Germination and Development of Two Inland Northwest Native Plants

A Thesis Presented in Partial Fulfillment of the Requirements for the Degree of Master of Science with a Major in Natural Resources

> In the College of Graduate Studies by

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Authorization to Submit Thesis

This thesis of Rebecca A. Sheridan, submitted for the degree of Master of Science with a Major in Natural Resources and titled "Germination and Development of Two Inland Northwest Native Plants," has been reviewed in final form. Permission, as indicated by the signatures and dates below, is now granted to submit final copies to the College of Graduate Studies for approval.

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Abstract

Native plant and forest nurseries must produce high-quality seedlings while facing changing environmental regulations and climatic conditions. Nursery growers continually test and adapt methods of seedling production to ensure consistent, sustainable outcomes. In Chapter 1, we tested the germination and growth of redosier dogwood in a subirrigation system. During germination, we compared treatments with and without overhead misting and germination cloth. We also tested two fertilizers applied in two ways— incorporated or top dressed. In Chapter 2, we modified environmental conditions of direct-seeded western redcedar: we altered (1) light with wire hardware cloth and (2) soil moisture with two types of mulch or no mulch. These experiment show that nursery cultural practices can be manipulated to improve germination rates, to reduce overall water and fertilizer use, and to adjust growth rates in the nursery and the field. Additional environmental factors and establishment strategies need to be considered for successful establishment.

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Chapter 1: Native Plant Germination and Growth in a Subirrigation System

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Abstract

Native plant and forest nurseries consume high amounts of water when irrigating crops with overhead and hand-watering systems. As water conservation continues to be an issue, subirrigation is being considered as an alternative watering method by growers and nursery owners. We tested the germination and growth of redosier dogwood (*Cornus sericea*) in a subirrigation system. During germination, we compared treatments with and without overhead misting and germination cloth in addition to subirrigation. We also tested two fertilizers-Osmocote[®] Pro 17- 5-11 and Nutri-Rich 8-2-4-applied in two waysincorporated or top dressed. Results showed that germination was successful using subirrigation only, but germination was highest in treatments that also had germination cloth and received overhead misting twice a day. The treatments with incorporated Osmocote® grew more in the nursery, but the treatments with top-dressed Osmocote[®] grew taller after outplanting. The Nutri-Rich fertilizer did not work in this experiment because of a pest infestation. The experiment showed that subirrigation can be used to successfully germinate seed and that nursery cultural practices can be manipulated to improve germination rates, to reduce overall water and fertilizer use, and to adjust growth rates in the nursery and the field. This paper was presented at the annual meeting of the Western Forest and Conservation Nursery Association (Eugene, OR, October 26–27, 2015).

Introduction

Forest tree and native plant nurseries grow seedlings for reforestation and restoration projects. Given that the seedlings are typically planted in projects that have environmental objectives, nursery growers want to ensure that the plants are grown in a sustainable manner. Growing seedlings in a nursery requires resources, including water. Developing more waterefficient practices to grow plants can reduce the nursery's water use (Landis 1989).

Subirrigation, or ebb-and-flood irrigation, is one system used in container nurseries to irrigate plants. In a subirrigation system, containers sit in a tray or reservoir, which is then flooded allowing water to enter into holes in the bottom of the containers. After a period of soaking, any unabsorbed water is drained. The volume of water movement is a balance between growing medium porosity and bulk density, container configuration, and water requirements of the plants (Ferrarezi et al. 2015). Subirrigation applies water directly to the medium, resulting in a higher water-use efficiency compared with overhead irrigation (Gent and McAvoy 2011). Holding tanks can be used to store the unabsorbed water for reuse. Subirrigation systems have been used to grow healthy seedlings while reducing overall water and fertilizer use and decreasing weeds (Bumgarner et al. 2008, Davis et al. 2011, Wilen et al. 1999); in these experiments, the seedlings were either transplanted into the containers or grown with overhead irrigation during the germination phase.

As growers' repertoires expand to include growing a variety species of using subirrigation, it becomes necessary to identify the most effective way to optimize water use. At the same time, growers need to determine best practices and successful protocols to produce healthy seedlings of each species (Schmal et al. 2011). Nurseries that currently use subirrigation to grow seedlings still use overhead irrigation during the germination phase, which requires additional infrastructure (e.g., Dumroese et al. 2011). If subirrigation systems are to be used independent of overhead irrigation infrastructure, subirrigation must meet the water needs of the plant throughout the growing season. Therefore, the irrigation system and the growing medium must provide sufficient water to the seed or seedling during each growing stage, particularly the germination phase. To properly grow high quality seedlings, it is likely that nursery cultural treatments, such as irrigation method and fertilizer application, may need to be adjusted.

The objective of our study was to determine whether it is possible to germinate redosier dogwood (*Cornus sericea* L.) seed in a subirrigation system and if fertilizer type and application method affect the growth of this species during its time in the nursery and its first year after planting. We also examined whether different nursery cultural practices affect germination.

