Investigating the Seed Bank of a Wetland Plant Community in a Long-Term Mowing and Fertilization Experiment

Peyton S. Harrelson

DEPARTMENT OF BIOLOLGY, EAST CAROLINA UNIVERISTY

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR UNIVERSITY HONORS

2023

Abstract

The seed bank of a plant community—the store of viable, dormant seeds below the soil surface—holds a legacy of its past composition and may have potential for restoration following habitat degradation. An ongoing 20-year fertilization and mowing experiment studies the plant community of a nutrient poor wetland in the coastal plain of North Carolina. The experiment is replicated on eight blocks and features a hydrology gradient that is caused by a drainage ditch. Since the start of the experiment, unmowed plots have become less diverse and are dominated by trees and shrubs. This long-term experiment provided the opportunity to explore the persistence of the seed bank through time. Soil samples from each treatment plot were collected and placed in a growth room to allow germination. The number of seedlings that emerged was recorded to test for the effect of fertilizer, mowing, and drainage on species richness and abundance of the seed bank and compare its composition to that of current and past aboveground vegetation. Multivariate statistics were used to look for treatment effects on the composition of the seed bank community. Fertilizer and drainage did not have a significant effect on the density or species richness of the seed bank, but it had a strong effect on its composition. In contrast, mowing had a significant positive effect on species richness and seed density but did not affect the composition of the seed bank. In comparison between the seed bank and the current aboveground vegetation composition, the mowed plots showed greater similarity, suggesting that the seed bank retains the species of the past plant community. As the aboveground vegetation changes, the seed bank may hold the ability to serve in future restoration projects.

INVESTIGATING THE SEED BANK OF A WETLAND PLANT COMMUNITY IN A LONG-TERM MOWING AND FERTILIZATION EXPERIMENT

Introduction

A seed bank is the collection of every viable seed in an area above and below the soil surface (Cavers 1994). Seed banks are further classified by the length of viability of the seeds once they are produced. Thompson and Grime (1979) make this distinction by designating seeds that are viable for less than a year as part of the transient seed bank, and those that are viable for more than a year as part of the persistent seed bank. Seeds are considered dormant until germination. Dormancy can be described as a state of “metabolic inactivity” that offers protection from unfavorable conditions (Lennon et al. 2021). For a seed in the bank to leave dormancy a variety of conditions such as light availability or grazing activity have inconsistent consequences on different species. This contributes to the vastly different seed banks based on length of viability as well as their patterns of dormancy (Cavers 1994).

There have been many studies regarding aboveground vegetation, but very few in comparison investigating what is occurring in underground seed banks. The plant community aboveground plays a role creating and maintaining the seed bank. It is important to take into account factors that shape the plant community, especially since these factors have lesser-known effects on the seed bank. Two factors that affect seed banks and shapes aboveground plant communities is disturbance and nutrient availability.

Above ground, disturbance of a forest provides the ability for light to reach the understory, allowing a diverse community of herbaceous species to thrive on the forest floor. When a forest is first burned, secondary succession begins. The first species to take over the landscape are typically herbaceous species, and woody trees come later during succession. The disturbance of fire leaves a bare landscape that is then repopulated by the seed bank that is left behind, determining the species that will be found in the aboveground vegetation (Carri et al. 2022). Changes in the aboveground vegetation due to disturbance are not immediately reflected in the seed bank (He et al. 2021). It is also hypothesized that after a disturbance, the soil seed bank can be the primary source of regeneration, particularly in grasslands (Bernard and Blaise 2004). After the disturbance of a fire, herbaceous and woody plants have many mechanisms to return to aboveground vegetation, including their seed bank (Carri et al. 2002).

Nutrient availability is another factor that affects the composition and diversity of plant communities. The amount and composition of soil nutrients affects what plants can grow there. Nitrogen deposition, resulting from combustion process and agricultural practices, generally leads to a decrease in biodiversity due to the resulting heightened fertilization (Field et al. 2014). As a result of this nutrient addition, the altered plant community can in turn lead to changes in the composition of the seed bank (Eskelinen et al. 2021). Fertilization tends to cause minor changes on the persistent seed bank composition, but fertilization causes more major changes in the aboveground composition (Zhang et al. 2019). The fertilization that results from anthropogenic addition of nitrogen can have even greater effects on plant communities that are nutrient-poor, as species that are nitrogen-tolerant with greater competitive ability can exclude specialists of low nutrient environments (Plassmann et al. 2008).

