AN ABSTRACT OF THE THESIS OF

J. A. GIL VERA-CASTILLO for the degree of Doctor of Philosophy

in Forest Science presented on April 24, 1995.

Title: The Influence of Antidesiccants on Field Performance and Physiology of 2+0 Ponderosa Pine (*Pinus ponderosa* Dougl.) Seedlings.

Abstract approved: Signature redacted for privacy.
Dr. Stephen D. Hobbs

This thesis research examines the effects of film-forming antidesiccants applied to dormant ponderosa pine (*Pinus ponderosa* Dougl.) seedlings after being lifted and to actively growing seedlings. The basic proposition was that antidesiccants would have a positive effect on reducing water loss in ponderosa pine seedlings. In order to evaluate the effects of six different antidesiccant treatments on the physiology, morphology, and phenology of ponderosa pine seedlings, four experiments were conducted and four hypotheses tested. Eleven variables were studied: survival, height and diameter growth, budbreak, budset, foliar damage, root growth potential, water loss, stomatal conductance, electrolyte leakage, and chlorophyll fluorescence.

None of the antidesiccants or concentrations tested affected survival or height and diameter growth. However, several antidesiccant treatment did cause temporary reductions in stomatal conductance of outplanted seedlings in June 1992. Seedlings treated with AntiStress® 2000 (1:20 and 1:40) had the lowest reduction in stomatal conductance. In another experiment in a controlled environment, antidesiccants did reduce water loss but failed to significantly affect height growth.

Antidesiccant application to actively growing seedlings subjected to different periods of wind exposure reduced foliar damage as measured by the amount of electrolyte leakage released from the fascicles but increased stomatal conductance. In a separate experiment under greenhouse conditions antidesiccants did not reduce stomatal conductance but they did damage seedling foliage.

The film-forming antidesiccants had a negative effect on the growth of new roots. A 46% reduction in new root growth was observed. When antidesiccants were used, budbreak activity was also delayed. Moreover, in several experiments antidesiccants had no effect on photochemical efficiency. Antidesiccants also did not affect light absorption or emission.

In this thesis research the proposition was that antidesiccants would have a positive effect on ponderosa pine seedlings. Based on the preponderance of evidence from the four experiments conducted, it is concluded that under the conditions of this study, antidesiccants tested had little overall effect on ponderosa pine seedlings. However, had seedlings been subjected to greater water stress, the outcome of this research might have been different.

THE INFLUENCE OF ANTIDESICCANTS ON FIELD PERFORMANCE AND PHYSIOLOGY OF 2+0 PONDEROSA PINE (*Pinus ponderosa*Dougl.) SEEDLINGS

by

J.A. Gil Vera-Castillo

A THESIS

submitted to

Oregon State University

in partial fulfillment of

the requirements for the degree of

Doctor of Philosophy

Completed April 24, 1995

Commencement June 1995

ACKNOWLEDGEMENTS

This thesis research could not have been accomplished without the support and help of many people. A few in particular kept me going when I had lost faith in myself, and thought I had lost track of what I had set out to accomplished. I would like to express my sincere appreciation to my major professor, Dr. Stephen Hobbs for his support during my stay at OSU and for his many important contributions to this thesis. Also, I would like to thank my committee members in this research project: Drs. Tony Chen, Les Fuchigami, David Hibbs, and John Tappeiner, for their advice during my academic program at OSU. I especially appreciated their help during my preparation for the preliminary written and oral examinations. Tom Sabin and Lisa Ganio provided important statistical advice since the beginning of this thesis research.

I would like to thank the Mexican National Council of Science and Technology (CONACYT) and the National Institute of Forest Research (INIFAP) for granting me a scholarship. Special thanks are due to in particular to Lic. Humberto Valencia Salazar and Ing. Carlos E. Gonzalez Vicente for their help and support during my doctoral program. Also, I would also like to thank the Department of Forest Science for financial support during my last term at Oregon State University.

Appreciation is also given to my friends André Laroze, Carlos L. and Ana Ballaré-Scopel, Ruben and Laura Cruz-Fuentes, Brad and Tina Withrow-Robinson, Nestor and Rosi Rojas-Steinbrecher. Among the friends that contributed to make our

stay more enjoyable in Corvallis were Cleo C. de Mueles, David and Sarah Hibbs, Bill and Eija Emmingham, and Terry, Sandy, and Gary Vaughn.

I also wish thanks my parents, Alberto and Guadalupe, and my brothers and sisters, David, Ana Margarita, Gabriel, Moises, Emma, Beatriz, Isaac, Virginia Eulalia, Alberto, Silvia, and Dario for their love and moral support.

Last, and certainly not least, I would like to thank my wife, Barbara, and children, Gil and Itzel for enduring sometimes difficult living conditions when I was often preoccupied with my classes and research project. Your effort, support, and understanding are a major part of my success.

TABLE OF CONTENTS

TABLE OF CONTENTS (Continued)

LIST OF FIGURES

LIST OF TABLES

LIST OF TABLES (Continued)

LIST OF TABLES (Continued)

THE INFLUENCE OF ANTIDESICCANTS ON FIELD PERFORMANCE AND PHYSIOLOGY OF 2+0 PONDEROSA PINE (*Pinus ponderosa* Dougl.) SEEDLINGS

CHAPTER 1
INTRODUCTION

The quality of bareroot seedling planting stock depends on many factors including the way it is lifted, graded, packaged, and stored (Garber and Mexal 1980; Burdett and Simpson 1984). In the nursery, the lifting operation is very stressful on seedlings because it damages both shoots and roots. Damage to the root system is particularly significant. Lifting can result in the loss of 50% of all roots (Wakeley 1965) and as much as 75% of the small roots (Rowan 1983). Moreover, the remaining fine roots are susceptible to desiccation before seedlings are outplanted. If the root system is unable to supply sufficient water to the seedling, water stress will develop and consequently other types of stress such as reduced nutrient uptake may occur (Coutts 1981).

Transplanting shock is an interruption of normal seedling growth which is caused by a complex of physiological changes that occur after outplanting (Cleary et al. 1988). There are three possible reasons for transplanting shock: 1) loss of fine roots on lifting; 2) desiccation during lifting, grading, storing, shipping, and planting; and 3) poor root to soil contact after outplanting (Sands 1984). Physical damage and desiccation of the root system are believed to be important features of transplanting shock which can result in reduced shoot growth and even mortality (Burdett and Simpson 1984; Molina and Trappe 1984; Johnson and Cline 1991).

Reducing water loss during lifting has been confirmed to be an important factor in reducing desiccation stress and increasing the survival of tree seedlings (Lefevre et al. 1991). Seedlings subjected to excess desiccation during any phase of nursery production will have reduced growth potential and be of poor quality (Chen et al. 1991). Also during cold storage, seedling quality can be reduced due to metabolic changes if the storage environment is other than optimal (Burdett and Simpson 1984). Freezing temperatures damage seedling roots, especially fine roots, when ice crystal formation occurs within the cells, rupturing the cell membrane (Levitt 1972), whereas warmer temperatures encourage increased maintenance respiration and the depletion of carbohydrate reserves (Johnson and Cline 1991). During cold storage, tree seedlings must depend upon their carbohydrate reserves to meet their physiological requirements (Marshall 1985). The rate of depletion of carbohydrate reserves is strongly influenced by the cold storage environment (Burdett and Simpson 1984). Moreover, cold storage can also affect seedling physiology by partially satisfying the chilling requirements necessary to break dormancy (Johnson and Cline 1991). Once planted, seedlings can lose water rapidly, and desiccation is the most common menace to survival during at least the first season after outplanting (van den Driessche 1989). Although seedlings are normally able to prevent desiccation by stomatal closure, they do so at the cost of preventing diffusion of atmospheric carbon dioxide into the leaf intercellular space, thus limiting photosynthesis and ultimately seedling growth (Simpson 1984).

For many years researchers have worked to reduce plant transpiration by reducing the permeability of the leaf surface to water vapor movement. One obvious

means of achieving this is to manipulate the stomatal apparatus inducing stomatal closure with substances known to induce stomatal closure, for example, paclobutrazol. An alternative procedure is to impose a low permeability film over the entire leaf with plastic films and wax emulsions commonly called antidesiccants.

From this perspective, it is important not only to understand the nature of transpiration suppression but also to investigate the effect of antidesiccant compounds on photosynthesis and plant growth. Since both transpiration and photosynthesis involve gaseous diffusion across the leaf-air interphase, it seems probable that an increase in the resistance across this zone would significantly affect both processes (Slatyer and Bierhuizen 1964). The available data suggest that the ultimate effects of antidesiccants on transpiration and photosynthesis depend to a degree on compound permeability and to a greater extent on environmental conditions (Davies and Kozlowski 1974).

Antidesiccants might be useful in improving conifer regeneration success through two mechanisms: the reduction of transpiration losses during the stressful period after outplanting and the extension of the planting season by pretreatment of seedlings to resist drought stress (Simpson 1984; Odlum and Colombo 1987).

This thesis research examines the effects of antidesiccants on survival, growth, and physiology of 2+0 ponderosa pine (*Pinus ponderosa* Dougl.) seedlings. The basic proposition is that antidesiccants have a positive effect on ponderosa pine seedlings. To explore this proposition, four hypotheses were tested. The first hypothesis is that antidesiccants applied to dormant seedling after lifting will not improve field survival and growth of ponderosa pine seedlings by influencing plant water relations. The loss of

water vapor from the shoot may be reduced by increasing stomatal resistance during the stressful first growing season after outplanting. Thus a high initial water potential facilitates photosynthesis and root growth during the seedling establishment phase provided other necessary conditions are favorable. Subsequently, root growth will increase water uptake by increasing the absorptive surface area and improving the root-soil contact.

To test this hypothesis an experiment was conducted in which the effects of three concentrations of two different antidesiccants were investigated (Figure 1-1). Field survival, growth, and stomatal conductance of treated seedlings were compared with an untreated control.

The second hypothesis is that antidesiccants do not reduce desiccation if applied to actively growing seedlings before outplanting. This practice could be helpful in some climates such as Mexico and Central America where actively growing seedlings are outplanted. During seedling growth, short periods of moisture stress result in reduced growth and wilting injury (Kramer 1983). Moreover, desiccation can damage plant cells, and result in increased leakage of solutes from leaf and root tissues. To test this hypothesis, an experiment was conducted in which two antidesiccants at three concentrations were applied to actively growing seedlings which were then subjected to different periods of wind exposure (Figure 1-1). The damage caused by desiccation was evaluated by measuring electrolyte leakage from the shoot and root systems, which was compared with untreated seedlings. Water loss and stomatal conductance were also measured.

The third hypothesis states that antidesiccants and cold-storage conditions do not affect subsequent physiology, phenology, and morphology of 2+0 ponderosa pine seedlings. Of particular interest was light absorption and the effect antidesiccants might have on photosynthesis. Since chlorophyll *a* fluorescence is commonly used as a measure of the photochemical efficiency of photosystem II (Krause and Weis 1984; Öquist and Malmberg 1989), a decrease in maximum fluorescence of seedlings treated with antidesiccants could be evidence that antidesiccants do affect light absorption. To test this hypothesis, an experiment was performed with two antidesiccants at three concentrations applied to dormant seedlings (Figure 1-1). Variable fluorescence was used as an indirect measure of photosynthesis inhibition . Also, other variables such as seedling survival, root growth potential, height growth, diameter growth, and stomatal conductance of treated seedlings were measured and compared with an untreated control. Seedlings were evaluated in the greenhouse during one growing season.

Finally, the fourth hypothesis states that antidesiccants applied to the whole seedling do not initially reduce the amount of water loss without affecting the photochemical efficiency of ponderosa pine seedlings. To test this hypothesis, an experiment was performed where the effects of three concentrations of two antidesiccants and their effects on water loss of seedlings was studied under controlled environmental conditions (Figure 1-1). Also height growth and chlorophyll fluorescence were measured.

Experiment 1

Ho: Antidesiccants applied to dormant seedlings after lifting do not improve field survival and growth of ponderosa pine seedlings.

Dormant seedlings lifted ⇨ Six antidesiccants treatments applied to whole seedlings. ⇨ Treated and untreated seedlings outplanted at Warm Springs, OR. ⇨ Field survival and height growth were evaluated during two growing seasons. Stomatal conductance was evaluated the first growing season.

Experiment 2

Ho: Antidesiccants do not reduce desiccation when applied to actively growing seedlings before outplanting.

Actively growing seedlings lifted. ⇨ Six antidesiccants treatments applied to whole seedlings. ⇨ Treated and untreated seedlings were desiccated in a growth growth room over different time periods. ⇨ Water loss, stomatal conductance, and electrolyte leakage were evaluated for each period of desiccation.

Figure 1-1. Schematic outline of the experiments used to test the four hypotheses.

Experiment 3

Ho: Antidesiccants and cold-storage conditions do not affect subsequent physiology, phenology and morphology of ponderosa pine seedlings.

Dormant seedlings lifted. ⇨ Six antidesiccants treatments applied to whole seedlings. ⇨ cold storage ↗ 20 days / ↘ 40 days ⇨ Greenhouse test over a six month period ⇨ Survival, foliar damage, height and diameter growth, stomatal conductance, and chlorophyll fluorescence and RGP were evaluated.

Experiment 4

Ho: Antidesiccants applied to dormant seedlings initially do not reduce the amount of water loss without affecting photochemical efficiency.

Dormant seedlings lifted. ⇨ Six antidesiccants treatments applied to whole seedlings. ⇨ Growth room test over a 40 days ⇨ Survival, height growth, chlorophyll fluorescence, and water loss analysis were evaluated.

Figure 1-1. Continued.

References

Burdett, A.N. and D.G. Simpson. 1984. Lifting, grading, packaging, and storing. In Duryea, M.L. and T.L. Landis (eds). Forestry Nursery Manual: production of bareroot seedlings. Martinus Nijhoff / Dr. W. Junk Publishiers, The Hague. pp. 227-234

Chen, T.H.H., P. Murakami., P. Lombard, and L.H. Fuchigami. 1991. Desiccation tolerance in bare-rooted apple trees prior to transplanting. J. Environ. Hort. 9(1):13-17.

Cleary, B.D., R.D. Greaves, and P.W. Owston. 1988. Seedlings. In Cleary, B.D., R.D. Greaves, and R.K. Hermann (eds). Regenerating Oregon's Forests. A guide for the regeneration forester. Oregon State University. Extension Service. Fifth printing. Corvallis, OR. pp. 63-97.

Coutts, M.P. 1981. Effect of root or shoot exposure before planting on the water relations, growth, and survival of sitka spruce. Can. J. For. Res. 11(3):703-709.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects of antitranspirants on water relations and photosynthesis of woody plants. J.Amer. Soc. Hort. Sci. 99(4):297-304.

Garber, M.P. and J.G. Mexal. 1980. Lift and storage practices: their impact on successful establishment of southern pine plantations. N.Z. J. For. Sci. 10(1):72-82.

Johnson, J.D. and M.L.Cline. 1991. Seedling quality of southern pines. In Duryea, M.L. and P.M. Dougherty (eds). Forest Regeneration Manual. Kluwer Academic Publishers. The Netherlands. pp. 143-159.

Kramer, P.J. 1983. Water relations of plants. Academic Press. New York. 489 p.

Krause, G.H. and E. Weis. 1984. Chlorophyll fluorescence as a tool in plant physiology. II. Interpretation of fluorescence signals. Photosynthesis Res. 5:139-157.

Lefevre, R.E., A.C. Cameron, and N.C. Peterson. 1991. Influence of moisture loss during storage and new growth of conifer seedlings. J. Environ. Hort. 9(2): 92-96.

Levitt, J. 1972. Responses of plants to environmental stresses. Academic Press. New York. 697 p.

Marshall, J.D. 1985. Carbohydrates status as a measure of seedling quality. In Duryea, M.L. (ed). Proceedings: evaluating seedling quality: principles, procedures, and predictive abilities of a major test. Forest Research Laboratory. Oregon State University. pp. 49-58

Molina, R. and J.M. Trappe. 1984. Mycorrhiza management in bareroot nurseries. In Duryea, M.L. and T.L. Landis (eds). Forestry Nursery Manual: production of bareroot seedlings. Martinus Nijhoff / Dr. W. Junk Publishiers, The Hague. pp. 211-223.

Odlum, K.D. and S.J. Colombo. 1987. The effect of three film-forming antitranspirants on moisture stress of outplanted black spruce seedlings. Tree Planters' Notes. 38(4):23-26.

Öquist, G. and G. Malmberg. 1989. Light and temperature dependent inhibition of photosynthesis in frost-hardened and un-hardened seedlings of pine. Photosynthesis Res. 20:261-277.

Rowan, S.J. 1983. Loss of feeder roots lower seedling survival more than severe black root rot. Tree Planter's Notes. 34:18-20.

Sands, R. 1984. Transplanting stress in radiata pine. Aust. For. Res. 14(1):67-72.

Simpson, D.G. 1984. Filmforming antitranspirants: their effects on root growth capacity, storability, moisture stress avoidance, and field performance of containerized seedlings. Forestry Chronicle. 60(6):335-339.

Slatyer, R.O. and J.F. Bierhuizen. 1964. The influence of several transpiration suppressants on transpiration, photosynthesis, and water-use efficiency of cotton leaves. Aust. J. Biol. Sci. 17(1):131-146.

van den Driessche, R. 1989. Changes in osmotic potential of Douglas-fir (*Pseudotsuga menziesii*) in relation to temperature and photoperiod. Can. J. For. Res. 19:413-421.

Wakeley, P.C. 1965. Results of southern pine planting experiments in the middle twenties. J .Forestry. 67:237-241.

CHAPTER 2

LITERATURE REVIEW

General considerations

Three quarters of the water vaporized on land is transpired by plants (Nobel 1991). Plants act as a conduit which allows for the transfer water from the soil to the atmosphere. Water enters the seedling through roots, crosses the endodermis to reach the root xylem and moves within the xylem to the mesophyll and evaporation sites (Jarvis 1975). The driving force to move water against the gravitational component of total water potential and fractional resistances is derived from the evaporation of water vapor from the leaves (Novel 1991).

When transpiration exceeds water absorption, water stress develops in seedlings and growth is diminished (Kramer 1983; Burdett 1990; Pallardy et al. 1991). Water from the mesophyll will be lost first followed by water located in the more distal portions of the seedlings hydraulic system. In response to this water deficit gradient, cellular water moves down the energy gradient toward the xylem in an attempt to equilibrate and eliminate the potential difference. The amount of water transpired from the leaf may be significantly reduced by:

a) limiting opening of the stomata, and

b) coating transpiring surfaces with antidesiccants.