Redosier dogwood is an appropriate species for this experiment. The species is often

found along margins of streams and wetlands, where the soils are saturated for a portion of the growing season but are dry by late summer (Stevens and Dozier 2000). In addition, redosier dogwood is a popular choice for landscaping in the Pacific Northwest, with many nurseries and growers in the area producing it often and in large quantities. The species develops a broad canopy that deflects water when overhead irrigation is used. Using subirrigation can ensure that each container receives the water it needs (Davis et al. 2008, Landis and Wilkinson 2004).

Material and Methods

The experiment was conducted at Oxbow Native Plant Nursery, located within the Oxbow Farm and Conservation Center in Carnation, WA (~47°41'N., 121°58'W.). Seed for this study was obtained in northern Idaho and was stratified for 136 days at 0 to 1.5 °C (32 to 35 °F) before sowing. Seed viability was tested by placing seed in a closed, clear plastic container under a full-spectrum light and recording germination for 4 weeks. The light was on an automatic timer for 16 hours of daylight and 8 hours of darkness. The seed were misted three times a day to keep them moist and were kept under ambient temperature, which was tracked with a temperature-humidity sensor (Decagon Devices, Inc., Pullman, WA). A seed was counted as germinated when 5 mm (0.2 in) of the radicle was visible (Baskin and Baskin 2014). Mean germination was 86 percent (n = 4, standard deviation = 3.63, descriptive statistics from R, version 3.1.1).

The greenhouse at the Oxbow Native Plant Nursery is a Cravo greenhouse (Cravo Equipment Ltd., Brandtford, ON, Canada) and regulates temperature automatically by opening and closing roof and side panels; there is no supplemental heating, cooling, or lighting in the greenhouse. For this experiment, Ray-Leach SC10 containers (Stuewe & Sons, Inc., Tangent, OR) were filled with Sunshine Mix #4 Aggregate Plus (Sun Gro® Horticulture, Agawam, MA, Lot S13-153). The medium was made of 65 to 75 percent Canadian Sphagnum peat moss; the remaining proportion was horticultural grade perlite and dolomitic limestone. The Ray-Leach cells were placed in racks that held 98 containers each. A total of 2,800 containers were used in the study, arranged in a split plot design with five replications. On June 11, 2014, three seeds were sown in each container. The seeds were then covered with approximately 0.5 cm (0.20 in) of medium.

Subirrigation

The subirrigation system consisted of cement mixing tubs measuring 0.61 x 0.91 x 0.20 m (24 in x 36 in x 8 in). Before sowing, all tubs were filled with 50 L (13 gal) of water and the trays were soaked for 2 hours. At this point, the containers were saturated, except for the unconsolidated peat at the top of the container. Any remaining water in each tub was drained into a 70-L (18-gal) container (Rubbermaid, Atlanta, GA) between subirrigations. The irrigation water was recycled for each irrigation, with a separate supply maintained for each subirrigation tub. Subsequent irrigations soaked for 1 hour (germination phase) or 15 minutes (growth phase). For each irrigation event, the supply was topped off to 50 L (13 gal) with fresh water from the greenhouse water supply. In the third replication, a temperature and humidity sensor (Decagon Devices, Pullman, WA) was installed in each irrigation treatment.

Fertilizer Treatments

Two nursery fertilizers were used: (1) Osmocote[®] Pro Control Release 17-5-11 NPK (3-4 month release, Everris, Geldermalsen, Netherlands) and (2) Nutri-Rich 8-2-4 NPK (Stutzman Environmental Products, Inc., Canby, OR). The Osmocote[®] product is a general purpose fertilizer made of coated prills and is acceptable for use in containerized nurseries. The Nutri-Rich product is an organic-certified, granulized product made primarily of chicken manure. The advertised applications of Nutri-Rich include use on trees and shrubs. Each fertilizer was applied in one of two ways: either (1) incorporated into the growing medium or (2) applied as a top dressing. The rate of fertilization was determined by the medium recommended application rate by Osmocote® (Dumroese et al. 2007). The amount of Nutri-Rich was adjusted to provide the same amount of nitrogen (N) as the Osmocote[®]. For the incorporated treatment, 96.36 g (3.40 oz) Osmocote[®] or 394.70 g (13.92 oz) Nutri-Rich were mixed into the medium before filling 140 containers. For the top-dressed treatment, 0.688 g (0.024 oz) of Osmocote[®] or 2.82 g (0.099 oz) of Nutri-Rich fertilizer were applied to the top of each container before sowing. In the top-dressed containers, care was taken to keep the seeds from contacting the fertilizer, though the higher top-dressing rate of Nutri-Rich fertilizer made this difficult. One-and-a-half container racks were placed in each of 20 subirrigation tubs (figure 1.1). From each of the four fertilizer treatments, 35 containers were put in a tub,

for a total of 140 containers per tub.

Germination Treatments

Four germination treatments were used during the emergence phase (first 6 weeks after sowing): (1) unmisted and uncovered, (2) unmisted and covered with a germination cloth, (3) overhead misted and uncovered, and (4) overhead misted and covered with a germination cloth. The germination cloth was 0.5 oz Plant and Seed Guard (DeWitt Company, Sikeston, MO), a lightweight, white fabric. The treatments that were overhead misted were misted twice a day, in the morning and evening. All treatments were subirrigated every other day in the morning during the emergence phase. Each germination treatment was applied to five subirrigation tubs (whole plots in a split plot design).

