5-2019

The Effect of a Surfactant Seed Coating on the Germination and Growth of Three Native Bulrushes

Anders Hart
Utah State University

THE EFFECT OF A SURFACTANT SEED COATING ON THE GERMINATION AND GROWTH OF THREE NATIVE BULRUSHES

by

Anders Hart

Capstone submitted in partial fulfillment of the requirements for graduation with

UNIVERSITY HONORS

with a major in

Conservation and Restoration Ecology in the Department of Wildland Resources

Approved:

Capstone Mentor
Dr. Karin Kettenring

Departmental Honors Advisor
Dr. Geno Schupp

University Honors Program Director
Dr. Kristine Miller

UTAH STATE UNIVERSITY
Logan, UT

Spring 2019

Abstract

Great Salt Lake (GSL) wetlands provide vital ecosystem services, including habitat for migratory birds. Alkali bulrush (*Bolboschoenus maritimus*), hardstem bulrush (*Schoenoplectus acutus*), and three-square bulrush (*Schoenoplectus americanus*) play an important role in providing these services, but invasion by *Phragmites australis* has reduced the extent these species in GSL wetlands. Restoring these native bulrushes following *Phragmites* removal is a primary goal for GSL managers. However, climate change and increasing human water demands upstream may alter the hydropattern of GSL wetlands, leading to lower soil moisture availability and potentially inhibiting germination and establishment of these species. Surfactant seed coatings (SSCs) have been effective in increasing soil moisture availability and plant establishment in dryland systems but have not been tested extensively on wetland species. To understand if an SSC could enhance wetland revegetation projects, we conducted two experiments to test the effect of an SSC on germination and seedling biomass of *B. maritimus*, *S. acutus*, and *S. americanus*. In one experiment, we tested whether the addition of an SSC at a low and high dose to seeds of *B. maritimus, S. americanus*, and *S. acutus* increased germination proportion, germination synchrony, and germination rate when moisture level was kept constant in growth chambers. *S. acutus* had a significantly higher germination proportion at the low and high doses of the SSC compared to uncoated control, while the respective germination proportions of the other species were not different from control. *S. acutus* had a significantly higher germination synchrony with the low-dose coating compared to control, while there were no significant differences in germination rate in any of the species. In a second experiment, we tested the effect of a low and high dose of the SSC on the per-seedling above- and belowground biomass of these species in growth chambers at three moisture levels. For the low-dose SSC treatment, *S. acutus* had higher per-seedling aboveground and belowground biomass than control at the intermediate moisture level, while the other species-treatments combinations did not show a clear pattern. These results suggest that this SSC may enhance the germination proportion and synchrony of *S. acutus*, and under certain conditions may lead to higher seedling biomass of *S. acutus*. Future research should seek to reproduce these findings and determine if an SSC is a useful tool in wetland revegetation projects.

Acknowledgements

I want to thank my faculty mentor, Dr. Karin Kettenring, for her guidance on this project. Her patience and expertise helped me to carry this research through to its completion. I also want to thank my collaborator, Emily Martin for her insight and problem-solving skills that contributed greatly to the success of this project. I'm grateful for the S.J. and Jessie E. Quinney College of Natural Resources for generously providing funding to support this research. Dr. Matthew Madsen at Brigham Young University provided invaluable help with the seed coating. I couldn't have completed this research without the help of technicians in the Kettenring Wetland Ecology Lab. Finally, I thank my family, friends, and professors for their support throughout my time at Utah State University.

Table of Contents

List of Figures and Tables

Final Written Product

Word Count: 5402

Introduction

The wetlands surrounding the Great Salt Lake (GSL) provide many ecosystem services. One such service is the provision of habitat and seeds which many species of migratory and nesting waterbirds use as a food source beginning in midsummer (Evans and Martinson, 2008). Three of the most important native plant species that characterize these wetlands are *Bolboschoenus maritimus* (alkali bulrush), *Schoenoplectus acutus* (hardstem bulrush), and *S. americanus* (three-square bulrush). However, in recent years an invasive species and increasing water demands on the rivers that supply water to GSL wetlands have posed serious challenges for the continued viability of this ecosystem and the services it provides (Downard et al., 2014).

The main invasive species that affects GSL wetlands is the grass *Phragmites australis*, which quickly invades wetlands and forms dense monocultures which prevent native species from reestablishing (Verlandier et al., 2013). *P. australis* has invaded 93 km^2 near the Great Salt Lake with the potential to spread to another 9.6 km^2 in the system (Long et al. 2016). The Kettenring Wetland Ecology Lab at Utah State University has produced recommendations for the restoration of wetlands invaded by *P. australis* (Rohal et al., 2016). However, restoring native vegetation, including the bulrush species mentioned above, has proven difficult.

One of the main limiting factors for revegetating GSL wetlands with native species is water availability. *Bolboschoenus maritimus*, *S. acutus*, and *S. americanus* seeds generally germinate at the highest rate under moist or flooded conditions, though they have other dormancy-breaking requirements; of these three species, *B. maritimus* seeds show the strongest positive response to flooding (Marty and Kettenring 2017). Due to the moisture requirements for germination, water stress may limit the germination and establishment of these species. This concern is especially salient because western U.S. snowpack is predicted to decline by up to 60% over the next 30 years as a consequence of climate change (Fyfe et al. 2017). The rapidly growing population of the Wasatch Front on the eastern shore of the Great Salt Lake also poses risks for water availability in GSL wetlands, as increased urban development eliminates the seasonal agricultural return flows that supply water to many GSL wetlands (Downard et al. 2014).

Surfactant seed coatings (SSCs) can protect seeds and seedlings from water stress and have proven useful in increasing the germination and seedling growth of upland plants in western U.S. rangeland ecosystems (Madsen et al. 2014). For the purposes of this research, an SSC is a nonionic alkyl ended block copolymer that is applied directly to the seed. SSCs reduce the hydrophobicity of soil microsites surrounding seeds and can increase water availability for germinating seeds and young seedlings (Madsen et al. 2016a). Madsen et al. (2016b) found that the application of an SSC to seeds of two turfgrasses improved germination synchrony, reduced time to germination, increased aboveground biomass and cover relative to untreated seed.

However, it is unknown if these coatings could enhance the germination and establishment of wetland plants. Therefore, we conducted two experiments to evaluate the effectiveness of a surfactant seed coating (SSC) for promoting germination and increased growth of *B. maritimus*, *S. acutus*, and *S. americanus*. More specifically, our goal was to test the effectiveness of an SSC (Aquatrols ASET 4001) in increasing the germination synchrony, reducing the time to germination, and increasing the aboveground and belowground biomass of seeds and seedlings, with a focus on aiding germination and establishment under water-stressed conditions. We hypothesize that this coating would increase germination synchrony, reduce the time to germination, and increase the biomass of these species, especially under water-stressed conditions.

