# Repetitive Reaction and Restitution (R<sup>3</sup>) Induction of Drought Hardiness in Conifer Container Seedlings

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**Abstract:** Planting failures are often attributed to unexpectedly harsh conditions after planting. Characterization of soil water at the planting site, along with associated influences of site preparation and soil texture, is recommended. Additionally, tree planting technique and seedling biology should be targeted to site conditions.

A nursery regime for induction of drought hardening is proposed for hot-lift summer- and fall-planted container conifers.

Keywords: seedling hardiness, nursery culturing, drought stress

## Introduction

Nursery growers, researchers, and field foresters have improved forest seedling quality, planting technology, and site preparation with a corresponding increase in survival and growth after outplanting. This expertise has been disseminated worldwide through publications and conferences. In the 1970s and 1980s, average survival for planted seedlings was 50-75% (Cayford 1978; Mexal and others 2008). Today we expect survival of 90% or greater, although particularly harsh sites may need to be planted more than once due to poor seedling survival where water is a limiting factor (Mexal and others 2008; Regan and Davis 2008).

# Characterizing the Site \_

Site preparation treatments such as vegetation control, scarification, and cultivation, along with annual weather patterns, result in a range of site conditions for outplanting. Lantz (1996) describes North Carolina standards for planting categories as normal, critical, and severe based temperature (Table 1), relative humidity, wind, and soil moisture on the day of planting. To further characterize the site, soil texture must be considered; the ability to hold water varies among soils (Table 2; Figure 1). When soil water potential is below -60 to -70kPa (-0.6 to -0.7 bar), root elongation ceases (Grossnickle 2005). In order to characterize the planting site for outplanting dates beyond the traditional outplanting windows, a soil moisture stress curve should be estimated based on a progression of moisture conditions at selected depths (Figure 2) using standard methods for determining soil hydraulic conductivity. This curve will suggest the moisture stress level likely to be encountered by a newly planted seedling before new root growth penetrates to a new source of moisture.

Table 1. North Carolina planting categories (Lanz 1990).			
	Normal	Critical	Severe
Temperature	2 to 24 °C (35 to 75 °F)	24 to 29 °C (76 to 85 °F)	> 29 °C (> 85 °F)
Relative Humidity	>50%	30-50%	<30%
Wind	<10mph	>10mph	>15mph
Soil moisture buildup	<30	30-80	>80

Table 1. North Carolina planting categories (Lantz 1996).

Table 2. Storage capacity for different soil texture classes (inches water/
feet depth). (1 in = 2.54 cm; 1 ft = 0.3 m)

Texture class	Field capacity	Permanent wilting point
Sand	1.2	0.3
Fine sand	1.4	0.4
Sandy loams	1.9	0.6
Fine sandy loams	2.6	0.8
Loams	3.2	1.2
Silt loams	3.4	1.4
Light clay loam	3.6	1.6
Clay loam	3.8	1.8
Heavy clay loam	3.9	2.1
Clay	3.9	2.5

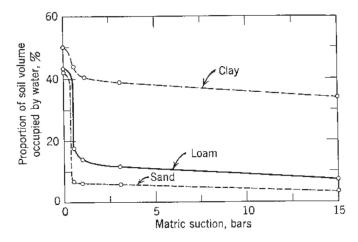


Figure 1. Volumetric water content of three soils at different matric suctions (from Black 1968).

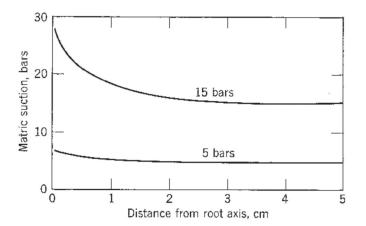


Figure 2. Matric suction versus distance from root in bulk sandy loam soil at -5 and -15 bars (Black 1968).

#### The Root/Soil Interface

Kramer and Kozlowski (1960) note that pines (Pinus spp.) have few growing root tips during winter, yet can absorb considerable quantities of water. Suberized roots can be a major route of water uptake in tree root systems, through lenticels on the surface of the root and by discontinuities in the periderm (bark) plates although unsuberized (white) root tips on growing roots have the highest uptake per unit of surface area (Carlson and others 1990). New root growth leads to an increase in daily minimum water potential  $(\Psi)$ for newly planted seedlings (Figure 3) (Grossnickle 2005). Given that the newly planted seedling may encounter a soil moisture stress condition that precludes new root growth, survival depends upon creating the water continuum with the existing root system-the hydrostatic system (Kramer and Koslowski 1960) or the soil-plant-atmosphere continuum (SPAC) (Grossnickle 2005).