In both cases, the stomata play a major role in controlling the loss of water vapor from leaves and the entry of carbon dioxide (Nobel 1991). Stomata are the guardians of gas

exchange, providing paths for carbon dioxide intake while minimizing the inevitable flux of water vapor under continuously changing ambient conditions. The stomatal pore is formed between two guard cells which are specialized cells of the epidermis. The guard cells alter in turgor and volume during stomatal movements (Weyers and Meidner 1990).

History of antidesiccants

Cultural practices to protect trees against desiccation date back 2 ,294 years when the Greeks used mud and sawdust to prevent loss of water and reduce stem temperature of *Ficus* and *Ulmus* (Theophrastus 300 BC).

Even though antidesiccants were used around 300 BC, the first article about antidesiccants and transpiration from leaves and how to reduce it was not written until 1727 by Stephen Hales. He measured the quantities of water imbibed and transpired by plants and trees such as *Quercus* and *Larix*. To reduce transpiration leaves were coated with a mixture of tobacco, clay and bean flower (Hales 1961). Hales, also, made several attempts to give an artificial taste to fruits by making trees imbibe some perfumed liquor. According to antidesiccants classification (Gale and Hagan 1966) this practice can be classified as film-forming antidesiccants (when plants absorb alcohol through the root system).

For more than two centuries, following Stephen Hales' work, there was little attention given to antidesiccants. However, after World War II, the vinyl plastic industry developed new films and the interest for using antidesiccants with tree seedlings reemerged (Nauth 1947; Lebovits 1966). In the 1960's several studies were written

about antidesiccants. Most were done to determine the best chemical compounds, their toxicity effects on gas exchange and growth of field crops and conifers (Poljakoff-Mayber et al.1967).

Antidesiccant theory

A good antidesiccant must meet certain characteristics such as forming a good quality film on the leaf. The film coverage must be uniform without abnormalities, and it needs to control transpiration and reduce water loss, be effective under varying environmental conditions and, not be phytotoxic or detrimental to plant growth. (Poljakoff-Mayber et al.1967).

Permeability is common to all plastics and is an important factor in determining its usefulness as an antidesiccant to protect seedlings against desiccation. Mass transport through polymeric materials occurs by activated diffusion, the permeability constant is defined as the rate of permeation multiplied by the thickness of the membrane and divided by the product of its area and the difference in pressure between two environments separated by the membrane (Lebovits 1966).

The transmission mechanism of a gas or vapor through a plastic film is, in the absence of cracks or pinholes, of the active diffusion type (Stannett and Yasuda 1964). The process takes place in three steps. First, the permeant dissolves in the permeable membrane on the side of its higher concentration. Second, it diffuses through the membrane towards the side of the lower concentration, a process which depends on the formation of holes in the plastic network due to thermal agitation of the chain segments.

Finally, the permeant becomes absorbed on the side of the lower concentration. In contrast to this, the permeating molecules do not change from undissolved to dissolved, and do not form transient holes in this passage when permeating through porus materials (Lebovits 1966).

To determine permeability (P) as a mathematical expression, a series of calculations must be done to estimate the diffusion constant (D) and solubility coefficient (S). Under steady-state conditions the rate of flow of the gas, can be expressed by Fick's Law:

$$P = -D(\delta c / \delta x)$$

where P is the amount of gas passing through a unit area of film in the unit time, D is the diffusion constant, and $\delta c / \delta x$ is the concentration gradient.

The gas concentration is usually expressed in terms of pressure and is related with Henry's Law, which states that the saturation partial pressure in the vapor phase is proportional to the mol fraction of the solute in the liquid phase (Nobel 1991):

$$C = (S)(p)$$

where C is gas concentration, and p is pressure. S is the solubility coefficient for the particular gas or vapor in the antidesiccant in question.

The temperature dependence of permeability, diffusion and solubility can be expressed by Arrhenius equation:

$$\text{rate constant} = B^{-A/RT}$$

where B is a constant, a plot of the logarithm of the rate constant versus 1 / T is commonly known as an Arrhenius plot and (-A / R) is the slope (Nobel 1991). Finally, the equation to calculate antidesiccant permeability is:

$$P = D_o S_o \, [\,-(Ed + Hcond + Hm) \,/\, RT\,]$$

Where P is permeability constant; D_o is diffusion coefficient; S_o is solubility coefficient; Ed is energy for diffusion process; Hcond is heat of condensation; Hm is heat of mixing; R is gas constant; and T is temperature (K^o).

The units used for permeability constants are cubic centimeters of gas at standard temperature and pressure (STP) passing per second under a gradient of one centimeter of mercury pressure per millimeter thickness and per square centimeter of area (Stannet and Yasuda 1964).

There are three methods for determining the maximum flux: a) The concentration method relies on measuring the increase in concentration of the penetrant in an isolated receiving section of the test cell, b) the volume method is based on measuring the volume of the penetrant either in the gas or in the liquid phase, and c) the pressure method consist of the measurements of the pressure increase in a known receiving volume on the low-pressure side of the cell (Stannett and Yasuda 1964).

The following information (Table 2-1) on permeability of plastic films to oxygen, carbon dioxide and water vapor at 30^0 C , was reported by Stannett and Yasuda (1964) and Lebovits (1966).

Table 2-1. Permeability of polymers to oxygen, carbon dioxide and water vapor .

Chemical compound	Permeability to (cc.STP /cm/mm/sec/cm Hg x 10^{10}): Oxygen	Carbon dioxide	Water vapor
Polyvinylidene chloride	0.05	0.29	14 - 1000
Rubber hydrochloride	0.25 - 5.4	1.7 - 18.2	250 - 19000
Polyvinyl chloride	1.20 - 6.0	10.2 - 37.0	2600 - 6300
Polystyrene	15 - 250	75 - 370	10000
Silicone rubber	1000 - 6000	6000 - 30000	106000

More recently, it has been determined that wettability of the leaf surface; surface tension of the compound; contact angle; and stomatal frequency, distribution, and morphology influence the effectiveness of growth regulator antidesiccants (Schönherr and Bukovac 1972; Weyers and Meidner 1990).

Stomata are important because most of the water lost by transpiration escapes through them, and most of the carbon dioxide used in photosynthesis enters through them (Kramer 1983).

The stomatal pore is formed between two guard cells which are specialized cells of the epidermis. Penetration of the stomata by liquids is based on the theory of capillary rise. To get the equation to estimate the penetration of stomata by growth regulators, it

is necessary to determine the pressure difference across the liquid meniscus, the surface tension of the chemical compound, the principal radii of curvature of the liquid meniscus, the radius of capillary; and the contact angle formed by the antidesiccant advancing over a dry surface (Schönherr and Bukovac 1972).

The equation for a conical converging capillary is defined as:

$$P = 2\Upsilon L \sin(\phi 1 + \theta a) / r$$

where P is the pressure difference, ΥL is the tension of the liquid, $\phi 1$ is the conical capillary wall, θa is the advancing contact angle, and r is the radius of capillary.

The same equation can be used to estimate diverging capillary but is necessary to change the term $\phi 1$ for $\phi 2$.

The stomatal density and leaf morphology are important factors to consider before applying any kind of antidesiccant to plants. The neighboring cells, sizes, arrangements and cell wall characteristics of the cell, other than guard cells, are also important in bringing about changes in stomatal aperture. These changes of volume are due to osmotic water movement following the increased solute content of guard cells (Weyers and Meidner 1990).

Stomatal distribution on the leaf surface, differences in leaf morphology and surface topography are important factors to ponder when measuring stomatal conductance or applying antidesiccants. Stomata may be located below the plane of the epidermis, either in grooves or over-arched by epidermal cells. Sunken or recessed stomata may trap air bubbles in epidermis strips, making observations of the pore very difficult. The reduction in air leaf conductance due to the constriction to the vapor flux

pathway may be taken into account.

The cuticular conductance of non-stomata-bearing leaf epidermis may be 1-2 % of an epidermis with open stomata. The occurrence of hairs and other epidermal outgrowth is of importance for investigations with porometers and experiments involving epidermal strips. Trichomes make it difficult to achieve airtight seals between the leaf and apparatus. The presence of trichomes on the epidermis may increase the boundary layer thickness and they may affect the leaf 's reflectivity (Weyers and Meidner 1990).

Antidesiccant classes

Antidesiccants have been defined as any materials applied to plants for the purpose of retarding transpiration (Gale and Hagan 1966; Noggle and Fritz 1983). The antidesiccants are usually sprayed on plants in order to form a film on the surface of the leaves that will be more permeable to carbon dioxide and oxygen than to water vapor (Gale and Poljakoff-Mayber 1967).

Antidesiccants have been divided into four categories: 1) film-forming, 2) growth regulating, 3) stomatal regulating, and 4) reflective materials (Gale and Hagan 1966; Martin 1974; Tracy and Lewis 1981).

1) The film-forming category is further divided into a) thin-film and b) thick-film types.

a) The thin-film type usually is absorbed through the root system and transported to the mesophyll reducing the transpiration rate. The chemical compounds that have been used are hexadecalon and silicons. The reason why higher alcohols are used is that

the highly polar alcohol molecules are attracted to the water and the hydrophobic ends are repulsed, so that Van der Waals forces cause a tightly and symmetrically arranged monomolecular layer to form, which is highly impermeable to water vapor (Gale and Hagan 1966).

The Van der Waals forces are the electrostatic attractions between electrons in one molecule and the nucleus of an adjacent molecule minus the molecules' interelectronic and internuclear repulsive forces. Therefore, Van der Waals forces result from random fluctuations of charge and are important only for molecules that are very close together, especially, for neighboring molecules (Nobel 1991).

b) Thick-film types cover the stomata with a film whose resistance to water vapor transmission is greater than its resistance to carbon dioxide and oxygen. Different compounds and formulations have been used to reduce water loss. The permeability characteristic for each compound used in early research were different. Some of the ingredients studied were copolymers of acrylonitrile, vinylidene chloride, silicone, polyvinyl acetate, and hexadecalone (Gale and Hagan 1966).

2) The growth regulating category is better known as plant hormones. Water stress affects hormone balances, which in turn control plant developmental patterns. All phytohormones are affected by water stress, the most common information supports the hypothesis that abscisic acid, cytokinins, and ethylene are the most important in controlling water balance (Hale and Orcutt 1987).

Abscisic acid (ABA) has been implicated in several physiological responses of trees and other plant responses to water stress (Johnson 1991). Water deficits cause an

increase in ABA which accumulates primarily in the leaves. Stomatal closure during water stress is the best known response to ABA increases and appears to cause the afflux of potassium (K^+) from the guard cell, resulting in turgor loss and subsequent stomatal closure (Davis and Kozlwski 1975a; Hale and Orcutt 1987; Johnson 1991; Nobel 1991).

The initial response of plants to drought is a decrease in leaf water potential, which results in the release of ABA from mesophyll chloroplasts followed by a rapid synthesis of ABA . Upon rehydration and release from drought stress the ABA levels decrease but at a slower rate than the initial rate of increase (Hale and Orcutt 1987).

There are other growth regulators that reduce transpiration such as paclobutrazol, daminozine and PP333. The main effect of these chemicals is a reduction on shoot / root ratio that leads to a reduction in water uptake (Swietlik and Miller 1983; Sterrett 1985).

3) The stomatal regulating class of compounds is related with carbon dioxide (CO_2) concentration. The degree of stomatal opening often depends on the CO_2 concentration in the guard cells, which reflects their own carbohydrate metabolism as well as the carbon dioxide in the air within the leaf (Nobel 1991).

Elevated CO_2 concentrations depress transpiration by closing stomata and at the same time increase photosynthesis when light and other factors are not limiting (Gale and Hagan 1966). The degree of stomatal opening often depends on the CO_2 concentration in the guard cells, which reflects their own carbohydrate metabolism as well as the CO_2 level in the air within the leaf. For instance, upon illumination, the CO_2 concentration in the leaf intercellular air space is decreased by photosynthesis, resulting in decreased CO_2

levels in the guard cells, which somehow triggers stomatal opening (Nobel 1991).

4) The reflective materials are effective in reducing water loss in an indirect way. They do not act as a physical barrier to water vapor, nor do they directly affect stomata. Rather, they reflect solar radiation back from plant parts by reducing the energy input to the plant, the transpiration rate is reduced. Most of these compounds are white spray materials such as kaolinite or lime, which form a coating with a high reflectivity upon drying (Martin 1974). Reflective materials have been used to reduce trunk temperatures of fruit trees. The application of reflective material can be either alone or mixed with other antidesiccants (Gale and Hagan 1966).

Antidesiccant chemistry

Different materials and formulations have been tested for their possible use as antidesiccants such as polyethylene, acrylic polymer, silicone, carboxymethylcellulose, natural rubber, wax emulsion, carnauba wax, polyvinyl chloride, polyvinyl alcohol, terpenic polymer, and styrene butadiene. Some of them are still used by horticulturalists to prevent desiccation of plants and trees (Gale 1961; Poljakoff-Mayber et al.1967).

In this thesis two antidesiccants were tested. The first is Moisturin, a vinyl chloride monomer and vinylidene chloride monomer (Badertscher 1991). The second is Anti-Stress 2000, an acrylic polymer (Englert 1992).

Acetylene and ethylene can be used to produce the compound vinyl chloride, $CH_2 : CHCl$. Vinyl chloride is a gas that can be manufactured by passing acetylene gas through a mixture of $CuCl_2$, NH_4Cl and concentrated hydrogen chloride (HCl). The

resulting reaction produces a gas, vinyl chloride, and some unreacted acetylene which passes through the autoclave where small molecular compounds of the monomer vinyl chloride are obtained.

On the other hand, petroleum and brine are used in fabricating vinylidene chloride polymers (CH_2:CCl). Ethylene is obtained by cracking petroleum and chlorine by the electrolysis of brine, which is combined in the presence of molten metal salts, to form trichloroethane ($CH_2Cl \cdot CHCl_2$) which is converted to vinylidene chloride when treated with lime [$Ca(OH)_2$]. To produce the vinylidene chloride monomer, asymmetric trichloroethane is reacted with alkali (Nauth 1947; Schildknecht 1952).

There are different antidesiccant and surfactant brands on the market that are used to increase resistance to moisture loss on shoots, roots or both (Table 2-2).

Antidesiccant phytotoxicity

A number of agricultural chemicals and biocides such as insecticides, herbicides and antitranspirants adversely affect the growth and development of plants. Commonly, some of these chemicals inhibit photosynthesis by blocking stomata or causing changes in optical properties of leaves, heat balance of leaves, leaf metabolism, leaf anatomy, or by any combination of these (Kozlowski and Mudd 1975).

Film-forming coatings can be either non-toxic, toxic or highly toxic to plants. Antidesiccant efficacy can be species specific and since toxic side-effects can occur, it is advisable to test any product on a per species basis before field use (Colombo and Odlum 1987). As an illustration, Tag® (polyethylene) antidesiccant is toxic to beans

Table 2-2. Antidesiccants, surfactants and principal ingredients commonly used to reduce transpiration and root desiccation on plants.

Commercial name	Main ingredient (s)	Reference
Antidesiccants:		
Adol 52	Cetyl alcohol, C_{14} and C_{18} alcohols	Slatyer and Bierhuisen 1964
Adkar Cloud Cover	Acrylic polymer	Perkins et al. 1991
AntiStress 2000	Acrylic polymers	Englert 1992
Clear Spray	Acrylic polymer	Simpson 1984
Folicote	Hydrocarbon wax-emulsion	Tracy and Lewis 1981
Keykote	Plastic-wax	Davies and Kozlowski 1974
Moisturin	Vinyl chloride monomer and Vinylidene chloride monomer	Badertscher 1991
Paclobutrazol	*	Swietlik and Miller 1983
PMA	Phenylmercuric acetate	Waisel et al. 1969
Saran	Polyvinylidene chloride	Poljakoff-Mayber et al. 1967
Tag	Polyethylene	Poljakoff-Mayber et al. 1967
Vapor gard	Poly-1-p-menthene-8,9-diyl	Shekour et al. 1991
Wilt Pruf	di-1-p-menthene	Steinberg et al. 1990
XF-4-3561 Fluid	Silicone emulsion	Lee and Kozlowski 1974
Surfactants:		
Atplus 401	Anionic surfactant blend	Colombo and Odlum 1987
Canplus 300	Polyethylene polyol fatty and acid esters	Colombo and Odlum 1987
Moisturite	Starch-grafted polyacrylate polymer	Ingram and Yeager 1987
Terra-Sorb	**	Ingram and Yeager 1987
Triton B-1956	Polymer	Ranney et al. 1984
Tween 20	Polyethylene polyol fatty and Sorbitan monolaurate	Colombo and Odlum 1987
Waterlock	Polymer	Magnussen 1985

*[(2RS,3RS)-1-(4-chlorophenyl)-4,4-dimetyl-2-1-(1,2,4-triazol-1-yl-) pentan-3-ol]
** Saponified starch-graft polyacrylonitrile copolymers

but is not toxic to bananas, vines, sugar beets and pines (Poljakoff-Mayber et al.1967).

Colombo and Odlum (1987) tested six antitranspirants on black spruce and concluded that 28 days after they applied Vapor Gard (di-1-p-menthene) to seedlings, the foliage was damaged due to browning of the needles, and in the most severe cases, terminal buds were dead. Moreover, Lee and Kozlowski (1974) studied the toxicity effect of silicone emulsion antitranspirant on *Fraxinus americana* L, *Acer saccarum* Marsh. and *Pinus resinosa* Ait. As a result, they concluded that seedlings of these species treated with 50% silicone showed severe toxicity symptoms and their leaves died within six weeks after treatment.

Antidesiccant effects on photosynthesis

Several researchers suggest that when applied, film antidesiccants reduce transpiration and photosynthesis by roughly the same amount because antidesiccants form an impermeable barrier to both carbon dioxide and water vapor (Gale and Poljakoff-Mayber 1967; Davies and Kozlowski 1974; Lee and Kozlowski 1974; Olofinboba et al. 1974; Ceulemans et al. 1983; Noggle and Fritz 1983).

Consequently, photosynthesis components (photochemical, electron transfer, and biochemical) can be affected by antidesiccants. The most obvious effects are on carbon dioxide and light absorption because some antidesiccants have low permeability to gases and high refractive indices.

Film-forming antidesiccants may reduce water loss from plants either by decreasing absorption of radiant energy, reflecting incident light or by forming thin

films on leaves which reduce cuticular and stomatal transpiration (Olofinboba et al. 1974).

