Seed Treatments for Container Seedling Production at the University of Idaho

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Introduction

Propagation from seeds is the most common method of plant production in forest and conservation nurseries (Landis and others 1999). Seed propagation is particularly widespread because this technique is relatively simple, inexpensive, and preserves genetic diversity in the crop – an important consideration in maintaining biodiversity and species persistence following outplanting. Other benefits of seed propagation include ease of shipping and long-term storage, especially for seeds with hard seed coats.

This article documents the techniques used to successfully propagate a wide variety of plants native to Idaho at the University of Idaho Franklin H. Pitkin Forest Nursery (UIPFN). Complete propagation protocols are being developed and will be posted on the Native Plant Network (www.nativeplantnetwork.org).

Disinfecting Seeds

Woody plant seeds, especially those with rough seed coats, harbor fungal and bacterial pathogens that can cause major problems during the seed treatment and germination process. This problem is acute for species that require several months of stratification; such seeds should be disinfected as a first step. One of the easiest and most effective treatments is a running water rinse (Figure 1) where seeds are placed under running water for 24 to 48 hours (James and Genz 1981). In addition to gently washing away pathogens, this treatment encourages full seed hydration, which is the first step in the germination process.

In cases when a water rinse is not sufficient, a solution of commercially available bleach and water can be used. Care should be taken with thin coated seeds, such as red alder (*Alnus rubra*), which can be damaged by prolonged exposure to concentrated bleach solutions. The most effective concentration (v:v) is species-dependent and can range from 1:8 to 2:3 (bleach:water) (Luna and others 2009). At the UIPFN, a 1:5 concentration is used to disinfect species particularly susceptible to fungal contamination, such as lodgepole, limber, and western white pines (*Pinus contorta, P. flexis, P. monticola, P. ponderosa*) and Douglas-fir (*Pseudotsuga menziesii*).



Figure 1 – A running water rinse accomplishes 2 prerequisites for effective seed treatments: cleansing the seedcoat and imbibing the seed with water.

Seeds can be placed in sacks of tulle or mesh bags, submerged in a bleach solution, and swirled around in order to allow bleach to coat seeds evenly. After several minutes, seeds are thoroughly rinsed in running water. In extreme situations, fungicide can be used to reduce mold development, but should only be done when absolutely necessary due to possible phytotoxicity and health concerns (Finnerty and Hutton 1993).

Seed Treatment by Dormancy Type

Seed dormancy is an adaptation that ensures seeds will germinate only when environmental conditions are favorable for survival. Some seeds require no treatment and will germinate immediately in the proper environment whereas most native plants have seeds with some type of dormancy. Determining the type and degree of seed dormancy is critical to successful propagation, and keys are available to determine which type of dormancy is exhibited by seeds (Luna and others 2009).

External Dormancy

The term external dormancy is synonymous with physical dormancy and is caused by the presence of a lignified layer of cells in the seed coat that prevents a dormant seed from taking up water and gases. However, once the seed is made permeable it can complete its germination within 2 weeks, if no additional dormancy exists (Baskin and Baskin 1998; Luna and others 2009).

Scarification

Physical dormancy is alleviated by scarification of the seed coat, which can be done in a number of ways including piercing, filing, nicking, sanding, burning, acid scarification, or wet heat. At the UIPFN, a concentrated solution of sulfuric acid (H₂SO₄) is used to treat species with especially thick seed coats or stony endocarps such as black hawthorn (Crataegus douglasii), redstem ceanothus (Ceanothus sanguineus), and oakleaf sumac (Rhus trilobata), with treatment durations ranging from 20 to 45 minutes (Table 1). For this procedure, seeds are placed in a glass container and covered with concentrated sulfuric acid. The seeds are gently stirred and allowed to soak for a predetermined duration of time (dependent on the species, seed size, and seed coat thickness). Sulfuric acid reacts strongly with water, thus it is important to remember to never add water to acid, as it can result in a powerful reaction and cause severe chemical burns. In addition, sulfuric acid can damage the seed embryo; therefore seeds must be closely monitored during the treatment. If working with a new species for which treatment duration is not known, monitoring can be done by removing seeds from the acid bath at regular intervals and cutting them. Once the seeds can be cut easily but maintain a firm embryo they have been sufficiently scarified, at which point all seeds should be removed from the acid. Sulfuric acid can be potentially dangerous to the applicator and requires protective wear (gloves, goggles, and aprons) and must be disposed of properly (Luna and others 2009).