A long-term fertilization and mowing study provided us an opportunity to investigate the persistence of seed bank through time and the effects of nutrient enrichment on seed bank composition. This ongoing 20-year fertilization and mowing experiment studies the plant community of a nutrient poor wetland in the coastal plain of North Carolina, found on East

Carolina University's West Research Campus (Goodwillie et al. 2020). The study site was historically maintained as an open landscape because of disturbance by wildfires. In more recent years prior to the experiment, the site was burned or mowed periodically to prevent encroachment of trees. Since the start of the experiment, unmowed plots have become less diverse and are dominated by trees and shrubs. Since the start of the long-term experiment, we have seen that fertilization has caused a decline in species richness in the aboveground vegetation (Goodwillie et al. 2020). The findings of this long-term experiment have led us to investigate three research questions through a study of its seed bank:

1. How do fertilization, disturbance, and drainage affect the diversity of the seed bank?
2. How do fertilization, disturbance, and drainage affect the composition of the seed bank?
3. How has the seed bank changed as the aboveground community has undergone succession?

Here I report the results of a growth room experiment to study the seed bank of a long-term fertilization and mowing experiment. The seed bank was characterized by utilizing a common approach known as the seed emergence method (Ter Heerdt et al. 1996), in which the number and identity of seeds in the seed bank is estimated by observing the seedlings that emerge. The effects of fertilization, disturbance and drainage were analyzed for the seed bank, then this data was then used to compare the current seed bank to the past and current aboveground vegetation.

Methods

Study Site

The long-term experiment that is the focus of this study is located at the West Research Campus of East Carolina University in Greenville, North Carolina. The West Research Campus lies on a poorly drained and nutrient poor wetland in the coastal plain. The 0.7-ha experimental site was mowed and burned in 2003 at the start of the long-term study. Four treatments are replicated on eight 20 x 30 m blocks and applied randomly to plots within the block. Mowing and fertilization are applied in a two x two factorial design to yield four treatments: no mowing and no fertilization, fertilization, mowing, and mowing and fertilization. The mowing is applied once each early winter by rotary mower and then the mowed litter is raked off to better simulate the effects of vegetation removed by a wildfire. Fertilizer (10-10-10 NPK, 45.4 kg/ha of each nutrient per year) is applied to the plots three times a year. Located along one side of the plots is a ditch that drains blocks nearby, creating a soil moisture gradient. The vegetation is sampled every year at three permanent randomly-placed 1 m^2 quadrats per treatment plot by undergraduates in a course-based research program at East Carolina University.

Seed Bank Study

Soil was collected on January 14th, 2022 at a 15 cm depth from each of the 32 plots (8 blocks x 4 treatments) using a soil auger. Samples were taken from three haphazardly located sites within each plot, avoiding the edges adjacent to other plots, and then pooled. The auger was wiped clean between each plot to ensure there was no contamination of seeds between plots. The pooled soil samples were then placed in sealed Ziploc bags and refrigerated for four days until potting.

Soils of each plot were placed in six replicate pots. Each pot received 300 mL of unfertilized potting soil with 300 mL of the collected soil sample layered on top. Equipment was rinsed between soil samples to avoid contamination of seeds. The pots were randomly arranged with trays in a growth room that included both artificial and natural lights. Throughout the experiment, trays were rotated around the growth room three times a week to minimize effects of variation in light conditions. The seed bank was quantified using the seedling emergence method, where the germination and growth of seedlings is used to estimate the composition of the seed bank (Ter Heerdt et al. 1996). Once seedlings emerged, they were identified when possible, recorded and manually removed from pots. After 7 weeks the soil surface was disturbed to allow more seeds to be brought closer to the soil surface for germination. All seedlings were identified and removed by April 19th, 2022.

Analysis Methods

Seed Bank Analysis

To calculate seedling abundance the total number of seedlings that emerged was recorded for each pot. Species richness—the number of different species emerging—was also calculated for each pot. A three-factor analysis of variance was used to test the effect of drainage, fertilization and mowing on the species richness and abundance of seedlings.

Non-metric multidimensional scaling (NMDS) analysis was used to provide a graphical representation of patterns in community composition. This approach reduces the 42 species into a two-dimensional plane where each symbol represents one of the treatment plots, producing an ordination where the symbol proximity is based on their similarity in species composition. Two symbols that are further away from one another represent two treatment plots that are more different in composition.