The transpiration process involves the evaporation of water from cell walls and its diffusion out of the leaves through the stoma into the turbulent air. Carbon dioxide diffuses across the same pathway as the water vapor does but in the reverse direction. Plants treated with film-forming antidesiccants have an additional source of resistance in the pathways for entry of carbon dioxide and the exit of water vapor. However, under conditions of water stress, seedlings treated with antidesiccants may in fact have increased photosynthesis compared to untreated seedlings grown under the same conditions (Gale and Poljakoff-Mayber 1967; Nobel 1991).

Normally, the pathway of water vapor movement is generally across a thin waxy layer on the cell walls within the leaf. After crossing the waxy layer, the water vapor diffuses through the intercellular air space and then through the stoma to reach the boundary layer adjacent to the leaf surface (Nobel 1991). However, with the film-forming antidesiccant , water vapor must diffuse through additional resistance (Poljakoff-Mayber et al. 1967).

Carbon dioxide diffuses from the air, across the boundary and film-forming, layers through the stomata, across the intercellular air space, into the mesophyll cells, and eventually into the chloroplast. Carbon dioxide diffusing from the air through the cell walls encounters 60% more resistance than water vapor diffusing in the opposite direction over the same pathway without the film-forming resistance (Nobel 1991).

There are some studies that have examined antidesiccant effects on transpiration and photosynthesis in conifers. Davies and Kozlowski (1975b), determined that *Pinus resinosa* Ait. seedlings sprayed with six different antidesiccants had reduced transpiration and net photosynthesis within 24 hours after treatment. Thirty-two days after treatment, all of the antidesiccants significantly reduced water loss. Dow Silicone had a 95 % reduction in comparison with an untreated control. Net photosynthesis rate was reduced significantly at 32 days after treatment. With the same antidesiccant, seedlings were photosynthesizing at approximately one tenth the rate of untreated control plants. With the same species, only treated with a 5 % Silicone emulsion antitranspirant, a reduction in transpiration by about 90% during ten days without affecting chlorophyll content of old needles (Lee and Kozlowski 1974).

The inhibition of transpiration and photosynthesis in azaleas treated with Folicote (hydrocarbon wax-emulsion) antidesiccant also was studied by Ceulemans et al. (1983). They concluded that during the first and second day following the antidesiccant treatment, stomatal conductance was lowered by 46% and the net carbon dioxide exchange rate decreased by 44%.

Antidesiccants effects on water efficiency

Optimizing water use efficiency has been of interest to physiologists in terms of photosynthesis for many years (Kramer 1983). Stomatal opening leads to the carbon dioxide uptake necessary for photosynthesis but in the process, plants lose water (Nobel 1991). There are several approaches to improving water use efficiency. These include

plant breeding, management of stress tolerance, and the application of antidesiccants (Kramer 1983). Water loss in plants can be reduced by growth regulating antitranspirants that induce stomatal closure or film-forming antitranspirants which coat the leaf surface and reduce cuticular and stomatal transpiration (Davies and Kozlowski 1974; Olofinboba et al. 1974; Nobel 1991). Moreover, it has been shown that film-forming polymer materials are more permeable to water than carbon dioxide by a factor of at least four (Davies and Kozlowski 1974).

Growth regulating antidesiccants such as paclobutrazol have an inhibitory effect on shoot and root growth of red maple, apple seedlings, yellow-poplar and American sycamore (Sterrett 1985). In response to the chemical, plants have an osmotic adjustment to lower water potential that enables turgor maintenance (Swietlik and Miller 1983).

The water use efficiency (WUE) can be calculated as:

$$WUE = \text{mol } CO_2 \text{ fixed / mol } H_2O \text{ transpired}$$

As previously noted azaleas treated with Folicote (hydrocarbon wax-emulsion) antidesiccant under a controlled environment showed a reduction in transpiration rate by about 45% and simultaneously the net carbon exchange rate decreased by about 44%. However, there was no influence of antidesiccant treatments on water use efficiency (Ceulemans et al. 1983). Contrary to these results, in another study antidesiccants reduced water use of potato plants grown in a greenhouse by 20-40 % depending on the antidesiccant concentration (Lipe and skinner 1979).

Antidesiccants and forest tree seedlings

During the 1960's interest in using antidesiccants to reduce transpiration of forest tree seedlings was renewed by Gale (1961), Slatyer and Bierhuizen (1964), Gale and Poljakoff-Mayber (1967). The main objectives were to study the effect antidesiccants had in reducing the water requirements and improving survival and growth of seedlings after outplanting.

A large number of film-forming antidesiccants were screened on forest tree seedlings. Of these a few were found which formed good, non-toxic films on plant leaves, and which also reduced transpiration by 20-40 %. Under controlled conditions *Pinus halepensis* Mill. seedlings sprayed with antidesiccants had reduced transpiration and increased the growth but the results in the field were disappointing (Poljakoff-Mayber et al.1967). Sometimes, however, there were no statistical differences among treatments (Williams et al. 1990).

Antidesiccant effects on leaf temperature, transpiration, photosynthesis, plant water balance, and soil moisture content have been studied in conifers. The results are not consistent for the same species and sometimes for the same antidesiccant compound (Gale and Poljakoff-Mayber 1965; Poljakoff-Mayber et al.1967; Davies and Kozlowski 1974; Davies and Kozlowski 1975b).

Aqueous emulsions containing 1 to 10 % silicone were effective in controlling transpiration of numerous woody angiosperms and gymnosperms (Lee and Kozlowski 1974). Furthermore, paclobutrazol is a potential injectable bioregulator for controlling transpiration and growth of woody plants such as red maple, yellow poplar, white ash,

and American sycamore. It is not phytotoxic even at high concentrations (Sterret 1985).

Antidesiccants and horticulture

As previously mentioned, early investigations on the effect of antidesiccants gave very inconsistent results (Gale 1961). Different types of compounds have been developed and tested on several crops and fruit trees and a few on ornamental plants (Ceulemans et al.1983). The results have varied widely with the same compound and dilution used. For example, polyvinyl coatings caused damage and decreased survival of transplanted citrus nursery stock. Hexadecalon reduced transpiration of tomato plants and doubled the yield. Silicone compounds reduced transpiration by about half in sun flower plants and increased the photosynthesis-transpiration ratio of sugar beets (Gale 1961; Lee and Kozlowski 1974).

Usually transpiration and the rate of carbon dioxide fixation are reduced 24 hours after applying the antidesiccants, reduction rates are between 26-55 % and 20-65 % respectively (Waisel et al.1969; Davies and Kozlowski 1974; Olofinboba et al.1974; Sinclair et al.1975; Tracy and Lewis 1981). For example in pepper, maize and tomato plants the rate of carbon dioxide fixation was decreased by 65.6 % , 50.7 % and 24.5 % respectively (Kastori et al.1991).

Applying growth regulators (Paclobutrazol) to apple seedlings showed that seedlings can adapt to water stress by osmotic adjustment and possibly through increased root dry weight and increased root to leaf ratio (Swietlik and Miller 1983). The same chemical injected into apple seedlings suppressed height growth and reduce leaf size

(Sterrett 1985).

Film-forming antidesiccants had a significant effect on protecting the leaves from powdery mildew *Sphaerotheca panossa* (Wallr. ex.Fr) var. *rosae* Wor. (Hagiladi and Ziv 1986). Moreover, film-forming antidesiccants increased fruit size (olives, cherries, and peaches), improved yields, and reduced moisture stress and irrigation requirements (Lipe and Skinner 1979).

Conclusions

There are four categories of antidesiccants. The efficiency of these varied greatly with species of plants, although research has shown positive results in some species. Antidesiccants have been used with different goals such as reduction of water loss in cold storage or after planting; improving survival and growth of transplanted forest tree seedlings as well as flowers; and increasing total yield of agricultural crops.
Both transpiration rate and carbon dioxide fixation have been reduced after applying antidesiccants (Olofinboba et al. 1974; Kastori et al. 1991). It is important to point out that antidesiccants increased leaf water potential of transplanted trees (Davenport et al. 1972) and potato plants (Kyaw et al. 1991). Also, Davenport et al. (1972), pointed out that reduced photosynthesis would be expected to reduce growth but growth is also a function of cell expansion which depends on maintenance of high turgor. Since antidesiccants act to increase turgor, it follows that applying antidesiccants could alter growth considerably. Taken together these observations suggest that antidesiccants could play an important role in reducing seedling transplanting shock.

References

Badertscher, D. 1991. Moisturin. Burke's protective coatings. Technical Bulletin. Washougal. WA. 5 p.

Burdett, A.N. 1990. Physiological process in plantation establishment and the development of specifications for forest planting stock. Can. J. For. Res. 20:415-427.

Ceulemans, R., R.Gabriels, and I. Impens. 1983. Antitranspirant effects on transpiration, net CO_2 , exchange rate and water-use efficiency of azalea. Scientia Horticulture. 19:125-131.

Colombo, S.J. and K.D. Odlum. 1987. Efficacy of six antitranspirants on black spruce container seedlings. New Forest. 3:239-244.

Davenport, D.C., P.E. Martin, and R.M. Hagan. 1972. Antitranspirants for conservation of leaf water potential of transplanted citrus trees. HortScience. 7(5):511-512.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects of antitranspirants on water relations and photosynthesis of woody plants. J. Amer. Soc. Hort. Sci. 99(4):297-304.

Davies, W.J. and T.T. Kozlowski. 1975a. Effect of applied abscisic acid and silicone on water relations and photosynthesis of woody plants. Can. J. For. Res. 5:90-96.

Davies, W.J. and T.T. Kozlowski. 1975b. Effects of applied abscisic acid and plant water stress on transpiration of woody angiosperms. For. Sci. 21(2):191-195.

Englert, J.M. 1992. Physiological and cultural conditions affecting postharvest handling of bare-rot nursery plants. MSc. Thesis. Oregon State University. 79 p.

Gale, J. 1961. Studies on plant antitranspirants. Physiol. Plant. 14:777-786.

Gale, J. and R.M. Hagan. 1966. Plant antitranspirants. Ann. Rev. Plant Physiol. 17:269-282.

Gale, J. and Poljakoff-Mayber, A. 1965. Effects of antitranspirants treatments on leaf temperatures. Plant and cell Physiol. 6:111-115.

Gale, J. and Poljakoff-Mayber, A. 1967. Plastic films on plants as antitranspirants. Science. 156:650-652.

Hagiladi, A. and O. Ziv. 1986. Use of antitranspirants for control of powdery mildew on field roses. J. Environ. Hort. 42(2):65-68.

Hale, M.G. and D.M. Orcutt. 1987. The physiology of plants under stress. John Willey and Sons. 206 p.

Hales, S. 1961. Vegetable staticks. First published 1727. Oldbourne Book Co. Great Britain. 214 p.

Ingram, D.L. and T.H. Yeager. 1987. Effects of irrigation frequency and water absorbing polymer amendment of Ligustrum growth and moisture retention by a container medium. J. Environ. Hort. 5(1):19-21.

Jarvis, P.G. 1975. Water transfer in plants. In de Vries, D.A. and N. Algan (eds) Heat transfer in the biosphere. I. Transfer processes in plant environment. John Wiley and Sons. New York. pp. 369-394.

Johnson, J.D. 1991. Measuring stress-related plant growth regulators. In Lassoie, J.P. and T.M. Hinckley (eds). Techniques and approaches in forest tree ecophysiology. CRC Press. Boca Raton, Florida. pp. 275-292.

Kastori, R., N. Petrovic., and Z. Stankovic. 1991. Possibilities of using antitranspirants in plant production. Bioloski-Vestnik. 39(1-2):109-113.

Kozlowski, T.T. and J.B. Mudd. 1975. Responses of plants to air pollution. In Kozlowski, T.T. and J.B. Mudd (eds). Responses of plants to air pollution. Academic Press. New York. pp. 1-8.

Kramer, P.J. 1983. Water relations of plants. Academic Press. New York. 489 p.

Kyaw, W., G.A. Berkowitz, and M. Henninger. 1991. Antitranspirants-induced increases in leaf water potential increase tuber calcium and decrease tuber necrosis in water-stressed potato plants. Plant Physiol. 96:116-120.

Lebovits, A. 1966. Permeability of polymers to gases, vapors, and liquids. Modern Plastics. 43(7):139-213.

Lee, K.J. and T.T. Kozlowski. 1974. Effects of silicone antitranspirant on woody plants. Plant and Soil. 40:493-510.

Lipe, W.N. and J.A. Skinner. 1979. Effect of an antitranspirant on water use, growth, and yield of greenhouse-grown potatoes. HortScience. 14(3):239-241.

Magnussen, S. 1985. Effects of root-coating with polymer water lock on survival and growth of drought-stressed bareroot seedlings of white spruce(*Picea glauca* Moench) and red pine (*Pinus resinosa* Ait). Tree Planters' Notes. 37(1):15-19.

Martin, J.D. 1974. Antitranspirants, possible uses in floriculture. Flor. Rev. 115(4051):19-21, 130-138.

Nauth, R. 1947. The chemistry and technology of plastics. Reinhold Publishing Co. New York. 522 p.

Nobel, P.S. 1991. Physicochemical and environmental plant physiology. Academic Press, Inc. 635 p.

Noggle, G.R. and G.J. Fritz. 1983. Introductory plant physiology. Prentice-Hall, Inc. New Jersey. 627 p.

Olofinboba, M.O., T.T. Kozlowski, and P.E. Marshall. 1974. Effects of antitranspirants on distribution and utilization of photosynthate in *Pinus resinosa* seedlings. Plant and Soil. 40:619-635.

Pallardy, S., J.S. Pereira, and W.C. Paerker. 1991. Measuring the state of water in tree systems. In Lassoie, J.P. and T.M. Hinckley (eds). Techniques and approaches in forest tree ecophysiology. CRN. Boca Raton, FL. pp. 27-74.

Perkins, T.G., G.T. Adams, and R.M. Klein. 1991. Desiccation or frezing?. Mechanisms of winter injury to red spruce foliage. Amer. J. Bot. 78(9):1207-1217.

Poljakoff-Mayber, A., J. Gale., H. Aluma., I. Kahane., H. Ginsburg., Z. Goldschmidt., I. Nir., and E. Shabi. 1967. Final technical report on effect of plant antitranspirants on certain physiological processes of forest seedlings and other plant material. Project No.A10-FS-10. The Hebrew University of Jerusalem. Jerusalem. 107 p.

Ranney, T.G., N.L. Bassuk, and T.H. Whitlow. 1989. Effect of transplanting practices on growth and water relations of 'Cold' cherry trees during reestablishment. J. Environ. Hort.7(1):41-45.

Schildknecht, C.E. 1952. Vinyl and related polymers. John Willey and Sons, Inc. New York. 723 p.

Schönherr, J. and M.J. Bukovac. 1972. Penetration of stomata by liquids. Plant Physiol. 49:813-819.

Shekour, G.M., C.R. McDavid, and R.A.I. Brathwaite. 1991. Response of sweetcorn to different rates and frequencies of application of PMA and Vapor Gard. Tropical Agriculture. 68(3):225-230.

Simpson, D.G. 1984. Filmforming antitranspirants: their effects on root growth capacity, storability, moisture stress avoidance, and field performance of containerized conifer seedlings. Forest Chronicle. 60(6):335-339.

Sinclair, T.R., G.E. Bingham., E.R. Lemon and L.H. Allen, Jr. 1975. Water use efficiency of field-grown maize during moisture stress. Plant Physiol. 56:245-249.

Slatyer, R.O. and J.F. Bierhuizen. 1964. The influence of several transpiration suppressants on transpiration, photosynthesis, and water-use efficiency of cotton leaves. Aust. J. Biol. Sci. 17(1):131-146.

Stannett, V. and H. Yasuda. 1964. Permeability. In Raff, R.A.V. and K.W. Doak (eds). Crystalline olefin polymers. Part II. High Polymers. Vol. XX. Interscience Publishers. New York. pp. 131-184.

Steinberg, S.L., M.J. McFarland, and J.W. Worthington. 1990. Antitranspirant reduce water use by peach trees following harvest. J. Amer. Soc. Hort. Sci. 115(1):20-24.

Sterrett, J.P. 1985. Paclobutrazol: a promising growth inhibitor for injection into woody plants. J. Amer. Soc. Hort. Sci. 110(1):4-8.

Swietlik, D. and S.S. Miller. 1983. The effect of paclobutrazol on growth and response to water stress of apple seedlings. J.Amer. Soc. Hort. Sci. 108(6):1076-1080.

Theophrastus. 300 BC. Enquiry into plants and minor work on odours and weather signs. Book IV: Of the trees and plants special to particular districts and position. William Heinemann. London.

Tracy, T.E. and A.J. Lewis. 1981. Effects of antitranspirants on hydrangea. HortScience. 16(1):87-89.

Waisel, Y., G.A. Borge, and T.T. Kozlowski. 1969. Effects of phenylmercuric acetate on stomatal movement and transpiration of excised *Betula papyrifera* Marsh. leaves. Plant Physiol. 44:685-690.

Weyers, J.B.D. and H. Meidner. 1990. Methods in stomatal research. Longman Scientific and Technical. Great Britain. 233 p.

Williams, P.A., A.M. Gordon and A.W. Moeller. 1990. Effects of five antitranspirants on white spruce and white pine seedlings subjected to greenhouse drought. Tree Planters' Notes. 41(1):34-38.

Chapter 3

THE INFLUENCE OF TWO ANTIDESICCANTS ON THE STOMATAL CONDUCTANCE AND FIELD PERFORMANCE OF 2+0 PONDEROSA PINE (*Pinus ponderosa* Dougl.) SEEDLINGS

Abstract

Seedling desiccation is a threat to newly planted seedlings. However, improvements in survival and growth of newly planted seedlings can potentially be achieved by modifying seedling water balance through a reduction in transpiration.

In this experiment, the effects of two antidesiccants on stomatal conductance, survival, and height growth of 2+0 ponderosa pine seedlings (*Pinus ponderosa* Dougl.) were evaluated over a two-year period near Warm Springs, Oregon. Three different concentrations of the antidesiccants Moisturin® and AntiStress 2000® were applied to seedlings prior to outplanting for a total of six treatments. A seventh treatment was used as an untreated control. Stomatal conductance was measured three times per day once in June, July, and August of the first year following outplanting. Seedling survival and height growth were measured annually for two years.

Stomatal conductance differed during each of the three months measured. In June, several antidesiccant treatments significantly reduced stomatal conductance. Also, there were differences in stomatal conductance within a measurement date due to time of measurement. Moreover, the interaction of time of measurement and antidesiccant treatment was highly significant. During July and August, no significant differences in stomatal conductance due to antidesiccant treatments but there were significant

differences due to time of measurement. Antidesiccants had no significant effects on seedling survival and height increment during the two growing seasons of this study.