Internal Dormancy

Seeds with internal dormancy have embryos that are either underdeveloped or exhibit a low growth potential (Baskin and Baskin 1998). A number of different types of internal dormancy exist, but all respond to some combination of specific temperature and moisture treatments. Stratification can be described as the process that uses high moisture and temperature or a series of temperatures to overcome seed dormancy. According to Luna and others (2009) the term "stratification" refers to cold, moist conditions, while the term "warm, moist treatment" means exactly that. Several native plants require some combination of stratification and warm, moist treatment. Stratification temperatures usually range between 1 and 3 °C (34 to 38 °F), while warm, moist treatment temperatures fall between 18 and 30 °C (65 to 86 °F).

A number of different techniques can be used for stratification and for warm, moist treatment. Seeds can be placed in plastic bags and mixed with a moist medium, such as Sphagnum moss, or wet burlap to maintain high humidity. At the UIPFN seeds are placed in nylon tulle, which can be purchased at any fabric store in a variety of mesh sizes. The nylon tulle is tied, labeled, buried in a bucket filled with moist media, and placed in the appropriate temperature conditions (Figure 2). This is a particularly useful technique for species that are transplanted into containers as germinants, such as black hawthorn, red currant (Ribes sanguineum), Rocky Mountain juniper (Juniperus scopulorum), and several rose species (Rosa nutkana and R. woodsii) (Table 1). The germinant sowing procedures are well described in Landis and others (1999).



Figure 2 - At the Pitkin Forest Nursery seeds are placed in nylon tulle, which is buried in a bucket filled with moist media and placed in the appropriate temperature conditions for stratification or warm, moist treatment.

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Another method, "naked" stratification, involves placing seeds in nylon tulle, which is positioned inside a plastic bag filled with a small amount of water in order to maintain high relative humidity (RH) inside each bag (Figure 3). The nylon tulle containing seeds should not come in contact with the water in the plastic bag. Bags can be tied and hung inside a cooler for the required duration of time. The seeds should be checked frequently to make sure there is no fungal presence and rinsed periodically to cleanse and aerate them. At the UIPFN, this method is used primarily for conifers, but is also suitable for the treatment of some native shrubs and hardwoods (Table 1).



Figure 3 - Naked stratification involves placing seeds in nylon tulle, which positioned inside a plastic bag above a supply of water, in order to maintain high relative humidity (RH) inside each bag. The nylon tulle containing seeds should not come in contact with the water in the plastic bag. Bags can be tied and hung inside a cooler for the required duration of time.

An alternative method involves stratifying seeds once they are in the growing containers. Dormant seeds are sown on the surface of the media and sometimes covered by a thin layer of media or grit that helps maintain constant conditions around the seed. Whether or not seeds are covered depends on their light requirement; for instance seeds of western syringa (*Philadelphus lewisii*) are stratified uncovered because they require light during germination. Once sown, containers are watered until the medium is brought to field capacity. Finally, the sown containers are placed into a cooler, or stored outside in a protected area during the winter (in areas where appropriate temperature conditions are met). If only a warm, moist treatment is required, seeds can be maintained under a moist cover in a greenhouse. It is important to note that the media moisture level should be maintained during the treatment period, which is of particular importance for warm, moist treatments. In addition, seeds treated in this manner must be protected against rodent predation.

At the UIPFN a number of native shrubs and hardwoods are stratified directly in containers (Table 1). This approach is especially useful for smaller seeds that are harder to transplant or those that are covered with mucilage [for example, serviceberry (Amelanchier alnifolia)] that prevents them from being effectively handled during naked or moist media stratification. In addition, container stratification allows for a quick transition from stratification into the greenhouse, thus saving the grower a substantial amount of time in the spring when time is limited. However, this approach requires considerably more area than other stratification methods and is not advisable when space constraints are an issue. To make certain that each container has a viable seed, several seeds are sown in each container and thinned after germination.