Methods

Seed sourcing and viability testing

The bulrush seeds used in these experiments were harvested from multiple GSL wetland sites in Utah in 2016. These seeds were tested in February 2018 (7 months before the first experiment) for viability using the tetrazolium method (Lakon 1949). The *B. maritimus* seed had an average viability of 82% (SD =8.4), the *S. acutus* seeds had an average viability of 73% (SD=4.9), and the *S. americanus* had an average viability of 55% (SD=1).

Viability tests on the same seed lots done in February 2019 showed that *B. maritimus* had an average viability of 88.4% (SD=3.1%), *S. acutus* had an average viability of 61.9% (SD=16.5%), and *S. americanus* had an average viability of 40.2% (SD=2.7%).

Experiment 1: Germination

Coating Preparation

This experiment ran from September 14 to November 7, 2018. Because *S. americanus* and *S. acutus* seeds require cold, moist stratification of at least 30 days for highest potential germination, we used seeds that had been in cold, moist stratification for about 4 months before this experiment took place (Marty and Kettenring 2017). The stratification treatment consisted of storage in a 5-gallon bucket with moist sand in a standard refrigerator at approximately 4 °C. We broke dormancy of the approximately 163 g of *B. maritimus* seeds by soaking them in a 3% bleach solution for approximately 30 hours before applying the SSC using a paint strainer bag (see Marty and Kettenring 2017).

We coated seeds separately for each experiment. Seeds were coated in the Madsen lab at Brigham Young University in Provo, Utah and were transported back to the Kettenring lab at Utah State University (USU) in Logan, Utah where the experiments were carried out. On the day we coated the seeds, we thoroughly rinsed the *B. maritimus* seeds with tap water and placed them in a plastic bucket. We placed the *S. americanus* and *S. acutus* seeds in a styrofoam container for transport to the Madsen lab at Brigham Young University. In the Madsen lab, we dried all the seeds (each species separately) in a seed drier for approximately 15 minutes. We then prepared to coat a portion of each species with a low dose of the surfactant, a high dose, and leave a portion as an uncoated control. The surfactant coating consisted of the active ingredient, ASET-4001 surfactant (85%), Selvol polyvinyl alcohol binder, and diatomaceous earth (DE).
The seed coater (30-cm rotary seed coater from Universal Coating Systems) required at least 100 g of seed to coat batches evenly. We used *Purshia tridentata* seed that was available in the Madsen lab as a filler to ensure that we had at least 100 g of seed to place in the seed coater. For the *S. acutus*, we used 67.7 g *P. tridentata* as filler. For the *S. americanus*, we used 84.4 g *P. tridentata* in each coating. For the *B. maritimus*, we used 80.43 g *P. tridentata* in each coating.

Low-Dose Coating

This dose consisted of 0.1 g of ASET-4001 surfactant per 100 g of seed. For this treatment, we made a 10x batch of polyvinyl alcohol binder and the surfactant (109 g binder and 1 g surfactant) for easier measurement of the surfactant. We then took a 1x portion of that mixture (10.9 g binder and 0.1 g surfactant) and applied the mixture to the seeds in the coater. We then applied diatomaceous earth and the remaining polyvinyl alcohol binder to the seeds to prevent them from

sticking to each other. The recipe for the low-dose coating for 100 g seed was: 16.4 g polyvinyl alcohol binder, .1 g ASET-4001 surfactant, and 7.0 g of diatomaceous earth. After coating, each batch of each treatment-species combination was dried for about 5 minutes in the seed dryer (35 °C) and placed in a paper bag.

High-Dose Coating

This coating consisted of 5 g ASET-4001 surfactant per 100 g of seed. Applying the high-dose coating involved three steps: two base coatings of polyvinyl alcohol binder and diatomaceous earth and a final coating of the surfactant with diatomaceous earth. For 100 g of seed, step one consisted of 14.0 g polyvinyl alcohol binder and 10.0 g diatomaceous earth; the second step consisted of 66 g polyvinyl alcohol binder and 53.0 g diatomaceous earth; step 3 consisted of 5.0 g ASET-4001 surfactant and 5.0 g diatomaceous earth. After coating, each batch of each treatment-species combination was dried for about 5 minutes in the seed dryer (35 °C) and placed in a paper bag.

Germination Trial

In the Kettenring lab, we filled 45 19.3×11.4×9.4 cm (1050 ml) rectangular clear plastic containers (5 replicates x 3 species x 3 coating treatments) each with 200 ml quartz sand and placed 50 seeds in each container for the corresponding treatment, being careful to discard the filler seeds. We then added 80 ml of tap water to each container and closed the snap-on lids. We weighed each container and recorded the total weight on the lid. We randomly placed the containers in a Conviron A1000 growth chamber with the temperature set to 18 °C (night, from 7:00 p.m. to 6:00 a.m.) and 33 °C (day, 6:00 a.m. to 7:00 p.m.), with humidity set to 80% (actual humidity rarely exceeded 40% at these settings).

We checked for germination in the containers every 2 days. We defined germination as the emergence of a radicle from the seed (Nonogaki et al. 2010). We counted and removed germinated seeds when we encountered them. Every 4 days, we weighed a random sample of 8 containers. If the average reduction in weight from the original weight exceeded 2%, we added water to bring the weight of each container back to the starting weight.

We stopped counting seedlings 47 days after beginning the experiment because we judged that germination was then resulting from seeds whose germination was broken during their time in the growth chamber (Baskin and Baskin 2014). Often the coating resulted in 2 or 3

seeds clumping together, so some of the containers with coated seeds contained more than 50 seeds. To have an accurate count of seeds in each container, we counted the remaining seeds in each container by combing through the top 1 cm of sand with tweezers and removing each seed that was found until no more seeds could be detected.

Data Analysis

We performed a one-way Analysis of Variance (ANOVA) on the final germination proportion, with seed coating (3 levels) as the fixed effect using JMP software. We also calculated germination synchrony and mean germination rate using the GerminaQuant R package. Germination synchrony is an indicator of how overlapping individual germination events are in time (Ranal and Santana 2006). The mean germination rate is the reciprocal of the mean germination time, which is calculated as the average time to germination weighted by the average number of seeds in each time interval (Labouriau 1983). We also performed Tukey HSD analyses of the species-treatment combinations, with alpha set to 0.10 for assessing significance because the Tukey HSD tends to be overly conservative (Abdi and Williams 2010).

Experiment 2: Biomass

This experiment ran from October 17 to December 5, 2018. We coated a fresh batch of seeds of each species from the original seed lots immediately before this trial. We followed the same procedure as the previous round of coating described above to obtain seeds coated with a low dose and a high dose of the surfactant, with the exception that we brought enough seed that filler seeds were not needed. Because some seeds clumped together after coating, we sieved all the coated seeds to reduce the proportion that was clumped.

