseed	Native	23%
Vulpia octoflora	sixweeks fescue	Native	13%

Soil Seed Bank Composition

Across the 118 sample points, seeds of 24 species germinated. The seed bank community included nine non-native annual species and 15 native species (**Appendix Table 5**). Total (0-5 cm) seed bank abundance averaged 5270 seeds m^{-2}. Seed from four species made up made up more than 75% of the seed bank abundance. As a percentage of the total (0-5 cm) seed bank, those included *Bromus tectorum* L. (cheatgrass - non-native annual grass, 32.5%), *Sporobolus cryptandrus* (Torr.) A. Gray (sand dropseed - native perennial grass, 32.5%), *Vulpia octoflora* (Walter) Rydb. (sixweeks fescue - native annual grass, 7.3%), and *Cryptantha minima* Rydb. (little cryptantha - native annual forb, 6.6%). Non-native annual seed (primarily *Bromus tectorum*) made up slightly more than a third of the seed bank, followed by native perennial grass seed (primarily *Sporobolus cryptandrus*) and native annual forb seed (**Table 4.2**). Although non-native annuals made up the largest proportion of seed by a single functional group, approximately two-thirds of the seed bank was composed of native species.

Table 4.2: Percent seeds by functional group at the Two Road Fire site.

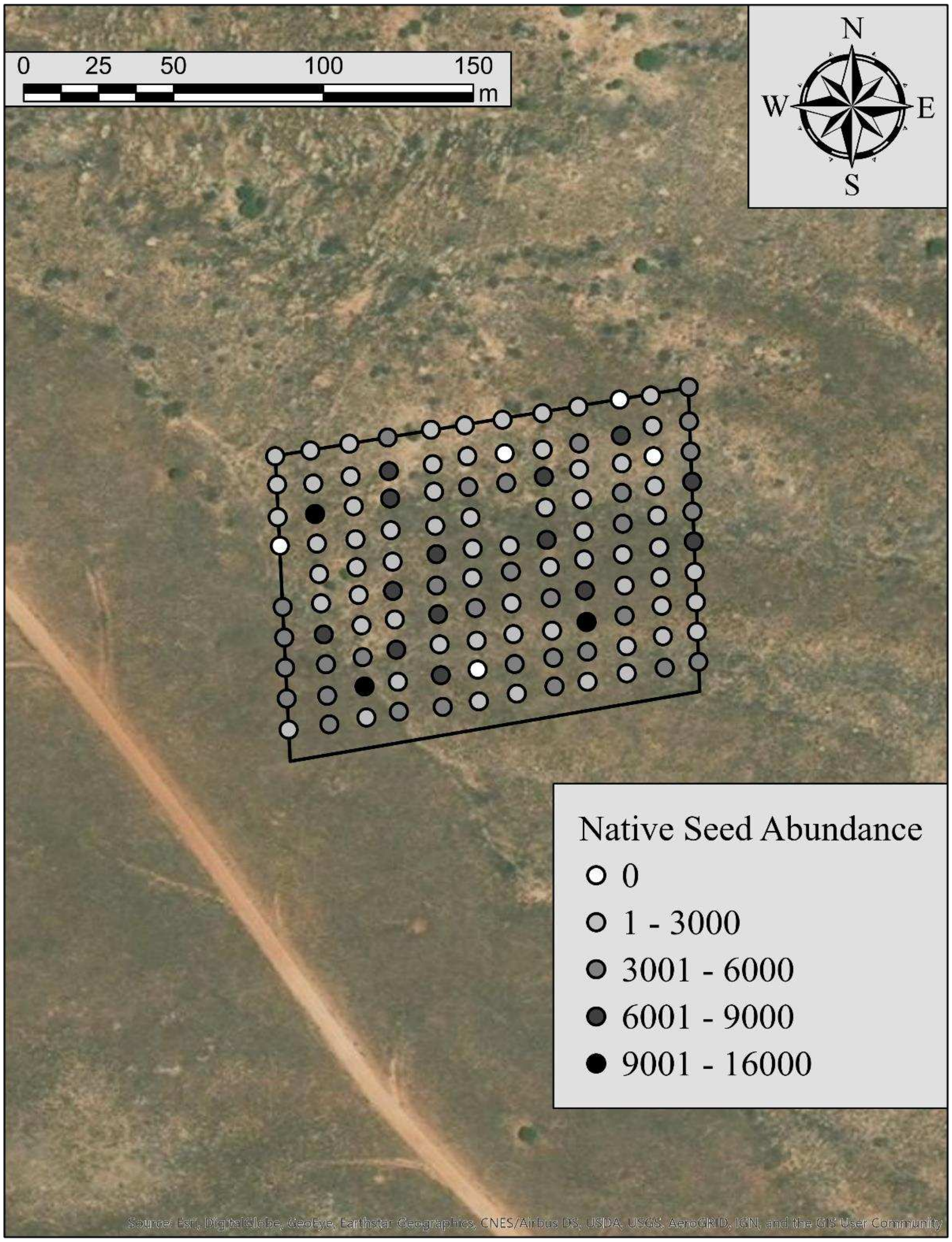
Soil Seed Bank Layer	Non-native Annual	Native Annual Grass	Native Perennial Grass	Native Annual Forb	Native Perennial Forbs & Subshrubs
0-2 cm	35.7	7.4	31.6	20.5	4.8
2-5 cm	35.6	7.0	34.7	17.2	5.5
0-5 cm	35.7	7.3	32.6	19.5	5.0

Spatial Variability of the Soil Seed Bank

Seeds of native species were more prevalent and evenly distributed than those of non-native species. Native species seeds were abundant across the landscape, with an average of 3390 seeds m^{-2} and a coefficient of variation (CV) of 75% compared to non-native species seed CV of 124% (**Table 4.3**; **Figure 4.3**). There were three native seed bank hotspots with abundances greater than 9000 seeds m^{-2}. Areas of moderate seed abundance (3000 - 9000 seeds m^{-2}) were relatively evenly dispersed throughout the site. There were five points that did not have any native species seed in the seed bank (**Figure 4.3**). Interestingly, the sample point that had the highest native and non-native seed abundance was the same, with a total seed abundance of more than 20,000 seeds m^{-2} (**Figure 4.3**, **Figure 4.4**).

Table 4.3: Mean native and non-native species seed abundance in the total (0-5 cm) seed bank across the Two Road Fire site. Means reported ± standard error. Statistical significance of paired sample mean difference in seed abundance using a paired t-test test: * $p \leq 0.05$; ** $p \leq 0.01$; † $p \leq 0.1$.

Native seeds m^{-2}	**Non-native seeds m^{-2}**
3391 ± 233	1880 ± 215
t = 5.279	
p = <0.0001**	



Source: Esri, DigitalGlobe, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AeroGRID, IGN, and the GIS User Community

Figure 4.3: Native species seed abundance (seeds m^{-2}) of the surficial soil seed bank (0-5 cm) across the landscape sampled at the Two Road Fire site in western Colorado. The area likely cross two different soil types as it is within a mapped unit of the Neiberger-Pariette complex, but it is unclear due to mapping resolution (Soil Survey Staff, 1999).

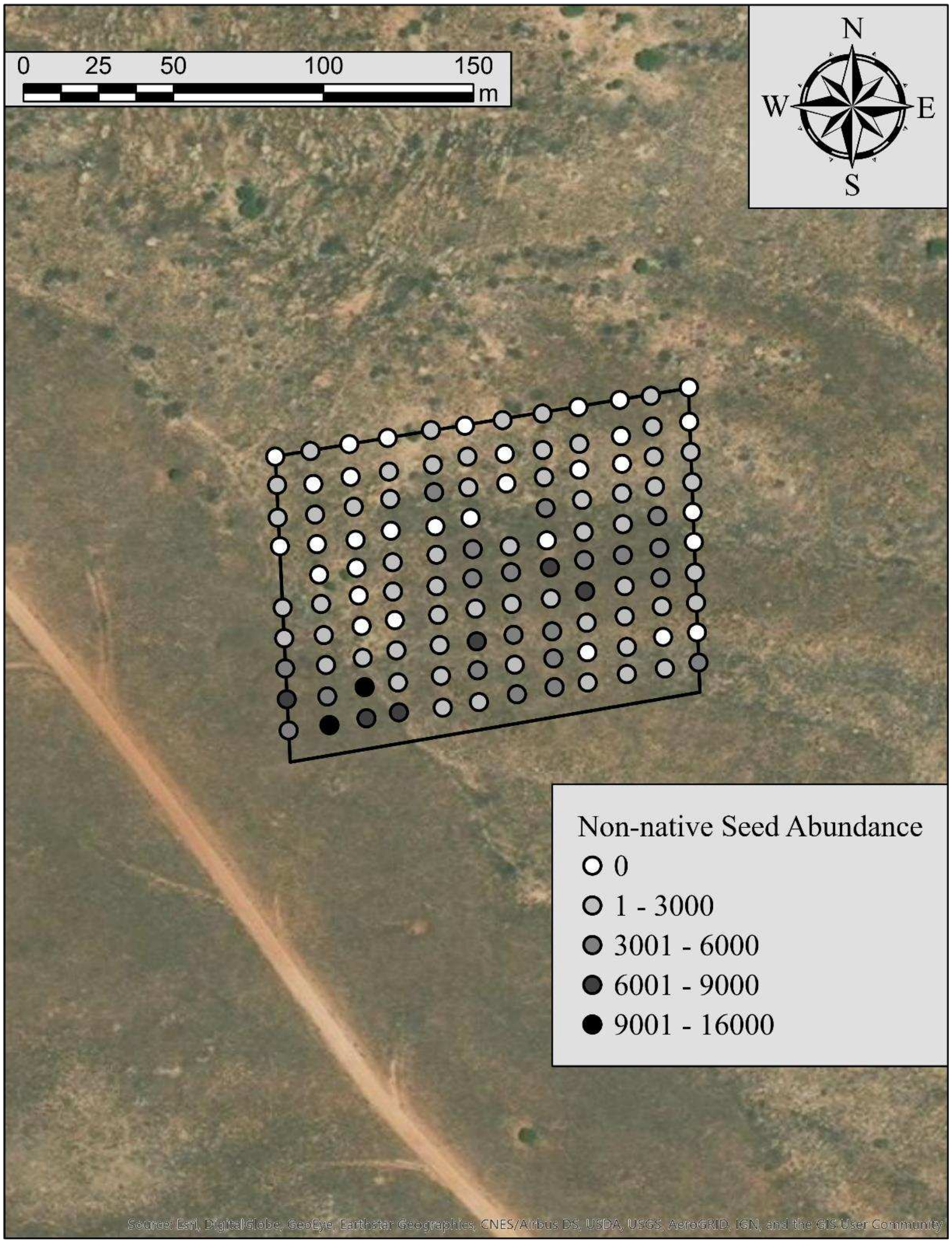


Figure 4.4: Non-native species seed abundance (seeds m^{-2}) of the surficial soil seed bank (0-5 cm) across the landscape sampled at the Two Road Fire site in western Colorado. The area likely cross two different soil types as it is within a mapped unit of the Neiberger-Pariette complex, but it is unclear due to mapping resolution (Soil Survey Staff, 1999).

Non-native species seeds, primarily *Bromus tectorum* L. (cheatgrass), were less abundant and more unevenly distributed across the landscape (**Table 4.3**; **Figure 4.4**). The mean abundance of non-native species seeds was 1880 seeds m^{-2} with a CV of 124%. *Bromus tectorum* L. (cheatgrass) made up 91% of the non-native seed bank across the site with a mean seed abundance of 1712 seeds m^{-2}. The remaining 9% (168 seeds m^{-2}) was made up by the eight other non-native annual forbs (**Appendix Table 5**). Points with the highest abundance (>9000 seed m^{-2}) of non-native species seed occurred nearest the road. Across the site, 31 sample points did not have any non-native seed in the seed bank (**Figure 4.4**). The paired t-test found that mean difference of native to non-native species seed abundance was significantly different than zero (p-value = <0.0001, **Table 4.3**) and that mean native seed abundance is greater across this cheatgrass invaded landscape.

Vertical Distribution of the Soil Seed Bank

Seeds of non-native and native species were found in both 0-2 cm (top) and 2-5 cm (bottom) layers of the soil seed bank. Both native and non-native seeds are concentrated in the top layer of the soil seed bank (**Table 4.4**). Seedlings from 22 species emerged from top (0-2 cm) layer seed bank samples, comprising 68.8% of the overall (0-5 cm) seed bank. On average the 0-2 cm seed bank contained 3625 seeds m^{-2} and the 2-5 cm seed bank contained 1645 seeds m^{-2}. There were seedlings from 22 species that emerged from the bottom layer seed bank samples, comprising 31.2% of the overall seed bank. The two layers shared 20 of the 24 species found in the total seed bank. Across the 118 sample points, there was an average of 2.6 species $point^{-1}$ in the top layer (range 0 – 7) and 1.7 species $point^{-1}$ in the bottom layer (range 0 – 4).

Table 4.4: Vertical distribution of non-native and native species seed in the top (0-2 cm) and bottom (2-5 cm) layers of the soil seed bank across the Two Road Fire site. Means reported plus or minus standard error. Statistical significance of paired sample mean difference in seed abundance using a paired t-test test: * $p \leq 0.05$; ** $p \leq 0.01$; † $p \leq 0.1$.

Seed bank layer	**Non-native seeds m^{-2}**	**Native seeds m^{-2}**
Top (0-2 cm)	1294 ± 155	2331 ± 197
Bottom (2-5 cm)	585 ± 91	1060 ± 95
	t = 5.30	t = 6.27
	p = <0.0001**	p = <0.0001**

The two species found only in the top seed bank layer were *Alyssum desertorum* Stapf (desert madwort – non-native annual forb) and *Amaranthus albus* L. (prostrate pigweed – non-native annual forb), each with only one seedling. The two species found only in the bottom seed bank layer were *Achnatherum hymenoides* (Roem. & Schult.) Barkworth (Indian ricegrass – native perennial grass) and *Chamerion angustifolium* (L.) Holub (fireweed – native perennial forb), each with only one seedling. The percentages of seeds in each functional group that make up the corresponding seed bank layers were similar, despite there being less than half of the overall seed present in the lower layer (**Table 4.2**, **Appendix Table 5**).

Non-native species seeds were concentrated in the top layer of the soil seed bank. On average there were 1294 non-native seeds m^{-2} in the 0-2 cm layer, and 585 non-native seeds m^{-2} in the 2-5 cm layer (p-value = <0.0001, **Table 4.4**). Though in much greater abundance in the top layer, *Bromus tectorum* L. (cheatgrass) and other non-native species seed are vertically mobile in the seed bank and made up a similar proportion of the lower seed bank in the top layer (**Table 4.2**). Interestingly, the non-native annual forbs *Chorispora tenella* (Pall.) DC. (blue mustard) and *Sisymbrium altissimum* L. (tall tumblemustard) were found to be more prevalent in the bottom layer than the top layer of the seed bank.

Discussion

The composition and distribution of the soil seed bank across the Two Road Fire site was more diverse than the aboveground plant community would suggest. Contrary to expectations, the soil seed bank (0-5 cm) of non-native species (primarily *Bromus tectorum* L.) was not more abundant or homogenously distributed than native species seed. Although cheatgrass and other non-native annual forb species dominated the aboveground vegetation of the site, native species seeds were more abundant and more evenly distributed. These data do not support the hypothesis that the surface (0-5 cm) soil seed bank of non-native species is more homogenous (less variable) than that of native species across the Two Road Fire site. These data support the hypothesis that the seed bank of non-native species is concentrated in the upper portion (0-2 cm) of the soil seed bank.

Spatial Variability of the Soil Seed Bank

The composition and distribution of the germinable soil seed bank across the landscape was more species rich and heterogeneous than the aboveground plant community. Such differences in the spatial distribution of the species in the seed bank versus the aboveground vegetation are supported by Vandvick et al. (2016), but this is unusual for an invaded site (Gioria and Pyšek 2015). Although cheatgrass and other non-native species were the most dominant above ground species, they did not dominant the seed bank.

Across the sampled area, non-native species seed made up 35% of the total (0-5 cm) seed bank and abundance varied greatly from point to point. This is counter to studies conducted in the Great Basin that have found cheatgrass seeds to make > 90% of the seed bank in cheatgrass-dominated sites (Humphrey and Schupp 2001, Boudell et al. 2002, Smith et al. 2008). In a review of seed banks of invaded ecosystems, Gioria and Pyšek (2015) found that a majority of invaded sites have >50% of the seed bank made up by non-native species seed, also counter to these findings. Interestingly, results similar to this study were found by Haight et al. (2019) nearby in Arches National Park, which is on the Colorado Plateau. They also found a similar mismatch between cheatgrass dominance in the aboveground plant community and the seed bank, with cheatgrass seed abundance decreasing with increasing sand content across a soil texture gradient. They found that native species not dominant in the aboveground vegetation, such as *Vulpia octoflora* (Walter) Rydb. (sixweeks fescue – native annual grass), *Sporobolus* spp. R. Br. (dropseed – native perennial grass), and *Plantago patagonica* Jacq. (woolly plantain – native annual forb), made up much larger relative abundances of the seed bank. Many of these same species were found at the Two Road Fire site and made up a relatively large part of the native seed bank.

In this study *Sporobolus cryptandrus* (Torr.) A. Gray (sand dropseed – native perennial grass) seed abundance across the site was identical to *Bromus tectorum* L. (cheatgrass – non-native annual grass); while Haight et al. (2019) found *Vulpia octoflora* (Walter) Rydb. (sixweeks fescue – native annual grass) to have similar seed abundance to cheatgrass. In this study, cheatgrass was tied for being the most abundant single species in the seed bank, alongside sand dropseed (both making up 32.5% of the seed bank, 65% of the total). However, sand dropseed was found to be only a minor component (24% relative frequency) of the aboveground vegetation. These data indicate that across a site impacted by wildfire and cheatgrass invasion, seed banks show high spatial heterogeneity and that the patchy nature of seed banks is often species-dependent (Gioria and Pyšek 2015, Vandvik et al. 2016).

If seeds of native species are more abundant and less variable, why do cheatgrass and other non-natives dominant above ground vegetation of the site? One potential explanation lies with the life history and reproductive strategy of the most prevalent native species in the seed bank, *Sporobolus cryptandrus* (Torr.) A. Gray (sand dropseed). Sand dropseed is a C4 warm-season grass that germinates and grows in the spring into the summer and does not actively compete with winter annuals like cheatgrass (Pellant 1996, Humphrey and Schupp 2001, Ferrari and Parera 2015). Cheatgrass is a C3 winter annual grass that initiates growth in the fall and spring, assimilates nutrients in the spring, and is nearing reproductive maturity when sand dropseed would normally germinate and initiate growth (Pellant 1996). Other non-native annuals found in the seed bank at the study site have temporal patterns similar to cheatgrass (Blaisdell and Holmgren 1984, Knapp 1996, Pellant 1996, Jonas et al. 2018). Additionally, sand dropseed is known to form a persistent seed bank that lies in the seed bank to wait for an optimal germination and growth window (Coffin and Lauenroth 1989, Humphrey and Schupp 2001, Sartor and Marone 2010). Thus, non-native annual species are likely suppressing the germination and growth of sand dropseed – although there were pockets of it found in the aboveground plant community. Species that can actively compete with the non-native annuals were not as large of a component of the aboveground vegetation or seed bank.