Emergence Phase

Following sowing, redosier dogwood seeds were tracked twice daily for emergence. A seed was considered to have emerged when its cotyledons fully cleared the surface of the medium (figure 1.2). When a seed emerged, it was marked with a ball-point pin; a different color was used for the first, second, and third seed to emerge in each container. Redosier dogwood seed is technically classified as a stone containing two embryos. Therefore, each of the seeds planted could potentially produce two seedlings. If two germinants emerged in close proximity or had visibly emerged from the same seed coat, they were classified as seedlings from the same stone. Emergence was scored by container, where at least one seedling had to emerge in a container for it to be counted as having had successful emergence. Emergence was tracked for 5 weeks after the first seedling emerged. If a seedling died during the emergence phase, it was removed, its place was marked with an additional pin, and a possible cause of death was recorded. At the end of the emergence phase, the quality of each remaining seedling was noted.

Growth Phase

After the 6-week emergence phase, each container was thinned to one seedling per cell and the reservoirs of irrigation water were emptied and refilled. The containers, originally organized within the tubs by germination treatment, were reorganized so that the same fertilizer treatments were grouped within the same subirrigation tub. This reorganization meant that seedlings would be exposed to only the assigned fertilizer type if the fertilizer leached into the recycled irrigation water during the experiment. The original plot and subplot identities, however, were tracked through the rest of the experiment.

During the growth phase, the irrigation schedule was based on gravimetric weights, in which the containers were allowed to dry to 80 percent of field capacity during August, then to 70 percent during September and October. The pH and electrical conductivity (EC—a proxy measurement for available fertilizer) of the irrigation water were measured weekly to monitor whether the water stayed within safe ranges for seedlings. The seedlings were not pruned. At the end of August, the seedlings were moved from the Oxbow Native Plant Nursery to the Franklin H. Pitkin Forest Nursery in Moscow, ID (46°43'N., 116°57'W.) and kept outside. Samples were taken from the recycled irrigation water in August before the move and again in October at the end of the growing season and were tested for nutrients. In addition to the scheduled irrigation events, the seedlings received 2.64 cm (1.04 in) of rain during the 2 months they were held outside at the Pitkin Forest Nursery before outplanting.

Outplanting

Seedlings were outplanted in the last week of October 2014 to a relatively flat, tilled agricultural field at the Pitkin Forest Nursery, with coarse loamy soil. No additional experimental treatments or irrigation were applied when the seedlings were planted. During the week after planting, 1.88 cm (0.73 in) of rain fell. Seedling survival was recorded in May 2015. Seedlings were considered dead if they failed to leaf out, if leaves were fully desiccated, or if the seedling was missing entirely. Seedling root collar diameter and height were measured on all outplanted seedlings in November 2014, when the seedlings' leaves had turned red and were beginning to senesce, and again in July 2015, at which time growth was ceasing due to the seasonal summer drought. Field growth was calculated as the difference in height and root collar diameter from the time of outplanting to the final measurements in July.

Statistical Methods

Statistical analyses were done in R, version 3.1.1. The experiment was a split-plot design, in which the whole plot level (germination treatment) was a randomized complete block design. There were five blocks consisting of four irrigation tubs grouped on a table. The

subplot level (fertilizer treatments) was also a randomized complete block design. The two phases of this experiment—(1) the emergence phase and (2) the growth and out- planting phase—were analyzed separately. Data collected during the emergence phase were subject to analysis of variance (ANOVA) to test the effects of fertilizer type and germination treatment on emergence. During the growth and outplanting phase, data were analyzed using a multivariate analysis of variance (MANOVA) with a Pillai's trace test to test the effects of fertilizer type and irrigation method on height and root collar growth. MANOVA was used because height and root collar diameter are dependent variables on the same experimental unit, a seedling. Significance was determined at the $\alpha \le 0.05$ level. The model assumptions of normality and constant variance were evaluated using diagnostic plots, and the assumptions were determined to hold, with no data transformations deemed necessary.

Results

Emergence

In the treatments with Nutri-Rich fertilizer, very few seedlings emerged, which appeared to be due to a fungus gnat infestation, in which the larvae ate germinating seed before seedlings emerged. Therefore, those data were eliminated from the study. Interaction between the Osmocote[®] fertilizer treatments and germination treatment was significant (p = 0.04) (figure 1.3). Emergence was greater in treatments with overhead misting (p < 0.001). The treatments without overhead misting trended toward higher emergence in seedlings with top-dressed fertilizer than those with incorporated fertilizer (p = 0.06).