Some species require exceptionally humid conditions in order to germinate. For such species the use of a "foghouse" or another high humidity environment may be necessary to promote germination following stratification or warm, moist treatment. Mountain huckleberry (Vaccinium membranaceum) is an example of a species with this requirement. Although generally considered to be difficult to propagate, it has been successfully cultivated at the UIPFN. Seeds are sown directly into hydrated Jiffy J2865 (3.1 in³) forestry peat pellets (Jiffy Products of America, Inc., Norwalk, OH), placed in corresponding trays, sealed in plastic bags to prevent moisture loss, and then stratified at 1.5 °C (35 °F) for 30 to 45 days. Following stratification, the trays are moved to the foghouse (at 85% RH) and placed on benches with heated-water circulation (21 °C [70 °F]). Germination occurs in 13 to 21 days. After 90 days seedlings are moved into a greenhouse and placed in front of moist cooling pads, where temperatures range between 26.5 °C (80 °F) and 10 °C (50 °F) (Regan and others 2012).

Scientific Name	Common Name	Sulfuric Acid	Gibberellic Acid	Running Water Soak	Warm Treatment	Stratification	Germinant Sowing	Running Water Rinse
Hardwoods								
Alnus incana	Thinleaf Alder	-	_	yes	_	180 d naked	-	yes
Betula occidentalis	Water Birch	-	-	-	-	-	-	-
Betula papyrifera	Paper Birch	-	-	-	-	_	-	-
Cornus sericea	Redosier Dogwood	-	-	yes	-	90 d naked	-	yes
Crataegus douglasii	Black Hawthorn	25 min	-	yes	30 d media	120 d media	yes	_
Prunus virginiana	Chokecherry	_	-	yes	_	140 d container	-	_
Sorbus scopulina	Mountain Ash	-	-	yes	_	150 d container	-	_
Shrubs								
Amelanchier alnifolia	Serviceberry	_	_	yes	_	120 d container	_	_
Ceanothus sanguineus	Redstem Ceanothus	20 min	_	ves	_	60 d naked	_	ves
Dasiphora fruticosa	Shrubby Cinquifoil	_	_	ves	_	60 d naked	_	ves
Philadelphus lewisii	Westem Syringa	_	_	ves	_	60 d container	_	_
Physocarpus malvaceus	Ninebark	_	_	ves	_	30 d naked	_	ves
Purshia tridentata	Antelope Bitterbrush	_	_	ves	_	60 d naked	_	ves
Rhamnus purshiana	Cascara	_	_	ves	_	115 d naked	_	ves
Rhus trilobata	Oakleaf Sumac	45 min	_	yes	_	60 d naked	_	yes
Ribes aureum	Golden Currant	_	_	ves	_	60 d container	_	-
Ribes sanguineum	Red Currant	_	_	yes	_	120 d media	yes	_
Rosa nutkana	Nootka Rose	_	_	yes	115 d media	115 d media	yes	_
Rosa woodsii	Woods' Rose	_	_	yes	30 d media	120 d media	yes	_
Salvia dorrii	Desert Purple Sage	_	_	yes	_	_	-	_
Sambucus nigra	Blue Elderberry	_	GA	yes	_	60 d tray	_	_
Vaccinium membranaceum	Mountain Huckleberry	_	_	yes	_	30 d Jiffy tray	-	_
Conifers								
Abies concolor	Concolor Fir	-	-	yes	-	28 d naked	-	yes
Abies grandis	Grand Fir	_	_	yes	_	28 d naked	-	yes
Abies lasiocarpa	Subalpine Fir	_	-	yes	_	28 d naked	-	yes
Juniperus scopulorum	Rocky Mountain Junipe	er –	-	yes	120 d media	120 d media	yes	-
Larix occidentalis	Western Larch	-	-	yes	-	28 d naked	-	yes
Picea engelmannii	Engelmann Spruce	-	-	yes	-	28 d naked	-	yes
Picea pungens glauca	Blue Spruce	-	-	yes	-	28 d naked	-	yes
Pinus contorta	Lodgepole Pine	-	-	yes	-	28 d naked	-	yes
Pinus flexilis	Limber Pine	-	-	yes	-	40 d naked	-	yes
Pinus monticola	Western White Pine	-	-	yes	_	120 d naked	-	yes
Pinus ponderosa	Ponderosa Pine	-	-	yes	-	46 d naked	-	yes
Pseudotsuga menziesii	Douglas-fir	-	-	yes	-	28 d naked	-	yes
Thuja plicata	Westem Redcedar	-	_	yes	_	28 d naked	-	yes
Tsuga heterophylla	Western Hemlock	-	-	yes	-	45 d naked	-	yes
All water rinses are 24 hours and occur after stratification; all soaks are 48 hours and occur before stratification or sowing (if no stratification is required).								