Seeds of the following native annual species comprised 26.8% of the (0-5 cm) seed bank: *Vulpia octoflora* (Walter) Rydb. (sixweeks fescue), *Chamaesyce serpyllifolia* (Pers.) Small (thymeleaf sandmat), *Cryptantha minima* Rydb. (little cryptantha), *Descurainia pinnata* (Walter) Britton (western tansymustard), *Draba reptans* (Lam.) Fernald (Carolina draba), *Draba cunefolia* Nutt. ex Torr. & A. Gray (wedgeleaf draba), and *Plantago patagonica* Jacq, (woolly plantain). Although these species share similar life history strategies as the dominant non-natives, they made up a smaller portion of the overall soil seed bank and aboveground vegetation (Gremer and

Venable 2014, Haight et al. 2019). These data indicate that native species are relatively ubiquitous but are unable to exclude non-native annuals at this site.

The patchy nature of non-native annual seeds and their co-occurrence with native species seeds poses an interesting management and restoration dilemma. If the seed bank were dominated by non-native annual species that were homogenously distributed across a landscape, a broad-scale treatment to deplete the entire seed bank and then re-seed would make sense (Sebastian et al. 2017a). However, these data suggest that a targeted approach is necessary in order to deplete the non-native seed bank and express the native seed bank.

As these data indicate, native species seeds are present in relatively high abundances across this landscape. If given the opportunity, these seeds could germinate and become part of the aboveground plant community. However, the competitive nature of cheatgrass and other non-native annuals is likely suppressing this seed bank (Knapp 1996, Sperry et al. 2006). Additionally, studies have shown that the results of re-seeding in the Colorado Plateau are highly variable and correlated with an increase of non-native species (Grant-Hoffman et al. 2018, Jonas et al. 2018).

One potential management opportunity would be to specifically target the non-native annual species in their vegetative stage so that they cannot replenish the seed bank (Sebastian et al. 2017a). Also, instances of large cheatgrass "die-offs" or stand replacing failures that have been observed in the Great Basin also present an interesting restoration opportunity (Baughman et al. 2017). However, these are unpredictable and likely due to stochastic processes. But, if the seed bank of non-native species could be depleted and the native species seed bank remain intact, then passive restoration using native seed *in situ* could be a viable option.

Vertical Distribution of the Soil Seed Bank

Both native and non-native seeds that germinated were most abundant in the upper two centimeters of the soil at the Two Road Fire site and the total number of seeds declined with depth, similar to the findings of others (Guo et al. 1998, Boudell et al. 2002, Wright and Zuur 2014, Vandvik et al. 2016). Across the Two Road Fire site, 68.8% of the seed bank (3625 seeds m^{-2}) was in the 0-2 cm layer of the seed bank and 31.2% of the seed bank (1645 seeds m^{-2}) was in the 2-5 cm layer of the seed bank. This proportion of the seed bank below 2 cm in the soil is greater than values reported in the literature, where often <20% of the seed bank of sites is located lower than 2 cm (Guo et al. 1998, Benvenuti 2007, Vandvik et al. 2016). Interestingly, the proportional make up of plant functional group seeds was almost identical between the upper and lower portions of the seed bank (**Table 4.2**).

Non-native species made up 35% of both the upper and lower portions of the seed bank, corresponding to an average of 1294 seeds m^{-2} and 585 seeds m^{-2}, respectively. The concentration of non-native species seed in the upper portion of the seed bank is similar to the findings of others, but the proportion of the lower seed bank made up by non-native seeds is higher than seen in the literature (Boudell et al. 2002, Gioria and Pyšek 2015, Vandvik et al. 2016).

The vertical movement of seed in the soil profile is influenced by several interacting factors including seed size (Guo et al. 1998, Chambers 2000), soil texture and other soil properties (Chambers and MacMahon 1994, Benvenuti 2007, Burmeier et al. 2010, Pakeman et al. 2012), precipitation (Guo et al. 1998, Gallagher 2014), and soil fauna (Chambers and MacMahon 1994). Additionally, the site is grazed by cattle, which has been found to bury grass seeds to depths greater than 2 cm through hoof trampling and may be an important biotic mechanism of vertical seed movement at this site (Winkel et al. 1991). This study did not attempt to describe the mechanisms influencing seed movement into the lower portion of the seed bank, only describe the composition and distribution.

The vertical distribution of different species seed in the soil column has effects on germination and establishment of plants and the resultant community structure (Chambers and MacMahon 1994, Hopfensperger 2007, Bossuyt and Honnay 2008). Seeds in the upper portion of the seed bank are more likely to receive necessary germination cues to initiate growth and establish, but are also more vulnerable to seed predation or seed mortality by other factors like fire (Chambers and MacMahon 1994, Guo et al. 1998, Meyer et al. 1998, Jankowska-Blaszczuk and Daws 2007, Smith et al. 2008, Wright and Zuur 2014). Seeds that make their way lower in the soil profile provide a more protected memory of a site's vegetation composition (Chambers and MacMahon 1994, Guo et al. 1998). These lower seeds can also be a persistent reservoir of seeds that could emerge following soil disturbance or other dormancy-breaking events (Pakeman et al. 2012, Gioria and Pyšek 2015, Vandvik et al. 2016). The finding that the seed bank functional make-up is similar between the upper and lower seed bank is a novel one. This indicates that both native and non-native species are affecting recent seed rain and the more protected portion of the seed bank at the Two Road Fire site.

Interestingly, few studies have analyzed the vertical distribution and movement of *Bromus tectorum* L. (cheatgrass) and other non-native species seeds in the seed bank in North American rangelands (Chambers and MacMahon 1994, Guo et al. 1998, Boudell et al. 2002). The presence of non-native species' seed, especially *Bromus tectorum* L. (cheatgrass) seeds, in the bottom layer of the seed bank is worrisome as cheatgrass is known to be able to successfully germinate from depths greater than 2 cm. These data also suggest that cheatgrass seeds may be longer-lived than thought or can move downward in the soil column relatively quickly, perhaps facilitated by cattle trampling (Winkel et al. 1991), but this is speculative. Further study is needed to assess the mechanisms and rate of vertical seed movement in rangelands, as well as the potential of these lower seeds to germinate and establish on a site, as the presence of both native and non-native seeds lower in the soil profile has important management and restoration implications.

There has been increased focus on the management and depletion of the cheatgrass seed bank to restore non-native annual dominated rangelands (Kyser et al. 2013, Sebastian et al. 2016, 2017a, 2017b). The data from this study indicate that herbicides that have highest efficacy in the top (0 – 2 cm) portion of the soil profile would be successful in depleting a majority of the non-native annual seed bank, but would not remove the *in situ* propagule pressure completely. This supports Sebastian et al.'s (2016) finding that multi-year cheatgrass seed bank control to have the greatest chance of restoration success. Further, herbicides such as indaziflam that have multi-year residence times in the soil thus provide an added layer of protection by being able to kill the seeds in top layer of the soil seed bank and successive inputs of seed into the seed bank from cheatgrass that would emerge from lower in the soil profile and reproduce (Hull 1964, Sebastian et al. 2017a). Further study and experiments on the regenerative potential of the cheatgrass seed bank across soil depths, the seed densities required to maintain cheatgrass dominance, and the impact of herbicides on seed banks across depths are warranted to help managers make better-informed decisions.

Conclusions and Management Implications

This study has helped to address the knowledge gap of the horizontal and vertical distribution of the soil seed bank in areas invaded by *Bromus tectorum* L. (cheatgrass) in the Colorado Plateau (Pekas and Schupp 2013, Gioria and Pyšek 2015, Jonas et al. 2018, Winkler et al. 2018, Haight et al. 2019). This study found that the germinable seed bank of non-native species had a patchy spatial distribution and were less abundant than native seeds across the sampled Two Road Fire site. I also found that the soil seed bank of non-native species is more concentrated to the top (0 – 2 cm) portion of the soil seed bank, but that non-native species seeds are also present in substantive abundances in the bottom (2 – 5 cm) portion of the seed bank.

This study suggests that the native seed bank of this rangeland invaded by *Bromus tectorum* L. (cheatgrass) in the Colorado Plateau is resilient. Native species seed were more abundant and evenly distributed in the soil profile across the landscape. Whereas non-native species seed are more patchily distributed, even though they may be dominant and homogenously distributed in the aboveground vegetation. Long-term targeted management of the non-native species vegetation and seed bank may help to express the native seed bank *in situ* and restore the biodiversity found in the seed bank to the aboveground plant community.

Ecological Restoration and Cheatgrass Control Options

The prevalence of native species seeds in the seed bank across the Two Road Fire site was encouraging. Though it has been approximately 25 years since the area burned and was invaded by *Bromus tectorum* L. (cheatgrass), native species continue to persist both in the aboveground vegetation and in the seed bank. Thus, the Two Road Fire site shows high potential for *in situ* passive restoration of a diverse native plant community if cheatgrass and it's seed bank could be specifically depleted (Sebastian et al. 2016, 2017a, Clark et al. 2019). The high abundance of *Sporobolus cryptandrus* (Torr.) A. Gray (sand dropseed) seed indicates that it could become dominant at the site, given a reduction in competition from cheatgrass and favorable germination and establishment conditions. But the phenological and competitive mismatch between this warm season grass and cheatgrass, poses a challenge. Native annual species were present that could potentially compete with *Bromus tectorum* (cheatgrass) include *Descurainia pinnata* (western tansymustard), *Draba reptans* (Carolina draba), *Plantago patagonica* (wooly plantain), *Cryptantha minima* (little cryptantha), and *Vulpia octoflora* (sixweeks fescue) (Herron et al. 2013).

However, seed of these native annual species comprise a smaller proportion of the above ground vegetation and seed bank than non-natives, and cannot out-compete the non-native species passively. Either a targeted reduction in cheatgrass competition or actively over-seeding seeding such annuals may help to tip the competitive balance and facilitate restoration (Herron et al. 2013, Barr et al. 2017, Sebastian et al. 2017a, Ott et al. 2019). However, this is speculative.

A challenge to the management of the non-native seed bank at this site is that the non-native seed bank was found to be patchily distributed across the landscape, contrary to expectations (Knapp 1996, Humphrey and Schupp 2001, Gioria and Pyšek 2015, Rayburn et al. 2016). Studies have found *Bromus tectorum* L. (cheatgrass) seed tend to primarily remain near its parent plant following seed rain (Chambers and MacMahon 1994, Knapp 1996, Smith et al. 2008), so targeting aboveground patches or dispersal corridors (like near to the road in this case) may be viable options (Chambers et al. 2017). This study suggests that wide-spread herbicide treatments could run the risk of applying too much product and that more targeted, finer-scale applications could be more effective.

The vertical stratification of the seed bank shows has implications for soil disturbance, herbicide use, and resulting plant community dynamics. A shallow soil disturbance or depletion of the top layer of the seed bank could result in the expression of bottom seed bank layer. But the expression of seed from lower in the soil profile is highly species-dependent and influenced by seed size (Chambers and MacMahon 1994, Benvenuti 2007). Larger-seeded species, including cheatgrass, may be able to successfully germinate and establish from depth (Hull 1964, Chambers and MacMahon 1994, Chambers 2000, Pakeman et al. 2012). Whereas small-seeded species (such as sand dropseed) would likely be unable to germinate and establish without a soil disturbance bringing them to the surface (Chambers 2000, Benvenuti 2007). It is important to note that non-native species seed like *Bromus tectorum* L. (cheatgrass), *Sisymbrium altissimum* L. (tall tumblemustard), *Chorispora tenella* (Pall.) DC. (blue mustard), *Salsola tragus* L. (prickly Russian thistle), etc. all occurred in the lower portion of the seed bank. The forbs *Chorispora tenella* (Pall.) DC. (blue mustard) and *Salsola tragus* L. (prickly Russian thistle) were in greater abundance in the bottom than the top portion. Thus, soil disturbance and expression of the lower seed bank could help to re-establish non-natives at the site from below in addition to propagule pressure from dispersal from the surrounding community.

The concentration of cheatgrass seeds in the top layer of the soil seed bank bodes well for use of pre-emergent herbicides such as indaziflam and others. Such herbicides that have the potential to provide up to three-year control of cheatgrass with a single application would also help to control non-native species' seeds subsequently dispersed from seedlings that germinate from the bottom layer of the seed bank (Hull 1964, Sebastian et al. 2016, 2017a). However, these herbicides may have the potential to negatively impact native grasses and forbs which were large components of the seed bank. Studies have found that pre-emergent herbicides do not have a significant impact on the standing perennial vegetation of a site (Clark et al. 2019), but native perennials were found infrequently across the Two Road Fire site. There is also limited research to determine the impact of such herbicides on native annual grasses and forb seed banks (Kyser et al. 2013, Sebastian et al. 2017a, Clark et al. 2019). Further research is needed to ascertain the impact these herbicides on the seed bank of native annual grasses and forbs in ecosystems like the Colorado Plateau.

REFERENCES

Alonso, D. G., R. S. Oliveira, K. E. Hall, W. C. Koskinen, J. Constantin, and S. Mislankar. 2015. Changes in sorption of indaziflam and three transformation products in soil with aging. Geoderma 239–240:250–256.

Barr, S., J. L. Jonas, and M. W. Paschke. 2017. Optimizing seed mixture diversity and seeding rates for grassland restoration. Restoration Ecology 25:396–404.

Baughman, O. W., R. Burton, M. Williams, P. J. Weisberg, T. E. Dilts, and E. A. Leger. 2017. Cheatgrass die-offs: A unique restoration opportunity in northern Nevada. Rangelands 39:165–173.

Benvenuti, S. 2007. Natural weed seed burial: effect of soil texture, rain and seed characteristics. Seed Science Research 17:211.

Bernstein, E. J., C. M. Albano, T. D. Sisk, T. E. Crews, and S. Rosenstock. 2014. Establishing cool-season grasses on a degraded arid rangeland of the Colorado Plateau. Restoration Ecology 22:57–64.

Blaisdell, J. P., and R. C. Holmgren. 1984. Managing intermountain rangelands - Salt-desert shrub ranges. General Technical Report - US Department of Agriculture, Forest Service.

Von Blanckenhagen, B., and P. Poschlod. 2005. Restoration of calcareous grasslands: the role of the soil seed bank and seed dispersal for recolonisation processes. Biotechnology, Agronomy, Society and Environment 9:143–149.

Bossuyt, B., and O. Honnay. 2008. Can the seed bank be used for ecological restoration? An overview of seed bank characteristics in European communities. Journal of Vegetation Science 19:875–884.

Boudell, J. A., S. O. Link, and J. R. Johansen. 2002. Effect of soil microtopography on seed bank distribution in the shrub-steppe. Western North American Naturalist 62:14–24.

Burmeier, S., R. L. Eckstein, A. Otte, and T. W. Donath. 2010. Desiccation cracks act as natural seed traps in flood-meadow systems. Plant and Soil 333:351–364.

Chambers, J. C. 2000. Seed movements and seedling fates in disturbed sagebrush steppe ecosystems: Implications for restoration. Ecological Applications 10:1400–1413.

Chambers, J. C., J. L. Beck, J. B. Bradford, J. Bybee, S. Campbell, J. Carlson, T. J. Christiansen, K. J. Clause, G. Collins, M. R. Crist, J. B. Dinkins, K. E. Doherty, F. Edwards, S. Espinosa, K. A. Griffin, P. Griffin, J. R. Haas, S. E. Hanser, D. W. Havlina, K. F. Henke, J. D. Hennig, L. A. Joyce, F. F. Kilkenny, S. M. Kulpa, L. L. Kurth, J. D. Maestas, M. Manning, K. E. Mayer, B. A. Mealor, C. McCarthy, M. Pellant, M. A. Perea, K. L. Prentice, D. A. Pyke, L. A. Wiechman, and A. Wuenschel. 2017. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior’s integrated rangeland fire management strategy to long-term strategic conservation actions Part 1. Science basis and applications. USDA Forest Service - General Technical Report RMRS-GTR.

Chambers, J. C., and J. A. MacMahon. 1994. A day in the life of a seed: Movements and fates of seeds and their implications for natural and managed systems. Annual Review of Ecology and Systematics 25:263–292.

Chambers, J. C., D. A. Pyke, J. D. Maestas, M. Pellant, C. S. Boyd, S. B. Campbell, S. Espinosa, D. W. Havlina, and K. E. Mayer. 2014. Using resistance and resilience concepts to reduce impacts of invasive annual grasses and altered fire regimes on the sagebrush ecosystem and greater sage-grouse: a strategic multi-scale approach. Gen. Tech. Rep. RMRS-GTR-326. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.

Clark, S. L., D. J. Sebastian, S. J. Nissen, and J. R. Sebastian. 2019. Effect of indaziflam on native species in natural areas and rangeland. Invasive Plant Science and Management 12:60–67.

Coffin, D. P., and W. K. Lauenroth. 1989. Spatial and temporal variation in the seed bank of a semiarid grassland. American Journal of Botany 76:53–58.

Cowan, O. S., and P. M. L. Anderson. 2014. The peninsula shale renosterveld of devil's peak, western cape: A study into the vegetation and seedbank with a view toward potential restoration. South African Journal of Botany 95:135–145.

Crist, M. R., J. C. Chambers, S. L. Phillips, K. L. Prentice, and L. A. Wiechman. 2019. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 2 . Management applications. General Technical Report RMRS-GTR-389. Fort Collins, CO.

Dear, B. S., G. A. Sandral, and B. C. D. Wilson. 2006. Tolerance of perennial pasture grass seedlings to pre- and post-emergent grass herbicides. Australian Journal of Experimental Agriculture 46:637–644.

Diamond, J. M., C. A. Call, and N. Devoe. 2012. Effects of targeted grazing and prescribed burning on community and seed dynamics of a downy brome (Bromus tectorum)– dominated landscape. Invasive Plant Science and Management 5:259–269.

Duniway, M. C., E. L. Geiger, T. J. Minnick, S. L. Phillips, and J. Belnap. 2018. Insights from long-term ungrazed and grazed watersheds in a salt desert Colorado Plateau ecosystem. Rangeland Ecology & Management 71:492–505.

Environmental Systems Research Institute (ESRI). (2019). ArcGIS Pro v.2.4.1. Redlands, CA.

Faist, A. M., S. Ferrenberg, and S. K. Collinge. 2013. Banking on the past: Seed banks as a reservoir for rare and native species in restored vernal pools. AoB PLANTS 5:1–11.

Ferrari, F. N., and C. A. Parera. 2015. Germination of six native perennial grasses that can be used as potential soil cover crops in drip-irrigated vineyards in semiarid environs of Argentina. Journal of Arid Environments.