Growth

Measurements of height and root collar diameter in November 2014 accounted for seedling growth through their time in the nursery. A significant interaction occurred between the germination and fertilizer treatments (p = 0.02). Seedlings with incorporated Osmocote[®] fertilizer were taller than seedlings with top-dressed fertilizer (figures 1.4 and 1.5). The seedlings with incorporated Osmocote[®] also had larger root collar diameters (data not shown). After planting, some seedlings suffered from herbivory, presumably by rabbits (Riley 2014), and some experienced frost heave; however, most were able to persist and grow in spite of

these challenges. Redosier dogwood has the ability to resprout, and this growth pattern was observed in some cases in the field.

Seedling growth after outplanting was significantly affected by fertilizer type (p < 0.001) and germination treatment (p < 0.02), with no significant interaction between the two treatments. Seedlings with top-dressed fertilizer had greater height and root collar diameter growth than did those with incorporated fertilizer (table 1).

Discussion

The first phase of this experiment showed that it is possible to germinate seed using subirrigation only, but the emergence rates were lower in this treatment compared with those that also received overhead misting. The emergence phase has been identified as a challenge to adoption of subirrigation in nurseries (Dumroese et al. 2007). Even without sufficient water, seed may still germinate, but the seed will be more vulnerable to disease and decay, and germination will be less uniform (Bewley and Black 1994). Nursery growers work to avoid these situations. Alternative nursery cultural techniques, such as a grit layer on the top of containers, could also be used in conjunction with subirrigation to create favorable germination conditions. As nursery growers make decisions about propagation protocols using subirrigation, they will need to consider the natural history of the species with which they are working (Schmal et al. 2011). The redosier dogwood seed used in this experiment is relatively large, and the species is a wetland plant. The size and germination characteristics of other species might affect their suitability for use in a subirrigation system.

The treatments with Nutri-Rich fertilizer did not produce many seedlings. Although fungus gnat larvae were seen in all the treatment types, a greater number of larvae were observed in the containers with Nutri-Rich. It is unknown whether this condition was a direct consequence of the fertilizer, or if it was due to changes in the physical characteristics of the medium resulting from the fertilizer. In another study, Nutri-Rich fertilizer increased the medium's water-holding capacity in a subirrigation system (Dunlap 2015). Fungus gnats were also a problem in a previous subirrigation study, and reducing irrigation frequency helped address the issue (Dumroese et al. 2006).

In subirrigation systems, N from the controlled-release fertilizer is primarily retained within the medium and plant, and little N is lost in runoff water (Morvant et al. 2001, Pinto et

al. 2008). EC is higher at the top of containers that are subirrigated when growing a species that does not have fibrous roots in the upper layer of medium (Davis et al. 2008). By contrast, subirrigated containers that are planted with a species that has shallow, fibrous roots do not show elevated EC in the upper medium (Pinto et al. 2008). In this experiment, the seedlings were not observed to have numerous shallow roots, and the seedlings grown with incorporated fertilizer probably had better access to the fertilizer while growing in the nursery. Fertilizer that is retained within the media is available to the plant for use after outplanting (Dumroese et al. 2006, 2011), which may explain why we observed greater growth of the top-dressed seedlings after outplanting. The top-dressed fertilizer, which stays relatively dry in a subirrigation system compared with the incorporated fertilizer, may have broken down more slowly and, therefore, may have been available to the seedling in greater amounts after outplanting. Improving seedling quality or outplanting success with fertilizer has its limits, and extremely high rates of fertilization can negatively impact seedling quality and survival (Bumgarner et al. 2015), especially on dry sites. The germination treatments also had significant effects on growth after outplanting, which demonstrates the importance of following seedlings through outplanting to determine if nursery cultural practices continue to affect seedlings after outplanting (Davis et al. 2011).

This experiment demonstrated that it is possible for seed to germinate using subirrigation. This finding leads to further questions about how to improve germination and what options are best for fertilizing seedlings in subirrigation. Subirrigation will not be the irrigation method of choice for every nursery or every species, but it is an important tool that nursery growers can consider among their propagation options.



Figure 1.1 Large cement mixing trays were used to subirrigate seedlings grown in Ray-Leach containers. One-and-a-half racks of Ray-Leach containers fit in the mixing tray. The tray had a plug at the bottom to facilitate draining irrigation water into a storage tub. The containers covered with germination cloth are visible in the background. (Photo by Rebecca Sheridan, 2014)



Figure 1.2. Newly emerged seedlings were marked with a ballpoint pin when the cotyledons cleared the media surface. At this time, the date of emergence was recorded. (Photo by Rebecca Sheridan, 2014)



Figure 1.3. Seedling emergence was tracked by irrigation and fertilization treatment. The highest rates of emergence occurred in treatments that received overhead irrigation in addition to subirrigation. A statistically significant interaction (p < 0.05) occurred between the irrigation type and the fertilization method. The bars show standard error for five replications.



Figure 1.4. The seedlings were germinated and grown using subirrigation. In this photo, seedlings on the left were grown with top-dressed Osmocote[®] fertilizer and those on the right were grown with incorporated Osmocote[®] fertilizer. By November 2014, the seedlings with incorporated fertilizer were significantly taller than the seedlings with top-dressed fertilizer. (Photo by Rebecca Sheridan, 2014)



Figure 1.5. Seedling height after the nursery growing season (measured in November 2014, after the seedlings had dropped their leaves). Seedlings grown with incorporated Osmocote[®] fertilizer were taller than those with top-dressed fertilizer (p<0.001). The bars show standard error for five replications.