Grossnickle (2005) provides an excellent discussion of the importance of root-soil contact in the planting of seedlings and points out that poor contact will increase the resistance to water movement from the soil to the plant. Landis and Dumroese (2009) emphasize the importance of ensuring the planting stock is fully hydrated at the time of planting. When the soil is dry or the soil texture is coarse, the South African "Puddle-Plant" technique may be employed, with the addition of 3 to 5 L (0.8 to 1.3 gal) of water per plant (Viero 2000). Seedling water demand in the first few days after outplanting can be estimated from irrigation studies in the nursery immediately before lifting. When outplanting takes place outside the traditional outplanting window, however, hydration at the time of outplanting may be insufficient to meet moisture needs until the next rainfall event. Water deficits can inhibit physiological processes such as photosynthesis with a consequent reduction in root growth (Joly 1985).

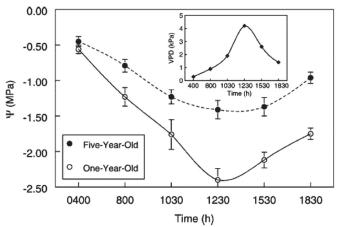


Figure 3. Diurnal y of newly planted and established lodgepole pine seedlings. Insert of vapor pressure deficit (Grossnickle 2005).

Water uptake can occur via an apoplastic or a symplastic pathway (Figure 4), both of which are influenced by solute concentrations and osmosis. As the distance from the root increases, soil moisture can be expected to increase. Water content in close proximity to the roots, however, can be increased due to production of mucigel. The peripheral cells of the root cap and the epidermal cells of the root produce and secrete large amounts of mucigel, a slimy substance made by dictyosomes. Mucigel is a hydrated polysaccharide containing sugars, organic acids, vitamins, enzymes, and amino acids (Walker and others 2003; Earley 2010). A prominent role of mucigel is to provide continuity between root surfaces and soil moisture.

## Proposed R<sup>3</sup> Protocol \_\_\_\_

In the nursery, mild moisture stress (-1.5 MPa [-15 bars]) is often used to induce dormancy, but foliar injury can occur at higher levels (-1.8 to -2.0 MPa [-18 to -20 bars]) (Landis 1999). Lopushinsky (1990) notes that drought hardiness can be increased by exposure to moderate water stresses (-0.5 to -1.0 MPa [-5 to -10 bars]) and cites work by Christersson showing that pot-grown Scots pine (Pinus silvestris L.) and Norway spruce (Picea abies [L] Karst.) seedlings could increase drought tolerance to -3.5 MPa (-35 bars), compared to -2.5 MPa (-25 bars) for unhardened seedlings. Joly (1985) suggested that the turgor pressure that leads to observable wilt probably changes as a result of water stress conditioning, and Stocker (1960) describes the "time course of changes in the plasmatic viscosity" as comprising two phases-Reaction phase (destruction of the plasma) and Restitution phase (reparation of the damage).

### **Reaction Phase**

At the beginning of the hardening regime, seedlings are leached with water (to reduce growing medium solution EC) and then dried down to the "Just Open Limit" (50% stomatal closure; temporary wilting). This increases respiration, decreases transpiration and phytosynthesis, and reduces growth. The expected  $\psi$  is approximately -2500 kPa on these temporarily wilted seedlings.

#### **Restitution Phase**

Seedlings are re-wetted with hardener fertigation. The irrigation system must be adequate to cool the entire crop within minutes. This increases photosynthetic rate. This regime is continued for 1 to 2 weeks.

### Repetition<sup>2</sup>

Reaction and Restitution phases are repeated. The temporary wilt will likely be observed on those seedlings that did not wilt on the first dry-down. Photosynthetic capacity may become greater after successive wilting periods (Stocker 1960).

#### Repetition<sup>3</sup>

Reaction and Restitution phases are repeated again. The temporary wilt will likely not be observed on this final dry down.

Once the steps have been completed, seedlings are rewetted with nutrient loading solution, then harvested and shipped.

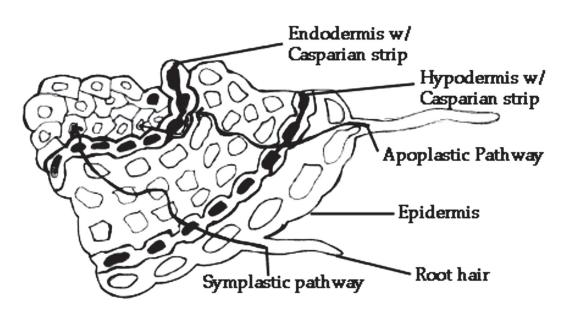


Figure 4. Water uptake pathways (Earley 2010).

Similar drought pre-conditioning protocols have been investigated over the past several decades with varying results. Any change to culturing regimes should first be tried on a small scale for each species, seed zone, and stocktype in question to determine its influence on subsequent crop quality and outplanting performance.

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The content of this paper reflects the views of the authors, who are responsible for the facts and accuracy of the information presented herein.