Seed Bank and Aboveground Vegetation Comparison

The similarity of the seed bank and the aboveground vegetation compositions were compared using the Sorensen's index of similarity (Sorensen 1948). Sorensen's index of similarity is $= 2w/(A + B)$, where w is the number of species shared by the seed bank and the aboveground vegetation, A is the number of species making up the aboveground vegetation, and B is the number of species making up the seed bank. The data from the long-term experiment from the years 2004 and 2021 were used for this analysis. The Sorensen's index of similarity was found for each plot using the species composition of the aboveground vegetation from the year 2004 and comparing it to the seed bank composition. The Sorensen's index of similarity was also found for each plot using the species composition of the aboveground vegetation from the year 2021 and comparing it to the seed bank composition. This was done to give the overall similarity of the composition between the seed bank and the aboveground vegetation to see if the treatments are reflected. To determine whether the treatments affected the similarity of seed bank and aboveground communities, analysis of variance was used, with fertilizer, mowing and drainage as independent variables and similarity index as the dependent variable.

A bivariate correlation test was also performed to test for the relationship between the abundance and species richness of seedlings in each pot to determine if the two were tightly related.

Results

The first seedling germinated on January 24th, 2022. Across all pots, 42 species in 17 families were found in the germinable seed bank (see Appendix I). The most common families according to the number of seedlings that germinated included Poaceae, Cyperacae, Campanulaceae, and Asteraceae. The two families with the highest diversity present included

Asteraceae and Poaceae, with 11 and 10 species respectively. Of the 42 species, 12 were not identified to species. The unidentified seedlings included four species of sedges, one rush, five grasses, one forb, and one species of moss. A total of 1362 seedlings emerged from the seed bank. Of the total number of seedlings that arose 46% of them emerged from the mowed and fertilized plots, and this number increases to 76% if the unmowed, fertilized plots are added. Most species found were perennials.

Mowing and fertilization had a significant ($p < 0.001$, $F_{1,7} = 22.171$; $p < 0.001$, $F_{1,7}=104.818$) positive effect on species richness, while drainage had no effect ($p < 0.245$; $F_{1,7} = 1.360$). There was a significant interaction where mowing increased the effect of fertilization ($p < 0.001$; $F_{1,7} = 14.125$). There was also a significant interaction were drainage decreased the effects of fertilization ($p < 0.001$; $F_{1,7} = 23.829$). The results were similar for abundance, and the two dependent variables were found to be highly correlated ($p < 0.001$), so only species richness is reported. The bivariate correlation for abundance and richness had a Pearson correlation of 0.770, which is a significant correlation that shows a positive linear relationship between the two variables.

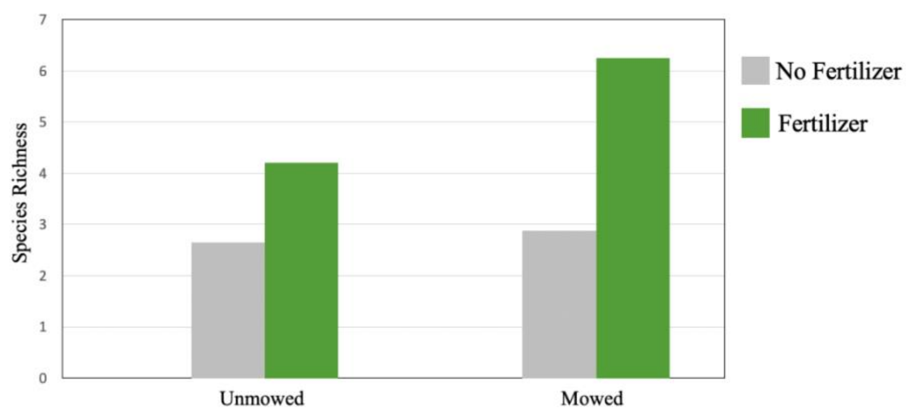

Figure 1 Effects of mowing and fertilization on seed bank diversity. Species richness is the average number of species emerging from the seed bank in replicate pots of a given treatment.

In the NMDS analysis of the seed bank data, the plots that were drained versus the plots that were not drained were separated along the x-axis. The plots that were fertilized versus those that were unfertilized were separated along the y-axis. There was no separation produced from the treatment of mowing. The NMDS of the seed bank suggests that the only factor that did not appear to affect the composition of the plots was mowing.