Table 1. Summary of seed treatment techniques used for native hardwoods, shrubs, and conifer species grown at the University of Idaho Pitkin Forest Nursery (Moscow, ID).

Gibberellic Acid

Gibberellic acid (GA₂) is a naturally occurring growth hormone that is often used to alleviate internal dormancy. It can be used alone or in combination with stratification and is sometimes substituted for a warm, moist treatment (Timson1966; Pinfield and others 1972; Luna and others 2009). GA₃ acts through increasing the growth potential of the embryo and by overcoming the mechanical constraints that prevent the emergence of the radicle (Hilhorst and Karssen 1992; Leubner-Metzger 2003). This chemical can be readily purchased from horticultural or chemical suppliers; however, the concentration of the effective GA₃ solution is species dependent, typically varying between 100 and 1000 ppm (Luna and others 2009). A treatment solution can be prepared by dissolving GA, in distilled water according to the concentration requirement. The UIPFN uses a 346 ppm solution to treat seeds of blue elderberry (Sambucus nigra spp. cerulea), which is prepared by mixing 51.9 mg of GA₃ with 0.15 L of water. GA₂ solutions can be applied by soaking seeds directly, thoroughly spraying them, or by placing seeds on germination paper saturated with the solution. With this latter technique, following treatment seeds are removed and then planted into growing containers (Figure 4). Because GA₃ is a powerful growth hormone, high concentrations can cause premature germination and poor seedling quality; when working with a new species, always start with dilute concentrations first (Luna and others 2009).



Editor's Note: Trimaco® Cone Paint Strainer bags come in 1 and 5 gallon sizes and are perfect for rinsing and stratifying seeds.



Figure 4 - The GA_3 solution can be applied with a spray bottle. This method requires seeds to be germinated in trays and then planted into growing containers.

Sowing Techniques in Container Nurseries

Direct Sowing

Direct sowing is a quick and economical method for sowing seeds because it minimizes handling and labor. It is by far the most common approach, especially for those seeds that require little to no pre-sowing treatment. Furthermore, direct sowing can be mechanized for larger scale operations, further expediting the process (Luna and others 2009). For this approach it is important for growers to have accurate germination information to set optimal sowing rates in order to insure adequate seedling emergence. If such information is not available, small-scale germination tests should be conducted in order to assess the germination rate of a particular seedlot (Luna and others 2009). Procedures for determining the optimal number of seeds necessary to obtain a target number of germinants are described in detail in Chapter 8 of the Nursery Manual for Native Plants (Luna and others 2009). However, many native plants have various dormancy requirements and often benefit from germinant sowing.

Germinant Sowing

Germinant sowing, sometimes referred to as planting "sprouts," is particularly useful for large seeds, seed lots of variable quality, or for those for which no germination test data are available (Landis and Simonich 1984; Finnerty and Hutton 1993). Germinant sowing is primarily used for seeds with internal dormancy that require stratification and/or warm, moist treatment, such as maples and junipers. The major advantage of this method is that it ensures a live seedling originates from each container (Landis and others 1999). At the UIPFN, this technique is used frequently, especially for seedlings planted into Jiffy forestry pellets.