We filled 135 1050-mL rectangular plastic containers with 150 g of SunGro Propagation Mix and placed them in a drying oven at 60 °C about 43 hours before sowing the seeds. Following the addition of seeds, we watered each pot with enough water to reach the required percentage of field capacity. The moisture treatments we used for this experiment were 100%, 72%, and 55% of field capacity. We had previously determined the field capacity of this brand of propagation mix using methods adapted from Madsen et al. (2016b). We poked several small holes in 3 of the same 1050-mL containers and filled each with about 100 g of the propagation mix. We soaked the soil in each container with tap water and allowed each container to drain for about 24 hours with the tops covered to prevent evaporative losses. After the draining period, we

placed the containers in a drying oven at 60°C for 48 hours. After that time, we measured the soil mass and subtracted the oven-dried weight of soil in each container from the weight at field capacity (obtained after draining) to obtain the weight of water that was required to bring the soil from oven-dryness to field capacity. We divided that quantity by the weight of oven-dried soil to know how much water per gram of soil was needed to bring oven-dry soil to field capacity. The average value was 6.47 grams of water per gram of oven-dry soil.

When we were sowing the seeds for this experiment, adding water to the soil disturbed the surface and caused some seeds to sink to the interior of the soil mass and the bottom of the container. To ensure that enough seeds were present on the soil surface, we added a small number of seeds (about 30) to the soil after watering and sprayed the soil with tap water from a spray bottle to ensure that water came in contact with the seed coating in a manner analogous to precipitation. After watering and adding the seeds again, we recorded the weight of each container. We placed a 1-gallon plastic bag with 3-4 small (~2 mm-diameter) holes in each side over each of the containers to promote moisture retention between waterings. We randomly placed the containers in two Conviron A1000 germination chambers with the same temperature, humidity, and photoperiod settings as Experiment 1. Twice a week, we weighed each container after removing the plastic bag from each and added water to bring the weight back to the correct weight.

Marking Seedlings and Biomass Harvest

We began recording the number of seedlings in each pot 18 days after beginning the experiment, and then did so every time we watered the pots (twice a week). We removed seedlings from the pots when they reached approximately 21 days old. Because we had not marked the seeds by the day of emergence until after many seeds had germinated, we had to estimate the age of each seedling relative to the other seedlings in the same container based on size. We marked all seedlings with colored flags that corresponded to the approximate day of emergence. After we began marking new seedlings, we marked new seedlings during the twice-weekly watering until 30 days after planting the seeds, after which we discarded any new seedlings because we determined that at that point germination was resulting from seeds whose dormancy was broken during their time in the growth chamber (Baskin and Baskin 2014). For the period between the beginning of marking the new seedlings and 30 days after planting, we assumed germination

occurred at the midpoint between watering periods because we only recorded germination two times per week. 14 pots did not have any seedlings in them when we stopped marking new seedlings and at that were removed from the growth chambers. For a small number of pots, we had to subtract from the total number of seedlings because seeds either died or were no longer visible near the flag marking their location in the container.

When seedlings reached 21 days old, we removed them from the containers and used scissors to collect the belowground and aboveground biomass for each container, with all the aboveground biomass for each container pooled in a coin envelope and all the belowground biomass for each container pooled in a coin envelope. After all seedlings were harvested, we dried the biomass in an oven at 60°C for 48 hours, after which we weighed each sample.

Data Analysis

We divided the total aboveground and total belowground biomass by the number of seedlings in each container and then performed a two-way ANOVA on the per-seedling aboveground and per-seedling belowground biomass data with coating (3 levels) and soil field capacity (3 levels) as fixed effects. For analyses where there were significant effects, we conducted Tukey's range tests to test the significance of interactions among effects, with alpha set to 0.10 for assessing significance because the Tukey test is overly conservative (Abdi and Williams 2010).

Results

Experiment 1: Germination

Final Germination Proportion

Schoenoplectus acutus final germination was higher overall, regardless of seed coating, than the other two species: 36.2% for *B. maritimus*, 75.8% for *S. acutus*, 30.1% for *S. americanus*. *S. acutus* germination with the seed coating (low and high dose) was higher than control (p=.04). No significant differences were seen in germination proportion between treatments in the other species.

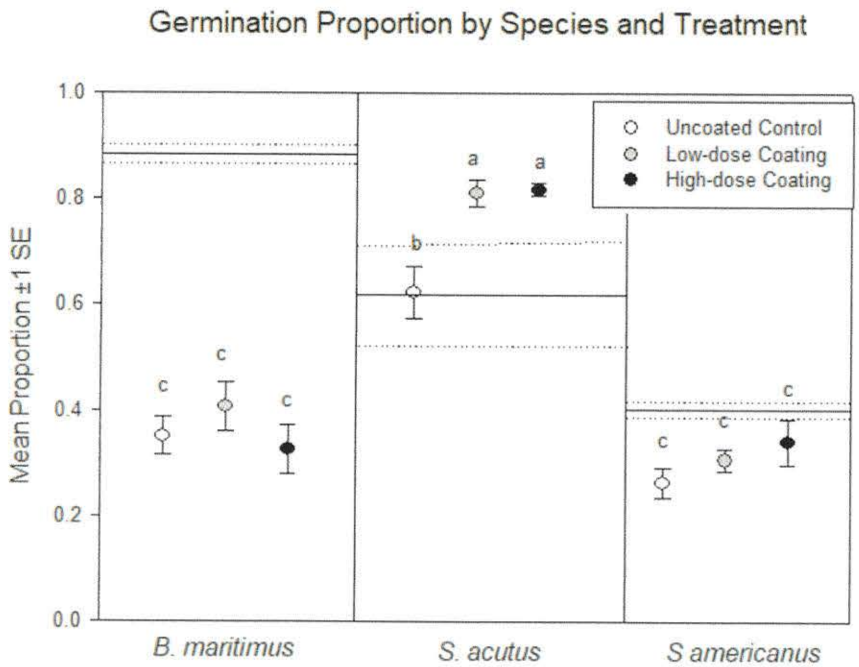

Figure 1. Final germination proportion results for all germination treatments. "C" means control, "LD" means low-dose, and "HD" means high-dose. Lines show mean seed viability ± 1 SE from the February 2019 test. Different letters indicate significant differences at the 0.10 level.

Germination Synchrony

S. acutus seeds with the low-dose coating had significantly higher germination synchrony than all of the other treatments except the *S. acutus* seeds at the high dose, which had lower but not statistically different synchrony ($p<0.01$).