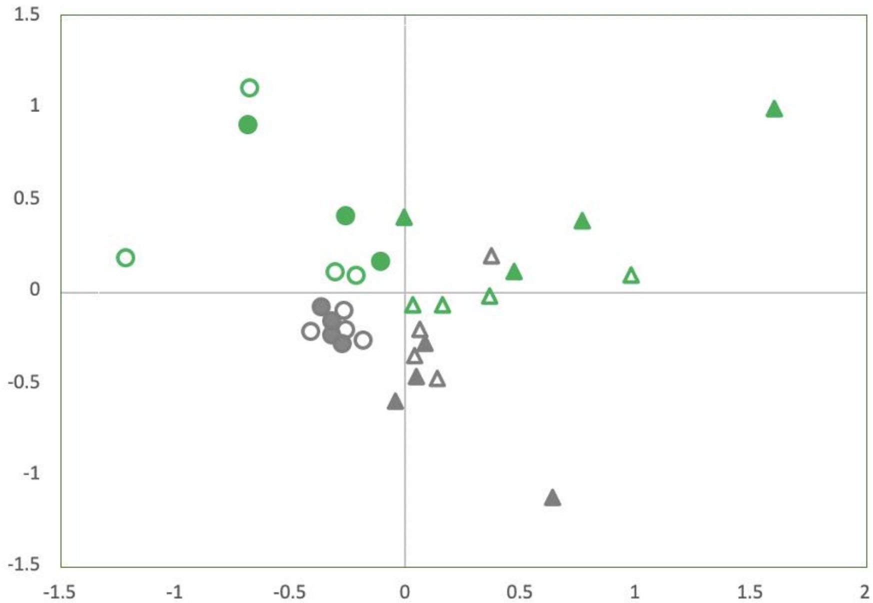

Figure 2 Non-metric multidimensional scaling plot of seed bank data. Each data point represents the summed species composition of the six replicate pots from each of the 32 treatment plots. Green symbols are fertilized plots while gray symbols are unfertilized plots. Circles represent wet plots, and triangles dry plots. Symbols that are filled represent mowed plots, and those that are not filled represent unmowed plots.

When the seed bank was compared to the 2004 aboveground vegetation using the Sorensen similarity index, no significant effects of drainage, fertilizer, or mowing were seen on similarity values in the results of analysis of variance. When the seed bank was compared to the 2021 aboveground vegetation using the Sorensen similarity index, mowing had a highly significant positive effect, increasing similarity. The mowing and fertilization interaction was also significant, with mowed plots increasing the effect of the fertilization. The seed bank was found to be the least similar to the 2021 aboveground vegetation in the unmowed plots. The aboveground vegetation in unmowed plots has changed dramatically since the start of the experiment. This suggests that the seed bank below ground has not changed as quickly as the aboveground plant community and has retained the landscape of the past. The mowed plots currently resemble an herbaceous landscape, which is similar to the herbaceous landscape that is produced by the seed bank; however, the aboveground forest is least similar to the herbaceous landscape that is sustained by the seed bank.

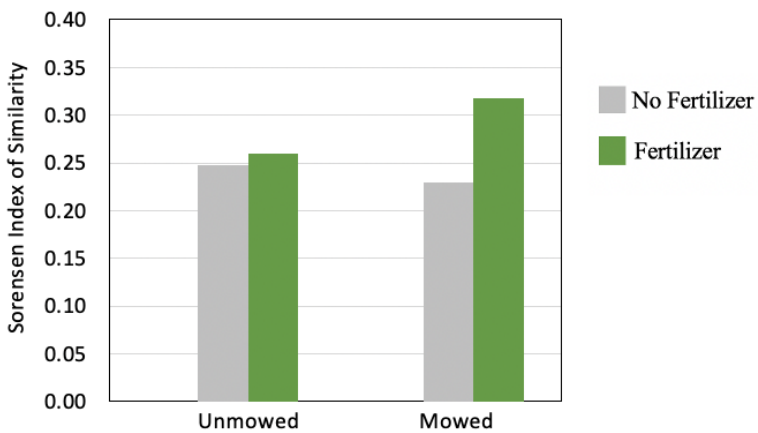

Figure 3 Similarity index of seed bank and 2004 aboveground vegetation.

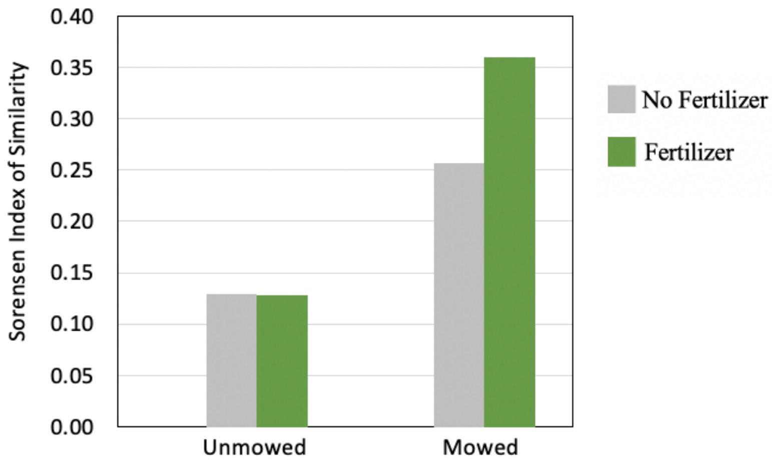

Figure 4 Similarity index of seed bank and 2021 aboveground vegetation.

