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Methods and Application for Tracking Seedling Fate on the Utah Test and Training Range

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on the Utah Test and Training Range

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ABSTRACT

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Remote sensing of the environment has become an effective and useful research approach applied across a wide range of scientific and professional disciplines. Generally remote sensing is used to evaluate patterns and processes at broad spatio-temporal scales, such as classifying landscape vegetation patterns or for creating digital surface models, however, there are increasing opportunities to expand the use of remotely sensed information to a wider range of applications at variable spatial and temporal scales. In the field of plant seedling and germination research methods are needed to improve plant establishment and restoration monitoring, particularly in areas that have historically low success rates such as in semi-arid and arid rangeland landscapes. The purpose of this research is to assess the efficacy of remote sensing for tracking seedling height, seedling density, and seedling fate, and determine the biotic causes of seedling mortality in a rangeland revegetation site in northwestern Utah. In Chapter 1, we use 28 time-lapse and motion sensing infrared cameras (Reconyx) to measure seedling density and height in fenced and unfenced plots during the initial four months of seedling establishment and growth. We compare imaged-based measurements of seedling height and density with similar measurements collected in the field and at different daylight hours to determine the accuracy and reliability of remotely sensed measurements. We found that the ideal sample periods for capturing the clearest images were at the time the sun passed zenith and shadows were minimized. Average seedling height was 14% lower in image-based versus field estimates. Seedling density was underestimated by approximately 30% when using cameras. Our study establishes that remote sensing of seedlings using time-lapse cameras is a method for seedling research and monitoring in restoration efforts which merits further research and development. In Chapter 2, we track biotic causes of seedling fate using the methods developed in Chapter 1, and compare seedling survival in fenced and unfenced plots. Fencing led to a four-fold increase in the number of seedlings emerged from the soil. Herbivory and damage caused by trampling and burial resulted in the death of 61.4 % of all unfenced seedlings. Fencing plots increased the probability of seedling survival by seven times. Using cameras to track seedling fate at two restoration sites revealed that small herbivores, including *Lepus californicus*, *Thomomys bottae*, and *Dipodomys* sp. drastically reduced seedling survival during the first year after planting. Effects of herbivores on seedling survival should be taken into consideration when planning revegetation operations, and further research can increase knowledge of how herbivory affects restoration efforts. Using cameras can provide meaningful information to managers and researchers about seedling status and fate.

Keywords: remote sensing, motion camera, seedling survival, herbivores, small mammals, time-lapse, rangeland, restoration

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CHAPTER 1

Evaluation of Remotely Triggered Cameras as a Method for
Measuring Seedling Emergence, Growth and Survival

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ABSTRACT

In the field of plant seedling and germination research, understanding the causes of plant

mortality is necessary to develop solutions that will improve the success of direct seeding and

restoration monitoring efforts. The purpose of this research is to assess the efficacy of time lapse

and motion sensing cameras for tracking seedling height and density, and the biotic causes of

seedling fate in a reseeded rangeland. The study sites were located on the Utah Test and Training

Range (UTTR), Utah in salt desert shrub plant communities. In spring 2017, we placed 28

cameras in fenced and unfenced plots that were seeded with Elymus elymoides (Raf.) Swezey

using a randomized split-plot study design. At each plot, we used a Reconyx PC 900 camera to

photograph emerged seedlings at hourly intervals throughout the daylight hours during the initial

four months of seedling growth. Seedling density and height were recorded in-field and

compared with camera images to determine accuracy and reliability of the remotely sensed

images. We found that the ideal sample period for capturing the clearest images occur when the

sun has recently passed its zenith and shadows are minimized. Average seedling height and

density were underestimated by 14% and 30% between camera and field estimates, respectively.

Reducing seedling density may improve measurement accuracy from images. Additional

research is needed to refine the use of cameras for seedling research and monitoring in

restoration efforts and provide meaningful information to managers and researchers about seedling survival potential and ultimately the fate of the individual plant.

INTRODUCTION

Rangelands account for a large proportion of terrestrial land surface (~51 %), including land that supports tremendous biodiversity and provides valuable ecological services [1]. Human use of rangelands, though varied, is extensive and often [1] these impacts can lead to plant community alterations and disturbance such as the reduction of native plant populations and impaired ecological processes [2]. Degraded lands often promote the colonization and dominance of invasive species and altered fire regimes [3-5]. Using technology can improve the ability of managers to monitor plant community changes and how they affect rangeland resources and ecological services.

In contrast to plant communities dominated by perennial vegetation, annual invasive weeds often facilitate impaired ecological processes that result from altered fire regimes, changed erosion dynamics, and changed biotic community dynamics [6-8]. Such degraded plant community states often experience decreased forage quality and cover for wildlife and a reduction in animal diversity [9]. Lower plant and animal diversity reduce biosphere integrity, which is already at risk worldwide [10].

To increase biotic integrity, improve plant community function, and reduce fire risk, land managers reseed degraded rangeland using desirable plant species, in particular, perennial grasses, forbs and shrubs [7]. Annually, more than US $100 million dollars are spent in an effort to restore degraded rangelands worldwide [11-13], however, these efforts have historically demonstrated low success rates [7,11,14]. Methods are needed to better monitor and assess

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reseeding efforts that can lead to extensive cost-savings, higher restoration success, and improved ecosystem function [7,11,12].

Technological advances are integral for improving restoration success through increasing our understanding of seed ecology and limitations in plant establishment. Developing improved monitoring techniques for characterizing demographic stages, including seed germination, seedling emergence, seedling establishment, and survival to an adult plant, will increase our understanding of limitations to revegetation success [15-18]. To improve our understanding of these processes we must determine how they relate to environmental variables (biotic and abiotic factors). Monitoring these characteristics requires frequent site visits to quantify numbers of seedlings (density), plant growth rates, and seedling survival. Restoration monitoring is often challenging because field-travel logistics and limited time and resource availability decrease sample collection frequency. This may be especially difficult in remote study areas or sites with poor access. Current monitoring methods can provide valuable information, but are on a limited temporal scale. Methods that can record greater detail of what transpires in reseeded areas will increase our ability to monitor restoration efforts Remote sensing technology, a research tool that has been widely accepted and applied across scientific and professional disciplines, has been developed to reduce the time and effort required for collection of vegetation data while increasing the ability of researchers to study ecological processes in greater detail [19].

Remote sensing is the collection of data with sensors from a distance [20]. Remote sensing for natural resource management is often applied at broad spatial scales, such as classifying landscape vegetation types or creating digital elevation models [21,22]. However, there are increasing opportunities to expand the use of remotely sensed data to multiple scales and applications, such as agricultural and wildland seedling monitoring [23]. One such application of

remote sensing is the use of camera traps, which were developed as a non-invasive method to remotely collect information using motion-sensing infrared cameras [24, 25]. Research using camera traps varies widely and includes applications such as estimation of tiger densities in India [26], monitoring wildlife interactions with feral horses at water sources in the Great Basin, USA [27], determining chukar watering patterns and water site selection [28], and monitoring invasive rodents and rodent granivory [29,30]. Research involving vegetation monitoring that utilizes camera traps is limited. Less than one percent of camera trap studies included vegetation sampling; however, vegetation measurement using other camera technologies is a rapidly expanding field of research [31].

The first use of cameras for quantifying vegetation characteristics was in the 1920's when an apparatus for photographing vegetation quadrats was developed [32]. Adjustments and developments to this method have been made over the years [33-35], including the use of unmanned aerial systems (UAS) for photographing vegetation [36], but the same basic concept of photographic measurements (photogrammetry) of vegetation from above is still used [37-39]. These methods have been demonstrated to be accurate for measuring mature plants in both rangeland and agricultural settings [39-41]. Photographic monitoring of vegetation is currently used for measuring plant canopy cover, species composition, plant health, and change in the plant community or individual plants over time for mature individuals [42-44]. The use of photogrammetry in seedling research is much more limited and focuses mainly on large seedlings in precision agriculture and forestry, or measuring seedling characteristics in a lab setting with specialized equipment [23,45,46].

The purpose of this study was to develop a vegetation monitoring system that can track seedling height, density, and mortality in rangeland revegetation efforts using remote sensing

technology. Specifically, we tested the efficacy of motion and timed cameras (Reconyx) for quantifying seedling density, measuring seedling height, and determining the cause of plant mortality. We also discuss the decision making process involved in selecting a camera, adjusting settings, and positioning the camera for optimal data capture. By developing this method that tracks seedling dynamics at a fine scale, optimizes the frequency of sampling, and reduces travel costs, this technique can be used to improve seedling research and be a valuable tool for managers to monitor seeding success with greater accuracy and time efficiency.

MATERIALS AND METHODS

Study Site Description

This study was conducted at two locations, Murray's Mesa (41.036394°N, -112.979465°W) and Arctic Road (41.078425°N, -112.927195°W), on the Utah Test and Training Range (UTTR) located in the West desert of Utah, United States. This military-managed land is in a relatively low precipitation area of the semi-arid Great Basin Region, receiving approximately 258 mm of precipitation annually [47]. Murray's Mesa is located at 1399 m elevation with <4% slope. We determined through Brigham Young University's Environmental Analytical Lab (Provo, UT, USA) that the top 15 cm of soil contained 37.4% silt, 22.4% clay, and 40.2% sand with a pH of 7.8 and 1.3% organic matter. The Arctic Road site is located at 1338 m elevation with <4% slope and soil containing 47.4% silt, 26.4% clay, and 26.2% sand, a pH of 7.6, and 2.7% organic matter. Both sites consist of a degraded salt desert shrub community. Remnant native perennial plants include *Sarcobatus vermiculatus* (Hook.) Torr. (greasewood), *Atriplex confertifolia* (Torr. & Frem.) S. Watson (shadscale), *Artemisia spinescens* D.C. Eaton (bud sagebrush) and *Elymus elymoides* (Raf.) Swezey (bottlebrush

squirreltail) and *Achnatherum hymenoides* (Roemer & J.A. Schultes) Barkworth. Within these communities, military activity has contributed to increased fire frequency and the invasion of annual grasses and forbs including *Bromus tectorum* L. (cheatgrass), *Halogeton glomeratus* (Bieb.) C.A. Mey (halogeton), *Salsola iberica* (Sennen & Pau) Botsch. (Russian thistle), and *Sisymbrium altissimum* L. (tumble mustard). Revegetation from seed with a mix of native and introduced species was attempted at the Arctic Road site in 2016 with very little success, and at the Murray's Mesa site in 2017 with limited plant establishment.