Table 1.1. Fertilizer treatment (p < 0.001) and germination treatment (p < 0.02) had a significant effect on height and root collar diameter growth.

Treatment	Height growth (cm) and (standard error)	Root collar diameter growth (mm) and (standard error)			
	Osmocote® incorporated				
Subirrigation only	15.9 (4.0)	1.57 (0.51)			
Germination cloth	21.5 (5.4)	2.03 (0.31)			
Overhead misting	18.2 (4.0)	1.73 (0.40)			
Germination cloth and overhead misting	15.1 (3.6)	1.78 (0.38)			
Osmocote [®] top dressed					
Subirrigation only	18.9 (4.2)	1.78 (0.43)			
Germination cloth	20.4 (4.0)	2.04 (0.43)			
Overhead misting	22.5 (5.0)	1.97 (0.46)			
Germination cloth and overhead misting	20.0 (3.8)	1.95 (0.40)			

cm = centimeter. mm = millimeter.

Note: The standard errors are for five replications.

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Chapter 2: Mulching and Shade Effects on Emergence and Survival of Direct-Seeded Western Redcedar (*Thuja plicata*)

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Abstract

Western redcedar (*Thuja plicata* Donn ex D. Don) is an important forest species valued by foresters for its timber value and by the public for its beauty. Regeneration of this tree species is threatened by difficulties in plantation establishment and by predicted climate change. Western redcedar is one of the most shade-tolerant species in northwestern forests, but regeneration requires sufficient light and moisture. Previous attempts at direct seeding the species have been mostly unsuccessful. We modified environmental conditions of directseeded western redcedar in two ways: we altered (1) light with wire hard-ware cloth and (2) soil moisture with one of two types of mulch or no mulch. The treatment without mulch had significantly higher emergence, but seedlings in all treatments did not survive through the first season. Additional environmental factors and establishment strategies need to be considered for successful direct seeding of western redcedar.

Introduction

Western redcedar (*Thuja plicata* Donn ex D. Don) is an ecologically important and economically versatile species. The species grows in a variety of forest types and provides habitat and browse for animals (Minore 1990). Western redcedar has long been an important timber species (Haig et al. 1941); the wood is workable and durable, making it useful in a wide variety of applications, from roofing shingles to decorative chests (Nystrom et al. 1984, Minore 1990). Western redcedar is also valued for its beauty by the general public (Sharpe 1974). Despite this species' environmental, economical, and aesthetic value, establishing plantations or managing natural stands to increase the number of western redcedar trees can be challenging (Nystrom et al. 1984). In addition, predicted climate changes will shift the region of suitable growing conditions for western redcedar, which will require careful consideration of replanting schemes involving this species (Hebda 2009). These changes will force foresters to plan for a dynamic context and may require assisted migration of some species (Williams and Dumroese 2014).

Western redcedar is found on the Pacific Coast and in the Inland Northwest, with little overlap between the two ranges. In the Inland Northwest, the species grows from lat. 54°30' N. in British Columbia and south into Montana and northern Idaho (Minore 1990). Along the coast, its range extends farther south into California (lat. 40°10' N.) and north into southeast Alaska (lat. 56°30' N.). In the central part of its Pacific range, the species grows inland as far as the western slopes of the Cascades (Minore 1990). Western redcedar is distributed across a range of environmental conditions but grows best on moist, humid sites (Fan et al. 2008), such as in stream bottoms, moist flats, and north-facing slopes (Brand and Schopmeyer 2008). Precipitation within the coastal range for western redcedar ranges from 890 mm to 6,600 mm (35 in to 260 in), mostly as winter rain; the interior range receives 710 to 1,240 mm (28 in to 49 in) annual precipitation, as snow and rain (Minore 1990). Western redcedar is one of the most shade-tolerant species in northwestern forests (Coates and Burton 1999, Ferguson et al. 1986) and can grow on a variety of soils across a range of elevations (Brand and Schopmeyer 2008), although sedimentary bedrock can increase mortality (Moore et al. 2004). Western redcedar does not commonly grow in pure stands but grows readily within mixed stands (Sharpe 1974).

Western redcedar is present in all stages of forest succession (McKenzie and Tinker 2013), but natural regeneration depends on well-disturbed mineral soil and canopy gaps in established stands (Clark 1970, Gray and Spies 1996). Remnant individuals in old-growth stands provide sources of seed for regeneration (Keeton and Franklin 2005). Western redcedar can be a prolific, although erratic, seed producer (Gashwiler 1970, Minore 1990). Survival of seed through its first winter can exceed 90 percent (Gashwiler 1967). The seed has low survival in storage for 3 months at 2.0 °C (35.6 °F), however, suggesting that naturally dispersed seed will not be viable for more than one season (Terskikh et al. 2008). Western redcedar seed is less susceptible to predation than other, larger conifer seeds (Gashwiler 1970). The seed may be less palatable because of its pungent odor (Gashwiler 1967).