Introduction

Seedling performance on a reforestation site depends on inherent growth potential at the time of planting and the degree to which field site environmental conditions allow this potential to be expressed (Grossnickle et al. 1991). Generally, seedling performance is diminished during the processes of lifting, grading, storing, shipping, and planting (Rietveld 1989). Following outplanting, seedlings can loss water rapidly. Desiccation has been identified as the most common problem in the survival of the seedlings (Burdett 1990). Transplanting stress describes the water-stressed condition of seedlings after outplanting. Seedlings need to recover from injuries caused during lifting and planting in order to reestablish the normal plant water relations (Sands 1984).

Although seedlings are able to reduce desiccation by stomatal closure, they do so at the cost of decreasing the inward diffusion of carbon dioxide with a subsequent reduction in photosynthesis (Gale 1961; Gale and Hagan 1966). Water stress induces stomatal closure in newly planted seedlings and can cause limited root growth because of reduced photosynthesis. Thus alleviating water stress can promote stomatal opening and the assimilation of carbon dioxide, photosynthesis and hence root growth through increased photosynthesis (Burdett 1990).

Antidesiccants are chemicals that reduce water loss from plants. Film-forming antidesiccants act as a barrier preventing water loss from the stomata. When soil water availability is low, antidesiccants might help to maintain plant water potential at a sufficiently high level, such that transpiration and photosynthesis may be greater than an untreated control (Gale 1961).

Although considerable work has been reported with antidesiccants on perennial tree fruit crops and flowers to increase the size of ripening fruits and durability of flowers, relatively less information is available on forest tree species (Martin 1974). Antidesiccants have also been shown to help plants reduce transpiration and minimize diseases (Martin 1974; Hagiladi and Ziv 1986; Tracy and Lewis 1981).

For forest tree seedlings, antidesiccants deserve consideration because of their potential benefit of minimizing moisture loss after planting (Colombo and Odlum 1987). Reduction of water loss after outplanting permits tree seedlings to maintain turgor until the root system can reestablish good soil contact and allows seedling survival with minimal injury (Gale and Hagan 1966; Colombo and Odlum 1987; Marshall et al. 1991). Antidesiccants such as latex emulsion, polyvinyl waxes, polyethylene, and higher alcohols such as hexadecalon have been applied with mixed results. Their effectiveness seems to depend on the nature of the chemical, the species, seedling age, plant growth rate, and atmospheric conditions (Kramer 1983).

The hypothesis tested in this experiment was that the application of antidesiccants on dormant seedlings after lifting will not improve field survival and growth of ponderosa pine (*Pinus ponderosa* Dougl.) seedlings by influencing plant water relations.

Methods

Plant Material and Experimental Treatments

Two-year-old (2+0) ponderosa pine seedlings were operationally grown at the USDA Forest Service Bend Pine Nursery in Bend, Oregon. A total of 500 seedlings were obtained from the nursery and transported to Oregon State University (OSU), Corvallis, Oregon. Seedlings were kept in a cold room at 5^0C for two weeks prior to outplanting. On March 18, seedlings were treated with two antidesiccants (Moisturin® and AntiStress 2000®), each with three different concentrations (Table 3-1). The concentrations tested were based on the manufacturer's recommendations. Treatments were applied to whole seedling. A group of randomly selected seedlings served as an untreated control. Seedlings were washed and then dipped in the different antidesiccants concentrations. After the antidesiccants dried on the seedlings (approximately 10 minutes), seedlings were put into plastic bags and placed in card board boxes in the cold room for two days.

Study establishment

The experiment was located on the Warm Springs Indian Reservation in Oregon on the east slope of the Cascade Mountains at a site called Wolford Canyon (Lat. 44^0 46' 24" N, Long. 121^0 19' 21" W, elevation 1324 m). The main plant community at the study area was dominated by ponderosa pine with an understory of grasses. The stand was clearcut harvested and the site broadcast burned in the fall 1990. The following planting season the site was reforested with P+1 ponderosa pine seedlings but

Table 3-1. Antidesiccant treatments applied to 2+0 ponderosa pine seedlings.

Treatment	Antidesiccant
1	Untreated control
2	Moisturin 1:3[1]
3	Moisturin 1:5
4	Moisturin 1:7
5	AntiStress 2000 1:20
6	AntiStress 2000 1:30
7	AntiStress 2000 1:40

[1] Antidesiccant:Water
Moisturin® (Vinyl chloride monomer and vinylidene chloride monomer). Burke's Protective Coatings, Washougal, WA.
AntiStress 2000® (Acrylic polymers). Polymer Ag, Inc., Fresno, CA.

more than 90% of the seedlings died. There was no additional site preparation before March 1992. Seedlings treated with antidesiccants and untreated seedlings were outplanted at 1.50 x 1.50 m spacing on March 25, 1992 under favorable conditions. The competing vegetation was not controlled after outplanting.

Measurements

Stomatal conductance (mmol $m^{-2}s^{-1}$) was measured once a month during June, July, and August 1992 with a portable LI-1600 steady state porometer (LI-COR, Inc. Lincoln, Nebraska). For each month of measurement, stomatal conductance was

monitored three times per day (0730, 0930 and 1130 h) on one day per month. To determine antidesiccant effects on stomatal conductance, two seedlings per treatment per block were measured. The same seedlings and fascicles were measured each time. Four fascicles per seedling per treatment per block were chosen to measure needle stomatal conductance. Needle surface area was determined measuring the projected leaf of each needle and was then multiplied by 5.0 cm (cuvette diameter). Therefore, the measured needle area was 7.72 cm^2.

Seedling height and survival were measured three times. Shoot height was first measured seven days after outplanting. This was defined as the vertical distance from the ground line to the tip of the terminal leader (Mexal and Landis 1990). The second height measurement was done at the end of the 1992 growing season. Finally, the last height measurement was taken on September 2, 1993. Seedling survival was recorded at the same time as seedling height.

Experimental design and statistical analysis

To test the effect of antidesiccants on stomatal conductance, survival and height growth, a randomized complete block design with four blocks was used. There were seven treatments per block and 16 seedlings per treatment. In total, 448 seedlings were evaluated. Treatments were randomly assigned within each block. Data were analyzed using SAS general linear procedure (SAS Institute Inc. 1993).

Because stomatal conductance was measured three times per day in each of three months, a repeated measurement analysis for a randomized complete block design was performed to analyze stomatal conductance data (Gumpertz and Brownie 1993). The

analyses were performed by date and time of measurement. The Least Significance Difference (LSD) test was used to determine significant differences among treatment means at the α=0.05 level for those effects found to be significant by analysis of variance.

Results

Stomatal conductance

The repeated analysis of variance for June 27 showed that there was a significant antidesiccant effect on stomatal conductance (p=0.0001)(Table 3-2). Stomatal conductance at time of measurement also was significantly different (p=0.0067). Moreover, the interaction of time of measurement and antidesiccant treatment was highly significant (p=0.0001). There was also a significant blocking effect that increased the precision of the analysis for the June data (Table 3-2).

In July, no significant antidesiccant effect on stomatal conductance was found but a significant effect on stomatal conductance due to the time of measurement was identified (p=0.0001)(Table 3-2). As with the June measurements, it was found that blocking increased the precision of the analysis (p=0.0084).

The analysis of data obtained in August showed that there was no significant antidesiccant effect on stomatal conductance (p=0.0854) but there was a significant effect due to the time of measurement (p=0.0001)(Table 3-2). Again, blocking was significant (p=0.0001).

The stomatal conductance patterns were different during the three-month measurement period in 1992. In June the analysis of variance by time of measurement, 0730, 0930, and 1130 h, showed that antidesiccant treatments had a significant effect on stomatal conductance regardless of time of measurements (Table 3-3). However, in July the opposite was true. Analysis of variance showed no significant effect due to antidesiccant treatments regardless of time of measurement. Whereas on August 29 analysis of variance showed that antidesiccant treatment had a significant effect on stomatal conductance ($p=0.0070$) but only at 0900 h.

On June 27 there was a significant reduction in stomatal conductance of seedlings coated with several of the antidesiccants treatments (Table 3-4). However, this varied by time of measurement. Moisturin (1:3 and 1:5) and AntiStress 2000 (1:20 and 1:40) were the most effective antidesiccant treatments in significantly reducing stomatal conductance at 0730 h with 2.39, 3.59, 3.93, and 2.91 mmol $m^{-2}s^{-1}$, respectively. AntiStress (1:20) was probably the most effective treatment in reducing the stomatal conductance during the day. For these seedlings stomatal conductance was never greater than 3.93 mmol $m^{-2}s^{-1}$.(Table 3-4). Seedlings treated with Moisturin (1:3) were the only treated seedlings that gradually increased stomatal conductance (6.05 mmol $m^{-2}s^{-1}$) through 1130 h. The other treatments, including the untreated control, had diminished stomatal conductance at this time (Table 3-4).

Making a comparison between the untreated control and Moisturin (1:3), the needle stomatal conductance was decreased by 69 % at 0730 h. When the seedlings were measured again at 0930 h they also showed statistically significant differences

Table 3-2. Summary of repeated measurements analyses of variance of effects of antidesiccants on stomatal conductance of 2+0 ponderosa pine measured in 1992. Values are probability of a greater F-value.

Source of variation	June 27	July 25	August 29
Block	0.0452	0.0084	0.0001
AT[1]	0.0001	0.2262	0.0854
Time (T)	0.0067	0.0001	0.0001
T x AT[1]	0.0001	0.7853	0.1070

Table 3-3. Summary of repeated measurements analyses of variance of effects of antidesiccants on stomatal conductance of 2+0 ponderosa pine seedlings by date and time of measurement in 1992. Values are probability of a greater F-value.

Time / source of variation	June 27	July 25	August 29
0730 h			
Block	0.5611	0.0014	0.0001
AT[1]	0.0001	0.2629	0.4820
0930 h			
Block	0.1543	0.1396	0.0001
AT[1]	0.0004	0.3968	0.0070
1130 h			
Block	0.0052	0.0001	0.0001
AT[1]	0.0046	0.5146	0.1234

AT[1] = Antidesiccant treatments.

Table 3-4. Treatment mean values for stomatal conductance (mmol $m^{-2}s^{-1}$) of 2+0 ponderosa pine seedlings on June 27,1992 by time of measurement.

Treatment	Time of measurement (h)		
	0730	0930	1130
Untreated control	7.83a	6.06b	4.86a
Moisturin (1:3)	2.39b	5.21b	6.05a
Moisturin (1:5)	3.59b	8.10a	4.59a
Moisturin (1:7)	6.69a	7.65a	4.95a
AntiStress (1:20)	3.93b	3.43b	3.68b
AntiStress (1:30)	8.53a	6.75a	4.19a
AntiStress (1:40)	2.91b	5.84b	3.75b

Means within the same time of measurement followed by the same letter do not differ significantly according to LSD (α=0.05).

among antidesiccant treatments. At this time the untreated control, Moisturin (1:3), and AntiStress 2000 (1:20 and 1:40) had the lowest stomatal conductance. When stomatal conductance was measured at 1130 h, there was still a significant treatment effect. Seedlings treated with AntiStress 2000 (1:20 and 1:40) had mean stomatal conductance values of 3.68 and 3.75 mmol $m^{-2}s^{-1}$, respectively which were significantly less than the other treatments. In July, stomata conductance of seedlings treated with antidesiccants and the untreated control were not statistically different (Tables 3-3). However, analysis of variance of stomatal conductance data collected on August 29 showed a

Table 3-5. Treatment mean values for stomatal conductance (mmol $m^{-2}s^{-1}$) of 2+0 ponderosa pine seedlings on August 29,1992 by the time of measurement.

Treatment	Time of measurement (h)		
	0730	0930	1130
Untreated control	2.17a	4.36a	3.04a
Moisturin (1:3)	1.60a	4.66a	2.78a
Moisturin (1:5)	2.24a	3.24b	2.59a
Moisturin (1:7)	1.90a	4.30a	3.09a
AntiStress (1:20)	2.26a	4.44a	2.82a
AntiStress (1:30)	1.11a	3.19b	2.71a
AntiStress (1:40)	1.57a	4.15a	2.28a

Means within the same time of measurement followed by the same letter do not differ significantly according to LSD (α=0.05).

significant treatment effect (p=0.0070) at 0930 h (Table 3-3). There were no significant treatment effects at 0730 or 1130 h (p=0.4820 and p=0.1234, respectively). Further analysis of mean separation for the 0930 h data showed that AntiStress 2000(1:30) and Moisturin (1:5) resulted in significantly lower stomatal conductance with 3.19 and 3.24 mmol $m^{-2}s^{-1}$ respectively (Table 3-5).

Survival

There were no significant treatment effects on the survival of ponderosa pine seedlings after outplanting. In the first growing season, 26 seedlings died (6% of the population) from deer browsing which was randomly distributed in all blocks and treatments. After two growing seasons, total mortality was 73 seedlings or 16%.

The animal damage identified in the second growing season was caused by deer and pocket gophers. The damage caused by the pocket gophers was characterized by basal stem debarking and stem clipping. Some seedlings were browsed twice. As a result of this damage, height was suppressed on some seedlings. As in the first year, damage was randomly distributed across all blocks and treatments. However, there were new buds growing in the places were the deer browse damage had occurred.

Height Increment

Analysis of height growth for each of the two growing seasons (1992 and 1993) showed that none of Moisturin or AntiStress 2000 treatments had a significant effect on height growth for the concentrations tested (Table 3-6). Moreover, there was no treatment effect on the total height increment over the two-year measurement period.

Discussion

Several of the antidesiccant treatments reduced stomatal conductance of 2+0 ponderosa pine seedlings. This difference occurred during June, only three months after seedlings were planted. There were no antidesiccant effects on stomatal conductance in July and August. In spite of the reduced stomatal conductance in June there were no

treatment effects on survival and growth. One possible explanation for this outcome could be that seedlings were not water stressed. Consequently, photosynthesis was not reduced and thus untreated seedlings could maintain survival and growth statistically equivalent to treated seedlings. It was found that fifteen days after planting (April 9, 1992) there were seven days of rainfall which totalled 36.3 mm at the nearest weather station. This value accounted 86% of the total precipitation recorded during that month. The 30 year average (1961-1990) for April is 22.9 mm. For the whole month of April 1992 precipitation was twice that at 42.2 mm. Also, there were three extreme rainfalls events recorded, April 9, 13, and 18 with 8, 11, and 8 mm respectively (Earth Info, Inc. 1994). It is possible that the increased precipitation following planting may have had a positive influence, improving seedling water potential and root growth, on the untreated control seedlings. Consequently, these seedlings were probably not under water stress.

Comparison of these results with previous work is difficult because other authors used different antidesiccants, species or experimental protocols. Also, it is important to note that this study represents the first time that tested Moisturin and AntiStress 2000 were tested on ponderosa pine seedlings. Nevertheless, a comparison with other studies is useful.

Kozlowski and Constantinidou (1986) have reported that antidesiccants applied to *Pinus resinosa* Ait. and *Fraxinus americana* L. showed a reduction in transpiration

Table 3-6. Analyses of variance of annual and total height increment of 2+0 ponderosa pine seedlings measured during two growing seasons.

Source of variation	df	MS	F	Prob>F
1992 increment				
Block	3	1.83	0.51	0.6750
AT[1]	6	10.29	1.18	0.3604
Error	18	8.72	2.42	0.0011
1993 increment				
Block	3	21.93	1.49	0.2171
AT[1]	6	1.52	0.04	0.9997
Error	18	41.10	2.79	0.0002
Total increment				
Block	3	58.52	1.17	0.3194
AT[1]	6	83.87	0.91	0.5069
Error	18	91.71	1.84	0.0199

AT[1] = Antidesiccant treatments.

for at least 32 days after seedlings were sprayed. Moreover, Lee and Kozlowski (1974) found that *Pinus resinosa* treated with silicone antidesiccant had a reduction in transpiration by about 90% during 10 days in a greenhouse experiment. These reductions most likely resulted from the physical blockage of stomata caused by the

antidesiccant (Davies and Kozlowski 1974; Noggel and Fritz 1983).

In particular, Moisturin (1:3) had a 69% reduction on stomatal conductance at 0730 h on June 27 when compared with the untreated control. These results agree with Ranney et al. (1989). They found similar responses in stomatal conductance of 'Colt' cherry trees after outplanting.

Colombo and Odlum (1987) studied the efficacy of six antidesiccants on transpiration of black spruce container seedlings and found that Vapor Gard (di-1-p-menthene) was the most effective antidesiccant in reducing transpiration. After 28 days, seedlings had lost only 22% of their total water content, whereas untreated control seedlings had lost 92%.

The fact that Moisturin and AntiStress 2000 did not increase the survival of 2+0 ponderosa pine seedlings after outplanting is consistent with work done by Alm and Stanton (1990). They found that survival of *Picea glauca* Vosh. was not increased when compared to an untreated control at the end of the first growing season. However, the survival results of this study do not agree with Marshall et al. (1991). They found that Folicote antidesiccant produced a significant delay in the mortality of *Pinus banksiana* Lamb., *Picea mariana* B.S.P., and *Picea glauca.* Recently, Williams et al. (1990), reported that *Picea glauca* and *Pinus resinosa* treated with antidesiccants had increased survival and reduced foliar damage.

The Moisturin and AntiStress 2000 treatments in this experiment had no significant effects on height growth during two growing seasons. The effect of other antidesiccants on the growth of tree seedlings have often given inconsistent results and

some times the antidesiccants reduced growth when compared with the untreated control (Gale and Hagan 1966). For example, under irrigated conditions *Prunus avium* x *pseudocerasus* seedlings sprayed with Folicote antidesiccant showed a reduced mean growth rate of 31% in comparison with the untreated control (Ranney et al. 1989). Furthermore, Magnussen (1986) working with *Pinus resinosa,* showed that seedlings treated with antidesiccants had 10% less growth than the untreated control.

Conclusions

Moisturin and AntiStress treatments temporarily reduced stomatal conductance of 2+0 ponderosa pine seedlings during the first growing season after outplanting. However, this did not improve seedling survival and growth. Two weeks after outplanting there was a period of unusually high rainfall. This may have resulted in a favorable water potential in the untreated control seedlings which in turn may have lead to a cycle of increased root growth and photosynthesis. In addition, the rainfall may have washed some of the antidesiccants from the leaves. As a result, the antidesiccant effects were perhaps, less effective.

Although antidesiccant treatments tested in this experiment did not increase seedling survival and growth, the favorable site conditions may have precluded an antidesiccant effect. Under these circumstances, additional research on the application of antidesiccants to ponderosa pine seedlings is justified, particularly under conditions of increased moisture stress.