Fuhlendorf, S. D., D. M. Engle, R. D. Elmore, R. F. Limb, and T. G. Bidwell. 2012. Conservation of pattern and process: Developing an alternative paradigm of rangeland management. Rangeland Ecology and Management 65:579–589.

Gaggini, L., H. P. Rusterholz, and B. Baur. 2019. The invasion of an annual exotic plant species affects the above- and belowground plant diversity in deciduous forests to a different extent. Perspectives in Plant Ecology, Evolution and Systematics 38:74–83.

Gallagher, R. S. 2014. Seeds: The ecology of regeneration in plant communities. Page (R. S. Gallagher, Ed.). CABI, Wallingford.

Gioria, M., and P. Pyšek. 2015. The legacy of plant invasions: changes in the soil seed bank of invaded plant communities. BioScience 66:40–53.

Grant-Hoffman, M. N., A. Lincoln, and J. Dollerschell. 2018. Post-fire native seed use in Western Colorado: A look at burned and unburned vegetation communities. Natural Areas Journal 38:286–297.

Grant-Hoffman, M. N., K. A. Stevens, M. H. DeChandenedes, and M. Preseton. 2012. Science plan for McInnis Canyon National Conservation Area. Science Plan for the McInnis Canyon National Conservation Area. Washington, D.C.

Gremer, J. R., and D. L. Venable. 2014. Bet hedging in desert winter annual plants: Optimal germination strategies in a variable environment. Ecology Letters 17:380–387.

Guo, Q., P. W. Rundel, and D. W. Goodall. 1998. Horizontal and vertical distribution of desert seed banks: Patterns, causes, and implications. Journal of Arid Environments 38:465–478.

Haight, J. D., S. C. Reed, and A. M. Faist. 2019. Seed bank community and soil texture relationships in a cold desert. Journal of Arid Environments 164:46–52.

Hassan, M. A., and N. E. West. 1986. Dynamics of soil seed pools in burned and unburned sagebrush semi-deserts. Ecology 67:269–272.

Haussmann, N. S., C. Delport, V. Kakembo, K. K. Mashiane, and P. C. le Roux. 2019. Restoration potential of invaded abandoned agricultural fields: What does the seed bank tell us? Restoration Ecology 27:813–820.

Herron, C. M., J. L. Jonas, P. J. Meiman, and M. W. Paschke. 2013. Using native annual plants to restore post-fire habitats in western North America. International Journal of Wildland Fire.

Hopfensperger, K. N. 2007. A review of similarity between seed bank and standing vegetation across ecosystems. Oikos 116:1438–1448.

Hull, A. C. 1964. Emergence of cheatgrass and three wheatgrasses from four seeding depths. Journal of Range Management 17:32.

Humphrey, L. D., and E. W. Schupp. 2001. Seed banks of Bromus tectorum-dominated communities in the Great Basin. Western North American Naturalist 61:85–92.

Jankowska-Blaszczuk, M., and M. I. Daws. 2007. Impact of red: Far red ratios on germination of temperate forest herbs in relation to shade tolerance, seed mass and persistence in the soil. Functional Ecology 21:1055–1062.

Jonas, J. L., M. N. Grant-Hoffman, and M. W. Paschke. 2018. Restoration of North American salt deserts: A look at the past and suggestions for the future. Ecological Restoration 36:177–194.

Keddy, P. A., I. C. Wisheu, B. Shipley, and C. Gaudet. 1989. Seed banks and vegetation management for conservation: Toward predictive community ecology. Pages 347–363 Ecology of Soil Seed Banks. Academic Press, Inc.

Knapp, P. A. 1996. Cheatgrass (Bromus tectorum L) dominance in the great basin desert. History, persistence, and influences to human activities. Global Environmental Change 6:37–52.

Kyser, G. B., R. G. Wilson, J. Zhang, and J. M. Ditomaso. 2013. Herbicide-assisted restoration of great basin sagebrush steppe infested with medusahead and downy brome. Rangeland Ecology and Management 66:588–596.

Lockwood, J. L., P. Cassey, and T. Blackburn. 2005. The role of propagule pressure in explaining species invasions. Trends in Ecology and Evolution 20:223–228.

Meyer, S. E., S. L. Carlson, and S. C. Garvin. 1998. Seed germination regulation and field seed bank carryover in shadscale (Atriplex confertifolia: Chenopodiaceae). Journal of Arid Environments 38:255–267.

Monitoring Trends in Burn Severity MTBS (2019). 1994 Colorado: Two Road. [Data set]. Fire ID: co3920910901919940712. https://www.mtbs.gov/

Moyes, A. B., M. S. Witter, and J. A. Gamon. 2005. Restoration of native perennials in a California annual grassland after prescribed spring burning and solarization. Restoration Ecology 13:659–666.

Ott, J. E., F. F. Kilkenny, D. D. Summers, and T. W. Thompson. 2019. Long-term vegetation recovery and invasive annual suppression in native and introduced postfire seeding treatments. Rangeland Ecology and Management 72:640–653.

Pakeman, R. J., J. L. Small, and L. Torvell. 2012. Edaphic factors influence the longevity of seeds in the soil. Plant Ecology 213:57–65.

Pekas, K. M., and E. W. Schupp. 2013. Influence of aboveground vegetation on seed bank composition and distribution in a Great Basin Desert sagebrush community. Journal of Arid Environments 88:113–120.

Pellant, M. 1996. Cheatgrass: The invader that won the West. Boise, Idaho.

Perryman, B. L., B. W. Schultz, J. K. McAdoo, R. L. Alverts, J. C. Cervantes, S. Foster, G. McCuin, and S. Swanson. 2018. Viewpoint: An alternative management paradigm for plant communities affected by invasive annual grass in the Intermountain West. Rangelands 40:77–82.

PRISM Climate Group, Oregon State University, http://prism.oregonstate.edu, created 4 Feb 2004. (39.1986°, -109.0316°)

R Core Team (2019). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Rawlins, J. K., B. A. Roundy, S. M. Davis, and D. Egget. 2012. Predicting germination in semi-arid wildland seedbeds. I. Thermal germination models. Environmental and Experimental Botany 76:60–67.

Rayburn, A. P., C. Schriefer, A. Zamora, and E. A. Laca. 2016. Seedbank-vegetation relationships in restored and degraded annual California grasslands: Implications for restoration. Ecological Restoration 34:277–284.

Sartor, C. E., and L. Marone. 2010. A plurality of causal mechanisms explains the persistence or transience of soil seed banks. Journal of Arid Environments 74:303–306.

Sebastian, D. J., S. J. Nissen, J. R. Sebastian, and K. G. Beck. 2017a. Seed bank depletion: The key to long-term downy brome (Bromus tectorum L.) management. Rangeland Ecology and Management 70:477–483.

Sebastian, D. J., S. J. Nissen, J. R. Sebastian, P. J. Meiman, and K. G. Beck. 2017b. Preemergence control of nine invasive weeds with aminocyclopyrachlor, aminopyralid, and indaziflam. Invasive Plant Science and Management 10:99–109.

Sebastian, D. J., J. R. Sebastian, S. J. Nissen, and K. G. Beck. 2016. A potential new herbicide for invasive annual grass control on rangeland. Rangeland Ecology and Management 69:195–198.

Shinn, S. L., and D. C. Thill. 2004. Tolerance of several perennial grasses to imazapic 1. Weed Technology 18:60–65.

Sleeter, B. M., T. S. Wilson, and W. Acevedo, editors. 2012. Status and trends of land change in the Western United States—1973 to 2000. U.S. Geological Survey Professional Paper 1794-A. U.S. Geological Survey.

Smith, D. C., S. E. Meyer, and V. J. Anderson. 2008. Factors affecting Bromus tectorum seed bank carryover in Western Utah. Rangeland Ecology & Management 61:430–436.

Soil Survey Staff, Natural Resources Conservation Service, United States Department of Agriculture. 1999. Official Soil Series Descriptions. Pariette series. Available online at https://soilseries.sc.egov.usda.gov/OSD_Docs/P/PARIETTE.html. Accessed [2/07/2020].

Sperry, L. J., J. Belnap, and R. D. Evans. 2006. Bromus tectorum invastion alters nitrogen dynamics in an undisturbed arid grassland ecosystem. Ecology 87:603–615.

Stier, M. P. 2012. Colorado Plateaus Ecoregion. Pages 219–227 *in* and W. A. Benjamin M. Sleeter, Tamara S. Wilson, editor. Status and Trends of Land Change in the Western United States—1973 to 2000. U.S. Geological Survey.

USDA Farm Service Agency (FSA) Aerial Photography Field Office. 2017 Release. Available at https://www.usgs.gov/centers/eros/science/usgs-eros-archive-products-overview?qt-science_center_objects=0#qt-science_center_objects

USDA, NRCS. 2019. The PLANTS Database (http://plants.usda.gov, 28 September 2019). National Plant Data Team, Greensboro, NC 27401-4901 USA.

Valkó, O., P. Török, B. Tóthmérész, and G. Matus. 2011. Restoration potential in seed banks of acidic fen and dry-mesophilous meadows: Can restoration be based on local seed banks? Restoration Ecology 19:9–15.

Vandvik, V., K. Klanderud, E. Meineri, I. E. Måren, and J. Töpper. 2016. Seed banks are biodiversity reservoirs: Species-area relationships above versus below ground. Oikos

125:218–228.

Vilà, M., and I. Gimeno. 2007. Does invasion by an alien plant species affect the soil seed bank? Journal of Vegetation Science 18:423–430.

Walck, J. L., S. N. Hidayati, K. W. Dixon, K. Thompson, and P. Poschlod. 2011. Climate change and plant regeneration from seed. Global Change Biology 17:2145–2161.

Welsh, S. L. 1978. Problems in plant endemism on the Colorado Plateau. Great Basin Naturalist Memoirs:191–195.

West, N. E. 1983. Great Basin-Colorado Plateau sagebrush semi-desert. Pages 331–349 *in* N. E. West, editor. Temperate Deserts and Semi-Deserts. Elsevier, Amsterdam.

Winkel, V. K., B. A. Roundy, and D. K. Blough. 1991. Effects of seedbed preparation and cattle trampling on burial of grass seeds. Journal of Range Management 44:171–175.

Winkler, D. E., D. M. Backer, J. Belnap, J. B. Bradford, B. J. Butterfield, S. M. Copeland, M. C. Duniway, A. M. Faist, S. E. Fick, S. L. Jensen, A. T. Kramer, R. Mann, R. T. Massatti, M. L. McCormick, S. M. Munson, P. Olwell, S. D. Parr, A. A. Pfennigwerth, A. M. Pilmanis, B. A. Richardson, E. Samuel, K. See, K. E. Young, and S. C. Reed. 2018. Beyond traditional ecological restoration on the Colorado Plateau. Restoration Ecology 26:1055–1060.

Wright, B. R., and A. F. Zuur. 2014. Seedbank dynamics after masting in mulga (Acacia aptaneura): Implications for post-fire regeneration. Journal of Arid Environments 107:10–17.

Young, J. A., R. A. Evans, and R. E. Eckert. 1969. Population dynamics of downy brome. Weed Science 17:20–26.

CHAPTER V PRELIMINARY RESULTS OF FIRST YEAR RESTORATION PLOT STUDY IN CALIFORNIA PARK

Introduction

Previous restoration treatments conducted in California Park have had mixed success (USDA Forest Service 2003). Land managers aim to find methods for increasing cover, diversity, and stability of a native plant communities in degraded areas of California Park in order to reduce soil loss and continued site degradation.

Heterogeneity in soils and microenvironments has been shown to increase plant community diversity and biodiversity is known to be a major influence of ecosystem resilience (Johnston 2014, Chambers et al. 2017, Naeth et al. 2018, Paschke et al. 2019). Given an uncertain future climate, it is important to establish and promote a diverse native plant community that will be resilience to extreme weather events and reduce soil erosion. Overall, I hypothesize that creating site heterogeneity in seeded restoration sites in California Park will increase resulting native plant diversity.

Restoration treatments have been shown to increase site heterogeneity and resultant diversity of native plant communities. These treatments include the creation of small slash piles (Chambers 2000, Boyd and Obradovich 2014, Castillo-Escrivà et al. 2019), small pits and mounds (Chambers 2000, Johnston 2014, Naeth et al. 2018), and excluding large herbivores (Aguilera et al. 2016, Naeth et al. 2018).

Slash is an abundant resource in forest and rangeland environments and represents a valuable resource for restoration of degraded sites (Chambers 2000, Boyd and Obradovich 2014). I predict that slash piles will provide favorable microsites. If true, this could be a practical method for increasing revegetation success in degraded areas of California Park. Thus, I hypothesize that small slash piles will result in greater diversity of seeded native plant species (Hypothesis 5.1).

Small-scale soil disturbances (such as those created by wildlife) are known to promote plant community diversity by creating heterogeneity in resource availability (Chambers and MacMahon 1994, Chambers 2000, Johnston 2014, Naeth et al. 2018). This small-scale heterogeneity creates more niches for diverse native plant species to establish, potentially resulting in a more diverse plant community (HilleRisLambers et al. 2012). Thus, I hypothesize that creating soil resource heterogeneity (small pits and mounds) in seeded restoration sites of California Park will increase resulting native plant diversity (Hypothesis 5.2).

Grazing and browsing by domestic livestock and wildlife are known to be a major impediment to revegetation projects (Yates et al. 2000, Jonas et al. 2018, Winkler et al. 2018). Given the grazing history at California Park and the abundance of large wild ungulates, it is likely that herbivores have exerted a strong filter on plant community diversity in California Park (Kachergis et al. 2012, 2014, Hulvey and Aigner 2014). However, fencing infrastructure is expensive and can attract detrimental attention and use by large ungulates and land users. Understanding the potential role of livestock grazing in native plant establishment will be critical for development future restoration plans. Thus, I hypothesize that excluding livestock from restoration treatments will increase resulting native plant diversity (Hypothesis 5.3).

Materials and Methods

Site Description and Restoration Plot Selection

California Park is located in Routt County, Colorado on the Medicine Bow-Routt National Forest and is managed by the USDA Forest Service, Hahns Peaks-Bears Ears Ranger District. California Park is an 11,000 hectare silver sagebrush (*Artemisia cana* Pursh) park tucked between the Elkhead Mountains and the Park Range 60 km northwest of Steamboat Springs (**Figure 5.1**). The Forest Service classified this area as a Special Interest Area due to its rich diversity of plant and animal life, cultural heritage, and archeological and geologic significance (USDA Forest Service 2003). Mean annual temperature and precipitation were 4.2°C and 718 mm, respectively (PRISM climate group 1981-2010 climate normals, 40.7431°N 107.1281°W).

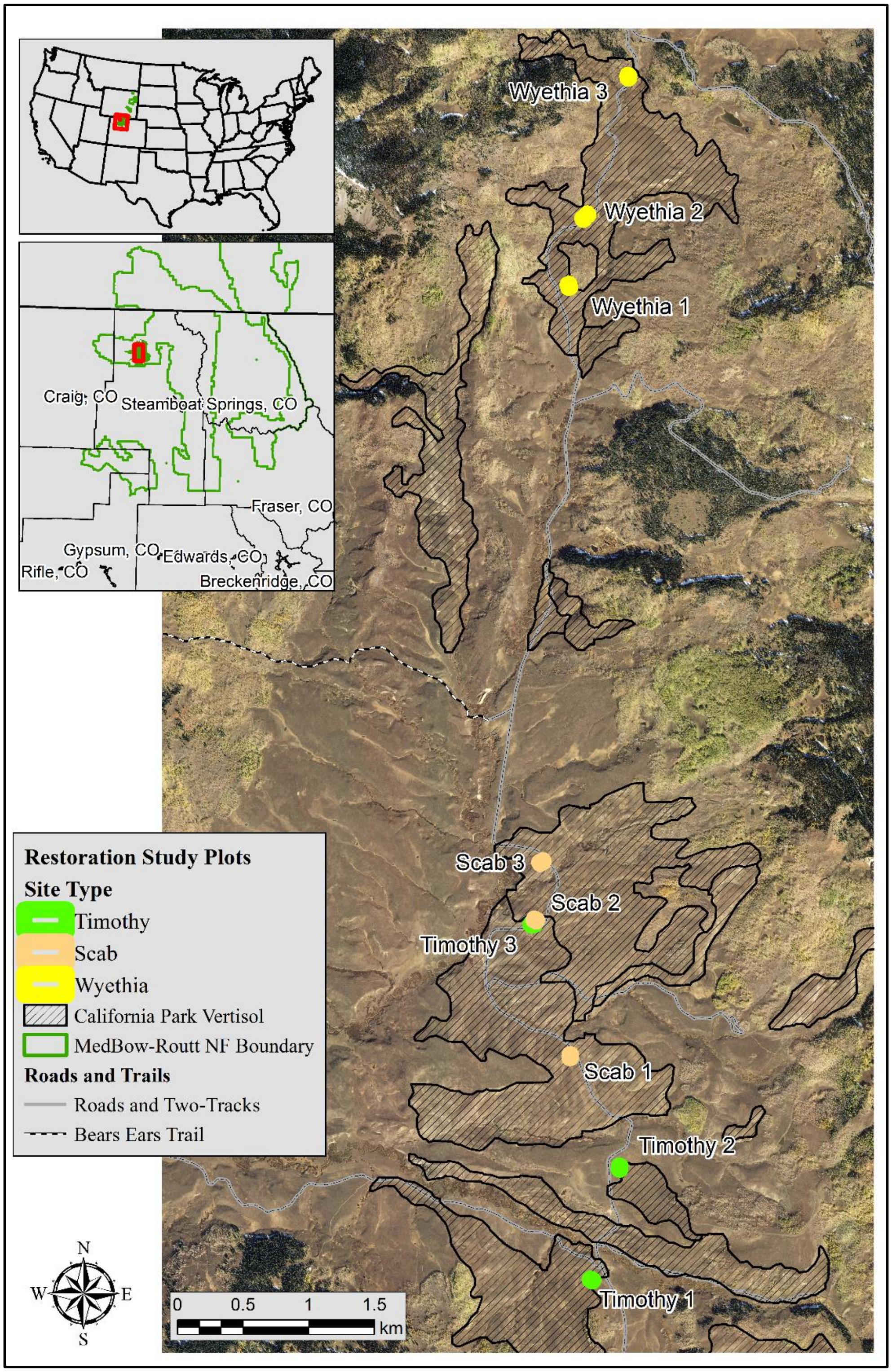


Figure 5.1: Locations of the nine study blocks (3 Timothy sites, 3 scab sites, and 3 Wyethia sites) located throughout California Park, Colorado established in 2018. Aerial imagery from fall of 2017, taken by the National Agriculture Imagery Program. (USDA-FSA, 2017)

The land use history of California Park is extensive and includes Paleo-Indian hunter-gatherers, Ute peoples, cattleman, sheepherders, homesteaders, Forest Service personnel, and recreationists. To understand the interest in ecological restoration being done in California Park, see Chapter I for additional context, as the land has a rich history of use and management.