Discussion

The most important finding of this study is that, after 20 years of succession, the seed bank retains the imprint of the original plant community. Based on the NMDS that investigated the composition of the seed bank, no real separation was found between the mowed and unmowed plots. This can be interpreted as mowing having no effect on the composition of the seed bank, which contrasts the current state of the aboveground vegetation where plant communities are distinctly different in mowed and unmowed plots. In a study of an ephemeral wetland, Faist et al. (2013) also found that the seed bank was more similar to the past aboveground vegetation than it was to the more recent aboveground vegetation, supporting the idea that seed banks holds the store of past species. In our study, the seed bank was equally similar to the 2004 aboveground vegetation in the unmowed and mowed plots, which at that early point in the experiment had similar plant species composition. In contrast, when comparing the composition of the seed bank to the 2021 aboveground vegetation, the seed bank had a much higher index of similarity to the mowed plots than the unmowed plots. A study by Dolle and Schmidt (2009) found that as disturbance, like mowing, increases there is an increase in the similarity between the seed bank and the aboveground vegetation. This finding agrees with the low index of similarity that was found between the current seed bank and the 2021 unmowed aboveground vegetation. The past state of the aboveground vegetation was similar in mowed and unmowed plots, which can be seen in the current seed bank which means that it may be reflecting the past plant community. Before the start of this experiment, this site was historically maintained by wildfires and mowing. Even though the aboveground vegetation in unmowed plots has changed immensely as succession to a woody community has occurred in the absence of disturbance, the seed bank is more similar to the plant community of the past.

In this seed bank study, fertilizer has also changed the composition of the seed bank, which can be seen by a distinct separation in plots of fertilized versus unfertilized plots in the NMDS. Findings from another long-term fertilization experiment also corroborates fertilizer affecting the composition of the persistent seed bank (Zhang et al. 2019). Fertilizer was found to increase both diversity and abundance in the seed bank, which contrasted with what was seen with the aboveground vegetation, where an overall decrease in species richness has been caused by fertilization. These contrasting results leave a variety of questions as to what roles fertilization plays in increasing seed bank species diversity. Zhang et al. (2019) explains that the effect of fertilization on seed bank composition can be due to its direct effects on soil pH as well as its indirect affects through altering the aboveground vegetation composition. The fertilizer may be acting to increase the fitness of the aboveground plant community, or it may even be acting to encourage germination of the seeds that are present, as has been demonstrated for some species in experimental studies (Sweeney et al. 2008).

Drainage has affected the composition of the seed bank, and this may be due to the length of time that the ditch causing the hydrology gradient has been present. The area of this site was drained by multiple ditches that were installed in 1960 as a method to prevent flooding for logging (Chester 2004). Since the installation of the drainage ditches, they have left a mark on the composition of the aboveground plant community and the seed bank. Plant communities away from the ditch's draining effects include many facultative and obligate wetland species, while those in the ditch's zone of influence are dominated by species that thrive in drier soils (Goodwillie et al. 2020). The effect that drainage has on the composition of the seed bank can be seen in a distinct separation in the drained versus the undrained plots in the NMDS. According to Faist et al. (2013), the germination of certain species may be driven by environmental factors. This could explain the varying effects from the presence or absence of moisture and nutrients on the composition of the aboveground vegetation.

The species that predominated in the seed bank included *Lobelia nuttallii, Eupatorium capillifolium,* and *Dichanthelium scoparium*. *Lobelia nuttallii* and *Eupatorium capillifolium* are both perennial forbs while *Dichanthelium scoparium* is a grass. Very few woody species were present in the seed bank. None of the species identified in the seed bank were true trees, only woody shrubs, which contrasts with the dominance of trees in the forest that can be seen in the unmowed plots. This again suggests that the seed bank is reflecting the past herbaceous composition of the aboveground vegetation.

The method of quantifying the seed bank created some limitations in this experiment. In the seed emergence method, the total number and identity of seeds present is unknown because some seeds may remain dormant in the soil and fail to germinate. A variety of factors can cause seeds to germinate, and the conditions in which our seed bank was kept in the growth room might have been more conducive to certain species while limiting the germination of other species. Another limitation of this experiment was the inability to identify some of the graminoid species, as well as one forb. During the growth period of the experiment, these seedlings did not present traits that allowed us to identify and distinguish them as a particular species.