Study Design

The study was implemented with 28 Reconyx PC900 (Reconyx, Holmen, WI, USA) remotely triggered cameras. We chose this camera model because it has factory-installed weatherproof protection, can be programmed to take both time-lapse and motion-triggered photos, is commonly used in wildlife research, can be modified to capture images at close range (≤60 cm), and is readily available to land managers and researchers in the wildlife and range disciplines. We placed 14 cameras at each site, arranged in a randomized split-plot design for a total of seven replications. Half of the plots were fenced to exclude herbivores. Each plot was hand-seeded on May 27 with *E. elymoides* in four 75 cm rows placed perpendicular to the camera position. The first row was placed 35 cm from the camera, and rows were spaced 20 cm apart to ensure visibility of individual rows on each image (Figure 1-1). Rows were marked on each end with a wood dowel to help in locating and counting seedlings both in the field and on images. Each row was seeded at a 0.5 cm depth with 50 pure live seeds, totaling 200 seeds per plot. Due to the dry climate at the study sites (~258 mm precipitation annually)[47], we watered plots daily to ensure sufficient soil moisture for seed germination and seedling emergence. One

plot was selected at each site to monitor soil moisture from 0-10 cm depths using Decagon MPS-6 dielectric water potential sensors (Meter Group Inc., Pullman, WA, USA). Plots were brought to field capacity (-33 kPa, 0.301 g of water 1g soil^{-1}) three days after planting and maintained at $\geq$50% of field capacity until all plots reached 50% emergence. Upon reaching 50% emergence, watering was reduced to only twice per week and completely discontinued five weeks after planting.

We placed cameras in each plot 10 cm above the soil surface and angled forward 15º. Keeping the cameras at this slightly elevated height reduced the amount of dust that collected on the lens and allowed for multiple rows to be visible in the camera's field of view. Had we left the camera level with the soil surface, the first row of seedlings would have blocked the seedlings in the rows behind it. Additionally, our preliminary work indicated that it was difficult to detect seedlings if the cameras were placed at nadir (directly above seedlings) because the area of the plant that was visible was much less than if viewed from the side. Camera focal length was factory adjusted to 61 cm. Cameras were programmed to capture one photo every hour from 8 AM to 7 PM (approximate light hours) daily. Cameras were also set to trigger with changes in infrared heat (caused by motion), taking three photos per trigger, with a wait period of 15 s between triggers. We checked cameras every two weeks for adequate battery life, proper functioning, and to replace memory cards. At these times, plots were cleared of weeds and camera lenses cleaned to maintain visibility of seedlings.

Using data collected in-field and on-camera, as well as practical experience, observations, and challenges encountered, we created a decision flow chart to inform camera setup and study design for using cameras in plant research. We divided the decision-making and study design into 6 main categories:

1. Primary focus of the study

2. Size/scale of the study subject

3. Primary features of subjects to be studied/measured

4. Plot setup

5. Camera settings

6. Camera maintenance

Measurements

We measured seedling height and density approximately 3 times per week from 10 June – 5 July, 2017, and collected a final measurement for both characteristics on 19 September, 2017. Seedling density and average height were measured in each plot by counting the number of seedlings per row and measuring the height of seedlings in each row. These measurements were taken in the field at the same time as photograph collection so the two measurements could be compared. Since photographs put seedlings on a one-dimensional image, seedlings farther back in the image appear smaller than seedlings closer to the camera. To calibrate image height measurements, we took images of seedlings and physically measured them concurrently to adjust for the amount of distortion to seedling height in images.

Motion-triggered images were collected continuously from 27 May - 20 September, 2017. Animals were identified as accurately as possible from the images, usually to the level of genus, and when possible to species. Images were used to determine whether the animal was grazing or otherwise damaging seedlings and how many seedlings had been damaged. The causes of damage to plants were quantified by tracking individual plants and documenting when entire plants or parts of plants failed to occur in subsequent images. If images showed an herbivore

consuming part or all of the plant, or if the plant was missing all or part of its vegetation directly after an herbivore was foraging at the plant, it was classified as a herbivory event for that animal. If no herbivore was detected when a plant was partially or wholly removed, it was classified as unknown herbivory. If a seedling was otherwise damaged by being buried or trampled, it was labeled accordingly.

Analysis

Seedling density and average seedling height measurements from images were compared to in-field measurements. Overall accuracy of seedling density and average seedling height measurements using remotely sensed images, and factors affecting measurement accuracy from images for these characteristics were analyzed using mixed model analysis of variance in SAS® (SAS Institute Inc., Cary, NC, USA), with an α=0.05. Factors included in the model for seedling height included date, row order from the camera, fencing, and the interaction of fencing and date. We performed the same analysis for seedling density including date, row order from the camera, fencing, and the interaction of date with fencing and with row as factors. After adjusting for these factors, we used a mixed model analysis of variance to determine whether time of day affected measurement accuracy from images. To determine the accuracy of cameras in detecting herbivory, we calculated frequency of herbivory events for each herbivore, and included a category for an unknown cause of herbivory.

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RESULTS

Average Height

Average seedling height was underestimated (p=0.03) by approximately 14% in remote

sensed images compared to field measurements. Factors affecting the accuracy of image

estimates for height included date (p<0.001), fencing (p<0.001), and the interaction of date and

fencing (p<0.001). Row order from the camera was the only factor that did not affect the

accuracy of seedling height estimates (p=0.4).(Table1-1). Measurements were underestimated

at earlier dates by as much as 27% (14 June) and became more accurate over time with a

difference of 8% on July 5 and no difference between field and image estimates at the last

measurement (19 September, Figure 1-2). Unfenced image estimates were 2.9 cm closer to field

measurements than fenced plot estimates, a difference of 16%. Fenced plot estimates of average

seedling height were overestimated by approximately 2 cm in June and became more accurate

over time (Figure 1-3B) while unfenced plot estimates were consistently overestimated by about

2.5 cm (Figure 1-3C). Accuracy of plant height from images did not differ between dates in

unfenced plots (Figure 1-3A). After adjusting for the effects of date, row, and fencing, time of

day had an effect on the accuracy of height estimates from images (p=0.026). No major patterns

were observed, but 3:00 and 4:00 PM were less accurate than 1:00, 5:00, and 7:00 PM but not

different from the other hours (Figure 1-4).

Seedling Density

Density estimates in images were different from field measurements and were

underestimated by approximately 30% (5.3 seedlings ¼ m^{-2}, p=0.019). Date (p<0.001), fencing

(p<0.001), the interactions of date with row order from the camera (p<0.001) and with fencing

(p=0.016),and time of day affected the accuracy of image estimates for seedling density. Row

order from the camera did not affect accuracy of image estimates for seedling density ($p=0.069$, Table 1-2). At earlier dates, seedling density was underestimated in images by approximately 16%, and increased with time until density estimates were accurate, then overestimated at the latest dates by 3% (July 5) and 31% (September 19, Figure 1-5). In fenced plots, seedling density was underestimated in images by 5.7%, whereas seedling density estimates were not affected in unfenced plots. Fenced plot densities were underestimated at earlier dates by up to 25% (June 14), and were not different from field measurements on July 5 and September 19 (Figure 1-6). At earlier dates, row order from the camera did not affect the accuracy of image density estimates compared to field measurements, but on September 19 rows 1 and 4 (the first and last rows) were overestimated compared to field measurements by 47% and 56%, respectively. After adjusting for date, row, and treatment, time of day had an effect on the accuracy of density estimates ($p<0.001$). Afternoon hours from 1:00 PM to 3:00 PM had more accurate seedling density estimates than morning hours (8:00 AM to 10:00 AM) and some evening hours (4:00 AM, 6:00 PM, and 7:00 PM, Figure 1-7).

Herbivory Detection

A large suite of herbivores was detected consuming seedlings in herbivory events (Table 1-3). Herbivory was detected and assigned to specific herbivores for 69.1% of damaged seedlings. Cause of seedling herbivory was unknown for 22.6% of seedlings, and 8.3% of seedlings were trampled or buried (Table 1-4). The smallest herbivore detected was *Acrididae* family (grasshoppers), which accounted for 5.6% of known herbivory to seedlings.

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DISCUSSION

Average Height

Researchers have used cameras to study plants [21,42,48] but they generally use satellites or cameras from ≥ 2 m distance where mature or large young plants like tree seedlings are subject [42,49]. The small size of *E. elymoides* seedlings relative to more mature plants creates a unique challenge in accurately assessing plant height in images. A small error in the calibration of seedling height measurements in images may have led to the 14% underestimation of seedling height. One method to improve the accuracy of image height estimates would be to place a ruler or small Robel pole style instrument [50] attached to a small dowel vertically next to seedlings at each row as a reference scale.

Fencing appeared to decrease the accuracy of height measurements from images. At later dates, height estimates from images were more accurate than at the beginning of the summer. A reasonable explanation for the patterns with date, fencing, and their interaction, is that higher seedling density made it more difficult to obtain accurate seedling measurements in images due to more visual obstruction from seedlings in the front rows. In unfenced plots, seedlings were often grazed by herbivores, maintaining or reducing the average height and density, and thus maintaining the accuracy of estimates. Since less-dense plants could lead to more accurate image estimates, this should be a consideration when measuring height on very small seedlings. When time of day analysis for height is considered, 1:00 PM, 5:00 PM, and 7:00 PM were the only hours that had more accurate image estimates than others. This is most likely due to the effects of shadows and the angle of the sun. To sample at the best time of day, researchers should consider the angle of the sun and visual obstructions that may cause shadows.

Seedling Density

The number of plants present tended to reduce accuracy of plant density estimates from images. For example, seedling densities were highest at earlier dates before seedlings had been grazed, which was also when seedlings densities were underestimated the most. Similar to seedling height, density estimates from images were underestimated more in the fenced plots, which had higher plant densities than unfenced plots. The high density averages were approximately 22 seedlings ¼ m^{-2}, and the low values ranged between 10-15 seedlings ¼ m^{-2}. Overall, density estimates became more accurate over time, which correlated with a reduction in seedling density. We felt that our ability to detect seedlings from images decreased with increasing seedling density because individual seedlings in rows closer to the camera would obstruct the view of other seedlings in rows farther from the camera. Additionally, seedlings growing close together were difficult to determine from the images if they were individual plants or tillers from the same plant.

Similar to aerial wildlife surveys reported in the literature [51], it appears that seedling density and height estimates could be influenced by different sightability factors (factors affecting the probability of seeing an individual) like number of seedlings in the image (analogous to group size in wildlife), visual obstruction (seedlings themselves, analogous to cover for wildlife), and size of the individuals [51]. Though this study did not calculate sightability adjustments for seedlings, models similar to wildlife sightability could be developed to adjust estimates based on probability of seeing individual seedlings [51].

Afternoon hours from 1:00 PM to 3:00 PM had the most accurate seedling density estimates. At the extremes of the day, the sun casts long shadows, affecting the visibility of seedlings in images. The sun is overhead at noon, but with no shadow the seedlings may be washed out in the

image and hard to see. As the sun passes its zenith, shadows may be cast from the seedlings, increasing visibility, but not being overcast from larger shadows as they are in the morning and evening. Best time of day for reducing the effect of shadows will also depend on season and location [52].