The geomorphology, landforms, and associated soils of this SIA are highly variable throughout the relatively small area of the park and present unique challenges. The dominant geology throughout California Park are landslide deposits and residuum from sedimentary rocks including the Lance Formation and Lewis shale. The soils derived from Lewis shale (a marine shale) tend to pose the greatest management concerns as the fine-grained material is highly erodible and susceptible to landslides and earth flows (USDA Forest Service 2003). The mineralogy of soils derived from Lewis shale is dominated by smectitic clays that can exhibit shrink-swell cracking and are pedologically classified as Vertisols (Soil Survey Staff, 2006). Historical overuse and denudation of vegetation caused soil compaction of these clay soils and exacerbated shrink-swell cracking due to the lack of ground cover (USDA Forest Service 2003).

There are three primary plant community degradation states that the Forest Service is interested in restoring to increase wildlife and grouse habitat potential, decrease bare soil cover, and reduce soil erosion and improve water quality. See Chapter I "Study Sites" for a more thorough description of the ecological characteristics. In summary, one degradation state includes large areas of uplands are dominated by *Phleum pratense* L. (Timothy) and other non-native pasture grasses like *Bromus inermis* Leyss. (smooth brome), with few forbs and shrubs to support upland birds (Crawford et al. 2004, Kachergis et al. 2012, 2014, Dumroese et al. 2016). Another degradation state includes sloping uplands that are dominated by bare ground and the short-lived native annual forb *Madia glomerata* Hook. (mountain tarweed) with sparse vegetation that frequently slumps that are termed “Scabs” (USDA Forest Service 2003). The third degradation state are sloping uplands dominated by a native forb species – *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears) – that has created large “monocultures” following historical overgrazing (Kachergis et al. 2012, 2014). These high sloping areas with relatively little vegetation or vegetation that senesces rapidly, such as *Madia* and *Wyethia*, can cause sedimentation into the Elkhead Creek that flows into the Yampa River (USDA Forest Service 2003, USDA Forest Service personnel, personal communication).

Starting in 2000, Forest Service resource managers have used numerous restoration methods to try to increase cover of native grasses, forbs, and sagebrush on degraded areas in California Park. Some techniques have included plowing, sub-soil ripping, direct seeding, fertilization, herbicide application, and limited grazing exclusion (USDA Forest Service 2003). Despite these efforts, relatively little success of increasing plant species richness has been seen in the uplands over the past two decades (USDA Forest Service personnel, personal communication).

Restoration Plot Study Establishment and Treatments

In the summer and fall of 2018, I established a multi-year study in California Park to test the three aforementioned hypotheses. The study involves replicated test plots (2- x 2-m) containing four treatments: an unseeded control, seeded only, seeded plus pits and mounds, and seeded plus slash. Each of these four treatments were either fenced to attempt and exclude livestock or unfenced to allow grazing and browsing. This resulted in eight possible treatment combinations. Treatments were arranged in a split-block design (**Figure 5.2**). In coordination with USDA Forest Service personnel, nine blocks were selected throughout California Park, three on each degraded state of interest (**Figure 5.1**). These include “Timothy” sites, those dominated by *Phleum pratense* L. (Timothy) and other non-native pasture grasses; “Scab” sites that have low plant cover; and “Wyethia” sites that are dominated by the native forb *Wyethia amplexicaulis* (Nutt.) Nutt. (mule-ears). Criteria for block selection included being located in a mapped unit of the Jokodowski soil series (Fine, smectitic Typic Humicryerts) and near to the road for relatively easy access for treatment application and follow-up vegetation sampling (Soil Survey Staff, 2006).

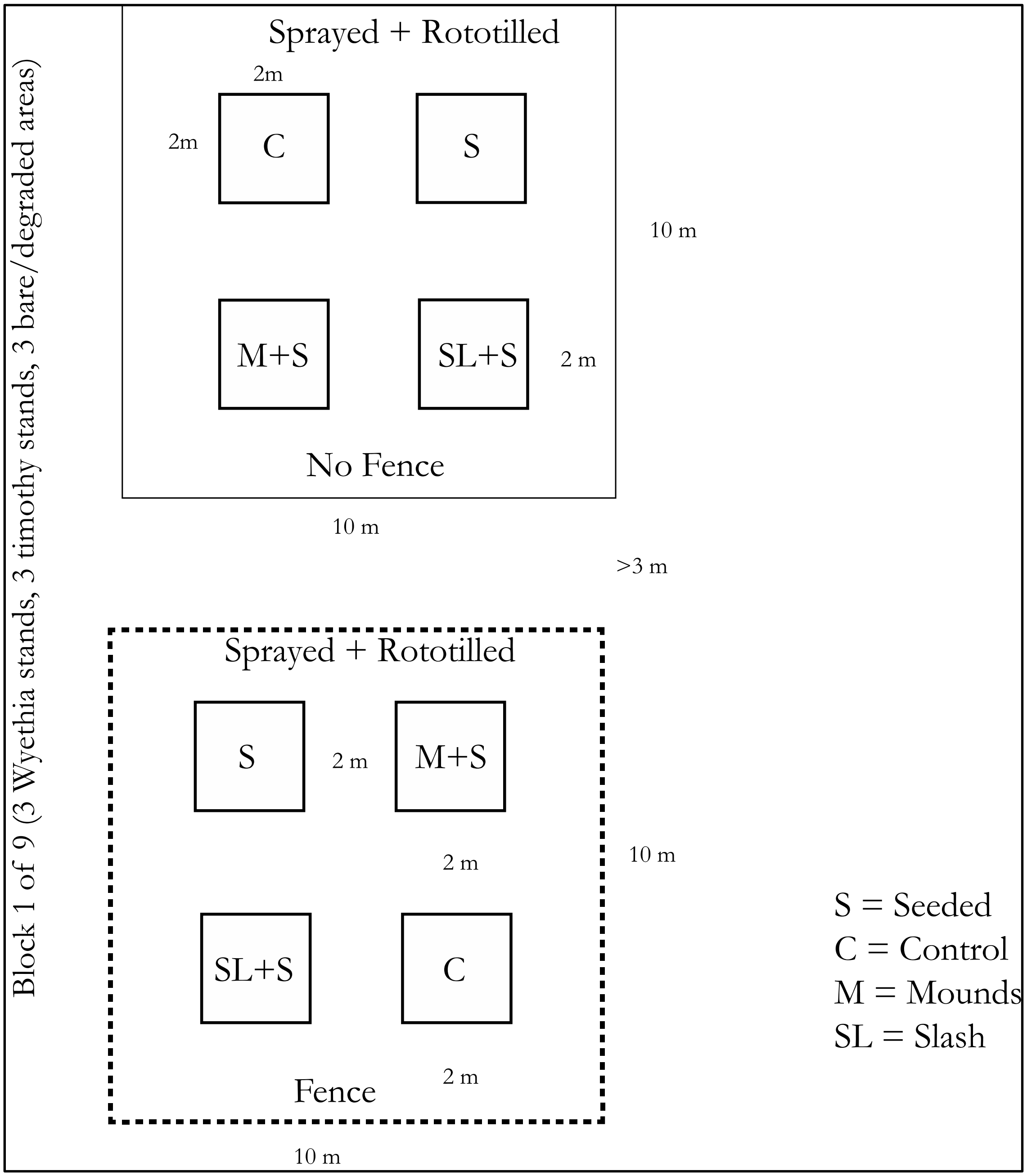


Figure 5.2: Diagram of one of the nine revegetation study blocks established in California Park in 2018. In each block, one sub block had a livestock exclusion fence installed around the 10- x 10-m area, and the other sub block was not fenced.

The locations of the nine study blocks (18 sub blocks) were marked on July 17, 2018. Each 10- x 10-m sub block was laid out using a triangulation method, marked using wooden stakes, and the GPS point of each corner recorded. In each block, the two sub blocks were located as close together as possible in order to reduce edaphic and other environmental differences between them. In order to buffer the distance between sub blocks for follow-up fencing installation and inspection by curious domestic and wild ungulates, a buffer of at least 3 m was set between each sub block. One half of each block (one sub block) was randomly assigned a fencing treatment (fenced of unfenced) by flipping a coin – fencing was installed following restoration treatment application.

On September 18, 2018, USDA Forest Service personnel applied a broad-spectrum herbicide (glyphosate) at a rate of 32 fl oz/ac (2338 ml/ha) using an All-Terrain Vehicle (ATV) mounted spray rig. This was done to attempt to kill or reduce the vigor of existing vegetation in the area of each block and reduce their competitive effect on follow-up seeding treatments. From October 15-18, 2018, each 10- x 10-m sub block was rototilled to a depth of approximately 10 cm to prepare the seedbed.

Study plots were established, and treatments applied from October 21-24, 2018. In each of the nine blocks, eight 2- x 2-m study plots were laid out using a triangulation method, marked with large metal plot nails and assigned a unique plot number. In total, this resulted in 72 plots, with three replicates in each stand, fencing, and treatment type. Soil pits and mounds were created by using a shovel and digging approximately 30- x 30-cm holes and placing the removed soil in a mound next to the pit. Nine pit and mound areas were created in each plot receiving this treatment. All plots, except controls, were then seeded (hand broadcast) with a diverse native seed mix (39 species, **Table 5.1**) at a rate of 1496 PLS m^{-2}. Plots were then raked lightly to incorporate the seed into the seedbed. Each seeded and control plot was then mulched with a light application of wood straw to improve seedling establishment in the study plots. In the 10- x 10-m area surrounding the plots in each sub block, extra seed was hand broadcast to provide an additional buffer. Following seeding and mulching, a seedbed roller was pulled across the plots and surrounding buffer area to increase seed-soil contact. One plot in each sub block then received a treatment of slash. Slash of *Artemisia tridentata* Nutt. (big sagebrush) from dozer line created during the nearby Silver Creek Fire was placed on the plot to a depth of approximately 0.5 m and to provide approximately 50% ground cover. Exclosures were constructed around the designated sub block of each of the nine blocks within a month following these treatments. Exclosures were 10-x 10-m and ~1.25m tall, constructed of wooden posts and top-rails with woven wire extending from the top rail to the soil surface to exclude large ungulates.

Table 5.1: Native species included in the seed mix applied on restoration study plots in California Park. Including 19 native forbs, 12 native grasses, and 7 native shrubs at a seeding rate of 1496 Pure Live Seed (PLS) m^{-2} (USDA NRCS PLANTS Database, 2019).

Common name	**Genus**	**Species**	**var**	**PLS m $^{-2}$**
Forbs:				
yarrow	*Achillea*	*millefolium*		45
nettelleaf hyssop	*Agastache*	*urticifolia*		45
western pearly everlasting	*Anaphalis*	*margaritacea*		38
arrowleaf balsamroot	*Balsamorhiza*	*sagittata*		45
Indian paintbrush	*Castilleja*	*spp*		89
fireweed	*Chamerion*	*angustifolium*		75
old man's whiskers	*Geum*	*triflorum*		3
fivenerve helianthella	*Helianthella*	*quinquenervis*		3
oneflower helianthella	*Helianthella*	*uniflora*		15
showy goldeneye	*Heliomeris*	*multiflora*		83
hairy false goldenaster	*Heterotheca*	*villosa*		38
Porter’s licorice-root	*Ligusticum*	*porterii*		38
flax	*Linum*	*lewisii*		8
biscuitroot	*Lomatium*	*dissectum*		45
silverleaf lupine	*Lupinus*	*argenteus*		45
silky lupine	*Lupinus*	*sericeus*		45
Rocky Mtn Penstemon	*Penstemon*	*strictus*	Bandera	45
Yampah	*Perideridia*	*gairdneri*		75
cinquefoil	*Potentillia*	*spp*		83
spreadfruit goldenbanner	*Thermopsis*	*divaricarpa*		2
Grasses:				
mountain brome	*Bromus*	*marginatus*		30
tufted hairgrass	*Deschampsia*	*caespitosa*		15
bottlebrush squirreltail	*Elymus*	*elymoides*		30
Idaho fescue	*Festuca*	*idahoensis*	Winchester	45
needle-and-thread	*Hesperostipa*	*comtata*		3
prairie Junegrass	*Koeleria*	*macrantha*		30
spike fescue	*Leucopoa*	*kingii*		30
purple oniongrass	*Melica*	*spectabilis*		30
green needlegrass	*Nasella*	*viridula*		30
western wheatgrass	*Pascopyrum*	*smithii*	Rosana	15
muttongrass	*Poa*	*fendleriana*	Ruin Canyon	15
bluebunch wheatgrass	*Pseudoroegneria*	*spicata*		30
Shrubs:				
mountain big sagebrush	*Artemisia*	*tridentata*	vaseyana	90
snowbrush ceanothus	*Ceanothus*	*velutinus*		1
winterfat	*Krascheninnikovia*	*lanata*		45
chokecherry	*Prunus*	*virginiana*		75
woods rose	*Rosa*	*woodsii*		46
thimbleberry	*Rubus*	*parviflorus*		38
Greene’s mountain ash	*Sorbus*	*scopulina*		38

Vegetation Sampling

Vegetation sampling of all 72 study plots was conducted twice during the summer of 2019, following a winter and spring with above-average precipitation (**Appendix Table 1**). These two sampling efforts were used to track the growth and survival of seeded plant species across the growing season. The first sampling effort was conducted from June 11-12, 2019 and the second sampling effort was conducted from July 22-23, 2019. Data collected in June included density counts of seeded plant species. Data collected in July included density counts and cover estimates of seeded plant species, along with cover estimates of unseeded plant species. This allows for the comparison of seeded species diversity, species richness, and plant abundance (plants m^{-2}) between June and July, and description of overall (seeded and unseeded) species diversity, species richness, and cover for July. Unseeded plant species were identified to the finest taxonomical level possible – mostly to the species level. Nomenclature for all plant species followed the USDA NRCS PLANTS Database (2019). Voucher specimens of unknown species were collected and brought back to the lab for identity verification.

Vegetation sampling of plots was conducted using a quadrat sampling method. Four 0.25- x 0.75-m (0.1875 m^2) sampling frames were placed in each 2- x 2-m plot, allowing for a 0.25 m buffer around the edge of the plot (**Figure 5.3**). To conduct density counts in June and July, the number of individual plants/seedlings of each seeded species rooted inside the frame were recorded. If seedlings of a particular seeded species in the frame were very dense, the number of individuals in a quarter of the frame were recorded. A modified Daubenmire (1959) method was used to collect aerial vegetation cover estimates in July for all species (seeded and unseeded) in each frame. The modification of the Daubenmire cover classes included an additional "Trace" cover class for the occurrence of single, small plants <1% cover. Bare ground cover (exposed soil) was also collected for each frame in July.

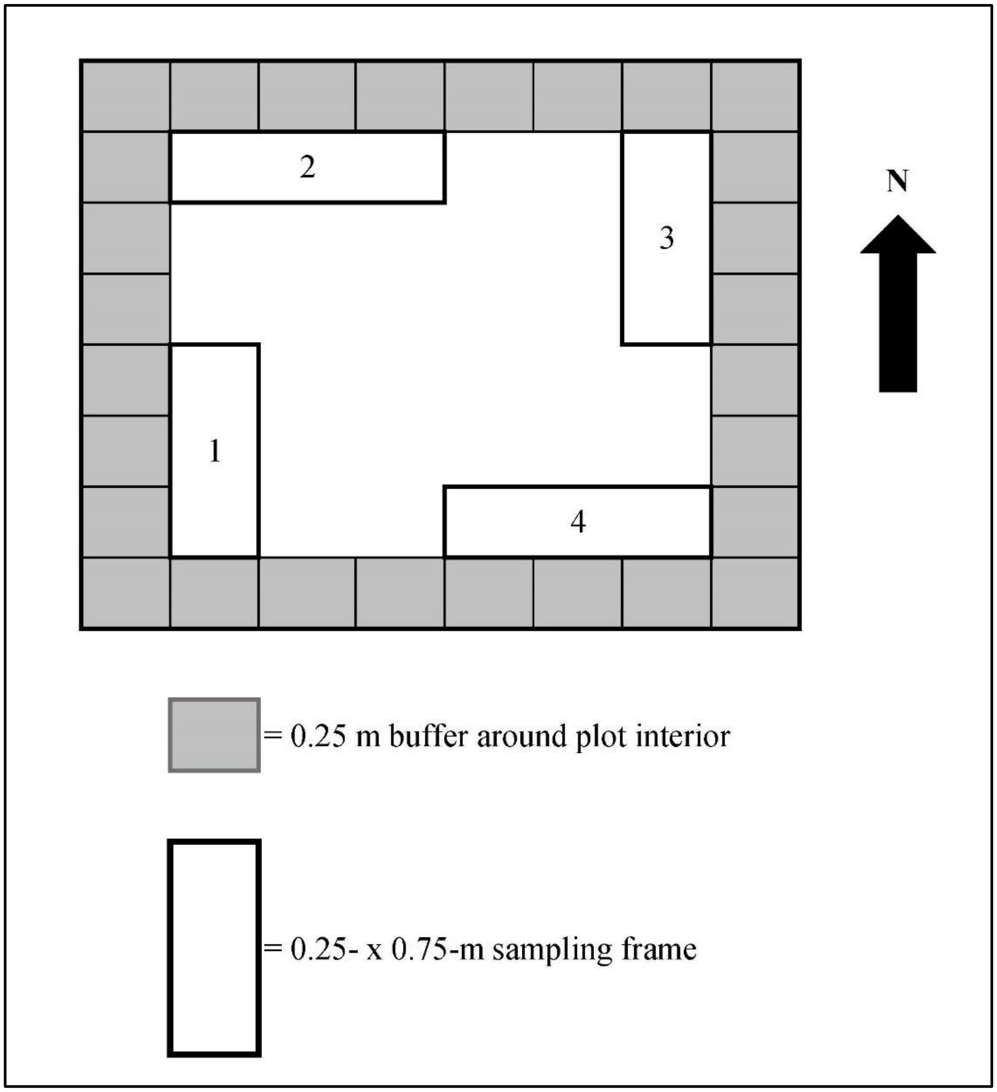


Figure 5.3: Diagram of the layout for the 4 sampling frames (0.25- x 0.75-m in each 2- x 2-m study plot, allowing for a 0.25 m buffer around the interior edge of the plot.

Statistical Methods

Analyses were conducted using the statistical program R v3.6.1 (R Core Team, 2019), along with functions in the packages Vegan (Oksanen et al. 2019), lmerTest (Kuznetsova et al. 2017), emmeans (Lenth 2019), Rmisc (Hope 2013), and ggplot2 (Wickham 2016). A linear mixed-effects modelling approach was used to analyze the effect of treatments (factors) on seeded species Shannon-Wiener diversity (H), species richness, and plant abundance (response variables). This modeling approach incorporated the effect of stand type, fencing treatment, and plot treatment type and their interactions as fixed effects, and the block and interaction of block and fence type as random effects. For each response variable for June and July, a 3-way linear mixed-effects model was fitted to the data. Normality of residuals was checked following model fit to determine if data met normality assumptions. This modeling approach helps to elucidate the effects of the different factors and their interactions on the response variables, and incorporates the variance introduced by the random effects (grouping variables) into the model. If variance of random effects in the model was greater than zero, than the degrees of freedom could be reduced from normal model degrees of freedom to account for the error of random variables.