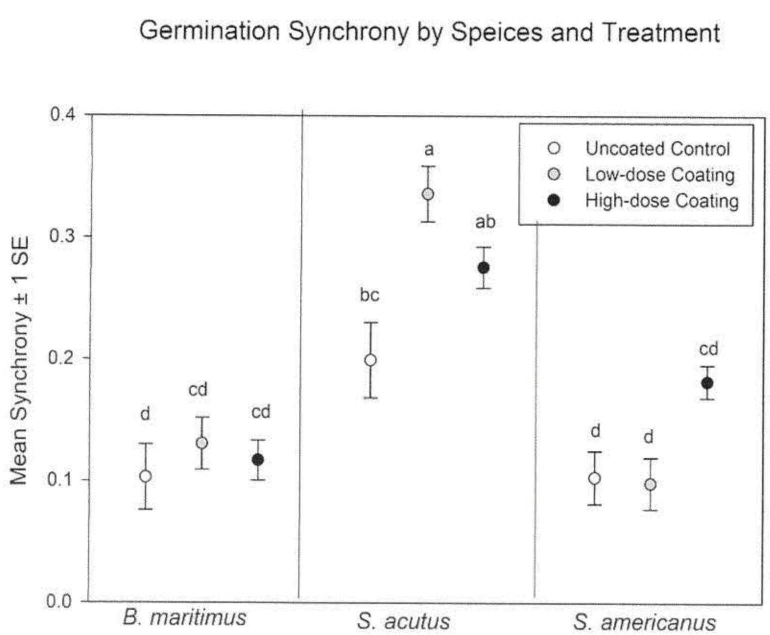

Figure 2. Germination synchrony results for all germination treatments. "C" means control, "LD" means low-dose, and "HD" means high-dose. Different letters indicate significant differences at the 0.10 level.

Germination Rate

The species-coating treatment interaction was not significant for germination rate (p=0.16), and the *S. acutus* seeds had a higher germination rate than the other two species (p<0.01).

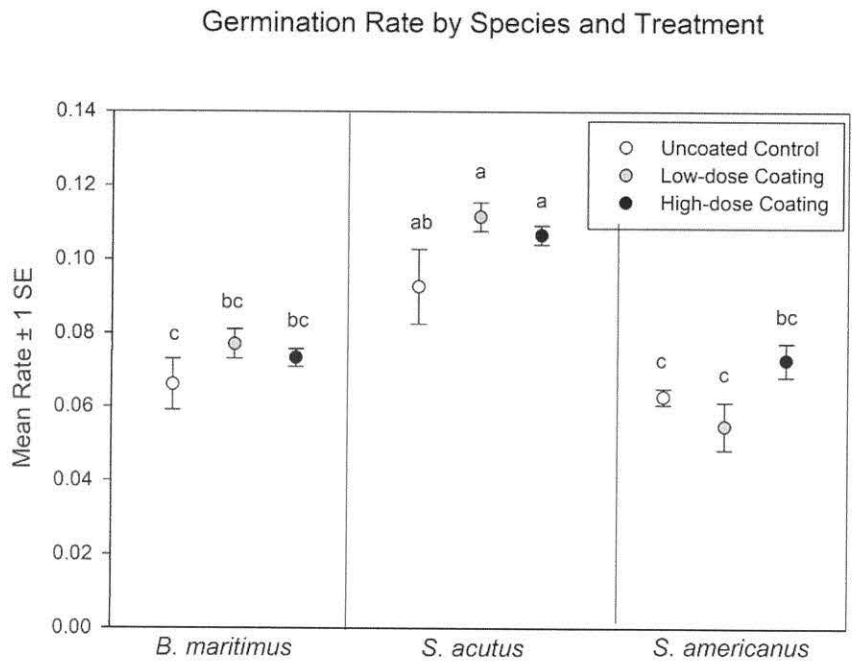

Figure 3. Mean germination rate for all germination treatments. The mean germination rate is the reciprocal of the mean germination time, which is calculated the average time to germination weighted by the average number of seeds in each time interval (Labouriau 1983). "C" means control, "LD" means low-dose, and "HD" means high-dose. Different letters indicate significant differences at the 0.10 level.

Germination Speed

S. acutus seeds germinated more quickly on average that *B. maritimus* seeds and *S. americanus* seeds. *B. maritimus* reached 90% of total eventual germination 27 days after sowing, *S. acutus* reached 90% germination 13 days after sowing, and *S. americanus* reached 90% germination 31 days after germination. Because only *S. acutus* showed significant differences in germination synchrony among coating treatments, here we only report time to 90% germination for coating treatments of that species. Time to 90% germination for the *S. acutus* control group seeds was 17 days, 11 days for the *S. acutus* low-dose seeds, and 13 days for high-dose *S. acutus* seeds.

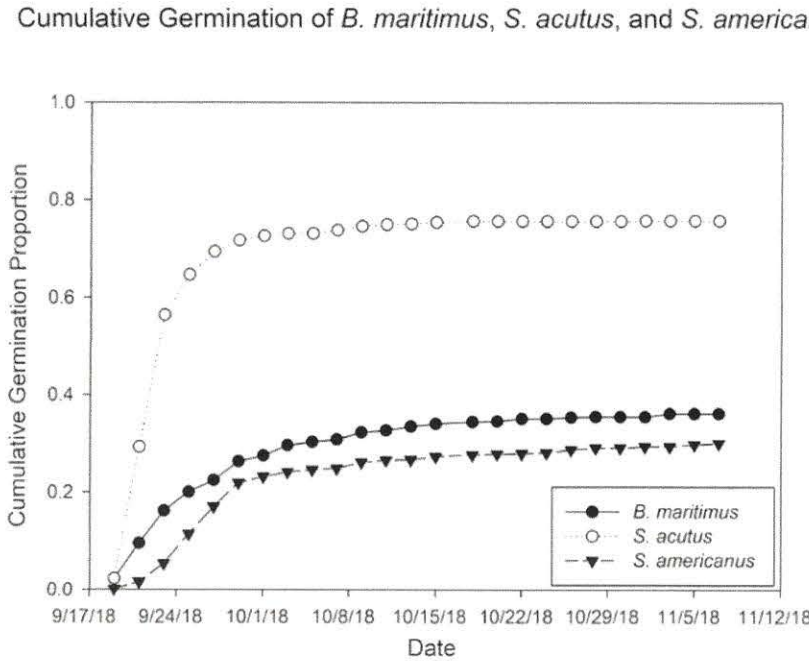

Figure 4. Cumulative germination proportion of each of the bulrush species.

Effect	F Ratio	DF	*p*-value
Germination Proportion			
Species	131.58	2, 35	0.00
Coating	6.01	2, 35	0.01
Container	0.01	1, 35	0.95
Species × Coating	2.73	4, 35	0.04
Germination Rate			
Species	44.88	2, 35	0.00
Coating	2.87	2, 35	0.07
Container	1.21	1, 35	0.28
Species × Coating	1.75	4, 35	0.16
Germination Synchrony			
Species	50.32	2, 35	0.00
Coating	6.89	2, 35	0.00
Container	2.90	1, 35	0.10
Species × Coating	5.02	4, 35	0.00

Table 1. ANOVA results from the germination experiment.

Experiment 2: Biomass

Aboveground Biomass

Species, coating treatment, the moisture level-species interaction, and the moisture level-coating treatment interaction were significant at the 0.05 level in our model for the aboveground biomass. For *B. maritimus*, none of the coated treatments were significantly different from control (p=0.43). For *S. acutus*, the only significant difference between a coated treatment and control was at the intermediate water level, where the low-dose treatment was significantly higher than control and the high-dose treatment (p=0.01, Figure 5). For *S. americanus*, none of the coating-moisture level treatments were significantly different from each other (p=0.38).