Herbivory Detection

Cameras were effective at capturing a majority of herbivory events (69%). It is likely that much of the unknown herbivory was caused by *Formicidae* family (ants), *Acrididae* family, or other small invertebrate herbivores that are too small with temperatures near ambient temperature to trigger the camera's infrared sensor. All but two (99.1 %) unknown herbivory events occurred during the day, which coincides with activity of diurnal species such as small invertebrate herbivores. A large suite of invertebrate herbivores such as *Formicidae* family, *Coleoptera* order (beetles), and *Acrididae* family can be encountered in the Great Basin which feed primarily on grasses like *E. elymoides* [53]. Additionally, 23.5 % of herbivory events occurred in fenced plots. In these fenced plots, herbivory events were from animals that were able to get past the boundary fence by flying over, burrowing under, or fitting through the spaces in the wire, such as *Eremophila alpestris* (horned lark), *Acrididae* family, and *Thomomys bottae* (Botta's pocket gopher). Unknown herbivores that were small enough to enter the fenced plots probably were in the same proportions as in the unfenced plots (22.7 %), adding to evidence that undetected herbivory events were by small invertebrate herbivores such as *Formicidae* family (Table 1-5).

Decision-Making Process

Camera Selection

One of the advantages of using motion sensitive cameras to track seedlings is the flexibility provided. In order to effectively utilize cameras to address unique research objectives, many decisions must be made based on specific research needs of individual studies. This study design was developed specifically using the Reconyx PC900 camera, but other cameras could be used. Considerations when selecting a camera other than the aforementioned include availability of the cameras, price/cost of using the cameras (this study used 28 cameras, which would have been a sizeable cost if they were not already available for use), durability and weather resistance, focal length and the ability to adjust focal length, field of view size, and type of trigger available (timed, motion, manual); [24].

Scale

Once a camera is selected, the user should determine the size of the plants to be studied. Studies of large plants can have a longer focal length and larger field of view (FOV), because the plants are more easily visible and a larger field of view may be necessary to capture images of larger plants. Conversely, small plants are more difficult to detect in images and the camera must be closer to the plants creating a narrower FOV. The appropriate focal length of the camera can be determined using these criteria (Figure 1-8).

Position

We recommend that primarily two factors should be used to determine the position that cameras should be placed: data to be collected and physical characteristics of the plant. If a study

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emphasizes vertical plant characteristics such as height or changes in height we recommend positioning the camera parallel to the ground allowing the camera focus to be perpendicular to the plant for adequate feature capture. Horizontal plant characteristics such as plant width, increases in foliage, and even biomass [50] can often also be determined with the camera parallel to the ground. If cameras are placed parallel to the ground, we recommend that they be elevated a minimum of approximately 10 centimeters off the ground and angled forward approximately 15 degrees. This helps prevent dust and debris buildup on the lens of the camera. If larger plants are being studied, the camera may be placed higher without an angle, since dust and debris will be less of a concern. One advantage of placing the camera parallel to the ground is that cameras will cast less shadow than a camera placed above the plants. If a study does not require height estimates, but requires cover or other similar estimates, we recommend placing the camera above the plants, perpendicular to the ground. Again, depending on the size of the plants, the camera height should be adjusted based on the size of the plants being studied (Figure 1-9). We also recommend that plant physical characteristics should be considered when determining camera position. *E. elymoides* seedlings are slender with much higher surface area visible from the side than from above, especially directly after seedling emergence. These characteristics of the plant make it important to place the camera parallel to the ground so that images capture the largest amount of surface area for easier identification. If the plant has more surface area visible from above and height measurements are not required, it may be better to place the camera above the plants (Figure 1-8).

Camera Settings

Perhaps the greatest flexibility in data collection can be acquired through the adjustment of camera settings. Temporal scale and type of data needed should be considered when selecting camera settings. With the cameras used in this study, time lapse images could be triggered as frequently as every 5 minutes or as infrequently as once a week. If research is concerned with frequent temporal changes, we recommend that the camera take frequent time lapse images. If temporal change is less frequent, adjust the camera accordingly. One consideration for the frequency of time lapse images is the amount of images that will be collected and require further processing. However, if there is doubt in the number of images needed to obtain a sufficient sample size, it is better to err on the side of more frequent images since large amounts of images can be culled if needed. If herbivory or other animal interactions with plants are of interest, then a motion trigger should be enabled on the camera to allow for observation of animal-plant interactions. Again, number of images per trigger should be determined based on the amount of detail required for analysis and the amount of images that will require processing after collection. If great amounts of detail for animal-plant interactions are needed, we recommend considering a camera with video capability (Figure 1-8).

CONCLUSIONS AND IMPLICATIONS

Remote sensing technology is a powerful tool for acquiring plant morphological and growth pattern data. Use of cameras in a near-ground setting offers the opportunity to collect details unavailable with higher-altitude sensors [41]. The ability to adjust camera position and settings allows for flexibility in creating the study design. Cameras can be set up and checked by one person in a few hours, and can collect data even when researchers are not present, which reduces the need to make frequent visits to the site. While cameras may require less fieldwork, the

amount of time required pre and post-collection can be substantial, and this trade-off should be considered before using cameras in research. A careful and thorough planning and decision-making process is imperative for effective data collection and post-collection processing.

This study focused on tracking one species (*E. elymoides*) in small, watered plots within the area of rangeland revegetation efforts. Future research should be conducted with cameras to determine if they can be successfully used to monitor multiple species in a rangeland reseeding efforts. Using cameras to monitor reseeding efforts may also require research on how camera measurements of height, density, and herbivory are affected in very low densities of seedlings, and the number of cameras required to achieve an acceptable statistical power with low densities of seedlings.

Though there are potential drawbacks to using remotely triggered cameras for research, creativity and thoughtfulness will allow cameras to be a powerful tool for researchers and land managers to study plants, especially seedlings. Potential areas of research or monitoring using remote cameras could include tracking seedling emergence, densities, demographics, and survival, among others. The ability to track specific causes of seedling death using direct photographic evidence could be useful for identifying causes of seedling death in restoration efforts. Using cameras for seedling monitoring and research during restoration will inform post-seeding management of rangeland restoration projects and possibly lead to more effective restoration efforts.

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TABLES

Table 1-1. Results of mixed model analysis for average seedling height.

Type 3 Tests of Fixed Effects for Avg Seedling Height				
Effect	Num DF	Den DF	F Value	Pr > F
Date	10	260	6.81	**<0.0001**
Row	3	81	0.98	0.4076
Treatment	1	26	34.77	**<0.0001**
Date*Treatment	10	260	12.66	**<0.0001**

Table 1-2. Results of mixed model analysis for seedling density.

Type 3 Tests of Fixed Effects for Seedling Density				
Effect	Num DF	Den DF	F Value	Pr > F
Date	10	260	8.02	**<0.0001**
Row	3	81	2.45	0.0698
Treatment	1	26	15.15	**0.0006**
Date*Row	30	813	2.38	**<0.0001**
Date*Treatment	10	260	2.24	**0.0160**

Table 1-3. Frequency and percent of herbivory events on seedlings separated by herbivore event types.

Herbivore/Type of Damage	Number of Events	Percent
Lepus californicus	338	36.15
Unknown Herbivory-Day	209	22.35
Thomomys bottae	161	17.22
Buried	65	6.95
Dipodomys sp.	63	6.74
Acrididae	52	5.56
Eremophila alpestris	24	2.57
Trampled	13	1.39
Urocitellus mollis	6	0.64
Unknown Herbivory-Night	2	0.21
Antilocapra americana	2	0.21
Grand Total	935	100.00

Table 1-4. Percent damage caused to seedlings separated by category. Herbivores damaged the largest proportion of seedlings (69 %).

Cause of Damage	Percent
Herbivores	69.09
Buried & Trampled	8.34
Unknown Herbivory-Day	22.36
Unknown Herbivory Night	0.21
Total	100.00

Table 1-5. Frequency of herbivory events on seedlings in fenced plots.

Fenced Herbivory	Number of Events	Percent
Unknown Herbivory	50	22.73
Acrididae	8	3.64
Eremophila. aplestris	1	0.45
Thomomys bottae	161	73.18
Grand Total	220	100.00

Figure 1-1. Photo of the plot setup with camera on left and seedlings in four rows marked by wooden dowels in the center. An unfenced plot is observed in the foreground, and a fenced plot in the background. This photograph was taken at the Murray's Mesa site located on the Utah Test and Training Range (UTTR), Utah.

Effect of Date on Picture Height Measurements
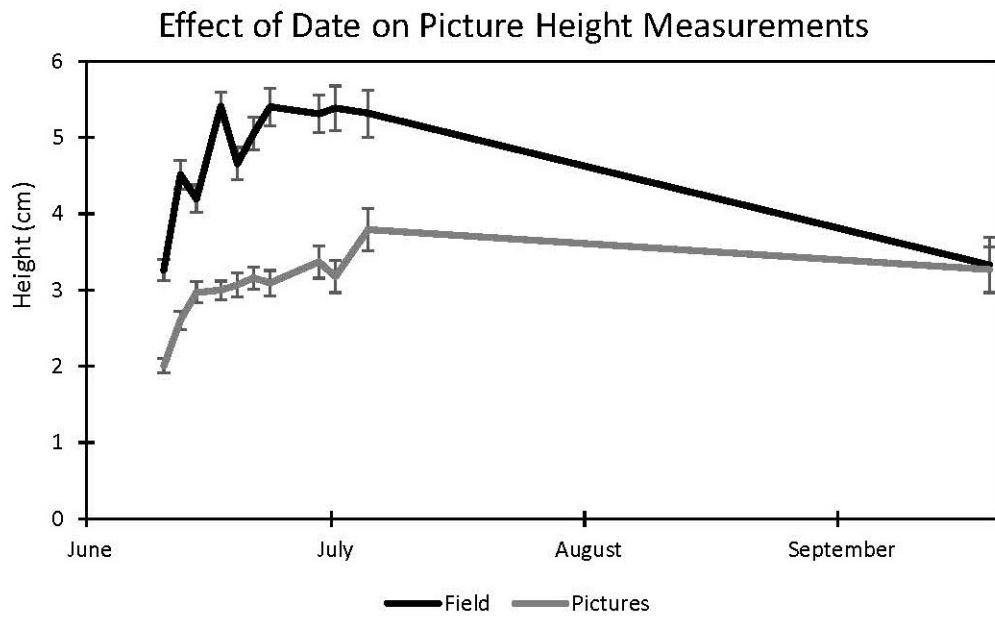
Figure 1-2. The difference (mean ± SE) in height measurements estimated from a image and measured in the field over the period of the study.

Figure 1-3. Interaction of date and fencing for height estimates in images vs. field (estimates ± SE). A) Shows the difference in height for fenced vs. unfenced plots. B) A comparison of image and field measurements for height in fenced plots, over time. C) A comparison of image and field measurements for height in unfenced plots, over time.