An α = 0.05 was used for significance testing as a threshold for assigning statistical differences. For factors found to have a significant effect on the response variables, *post hoc* tests were conducted to analyze differences of least squared means within groups. Tukey's Honest Significant Differences (HSD) adjustment of p-values was used for multiple comparisons.

To facilitate statistical analyses, seeded species density counts were converted to plants m^{-2} for each frame sampled, and the mean of the four quadrats calculated to give a mean plants m^{-2} of each species in each plot. From these plant abundance values, Shannon-Winer diversity (H) and species richness were calculated for each plot using the diversity and specnumber functions in the Vegan R package were used (Oksanen et al. 2019). Seeded plant species abundance of each species was summed for each plot to give total seeded species plant abundance. Analyses presented here for June and July focused on seeded species Shannon-Wiener diversity (H), species richness, and plant abundance (plants m^{-2}).

It should be noted that some species that were seeded could have also been present in the surrounding vegetation or seed bank of the study site. Such species include *Melica spectabilis* Scribn. (purple oniongrass), *Perideridia gairdneri* (Hook. & Arn.) Mathias (Gardner's yampah), *Festuca idahoensis* Elmer (Idaho fescue), *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass), *Rosa woodsii* Lindl. (Wood's rose), and *Potentilla* sp. L. (cinuquefoil). Therefore, it is possible for control plots to have seeded species present, either through seed movement from other plots, emergence from the seed bank, or re-sprouting from perennial plants.

Results

In both June and July, diversity and species richness of seeded species in treated plots were greater than control plots. Seeded species plant abundance was found to be highly variable. No effect of the fencing treatment was detected for these response variables. No significant effect of site type (Timothy, Scab, or Wyethia) was observed in the June dataset, but site type became a significant factor in the July data. Scab sites had lower overall seeded species diversity and richness compared to Wyethia sites, with Wyethia and Timothy sites not differing. No interaction terms (i.e. Site*Fence, Treatment*Site, Treatment*Fence, or Treatment*Site*Fence) were found to be significant. Similar trends were observed between June and July, with general increases in seeded species diversity (H) and species richness values in July, indicating that mortality of seeded plant species did not occur to a noticeable degree and that additional species germinated and grew between the two sampling periods.

June Seeded Species Data

Sampling conducted in June found 21 of 39 seeded species present in study plots across California Park. The average seeded species richness of all plots was 5.4 and ranged from 0 to 14. The average Shannon-Wiener diversity (H) value of plots was 1.09 and ranged from 0 to

2.19. The average abundance of seeded species of plots was 93 plants m^{-2} and ranged from 0 – 353 plants m^{-2}. Results of the 3-way linear mixed-effects models for seeded species diversity (H), seeded species richness, and seeded species abundance (plants m^{-2}) are shown in **Table 5.2**. For these data, plot-level treatment was the only factor found to have a significant effect on seeded species diversity (H), richness, and plant abundance (plants m^{-2}) at an $\alpha = 0.05$. Thus, data between stand types and fencing treatments were aggregated to display differences between treatments performed throughout California Park.

Table 5.2: Results of mixed effects 3-way analyses for seeded species in June. Models reported include those for Shannon-Wiener diversity (H) of seeded species, species richness of seeded species, and plant abundance (plants m^{-2}) of seeded species. DF degrees of freedom. Full model: plot Treatment x sub block Fencing type x block Site type. Two random effects included, including a blocking factor and interaction factor of block and fence type (block*fence). If variance of block or stand*fence random effects were greater than zero, the resultant model's denominator degrees of freedom (Den DF) were calculated differently with Satterthwaite's method and lowered to account for the error introduced by these grouping variables. Numerator degrees of freedom (Num DF) were calculated as the number of factor categories minus 1.

		Shannon-Wiener diversity (H) of seeded species			**Species richness of seeded species**			**Plant abundance (plants m^{-2}) of seeded species**		
Source of Variance	**Num DF**	**Den DF**	**FValue**	**ProbF**	**Den DF**	**FValue**	**ProbF**	**Den DF**	**FValue**	**ProbF**
Site type (S)	2	6	4.59	0.0617	6	3.77	0.0868	5.99	0.97	0.4315
Fence (F)	1	42	1.09	0.3026	42	0.32	0.5723	6	0.41	0.5436
S*F	2	42	0.02	0.9818	42	0.003	0.9973	6	0.25	0.7839
Treatment (T)	3	42	6.18	0.0014	42	8.87	0.0001	36	2.89	0.0485
T*S	6	42	1.02	0.4248	42	0.44	0.8454	36	1.69	0.1519
T*F	3	42	0.96	0.4214	42	0.60	0.6171	36	0.47	0.7032
T*S*F	6	42	0.83	0.5509	42	1.27	0.2935	36	1.31	0.2785

Plot-level treatments were found to have a significant effect on Shannon Wiener diversity (H) of seeded species (**Table 5.2;** F = 6.18, ProbF = 0.001). Tukey's HSD found that the seeded only, seeded plus pits and mounds, and seeded plus slash treatments had significantly greater seeded species diversity (H) than control plots (p-adj < 0.05; **Figure 5.4**). Diversity was not different among the three treatments that received the diverse seed mix. Site type had a moderate effect on diversity of seeded species (**Table 5.2**; F = 4.59, ProbF = 0.062). Scab sites tended to have lower diversity values (H = 0.68 ± 0.11) across all plots than Timothy (H = 1.14 ± 0.11) and Wyethia (H = 1.19 ± 0.09) sites.

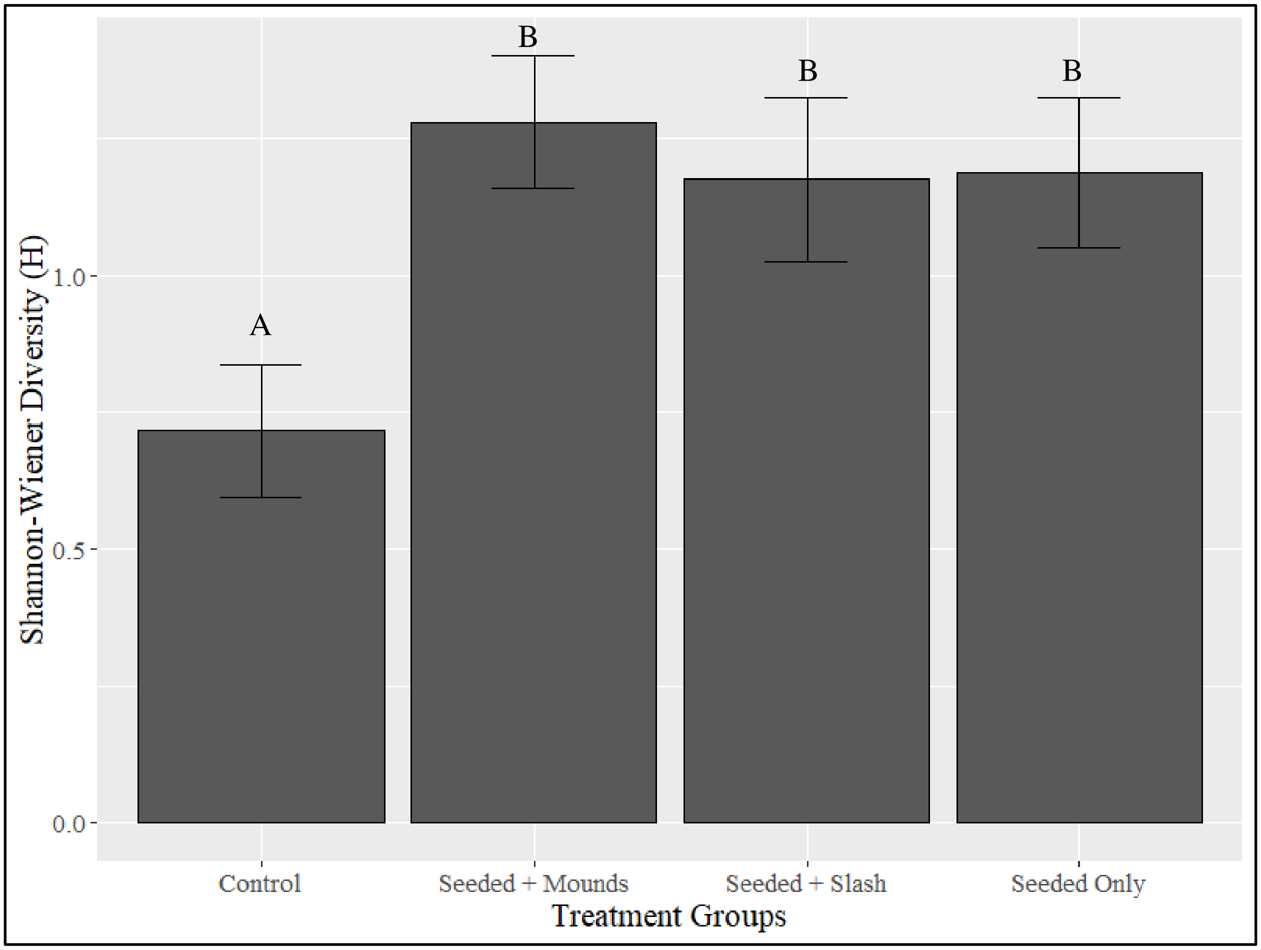


Figure 5.4: Shannon-Wiener diversity (H) of seeded species measured in June across treatments in restoration study plots. All treatments were statistically greater than the control (p-adj < 0.05). Error bars = SE. Letters indicate Least Squared Means differences calculated post-hoc with Tukey's HSD at α =0.05.

Plot-level treatments were found to have a significant effect on seeded species richness (**Table 5.2**; F = 8.87, ProbF = 0.0001). Tukey's HSD found that the seeded only, seeded plus pits and mounds, and seeded plus slash treatments had significantly greater seeded species richness than control plots (p-adj < 0.05; **Figure 5.5**). Species richness was not different between the three treatments that received the diverse seed mix. Site type had a moderate effect on species richness of seeded species (**Table 5.2**; F = 3.77, ProbF = 0.0868). Scab sites tended to have lower richness values (3.25 ± 0.42) across all plots than Timothy (6.5 ± 0.68) and Wyethia (6.5 ± 0.09) sites.

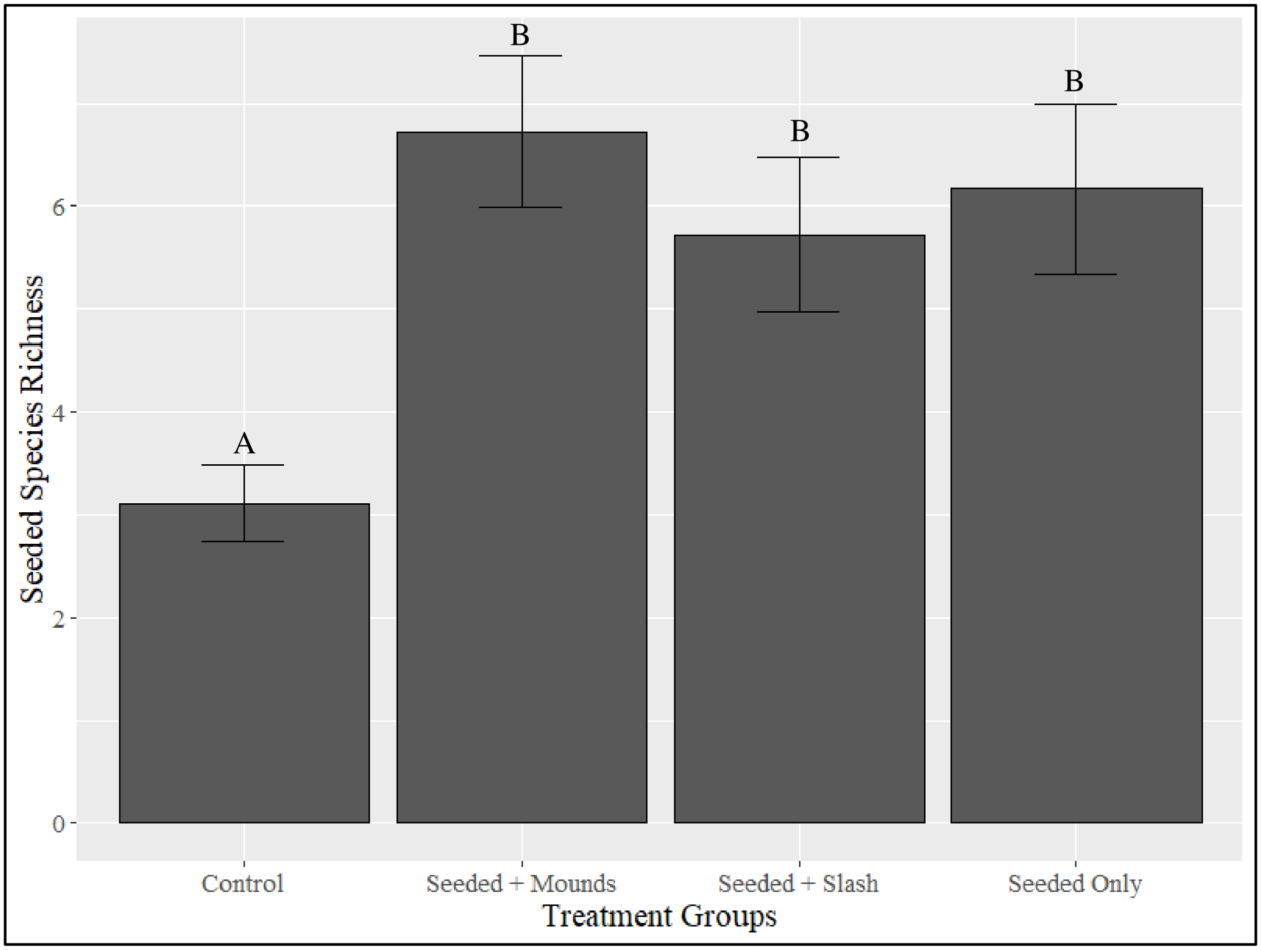


Figure 5.5: Species richness of seeded species in June across treatments in restoration study plots. All treatments were statistically greater than the control. Error bars = SE. Letters indicate Least Squared Means differences calculated post-hoc with Tukey's HSD at an α=0.05. Species that were often present in control plots included *Achillea millefolium* L. (common yarrow), *Melica spectabilis* Scribn. (purple oniongrass), and *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass).

Seeded species plant abundance was highly variable across study blocks in California Park (**Figure 5.6**). This high variability introduced additional error into the linear mixed effects model, resulting in reduced denominator degrees of freedom (**Table 5.2**). Plot-level treatments were found to have a significant effect on seeded species abundance (**Table 5.2**; F = 2.89, ProbF = 0.0485). However, Tukey's HSD did not find a significant difference between treatments at an $\alpha = 0.05$, and treatments did not differ from control plots (**Figure 5.6**). Interestingly, plots with slash piles tended to have lower seeded species plant abundance than plots with pits and mounds (p-adj = 0.0639).

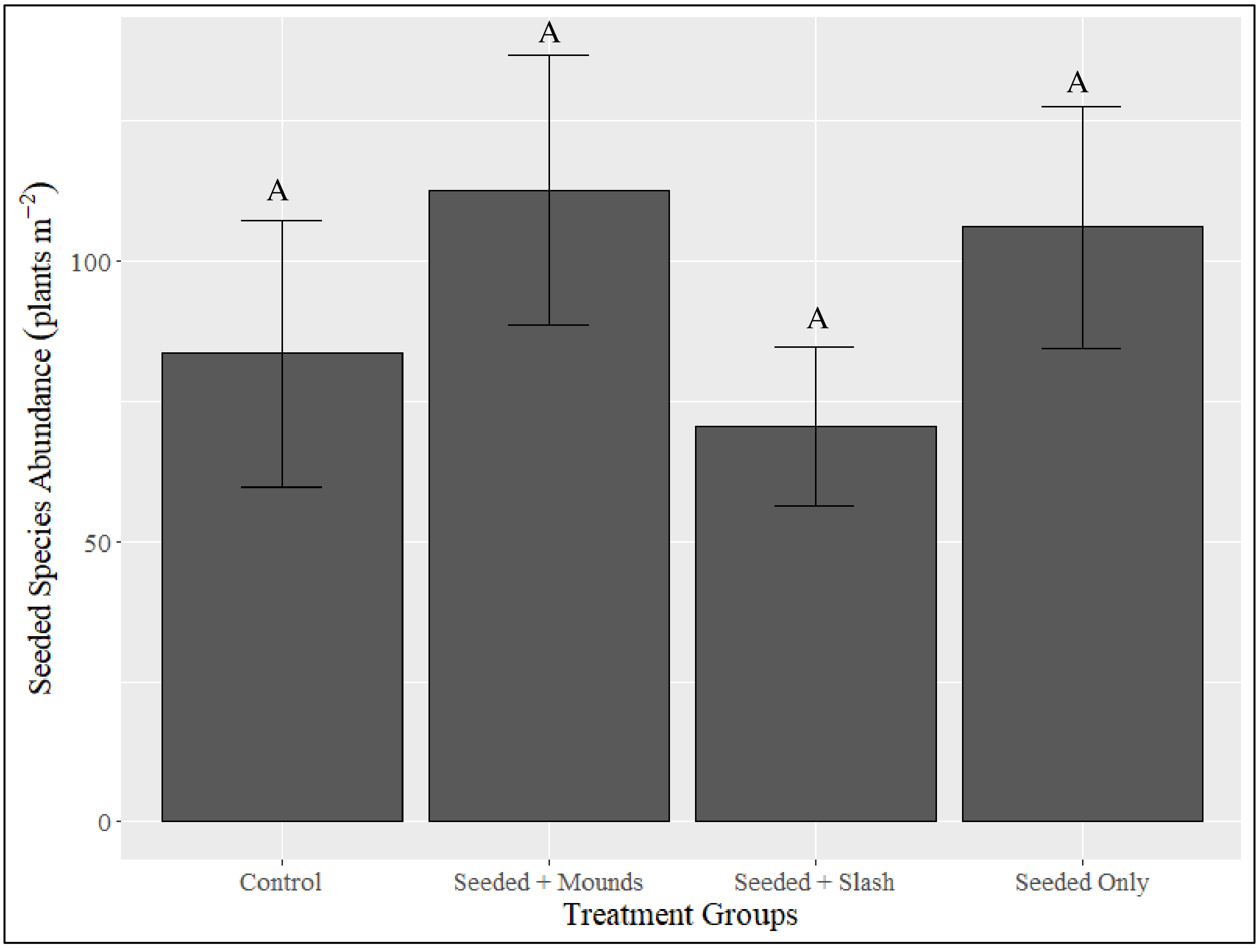


Figure 5.6: Mean plant abundance (plants m^{-2}) of seeded species in June across treatments in restoration study plots. Error bars = SE. Although the 3-way mixed linear model indicated Treatment having a significant effect (Fprob = 0.048), post-hoc tests of Least Squared Means adjusted Tukey's HSD found no significant differences between treatments at $\alpha = 0.05$. Species that were often present in control plots included *Achillea millefolium* L. (common yarrow), *Melica spectabilis* Scribn. (purple oniongrass), and *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass).