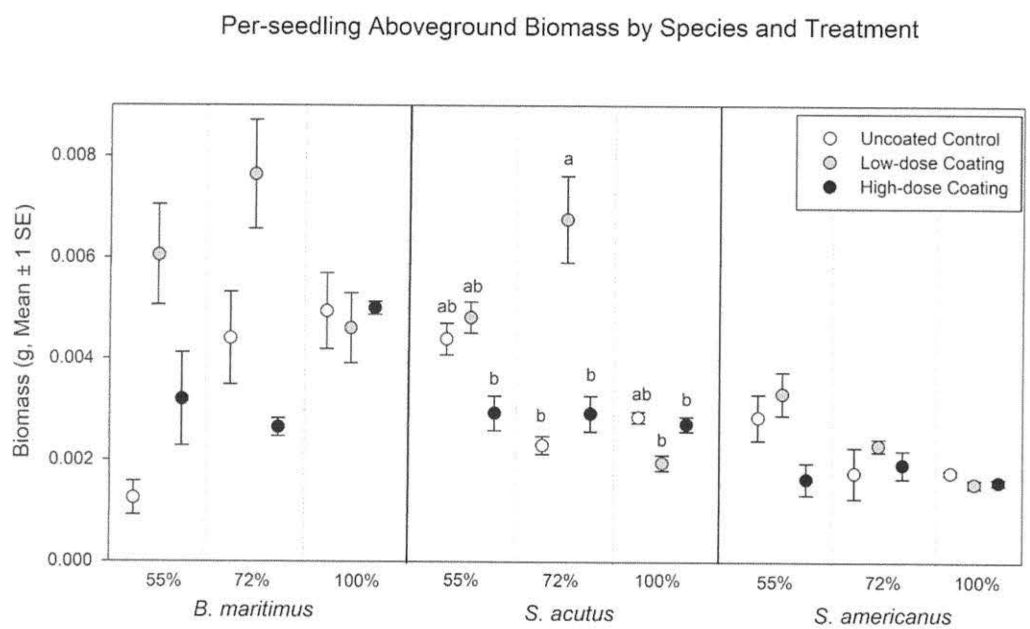

Figure 5. Aboveground biomass for each species by treatment.

<u>Belowground Biomass</u>

Species was the only factor that was significant at the 0.05 significance level in our model for the per-seedling belowground biomass (p<0.01). For *B. maritimus*, there was no clear pattern or statistical significance in the per-seedling belowground biomass results, though the uncoated treatment at the moderate moisture level was considerably lower than the low-dose and high-dose treatments at that moisture level. The coating treatment-moisture level interaction was significant for *S. acutus* (p<0.01). For *S. acutus*, per-seedling belowground biomass was significantly higher with the low-dose coating at the intermediate moisture level than the high dose at the low moisture level, low-dose treatment at the highest moisture level, and the control treatment at the medium moisture level. For *S. americanus*, none of the coating-moisture level

combinations were significantly different from each other for belowground biomass, though the low-dose treatment was considerably higher than the control and high-dose treatments at the lowest moisture level.

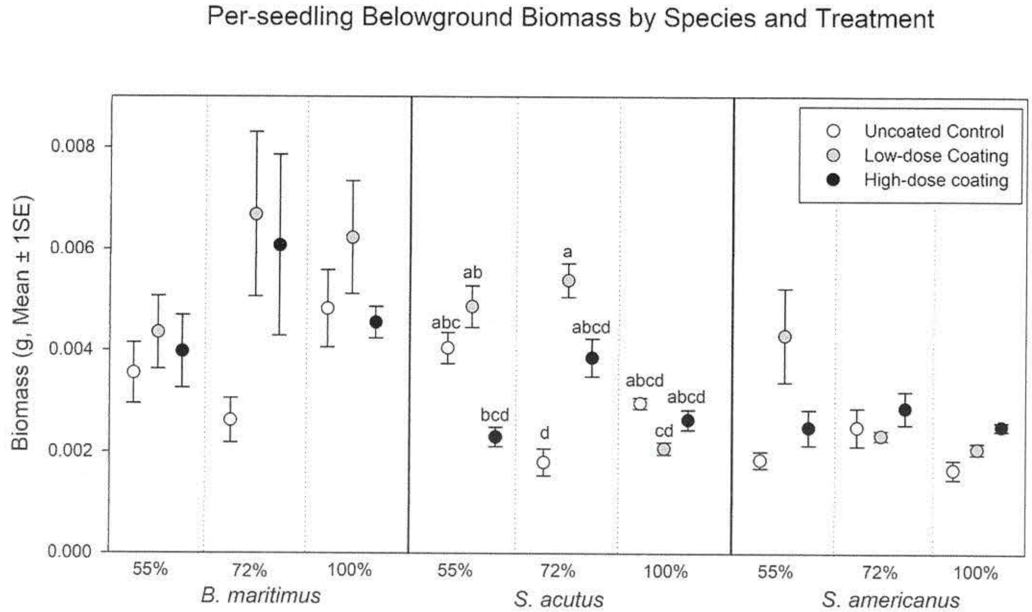

Figure 6. Belowground biomass for each species by treatment.

Effect	**F Ratio**	**DF**	***p*-value**
Aboveground *B. maritimus* Biomass			
Seed coating treatment	2.10	2, 36	0.14
Moisture level	1.55	2, 36	0.23
Moisture × Coating	0.99	4, 36	0.43
Belowground *B. maritimus* Biomass			
Seed coating treatment	0.37	2, 36	0.70
Moisture level	0.29	2, 36	0.75
Moisture × Coating	0.24	4, 36	0.91
Aboveground *S. acutus* Biomass			
Seed coating treatment	2.94	2, 36	0.07
Moisture level	3.53	2, 36	0.04
Moisture × Coating	3.79	4, 36	0.01
Belowground *S. acutus* Biomass			
Seed coating treatment	3.21	2, 36	0.05
Moisture level	2.65	2, 36	0.08
Moisture × Coating	5.77	4, 36	0.00
Aboveground *S. americanus* Biomass			
Seed coating treatment	1.55	2, 36	0.23

Moisture level	1.58	2, 36	0.22
Moisture × Coating	1.09	4, 36	0.38
Belowground *S. americanus* Biomass			
Seed coating treatment	1.77	2, 36	0.19
Moisture level	1.21	2, 36	0.31
Moisture × Coating	1.12	4, 36	0.37

Table 2. ANOVA results from the germination experiment.

Discussion

We sought to understand the effect of a surfactant seed coating (SSC) on the germination and biomass of three important wetland plants in this study. Increased germination and early growth of *B. maritimus*, *S. acutus*, and *S. americanus* with an SSC will potentially increase the resistance of revegetation projects to water stress in the face of climate change and increasing water withdrawals. We found that the low-dose treatment of the surfactant seed coating was beneficial for the germination of *S. acutus* seeds. Out of the coating treatments, the low-dose treatment showed the most consistent benefit for per-seedling biomass, though results were not consistent across species. This effect was evident for aboveground and belowground biomass of *S. acutus* at the intermediate water level. The pattern that emerges from these results is that a low-dose application of the SSC may aid the germination of *S. acutus* and enhance its seedling growth under moderate moisture conditions. However, the benefits seen for *B. maritimus* and especially *S. americanus* were limited.