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Accuracy of Picture Height Estimates by Time of Day
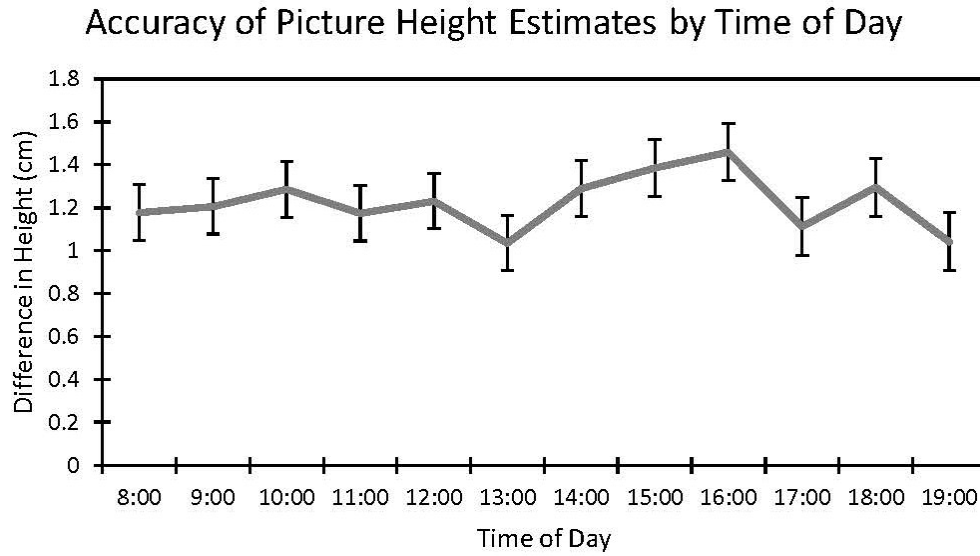
Figure 1-4. Effect of time of day on accuracy of image height estimates (estimate ± SE).

Effect of Date on Picture Density Estimates
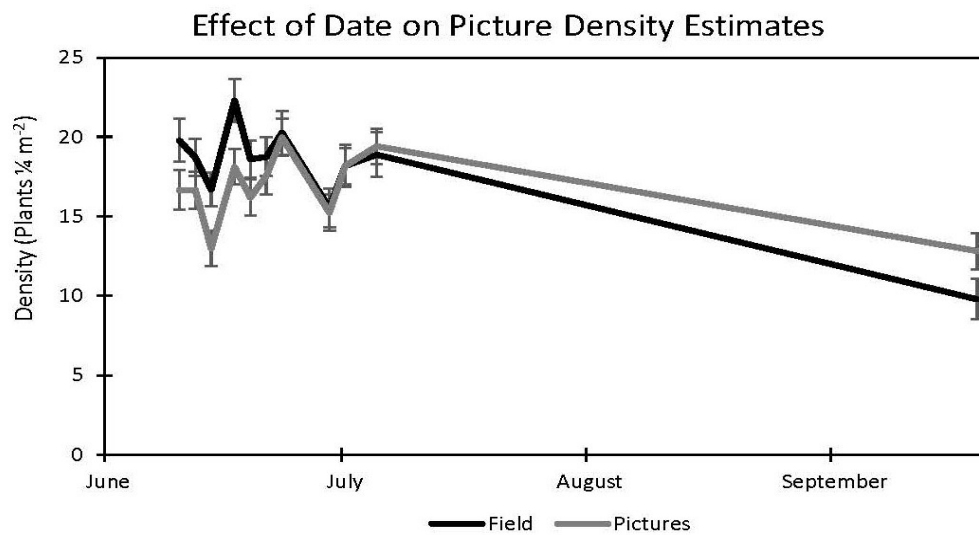
Figure 1-5. The difference (mean ± SE) in density measurements estimated from a image and measured in the field over the period of the study.
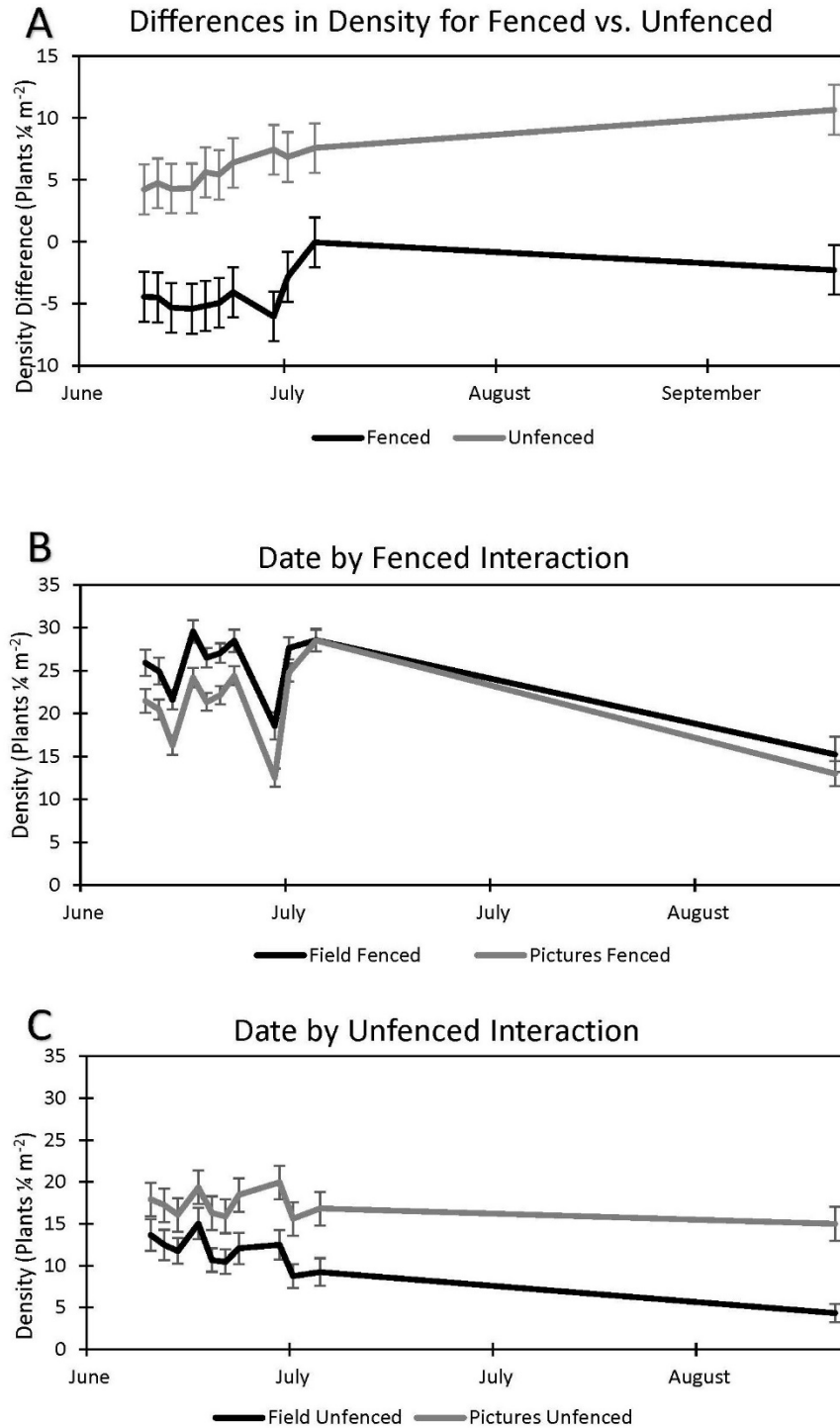
Figure 1-6. Interaction of date and fencing for density estimates in images vs. field (estimates ± SE). A) Shows the difference in density for fenced vs. unfenced plots. B) A comparison of image and field measurements for density in fenced plots, over time. C) A comparison of image and field measurements for density in unfenced plots, over time.
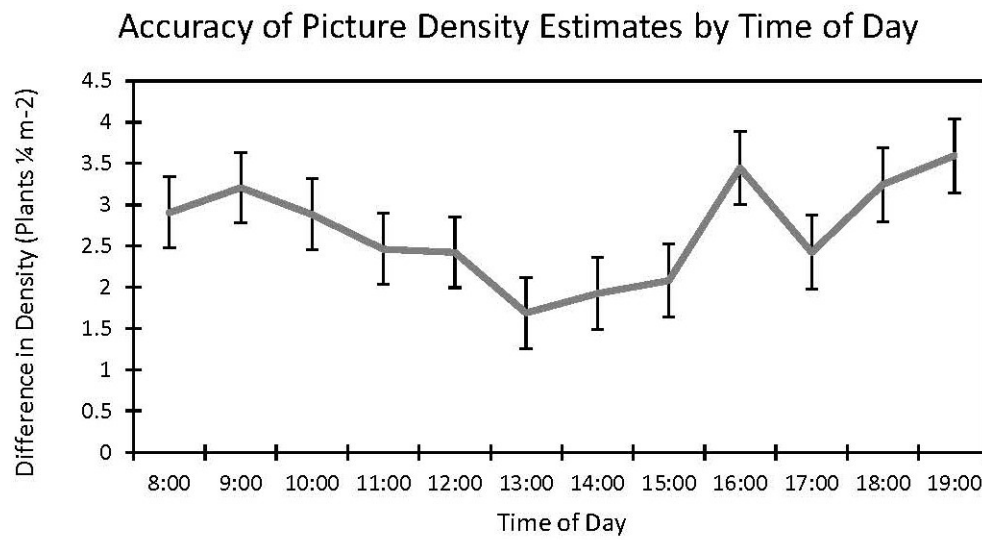
Figure 1-7. Effect of time of day on seedling density estimates in images vs. field (estimate ±SE).

Decision Flowchart

Step 1: Primary Focus of Study

Plants → Use time-lapse camera

Plants and Animals → Use camera with both time-lapse and motion triggers

Animals → Use motion-trigger camera

Step 2: Size/Scale of Study Subjects

Small (e.g. seedlings, burr buttercup, etc.) → Short focal length on camera (60 cm or less)

Large (e.g. mature plant, shrub, etc.) → Long focal length on camera (1m or more)

Step 3: Primary Features of Subjects to be Studied/Measured

Height, Frequency → Camera parallel to ground

Frequency, Density, Cover → Camera perpendicular to ground

Step 4: Plot Setup

Place camera in proper position. → Measure field of view and mark in plot. → Adjust camera focal length and distance from subjects as needed. → Re-measure and mark F.O.V. → Consider placing a ruler or other reference scale in the plot for accuracy and ease of measuring.

Step 5: Camera Settings

Timelapse → Determine frequency of data collection desired → Calculate amount of pictures to be processed → If too much → Determine a method to randomly sample from collected pictures; Adjust frequency of data collection

Motion → No animals → Don't use infrared motion sensing; Animals → Use infrared motion sensing

Step 6: Maintenance

Clean camera lenses and check battery periodically → If necessary, remove weeds or other obstructions from camera F.O.V.