Vegetative reproduction can also occur in some stands (Parker 1986).

Understanding the conditions under which western redcedar regenerates requires consideration of both the establishment phase and the growth phase (Ferguson et al. 1986). Natural regeneration can occur on disturbed areas, indicating that western redcedar is exposure tolerant (Wang et al. 1994). Initial seedling survival, however, requires a balance between light and moisture (Carter and Klinka 1992). Mortality of naturally regenerating seed can be high soon after peak emergence, but, after September, additional losses are minimal (Gashwiler 1971). The seedling first grows primary needle leaves before growing secondary, scale-like foliage, which may correspond to decreased mortality later in the growing season (Weber et al. 2003). If seedlings establish in full sunlight, abundant moisture is required for survival (Weber et al. 2003). Conversely, western redcedar seedlings exhibit greater shade tolerance on sites of low water availability (Harrington 2006). High temperatures, drought, and frost-heaving are major causes of seedling mortality (Brand and Schopmeyer 2008, Gashwiler 1971, Soos and Walters 1963).

Some western redcedar seedlings can survive at 10 percent of full sunlight, but seedling mortality tends to be higher at low light levels (Harrington 2006, Soos and Walters 1963). Seedling growth responds positively to increasing light and soil disturbance (Carter and Klinka 1992, Weber et al. 2003), with maximum growth rates occurring at 30 percent to more than 40 percent full sunlight (Harrington 2006, Wang et al. 1994). At high light, however, seedlings are susceptible to sun scorching (Wright et al. 1998). Western redcedar seedlings are particularly vulnerable to drought during the first 2 years (McKeever 1942). Height growth is slow during the seedling's first 5 years and peaks during the sapling's second decade (Nystrom et al. 1984). Ungulates are known to browse western redcedar repeatedly and severely, dramatically decreasing the number of leaved shoots per individual and increasing mortality (Burney and Jacobs 2010, Martin and Baltzinger 2002). Once established, western redcedar stands can have low mortality for several decades (Lutz and Halpern 2006).

Public concern about the decline of western redcedar in the Northwest has existed since the early 1970s (Sharpe 1974). Foresters are keen to promote western redcedar regeneration because of the tree's value. In intact stands, however, intense competition from overstory trees and understory vegetation limits seedlings' access to light, soil water, and nutrients (Harrington 2006). In gaps and larger openings such as clear cuts, natural regeneration requires seed sources that are within 100 m (330 ft), and several seed crops may be needed to fully stock the site; good seed crops can be expected only every few years (Clark 1970). Open environments present other challenges to the seedling, including competition, browsing, and sun scorching. Artificial regeneration using direct seeding or planting may be required to achieve reforestation objectives. Planting seedlings can be a way to avoid the stochastic events surrounding natural seedling establishment (Coates 2000). Seedlings need to be appropriately hardened for field conditions (Major et al. 1994). Direct seeding may be a low-cost option for regenerating western redcedar if successful techniques can be developed.

Successful direct seeding for any species requires proper timing, sufficient seed, predation and competition control, a suitable seedbed, and adequate soil moisture (Farmer 1997). Direct seeding has been used to reforest large areas of land in the American Southeast and has been particularly useful in large, remote, or low-productivity sites (Barnett 2014). Efforts to direct-seed western redcedar have generally proved unsuccessful, with lower germination and survival in western redcedar than Douglas-fir (*Pseudotsuga menziesii* [Mirb.] Franco), ponderosa pine (*Pinus ponderosa* Lawson & C. Lawson), and grand fir (*Abies grandis* [Douglas ex D. Don] Lindl.) (Engstrom 1955, Loewenstein and Pitkin 1966). Direct seeding has been most successful on north-facing sites with some shade and little competition; even under these conditions, however, the results have been only moderately successful (McKeever 1942). Direct seeding in fall may result in higher survival than in spring, although total survival through the first growing season was low in both treatments (Loewenstein and Pitkin 1966).

The objective of this study was to evaluate environmental influences on establishment success of direct-seeded western redcedar. We modified the environment using wire hardware cloth and mulch. Wire hardware cloth limits access by herbivores to the seeds and small seedlings (McKeever 1942) and hardware cloth increases shading on the seed by 15 to 21 percent (Minore 1972, Strothman 1972), which may help reduce mortality caused by high surface temperatures (Fowells and Arnold 1939). Mulch has a lower thermal admittance than bare soil, thereby helping to mitigate soil temperature and moisture stresses to newly germinated seedlings (Campbell and Norman 1998). The mulch retains moisture, which may also reduce water stress in the seedling. We hypothesized that seeds in the mulch and

hardware cloth treatments would have higher emergence than the treatment with no environmental modifications.

Materials and Methods

This study was conducted on a relatively level, tilled agricultural field with coarse, loamy soil at the University of Idaho's Pitkin Forest Research Nursery (46°43' N, 116°57' W). The site receives an average of 600 mm (23.6 in) of precipitation annually, and the average summer and winter temperatures are 18 °C and 0 °C (64 °F and 32 °F), respectively (Western Regional Climate Center 2005). No persistent vegetation existed at the site.