Our study suggests that seed banks may be used for restoration purposes because the seed bank does not change as quickly as the vegetation above it, retaining species of at least the past 20 years according to this study. The seed bank may retain species for even more years to come, but a future study of the seed bank will be needed to test the length of seed bank persistence. The species that are kept in the seed bank may be used to increase future diversity. The dynamics that are occurring in the seed bank are not necessarily the same as those that are occurring in the aboveground vegetation which may lead to its ability to restore degraded habitats (Zhang et al. 2019). One instance where a seed bank may be used is to restore diversity is in areas of encroachment by woody species that is due to suppressed disturbance. In these instances, a disturbance, like wildfire, may have been prevented for many years which increases the amount of woody species encroachment. To restore the plant community to its original state that was present before encroachment, a prescribed fire might be used. In a study by Webster and Halpern (2010), it was found that in a mixed conifer forest a prescribed fire in a forest that has been suppressed of wildfires had an increase in species richness and abundance. The restoration of diversity will be facilitated by the presence of long-term retention of seeds from species present in communities in the past. Recently, in 2014 the coastal plain featured in the study was declared a diversity hotspot and this declaration was largely based on the amount of plant diversity found in the area (Noss et al. 2015). The legacy of this diversity will remain in the seed bank for years to come, even when it is not seen above ground.

Literature Referenced

Bernard, A., and Blaise, T. 2004. The relationships between soil seed bank, aboveground vegetation and disturbances in old, embanked marshlands of Western France. *Flora*. 199:25-35.

Carri, E., P. Biagini, and F. Selvi. 2022. Early vegetation recovery of a burned Mediterranean forest in relation to post-fire management strategies. *Forestry: An International Journal of Forest Research. 95:548-561.*

Cavers, P.B. 1994. Seed banks: Memory in soil. *Canadian Journal of Soil Science*. 75:11-13

Chester, B. 2004. Floristic assessment of a wet mineral flat at the East Carolina University West Research Campus and investigation of influential, human-mediated factors on the plant community. *East Carolina University, Masters thesis*

Dolle, M., and Schmidt, W. 2009. The relationship between soil seed bank, above-ground vegetation and disturbance intensity on old-field successional permanent plots. *Applied Vegetation Science*. 12:415-428.

Eskelinen, A., E. Elwood, S. Harrison, E. Beyen and J. R. Gremer. 2021. Vulnerability of grassland seed banks to resource-enhancing global changes. *Ecological Society of America.* 102.

Faist A. M. 2013. Banking on the past: seed banks as a reservoir for rare and native species in restored vernal pools. *AoB Plants*. 5:1-11.

Field C. D., N. B. Dise and R. J. Payne et al. 2014. The role of nitrogen deposition in widespread plant community change across semi-natural habitats. *Ecosystems*. 17:864-877.

Goodwillie, C., M. McCoy and A. L. Peralta. 2020. Long-term nutrient enrichment, mowing, and ditch drainage interact in the dynamics of a wetland plant community. *Freshwater Ecology*. 11(10):1-16.

He, M., C. Xin and C. Baskin et al. 2021. Different response of transient and persistent seed bank of alpine wetland to grazing disturbance on the Tibetan Plateau. *Plant and Soil*. 459:93-107.

Lennon J. T., F Hollander, M. Wilke-Berenguer, and J. Blath. 2021. Principles of seed banks and the emergence on complexity from dormancy. *Nature Communications*. 12:1-16.

Noss R.F., W. J. Platt, B. A. Sorrie, A. S. Weakley, D. B. Means, J. Costanza, and R. K. Peet. 2015. How global biodiversity hotspots may go unrecognized: Lessons from the North American Coastal Plain. *Diversity and Distributions*. 21:236-244.

Plassmann K., N. Brown, M. Laurence, M. Jones, and G. Edward-Jones. 2008. Can atmospheric input of nitrogen affect seed bank dynamics in habitats of conservation interest? The case of dune slacks. *Applied Vegetation Science.* 11:413-420.

Sorensen, T. 1948. A method of establishing groups of equal amplitude in plant sociology based on similarity of species content and its application to analysis of the vegetation on Danish commons. *Biol. Skr*. (K. Danske Vidensk. Selsk. NS) 5:1-34.

Sweeney A. E., K. A. Renner, C. Laboski and A. Davis. 2008. Effect of fertilizer nitrogen on weed emergence and growth. *Weed Science*. 56(5):714-721.