Figure 1-8. Decision-making process flowchart, arranged by six steps for planning, camera setup, and camera maintenance.

CHAPTER 2

Biotic Causes of Seedling Mortality for the Native Species, *Elymus elymoides,*
(Bottlebrush Squirreltail) (Raf.) Swezey in a Drill-seeded Rangeland Environment

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ABSTRACT

Human activities have impacted rangelands and facilitated the colonization of invasive

annual grass and forb species worldwide. Generally, areas dominated by invasive annual species

fail to provide high quality habitat for wildlife and increase the frequency of wildfires by

producing abundant, continuous fuels that are dry earlier in the year compared to areas

dominated by native plants. Subsequently, efforts to restore degraded areas often fail.

Understanding processes involved in plant establishment can improve the ability to predict the

outcome of restoration practices and create effective solutions for rangeland restoration. The

purpose of this study was to identify biotic causes of plant mortality for species seeded during

rangeland revegetation. This study was conducted on the Utah Test and Training Range (UTTR)

in western Utah. This assessment includes tracking herbivory, seedling emergence from soil, and

timing and cause of seedling death. We placed cameras in 28 plots arranged in a randomized

split-plot design with fenced and unfenced plots and seeded with two rows of *Elymus elymoides*

(bottlebrush squirreltail) (Raf.) Swezey. We tracked individual seedlings and recorded their

status (alive, dead, grazed or damaged), comparing initial seedling establishment and seedling

survival between fenced and unfenced plots. Seed predators reduced initial seedling

establishment in unfenced plots by 4 times (p= 0.0002). Seedlings were 7 times more likely to

survive in fenced vs. unfenced plots. Of total seedling mortality, 73.6 % of seedling death was caused by herbivory from *Thomomys bottae* (Botta's pocket gopher), invertebrate herbivores, and *Lepus californicus* (black-tailed jackrabbit). Continued research should be conducted at larger scales to determine the effect of small herbivores on rangeland reseeding efforts. Strategies to mitigate the effect of herbivores should be considered to increase seeded plant establishment during restoration efforts.

INTRODUCTION

Rangelands cover approximately 50 % of the earth's landmass, consisting mostly of natural vegetation that is dominated by grasses, shrubs, and forbs (Institute, 1986). Rangelands provide the natural resources and ecological services that support a wide range of uses including wildlife habitat, livestock grazing, and watershed maintenance and sustainability (Institute, 1986). Exotic species that invade rangeland ecosystems are increasingly common worldwide, threatening biosphere integrity and ecosystem function (Duraiappah et al., 2005; Steffen et al., 2015). Invasive species can degrade plant community structure, decrease ecological resilience, and impair ecological processes that promote the ability for self-repair (Stringham et al., 2003). Additionally, invasive plants can alter fire regimes, watershed function, and plant and animal community diversity and health (Young & Evans, 1973; D'Antonio & Vitousek, 1992; Humphrey & Schupp, 2004). In addition to vegetative and watershed changes, invasive species can alter the animal community and food chain (Stringham et al., 2003; Freeman et al., 2014; Lucero et al., 2015). An altered animal community could impact native plant establishment leading to a positive feedback increasing the ability of exotic plants to invade (St. Clair et al., 2016).

To increase biotic integrity, enhance plant and animal community characteristics, improve ecosystem function, and reduce fire risk following disturbance, land managers often reseed rangelands with seed mixes that include desirable perennial plant species (Svejcar et al., 2017). These efforts to restore functional ecological characteristics are often expensive, costing land managers millions of dollars every year globally (Office, 2003; Hardegree et al., 2011; Merritt & Dixon, 2011). The success of reseeding has historically been low in arid and semi-arid environments, usually because of poor seed germination, limited establishment, or low survival (Lysne & Pellant, 2004; Hardegree et al., 2011; Sheley et al., 2011). Improving plant establishment from seeding efforts can lead to greater biodiversity, healthier ecosystem function, and extensive cost savings for restoration efforts (Hardegree et al., 2011; Merritt & Dixon, 2011).

Increasing plant establishment from seed requires a greater understanding of seedling establishment and survival, which have been described as "bottlenecks" to plant recruitment (Boyd & James, 2013; Hardegree et al., 2013; Bosco et al., 2015). Though the effects of failure to germinate or emerge from the soil are well documented (Belnap, 2003; Clark & Wilson, 2003; James & Svejcar, 2010; Bosco et al., 2015), the biotic causes of seedling stress and/or death in the first few months of life are not well documented. While lack of soil moisture is one cause of seedling stress/death, other potential causes include herbivory and seed predation (Bestelmeyer et al., 2007; Boyd & James, 2013; Bosco et al., 2015; Sharp Bowman et al., 2017). In both intact and disturbed, unrestored habitats, the effect of keystone guilds of small mammals such as heteromyid rodents have been identified as major drivers of plant establishment and succession (Brown & Heske, 1990; Kerley & Whitford, 2009; St. Clair et al., 2016; Bowman et

al., 2017). The effects of herbivores on seedlings during restoration efforts, however, are not well-studied.

Camera traps are one commonly used method of directly observing animal-animal and animal-plant interactions (Kays et al., 2009; Burton et al., 2015). Camera traps can be used to study numerous different life forms including large to small mammals, reptiles, amphibians, birds, and arthropods (McCallum, 2013; Burton et al., 2015; Welbourne et al., 2015). Camera traps are used for a wide variety of research objectives including estimating animal abundances and distributions (Karanth & Nichols, 1998; Cusack et al., 2015), documenting behaviors and interactions (Larsen et al., 2007; Hall et al., 2018), and tracking seed predation and herbivory of plants by animals (White et al., 2017). Cameras offer an opportunity to directly observe the effects of herbivores on seedlings in re-seeded rangeland areas.

The purpose of this study is to identify the biotic causes and timing of seedling stress and death, and quantify their potential effect on newly established seedlings in a tilled and drill-seeded rangeland revegetation site. Specifically, using camera traps we will identify herbivores that consume or damage seedlings and record the amount of seedling damage and death caused by each herbivore species. Understanding how herbivory and other stressors affect seedling establishment at revegetation sites will improve the ability of managers to account for and/or control seedling damage when attempting to establish desirable plant communities during restoration efforts.

MATERIALS AND METHODS

Study Site Description

Our study was conducted at two locations, Murray's Mesa (MM, 41.036394°N, -112.979465°W) and Arctic Road (AR, 41.078425°N, -112.927195°W), on the Utah Test and Training Range (UTTR) located in the West desert of Utah, United States. This military-managed land is in a relatively low precipitation area of the semi-arid Great Basin Region, receiving approximately 258 mm of precipitation annually (30 year norm; PRISM Climate Group, 2016). Murray's Mesa is located at 1399 m elevation with <4% slope. We determined through Brigham Young University's Environmental Analytical Lab (Provo, UT, USA) that the top 15 cm of soil contained 37.4% silt, 22.4 % clay and 40.2% sand with a pH of 7.8 and 1.3% organic matter. The Arctic Road site is located at 1338 m elevation with <4% slope and loam soil containing 47.4% silt, 26.4% clay, and 26.2% sand, a pH of 7.6, and 2.7% organic matter. Both sites consist of a degraded salt desert shrub community.

Military activity at these sites has contributed to increased fire frequency and the invasion of *Bromus tectorum* L. (cheatgrass) and a large number of invasive annual forbs such as *Halogeton glomeratus* (Bieb.) C.A. Mey (halogeton), *Salsola iberica* (Sennen and Pau) Botsch. (Russian thistle), and *Sisymbrium altissimum* L. (tumble mustard), leading to the need to restore degraded lands. Remaining desirable perennial plants at both sites include *Sarcobatus vermiculatus* (Hook.) Torr. (greasewood), *Atriplex confertifolia* (Torr. & Frem.) S. Watson (shadscale), *Artemisia spinescens* D.C. Eaton (bud sagebrush) and *Elymus elymoides* (Raf.) Swezey (bottlebrush squirreltail) and *Achnatherum hymenoides* (Roemer & J.A. Schultes) Barkworth. As with other low precipitation areas the establishment of seeded plant species has been marginal (Seabloom et al., 2003; Fay & Schultz, 2009; Robins et al., 2013). *E. elymoides* and other native

perennial grasses are commonly used species in seed mixes for restoration on the UTTR, but typically have low establishment rates.

Study Design

Our study was conducted with 7 replications at each site. The MM site was reseeded the year this study was conducted (2017) with limited plant establishment, and the AR site was reseeded the previous year (2016) with very little plant establishment. Each restoration site, covering approximately 162 hectares, was tilled at the time of restoration to a depth of approximately 20-30 cm to reduce *B. tectorum* recruitment from the seedbank, consistent with current management practices on the UTTR. Replications were organized by distance from the edge of the reseeded area in four blocks. Block one was 45 m from the edge of the reseeded area, block two was 70 m away, block three 95 m, and block four 120 m. Each replication was arranged in a randomized split-plot design with either an unfenced plot or a fenced plot that excluded mammalian herbivores (Figure 2-1). Fences were built using 1 m tall hardware cloth, with the bottom buried approximately 15 cm below the soil surface to prevent burrowing under the fence. At the top of the fence, 25 cm of metal flashing were attached to prevent small mammals from climbing over the fence. Fences were tall enough that resident large herbivores (i.e. *Antilocapra americana* (pronghorn)), would not reach over the fence to graze on seedlings. In each plot, we seeded *E. elymoides* in two rows on May 27 at a depth of 0.5 cm. Rows were placed 20 cm apart and each row contained 50 pure live seeds, for a total of 100 seeds per plot. This seeding design allowed optimal seedling image capture by cameras. Plots were watered to ensure seed germination and seedling emergence from the soil. Soil in plots was brought to field capacity (-33 kPa, 0.301 g of water 1g $soil^{-1}$) at planting, and then maintained at a minimum of 50 % of field capacity until at

least 50 % seedling emergence from the soil had been achieved in each plot. After plots reached 50 % emergence, watering was reduced to 2 times a week. Watering was terminated 5 weeks after planting. Soil moisture was tracked at each site at 1 cm and 10 cm depths using Decagon MPS-6 dielectric water potential sensors (Meter Group Inc., Pullman, WA) to ensure adequate soil moisture for germination and growth of young seedlings (Atwater et al. 2015).

Reconyx PC900 (Reconyx, Holmen, WI, USA) cameras were placed in each plot 10 cm above the soil surface and angled forward 15° and programmed to trigger with changes in infrared reflection. For each trigger, the cameras would capture three images, with a five second waiting period between each image and a 15 second waiting period between triggers. Cameras were also set to capture time lapse photographs daily to track any changes in seedlings which were missed by motion. We analyzed camera images to determine species of herbivores, as well as other animal species damaging seedlings at the sites. We sampled seedling density and seedling height per row to track seedling growth and survival. Individual seedlings were tracked on camera images over the course of the growing season, and the condition of each seedling was recorded daily (live vs. dead vs. grazed) to identify exact cause and timing of stress and/or death.