July Seeded Species Data

Sampling conducted in July found 31 of the 39 seeded species across restoration plots across California Park (10 more than sampling in June). The average seeded species richness of all plots was 7.2 and ranged from 1 to 15. The average Shannon-Wiener diversity (H) value of plots was 1.34 and ranged from 0 to 2.19. The average abundance of seeded species of plots was

85 plants m^{-2} and ranged from 8 – 272 plants m^{-2}. Results of the 3-way linear mixed-effects models for seeded species Shannon-Wiener diversity (H), seeded species richness, and seeded species abundance (plants m^{-2}) are shown in **Table 5.3**. Treatment and stand type (but not their interaction) had significant effects on seeded species diversity (H) and richness at an $\alpha = 0.05$. Treatment was the only factor that had a significant effect on seeded species plant abundance (plants m^{-2}) at an $\alpha = 0.05$. Treatment had the greatest effect on seeded species diversity, richness, and plant abundance, thus data were aggregated to show the differences between treatments across California Park.

Table 5.3: Results of mixed effects 3-way analyses for seeded species in July. Models reported include those for Shannon-Wiener diversity (H), species richness, and plant abundance (plants m^{-2}). DF degrees of freedom. Full model: plot Treatment x sub block Fencing type x block Site type. Two random effects included, including a blocking factor and an interaction factor of block and fence type (block*fence). If variance of block or stand*fence random effects were greater than zero, the resultant model's denominator degrees of freedom (Den DF) were calculated differently with Satterthwaite's method and lowered to account for the error introduced by these grouping variables. In all models for July data, high variance contributed by the block and block*fence error terms was greater than zero and thus incorporated into the model, reducing Den DF compared to the models for June data. Numerator degrees of freedom (Num DF) were calculated as the number of factor categories minus 1.

		Shannon-Wiener diversity (H) of seeded species			**Species richness of seeded species**			**Plant abundance (plants m^{-2}) of seeded species**		
Source of Variance	**Num DF**	**Den DF**	**FValue**	**ProbF**	**Den DF**	**FValue**	**ProbF**	**Den DF**	**FValue**	**ProbF**
Site type (S)	2	6	5.71	0.0409	6	14.49	0.0051	6	2.96	0.1278
Fence (F)	1	6	0.76	0.4179	6	0.49	0.5104	6	0.10	0.7588
S*F	2	6	0.53	0.6122	6	0.27	0.7751	6	0.17	0.8441
Treatment (T)	3	36	20.39	<0.0001	35.99	20.15	<0.0001	36	5.50	0.0033
T*S	6	36	0.57	0.7536	35.99	0.20	0.9758	36	0.53	0.7787
T*F	3	36	0.89	0.4545	35.99	2.16	0.1102	36	0.65	0.5902
T*S*F	6	36	0.54	0.7721	35.99	0.49	0.8102	36	0.40	0.8733

Plot-level treatments were found to have a significant effect on Shannon-Wiener diversity (H) of seeded species (**Table 5.3**; F = 20.39, ProbF = <0.0001). Tukey's HSD found that the seeded only, seeded plus pits and mounds, and seeded plus slash treatments had significantly greater seeded species diversity (H) than control plots (p-adj < 0.05; **Figure 5.7**). Diversity (H) was not different between the three treatments that received the diverse seed mix. Site type also had a significant effect on diversity of seeded species (**Table 5.3**; F = 5.71, ProbF = 0.041). Scab sites had lower diversity values (H = 1.04 ± 0.13, p-adj = 0.0346) across all plots than Wyethia (H = 1.66 ± 0.09) stands. Diversity of Timothy sites (H = 1.31 ± 0.11) was not different from either Scab or Wyethia sites.

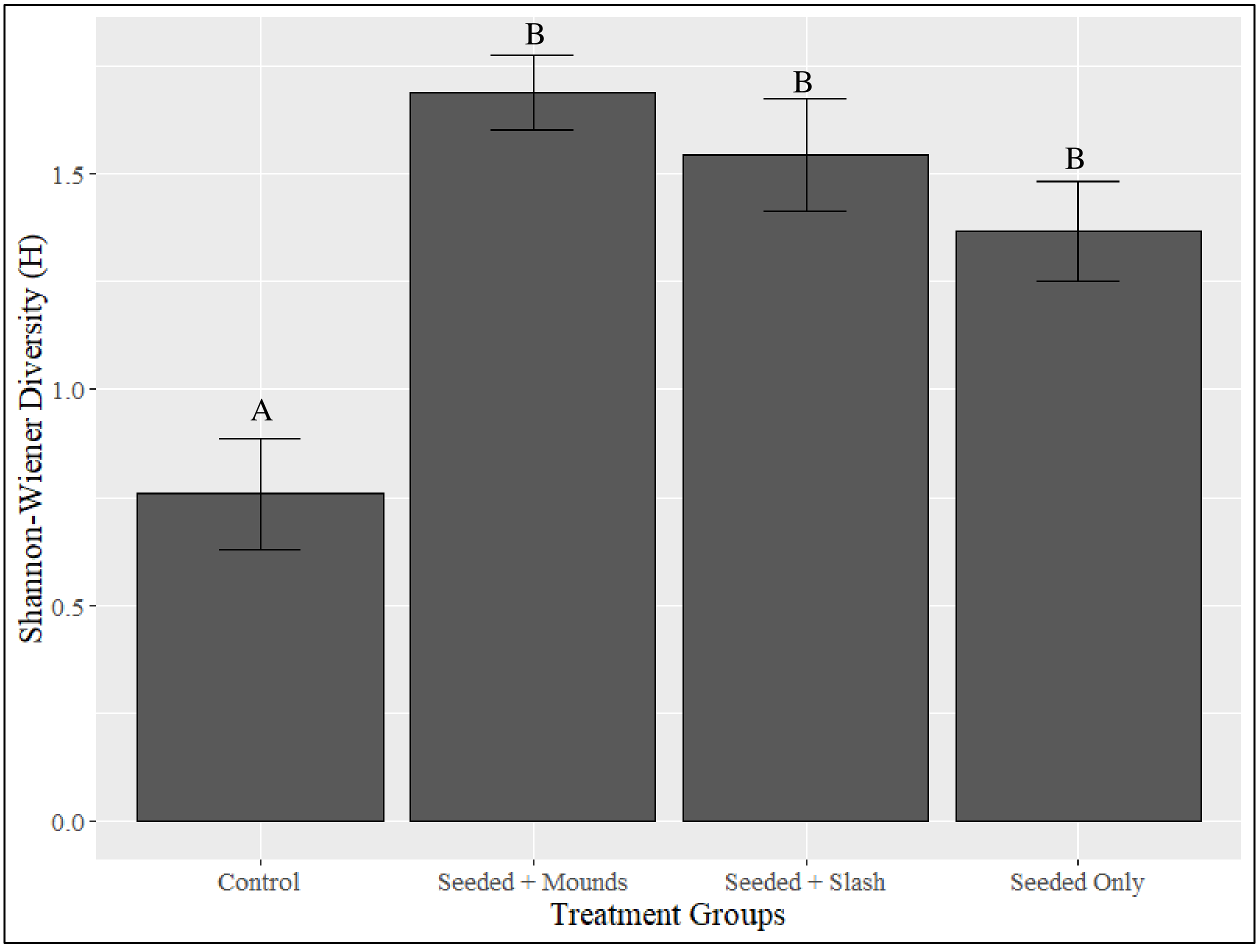


Figure 5.7: Mean Shannon-Wiener diversity (H) of seeded species measured in July across treatments in restoration study plots. All treatments were statistically greater than the control. Error bars = SE. Letters indicate Least Squared Means differences calculated post-hoc with Tukey's HSD at an α =0.05.

Plot-level treatments were found to have a significant effect on seeded species richness (**Table 5.3**; F = 20.15, ProbF = <0.0001). Tukey's HSD found that the seeded only, seeded plus pits and mounds, and seeded plus slash treatments had significantly greater seeded species richness than control plots (p-adj < 0.05; **Figure 5.8**). Species richness was not different between the three treatments that received the diverse seed mix. Site type also had a significant effect on species richness of seeded species (**Table 5.3**; F = 14.49, ProbF = 0.0051). Scab sites had lower richness values (4.8 ± 0.5, p-adj = 0.004) across all plots than Wyethia sites (9.5 ± 0.7). Richness of Timothy sites (6.5 ± 0.7) was not different from either Scab or Wyethia sites.

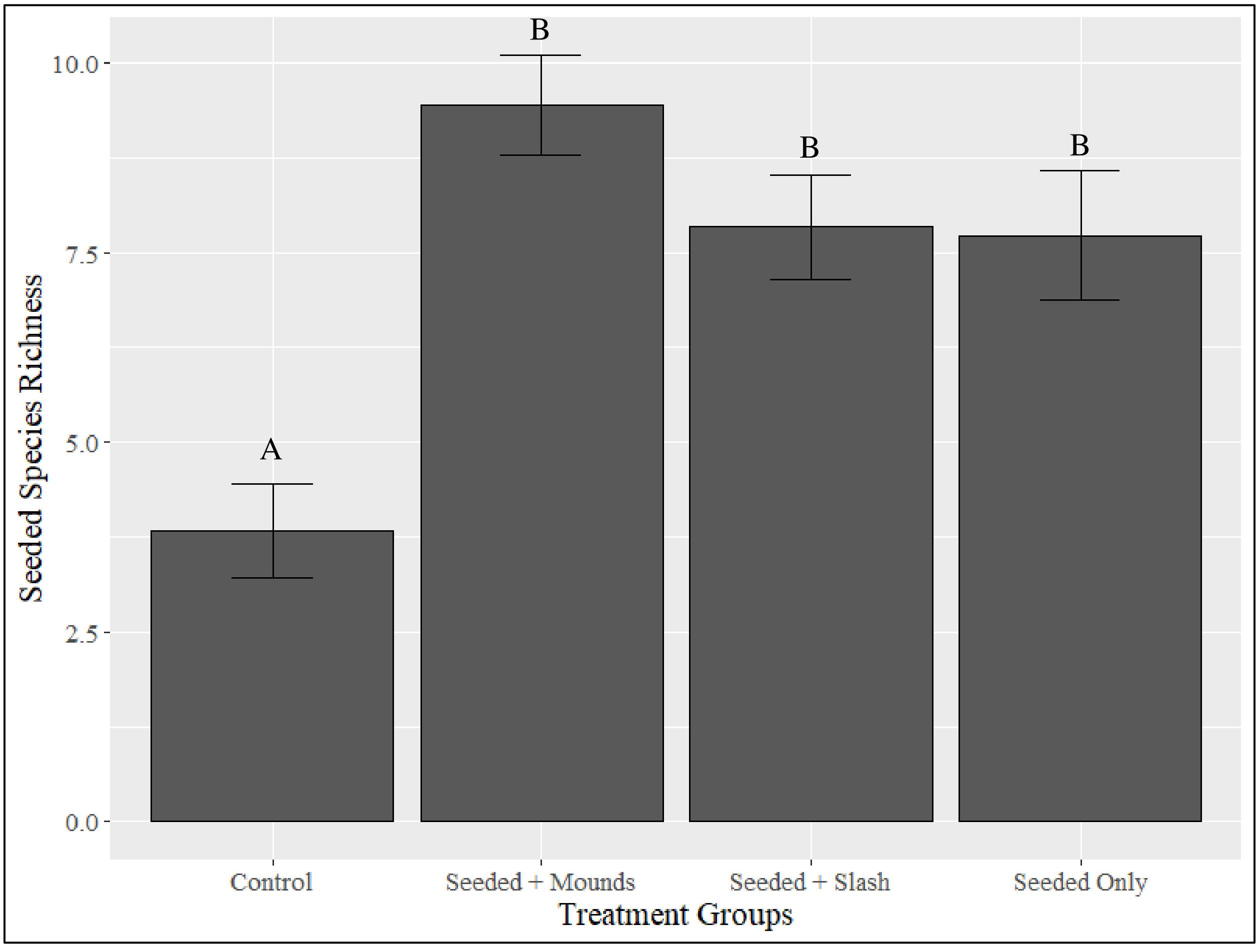


Figure 5.8: Mean species richness of seeded species measured in July across treatments in restoration study plots. All treatments were statistically greater than the control (p-adj < 0.05). Error bars = SE. Letters indicate Least Squared Means differences calculated post-hoc with Tukey's HSD at α = 0.05. Species that were often present in control plots included *Achillea millefolium* L. (common yarrow), *Melica spectabilis* Scribn. (purple oniongrass), *and Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass).

Plot-level treatments were found to have a significant effect on seeded species abundance (**Table 5.3**; F = 5.50, ProbF = 0.003). The seeded only treatment was the only treatment that had greater seeded plant abundance than the unseeded control (p-adj = 0.03; **Figure 5.9**). Interestingly, plots that received slash treatments had significantly lower seeded species plant abundance than plots with pits and mounds (p-adj = 0.041) and seeded only (p-adj = 0.013).

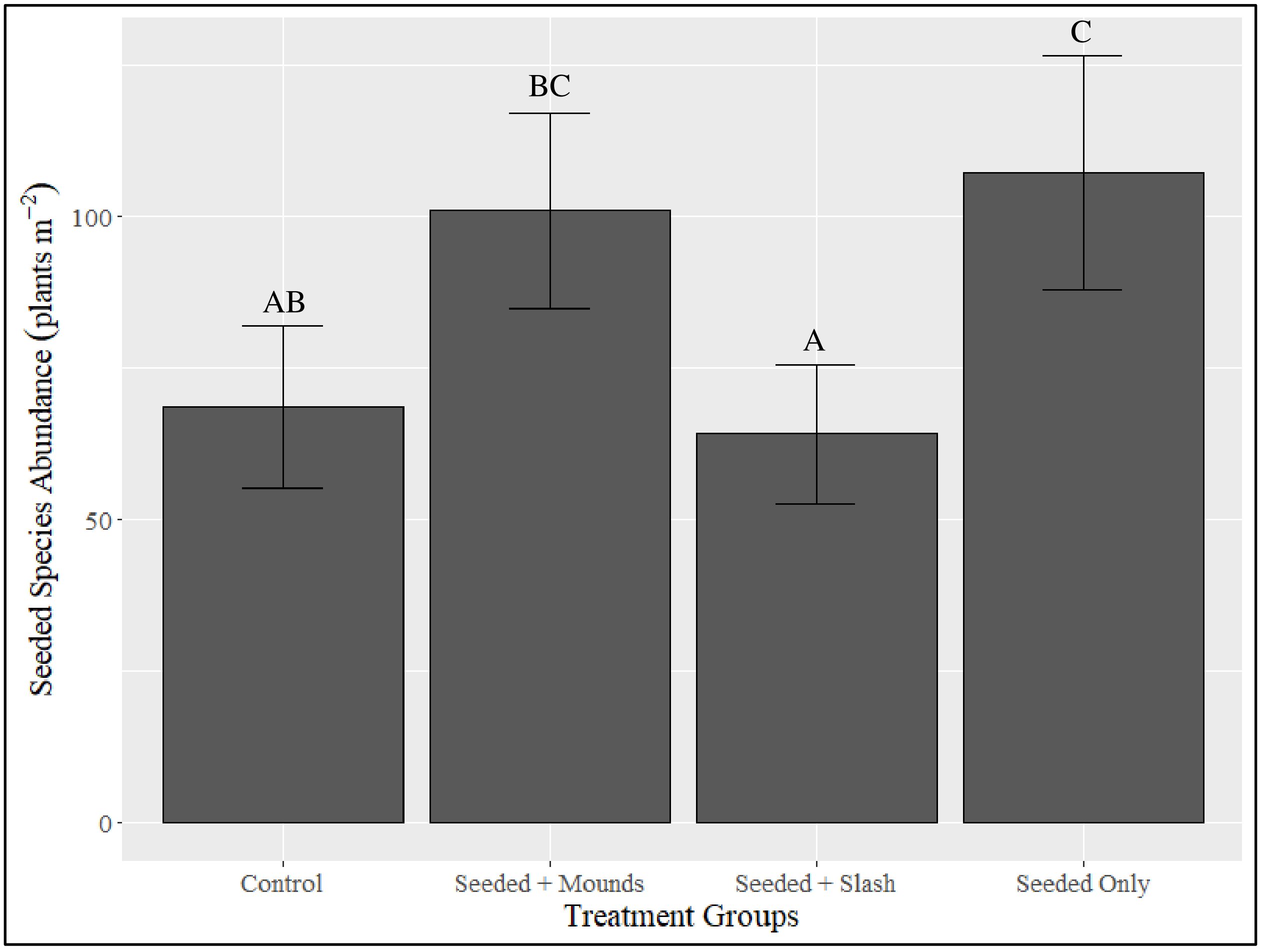


Figure 5.9: Mean plant abundance (plants m^{-2}) of seeded species in July across treatments in restoration study plots. Error bars = SE. Letters indicate Least Squared Means differences calculated post-hoc with Tukey's HSD at an α = 0.05. The seeded only treatment was greater than all other treatments, and seeded plus slash was significantly lower than the two other treatments that received the diverse seed mix, but not different from the control. Species that were often present in control plots included *Achillea millefolium* L. (common yarrow), *Melica spectabilis* Scribn. (purple oniongrass), and *Pascopyrum smithii* (Rydb.) Á. Löve (western wheatgrass).

Discussion and Conclusions

These first-year results show that restoration treatments that received the diverse seed mix had greater species diversity and richness of seeded species. Across all study blocks, species diversity and richness were greater in both June and July for plots that received the seeding only, seeding plus pits and mounds, and seeding plus slash pile treatments, compared to unseeded control plots. Fences to exclude livestock from restoration study plots did not have a significant effect on resultant seeded species diversity, richness, or plant abundance.

Site type had a moderate effect on seeded species diversity and richness in both June and July. Scab sites tended to have lower overall seeded species diversity, richness, and plant abundance, which is not surprising. The soil of plots in Scab sites were observed to be drying-out and exhibit surface soil cracking in June, while plots in Timothy and Wyethia stands were not exhibiting such cracking. Scab stands have been the most difficult for the resource managers to restore, so it is not surprising that these treatments are experiencing similar challenges (USDA Forest Service personnel, personal communication).