Seeds that will be sown as germinants are place in stratification or warm, moist treatment and monitored every few days to gauge germination. Seeds with an emerged radicle are separated out and sown. Larger seeds can be planted by hand, but smaller ones require tweezers, although planting method is subject to preference (Landis and others 1999). For tap-rooted species such as some nut crops, the radicle of the dominant root is sometimes pruned to encourage the establishment of a more fibrous root system. However, the pruned amount should not exceed 3.0 mm (0.12 in) (Emery 1988).

One of the major drawbacks of germinant sowing is the need for precision in germinant placement. The seeds must be sown with the radicle extending downward as inadequately placed seeds will be deformed, brittle, or break when they become larger. Another disadvantage is that sowing may take several weeks or months, resulting in an uneven-aged crop and may not be beneficial when labor is an operational limitation. In order to reduce sowing time, seeds can be germinated and the germinants transplanted into containers. At the UIPFN, this technique is used for black hawthorn, red currant, Rocky Mountain juniper, and several rose species (Table 1). Because these species are stratified in moist media, the entire stratification bucket is moved from the cool into a warm environment (21 °C [70 °F]) and remains there for 3 to 5 days. During this process media moisture levels match those maintained during stratification. Germination is monitored daily. Seeds with an emerged radicle are separated out and sown. The major advantage of this treatment is that it requires little additional effort, but ensures more uniform and timely germination. However, not all species respond well to this technique so some experimentation is needed when working with new species. It is also important to note that sowing may sometimes take several weeks or months, which may not be beneficial when labor is an operational limitation.

Sources

Baskin CC, Baskin JM. 1998. Seeds: ecology, biogeography, and evolution of dormancy and germination. San Diego (CA): Academic Press.

Emery DE. 1987. Seed propagation of native California plants. Santa Barbara (CA): Santa Barbara Botanic Garden.

Finnerty TL, Hutton KM. 1993. Wood shrub propagation: a comprehensive approach. In: Landis TD, technical coordinator. Proceedings, Western Forest Nursery Association. Fort Collins (CO): USDA Forest Service Rocky Mountain Forest and Range Experiment Station. General Technical Report RM-221: 82-91.

Hilhorst HW, Karssen CM. 1992. Seed dormancy and germination - the role of abscisic acid and gibberellins and the importance of hormone mutants. Plant Growth Regulation 11:225-238.

James RL, Genz D. 1981. Ponderosa pine seed treatments: effects on seed germination and disease incidence. Missoula (MT): USDA Forest Service Timber, Cooperative Forestry and Pest Management. Northern Region Report 81-16. 13 p.

Landis TD, Tinus RW, McDonald SE, Barnett JP. 1999. Seedling propagation, Vol. 6, The Container Tree Nursery Manual. Washington (DC): USDA Forest Service, Agricultural Handbook 674. p 35-92.

Landis TD, Simonich EJ. 1984. Producing native plants as container seedlings. In: Murphy PM, compiler. The challenge of producing native plants for the Intermountain Area. Proceedings, Intermountain Nurserymen Association Conference. Ogden (UT): USDA Forest Service, Intermountain Forest and Range Experiment Station. General Technical Report 168. 96 p.

Leubner-Metzger G. 2003. Functions and regulation of beta-1,3-glucanases during seed germination, dormancy release and after-ripening. Seed Science Research 13:17-34.

Luna T, Wilkinson K, Dumroese RK. 2009. Seed germination and sowing options. In: Dumroese RK, Luna T, Landis TD, editors. Nursery manual for native plants: A guide for tribal nurseries - Volume 1: Nursery management. Washington (DC): USDA Forest Service. Agriculture Handbook 730. p 133-151.

Pinfield NJ, Stobart AK, and Martin MH. 1972. Control of germination in *Stachys alpina* L. New Phytologist 71:99-104.

Regan DJ, Woodruff K, Davis AS. 2012. Propagation protocol for mountain huckleberry (*Vaccinium membranaceum*). Native Plants Journal 13(1): 14-18.

Timson J. 1966. Germination of *Polygonum convolvulus* L. New Phytologist 65:423-428.