References

Alm, A. and J. Stanton. 1990. Field trials of root dipping treatments for red pine, jack pine, and white spruce nursery stock in Minnesota. Tree Planters' Notes. 41(3):18-20.

Burdett, A. N. 1990. Physiological process in plantation establishment and the development of specifications for forest planting stock. Can. J. For. Res. 20:415-427.

Colombo, S.J. and K.D. Odlum. 1987. Efficacy of six antitranspirants on black spruce container seedlings. New Forests. 3:239-244.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects of antitranspirants on water relations and photosynthesis of woody plants. J. Amer. Soc. Hort. Sci. 99(4):297-304.

Earth Info, Inc. 1994. U.S. Weather data to 1993-Western states on CD-Rom.

Gale, J. 1961. Studies of plant antitranspirants. Physiol. Plant. 14:777-786.

Gale, J. and R.M. Hagan. 1966. Plant antitranspirants. Ann. Rev. Plant Physiol. 17:269-282.

Grossnickle, S.C., J.E. Major, J.T. Arnott, and V.M. Lemay. 1991. Stock quality assessment through an integrated approach. New Forests. 5:77-91.

Gumpertz, M.L. and C. Brownie. 1993. Repeated measures in randomized block and split-plot experiments. Can. J. For. Res. 23:635-639.

Hagiladi, A. and O. Ziv. 1986. Use of antitranspirants for control of powdery mildew on field grown roses. J. Environ. Hort. 4(2):65-68.

Kozlowski, T.T. and H.A. Constantinidou. 1986. Responses of woody plants to environmental pollution. Part I. Sources and types of pollutants and plant responses. Forestry Abstracts. 47:5-51.

Kramer, P.J. 1983. Water relations of plants. Academic Press. New York. 489 p.

Lee, K.J. and T.T. Kozlowski. 1974. Effects of silicone antitranspirant on woody plants. Plant and Soil. 40:493-510.

Magnussen, S. 1985. Effects of root-coating with the polymer Waterlock on survival and growth of drought-stressed bareroot seedlings of white spruce (*Picea glauca* (Moench) Voss) and red pine (*Pinus resinosa* Ait.). Tree Planters' Notes. 37(1):15-19.

Marshall, J.G., D.R. Cyr. and E.B. Dumbroff. 1991. Drought tolerance and the physiological mechanisms of resistance in northern coniferous seedlings. For. Can., Ont. Region, Sault Ste. Marine, Ont. COFRDA. rep 3314. 17 p.

Martin, J.D. 1974. Antitranspirants, possible uses in floriculture. Flor. Rev. 155(4015):19-21, 130-138.

Mexal, J.G. and T.D. Landis. 1990. Target Seedling Concepts: height and diameter. In Rose, R., S. Campbell., T. Landis (eds). Target seedling symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report RM-200. pp. 17-35.

Noggle, G.R., and G.J. Fritz. 1983. Introductory plant physiology. Prentice-Hall, New Jersey. 627 p.

Ranney, T.G., N.L Bassuk, and T.H. Whitlow. 1989. Effect of transplanting practices on growth and water relations of 'Colt' cherry trees during reestablishment. J. Environ. Hort. 7(1):41-45.

Rietveld, W.J. 1989. Transplanting stress in bareroot conifer seedlings: its development and progression to establishment. North. J. Appl. For. 6(3):99-107.

Sands, R. 1984. Transplanting stress in radiata pine. Aust. For. Res. 14(1):67-72.

SAS Institute Inc., 1993. SAS companion for the microsoft windows environment. Version 6. First edition. Cary, NC. 356 p.

Tracy, T.E. and A.J. Lewis. 1981. Effects of antitranspirants on hydrangea. HortScience. 16(1):87- 89.

Williams, P.A., A.M. Gordon, and W. Moeller. 1990. Effects of five antitranspirants on white spruce pine seedlings subjected to greenhouse drought. Tree Planters' Notes. 4(1):34-38.

Chapter 4

EFFECT OF ANTIDESICCANTS ON DAMAGE RESULTING FROM LEAF AND ROOT DESICCATION OF 2+0 PONDEROSA PINE (*Pinus ponderosa* Dougl.) SEEDLINGS

Abstract

In this study the actively growing shoots and roots of 2+0 ponderosa pine (*Pinus ponderosa* Dougl.) seedlings were treated with two antidesiccants (Moisturin® and AntiStress 2000®) each at three different concentrations and then subjected to four wind exposure treatments (0, 60, 120, 180 minutes) to evaluate the effects of antidesiccants on desiccation. All antidesiccant treatments reduced damage to fascicles caused by desiccation as measured by the amount of electrolytes released. Wind stress treatments of 60 and 120 minutes significantly increased fascicle damage although surprisingly, 180 minutes of wind exposure did not. However, analysis of variance showed no significant effects for either type of treatment or their interaction on solute release from the roots.

Seedlings not exposed to wind but treated with Moisturin 1:5 and AntiStress 1:30 showed the highest stomatal conductance. Seedlings exposed for 120 minutes to wind stress and treated with AntiStress 1:30 or untreated control had reduced stomatal conductance. The same is true when seedlings were wind exposed for 180 minutes. Wind stress treatments had a cubic effect on stomatal conductance. Seedlings treated with antidesiccants did not reduce water loss during 60 or 120 minutes of wind exposure but at 180 minutes Moisturin 1:7 was effective in reducing water loss.

When antidesiccants were applied on foliage of actively growing seedlings, foliar damage was decreased. But in general, however, they also increased stomatal conductance and water loss.

Introduction

The fall, winter and spring planting seasons are commonly used in most reforestation programs in the United States and Canada but usually spring planting result in better survival than fall planting because of frost heaving and desiccating winds (Wilson 1968).

When spring planting, seedlings are lifted in late fall or early winter when the stock is dormant and then graded, packaged, and placed in cold storage until required the following spring (Wilson 1968; Wood 1990). Summer planting is performed in Canada and Mexico. In Canada, summer planting stock is lifted in June, July and August. Although top growth is partially dormant, roots are particularly active and highly susceptible to damage by desiccation during lifting and planting (Mitchell et al. 1990). In Mexico, seedlings used in reforestation programs are actively growing when they are planted. The wet season in Mexico usually occurs during the summer (Vera-Castillo 1986).

Partially dormant and actively growing seedlings are more susceptible to desiccation. A common method used in Latin America to reduce transpiration stress at planting is to remove leaves or prune the top shoot (Evans 1982). Another is the gradual reduction in watering rates during the last few weeks in the nursery. This reduction of watering helps prepare seedlings for the reduced water supplies they are likely to receive in the field (Weber and Stoney 1986). In spite of these procedures seedling mortality is high. For example, in Mexico seedling mortality is more than 60% because of desiccation (Vera-Castillo, 1986).

Desiccation can result in cellular membrane damage (Leopold et al. 1981; Blum and Ebercon 1981; Shanahan et al. 1990). Seedlings become more resistant to water stress by dehydration treatments which change the colloidal chemical state of the protoplasm and plasmalemma. This change results in an increased hydration of the protoplasm colloids (Henckel 1964).

Antidesiccants are thought to have a positive, temporary effect on seedling water status by reducing transpiration, especially in species sensitive to water loss (Poljakoff-Mayber et al.1967). The loss of water vapor from leaves is limited by diffusion either in stomatal pores themselves or in boundary layers covering the leaf surface. The total resistance for the flow of water vapor from the site of evaporation to the air surrounding a leaf is equal to the resistance of water vapor of the leaf plus the resistance of water vapor of the boundary layer (Nobel 1991). An antidesiccant is an additional source of resistance in the water vapor pathway (Poljakoff-Mayber et al. 1967) which may further reduce water loss.

The hypothesis tested of this experiment is that antidesiccants and wind stress treatments do not reduce desiccation when applied to actively growing seedlings before outplanting.

Methods

Plant material

Ponderosa pine (*Pinus ponderosa*) seedlings (2+0) were grown at the USDA Forest Service the Bend Nursery, OR., and lifted on February 20, 1992. The seedlot was identified as BIA, Warm Springs 122-57-38-30-88-3. Seedlings were brought to Oregon State University (OSU) in July 1992 and transplanted to pots for two months until they were actively growing. Seedlings were grown in a greenhouse at the Forest Research Laboratory. The seedlings were divided into two groups. The first group consisted of 84 seedlings that were used to study the effects of two antidesiccants, each at three different concentrations on shoot and root desiccation. The second group consisted of 21 seedlings and was used to evaluate the effects of the same antidesiccant treatments on water loss.

Study establishment

The study was conducted in the Forest Research Laboratory at OSU. Antidesiccants treatments were applied in the same greenhouse where seedlings were grown. Before seedlings were treated with antidesiccants, the shoot and root systems were washed with tap water to remove electrolytes adhering to the seedlings. Seedlings were then dipped in the different antidesiccant treatments and dried for 20 minutes in the greenhouse at 20°C. After the antidesiccants were dried, seedlings were moved to the walk-in growth room. Room temperature (°C) and relative humidity (%) were recorded continuously during the experiment. Twelve seedlings per treatment were randomly

selected for each treatment.

Antidesiccant treatments

Two antidesiccants, Moisturin® (Burke's Protective Coatings, Washougal, WA) and AntiStress® 2000 (Polymer Ag, Inc., Fresno, CA), each at three concentration, were used in this experiment. The treatments included an untreated control; Moisturin (1:3); Moisturin (1:5); Moisturin (1:7); AntiStress 2000 (1:20); AntiStress 2000 (1:30); and AntiStress 2000 (1:40). The numbers in parentheses refer to antidesiccant to water ratio.

Wind exposure treatments

A commercial fan was used to apply the wind exposure treatments. There were four treatments. The first treatment was an untreated control without wind exposure. In treatment two seedlings were wind exposed for 60 minutes. In treatment three seedlings were exposed to wind for 120 minutes. Finally, in treatment four seedlings were wind exposed for 180 minutes.

Treatments were applied in a 2.5 x 2.5 x 2.5 m walk-in growth room in September 1992. Light was provided by a mixture of 110-W F96T12/CW/HO fluorescent tubes and 300-W incandescent lamps. The photosynthetic photon flux density at plant level was 160 μ mol m^{-2} s^{-1}. Before the seedlings were exposed to wind, they were randomly distributed on the tables. The temperature and relative humidity were recorded continuously during the experiment and were 25-27°C and 40% respectively.

Measurements

The first set of 84 seedlings was used to measure electrolyte leakage and stomatal conductance. The plant material used to measure electrolyte leakage, and ultimately estimates of damage, were the fascicles and roots. The electrolyte leakage test is a measure of cellular membrane damage. The leakage of the protoplasm is calculated from the release of electrolytes (Martineau et al. 1979). The total electrolyte leakage is expressed as specific conductance of the aqueous bathing solution in which the tissues were immersed (Whitlow et al. 1991). Ten fascicles per seedling per treatment were collected for each wind exposure treatment (Peck and Wallner 1982). From the root system, one gram per treatment was taken from 15 to 20 cm of actively growing roots (Ingram and Buchanan 1981).

Fascicles and roots were cut into 1cm length sections and placed separately in a vial containing 10 ml of distilled water to allow diffusion of electrolytes. The vials were left at room temperature for 24 hours. Before measuring the conductance, the vials were vigorously shaken by hand for 30 seconds. The initial conductance was determined with a conductivity bridge (Beckman model RC-16C). After that, the samples were placed in an oven at 90°C for two hours to kill the cells. The vials were left overnight at room temperature, and then shaken by hand 30 seconds before measuring conductance. The final step was to calculate the index of damage. This was determined using the following equations (Colombo et al. 1984):

Equation 1. RC untreated control = (EC untreated control / EC killed untreated control) x 100

Where RC untreated control = relative conductivity of the untreated control samples; EC untreated control = electrical conductivity of the water in which non-desiccated fascicles and roots were immersed before killing; EC killed untreated control = electrical conductivity of water in which non-desiccated fascicles and roots were immersed after killing.

Equation 2. RC desiccated = (EC desiccated / EC killed desiccated) x 100

Where RC desiccated = electrical conductivity of the water in which desiccated fascicles and roots were immersed before killing; EC desiccated = electrical conductivity of the water in which desiccated fascicles and roots were immersed before killing; EC killed desiccated = electrical conductivity of the water in which desiccated fascicles and roots were immersed after killing.

Equation 3. D = (RC desiccated - RC untreated control) / (1-(RC untreated control / 100))

Where D = is an expression of the amount of damage induced by desiccation. Equation one was used to determine the initial electrolyte leakage from the untreated control. The proportion of the total foliage and root electrolytes released due to desiccation was calculated with equation two. Finally, the damage was calculated using the third equation.

Leaf stomatal conductance was measured with a LI-1600 steady state porometer (LI-COR, Inc., Lincoln, Nebraska) at the beginning of each wind exposure treatment. Before measuring stomatal conductance, the porometer was turned on and left in the growth room for 30 minutes to allow self adjustment to temperature and humidity conditions. Four fascicles per seedling per treatment were chosen to measure stomatal conductance. Needle surface area was determined measuring the projected leaf area of each needle. The measured needle area of the 12 needles was 7.72 cm^2. Stomatal conductance was measured in mmol $m^{-2}s^{-1}$.

The second set of 21 seedlings was used to determine water loss from the 0, 60, 120, and 180 minutes wind exposure treatments. Seedlings were randomly distributed on the benches and then exposed to wind continuously during 180 minutes. Every sixty minutes seedlings were weighed. Water loss was determined gravimetrically and was expressed on a fresh weight basis, using the following equation:

$$\text{Water loss} = \text{Initial weight} - \text{Final weight} / \text{Initial weight}.$$

Where the final weights refer to the times that seedlings were wind stressed (0, 60, 120, and 180 minutes).

Experimental design and statistical analyses

This experiment had a completely randomized design with a factorial treatment structure: 7 antidesiccant treatments and 4 wind exposure treatments. Each antidesiccant treatment had 12 seedlings that were subjected to four wind exposures: 3

seedlings per wind exposure treatments (wind treatments had no true replication). So, there were a total of 84 seedlings to study desiccation damage as measured by electrolyte leakage.

The second group consisted of 21 seedlings (three seedlings per each antidesiccant treatment) and was used to measure water loss during each wind exposure. A repeated measurement analysis of variance was performed for a completely randomized design. Treatment differences were analyzed using the Statistical Analysis System (SAS, Institute Inc., 1993) software. A log transformation of damage and water loss data was used to correct for heteroscedasticity. Because of data outliers on damage, the method of weighted least square was used to obtain parameter estimators. Type III sums of squares were used to test the hypotheses stated in this study. The Least Significant Difference (LSD) was used to determine significant differences among treatment means at the α=0.05 level for those effects found to be significant by analysis of variance.

Results

Damage from fascicles

Antidesiccants treatments had a highly significant effect in reducing damage to fascicles of 2+0 ponderosa pine seedlings. The weighted analysis of variance (Table 4-1a) showed that antidesiccant treatments had a significant effect on the amount of electrolytes released from fascicles (p=0.0185). The interaction term wind exposure and

antidesiccant treatments was not significant (p=0.0748). Multiple mean comparisons clearly showed that the untreated control had the greatest damage as measured by electrolyte leakage (Table 4-2a). There were no differences among antidesiccant treatments. All the antidesiccants treatments were effective in reducing electrolyte leakage caused by wind stress treatments.

There were significant effects due to wind stress treatments (p=0.0289) (Table 4-1a) Damage to seedlings exposed to wind for 60 or 120 minute was significantly greater than damage to the untreated control or the 180 minute treatment (Table 4-2b).

Damage to roots

No significant differences were found among antidesiccant treatments (p=0.3343), wind stress treatments (p=0.6260), or the interaction term of antidesiccant and wind exposure treatments (p=0.1866) on electrolyte leakage of actively growing roots (Table 4-1b).

Stomatal conductance

The repeated measurements analysis of variance showed that there was a significant antidesiccant effect on stomatal conductance (p=0.0001). The effect of wind stress treatments was highly significant (p=0.0001) and it had a cubic effect (p=0.0001) (Table 4-3). The interaction of antidesiccants and wind stress was highly significant (p=0.0001) and it had a linear (p=0.0001) and quadratic effect (p=0.0001) but these terms differ among the antidesiccant treatments.

Separation of means for the no wind stress treatment (0 minutes) showed that seedlings treated with Moisturin 1:5 and AntiStress 1:30 had the highest stomatal

Table 4-1. Weighted analysis of variance (a) and analysis of variance (b) of effects on damage due to antidesiccants and wind exposure treatments for fascicles and roots.

Source of variation	df	MS	F	Prob>F
(a) Fascicles				
AT[1]	6	2.380	2.81	0.0185
WS[2]	3	2.743	3.24	0.0289
AT x WS	18	1.419	1.67	0.0748
Error	56	0.848		
(b) Roots				
AT[1]	6	0.466	1.17	0.3343
WS[2]	3	0.233	0.59	0.6262
AT x WS	18	0.542	1.36	0.1866
Error	56	22.272		

AT[1] = Antidesiccant treatments.
WS[2] = Wind stress treatments.

conductance, 6.03 and 5.70 mmol $m^{-2}s^{-1}$ respectively (Table 4-4). On the other hand, the untreated control and seedlings treated with AntiStress 1:40 showed the lowest stomatal conductance with 3.15 and 3.58 mmol $m^{-2}s^{-1}$ respectively. Seedlings treated with Moisturin 1:3 and exposed to wind for 60 minutes showed the highest mean stomatal conductance (7.48 mmol $m^{-2}s^{-1}$). This value was statistically different from the rest of the treatments. With the exception of Moisturin 1:3 and AntiStress 1:40, all other treatments showed no significant differences in stomatal conductance (Table 4-4).

Table 4-2. Log mean values of damage to fascicles by antidesiccant treatments (a) and wind stress treatments (b) of 2+0 ponderosa pine seedlings.

Treatments	Log mean
(a) Antidesiccant treatments	
Untreated control	4.85a
Moisturin (1:3)	4.58b
Moisturin (1:5)	4.59b
Moisturin (1:7)	4.51b
AntiStress 2000 (1:20)	4.53b
AntiStress 2000 (1:30)	4.54b
AntiStress 2000 (1:40)	4.57b
(b) Wind stress treatments	
0 (Untreated control)	4.53b
60 Minutes	4.63a
120 Minutes	4.63a
180 Minutes	4.59ab

Means in the same column followed by the same letter do not differ significantly according to LSD (α= 0.05%).