Responses of **B. maritimus** *and* **S. americanus**

We are unsure as to why the *B. maritimus* seed displayed a low germination proportion in the germination trial, given the high viability of the seed lot seen in both tetrazolium tests. We also observed a generally low apparent germination proportion in the biomass trial, though we could not calculate the proportion in that trial because we did not record the exact number of seeds sown in each container. It is possible that soaking in 3% bleach for about 30 hours was too long and damaged embryos, as Kettenring (2016) found that *B. maritimus* had higher germination when soaked in bleach for 24 hours compared to 48 hours.

The lack of strong responses of *S. americanus* in both the germination and biomass experiments may be due to the difficulty in germinating *S. americanus* seeds in laboratory conditions in general (Marty and Kettenring 2017). Additionally, Keddy and Ellis (1985) found

that *S. americanus* did not show differential recruitment responses to a gradient of water availability, which ranged from water 10 cm above the soil surface to 5 cm below the soil surface. However, in Karagatzides and Hutchinson's (1991) evaluation of *S. americanus* biomass along an elevational gradient in a Canadian tidal marsh, *S. americanus* had higher aboveground and belowground biomass in the higher elevation location compared to the lower elevation. The main limitation of that study is that it did not explicitly examine water potential along the elevational gradient, and water availability may not have been a limiting factor for growth of *S. americanus* in the study area. For *S. americanus*, water availability may not be the most important factor driving germination and early establishment, and further research is needed to understand the optimal conditions for its germination and establishment (Marty and Kettenring 2017).

Mode of Action of the Surfactant Seed Coating

When interpreting these results, it is important to consider the mode of action of the SSC, which is reducing soil water repellency (Madsen et al. 2016a). Potting soil has a high water retention capacity, but Guatam and Ashwath (2012) found that many potting media become hydrophobic when oven-dried. Since we used oven-dried soil in both the germination and biomass experiments, this study provided a good test of the SSC, as the mode of action for this SSC is to reduce soil water repellency and allow more water to reach seeds and seedling roots (Madsen et al. 2016a). Guatam and Ashwath (2012) also found that when water content in the media was maintained at 15%, few of the media exhibited strong hydrophobicity. However, the reduction in water repellency from the SSC may have only occurred after the first watering in both experiments. Also, it is unclear if natural wetland soils would exhibit hydrophobicity and if so, at what water potentials.

Limitations

While these results highlight the potential effect of this SSC for enhancing germination and establishment of *S. acutus*, this study has several limitations. First, because we did not record the germination rate in the biomass experiment, we cannot know if the patterns we observed in the germination experiment were replicated in the biomass trial. As a consequence of not recording the exact number of seeds in each container in the biomass trial, we do not know if differences in

total (not per-seedling) biomass were due to explanatory variables or minor differences between containers in the number of seeds sown.

Another limitation of this study is the difference in seedling density among containers. Some containers had only one seedling in them, while several had more than 20 seedlings in them. In the containers with many seedlings, competition may have taken place and limited the growth of individual seedlings, even though total biomass in containers with many seedlings was high. Our method of pulling seedings based on age in the biomass trial is another limitation. We had to make subjective judgments based on height about the age of seedlings for individuals that germinated before we began marking newly germinated seedlings. For that reason, we introduced some uncertainty into the biomass results, given that other factors than age could have affected the size of seedlings.

Avenues of Future Research

This study is preliminary and did not replicate the more variable conditions, such as fluctuations in temperature, salinity, and water potential that would affect seeds and seedlings in wetlands. Therefore, further research is needed to determine if the possible benefits of this SSC persist in outdoor conditions. Future studies should use true wetland soil in laboratory or field experiments for a closer approximation of wetland conditions. Further research on a larger scale should also elucidate any interaction between geographic source of seed and the effect of the SSC. This concern is relevant because Kettenring (2016) found differential responses of *B. maritimus* to bleach dormancy-breaking treatments based on the geographic origin of seeds. Kettenring et al. (2016) also determined that a high degree of genetic diversity exists in *B. maritimus* populations, probably due to the role of birds in the dispersal of seeds. Further research should examine the possible effect of genotype and geographic origin on germination moisture requirements and seedling drought tolerance of all three bulrush species used in this study.

Other avenues of future research on this topic could investigate whether any possible benefit due to the SSC on germination proportion and seedling biomass persists in the presence of competition from invasive species such as *P. australis*. *P. australis* readily colonizes exposed mudflats that result from receding water levels and often competes directly with native species, including the three bulrushes considered here (Kettenring et al. 2016; Wilcox 2012). Increasing water availability via an SSC for native species could give native bulrushes a competitive advantage in the face of competition from *P. australis*. The SSC may confer another competitive

advantage to seeds given the fact that it increased the germination synchrony and reduced the time to 90% germination of the low-dose *S. acutus* seeds relative to control. Many invasive plant species emerge earlier than their native competitors (Schlaepfer et al. 2010; Van Kleunen and Johnson 2007), so reducing time to germination and increasing germination synchrony could allow *S. acutus* to germinate closer in time to invasive species and potentially have a leg up in the early growing season.

However, high nutrient concentrations (which are common in Great Salt Lake wetlands due to wastewater inputs) and sexual reproduction feedbacks based on genetic diversity can aid the spread of *P. australis* (Minchinton and Bertness 2003; Kettenring et al. 2011), so the magnitude of an SSC's effect in reducing the competitive advantage of *P. australis* warrants more study. Based on *P. australis*' life history and functional traits, the ideal situation for implementing an SSC technology would be a prepared seedbed that has been cleared of *P. australis* and in which the seed bank has reduced numbers of *P. australis* seed. Implementation of such a situation would require several years of intensive management per the recommendations of Rohal et al. (2017).

Finally, it remains unknown whether water availability is the only major constraint on the establishment of *B. maritimus*, *S. acutus*, and *S. americanus*, or whether other abiotic and biotic factors may constitute important demographic bottlenecks. The possible benefit of the low-dose coating relative to control and the high-dose coating may be due in part to the requirement of light for germination of *S. acutus* and *B. maritimus* (Marty and Kettenring 2017; Clevering 1995). A low-dose coating, which is thinner than the high-dose coating, may provide the benefit of the surfactant and allow light to reach the seeds.

