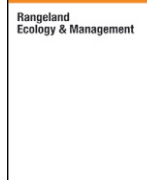




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Contents lists available at ScienceDirect

Rangeland Ecology & Management

journal homepage: <http://www.elsevier.com/locate/rama>

Germination and Seedling Emergence of Three Semiarid Western North American Legumes^{☆,☆☆}

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ARTICLE INFO

Article history:

Received 15 December 2014

Received in revised form 30 July 2015

Accepted 10 August 2015

Available online xxxxx

Keywords:

*Astragalus filipes**Dalea ornata**Dalea searlsiae*

revegetation

seed germination

seed scarification

ABSTRACT

Few seed sources of North American forbs are available for revegetation/restoration of degraded western rangelands adapted to annual precipitation zones less than 300 mm, and those that are available are mainly wildland collected. The amount of time and resources necessary to make wildland collections in quantity results in high seed prices and variable seed quality, such that forbs have been under-represented in rangeland seeding mixes. We have previously identified western prairie clover (*Dalea ornata* Douglas ex Hook.), Searls' prairie clover (*Dalea searlsiae* A. Gray), and basalt milkvetch (*Astragalus filipes* Torr. ex A. Gray) as native species adapted to low precipitation zones in the western United States for which field-grown seed production would potentially reduce seed costs and increase availability. A series of glasshouse experiments were conducted to determine the effects of scarification, planting depth, and soil composition on germination and seedling emergence of these species. All three species produce hard seeds, and scarification was necessary to increase germination and seedling emergence. Compared with a 6-mm planting depth, a planting depth of 19 mm retarded the rate of emergence for all species but only reduced the total seedling emergence for basalt milkvetch. With seed scarification in sandy soils, prairie clover seedling emergence exceeded 80% while basalt milkvetch was less than 10%. With seed scarification in soils with higher clay content, prairie clover total seedling emergence reduced to 58–70% while basalt milkvetch increased to approximately 30%. Along with enhancing stand establishment in seed production fields, these data will assist land managers in planning for optimal establishment of these species in rangeland revegetation/restoration projects.

Published by Elsevier B.V.

Introduction

Western rangelands in semiarid climates are exposed to periodic disturbances, such as wildfires, that often require revegetation to reestablish plant communities, avoid soil loss, deter the spread of exotic weeds, prevent impairment of ecosystem structure and function, and mitigate significant threats to human health, safety, life, and property (USDA-FS, 2000). Grasses typically comprise the core of these revegetation/restoration efforts, and seed growers have been successful at producing large volumes of high-quality seed for many grass species. Other plant species, especially native forbs, are disproportionately under-represented in revegetation/restoration projects, both in the number of species and in the volume of seed available for each species (Shaw et al., 2005; USDI-PCA, 2015).

Establishing indigenous forbs during revegetation/restoration is central to achieving diverse systems that function sustainably and resist invasion of exotic weeds (Pokorny et al., 2004; Cane, 2011).

Although federal land management agencies are encouraged to use native North American species for restoration and rehabilitation to the extent practical (USDI-PCA, 2015), seed cost and availability have limited their use (Richards et al., 1998). In particular, few seed sources of forbs native to the semiarid western regions of the United States are available for revegetation/restoration of degraded western rangelands that are adapted to annual precipitation zones < 300 mm. These forbs are often available as wildland-collected seeds rather than field-grown seeds, and many forb species are indeterminate, shatter during seed ripening, and are difficult to establish such that seed production is problematic and costly. The amount of time and resources necessary to make wildland collections in quantity results in relatively high seed prices and variable seed quality for for species. As a result, there is a need to identify forbs from western North America that are adapted to low precipitation zones and amenable to field seed production so that these species can be more fully utilized in revegetation/restoration efforts (USDA-FS and USDI-BLM, 2014).

Three such species are western prairie clover (*Dalea ornata* [Douglas ex Hook.] Eaton & J. Wright), Searls' prairie clover (*Dalea searlsiae* [A. Gray] Barneby), and basalt milkvetch (*Astragalus filipes* Torr. ex

[☆] Research was partially funded by the Great Basin Native Plant Project through the Department of the Interior/Bureau of Land Management Great Basin Restoration Initiative and the USDA Forest Service Rocky Mountain Research Station.

^{☆☆} Mention of a proprietary product does not constitute a guarantee or warranty of the product by USDA or the authors and does not imply its approval to the exclusion of other products that may also be suitable.

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A. Gray). These three forb species are legumes that biologically fix nitrogen in association with rhizobial symbionts, have distributions across the semiarid western United States, and are adapted to sites with 200 to 600 mm annual precipitation (Bhattarai et al., 2008, 2010, 2011). Additionally, they produce numerous inflorescences at a height sufficient for mechanical harvest. Collections of these three species were made previously, and prevaryety germplasm sources have been released for western prairie clover and basalt milkvetch (Johnson et al., 2008, 2011). Establishing stands of these species for field seed production, however, has been challenging due to poor germination and seedling emergence.

Additional research of these species is needed to improve establishment in seed production fields, as well as on rangeland sites. Although data specific to these species are lacking, studies from other species in the same genera have highlighted the presence of hard seed coats and the possible benefits of seed scarification (Eisvand et al., 2006; Rawlins et al., 2009; Molano-Flores et al., 2011). We hypothesized that seed scarification and a shallow planting depth would improve germination and emergence of western prairie clover, Searls' prairie clover, and basalt milkvetch. In addition, we hypothesized that planting in soils with a high sand content would increase the rate of germination and emergence in these three leguminous species. Our results evaluated germination and seedling emergence responses of these three species when exposed to seed scarification methods, planting depths, and soil compositions.