Plots that received treatments in addition to seeding (pits and mounds or slash) were not found to have higher seeded species diversity or richness than plots that only received the diverse seed mix after one growing season. Other studies have found that large holes, pits and mounds, or "potholes" have increased seeded species success in shrublands and grasslands and may require multiple growing seasons in order to detect a significant effect on plant community composition (Chambers 2000, Johnston 2014, Naeth et al. 2018). Thus continued monitoring will be important to determine if these treatments improve restoration outcomes of seeding in California Park.

Interestingly,plots that were seeded and had slash piles place on top tended to have lower overall abundance of seeded plants, similar to the levels of control plots. However, the seeded species diversity and richness of these slash treatments was similar to the seeded only and seeded plus pits and mounds treatments. This drop in observed plant abundance may either be an artifact of the sagebrush slash obscuring the observers' view of the plants in the plot, or that the slash was occupying more of the soil and canopy area and inhibited plant growth by physical obstruction.

Some notable observations to mention include the occurrence of elk jumping into some of the livestock exclosures and potentially impacting the restoration treatments. Forest Service personnel installed camera traps inside of a handful of livestock exclosures to attempt and assess the potential impact of these wildlife on the plots, but these camera trap data have not been analyzed. Additionally, the posts and rails of the livestock exclosures were found to be heavily used by songbirds, as there were large amounts of bird droppings on the wooden infrastructure. These birds may be dispersing plant seeds into the area of the exclosures – a potential confounding factor in the future. Also, rodents such as pocket gophers were found in several of the plots that had slash piles. These rodents could pose another confounding factor in these plots. However, it does show how habitat heterogeneity of restoration treatments can influence the plant and animal community.

These results show promising positive impacts of these restoration treatments on the plant communities of the three different degraded plant community states throughout California Park. It is apparent that the application of a highly diverse native species seed mix at a high seeding rate (1496 PLS m^{-2}) can increase the native plant species richness and diversity (Barr et al. 2017). Monitoring efforts should be continued to determine if these treatments continue to increase the native species diversity and cover in these degraded plant community types.

REFERENCES

Aguilera, L. E., C. Armas, A. P. Cea, J. R. Gutiérrez, P. L. Meserve, and D. A. Kelt. 2016. Rainfall, microhabitat, and small mammals influence the abundance and distribution of soil microorganisms in a Chilean semi-arid shrubland. Journal of Arid Environments 126:37–46.

Barr, S., J. L. Jonas, and M. W. Paschke. 2017. Optimizing seed mixture diversity and seeding rates for grassland restoration. Restoration Ecology 25:396–404.

Boyd, C. S., and M. Obradovich. 2014. Is pile seeding wyoming big sagebrush (Artemisia tridentata subsp. wyomingensis) an effective alternative to broadcast seeding? Rangeland Ecology and Management 67:292–297.

Castillo-Escrivà, A., G. M. López-Iborra, J. Cortina, and J. Tormo. 2019. The use of branch piles to assist in the restoration of degraded semiarid steppes. Restoration Ecology 27:102–108.

Chambers, J. C. 2000. Seed movements and seedling fates in disturbed sagebrush steppe ecosystems: Implications for restoration. Ecological Applications 10:1400–1413.

Chambers, J. C., J. L. Beck, J. B. Bradford, J. Bybee, S. Campbell, J. Carlson, T. J. Christiansen, K. J. Clause, G. Collins, M. R. Crist, J. B. Dinkins, K. E. Doherty, F. Edwards, S. Espinosa, K. A. Griffin, P. Griffin, J. R. Haas, S. E. Hanser, D. W. Havlina, K. F. Henke, J. D. Hennig, L. A. Joyce, F. F. Kilkenny, S. M. Kulpa, L. L. Kurth, J. D. Maestas, M. Manning, K. E. Mayer, B. A. Mealor, C. McCarthy, M. Pellant, M. A. Perea, K. L. Prentice, D. A. Pyke, L. A. Wiechman, and A. Wuenschel. 2017. Science framework for conservation and restoration of the sagebrush biome: Linking the Department of the Interior's integrated rangeland fire management strategy to long-term strategic conservation actions Part 1. Science basis and applications. USDA Forest Service - General Technical Report RMRS-GTR.

Chambers, J. C., and J. A. MacMahon. 1994. A day in the life of a seed: Movements and fates of seeds and their implications for natural and managed systems. Annual Review of Ecology and Systematics 25:263–292.

Crawford, J. A., R. A. Olson, N. E. West, J. C. Mosley, M. A. Schroeder, T. O. M. D. Whitson, R. F. Miller, M. A. Gregg, C. S. Boyd. 2004. Ecology and management of sage-grouse and sage-grouse habitat Ecology and management of sage-grouse and sage-grouse habitat. Journal of Range Management 57:2–19.

Daubenmire, R. 1959. A canopy-coverage method of vegetational analysis. Northwest Science 33:43-66.

Dumroese, R. K., T. Luna, J. R. Pinto, and T. D. Landis. 2016. Forbs: Foundation for restoration of monarch butterflies, other pollinators, and greater sage-grouse in the western United States. Natural Areas Journal 36:499–511.

H. Wickham. ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York, 2016.

HilleRisLambers, J., P. B. Adler, W. S. Harpole, J. M. Levine, and M. M. Mayfield. 2012.

Rethinking community assembly through the lens of coexistence theory. Annual Review of Ecology, Evolution, and Systematics 43:227–248.

Hulvey, K. B., and P. A. Aigner. 2014. Using filter-based community assembly models to improve restoration outcomes. Journal of Applied Ecology 51:997–1005.

Johnston, D. B. 2014. Rangeland restoration with super-absorbent polymoer and pothole surface at Horsethief State Wildlife Area.

Jonas, J. L., M. N. Grant-Hoffman, and M. W. Paschke. 2018. Restoration of North American salt deserts: A look at the past and suggestions for the future. Ecological Restoration 36:177–194.

Kachergis, E., M. E. Fernandez-Gimenez, and M. E. Rocca. 2012. Differences in plant species composition as evidence of alternate states in the sagebrush steppe. Rangeland Ecology & Management 65:486–497.

Kachergis, E., M. E. Rocca, and M. E. Fernández-Giménez. 2014. Long-term vegetation change provides evidence for alternate states in silver sagebrush. Rangeland Ecology & Management 67:183–194.

Kuznetsova, A., Brockhoff, PB., Christensen RHB (2017). lmerTest Package: Tests in Linear Mixed Effects Models. *Journal of Statistical Software*. 82(13), 1-26. DOI: 10.18637/jss.v082.i13 (URL: https://doi.org/10.18637/jss.v082.i13).

Naeth, M. A., A. C. Cohen Fernández, F. P. O. Mollard, L. Yao, S. R. Wilkinson, and Z. Jiao. 2018. Enriched topographic microsites for improved native grass and forb establishment in reclamation. Rangeland Ecology & Management 71:12–18.

Paschke, M. W., L. B. Perkins, and K. E. Veblen. 2019. Restoration for multiple use. Restoration Ecology 27:701–704.

PRISM Climate Group, Oregon State University, http://prism.oregonstate.edu, created 4 Feb 2004. (40.7431°N 107.1281°W)

Russell Lenth (2019). emmeans: Estimated Marginal Means, aka Least-Squares Means. R package version 1.4.1. https://CRAN.R-project.org/package=emmeans

Ryan M. Hope (2013). Rmisc: Rmisc: Ryan Miscellaneous. R package version 1.5. https://CRAN.R-project.org/package=Rmisc

Soil Survey Staff, Natural Resources Conservation Service, United States Department of Agriculture. 2006. Official Soil Series Descriptions. Jokodowski Series. Available online at https://soilseries.sc.egov.usda.gov/OSD_Docs/J/JOKODOWSKI.html. Accessed [8/27/2019].

USDA Forest Service. 2003. Integrated management plan for the California Park Special Interest Area. Steamboat Springs, Colorado.

USDA Farm Service Agency (FSA) Aerial Photography Field Office. 2017 Release. Available at https://www.usgs.gov/centers/eros/science/usgs-eros-archive-products-overview?qt-science_center_objects=0#qt-science_center_objects

USDA, NRCS. 2019. The PLANTS Database (http://plants.usda.gov, 28 September 2019).

National Plant Data Team, Greensboro, NC 27401-4901 USA.

Winkler, D. E., D. M. Backer, J. Belnap, J. B. Bradford, B. J. Butterfield, S. M. Copeland, M. C. Duniway, A. M. Faist, S. E. Fick, S. L. Jensen, A. T. Kramer, R. Mann, R. T. Massatti, M. L. McCormick, S. M. Munson, P. Olwell, S. D. Parr, A. A. Pfennigwerth, A. M. Pilmanis, B. A. Richardson, E. Samuel, K. See, K. E. Young, and S. C. Reed. 2018. Beyond traditional ecological restoration on the Colorado Plateau. Restoration Ecology 26:1055–1060.

Yates, C. J., D. A. Norton, and R. J. Hobbs. 2000. Grazing effects on plant cover, soil and microclimate in fragmented woodlands in south-western Australia: implications for restoration. Austral Ecology 25:36–47.

APPENDIX: SUPPLEMENTAL INFORMATION & DATA

Appendix Table 1: PRISM Climate Data for California Park and Grand Junction, Colorado. Thirty-year climate normals presented for 1981-2010 and yearly precipitation and temperature for each year of the study (2017-2019). From PRISM Climate Group, Oregon State University, http://prism.oregonstate.edu.

California Park, Colorado (40.7431°N 107.1281°W)								
	1981 - 2010		2017		2018		2019	
	ppt (mm)	MAT (°C)	ppt (mm)	MAT (°C)	ppt (mm)	MAT (°C)	ppt (mm)	MAT (°C)
January	71.4	-6.7	154.2	-6.5	55.4	-3.5	97.3	-6.1
February	71.5	-5.4	98.8	-1.1	67.8	-4.9	80.3	-7.2
March	69.3	-1.3	22.1	2.9	59.7	-0.9	118.9	-2.4
April	74.0	3	84.9	3.4	49.4	4	66.3	4
May	66.9	7.8	60.8	7.8	24.0	10.8	101.4	6.1
June	40.4	12.7	6.9	14.9	9.4	15.6	82.4	12.1
July	36.1	16.7	37.1	18	9.2	18.9	13.0	18.1
August	35.8	15.8	21.3	16.3	41.6	16.8	11.1	17.5
September	50.2	11.4	57.3	12.4	8.9	14.9	23.4	13.7
October	58.3	5.3	72.2	5	70.2	4.7	N/A	N/A
November	69.5	-1.9	48.0	2.7	82.6	-3.4	N/A	N/A
December	74.7	-6.7	47.2	-3.2	46.7	-6.7	N/A	N/A
Annual	718.2	4.2	710.7	6.1	524.6	5.5		

Grand Junction, Colorado (39.0734° N, 108.5373° W)								
	1981 - 2010		2017		2018		2019	
	ppt (mm)	MAT (°C)	ppt (mm)	MAT (°C)	ppt (mm)	MAT (°C)	ppt (mm)	MAT (°C)
January	12.6	-2	24.2	-0.9	13.0	0.2	12.8	-2.6
February	12.0	1.7	15.0	5.4	24.8	2.3	15.3	0.3
March	19.3	7	6.9	10.0	12.3	7.1	56.1	6.4
April	22.4	11.2	14.7	11.1	16.1	12.3	20.6	12.8
May	18.1	16.7	19.6	15.8	3.4	18.6	26.6	13.6
June	12.8	22	1.4	24.6	3.8	24.5	18.8	21.2
July	18.3	25.6	28.7	26.7	3.3	27.4	9.6	26.7
August	26.2	24.1	21.5	24.5	24.8	24.8	3.0	25.9
September	26.6	19	17.2	19.8	0.6	21.6	8.2	20.7
October	27.9	11.8	11.3	10.9	79.8	11	N/A	N/A
November	21.0	4.6	1.7	8.0	3.5	2.4	N/A	N/A
December	13.4	-1.4	0.5	0.2	19.3	-0.7	N/A	N/A
Annual	230.5	11.7	162.6	13.0	204.6	12.6		

Appendix Table 2a: Soil data for the upper 5cm of each California Park soil seed bank study sites sampled in 2017. Texture fractions by hydrometer, pH and EC by paste, and soil nutrients analyzed with AB-DTPA at the Colorado State University Soil, Water, and Plant Testing Lab. Undesirable sites (D) and Desirable or Reference sites (R) noted.

California Park Shrubland Sites							
	D1	D2	D3	D4	R1	R2	R3
Texture Class	sandy loam	sandy loam	sandy clay loam	sandy clay loam	sandy clay loam	sandy clay loam	sandy loam
Sand	58	62	60	55	61	59	57
Silt	27	21	19	18	17	14	25
Clay	15	17	21	27	22	27	18
pH	6.0	5.9	5.9	6.3	5.9	6.3	6.1
EC (mmhos/cm)	0.3	0.4	0.3	0.4	0.5	0.4	0.3
% O.M.	6.1	6.6	6.6	6.4	7.0	6.8	9.9
NO3-N mg/kg	2.5	3.6	4.6	7.1	2.3	3.1	3.7
P mg/kg	13.8	14.2	12.7	13.4	11.8	13.0	17.1
K mg/kg	225.9	222.5	237.7	410.0	257.9	240.2	478.8
Zn mg/kg	2.8	3.1	3.0	3.4	3.9	2.3	3.9
Fe mg/kg	61.9	53.9	63.7	47.3	68.9	50.6	80.9
Mn mg/kg	8.2	6.0	7.1	3.6	6.6	5.9	6.7
Cu mg/kg	4.2	4.1	4.7	3.4	3.5	4.0	2.4

Appendix Table 2b: Soil nutrient data for California Park soil seed bank study sites sampled in 2017 analyzed using a Mehlich III extraction procedure and ICP-AES. Thirty-three elements analyzed for, those with <1 ppm (mg/kg) concentrations removed due to detection limits (including Ag, As, B, Cd, Co, Cr, Hg, Li, Mo, Sb, Sn, Ti, Tl, U, and W).

California Park Shrubland Sites							
	D1	D2	D3	D4	R1	R2	R3
Al mg/kg	481.9	540.9	360.4	412.2	380.6	380.1	500.1
Au mg/kg	3.4	1.5	4.5	5.5	5.3	5.4	8.1
Ba mg/kg	56.9	155.5	37.5	62.8	61.6	40.0	66.6
Ca mg/kg	3751.6	4412.5	3299.3	3070.2	4358.2	1981.9	3574.9
Cu mg/kg	3.6	3.6	2.1	2.3	2.7	2.3	2.5
Fe mg/kg	229.7	255.1	143.1	184.5	201.4	166.2	201.4
K mg/kg	434.6	499.8	325.1	488.4	421.9	309.0	800.1
Mg mg/kg	1128.3	1253.4	826.0	785.2	899.0	1047.6	612.0
Mn mg/kg	43.5	26.0	18.0	27.0	18.6	50.7	40.0
Na mg/kg	18.3	17.6	12.7	15.3	17.3	25.7	9.0
Ni mg/kg	1.7	2.9	0.8	1.0	1.8	0.9	0.7
P mg/kg	50.2	59.4	41.0	52.5	46.7	52.5	119.6
Pb mg/kg	1.8	4.3	2.0	2.7	3.1	1.9	2.2
S mg/kg	19.8	19.8	11.5	15.0	8.8	9.1	19.6
Se mg/kg	3.5	3.1	2.5	2.9	2.8	2.2	4.6
Si mg/kg	119.5	125.9	79.8	96.6	90.7	91.9	67.3
Sr mg/kg	33.4	28.9	23.4	20.6	30.9	15.0	26.6
V mg/kg	8.2	8.5	7.9	9.1	7.4	10.3	9.8
Zn mg/kg	12.4	13.1	9.1	11.7	12.0	12.1	15.2

Appendix Table 2c: Soil data for the upper 5cm of each salt desert shrubland soil seed bank study site sampled in 2017. Texture fractions by hydrometer, pH and EC by paste, Sodium Absorption Ratio (SAR), and soil nutrients analyzed with AB-DTPA at the Colorado State University Soil, Water, and Plant Testing Lab. Undesirable sites (Undes.) and Desirable sites (Des.) noted.

Salt Desert Shrubland Sites							
	2 Road Fire (Undes.)	2 Road Seeded (Undes.)	Alkali Exclosure (Undes.)	Badger Wash Degraded (Undes.)	Badger Wash Reference (Des.)	Peeples Fire (Undes.)	Relic Exclosure (Des.)
Texture Class	sandy loam	sandy loam	clay loam	clay loam	sandy clay loam	sandy clay loam	sandy clay loam
Sand	59	61	42	43	57	50	55
Silt	22	21	23	24	16	22	16
Clay	19	18	35	33	27	28	29
pH	7.71	8.03	7.86	7.79	7.8	8.46	7.8
EC (mmhos/cm)	0.5	0.6	5.7	2.6	2.6	1.4	3.2
SAR	0.1	0.2	6.8	1.2	1.1	7.1	2.6
% O.M.	1.8	1.8	2.2	1.7	1.7	2.1	2
NO3-N mg/kg	4.3	3.7	28.5	3.3	5.1	9.2	8.9
P mg/kg	13.19	13.01	15.16	9.57	10.15	12.98	9.11
K mg/kg	302.2	430.9	355	140.6	173.8	991.3	257.7
Zn mg/kg	0.21	0.08	0.32	0.1	0.44	0.5	0.09
Fe mg/kg	2.3	2.04	2.98	3.67	3.94	3.91	2.44
Mn mg/kg	2.98	4.74	1.6	1.71	2.19	1.89	1.31
Cu mg/kg	0.93	1.17	2.49	1.89	1.4	3.7	1.5

Appendix Table 2d: Relative frequency of the dominant cover type recorded during soil seed bank sampling in California Park in 2017.

California Park Shrubland Sites							
	D1	D2	D3	D4	R1	R2	R3
Bare	0.08	0.07	0.23	0.13	0.12	0.15	0.05
Grass	0.5	0.42	0.25	0.1	0.62	0.42	0.37
Forb	0.42	0.52	0.47	0.77	0.27	0.23	0.23
Shrub	0	0	0.05	0	0	0.2	0.35

Appendix Table 2e: Relative frequency of the dominant cover type recorded during soil seed bank sampling in the Salt Desert in 2017.