Northwest Seed (IFA Nurseries, Canby, OR) supplied the seed on behalf of Potlatch Corporation. The seed was collected at 883 m (2,900 ft). The seed arrived at Pitkin Forest Research Nursery in sealed pouches on October 16, 2013, and was stored dry in a cooler at 0 to 1.5° C (32 °F to 35 °F) for 4 weeks until direct seeding. The seed was not soaked or cold stratified before direct seeding, because stratification does not change germination capacity in western redcedar (Khadduri 2007).

Five frames were constructed from plywood and placed on top of the soil at the research site (figure 2.1). Each frame was divided into six 15-by-15 cm (5.9-by-5.9 5.9 in) sections. Within each frame, six treatments were randomly assigned to the sections (three mulch treatments by two screening treatments). Mulch treatments consisted of no mulch, pine mulch, or straw mulch. Screening treatments consisted of wire hardware cloth or no wire hardware cloth. The pine mulch was aged pine needles collected from a stand of ponderosa pine adjacent to the field site. The straw mulch was from baled wheat straw. The pine and straw mulch pieces were similar in size with a maximum length of 12.7 cm (5 in) and interspersed smaller pieces. The screened sections were covered with 6.35-mm (0.25-in) hardware cloth, which sat on top of the frame, about 10 cm (3.9 in) above the soil. The unscreened sections were left uncovered.

Before direct seeding, a minimal number of weeds were hand weeded from the site and the ground was lightly scarified with a rake. The seeds were sown on November 15 and 17, 2013. In each section, 100 seeds were surface sown in a 10-by-10 grid, spaced 1.27 cm (0.5 in) apart. In the mulched treatments, the respective mulch was spread across the section approximately 2 cm (0.78 in) deep. No follow-up treatment was done to ensure seed-soil contact; however, the soil was wet at the time of sowing, and the seed stayed in contact with the soil once sown. The site received no maintenance from the time of seeding until the seed began to germinate. The plots were hand weeded through the spring and summer.

Because western redcedar germination is epigeal, seedling emergence was defined in this study as the presence of the hypocotyl hook above the soil surface (figure 2.2). In April and May 2014, the plots were checked weekly for newly emerged and newly dead seedlings. From May to October 2014, the plots were checked monthly. Each newly germinated seedling was marked with a colored, ballpoint pin, with a different color used each week. When a seedling died, it was marked with a black pin (figure 2.3).

In addition to the field study, a germination test was conducted with four replications of 100 seeds each. The seed was soaked in cold, running water for 24 hours and then was cold stratified for 1 month at 0 to 1.5 °C (32 to 35 °F) (December 18, 2013 to January 15, 2014). Seeds were then placed on moist germination paper under a full-spectrum light for approximately 12 hours daily (Karrfalt 2008). The temperature fluctuated several degrees around 21 °C (70 °F). The seed was misted three times per day. Germinated seeds were counted every 7 days for 28 days. Germination was defined as the presence of a 5 mm (0.2 in) radicle (Baskin and Baskin 2014).

Statistical analyses were done in R, version 3.1.1 (R Core Team 2015). The experimental design consisted of a factorial (three mulch treatments by two hardware cloth treatments) completely randomized design with five replications. An analysis of variance was performed to test the treatment effects on the total number of emerged seeds in the field trial. Differences among treatment means were determined using Tukey's range test at the $\alpha \leq 0.05$ level. Diagnostic plots for equal variance and normality were examined and no data transformations were determined necessary. Overall germination average and standard error were determined on the germination test data using Microsoft Excel statistical tools.

Results

In the germination test, the average germination was 81 percent (n = 4, standard deviation = 6.7 percent). In the field planting, however, average emergence across all treatments was 31 percent (n = 30, standard deviation = 7.5 percent). Emergence was quantified in the field planting rather than germination because the radicle was not visible on

seeds in the field.

The first seedlings emerged by April 12, 2014, which was defined as week 1. Seedling emergence occurred earlier in the bare soil plots than the plots with mulch (figure 2.4). Seedlings began dying by the second week of observation, well before emergence was complete (figure 2.5). More than one-half of the seedlings were dead by week 8 (May 27, 2014). Some seedlings survived into September, but by week 27 (October 20, 2014), all seedlings in all treatments died and monitoring ceased. Dead seedlings were most often found intact and standing upright, with no sign that the cause of death was a pathogen or herbivore.

The highest total emergence occurred in the nonmulched with wire screening treatment (38.6 percent) and the lowest total emergence occurred in the straw mulch with no wire screening treatment (24.4 percent) (table 2.1). Seed in the nonmulched treatments had significantly higher total emergence than seed in the needle mulch or straw mulch treatments (p < 0.01). Emergence did not differ significantly between the two mulch types. No significant interactions occurred between the screening and mulching treatments nor was a significant difference observed between total emergence in screened and nonscreened treatments.