Methods

Germplasm Sources

Two glasshouse experiments were conducted using Majestic and Spectrum western prairie clover germplasms (Johnson et al., 2011), Ds-23 and Ds-26 Searls' prairie clover collections (Bhattarai et al., 2011), and NBR-1 basalt milkvetch germplasm (Johnson et al., 2008; Bushman et al., 2010). Seed of Majestic and Spectrum for Experiment 1 were produced at BFI Native Seeds (Moses Lake, Washington), whereas in Experiment 2 seed of Majestic originated from its original wildland collection site near Madras, Oregon (lat 45°17'N, long 121°01'W, elevation 258 m). Seed of Ds-26 was produced at the USDA-NRCS Plant Materials Center at Aberdeen, Idaho. Seeds of Ds-23 and NBR-1 were produced at the Millville Research Farm (lat 41°39'N, long 111°48'W, elevation 1350m) in 2010. Seed from each germplasm source (except Ds-26) was hand-threshed and cleaned at the USDA-ARS Forage and Range Research Laboratory; Ds-26 was threshed and cleaned at the USDA-NRCS Plant Materials Center at Aberdeen, Idaho.

Seeds were tested for viability and germination at the Utah State Seed Laboratory (Salt Lake City, UT). Viability was assayed using tetrazolium staining on 100 seeds from each entry, and total viable seeds were the sum of viable and viable-hard seeds. Seeds that imbibed and stained with tetrazolium were considered viable while seeds that required nick scarification to imbibe and stain were considered viable-hard. This test was conducted for nonscarified and previously scarified seeds. Germination was separately measured on four replications of 78 seeds of each entry, where seeds that germinated within 21 days were summed together. Seedling emergence data in the two glasshouse experiments were adjusted on the basis of total viable seed.

Experiment 1

The purpose of Experiment 1 was to test the effects of seed scarification methods and planting depths. The seeds were planted into sand-filled benches with a soil medium composed of one part peat moss to four parts Kidman fine sandy loam (coarse-loamy, mixed, mesic Calcic Haploxeroll) that was steam treated at 71°C for 2.5 hours. Seeds were placed on the sand surface in 43-cm rows utilizing hardboard templates and covered to either 6- or 19-mm depths with sifted sand. Fifty seeds

were planted for each of the five entries (Majestic, Spectrum, Ds-23, Ds-26, and NBR-1), and four replications of each entry were planted in a randomized complete block design in a glasshouse at Logan, Utah. Two separate runs of the experiment were conducted during March and April 2011, respectively, with each run continuing for 28 days after planting.

Three seed treatments were tested: acid scarification, mechanical scarification, and untreated control. For acid scarification, the seeds were soaked in 98% sulfuric acid for 5 minutes, rinsed in tapwater for 3 minutes, and air dried. For mechanical scarification, the seeds were hand rubbed between two pieces of 120-grit sand paper for 30 seconds in small batches. Glasshouse air temperatures were 25:20°C day:night with a 12-h day length maintained with supplemental high-pressure sodium lamps that provided approximately 500 μmol photons m⁻²s⁻¹ daily photosynthetically active radiation. The plots were watered to saturation three times a week using a fine mist. Seedlings were counted as "germinated" as soon as cotyledons, seed coat, or hypocotyls were visible. If the soil surface was pushed up or cracked open by a seedling, but the seedling was not visible, it was not counted as emerged. The number of seedlings that emerged was counted three times a week for the duration of each run. Total emergence (TEM) was determined as the number of seedlings that emerged by the end of the experiment. The rate of emergence (RATE) was calculated as described by Maguire (1962):

$$\text{RATE} = E_1/D_1 + (E_2 - E_1)/D_2 + \dots + (E_n - E_{n-1})/D_n \quad (1)$$

where E is the number of seedlings emerged on a counting date and D is the number of days since planting.

Experiment 2

The purpose of Experiment 2 was to confirm the effects of scarification and test the effects of soil composition on seedling emergence. Three entries were included in this experiment: Majestic western prairie clover, Ds-23 Searls' prairie clover, and NBR-1 basalt milkvetch. Fifty seeds were planted for each entry in a replicated split-plot design with soil compositions as whole plots and seed treatments as subplots. Experiments were conducted in the same glasshouse as Experiment 1, and two separate runs of the experiment were conducted in March and April 2012. Seeds were planted at a 6-mm soil depth. Two soil types were used: Kidman fine sandy loam (FSL, as described earlier) and Nibley silty clay loam (SCL, fine, mixed, active, mesic Aquic Argixerolls). These two soils were partitioned into 54 × 54 cm whole plots as follows: 100% FSL, 33% SCL/67% FSL, 67% SCL/33% FSL, and 100% SCL, which from here on will be referred to as FSL (sand), 33-SCL, 67-SCL, and SCL (clay), respectively. Each plot was watered every 6 days with 4 L of tap water, saturating the soil for each plot. Seed treatments within each whole plot included acid-scarified seed (as described in Experiment 1) and untreated seed. Seedling TEM and RATE were determined as described in Experiment 1.

Statistical Analyses

Analysis of variance was assessed using the MIXED procedure of SAS (SAS Institute, 2004), where TEM and RATE were dependent variables. Entry, seed treatment, and planting depth or soil composition were considered fixed effects, while replication, run, rep by depth interaction (Experiment 1), and rep by soil type interaction (Experiment 2) were considered random effects. Residuals were checked for normality and homoscedasticity using Shapiro-Wilk and Kolmogorov-Smirnov tests implemented in the Univariate and Transreg procedures of SAS. In Experiment 2 a natural log transformation was necessitated to normalize the data, and means were back-transformed for presentation. The Tukey-Kramer experiment wise test at a $P < 0.05$ level of significance was used to determine significance of mean separations, and alphabetical

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