Salt Desert Shrubland Sites							
	2 Road Fire	2 Road Seeded	Alkali Exclosure	Badger Wash Degraded	Badger Wash Reference	Peeples Fire	Relic Exclosure
BSC	0	0	0	0.18	0.18	0	0.08
Bare	0.3	0.1	0.7	0.43	0.25	0.43	0.33
Annual Grass	0.53	0.83	0.05	0.08	0.02	0	0
Perennial Grass	0.03	0.02	0.02	0.12	0.13	0.03	0.45
Forb	0.05	0.02	0.13	0.05	0.1	0.42	0.02
Shrub	0.08	0.03	0.1	0.13	0.32	0.12	0.12

Appendix Table 3: California Park soil seed bank species abundance by site (seeds m^{-2}) (USDA NRCS 2019)

USDA Code	Species Name	Common Name	Native or Introduced	Functional Group	Duration	D1	D2	D3	D4	R1	R2	R3
ACLE9	*Achnatherum lettermanii* (Vasey) Barkworth	Letterman's needlegrass	Native	Graminoid	Perennial	37	111	16	5	58	0	0
ACMI2	*Achillea millefolium* L.	common yarrow	Native	Forb/herb	Perennial	47	5	0	0	0	11	32
ALLIU	*Allium* L.	onion	Native	Forb/herb	Perennial	0	11	0	0	0	0	0
ANSE4	*Androsace septentrionalis* L.	pygmyflower rockjasmine	Native	Forb/herb	Annual, Perennial	5	5	280	5	26	0	100
ARAR8	*Artemisia arbuscula* Nutt.	little sagebrush	Native	Shrub, subshrub	Perennial	0	0	0	0	0	5	0
ARCA13	*Artemisia cana* Pursh	silver sagebrush	Native	Shrub, subshrub	Perennial	0	0	0	0	0	0	26
ARCO5	*Arenaria congesta* Nutt.	ballhead sandwort	Native	Forb/herb, subshrub	Perennial	0	0	5	0	0	5	0
ARHO2	*Arabis holboellii* Hornem.	Holboell's rockcress	Native	Forb/herb	Perennial	5	0	5	0	0	32	0
ARTR2	*Artemisia tridentata* Nutt.	big sagebrush	Native	Shrub	Perennial	0	0	11	0	0	0	0

USDA Code	Species Name	Common Name	Native or Introduced	Functional Group	Duration	D1	D2	D3	D4	R1	R2	R3
ASMIO	*Astragalus miser* Douglas ex Hook. var. *oblongifolius* (Rydb.) Cronquist	timber milkvetch	Native	Forb/herb	Perennial	5	0	0	0	5	0	0
ASTER	Unknown Aster	unknown aster	Native	Forb/herb	Perennial	0	5	0	0	0	0	0
ASTRA	*Astragalus* sp. L.	milkvetch	Native	Forb/herb	Perennial	0	0	5	0	0	11	0
BRIN2	*Bromus inermis* Leyss.	smooth brome	Introduced	Graminoid	Perennial	0	0	0	16	0	0	0
BRMA4	*Bromus marginatus* Nees ex Steud.	mountain brome	Native	Graminoid	Perennial	16	0	5	47	0	0	185
CAHO5	*Carex hoodii* Boott	Hood's sedge	Native	Graminoid	Perennial	0	0	0	21	5	0	16
CAOL	*Cardamine oligosperma* Nutt.	little western bittercress	Native	Forb/herb	Annual, biennial, perennial	0	5	0	0	0	0	0
CLLA2	*Claytonia lanceloatum* Pall. Ex. Pursh	lanceleaf springbeauty	Native	Forb/herb	Perennial	0	0	0	5	0	0	0
COLI2	*Collomia linearis* Nutt.	tiny trumpet	Native	Forb/herb	Annual	0	26	100	32	5	0	53

USDA Code	**Species Name**	**Common Name**	**Native or Introduced**	**Functional Group**	**Duration**	**D1**	**D2**	**D3**	**D4**	**R1**	**R2**	**R3**
COPA3	*Collinsia parviflora* Lindl.	maiden blue eyed Mary	Native	Forb/herb	Annual	0	0	5	5	0	148	0
DAGL	*Dactylis glomerata* L.	orchardgrass	Introduced	Graminoid	Perennial	0	0	0	0	0	16	0
DECE	*Deschampsia cespitosa* (L.) P. Beauv.	tufted hairgrass	Native	Graminoid	Perennial	16	0	5	0	0	311	11
DRNE	*Draba nemorosa* L.	woodland draba	Native	Forb/herb	Annual	0	0	0	0	0	0	11
ELLA3	*Elymus lanceolatus* (Scribn. & J.G. Sm.) Gould	thickspike wheatgrass	Native	Graminoid	Perennial	0	0	0	37	0	0	0
ELTR7	*Elymus trachycaulus* (Link) Gould ex Shinners	slender wheatgrass	Native	Graminoid	Perennial	0	0	0	0	0	127	0
EPBR3	*Epilobium brachycarpum* C. Presl	tall annual willowherb	Native	Forb/herb	Annual	5	37	0	5	0	0	0
EPLA3	*Epilobium lactiflorum* Hausskn.	milkflower willowherb	Native	Forb/herb	Perennial	142	0	0	0	0	0	5

USDA Code	Species Name	Common Name	Native or Introduced	Functional Group	Duration	D1	D2	D3	D4	R1	R2	R3
ERIGE2	*Erigeron* sp. L.	fleabane	Native	Forb/herb	Perennial	0	5	0	0	0	5	0
ERPE3	*Erigeron peregrinus* (Banks ex Pursh) Greene	subalpine fleabane	Native	Forb/herb	Perennial	0	0	0	0	0	16	0
ERSP4	*Erigeron speciosus* (Lindl.) DC.	aspen fleabane	Native	Forb/herb, subshrub	Perennial	5	0	0	0	0	37	0
ERUR2	*Erigeron ursinus* D.C. Eaton	Bear River fleabane	Native	Forb/herb	Perennial	0	0	21	5	0	0	84
FEID	*Festuca idahoensis* Elmer	Idaho fescue	Native	Graminoid	Perennial	0	0	132	0	21	90	5
FETH	*Festuca thurberi* Vasey	Thurber's fescue	Native	Graminoid	Perennial	0	0	11	0	0	0	0
IRMI	*Iris missouriensis* Nutt.	Rocky Mountain iris	Native	Forb/herb	Perennial	0	5	0	0	0	0	0
KOMA	*Koeleria macrantha* (Ledeb.) Schult.	prairie Junegrass	Native	Graminoid	Perennial	0	5	0	0	0	0	0
MAGL2	*Madia glomerata* Hook.	mountain tarweed	Native	Forb/herb	Annual	0	0	5	0	0	0	0

USDA Code	Species Name	Common Name	Native or Introduced	Functional Group	Duration	D1	D2	D3	D4	R1	R2	R3
MESP	*Melica spectabilis* Scribn.	purple oniongrass	Native	Graminoid	Perennial	0	0	0	0	190	42	26
NOFE3	*Noccaea fendleri* (A. Gray) Holub	Fendler's pennycress	Native	Forb/herb	Perennial	0	0	0	5	0	5	0
ORLU2	*Orthocarpus luteus* Nutt.	yellow owl's-clover	Native	Forb/herb	Annual	0	0	0	0	0	5	0
PASM	*Pascopyrum smithii* (Rydb.) Á. Löve	western wheatgrass	Native	Graminoid	Perennial	21	11	5	0	0	153	26
PHPR3	*Phleum pratense* L.	Timothy	Introduced	Graminoid	Perennial	396	37	69	16	1214	0	142
POPR	*Poa pratensis* L.	Kentucky bluegrass	Introduced	Graminoid	Perennial	53	0	137	5	11	32	174
POTEN	*Potentilla* sp. L.	cinquefoil	Native	Forb/herb	Perennial	132	32	74	0	116	0	322
PSJA2	*Psuedostellaria jamesiana* (Torr.) W.A. Weber & R.L. Hardman	sticky starwort	Native	Forb/herb	Perennial	0	5	0	0	0	0	0
RAIN	*Ranunculus inamoenus* Greene	graceful buttercup	Native	Forb/herb	Perennial	0	0	0	0	0	0	132
SIDR	*Silene drummondii* Hook.	Drummond’s campion	Native	Forb/herb	Perennial	0	0	0	5	0	0	0

USDA Code	Species Name	Common Name	Native or Introduced	Functional Group	Duration	D1	D2	D3	D4	R1	R2	R3
SOCA6	*Solidago canadensis* L.	Canada goldenrod	Native	Forb/herb	Perennial	0	0	0	0	0	0	26
STCA	*Stellaria calycantha* (Ledeb.) Bong.	northern starwort	Native	Forb/herb	Annual, Perennial	0	0	5	0	0	0	0
TAOF	*Taraxacum officinale* F.H. Wigg.	common dandelion	Introduced	Forb/herb	Perennial	21	0	16	0	47	79	0
TRDU	*Tragopogon dubius* Scop.	yellow salsify	Introduced	Forb/herb	Annual, Biennial	5	0	5	0	0	0	0

Appendix Table 4: Salt desert shrubland soil seed bank species abundance (seeds m^{-2}) (USDA, NRCS 2019). Site names: Alkali Exlcosure (AE – Undesirable), Badger Wash Degraded (BWD – Undesirable), Badger Wash Reference (BWR- Desirable), Peeples Fire (PF – Undesirable), Relix Exclosure (RE – Desirable), Two Road Fire (2RF – Undesirable), Two Road Seeded (2RS – Undesirable).

USDA Codes	**Species Name**	**Common Name**	**Native / Introduced**	**Functional Group**	**Duration**	**AE**	**BWD**	**BWR**	**PF**	**RE**	**2RF**	**2RS**
ALAN3	*Aletes anisatus* (A. Gray) W.L. Theobald & C.C. Tseng	Rocky Mountain Indian parsley	Native	Forb/herb	Perennial	11	0	0	0	0	0	0
ALDE	*Alyssum desertorum* Stapf	desert madwort	Introduced	Forb/herb	Annual	0	0	0	0	0	0	26
ARLI	*Arabis lignifera* A. Nelson	desert rockcress	Native	Forb/herb	Perennial	0	0	5	0	0	0	0
ASFL2	*Astragalus flexuosus* Douglas ex G. Don	flexile milkvetch	Native	Forb/herb	Perennial	0	5	0	0	0	0	0
ATCA2	*Atriplex canescens* (Pursh) Nutt.	fourwing saltbush	Native	Shrub	Perennial	0	0	0	0	5	0	0
ATGA	*Atriplex gardneri* (Moq.) D. Dietr.	Gardner's saltbush	Native	Shrub, Subshrub	Perennial	0	5	0	0	0	0	0
BASC5	*Bassia scoparia* (L.) A.J. Scott	burningbush, kochia	Introduced	Forb/herb	Annual	84	0	0	0	0	0	0
BRTE	*Bromus tectorum* L.	cheatgrass	Introduced	Graminoid	Annual	5	26	142	0	195	211	422
CETE5	*Ceratocephala testiculata* (Crantz) Roth	curveseed butterwort	Introduced	Forb/herb	Annual	16	47	0	0	0	0	5

USDA Codes	**Species Name**	**Common Name**	**Native / Introduced**	**Functional Group**	**Duration**	**AE**	**BWD**	**BWR**	**PF**	**RE**	**2RF**	**2RS**
CHER2	*Chaetopappa ericoides* (Torr.) G.L. Nesom	rose heath	Native	Forb/herb	Perennial	0	0	0	0	0	5	0
CHTE2	*Chorispora tenella* (Pall.) DC.	crossflower	Introduced	Forb/herb	Annual	32	0	0	0	0	5	32
COCA5	*Conyza canadensis* (L.) Cronquist	Canadian horseweed	Native	Forb/herb	Annual, Biennial	0	0	0	0	5	0	0
CRAM3	*Cryptantha ambigua* (A. Gray) Greene	basin cryptantha	Native	Forb/herb	Annual	0	0	16	32	16	0	0
CRHU2	*Cryptantha humilis* (A. Gray) Payson	roundspike cryptantha	Native	Forb/herb, subshrub	Perennial	0	0	5	0	0	0	0
CRMI5	*Cryptantha minima* Rydb.	little cryptantha	Native	Forb/herb	Annual	11	16	0	0	0	5	5
DEPI	*Descurainia pinnata* (Walter) Britton	western tansymustard	Native	Forb/herb	Annual, Biennial, Perennial	42	5	53	475	63	37	0
DRRE2	*Draba reptans* (Lam.) FERNA10ld	Carolina draba	Native	Forb/herb	Annual	0	0	0	0	0	69	0
ELEL5	*Elymus elymoides* (Raf.) Swezey	squirreltail	Native	Graminoid	Perennial	16	16	11	0	53	0	0
ENDI	*Endolepis dioica* (Nutt.) Standl.	Suckley's endolepis	Native	Forb/herb	Annual	5	0	0	0	0	0	0
EPLA3	*Epilobium lactiflorum* Hausskn.	milkflower willowherb	Native	Forb/herb	Perennial	0	0	5	0	0	0	0

USDA Codes	**Species Name**	**Common Name**	**Native / Introduced**	**Functional Group**	**Duration**	**AE**	**BWD**	**BWR**	**PF**	**RE**	**2RF**	**2RS**
ERCA14	*Erysimum capitatum* (Douglas ex Hook.) Greene	sanddune wallflower	Native	Forb/herb, subshrub	Biennial, Perennial	32	0	0	5	84	0	0
ERCI6	*Erodium cicutarium* (L.) L'Hér. ex Aiton	redstem stork's bill	Introduced	Forb/herb	Annual, Biennial	0	0	5	0	0	11	16
ERTR13	*Eremopyrum triticeum* (Gaertn.) Nevski	annual wheatgrass	Introduced	Graminoid	Annual	375	5	0	0	0	0	0
GISI	*Gilia sinuata* Douglas ex Benth	rosy gilia	Native	Forb/herb	Annual	0	0	5	0	0	0	0
GUSA2	*Gutierrezia sarothrae* (Pursh) Britton & Rusby	broom snakeweed	Native	Forb/herb, Shrub, Subshrub	Perennial	21	0	0	0	42	26	5
HAGL	*Halogeton glomeratus* (M. Bieb.) C.A. Mey.	saltlover, halogeton	Introduced	Forb/herb	Annual	190	0	0	697	16	0	0
HECO26	*Hesperostipa comata* (Trin. & Rupr.) Barkworth	needle and thread	Native	Graminoid	Perennial	0	0	0	0	5	0	0
JUAR2	*Juncus arcticus* Willd.	arctic rush	Native	Graminoid	Perennial	11	0	0	0	0	0	0
LECI4	*Leymus cinereus* (Scribn. & Merr.) Á. Löve	basin wildrye	Native	Graminoid	Perennial	0	0	5	0	0	0	0

USDA Codes	Species Name	Common Name	Native / Introduced	Functional Group	Duration	AE	BWD	BWR	PF	RE	2RF	2RS
MAAF	*Malcolmia africana* (L.) W.T. Aiton	African mustard	Introduced	Forb/herb	Annual	0	95	47	0	16	5	0
MIMU	*Mirabilis multiflora* (Torr.) A. Gray	Colorado four o'clock	Native	Forb/herb, subshrub	Perennial	0	0	16	0	0	0	0
NALE	*Navarretia leucocephala* Benth.	whitehead navarretia	Native	Forb/herb	Annual	0	0	0	0	5	0	0
OECA10	*Oenothera caespitosa* Nutt.	tufted evening primrose	Native	Forb/herb, subshrub	Perennial	0	0	0	0	16	0	0
OREO	*Oreocarya* sp. Greene	cryptantha	Native	Forb/herb	Perennial	0	5	0	0	5	0	0
PHCR	*Phacelia crenulata* Torr. ex S. Watson	cleftleaf wildheliotrope	Native	Forb/herb	Annual	0	0	0	0	11	0	0
PIDE4	*Picrothamnus desertorum* Nutt.	bud sagebrush	Native	Shrub, Subshrub	Perennial	0	5	0	0	0	0	0
PLIN7	*Platyschkuhria integrifolia* (A. Gray) Rydb.	basindaisy	Native	Forb/herb, subshrub	Perennial	0	0	5	0	0	0	0
PLJA	*Pleuraphis jamesii* Torr.	James' galleta	Native	Graminoid	Perennial	0	0	0	0	5	0	0
PLPA2	*Plantago patagonica* Jacq.	wooly plantain	Native	Forb/herb	Annual	0	0	0	0	0	32	0
POFE	*Poa fendleriana* (Steud.) Vasey	muttongrass	Native	Graminoid	Perennial	0	0	26	0	0	0	0

USDA Codes	Species Name	Common Name	Native / Introduced	Functional Group	Duration	AE	BWD	BWR	PF	RE	2RF	2RS
SATR12	*Salsola tragus* L.	prickly Russian thistle	Introduced	Forb/herb	Annual	11	0	0	0	0	16	16
SPCO	*Sphaeralcea coccinea* (Nutt.) Rydb.	scarlet globemallow	Native	Forb/herb, subshrub	Biennial, Perennial	0	0	0	0	0	21	0
SPCR	*Sporobolus cryptandrus* (Torr.) A. Gray	sand dropseed	Native	Graminoid	Perennial	11	0	0	0	5	222	37
STAC	*Stenotus acaulis* (Nutt.) Nutt.	stemless mock goldenweed	Native	Forb/herb, subshrub	Perennial	0	42	37	0	5	0	0
STAL2	*Stanleya albescens* M.E. Jones	white princesplume	Native	Forb/herb	Biennial, Perennial	5	5	42	0	0	0	0
VUOC	*Vulpia octoflora* (Walter) Rydb.	sixweeks fescue	Native	Graminoid	Annual	0	0	26	0	0	248	26

Appendix Table 5: Seedling counts and distribution between soil layers of 24 species found in the seed bank of the Two Road Fire site sampled in June 2018. Nomenclature follows USDA NRCS PLANTS database (2019).

Species name	**Common name**	**Native or Non-native**	**Top (0 - 2 cm)**	**Bottom (2 - 5 cm)**	**Total (0 - 5 cm)**
Achnatherum hymenoides	Indian ricegrass	Native	0	1	1
Alyssum desertorum	desert madwort	Non-native	1	0	1
Amaranthus albus	prostrate pigweed	Non-native	1	0	1
Bromus tectorum	cheatgrass	Non-native	249	108	357
Chamerion angustifolium	fireweed	Native	0	1	1
Chaetopappa ericoides	rose heath	Native	6	2	8
Chamaesyce serpyllifolia	thymeleaf sandmat	Native	1	1	2
Chorispora tenella	blue mustard	Non-native	1	3	4
Cryptantha minima	little cryptantha	Native	52	20	72
Descurainia pinnata	western tansymustard	Native	30	21	51
Draba cunefolia	wedgeleaf draba	Native	5	1	6
Draba reptans	Carolina draba	Native	31	10	41
Epilobium ciliatum	fringed willowherb	Native	1	2	3
Erodium cicutarium	redstem stork's bill	Non-native	4	1	5
Gutierrezia sarothrae	broom snakeweed	Native	8	5	13
Lactuca serriola	prickly lettuce	Non-native	1	1	2
Lepidium huberii	Huber's pepperweed	Native	18	3	21
Malcolmia africana	African mustard	Non-native	4	1	5
Plantago patagonica	wooly plantain	Native	36	6	42
Salsola tragus	prickly Russian thistle	Non-native	4	1	5
Sisymbrium altissimum	tall tumblemustard	Non-native	5	7	12
Sphaeralcea coccinea	scarlet globemallow	Native	3	6	9
Sporobolus cryptandrus	sand dropseed	Native	239	118	357
Vulpia octoflora	sixweeks fescue	Native	56	24	80
		Total seedling count	756	343	1099