Seedlings exposed for 120 minutes of wind and treated with Moisturin 1:3 showed the highest stomatal conductance (5.29 mmol $m^{-2}s^{-1}$). While seedlings treated with AntiStress 1:30 and untreated control seedlings showed the lowest response (0.66 and 0.25 mmol $m^{-2}s^{-1}$ respectively). Seedlings exposed for 180 minutes to wind and treated with Moisturin 1:3 again showed the highest mean while untreated control seedlings and those treated with AntiStress 1:30 showed the lowest response.

Earlier, it was mentioned that the effect of wind stress on stomatal conductance had a cubic effect. The regression equations used to describe this data follow:

$$SCT1 = -5.643 + 15.088*WS - 7.166*WS^2 + 0.936*WS^3$$

$$SCT2 = -9.468 + 20.784*WS - 8.074*WS^2 + 0.936*WS^3$$

$$SCT3 = -1.146 + 12.787*WS - 6.669*WS^2 + 0.936*WS^3$$

$$SCT4 = -3.225 + 14.326*WS - 6.931*WS^2 + 0.936*WS^3$$

$$SCT5 = -5.199 + 16.184*WS - 7.241*WS^2 + 0.936*WS^3$$

$$SCT6 = 0.112 + 11.586*WS - 6.614*WS^2 + 0.936*WS^3$$

$$SCT7 = -8.800 + 19.305*WS - 7.795*WS^2 + 0.936*WS^3$$

Where SCT1 is the stomatal conductance of the untreated control seedlings.

WS is the wind stress treatments.

WS^2 is the square wind stress and WS^3 is the cubic wind stress.

SCT2 is the stomatal conductance of seedlings treated with Moisturin 1:3.

SCT3 is the stomatal conductance of seedlings treated with Moisturin 1:5.

SCT4 is the stomatal conductance of seedlings treated with Moisturin 1:7.

SCT5 is the stomatal conductance of seedlings treated with AntiStress 1:20, and so on.

The r^2 from the model accounted for 95% of the total variation.

Water loss

The repeated measurements analysis of variance showed that there was a significant antidesiccant treatment effect on water loss (p=0.0001) (Table 4-5). The analyses of variance for each time period that seedlings were stressed with wind: 60, 120, and 180, minutes, showed that antidesiccant treatments had a significant effect on water loss with p-values of 0.0001, 0.0001, and 0.0189 respectively (Table 4-6). Moreover, the repeated analysis of variance showed that wind exposure treatments had a linear effect on water loss (p=0.0001). The longer the seedlings were exposed the more water they lost. The interaction term for wind exposure and antidesiccant treatments was not significant (p=0.6578) (Table 4-5).

Separation of means test on water loss showed that after 60 minutes of wind stress the untreated control, Moisturin (1:7) and AntiStress 2000 (1:20 and 1:30) had significantly less water loss than seedlings treated with higher antidesiccant concentrations such as Moisturin 1:3 and 1:5 (Treatments 2 and 3 respectively) and the low concentration of AntiStress 2000 (1:40) (Table 4-7). After 120 minutes of wind exposure the antidesiccant treatments had the same mean separation grouping as at 60 minutes with one exception. Seedlings treated with AntiStress 2000 (1:20) moved into the higher stomatal conductance group. However, seedlings exposed to wind for 180 minutes showed that Moisturin 1:7 had significantly lower water loss than other treatments including the untreated control.

Table 4-3. Repeated measurements analysis of variance of stomatal conductance of 2+0 ponderosa pine seedlings.

Source of variation	df	MS	F	Prob>F
AT[1]	6	16.42	44.23	0.0001
Error (a)	14	0.37		
WS[2]	3	38.81	230.40	0.0001
Linear	1	80.19	10.97	0.0002
Quadratic	1	3.14	32.51	0.0001
Cubic	1	33.11	100.96	0.0001
AT x WS	18	3.53	22.11	0.0001
Linear	6	6.57	81.31	0.0001
Quadratic	6	3.64	37.65	0.0001
Cubic	6	0.40	1.23	0.3504
Error (b)	42	0.1684		

AT[1] = Antidesiccant treatments.
WS[2] = Wind stress treatments.

Discussion

Fascicles of actively growing ponderosa pine seedlings responded to two of the wind exposure treatments by releasing a greater amount of electrolytes. This result agrees with previous research on electrolyte leakage with different plants (Blum and Ebercon 1981; Leopold et al. 1981; Whitlow et al. 1991). They found that the

Table 4-4. Mean stomatal conductance by antidesiccant treatments of 2+0 ponderosa pine seedlings after 0, 60, 120, and 180 minutes of wind exposure.

Antidesiccant Treatments	Wind stress (minutes)			
	0	60	120	180
Untreated control	3.15d	3.50c	0.25d	0.00c
Mosturin (1:3)	4.11c	7.48a	5.29a	4.44a
Moisturin (1:5)	5.70a	4.58c	2.48c	2.77b
Moisturin (1:7)	5.03b	5.40c	2.43c	3.15b
AntiStress (1:20)	4.79b	5.35c	3.79b	3.47b
AntiStress (1:30)	6.03a	4.27c	0.66d	0.52c
AntiStress (1:40)	3.58d	6.28b	4.05b	3.64b

Means in the same columns followed by the same letter do not differ significantly according to LSD (α= 0.05%).

permeability of the protoplasm calculated from the release of electrolytes becomes much higher as a result of desiccation.

However, the use of Moisturin and AntiStress 2000 in this study seem to reduce the amount of electrolyte leakage from fascicles. As a result the damage to cell membranes of treated seedlings was diminished. On the other hand, antidesiccants did not reduced stomatal conductance but in some instances increased it.

In a study by Davies and Kozlowski (1974) it was found that antidesiccants significantly reduced water loss over untreated control plants at least 12 days after applying antidesiccants. Furthermore, foliar damage as measured by electrolyte leakage was increased after one hour of wind exposure.

Table 4-5. Repeated measurements analysis of variance of water loss of 2+0 ponderosa pine seedlings.

Source of Variation	df	MS	F	Prob>F
AT[1]	6	0.876	12.53	0.0001
Error (a)	14	0.069		
WS[2]	2	1.291	34.80	0.0001
Linear	1	2.582	45.22	0.0001
Quadratic	1	0.001	0.08	0.7808
AT x WS	12	0.029	0.79	0.6578
Linear	6	0.048	0.84	0.5605
Quadratic	6	0.011	0.62	0.7097
Error	28	0.037		

AT[1] = Antidesiccant treatments.
WS[2] = Wind stress treatments.

Levels of root damage caused by drought have been investigated using the electrolyte leakage test (Martin et al. 1987). The rate of electrolyte leakage was suggested as a possible indicator of seedling performance (McKay 1992). However, in this study it was found that root damage as measured by electrolyte leakage was not significantly affected by either the antidesiccants or wind exposure treatments. It is suggested that the root tissues used in this study could have had suberized root epidermal layers which acted to inhibited the free flow of electrolytes from the

Table 4-6. Analysis of variance for each time of wind stress treatments on water loss of 2+0 ponderosa pine seedlings treated with antidesiccants.

Source of variation	df	MS	F	Prob>F
(a) 60 minutes				
AT[1]	6	0.247	16.72	0.0001
Error	14	0.014		
(b) 120 minutes				
AT[1]	6	0.261	15.59	0.0001
Error	14	0.016		
(c) 180 minutes				
AT[1]	6	0.426	3.78	0.0189
Error	14	0.112		

AT[1] = Antidesiccant treatments.

intercellular spaces.

The results found in this study for stomatal conductance were contrary to expectations although they did support the hypothesis posed for this research. Seedlings treated with antidesiccants had higher stomatal conductances than the untreated control. Davies and Kozlowski (1974) found that 3+0 *Fraxinus amaricana* L. treated with Clear Spray antidesiccant did not show reduced transpiration within 24 hours after application

Table 4-7. Log mean water loss by antidesiccant treatments of 2+0 ponderosa pine seedlings after 60, 120, and 180 minutes of wind exposure.

Antidesiccant Treatments	Wind stress (minutes) 60	120	180
Untreated control	4.76b	4.59b	4.47a
Mosturin (1:3)	5.07a	4.86a	4.67a
Moisturin (1:5)	5.22a	4.88a	4.66a
Moisturin (1:7)	4.47b	4.17b	3.62b
AntiStress (1:20)	4.98b	4.80a	4.54a
AntiStress (1:30)	4.55b	4.25b	4.15a
AntiStress (1:40)	5.08a	4.77a	4.55a

Means in the same column followed by the same letter do not differ significantly according to LSD (α= 0.05%).

and it was not significantly different from the untreated control. Twelve days later treated seedlings showed increased water loss over the untreated control. They also determined that 3+0 *Pinus resinosa* Ait. treated with Folicote antidesiccant had greater transpiration than the untreated control.

Manufacturers recommend that antidesiccants be applied when plants are dormant. Perhaps this recommendation is made because leaves of dormant plants are associated with relatively low levels of stomatal activity and consequently, little transpiration. In these circumstances water vapor movement would probably have little impact on foliar coverage by film-forming antidesiccants. In this research water vapor

movement from the foliage of the actively growing seedlings may not have allowed adequate coverage of the stomata by the antidesiccants.

According to Davenport et al. (1972) this result might indicate that partial covering of the fascicles with the film-forming antidesiccant led to a general increase in leaf turgor with resultant stomatal opening. Another explanation supported by Gale and Hagan (1966) is that the increased resistance formed by the antidesiccant tended to raise the leaf temperature and thus increase the transpiration rate. This result disagrees with Poljakoff-Mayber et al. (1967) and Davies and Kozlowski (1974) since film-coating might be expected to decrease water loss.

The two antidesiccants used in this study reduced foliar damage, an indirect indication of reduced water stress, and they by and large increased stomatal conductance and water loss. However, it is speculated that antidesiccants may have increased leaf turgor and leaf temperature which may have influenced the stomatal opening of seedlings.

Conclusions

Foliar damage as measured by the amount of electrolytes released was reduced by all the antidesiccants tested. However, the antidesiccants and concentrations tested did not have an effect on root damage. Damage to seedlings exposed to wind for 60 or 120 minutes was greater than damage to the untreated control.

The highest stomatal conductance was obtained when ponderosa pine seedlings were wind exposed for 60, 120, or 180 minutes. While the no wind stress treatment

showed that seedlings treated with Moisturin 1:5 and AntiStress 1:30 had the highest stomatal conductance. The effect of wind stress on stomatal conductance of ponderosa pine seedlings had a cubic effect. Seedlings treated with Moisturin 1:7 had reduced water loss but only after 180 minutes of wind exposure.

According to the results obtained in this experiment the null hypothesis tested is accepted. However, the results were contradictory. The antidesiccants and concentration tested reduced damage to foliage as measured by the amount of electrolyte leakage. On the other hand, antidesiccants increased stomatal conductance and water loss. Consequently, further research on the application of antidesiccants to actively growing seedlings seems justified.

References

Blum, A. and A. Ebercon. 1981. Cell membrane stability as a measured of drought and heat tolerance in wheat. Crop Science. 21:43-47.

Colombo, S.J., D.P. Webb, and C. Glerum. 1984. Frost hardiness testing: An operational manual for the use with extended greenhouse culture. Ontario Minister of Natural Resources. Forest Research Report No.110. 14 p.

Davenport, D.C., M.A. Fisher and R.M. Hagan. 1972. Some counteractive effects of antitranspirants. Plant Physiol. 49:722-724.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects of antitranspirants on water relations and photosynthesis of woody plants. J. Amer. Soc. Hort. Sci. 99(4):297-304.

Evans, J. 1982. Plantation forestry in the tropics. Clarendon Press. Oxford. 472 p.

Gale, J. and R.M. Hagan. 1966. Plant antitranspirants. Ann. Rev. Plant Physiol. 17:269-282.

Henckel, P.A. 1964. Physiology of plants under drought. Ann. Rev. Plant Physiol. 15:363-386.

Ingram, D.L. and D. Buchanan. 1981. Measurement of direct heat injury of roots of three woody plants. HortScience. 16(6):769-771.

Leopold, A.C., M.E. Musgrave, and K.M. Williams. 1981. Solute leakage resulting from leaf desiccation. Plant Physiol. 68:1222-1225.

Martin, U., S.G. Pallardy and Z.A. Bahari. 1987. Dehydration tolerance of leaf tissues of six woody angiosperm species. Physiol. Plantarum. 69:182-186.

Martineau, J.R., J.E. Specht., H. Williams and C.Y. Sullivan. 1979. Temperature tolerance in soybeans. I. Evaluation of a technique for assessing cellular membrane thermostability. Crop Science. 19:75-78.

McKay, H.M. 1992. Electrolyte leakage from fine roots of conifer seedlings: a rapid index of plant vitality following cold storage. Can. J. For. Res. 22:1371-1377.

Milburn, J.A. 1979. Water flow in plants. Longman. London. 225 p.

Mitchell, W.K., G. Dunsworth., D.G. Simpson, and A. Vyse. 1990. Planting and seeding. In Lavender, D.P., R. Parish., C.M. Johnson., G. Montgomery., A. Vyse., R.A. Willis, and D. Winston (eds). Regenerating British Columbia's Forests. University of British Columbia. Vancouver. 235-253.

Nobel, P.S. 1991. Physicochemical and environmental plant physiology. Academic Press. Inc. San Diego, California. 635 p.

Peck, K.M., and S.J. Wallner. 1982. Ecotypic differences in heat resistance of aspen leaves. HortScience. 17(1):52-53.

Poljakoff-Mayber, A., J. Gale., H. Aluma., I. Kahane., H. Ginsburg., Z. Goldschmidt., I. Nir., and E. Shabi. 1967. Final technical report on effect of plant antitranspirants on certain physiological processes of forest seedlings and other plant material. Project No.A10-FS-10. The Hebrew University of Jerusalem. Jerusalem. 107 p.

SAS Institute Inc., 1993. SAS companion for the microsoft windows environment. Version 6. First edition. Cary, NC. 356 p.

Shanahan, J.F., I.B. Edwards., J.S. Quick and J.R. Fenwick. 1990. Membrane thermostability and heat tolerance of spring wheat. Crop Science. 30:247-254.

Vera-Castillo, J.A.G. 1986. Estudios de algunos factores que influyen en la produccion de *Pinus montezumae* Lamb., en vivero. Tesis de Meastria en Ciencias. Colegio de Postgraduados. Chapingo, Mexico. 141 p.

Weber, F.R. and C. Stoney. 1986. Reforestation in arid lands. Volunteers in Technical Assistance. Arlington, VA. 335 p.

Whitlow, T.H., N.L. Bassuk., T.G. Ranney, and D.L. Reichert. 1992. An improved method for using electrolyte leakage to assess membrane competence in plant tissues. Plant Physiol. 98:198-205.

Wilson, V. 1968. Planning rework programs on nonstocked plantations on the Umpqua National Forest. In U.S. Forest Service (ed). First Reforestation Workshop. Proceedings. 164-179.

Wood, J.E. 1990. Black spruce and jack pine plantation performance in boreal Ontario: 10-year results. North. J. Appl. For. 7(4):175-179.

CHAPTER 5

ANTIDESICCANT AND STORAGE EFFECTS ON PHYSIOLOGICAL, PHENOLOGICAL, AND MORPHOLOGICAL CHARACTERISTICS OF 2+0 PONDEROSA PINE (*Pinus ponderosa* Dougl.) SEEDLINGS

Abstract

The antidesiccants Moisturin® and AntiStress 2000®, each at three different concentrations, were applied to 2+0 ponderosa pine (*Pinus ponderosa* Dougl.) seedlings before cold storage. Seedlings were also subjected to two different storage conditions (dry and moist) and storage lengths (20 and 40 days). The effects of these treatments on seedling physiology, phenology, and morphology were evaluated in a greenhouse experiment over four months.

Survival of seedlings treated with antidesiccants and then stored in a cold room for 20 or 40 days was not affected during the experiment. On the other hand, budbreak and budset activity were delayed on seedlings treated with antidesiccants and then placed in cold storage in a dry condition. New root production was diminished 46% by Moisturin® (1:3). Storage for 40 days reduced root production by 24% as compared to seedlings stored 20 days. Chlorophyll fluorescence was reduced when seedlings were stored dry and the storage length was longer than 20 days. However, stomatal conductance was not affected by antidesiccant or storage treatments.

In this experiment it was hypothesized that Moisturin® and AntiStress 2000® , cold storage condition, and cold storage length treatments would not affect selected physiological, phenological, and morphological characteristics. This was not the case,

however, as the antidesiccants tested had a negative effect on budbreak, budset, and foliar damage. In addition, storage condition had a negative effect on foliar damage, budbreak, and budset although it did have a positive effect on height and diameter increment. Finally, storage length had negative effects on budset, foliar damage, root growth potential and chlorophyll fluorescence.

Introduction

Nursery and reforestation managers work continuously to improve the quality of seedlings and to define seedling targets for different species on different sites. A seedling target embodies those structural and physiological traits that can be quantitatively linked to successful reforestation (Rose et al. 1990). It is understood that the quality of seedlings can be diminished during the process of lifting, grading, storing and planting (Garber and Mexal 1980).

Current regeneration practices often necessitate prolonged storage of seedlings. Frequently, lifting and planting dates cannot be synchronized. Consequently, a large percentage of seedlings are stored prior to planting (Garber and Mexal 1980). Some researchers have found that increased storage length drastically decreases seedling quality and survival after outplanting (Garber and Mexal 1980, Simpson 1984, Balneaves and Menzies 1990, Omi 1991). Loss of water by seedlings, either during cold storage or after field planting, may result in growth-limiting or even lethal levels of plant moisture stress (Simpson 1984). Moreover, cold storage can decrease the ability of seedlings to initiate new roots or alter shoot height and diameter growth (Balneaves 1988).

Antidesiccants have shown promise for preventing seedling desiccation after lifting (Owston and Stein 1972). Although antidesiccants induce a less negative leaf water potential (Win et al. 1991), they can inhibit photosynthesis by reducing absorption of carbon dioxide (Kozlowski and Constantinidou 1986). Nevertheless, the use of antidesiccants is an alternative method for reducing water loss and improving seedling water status during transplanting (Ranney et al. 1989).

The hypothesis tested in this experiment was that antidesiccants and cold storage conditions do not affect subsequent physiology, phenology, and morphology of 2+0 ponderosa pine (*Pinus ponderosa* Dougl.) seedlings grown in a greenhouse.

Methods

Plant material

Two-year-old ponderosa pine seedlings were grown at the J. Herbert Stone Nursery, Central Point, OR, and lifted, graded, and stored on February 18, 1993. The seedlot identification number was 479-1. Cones were collected in the Mt. Thielsen area of the Winema National Forest in 1978, at an elevation of 1672 m and seed zone identification number 701 (State of Oregon Tree Seed Zone Map. Western Forest Tree Seed Council). On April 2, 400 seedlings were transported to Corvallis, OR., and stored at 5^0C in the cold room at the Forest Research Laboratory (FRL) for 18 days prior to starting the experiment. One-hundred and forty seedlings were used to evaluate root growth potential (RGP), and 224 were used to evaluate other physiological, morphological, and phenological variables.