Other constraints on the establishment of these species could include hydropatterns, as natural wetlands have characteristic hydroperiods that are subject to weather, climate, and human alterations to hydrology, and different hydropattern regimes are associated with specific vegetation communities (Foti et al. 2012). Further research should examine the effect of hydroperiods on the germination and establishment of *B. maritimus*, *S. acutus*, and *S. americanus* and account for the fact that Great Salt Lake levels peak in spring or early summer when these species germinate (Arnow and Stevens 1990), though climate change and human water use may reduce peak inflows from spring runoff (Downard et al. 2014). While the threats of climate change and water withdrawals are serious, managers have some flexibility in impounded

wetlands, as they can control the flow and depth of water in impoundments, though the total amount of water can limit that flexibility (Downard et al. 2014). Further research should examine what other bottlenecks may hinder bulrush establishment and the timing of water shortage bottlenecks in the establishment of these three species to put any potential benefit of the SSC into context.

Conclusion

Developing new technologies for wetland revegetation projects is an urgent research need to combat the effects of climate change, invasive species, and changes in hydrology. The data presented here offer preliminary evidence for a surfactant seed coating technology to enhance the germination and establishment of *S. acutus*, with less conclusive results for *B. maritimus* and *S. americanus*. In this study, we found that at the low dose of the SSC, *S. acutus* germination was significantly higher than control and the high-dose treatment. The low dose of the SSC also increased germination synchrony of the *S. acutus* seeds. Furthermore, we found that at the intermediate moisture level and with the low dose of the surfactant seed coating, per-seedling *S. acutus* aboveground and belowground biomass was significantly higher than control.

Coating seeds in a surfactant coating entails additional cost and time for revegetation projects. Therefore, future investigations should confirm the tentative evidence for a beneficial effect of this surfactant. Specifically, further research should (1) clarify the magnitude of the effect of surfactant seed coatings in aiding the restoration of these three species, (2) better replicate conditions that more closely approximate field conditions, (3) examine differences between populations from which experimental plant materials are taken, and (4) investigate whether the SSC is beneficial when bulrush seeds and seedlings face competition from invasive species.

Reflective Writing

Word Count: 1004

The experience of planning, implementing, and writing this capstone has taught me a variety of skills and expanded my academic horizons. I improved my scientific writing ability, my understanding of plant biology and ecology, and my ability to work with and manage people. This capstone experience has given me greater confidence in my abilities and prepared me for future professional and graduate school experiences.

Completing this project was indeed a capstone experience for my undergraduate education. It complemented and expanded on the information and skills I've learned in many classes, including Statistics for Scientists, Principles of Aquatic Ecosystem Restoration, General Ecology, and Wetland Ecology and Management. It was gratifying to have the opportunity to apply the knowledge I had gained from academic courses in a research project. This project broadened my knowledge of ecology because many of the classes I had taken were focused on terrestrial ecosystems, and this project allowed me to gain a deeper understanding of wetland ecology and watershed science in general. Much of that new knowledge came from my literature review when I was writing the capstone manuscript. I also gained valuable experience applying for grants from the College of Natural Resources to support my research. I have been able to present my research at the USU Student Research Symposium and will present it at the Society of Wetland Scientists annual meeting in Baltimore, Maryland at the end of May 2019. Taken together, all of these experiences broadened my academic experience at USU and have prepared me for graduate school and future jobs.

This project was exciting because it presented a potential avenue to increase the effectiveness of wetland restoration projects. I have always been interested in making the world a better place through improved stewardship of natural systems, and I carried that passion with me when I came to USU. Undergraduate research, and this capstone project specifically, have allowed me to contribute new knowledge to the field of ecology with implications for management activities. While the results presented here are preliminary and not totally conclusive, they show that this surfactant seed coating technology may benefit wetland plant seedlings and provide a foundation for further research. I hope others will carry on this line of research and help determine if this technology is a viable option for wetland reseeding projects in Great Salt Lake wetlands and beyond.

One of the most rewarding parts of this experience has been working with my faculty mentor, Dr. Karin Kettenring, and a doctoral student in her lab, Emily Martin. They both provided me with advice, encouragement, and technical expertise. I learned how to better work independently with their advice, and this experience will be a model for me as I prepare to pursue a graduate degree in 2020. Working with Dr. Kettenring also afforded me the opportunity to attend her weekly lab meetings. In those meetings, we discussed a variety of topics related to academic and professional development, work-life balance, and productivity. These meetings have enriched my educational experience at USU and broadened my horizons as a student and future professional.

Perhaps the greatest benefit of completing this project consisted of opportunities it gave me to confront unforeseen challenges and persevere to achieve a goal. I traveled to Brigham Young University (BYU) twice to coat bulrush seeds with Dr. Matthew Madsen before planting them in growth chambers at USU. The first time I went to BYU I went with Dr. Kettenring, and we realized we didn't have enough seeds to use the coating equipment. Working with Dr. Madsen, Dr. Kettenring and I figured out how to use filler seeds of other species to make sure we could coat the bulrush seeds effectively. During the second experiment, I realized that I should have started marking each seedling when it emerged after many had already done so. Working with my mentor, I had to devise a way to mark when seedlings had emerged before marking the newly emerging ones. These situations taught me the importance of planning ahead and of being able to come up with solutions when things don't go as planned.

I also learned how to better work with and organize other people as I worked on this project. I organized trips to BYU, and I worked with amazing technicians in the Kettenring Wetland Ecology Lab who assisted with planting seeds, marking seedlings, and harvesting biomass. After completing the laboratory portion of the project, I worked with Dr. Kettenring and Emily to analyze the data using R, SigmaPlot, and JMP software. I especially appreciated the opportunity to use R because I was taking a Geospatial Analysis with R course at the same time as I was analyzing the germination data, and I was able to practice my R skills in an applied way as part of this project.

For students thinking of joining Honors or are who are beginning their capstone projects, I would encourage you to find a topic that you're passionate about and that will make the world a better place. Don't wait to plan your project, and make sure your faculty mentor has enough time

to help you with carry out your project. Be prepared to write a lot, and view that task as an opportunity to improve your writing ability. Your Honors capstone experience will challenge you and lead to personal growth. Whether you pursue an academic career or not, completing an Honors capstone project will prepare you for future professional responsibilities and help you figure out what career path you want to pursue after you graduate.

I'm grateful for the opportunity to carry out this capstone project. I'm also thankful for my mentor, Emily Martin, the College of Natural Resources, and the USU Honors Program for helping me make it a reality. Research is one of the principal missions of Utah State University, and it has been a privilege to participate in it during my time here. I look forward to using what I've learned in this process in my future endeavors, and I will always remember this experience.

Works Cited

Abdi, H., & Williams, L. J. (2010). Tukey's honestly significant difference (HSD) test. *Encyclopedia of Research Design. Thousand Oaks, CA: Sage*, 1-5.

Arnow, T., & Stephens, D. W. (1990). Hydrologic characteristics of the Great Salt Lake, Utah, 1847-1986 (No. 2332). US Government Printing Office.

Baskin, C. C., & Baskin, J. M. (2014). Seeds: ecology, biogeography, and, evolution of dormancy and germination. Elsevier.

Clevering, O. A. (1995). Germination and seedling emergence of Scirpus lacustris L. and Scirpus maritimus L. with special reference to the restoration of wetlands. Aquatic Botany, 50(1), 63-78.