Analysis

We recorded the number of seedlings that emerged from the soil in each plot to determine total seedling emergence. These values were compared between fenced and unfenced plots to determine the influence of seed predation on seedling establishment. We used mixed model analysis using least squares means with $\alpha = 0.05$ to determine if fencing, distance from the edge of the reseeded area, site, or their interactions had an effect on seedling establishment. The probability of seedlings surviving in fenced versus unfenced plots was calculated using binary

logistic regression, calculating odds ratios for fenced vs. unfenced plots. The frequencies of herbivory events and the number of times each seedling was grazed by each type of herbivore was calculated. Herbivory was determined by tracking individual plants (Figure 2-2) and documenting when entire plants or parts of plants disappeared in images. If images showed an herbivore consuming part or all of the plant, or if the plant was missing all or part of its vegetation directly after an herbivore was foraging at the plant, it was classified as an herbivory event (grazed) for that animal. If no herbivore was detected when a plant was partially or wholly removed, and the removal occurred during the day, it was classified as invertebrate herbivory since this class of herbivores is mostly comprised of *Formicidae* family, *Acrididae* family, and *Coleoptera* order, which exhibit a diurnal activity pattern, and would not trigger the infrared motion sensors of our cameras. *Acrididae* did occasionally trigger the infrared motion sensor when in the camera's field of view, but were grouped with invertebrate herbivores since infrared detectors would not be triggered by every *Acrididae*. All herbivores were recorded in unfenced and fenced plots to determine all causes of seedling death, and percentages that each herbivore contributed to seedling fate. If a seedling was otherwise damaged by being buried or trampled, it was labeled as incidental damage. A seedling was considered dead if all above-ground tillers were removed down to the stem at ground level and it did not regrow tillers after grazing. Individual seedlings did not always die after being grazed, therefore the frequency of fatal and non-fatal herbivory events was calculated.

RESULTS

Seedling Emergence

Fenced plots had greater seedling emergence compared to unfenced plots ($P < 0.01$). Distance from the edge of the reseeded area, fencing, and the interactions of distance from edge*fencing and distance from edge*site exhibited a significant effect on seedling emergence ($P < 0.01$, 0.01, 0.01, 0.05, respectively; Table 2-1). The ls means of seedlings emerged in fenced and unfenced plots were 58.3 and 14.5 seedlings per plot, respectively, reflecting a four-fold increase in seedling emergence in fenced vs. unfenced plots ($P < 0.01$; Figure 2-3). Means for seedlings emerged, based on distance from the edge of the reseeded area (block), increased between block two (70 m) and block three (95 m) by a difference of 26.1 seedlings per plot ($P < 0.01$; Figure 2-4). Mean number of seedlings emerged per plot diverged between sites with increasing distance from the edge ($P < 0.05$; Figure 2-5). The difference between fenced and unfenced mean seedlings emerged decreased between block two (70 m) and block three (95 m) by 19.9 seedlings per plot ($P < 0.01$; Figure 2-6), leaving no difference between fenced and unfenced plots in block three (95 m from the edge).

Seedling Survival

Seedlings were 7 times more likely ($p<0.0001$) to survive in fenced plots than in unfenced plots (Wald's 95 % confidence interval= 5.3 to 9.2, Table 2-2). In unfenced plots, seedlings were often grazed multiple times, causing seedling stress but not always death (Figure 2-7). The maximum amount of times a seedling was grazed was five times, which only happened with two seedlings that both died. The maximum amount of times a seedling was grazed and survived was four times (Figure 2-8). Over half (61%) of seedlings in unfenced plots died. Seedling death was

concentrated in the first half of the summer, with 77.3 % of seedling death occurring within 60 days of planting, though a large amount also occurred from August 23 to September 9 (16.7 %). Overall, herbivory decreased over time, leading to decreased death rates in later months (Figures 2-9, 2-10).

Herbivory

Herbivory accounted for 89.2 % of seedling death in all plots. In unfenced plots, damage was caused to seedlings 876 separate times. Of those, 89.1 % was caused by herbivory, and 10.9 % was caused by incidental damages (e.g. trampled, buried). Most seedlings (38.6 %) were damaged by *L. californicus* grazing. Invertebrate herbivores (insects) were the second leading cause of plant damage (23.2 %). *T. bottae* caused 18.4 % of plant damage and *Dipodomys* sp. caused 8.8 % of damage (Table 2-3). Out of the 876 damage events, 44.2 % resulted in death of the seedling, for a total of 387 seedlings which died. Invertebrate herbivores caused the death of the most seedlings (32 %), followed by *T. bottae* (29.7 %), *L. californicus* (16.8 %) and *Dipodomys* sp. (9.6 %). Burial and trampling caused 10.9 % of seedling deaths. Other herbivores which caused seedling death included *E. alpestris*, and *A. americana* (Table 2-4). The only herbivore which grazed seedlings, but did not cause any seedling death was *Urocitellus mollis* (Piute ground squirrel). Substantial variation in herbivores present and cause of death was observed between sites (Table 2-5).

Camera images revealed that the AR site had frequent visitation by a number of mesocarnivores: *Canis latrans* (coyote), *Vulpes macrotis* (kit fox), and *Taxidea taxus* (badger), and MM had only a single visit by a *T. taxus*. Also, the MM plots had visits from *Dipodomys* sp., and *Peromyscus maniculatus* (deer mice) (Figure 2-11) while AR did not.

DISCUSSION

Initial Seedling Establishment

Overall, the drastic increase in seedling emergence from fenced plots (four times greater emergence, Figure 2-12) was consistent with results of Connolly (Connolly et al., 2014), who found that seed predators reduced emergence, establishment, and seed bank size of native species, and Sauzo (Suazo et al., 2013), who found rodent exclusion increased seedling emergence more than threefold. The initial seedling emergence in unfenced plots was most likely affected by different factors at the two different sites. Though both sites have similar plant communities, MM was visited by animals that consume seeds (*Dipodomys* sp., *P. maniculatus*). This difference between sites is likely because these rodents avoid foraging in areas with high risk of predation due to low shrub and perennial plant cover and abundant carnivores (Pearson, 1964; Newsome et al., 1989). Fenced plots at both sites had higher seedling establishment than unfenced plots; However, AR did not have the suite of seed predators experienced by MM. A probable explanation for the low unfenced seedling emergence at AR was the high level of incidental damage in these plots, which accounted for 27.2 % of seedling deaths. *A. americana*, *T. taxus*, and *V. macrotis* disturbed plots by laying and digging, most likely due to high soil moisture and/or standing water from watering treatments. Without watering, these plots may not have attracted as much soil-disturbing activity and had higher seedling emergence. There was substantial evidence at MM of seed predation limiting seedling establishment, detected by plot visits by seed predators. At 45 m and 70 m from the edge of the reseeded area plots exhibited lower unfenced seedling emergence, and these blocks were also frequently visited by seed predators before the seedlings emerged from the soil. At 95 m and 100 m there was no difference in seedling establishment between fenced and unfenced plots and fewer visits from seed

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predators (Figure 2-13). This pattern agrees with seed predation studies which have demonstrated that seed predators can remove up to 85 % of seeds (Hulme, 1998; Edwards & Crawley, 1999). An examination of the interaction between site and distance from the edge reveals that the site measurements diverged between 95 m and 120 m, though there was no readily available explanation for this pattern (Figure 2-8).

Seedling Survival

Similar to initial establishment, seedling survival was higher in fenced plots ($P < 0.01$). The higher survival in fenced plots resulted from the protection from herbivory and incidental damage. Studies in unrestored burned and unburned areas observed a similar pattern of seedling survival in fenced plots (St. Clair et al., 2016; Sharp Bowman et al., 2017). The concentration of seedling death in the first 60 days after planting is most likely because seedlings were younger and more fragile at that time, and became more resilient over time. Since many seedlings were grazed or damaged multiple times, it is possible that the proximate cause of death (e.g. being eaten by a *Dipodomys* sp.) was not the ultimate cause. Rather, cumulative stress from multiple damage events contributed to higher seedling death than a single damage event. When damage did occur, 65 % of seedlings survived after being grazed two or more times, and 5.7 % survived after being grazed four times (Figure 2-11). In reseeded areas where seedlings are not watered, seedlings may be less resilient, decreasing the survival rates of seedlings compared to this study. The response of seedlings to grazing has been documented (Briske, 1996), but the level of resilience in seedlings is less known. One study tracked overall seedling survival after cattle grazing, but not individual seedling responses or survival (Salihi & Norton, 1987).

Herbivory

Invertebrates and small mammals contributed most to the death of seedlings. *L. californicus* grazed seedlings the most, but they were less lethal to seedlings than *T. bottae* or invertebrate herbivores. This is most likely because *L. californicus* did not remove all aboveground tissue when grazing like invertebrates and rodents did, leaving plant material that could photosynthesize and recover (Crawley, 1990). The total proportion of seedlings grazed by herbivores did not necessarily reflect how lethal these herbivores are on plants; however, in un-watered reseeding efforts small mammals and invertebrate herbivores may contribute even more to seedling death. Overall, *T. bottae*, invertebrate herbivores, and *L. californicus* reduced seedling survival the most, as the proximate cause of 73.6 % of all seedling death. We expected *A. americana* to graze seedlings more since they were frequently observed in unfenced plots, but they grazed < 0.5 % of seedlings. This is most likely because as large herbivores, they target larger mature plants to fill their forage biomass needs (Belovsky, 1997).

CONCLUSIONS AND IMPLICATIONS

Plant establishment during restoration efforts often faces the same challenges in unrestored areas, namely granivory and folivory (Heske et al., 1993; Hulme, 1998; Sharp Bowman et al., 2017). The animal communities found within or proximate to restoration sites have a large impact on the establishment of *E. elymoides* in drill-seeded areas. Survival of seeded plants is reduced both as seed and seedlings, similar to the effects of herbivores in unrestored habitats (Heske et al., 1993; Hulme, 1998; Bowman et al., 2017). Seed predators reduce the initial establishment of new seedlings. The effect of seed predators may be even more significant when seed is broadcast on the soil surface, where it is readily available and easy to detect to birds, ants,

and rodents compared to seed buried during drill-seeding (MacMahon et al., 2000). Once seedlings emerge from the soil, they are subject to intense pressure from folivory. Stress caused by being grazed multiple times may be the ultimate factor leading to seedling death, though the seedlings are remarkably resilient to above-ground tissue removal. While large herbivores probably impact mature plants, they do not appear to pose a significant threat for initial establishment and growth of seedlings in this setting. Small mammals and invertebrates, however, were important biotic factors limiting seedling survival in this study.