Study establishment

The study was conducted at the FRL in a greenhouse in April 1993. Temperatures were maintained at 30:18^0 C (day:night) until July. During August and September temperatures were 37:18^{0}C. Before applying the antidesiccant treatments, seedlings were washed and tagged. Then 364 seedlings were dipped in Moisturin® or

Table 5-1. Antidesiccant , storage condition and storage length treatments applied to 2+0 ponderosa pine seedlings.

Treatment	Antidesiccant	Storage Condition	Storage Length[4]
1	Untreated control	D[2]- W[3]	20-40
2	Moisturin 1:3[1]	D - W	20-40
3	Moisturin 1:5	D - W	20-40
4	Moisturin 1:7	D - W	20-40
5	AntiStress 2000 1:20	D - W	20-40
6	AntiStress 2000 1:30	D - W	20-40
7	AntiStress 2000 1:40	D - W	20-40

Moisturin® (Vinyl chloride monomer and vinylidene chloride monomer). Burke's Protective Coatings, Washougal, WA. AntiStress 2000® (Acrylic polymers). Polymer Ag, Inc., Fresno, CA.
[1]Antidesiccant:Water. [2] Dry condition. [3] Wet condition. [4] Days.

Anti-Stress® 2000 (Table 5-1). After dipping, 182 seedlings were dried in the sun for ten minutes (dry condition), put into plastic bags, placed in cardboard cartons and then stored in a cold room at 5°C. The remaining 182 seedlings were stored immediately after dipping in antidesiccants (wet condition). The untreated control seedlings were dipped in water and then dried as were the antidesiccant-treated seedlings or stored wet after being dipped in water. Untreated control seedlings were packaged in the same manner as the treated seedlings.

Following dipping, seedlings were stored for either 20 or 40 days. Seedlings that were stored for 20 days, were potted on April 20, 1993 and those stored for 40 days were potted on May 10, 1993. The seedlings were then grown in the greenhouse in one gallon pots (C-700 El Campo, TX) in a media consisting of 1:1:1:2 mixture of sandy loam: loam:sand: pumice. Seedlings were watered with 200 ml of water every third day.

Measurements

Survival was recorded every week until the end of the experiment at 18 weeks. Budbreak was recorded daily during the first three weeks after seedlings were potted, while budset was recorded daily during the last two weeks of July. The root growth potential (RGP) test was begun 30 days after seedlings were potted for each antidesiccant-storage treatment combination following a procedure recommended by Ritchie and Tanaka (1990). The total number of new roots were counted after each storage time studied. Seedling height and stem diameter were measured twice, after seedlings were potted and at the end of the growing season (August 1993) to calculate increments. Assessing these morphological variables was done according to the suggestions of Mexal and Landis (1990).

Foliar damage was monitored weekly for 6 weeks until seedlings showed new leaves. Needle damage was scored on old needles as follows: 1 = healthy needles, 2 = first 10 mm of the needles tips reddish brown, 3 = between 1 to 10 needles dead, 4 = between 11 to 20 needles dead, 5 = all needles yellow, 6 = all needles brown.

Stomatal conductance was measured with a LI-1600 steady state porometer (LI-COR, Inc. Lincoln, Nebraska) three times per day (0930, 1130 and 1330 h) once a

month for four months (April-July). Before measuring stomatal conductance, the porometer was turned on and left in the greenhouse for 30 minutes to allow self adjustment to temperature and humidity conditions. Four fascicles per seedling per treatment were chosen to measure stomatal conductance. Needle surface area was determined measuring the projected leaf area of each needle and was then multiplied by five. Therefore, the measured needle area was 7.72 cm^2. Stomatal conductance was measured in mmol $m^{-2}s^{-1}$.

Chlorophyll fluorescence emissions were also measured five times once a month using an integrating fluorometer (Pacific Fluorotec, Burnaby, British Columbia, Canada) interfaced to a personal computer (286 MB-25 MHz) for data acquisition and processing. Four days were required to measure all the treatments combinations. A set of 28 seedlings were recorded per day, from 0900 to 1300 h. Prior to scanning for fluorescence, seedlings were preconditioned as described by Binder and Fielder (1991) to standardize the photosynthetic system and dark adaption of the seedlings. The standard preconditioned protocol consisted of five steps: 1) seedlings were watered to field capacity the day before measurement. 2) seedlings were then placed into a plastic chamber at 1730 h and illuminated with 250 μ photons $m^{-2}s^{-1}$ for 90 minutes. 3) seedlings were maintained in the dark for 11 h., 4) at 0600 h the light was turned on for 150 minutes, and 5) finally, seedlings were dark adapted for 30 minutes prior to measurement.

The measured parameters were maximum fluorescence (Fm) where the electron acceptors are reduced, variable fluorescence (Fv) defined as Fm minus ground

fluorescence (Fo). Ground fluorescence is a measure of the amount of chlorophyll measured in the sphere, where the electron acceptors are fully oxidized. Finally, the ratio of Fv / Fm was determined as a quantitative measure of photochemical efficiency. The Fv / Fm ratio has become an important measurable parameter of the physiological state of the photosynthetic apparatus of intact plant leaves (Vidaver et al. 1990; Binder and Fielder 1991). The fluorometer scanned the fluorescence emissions of each seedling for three minutes. The scan environment was characterized by a target light intensity of 150 $\mu E\ m^{-2}s^{-1}$ in the integrating sphere provided by a tungsten lamp.

Measurements for both stomatal conductance and chlorophyll fluorescence emissions began two weeks after the seedlings were potted for each treatment combination.

Experimental design and statistical analysis

The experimental design used in this study was a 7 x 2 x 2 factorial with seven antidesiccant treatments (Table 5-1), two cold storage periods (20 and 40 days after applying antidesiccants), and two seedling storage conditions (dry and wet). There were 13 seedlings in each treatment combination. From these, 8 were used to measure height, stem diameter, budbreak, budset and foliar damage. In addition, four of these eight were randomly selected for stomatal conductance and chlorophyll fluorescence testing. The remaining five were used to measure RGP.

SAS software was used for all the statistical analyses (SAS Institute Inc. 1993). Because stomatal conductance and chlorophyll fluorescence were measured repeatedly on the same seedlings, a repeated measures analysis of variance was performed on these

two variables (Gumpertz and Brownie 1993). Huynh-Feldt conditions was met. The Least Significant Differences test was used to determine differences among treatment means at the α=0.05 level.

Results

Survival

There was no mortality of ponderosa pine seedlings in any of the treatment combinations.

Budbreak

Budbreak was significantly affected by antidesiccant applications (p=0.0002) and storage condition (p=0.0143) (Table 5-2). In addition there were significant interactions between antidesiccant treatments and storage condition (P=0.0009). Moisturin (1:3) and AntiStress 2000 (1:40) delayed budbreak activity by 2 and 3 days respectively. There were no statistical differences for other antidesiccant treatments and the untreated control (Table 5-3). Seedlings treated with Moisturin (1:3) and then stored wet, delayed budbreak for three days. Moreover, seedlings dipped in low concentrations of AntiStress 2000 (1:30 and 1:40) and then stored dry, had a three day delay in budbreak in comparison with the untreated control (Table 5-4).

Budset

Antidesiccant treatments, storage condition, storage length, and all interactions were highly significant (p=0.0001) (Table 5-2). AntiStress 2000 (1:20 and 1:40) delayed budset three days, however there were no statistical differences among the rest of antidesiccant treatments (Table 5-3). Seedlings that were stored wet had an average of 84.80 days to budset. In addition, seedlings stored in the cold room 20 days had 90 days from budbreak to bud set. On the other hand, the seedlings stored 40 days had 77 days from budbreak to budset.

The mean values for the interaction of antidesiccant and storage condition treatments showed that seedlings dipped in Moisturin (1:5) and AntiStress 2000 (1:30 and 1:40) and then stored wet, had delays of more than five days to budset in comparison with the untreated control stored wet (Table 5-5). The interaction of antidesiccant and storage length treatments demonstrated that in general, seedlings stored 20 days had the highest mean values for days until budset.

Foliar damage

Antidesiccants significantly affected foliar damage (P=0.0015) (Table 5-2). Furthermore, storage condition and storage length also significantly affected foliar damage (p=0.0003 and p=0.0001 respectively). Foliage of seedlings stored dry were damaged more than those stored wet. Seedlings stored for 20 days had more damage than those stored 40 days. Moisturin (1:5) was associated with significantly more foliar damage (25 %). Moreover, all the concentrations tested for AntiStress 2000 damaged the fascicles (Table 5-3).

Table 5-2. Summary of analyses of variance of effects of antidesiccants, storage condition and storage length on budbreak, budset, foliar damage, height increment, diameter increment, and root growth potential of 2+0 ponderosa pine seedlings. Values are probability of a greater F-value.

Source of variation	Budbreak	Budset	Foliar Damage	Height increment	Diameter increment	RGP
AT[1]	0.0002	0.0001	0.0105	0.6038	0.6809	0.0002
SC[2]	0.0143	0.0001	0.0003	0.0176	0.0003	0.8660
SL[3]	0.5414	0.0001	0.0001	0.0876	0.2361	0.0001
AT x SC	0.0009	0.0001	0.3976	0.2750	0.9556	0.4858
AT x SL	0.9094	0.0001	0.9199	0.3011	0.8859	0.0471
SC x SL	0.1371	0.0001	0.3981	0.9915	0.4274	0.3826
AT x SC x SL	0.8059	0.0003	0.8612	0.9504	0.3264	0.0021

AT[1] = Antidesiccant treatments. SC[2] = Storage condition. SL[3] = Storage length.

Table 5-3. Summary of mean values of effect of antidesiccant treatments on budbreak, budset, foliar damage, and root growth potential (RGP) by antidesiccant treatments of 2+0 ponderosa pine seedlings.

Antidesiccant Treatments	Budbreak	Budset	Foliar damage	RGP
Untreated control	11.84b	81.53b	1.72b	109.1a
Moisturin (1:3)	13.96a	83.87b	1.78b	58.9b
Moisturin (1:5)	13.15b	83.84b	2.15a	89.7a
Moisturin (1:7)	13.00b	84.06b	1.56b	68.3b
AntiStress (1:20)	12.56b	84.84a	1.84a	80.1b
AntiStress (1:30)	13.12b	84.03b	1.96a	74.6b
AntiStress (1:40)	15.02a	85.87a	1.93a	83.4b

Means in the same column followed by the same letter do not differ significantly according to LSD (α= 0.05%).

Height increment

Antidesiccant and storage length treatments did not affect height increment (Table 5-2). However, this variable was affected by storage condition (p=0.0176). Mean height increment was greater on seedlings stored dry (6.59 cm) than those stored wet (5.99 cm) (p=0.0176).

Diameter increment

Antidesiccant and storage length treatments did not affect diameter increment (p=0.6809 and 0.2361 respectively) (Table 5-2). However, storage condition had a significant effect on diameter increment (p=0.0003). The stem diameter increment had

Table 5-4. Mean number of days until budbreak by antidesiccant and storage condition treatments of 2+0 ponderosa pine seedlings grown in a greenhouse.

Antidesiccant Treatments	Storage Condition	Mean Values
Untreated control	D[1]	11.93b
Untreated control	W[2]	11.75b
Moisturin (1:3)	D	13.25b
Moisturin (1:3)	W	14.68a
Moisturin (1:5)	D	12.81b
Moisturin (1:5)	W	13.50b
Moisturin (1:7)	D	13.87b
Moisturin (1:7)	W	12.12b
AntiStress 2000 (1:20)	D	12.50b
AntiStress 2000 (1:20)	W	12.62b
AntiStress 2000 (1:30)	D	15.00a
AntiStress 2000 (1:30)	W	11.25b
AntiStress 2000 (1:40)	D	16.50a
AntiStress 2000 (1:40)	W	13.62b

Means in the same column followed by the same letter do not differ significantly according to LSD (α= 0.05%).
D[1] = Dry condition. Wet[2] = Wet condition.

Table 5-5. Mean numbers of days until budset by antidesiccant treatments and storage condition and by antidesiccant treatments and storage length of 2+0 ponderosa pine seedlings grown in a greenhouse.

Antidesiccant Treatments	Storage Condition	Mean Values	Antidesiccant Treatments	Storage Length	Means Values
U N	D[1]	83.12b	U N	20	88.06b
U N	W[2]	79.93b	U N	40	75.00b
M1:3	D	83.75b	M1:3	20	86.87b
M1:3	W	84.00b	M1:3	40	80.87b
M1:5	D	82.12b	M1:5	20	92.94a
M1:5	W	85.56a	M1:5	40	75.25b
M1:7	D	83.12b	M1:7	20	90.56b
M1:7	W	85.00b	M1:7	40	77.56b
A1:20	D	82.62b	A1:20	20	91.43a
A1:20	W	87.06a	A1:20	40	88.25b
A1:30	D	81.56b	A1:30	20	91.87a
A1:30	W	86.50a	A1:30	40	76.18b
A1:40	D	86.18a	A1:40	20	91.06a
A1:40	W	86.56a	A1:40	40	80.60b

Means in the same column followed by the same letter do not differ significantly according to LSD (α= 0.05%).
D[1] = Dry condition, W[2] = Wet condition.

mean values of 7.42 and 6.93 mm for seedlings stored dry and wet respectively.

Root growth potential (RGP)

The total number of new roots was significantly affected by antidesiccant treatments ($p=0.0002$), storage length ($p=0.0001$), antidesiccant and storage length treatment interaction ($p=0.0471$), and by antidesiccant and storage condition and storage length treatment interaction ($p=0.0021$) (Table 5-2). The total number of new roots was diminished by the antidesiccant treatments. There was an exception with Moisturin 1:5, however, which was not significantly different from the untreated control (Table 5-3).

For total new roots, the interaction of antidesiccant and storage length treatments showed that the untreated control had the highest mean values for each one of the storage lengths tested (20 and 40 days). But storage length did not significantly affect the growth of new roots of the untreated control. Although the untreated control had mean values greater than 100, the seedlings treated with Moisturin 1:5, stored 40 days and AntiStress 2000 (1:20, 1:30 and 1:40) stored 20 days were not significantly different from the untreated control (Table 5-6).

Chlorophyll fluorescence

There were no antidesiccant or storage condition effects on maximum fluorescence (Fm), variable fluorescence (Fv) or photochemical efficiency (Fv / Fm) (Table 5-7). However, Fm, Fv, and Fv / Fm were significantly affected by storage length. Maximum fluorescence, variable fluorescence, and photochemical efficiency were also significantly affected by the time of measurement ($p=0.0001$) (Table 5-7). Also, the Fv / Fm was affected by the interaction of time of measurement with storage

condition (p=0.0295) (Tables 5-7).

The variable fluorescence of seedlings stored 40 days had the highest mean value with an increase of 14% over those stored 20 days. Mean variable florescence by values of storage length was 1.24 and 1.32 for 20 and 40 days respectively.

Stomatal conductance

The repeated measures analysis of variance showed that antidesiccants and storage condition did not significantly affect stomatal conductance during the study (Table 5-8). However, storage length had a significant effect on stomatal conductance for measurements taken at 0930 h (p=0.0001) and 1130 h (p=0.0196). There were also significant effects on stomatal conductance due to the time of measurement. The interaction of time and storage length was significant for the three measurement times (p=0.0001). There was also a significant four-way interaction between time, antidesiccant treatment, storage condition, and storage length at 0930 h (p=0.0193).

Discussion

The survival results in this study with Moisturin and AntiStress 2000 were consistent with those of Davies and Kozlowski (1974), Magnussen (1986), and Colombo and Odlum (1987). They found that antidesiccant treatments did not affect survival of conifer seedlings. In contrast, Odlum and Colombo (1987) where they found that 81% of *Picea mariana* seedlings were dead 28 days after applying Dow X2-1337 antidesiccant.

The two antidesiccants, Moisturin and AntiStress 2000, applied to seedlings after lifting,

Table 5-6. Mean values on total new roots by antidesiccant and storage length treatments of 2+0 ponderosa pine seedlings grown in a greenhouse.

Antidesiccant Treatment	Storage Length	Mean
Untreated control	20	112.7a
Untreated control	40	105.6a
Moisturin (1:3)	20	74.6b
Moisturin (1:3)	40	43.2b
Moisturin (1:5)	20	79.8b
Moisturin (1:5)	40	99.7a
Moisturin (1:7)	20	83.2b
Moisturin (1:7)	40	53.5b
AntiStress 2000 (1:20)	20	102.5a
AntiStress 2000 (1:20)	40	57.7b
AntiStress 2000 (1:30)	20	86.7a
AntiStress 2000 (1:30)	40	62.6b
AntiStress 2000 (1:40)	20	100.6a
AntiStress 2000 (1:40)	40	66.3b

Means in the same column followed by the same letter do not differ significantly according to LSD (α= 0.05%).

Table 5-7. Summary of repeated measurements analysis of variance of antidesiccants, storage condition and storage length on maximum fluorescence, variable fluorescence and Fv / Fm of 2+0 ponderosa pine seedlings. Values are probability of a greater F-value.

Source of variation	Maximum fluorescence (Fm)[1]	Variable fluorescence (Fv)[1]	Fv / Fm
AT[2]	0.9437	0.9298	0.9846
Storage C(SC)	0.8170	0.4157	0.9859
Storage L(SL)	0.0173	0.0012	0.0192
AT x SC	0.9260	0.6322	0.9832
AT x SL	0.2996	0.3001	0.1508
SC x SL	0.5187	0.8377	0.6067
AT x SC x SL	0.2498	0.0590	0.2337
Time (T)	0.0001	0.0001	0.0001
T x AT	0.9240	0.5915	0.9869
T x SC	0.0819	0.0942	0.0295
T x SL	0.5777	0.8827	0.2559
T x AT x SC	0.1886	0.1114	0.9017
T x AT x SL	0.2797	0.7807	0.3114
T x SC x SL	0.6026	0.6919	0.8010
TxATxSCxSL	0.3080	0.4210	0.1593

[1] Normalized data (Fo=0). AT[2] = Antidesiccant treatments.

Table 5-8. Summary of repeated measurements analysis of variance of effects of antidesiccants, storage condition and storage length on stomatal conductance by time of measurement of 2+0 ponderosa pine seedlings. Values are probability of a greater F-value.