Discussion

This experiment modified the seedbed environment to reduce light (screening treatment) and increase available soil moisture (mulch treatment). These modifications, however, were not sufficient to ensure western redcedar seedling survival past the establishment phase. More than one-half of the seedlings died before July and August, the hottest months of the year, at the field site (Western Regional Climate Center 2005). Soil temperature, soil moisture, and shade levels were not directly measured, but the seedlings likely died from high temperatures and low soil moisture. In a similar way, natural regeneration of western redcedar has been unsuccessful on high fire-severity sites, with high temperatures and low moisture conditions (Larson and Franklin 2005).

The wire hardware cloth was intended to provide some shade and also to limit access by herbivores to the seeds and seedlings. In southern pine forests, seed predation by rodents and birds is a major challenge to successful direct seeding (Barnett 2014). No significant effect of the wire hardware cloth was observed, however, suggesting seed herbivory did not occur in this experiment. Some seedlings were observed with damage from invertebrates, but no evidence suggested damage by vertebrate herbivore. If the seedlings had survived, however, herbivory would be a matter of concern for larger western redcedar seedlings (Stroh et al. 2008). In such cases, fertilization may aid in recovery from browse (Burney and Jacobs 2010).

Mulch can also help reduce the number of weeds on a site. In this experiment, the site was routinely weeded, so the ability of the mulch to suppress weeds was not quantified. If weeds had been present, they would have competed with seedlings for soil moisture. Removal of competing vegetation can lead to greater height growth of western redcedar than only removal of light competition, suggesting competition for water is more important than competition for light (Adams and Mahoney 1991). Weedy vegetation can also compete for soil nutrients, but western redcedar have deep-rooted, fine roots, which can reduce competition for nutrients (Messier 1993).

Both seedling emergence and mortality were observed earlier and at higher levels in the bare soil plots compared with the mulched plots (figures 2.4 and 2.5). However, it is important to note that the absence of mulch in the bare soil plots made it easier to observe emerging seedlings. For eastern white-cedar (*Thuja occidentalis* L.), seed that falls on forest floor litter has a lower chance of survival than seed that falls on nurse logs or mineral soil (Simard et al. 2003). In this experiment, the seed was in direct contact with mineral soil and then was covered in mulch. Although great care was taken to count seedlings within the mulch, additional seeds may have emerged below the mulch and died before they were observed. Alternatively, the bare soil may have warmed earlier in the spring, allowing for earlier germination and emergence.

The seed was not stratified before planting but was subjected to cold, moist temperatures through the winter months. The need for cold stratification in western redcedar is debated, with some authors suggesting no stratification is needed (Brand and Schopmeyer 2008). Kolotelo (1996) observed no effect of a 3-week stratification period. We do not believe the lack of artificial cold stratification affected the experimental results.

Conclusions

It is important to understand the whole-plant response of seedlings to environmental factors such as light levels, water stress, and competition to choose the best method, species, and site combinations for successful regeneration projects (Coates and Burton 1999). These factors interact with one another in the field, impacting seedling development in complicated ways (Harrington 2010). In this experiment, germinated seedlings did not survive in spite of modifications to the microenvironment. If direct seeding is to be successful with western redcedar, it can be considered only on carefully selected sites, and, even then, success is not guaranteed. Based on current approaches, planting seedlings is still the most successful method to ensure western redcedar establishment. Further investigation to develop strategies for direct seeding of this species, such as the use of pelletized seed (Khadduri 2007), is needed if direct seeding continues to be a desirable approach.



Figure 2.1. To assess environmental influences on direct seeding of western redcedar, five wooden frames were constructed, each with six treatment sections. After sowing, seed were subjected to three mulching treatments (pine, straw, or no mulch), with or without wire hardware cloth screening. (Photo by Rebecca Sheridan, 2013)



Figure 2.2. Seedlings were counted as emerged when the hypocotyl hook was visible above the soil surface. Emerged seedlings were marked with color-coded pins. (Photo by Rebecca Sheridan, 2014)



Figure 2.3. Seedling emergence was monitored from March through October 2014. On each monitoring date, different colored pins were used to mark newly emerged seedlings. Dead seedlings were marked with a black pin. By October 2014, all seedlings in the experiment had died. (Photo by Rebecca Sheridan, 2014)

Table 2.1. Average total seedling emergence percent and standard deviation by treatment (n = 5). Seed in the nonmulched treatments had significantly higher total emergence than those in the mulched treatments (p < 0.01). Emergence did not differ significantly between the two mulch types or between screened and nonscreened treatments. No significant interactions occurred between the screening and mulching treatments.

Variable	No mulch		Pine mulch		Straw mulch	
	Without screening	With screening	Without screening	With screening	Without screening	With screening
Percent emergence	36.40	38.60	31.00	28.60	24.40	29.00
Standard deviation	1.95	9.02	4.95	8.56	1.95	6.44



Figure 2.4. Number of newly emerged seedlings from April through May 2014, as affected by mulching and screening treatments. Seedlings were marked with a pin to ensure they were not recounted.



Figure 2.5. Number of seedlings that died, by week, starting on April 18, 2014. Dead seedlings were marked with a black pin to ensure they were not recounted.

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