Seedlings in this study were watered to ensure sufficient germination and growth to provide results. This study may have amplified biotic effects by drawing herbivores to the plots with higher moisture and more seedlings, or reduced biotic effects by providing a high density of seedlings so that herbivores were flooded with plants and were satiated, reducing the overall percentage of seedlings consumed according to predator satiation theory (Williams et al., 1993). It does however illustrate the potential effects herbivores can have in a restoration setting, and further research should be conducted at a larger scale and without watering to determine exactly how small mammal communities affect seedling establishment during reseeding efforts.

When restoration efforts are being planned, managers should carefully consider the herbivore community that occurs in the area. For example, years with lower small mammal abundance, or sites that have a healthy carnivore population may reduce the effects of small mammals, providing an opportunity to reseed with less granivory and herbivory pressure from the small herbivore population. Even slight reductions in the density of herbivores may increase seedling survival for *E. elymoides* and possibly other similar plant species since the seedlings can be resilient to grazing. This study only tracked seedling survival for *E. elymoides*, so further

research could illustrate the effects of herbivory on other plant species commonly used in restoration efforts.

ACKNOWLEDGEMENTS

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Table 2-1. Model for initial seedling establishment. Treatment, Block, Site*Block, and Block*Treatment were significant in the model.

Source	DF	Sum of Squares	F Ratio	Prob > F
Treatment	1	3828.1250	23.0762	**0.0002**
Block	3	2978.1786	5.9842	**0.0062**
Site	1	15.1250	0.0912	0.7666
Site*Block	3	1882.6786	3.7830	**0.0317**
Block*Treatment	3	2785.1786	5.5964	**0.0081**

Table 2-2. Percentages of living and dead seedlings in fenced and unfenced plots.

	Fenced	Percent	Unfenced	Percent	Total	Percent
Alive	599	81.50	158	38.63	757	66.17
Dead	136	18.50	251	61.37	387	33.83
Total	735	100.00	409	100.00	1144	100.00

Table 2-3. Frequency of damage to seedlings in plots by species/category, organized from highest to lowest.

Cause of Damage	Frequency	Percent
Lepus californicus	338	38.58
Invertebrate Herbivores	203	23.17
Thomomys bottae	161	18.38
Incidental Damage	78	8.90
Dipodomys sp.	63	7.19
Eremophila alpestris	23	2.63
Urocitellus mollis	6	0.68
Unknown	2	0.23
Antilocapra americana	2	0.23
Grand Total	876	100.00

Table 2-4. Cause of death of seedlings in unfenced plots, organized from highest to lowest.

Cause of Death	Frequency	Percent
Invertebrate Herbivores	124	32.04
Thomomys bottae	115	29.72
Lepus californicus	65	16.80
Incidental Damage	42	10.85
Dipodomys sp.	37	9.56
Eremophila alpestris	3	0.78
Antilocapra americana	1	0.26
Grand Total	387	100.00

Table 2-5. Cause of death of seedlings by site in unfenced plots.

	Site				
Cause of Death	AR	Percent	MM	Percent	Grand Total
Thomomys bottae	0	0.00	115	47.92	115
Invertebrate Herbivores	99	67.35	25	10.42	124
Lepus californicas	5	3.40	60	25.00	65
Incidental Damage	40	27.21	2	0.83	42
Dipodomys sp.	0	0.00	37	15.42	37
Eremophila alpestris	3	2.04	0	0.00	3
Antilocapra americana	0	0.00	1	0.42	1
Grand Total	147	37.98	240	62.02	387

Figure 2-1. Image showing randomized, split-plot design in two replications, each with a fenced plot, an un-fenced plot, and a camera in each plot.

Figure 2-2. Image showing individual seedlings being tracked in time-lapse photograph. Numbered markers were placed on the screen next to each seedling for tracking herbivory events, regrowth, and survival over time.
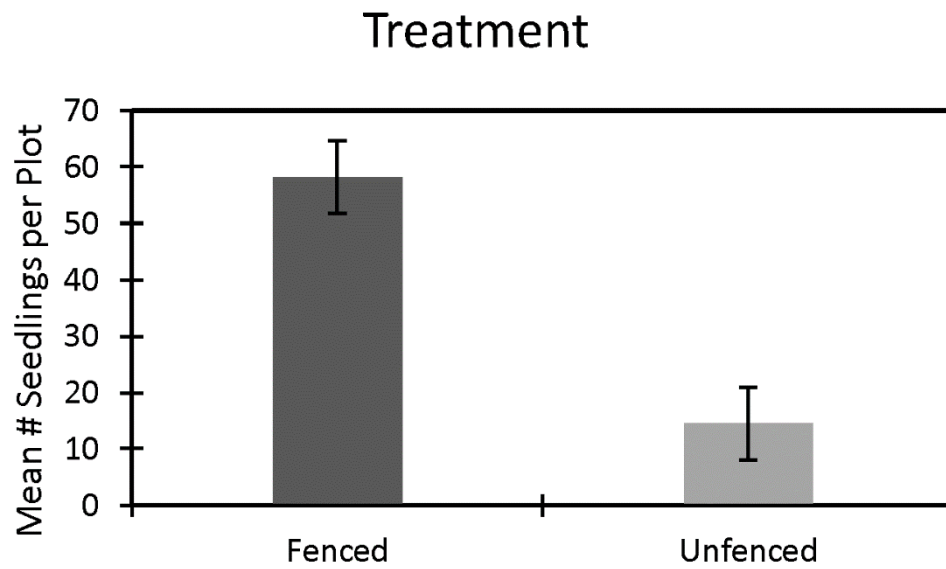
Figure 2-3. Difference between mean number of seedlings in fenced and unfenced plots (estimate ± SE). Mean number of seedlings per plot was estimated after all seedling emergence from the soil had occurred.
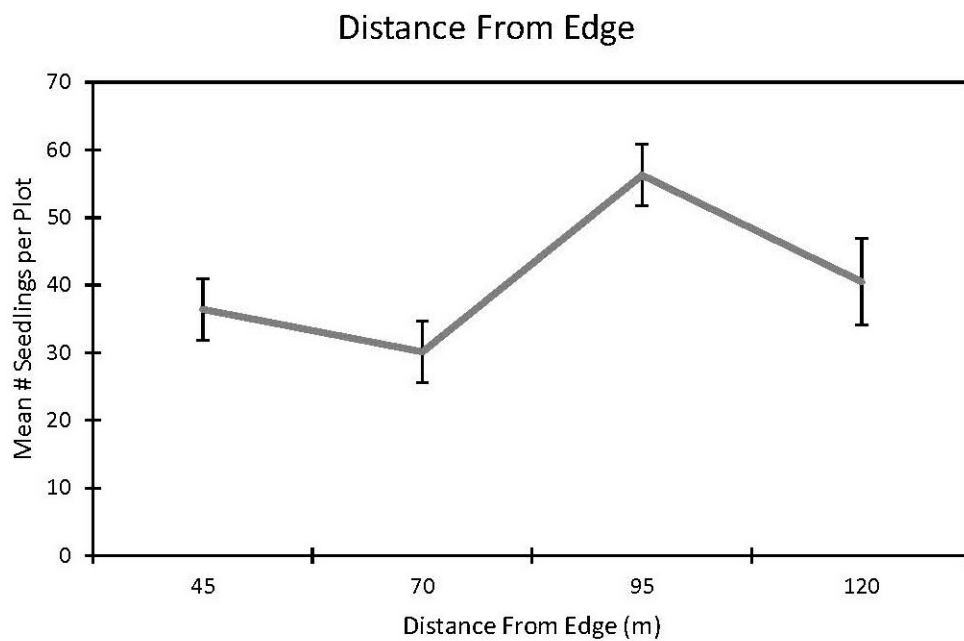
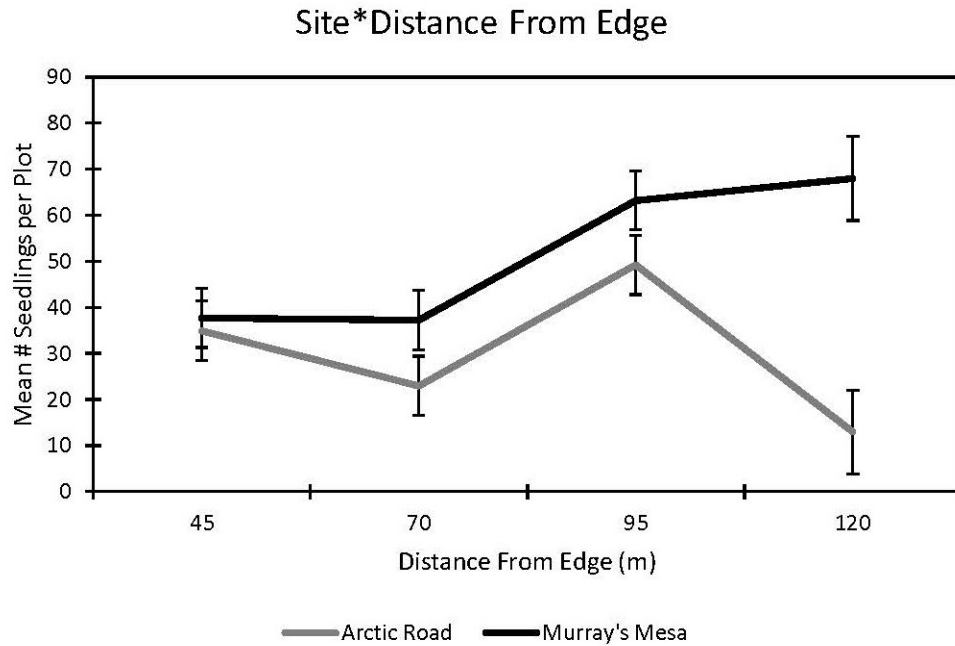Figure 2-4. Mean number of seedlings per plot at each distance from the edge of the reseeded area (estimate ± SE).