Ter Heerdt, G. N. J., G. L. Verweij, R. M. Bekker, and J. P. Bakker. 1996. An improved method for seed-bank analysis: Seedling emergence after removing the soil by sieving. *Functional Ecology* 10:144-151.

Thompson K., and J. P. Grime. 1979. Seasonal variation in the seed banks of herbaceous species in ten contrasting habitats. *Journal of Ecology* 67:893-921.

Webster, K. M., and C. B. Halpern. 2010. Long-term vegetation responses to reintroduction and repeated use of fire in mixed conifer forests of the Sierra Nevada. *Ecosphere* 1(5):1-27.

Zhang, C., C. G. Willis, Z. Ma, et al. 2019. Direct and indirect effects of long-term fertilization on the stability of the persistent seed bank. *Plant Soil* 438:239-250.

Appendix I

Family	Species	C	F	M	MF	Near Ditch	Away from Ditch
Anacardiaceae	*Rhus copallinum* L.	1	1	0	0	0	2
Asteraceae	*Baccharis halimifolia* L.	1	1	0	0	0	2
Asteraceae	*Erechtites hieraciifolius* (L.) Raf. ex DC.	0	1	0	1	2	0
Asteraceae	*Eupatorium capillifolium* (Lam.) Small	6	14	5	139	42	122
Asteraceae	*Eupatorium mohrii* Greene	2	1	0	2	0	5
Asteraceae	*Eupatorium rotundifolium* L.	1	0	3	8	1	11
Asteraceae	*Eupatorium semiserratum* DC.	2	5	2	11	13	7
Asteraceae	*Euthamia caroliniana* (L.) Greene ex Porter & Britton	4	2	10	13	6	23
Asteraceae	*Gamochaeta purpurea* (L.) Cabrera	1	0	0	4	2	3
Asteraceae	*Solidago pinetorum Small*	0	0	2	0	2	0
Asteraceae	*Solidago rugosa Mill.*	1	0	6	7	7	7
Asteraceae	*Solidago stricta Aiton*	0	0	2	0	0	2
Campanulaceae	*Lobelia nuttallii Schult.*	30	72	17	118	217	20
Clusiaceae	*Hypericum crux-andreae* (L.) Crantz	0	0	1	0	0	1
cyperacae	Unknown 1	5	5	16	20	32	14
cyperacae	Unknown 1	0	42	2	21	1	64
cyperacae	Unknown 3	1	9	5	1	1	15
cyperacae	Unknown 4	4	58	8	89	25	134
Fabaceae	*Lespedeza hirta* L.	0	0	0	1	0	1
juncacae	Unknown 1	0	4	0	3	0	7
Loganiaceae	*Gelsemium sempervirens* (L.) W.T. Aiton	1	1	0	0	0	2
Melastomataceae	*Rhexia mariana* L.	1	11	1	10	4	19
Onagraceae	*Ludwigia alternifolia* L.	0	0	0	2	0	2
Poaceae	*Amphicarpum purshii* Kunth	0	2	0	0	2	0
Poaceae	*Andropogon virginicus* L.	0	1	6	1	5	3
Poaceae	*Chasmanthium laxum* (L.) Yates	26	36	7	23	42	50
Poaceae	*Dichanthelium dichotomum* (L.) Gould var. dichotomum	13	9	2	21	15	30
Poaceae	*Dichanthelium scoparium* (Lam.) Gould	42	18	70	35	143	22
Poaceae	Unknown 1	14	9	11	17	33	18
Poaceae	Unknown 2	4	2	2	1	3	6
Poaceae	Unknown 3	4	0	9	4	6	11
Poaceae	Unknown 4	1	4	0	19	5	19
Poaceae	Unknown 5	0	1	2	0	3	0

Rosaceae	*Aronia arbutifolia* (L.) Pers.	0	0	1	1	0	2
Rosaceae	*Rubus argutus* Link	1	3	1	0	3	2
Rubiacae	*Gallium* sp.	0	0	0	1	0	1
Scrophulariaceae	*Gratiola pilosa* Michx.	0	0	1	8	1	8
Smilacaceae	*Smilax glauca* Walter	0	2	0	0	2	0
Smilacaceae	*Smilax rotundifolia* L.	3	0	0	0	3	0
Sphagnaceae	Unknown 1	6	35	17	42	49	51
unkown forb	Unknown 1	0	0	0	1	0	1
Violaceae	*Viola ×primulifolia* L. (pro sp.) [lanceolata × macloskeyi]	3	1	0	1	2	3