Downard, R., Endter-Wada, J., & Kettenring, K. M. (2014). Adaptive wetland management in an uncertain and changing arid environment. Ecology and Society, 19(2).

Evans, K. E., & Martinson, W. (2008). *Utah's featured birds and viewing sites: a conservation platform for IBAs and BHCAs*. Sun Litho.

Foti, R., del Jesus, M., Rinaldo, A., & Rodriguez-Iturbe, I. (2012). Hydroperiod regime controls the organization of plant species in wetlands. Proceedings of the National Academy of Sciences, 109(48), 19596-19600.

Fyfe, J. C., Derksen, C., Mudryk, L., Flato, G. M., Santer, B. D., Swart, N. C., . . . Jiao, Y. (2017). Large near-term projected snowpack loss over the western United States. Nature Communications, 8, 14996. doi:10.1038/ncomms14996

Gautam, R., & Ashwath, N. (2012). Hydrophobicity of 43 potting media: Its implications for raising seedlings in revegetation programs. Journal of Hydrology, 430, 111-117.

Karagatzides, J. D., & Hutchinson, I. (1991). Intraspecific comparisons of biomass dynamics in Scirpus americanus and Scirpus maritimus on the Fraser River Delta. The Journal of Ecology, 459-476.

Keddy, P. A., & Ellis, T. H. (1985). Seedling recruitment of 11 wetland plant species along a water level gradient: shared or distinct responses?. Canadian Journal of Botany, 63(10), 1876-1879

Kettenring, K. M. (2016). Viability, dormancy, germination, and intraspecific variation of Bolboschoenus maritimus (alkali bulrush) seeds. Aquatic Botany, 134, 26-30.

Kettenring, K. M., McCormick, M. K., Baron, H. M., & Whigham, D. F. (2011). Mechanisms of Phragmites australis invasion: feedbacks among genetic diversity, nutrients, and sexual reproduction. Journal of Applied Ecology, 48(5), 1305-1313.

Kettenring, K. M., Mock, K. E., Zaman, B., & McKee, M. (2016). Life on the edge: reproductive mode and rate of invasive Phragmites australis patch expansion. Biological Invasions, 18(9), 2475-2495.

Kettenring, K. M., Mossman, B. N., Downard, R., & Mock, K. E. (2018) Fine-scale genetic diversity and landscape-scale genetic structuring in three foundational bulrush species: implications for wetland revegetation. Restoration Ecology.

Labouriau, L. G. (1983). Uma nova linha de pesquisa na fisiologia da germinação das sementes. In Anais do XXXIV Congresso Nacional de Botânica. SBB, Porto Alegre (pp. 11-50).

Lakon, G. (1949). The topographical tetrazolium method for determining the germinating capacity of seeds. Plant Physiology, 24(3), 389.

Long, A. L., Kettenring, K. M., Hawkins, C. P., & Neale, C. M. (2017). Distribution and drivers of a widespread, invasive wetland grass, Phragmites australis, in wetlands of the Great Salt Lake, Utah, USA. Wetlands, 37(1), 45-5

Madsen, M. D., Davies, K. W., Boyd, C. S., Kerby, J. D., & Svejcar, T. J. (2016). Emerging seed enhancement technologies for overcoming barriers to restoration. Restoration Ecology, 24, S77-S84.

Madsen, M., Zvirzdin, D., Roundy, B., & Kostka, S. (2014). Improving reseeding success after catastrophic wildfire with surfactant seed coating technology. In Pesticide Formulation and Delivery Systems: 33rd Volume,"Sustainability: Contributions from Formulation Technology". ASTM International.

Madsen, M. D., Fidanza, M. A., Barney, N. S., Kostka, S. J., Badrakh, T., & McMillan, M. F. (2016). Low-dose Application of Nonionic Alkyl Terminated Block Copolymer Surfactant Enhances Turfgrass Seed Germination and Plant Growth. HortTechnology, 26(4), 379-385.

Marty, J. E., & Kettenring, K. M. (2017). Seed Dormancy Break and Germination for Restoration of Three Globally Important Wetland Bulrushes. Ecological Restoration, 35(2), 138-147.

Minchinton, T. E., & Bertness, M. D. (2003). Disturbance-mediated competition and the spread of Phragmites australis in a coastal marsh. Ecological Applications, 13(5), 1400-1416.

Nonogaki, H., Bassel, G. W., & Bewley, J. D. (2010). Germination—still a mystery. *Plant Science*, *179*(6), 574-581.

Ranal, M. A., & Santana, D. G. D. (2006). How and why to measure the germination process?. Brazilian Journal of Botany, 29(1), 1-11.

Rohal, C., Hambrecht, K., Cranney, C., & Kettenring, K. M. (2017). How to Restore Phragmites-Invaded Wetlands (Rep.). Retrieved February 24, 2019, from Utah Agricultural Experiment Station website: https://digitalcommons.usu.edu/cgi/viewcontent.cgi?article=1001&context=uaes_pu

Schlaepfer, D. R., Glättli, M., Fischer, M., & van Kleunen, M. (2010). A multi-species experiment in their native range indicates pre-adaptation of invasive alien plant species. New Phytologist, 185(4), 1087-1099.

Vanderlinder, M. S., Neale, C. M., Rosenberg, D. E., & Kettenring, K. M. (2013). Use of remote sensing to assess changes in wetland plant communities over an 18-year period: a case study from the Bear River Migratory Bird Refuge, Great Salt Lake, Utah. Western North American Naturalist, 74(1), 33-46.

Van Kleunen, M., & Johnson, S. D. (2007). South African Iridaceae with rapid and profuse seedling emergence are more likely to become naturalized in other regions. Journal of Ecology, 95(4), 674-681.

Wilcox, D. A. (2012). Response of wetland vegetation to the post-1986 decrease in Lake St. Clair water levels: seed-bank emergence and beginnings of the Phragmites australis invasion. Journal of Great Lakes Research, 38(2), 270-277.

Author Biography

Anders is graduating with a major in Conservation and Restoration Ecology and a minor in Watershed Science in the S.J. and Jessie E. Quinney College of Natural Resources. He is a USU Presidential Scholar, Undergraduate Research Fellow, USU Writing Fellow, recipient of the Quinney Scholarship, and the 2019 S.J. and Jessie E. Quinney College of Natural Resources valedictorian. He has completed three undergraduate research projects and presented results at many venues, including the National Conference on Undergraduate Research, Research on Capitol Hill at the Utah State Capitol, the USU Student Research Symposium, the USU Spring Runoff Conference, and the Great Salt Lake Issues Forum. He has participated in the Plant Identification competition at the 2016 and 2017 Society for Range Management annual meetings. Anders studied anthropological aspects of medicinal plants in Peruvian culture as part of a study abroad program in the summer of 2017. Following graduation, Anders will work for the Utah National Guard's Environmental Resources Management program. He plans to begin a graduate program in ecology or environmental planning in 2020.