Site*Distance From Edge

Figure 2-5. Interaction of site and distance from the edge of the reseeded area for the mean number of seedlings per plot (estimate ± SE). Mean number of seedlings per plot was estimated after all seedling emergence from the soil had occurred.
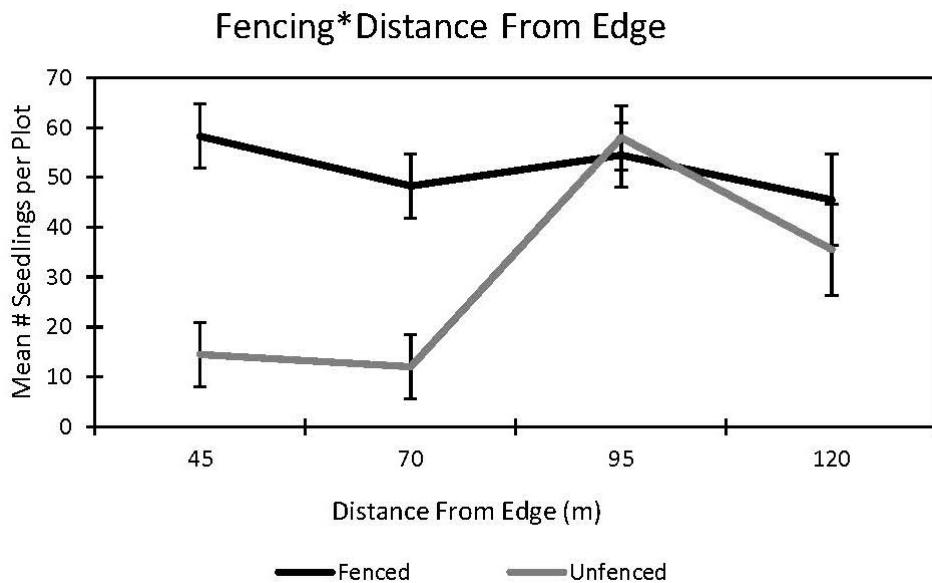

Fencing*Distance From Edge

Figure 2-6. Interaction of treatment and block for the mean number of seedlings per plot (estimate ± SE). At block three, there was no difference between fenced and unfenced plots. Mean number of seedlings per plot was estimated after all seedling emergence from the soil had occurred.
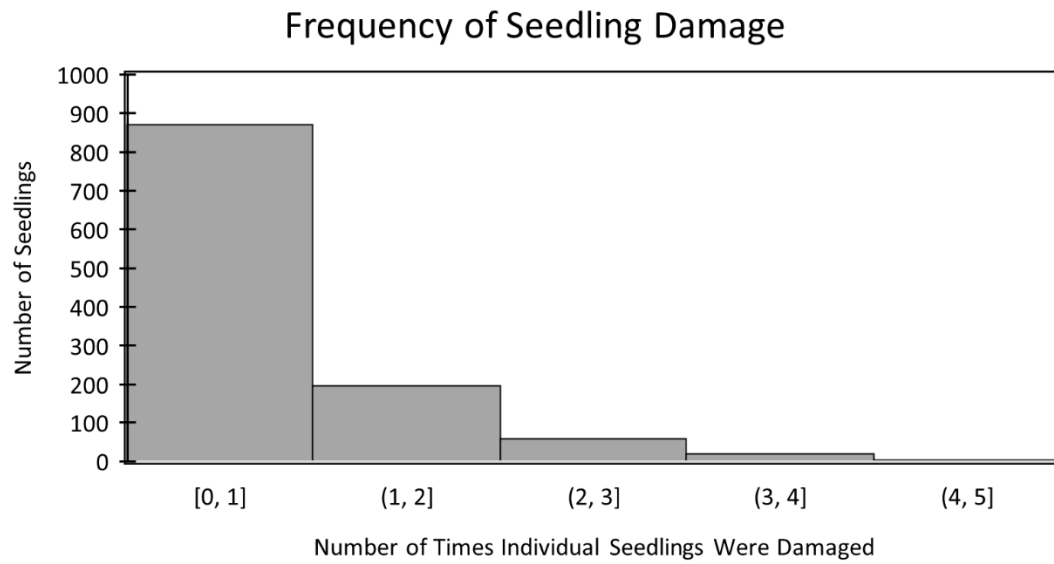
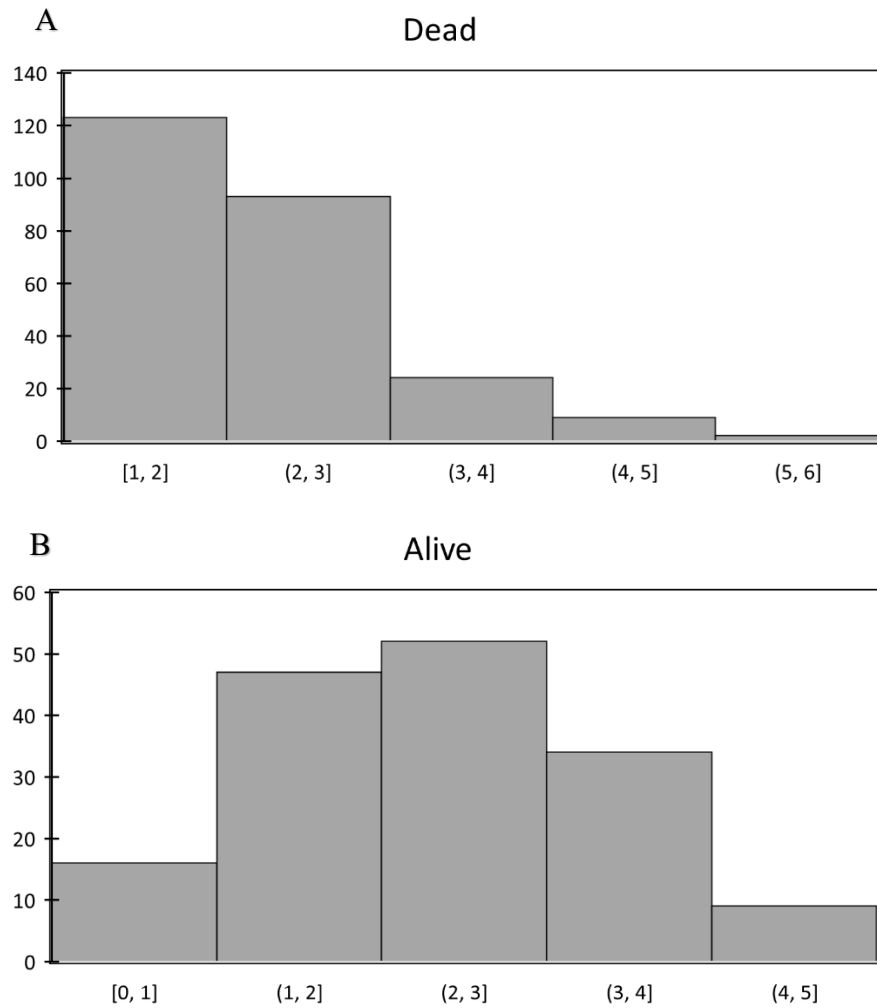Figure 2-7. Histogram showing distribution of the number of times seedlings were damaged.

Figure 2-8. Histograms showing the distribution of number of times seedlings were damaged for both A) seedlings that died and B) seedlings that survived through the summer in unfenced plots.
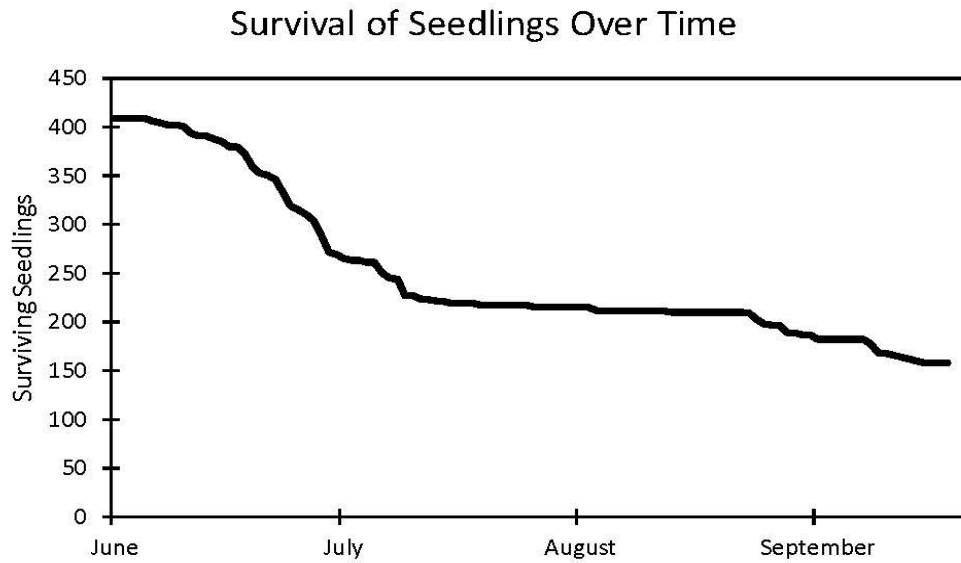
Survival of Seedlings Over Time

Figure 2-9. Survival of seedlings from May 27 to September 18, 2017.

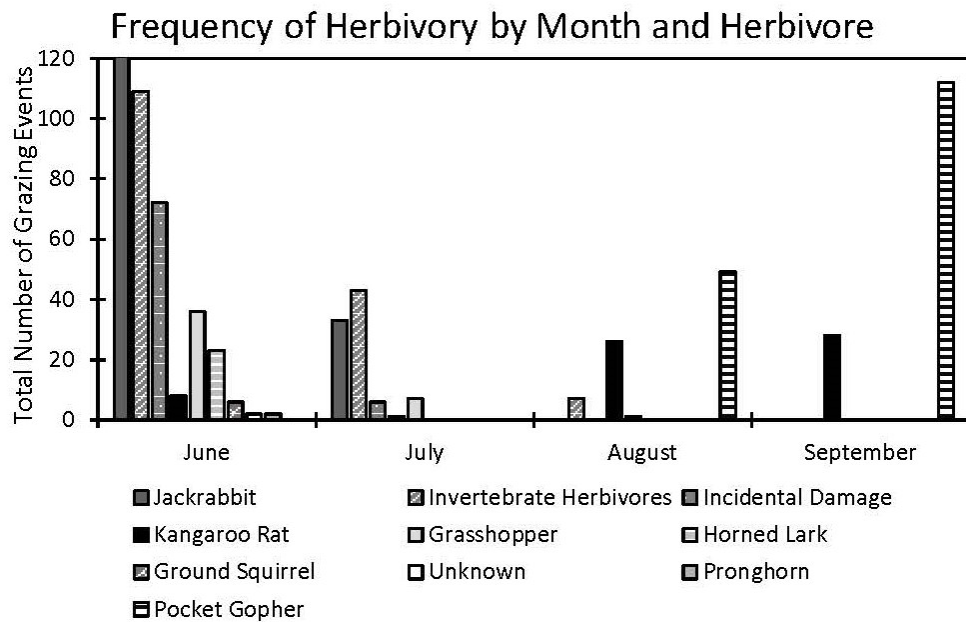## Frequency of Herbivory by Month and Herbivore

Figure 2-10. Frequency of herbivory events by month and by herbivore all plots. May was not included since planting was May 27, and no seedlings had emerged from the soil before June.

Figure 2-11. *P. maniculatus* (left) and *Dipodomys* sp. (right) observed at the Murray's Mesa site before seedlings had emerged from the soil.

Figure 2-12. Image showing low seedling survival in unfenced plots (left) compared to fenced plots (right). Notice evidence of small mammals digging in unfenced plots.