Family	Species	Growth Habitat	Wetland Status	Reproduction	Annual/ Perennial
Anacardiaceae	*Rhus copallinum* L.	Shrub Tree	UPL	fruit and rhizomes	Perennial
Asteraceae	*Baccharis halimifolia* L.	Shrub Tree	FAC	dioecious	Perennial
Asteraceae	*Erechtites hieraciifolius* (L.) Raf. ex DC.	Forb/herb	-	flower	Annual
Asteraceae	*Eupatorium capillifolium* (Lam.) Small	Forb/herb	FACU	flower	Perennial
Asteraceae	*Eupatorium mohrii* Greene	Forb/herb	FACW	flower and rhizome	Perennial
Asteraceae	*Eupatorium rotundifolium* L.	Forb/herb	FACW	flower	Perennial
Asteraceae	*Eupatorium semiserratum* DC.	Forb/herb	FACW	flower	Perennial
Asteraceae	*Euthamia caroliniana* (L.) Greene ex Porter & Britton	Forb/herb	FAC	rhizome	Perennial
Asteraceae	*Gamochaeta purpurea* (L.) Cabrera	Forb/herb	UPL	seeds	Annual
Asteraceae	*Solidago pinetorum Small*	Forb/herb		rhizome	Perennial
Asteraceae	*Solidago rugosa Mill.*	Forb/herb	FAC	rhizome	Perennial
Asteraceae	*Solidago stricta Aiton*	Forb/herb	OBL	rhizome	Perennial
Campanulaceae	*Lobelia nuttallii Schult.*	Forb/herb	FACW	-	Perennial
Clusiaceae	*Hypericum crux-andreae* (L.) Crantz	Shrub Subshrub	FACW	-	Perennial
cyperacae	Unknown 1	-	-	-	-
cyperacae	Unknown 1	-	-	-	-
cyperacae	Unknown 3	-	-	-	-
cyperacae	Unknown 4	-	-	-	-

Fabaceae	*Lespedeza hirta* L.	Forb/herb	FAC	flower	Perennial
juncacae	Unknown 1	-	-	flower	Perennial
Loganiaceae	*Gelsemium sempervirens* (L.) W.T. Aiton	Shrub Vine	FAC	seeds	Perennial
Melastomataceae	*Rhexia mariana* L.	Forb/herb	FACW	colonial hermaphrodite	Perennial
Onagraceae	*Ludwigia alternifolia* L.	Forb/herb	OBL		Perennial
Poaceae	*Amphicarpum purshii* Kunth	Graminoid	FACW	selfing	Annual
Poaceae	*Andropogon virginicus* L.	Graminoid	FAC	rhizome	Perennial
Poaceae	*Chasmanthium laxum* (L.) Yates	Graminoid	FACW		Perennial
Poaceae	*Dichanthelium dichotomum* (L.) Gould var. dichotomum	Graminoid	FAC	selfing	Perennial
Poaceae	*Dichanthelium scoparium* (Lam.) Gould	Graminoid	FACW	rhizomes	Perennial
Poaceae	Unknown 1	-	-	-	-
Poaceae	Unknown 2	-	-	-	-
Poaceae	Unknown 3	-	-	-	-
Poaceae	Unknown 4	-	-	-	-
Poaceae	Unknown 5	-	-	-	-
Rosaceae	*Aronia arbutifolia* (L.) Pers.	Shrub	FACW	rhizomes	Perennial
Rosaceae	*Rubus argutus* Link	Subshrub	FAC	fruit and rhizomes	Perennial
Rubiacae	*Gallium* sp.	Forb/herb	-	fruit	Perennial
Scrophulariaceae	*Gratiola pilosa* Michx.	Forb/herb	FACW	flowers; heteromorphic diaspores	Perennial
Smilacaceae	*Smilax glauca* Walter	Shrub Vine	FAC	fruit dioecious	Perennial
Smilacaceae	*Smilax rotundifolia* L.	Shrub Vine	FAC	fruit dioecious	Perennial
Sphagnaceae	Unknown 1	-	-	-	-
unkown forb	Unknown 1	Forb/herb	-	-	-
Violaceae	*Viola ×primulifolia* L. (pro sp.) [lanceolata × macloskeyi]	Forb/herb	-	rhizomes	Perennial

Family	Number of Seedlings
Anacardiaceae	2
Asteraceae	257
Campanulaceae	237
Clusiaceae	1
cyperacae	286
Fabaceae	1
juncacae	7
Loganiaceae	2
Melastomataceae	23
Onagraceae	2
Poaceae	416
Rosaceae	7
Rubiacae	1
Scrophulariaceae	9
Smilacaceae	5
unkown forb	1
Violaceae	5