Source of variation	Time of measurement (hours)		
	0930	1130	1330
AT[1]	0.2293	0.6942	0.2128
Storage C(SC)	0.1602	0.4678	0.7399
Storage L(SL)	0.0001	0.0196	0.2314
AT x SC	0.0947	0.4840	0.8115
AT x SL	0.8590	0.3250	0.8875
SC x SL	0.9269	0.9856	0.4375
AT x SC x SL	0.2995	0.9521	0.8949
Time (T)	0.0001	0.0001	0.0001
T x AT	0.0616	0.8388	0.3921
T x SC	0.1036	0.8653	0.5760
T x SL	0.0001	0.0001	0.0001
T x AT x SC	0.0001	0.3979	0.0554
T x AT x SL	0.0774	0.8023	0.1830
T x SC x SL	0.3345	0.3157	0.5021
TxATxSCxSL	0.0193	0.8029	0.6818

AT[1] = Antidesiccant treatments.

delayed budbreak and budset activity 15 % and 4 % respectively in comparison with the untreated control. These results are not consistent with those of Englert (1992) who found that *Quercus rubra* L. and *Acer platanoides* L. seedlings treated with Moisturin broke bud approximately one week early than untreated control seedlings. However, Englert's seedlings were air-dried in a laboratory for different time intervals up to 48 hours and then potted.

Terminal bud of seedlings treated with Anti-Stress 2000 (1:40) were so tight that seedlings could not emerge from the bud. A possible explanation was that the emerging bud could not break the physical barrier. As a result, the seedlings delayed budbreak activity for 5 days.

The greatest foliar damage was exhibited by seedlings dipped in Moisturin (1:5) and by all the concentrations of AntiStress 2000 which had 20 % greater foliar damage than the untreated control. Perhaps, because of the viscosity of those concentrations, penetration of the stomata may have occurred resulting in foliar damage. Similarly, Gale (1961), Davis and Kozlowski (1974), Olofinboba et al. (1974), Kozlowski and Constantinidou (1986), and Williams et al. (1990) found that the occlusion of stomatal pores by antidesiccants was often followed by changes in leaf metabolism due to toxicity.

Storage condition and length treatments increased foliar damage. When seedlings were dried after being dipped in antidesiccants, they showed an 18% increase in foliar damage over seedlings stored wet. Seedlings stored wet for 20 days had 22% greater foliar damage than those stored 40 days. These results are difficult to interpret

because these are no obvious causes for the increased foliar damage from a physiological perspective.

Simpson (1984) found that RGP of *Picea glauca* Voss. and *Tsuga heterophylla* were unaffected by antidesiccant treatments. In this study some antidesiccant treatments reduced the growth of new roots. Moisturin (1:3) treated seedlings had the lowest number of new roots, a 53% reduction over the untreated control. According to Krugman and Stone (1966), 1+1 ponderosa pine seedlings grown in optimum conditions can initiate and elongate new roots in a range between 172 to 534. Because the whole seedling was dipped in the antidesiccants, probably the antidesiccant film did not allow a good root-soil contact. This lack of contact may have affected water and nutrients uptake. As a result, photosynthesis and growth of new roots may have been decreased. Another possibility could be that the antidesiccants tested were toxic the to seedlings.

Storage condition had a negative effect on height and diameter increments. In general, seedlings stored wet had a reduction of 9 and 6 % on height and diameter increments respectively. Consequently, seedlings stored dry had greater growth. According to these results, it is suggested that seedlings treated with antidesiccants and then stored dry in the cold room may have had increased protected against desiccation and which in turn may have contributed to increased growth.

Antidesiccants did not affect chlorophyll fluorescence of ponderosa pine seedlings. This is not consistent with work reported by Kozlowski and Constantinidou (1986) who confirmed that antidesiccants alter the rate of photosynthesis by altering the optical properties of leaves by changes in reflectance and a decrease in light absorption.

However, chlorophyll fluorescence was affected by the storage length treatments. Previous research showed that exposure of conifers to low temperatures inhibits photosynthetic electron transport. This result agrees with Öquist and Ögren (1985). They determined that *Pinus sylvestris* L. seedlings exposed to low temperatures in the field and cold storage causes a more or less complete inhibition of photosynthesis.

According to Krause and Weis (1991) environmental stresses that affect the photosystem II efficiency lead to a characteristic decrease in the photochemical efficiency (Fv / Fm). In this experiment, seedlings stored dry for 20 days had a reduction in Fv / Fm. This result can be explained as follows: when seedlings were dormant, they were lifted, graded and then stored in the cold room. At the time that antidesiccant treatments were applied, they suddenly were exposed to sun light. So seedlings were subjected to low temperatures and high light. This process is called photochilling and is photoinhibitory (Bolhar-Nordenkampf et al. 1991). This may explain the decrease in Fv / Fm. On the other hand, seedling that were stored for more than 20 days in the cold room increased photochemical efficiency whereas Fv and Fm were diminished when the seedlings were stored only 20 days.

Application of Moisturin and AntiStress 2000 did not reduce the stomatal conductance of 2+0 ponderosa pine seedlings in the greenhouse. Because seedlings were not stressed, they may have increased leaf turgor after potting thus causing the antidesiccants were peeled off prematurely. This result was not the same as that obtained by Ceulemans et al. (1983) with azaleas where the plants had a 10% reduction in stomatal conductance. Moreover, Davies and Kozlowski (1974) found that *Pinus*

resinosa Ait. seedlings treated with Silicone had reduced transpiration compared to an untreated control.

Conclusions

The efficacy of both antidesiccants used in this experiment depended on their concentration. Antidesiccant applications did not effect survival. However, low concentrations of both antidesiccants delayed budbreak and budset. Just one treatment (Moisturin 1:5) caused foliar damage but without affecting survival. Height and diameter increments were not affected by antidesiccant treatments. The RGP was adversely affected by antidesiccants treatments. A high concentration of antidesiccants had a strong negative effect on RGP, however, the antidesiccant Moisturin 1:5 had the same effect as the untreated control. Chlorophyll fluorescence was not affected by the antidesiccants tested but was affected by storage length. Stomatal conductance was not affected by either antidesiccant, storage length or storage condition treatments. Time of measurement did however, significantly affect stomatal conductance.

The hypothesis tested in this experiment was rejected because it was found that some significant differences in the morphological, phenological, and physiological characteristics resulted from the treatments imposed. However, the seedlings used in this experiment were not subject to a moisture stress treatments. If potted seedlings had been subjected to increasing levels of moisture stress, antidesiccant effects might have been different. It is suggested that further research on antidesiccant, specifically including different levels of moisture stress, is warranted.

References

Balneaves, J.M. 1988. Packaging and cool-storage effects on growth of *Cupressus macrocarpa* seedlings. N. Z. For. Sci. 18(3):297-303.

Balneaves, J.M. and M.I. Menzies. 1990. Water potential and subsequent growth of *Pinus radiata* seedlings: influence of lifting, packaging, and storage conditions. N. Z. For. Sci. 20(3):57-267.

Binder, W.D. and P. Fielder. 1991. A standardized environmental preconditioning protocol for making variable chlorophyll fluorescence measurements. British Columbia. Ministry of Forestry. FRDA. Project 2.19. Extension. 9 p.

Bolhar-Nordenkampf, H.R., M. Hofer and E.G. Lechner. 1991. Analysis of light-induced reductions of the photochemical capacity in field-grown plants. Evidence for photoinhibition?. Photosynthesis Research. 27:31-39.

Burdett, N.A. 1990. Physiological process in plantation establishment and the development of specifications for forest planting stock. Can. J. For. Res. 20:415-427.

Ceulemans, R., R. Gabriels and I. Impens. 1983. Antitranspirants effects on transpiration, net CO_2 exchange rate and water-use efficiency of azalea. Scientia Horticulturae. 19:125-131.

Colombo, S.J. and K.D. Odlum. 1987. Efficacy of six antitranspirants on black spruce container seedlings. New Forests. 3:293-244.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects of antitranspirants on water relation and photosynthesis of woody plants. J. Amer. Soc. Hort. Sci. 99(4):297-304.

Englert, J. 1992. Physiological and cultural conditions affecting postharvest handling of bare-root nursery plants. M.S. Thesis. Oregon State University. Corvallis. 79 p.

Gale, J. 1961. Studies on plant antitranspirants. Physiol. Plant. 14:777-786.

Garber, M.P. and J. G. Mexal. 1980. Lift and storage practices: their impact on successful establishment of southern pine plantations. N. Z. For. Sci. 10(1):72-78.

Gumpertz, M.L. and C. Brownie. 1993. Repeated measures in randomized block and split-plot experiments. Can. J. For. Res. 23:635-639.

Kozlowski, T.T. and H.A. Constantinidou. 1986. Responses of woody plants to environmental pollution. Part I. Sources and types of pollutants and plant responses. Forestry Abstracts. 47(1):5-51.

Krugman, S.L. and E.C. Stone. 1966. The effect of cold nights on the root-regenerating potential of ponderosa pine. Forest Science. 12:451-459.

Magnussen, S. 1986. Effects of root-coating with the polymer waterlock on survival and growth of drought-stressed bareroot seedlings of white spruce (*Picea glauca* (Moench) Voss) and red pine (*Pinus resinosa* Ait.). Tree Planters' Notes. 37(1):15-19.

Mexal, J.G. and T.D. Landis. 1990. Target seedling concepts: height and diameter. In Rose, R., S. Campbell., T. Landis (eds). Target seedling symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report RM-200. pp. 17-35.

Odlum, K.D. and S.J. Colombo. 1987. The effect of three film-forming antitranspirants on moisture stress of outplanted black spruce seedlings. Tree Planters' Notes. 38(4):23-26.

Olofinboba, M.O., T.T. Kozlowski, and P.E. Marshall. 1974. Effects of antitranspirants on distribution and utilization of photosynthate in *Pinus resinosa* seedlings. Plant and Soil. 40:619-635.

Omi, S.K. 1991. Effects of fall lifting and long term storage on ponderosa pine seedlings and quality. Ph.D. Thesis. Oregon State University. Corvallis. 205 p.

Öquist, G. and E. Ögren. 1985. Effects of winter stress on photosynthetic electron transport and energy distribution between the two photosystems of pine essayed by chlorophyll fluorescence kinetics. Photosynthesis Research. 7:19-30.

Owston, P.W. and W.I. Stein. 1972. Coating material protect Douglas-fir, nobel fir seedlings against drying conditions... not recommended for extended storage-more test needed. Tree Planter's Notes. 23(3):21-23.

Ranney, T.G., N.L. Bassuk, and T.H. Whitlow. 1989. Effect of transplanting practices on growth and water relations of 'colt' cherry trees during reestablishment. J. Environ. Hort. 7(1):41-45.

Ritchie, G.A. 1985. Root growth potential: principles, procedures and predictive ability. In Duryea, M.L. (ed). Proceedings: Evaluating seedling quality: principles, procedures, and predictive abilities of major test. Forest Research Laboratory, Oregon State University, Corvallis. pp. 93-104.

Ritchie, G.A. and Y. Tanaka. 1990. Root growth potential and the target seedling. In Rose, R., S. Campbell., T. Landis (eds). Target seedling symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report RM-200. pp. 37-51

Rose, R., W.C. Carlson, and P. Morgan. 1990. The target seedling concept. In Rose, R., S. Campbell., T. Landis (eds). Target seedling symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report RM-200. pp. 2-8.

SAS Institute Inc., 1993. SAS companion for the microsoft windows environment Version 6. First edition. Carry, NC. 356 p.

Simpson, D.G. 1984. Filmforming antitranspirants: their effects on root growth capacity, storability, moisture stress avoidance, and field performance of containerized conifer seedlings. Forestry Chronicle. 60(6):335-339.

Vidaver, W.E., G.R. Lister, R.C. Brooke, and W.D. Binder. 1990. A manual for the use of variable chlorophyll fluorescence in the assessment of the ecophysiology of conifer seedlings. Canada-British Columbia Forest Resources Development Agreement. Project 2.19. 60 p.

Williams, P.A., A.G. Gordon, and A.W. Moeller. 1990. Effects of five antitranspirants on white spruce and white pine seedlings subjected to greenhouse drought. Tree Planters' Notes. 41(1):34-38.

Win, K., G.A. Berkowitz, and M. Henninger. 1991. Antitranspirant-induced increased in leaf water potential increases tuber calcium and decreases tuber necrosis in water stressed potato. Plant Physiol. 96:116-120.

CHAPTER 6

ANTIDESICCANT EFFECTS ON WATER LOSS OF 2+0 PONDEROSA PINE (*Pinus ponderosa* Dougl.) SEEDLINGS

Abstract

The effects of two antidesiccants: Moisturin® and AntiStress® 2000, each at three different concentrations, on survival, height growth, photochemical efficiency and water loss of 2+0 ponderosa pine seedlings were evaluated over a 40 day period in a walk-in growth room. Six antidesiccant treatments were compared to an untreated control.

Seedling survival was recorded every two days for the 40 day measurement period. Seedling height was measured at the beginning and end of the experiment and chlorophyll fluorescence was scanned one week after the experiment began and at day 40. Water loss was determined by weighing potted seedlings every two days.

Seedling survival, growth and photochemical efficiency were not affected by antidesiccant treatments. Several antidesiccants treatments did significantly reduce water loss. Moisturin® (1:3 and 1:5) and AntiStress® 2000 (1:20) reduced the amount of water loss for at least the first 36 days without affecting photochemical efficiency. Regression equations were developed to estimate the predictive values of water loss for each of the antidesiccant treatments. The model r-square accounted for 81% of the total variation in the data.

Introduction

The amount of water that can be absorbed by the root system is a function of water availability, environmental conditions, demands of the shoot system, and the physiological and morphological characteristics of the root system (Teskey 1991). Continuous water uptake is essential to the growth and survival of plants. If water uptake does not balance water loss, reduction in turgor occurs, causing cessation of growth and eventual death by dehydration (Kramer 1983).

Bareroot seedlings are susceptible to water loss after outplanting even under ideal planting conditions. Thus water stress in newly planted seedlings can lead to a cycle of reduced root growth limited by a lack of photosynthesis and photosynthesis in turn, limited by a lack of water uptake (Burdett 1990). Transplanting shock of newly planted seedlings is often used to describe the water-stressed condition of seedlings following planting (Sands 1984).

To survive transplanting shock seedlings must resist desiccation stresses and adapt to a more hostile environment (Rietveld 1989). When seedlings develop desiccation, reduced shoot growth and even mortality may result (Kramer 1983, Burdett and Simpson 1984, Burdett 1990).

Ponderosa pine seedlings have been planted in different types of environments. When planted on hot and droughty sites, seedlings exist in conditions of low moisture and high evaporative demand (Duryea and Lavender 1982). As a result, seedling mortality at the end of the first growing season can be high if seedlings are not protected against adverse environmental conditions. Different methodologies have been used to

improve seedling survival on hot and dry sites such as manipulating nursery stock so that seedlings are better able to withstand drought (Duryea and Lavender 1982), site preparation (Helgerson et al. 1992), and the use of antidesiccants (Gale and Poljakoff-Mayber 1965; Davenport et al. 1972; Martin 1974).

Film-forming antidesiccants have been applied to reduce transpiration and to improve seedling water status before and after planting (Davies and Kozlowski 1974). Seedlings treated with such antidesiccants have shown a decrease in the transpiration:photosynthesis ratio, thus reducing the irrigation requirements and alleviating the effects of water stress under dry conditions (Gale and Poljakoff-Mayber 1965).

The hypothesis tested in this experiment was that the application of antidesiccants to the whole seedling initially did not reduce the amount of water loss without affecting the photochemical efficiency of ponderosa pine seedlings.

Methods

Plant material

Two-years-old ponderosa pine seedlings (*Pinus ponderosa* Dougl.) were grown at the J. Herbert Stone Nursery, Central Point, OR., and lifted, graded, and stored on February 18, 1993. The seedlot identification number was 479-1. The cones were collected at the Mt. Thielsen area in the Winema National Forest in 1978, at an elevation of 1672 m and seed zone identification number 701(State of Oregon Tree Seed Zone

Map. Western Forest Tree Seed Council). On April 2, seedlings were transported to Corvallis, OR., and then stored in the cold room at the Forest Research Laboratory (FRL) until the experiment was established.

Study establishment

The study was performed on May 15, 1993 at the FRL in a 2.5 x 2.5 x 2.5 m controlled environment walk-in growth room. Light was provided by a mixture of 110-WF96T12/CW/HO fluorescent tubes and 300-W incandescent lamps. The photoperiod was 14 h day^{-1} and the photosynthetic photon flux density at plant level was 160 μ mol m^{-2} s^{-1}. The day:night temperatures were 22:16^{0}C respectively.

Antidesiccant treatments

There were six antidesiccant treatments. Treatment one was the untreated control. Treatments two, three and four were Moisturin® (Burke's Protective Coatings, Washougal, WA.) at 1:3 (antidesiccant:water), 1:5 and 1:7 dilution respectively. Treatments five, six and seven were AntiStress® 2000 (Polymer Ag, Inc., Fresno, CA) at dilutions of 1:20, 1:30 and 1:40 respectively.

There were four seedlings subjected to each antidesiccant treatment as well as the untreated control. In total, 28 seedlings were evaluated. Whole seedlings were dipped in the different antidesiccant treatments and then dried for 10 minutes. The root system of each seedling was then submerged in a half-gallon plastic container with 1.5 liters of water. A rubber stopper that was bored with a 8 mm hole and incised vertically to seal the container. Transparent silicone was used to seal around the seedling stem to prevent water vapor loss. Aluminum foil was then wrapped around the containers to

keep the water cool. Seedlings and containers were weighed with an electronic scale.

Measurements

Seedling survival was recorded every two days until the end of the experiment on day 40. Seedling height was measured at the beginning and at the end of the study period. Variable chlorophyll fluorescence was measured twice, one week after the experiment was started and at the end of the study using an integrating fluorometer (Pacific Fluorotec, Burnaby, British Columbia, Canada). The measured parameters were the maximum fluorescence (Fm) where the electron acceptors are reduced. Variable fluorescence (Fv) is defined as Fm minus ground fluorescence. Ground fluorescence is the amount of chlorophyll measured in the sphere, when the electron acceptors are fully oxidized. The ratio Fv / Fm is a measure of the photochemical efficiency of photosystem II (Öquist and Ögren 1985; Krause and Weis 1991).

Water loss was determined by weighing the seedlings and containers every two days for forty days with a electronic scale (Ohaus-Galaxy 4000D). The change in weight between measurement periods provides an estimate of transpired water vapor (Meyer and Anderson 1952; Teskey 1991).

Experimental design and statistical analyses

Treatment differences in seedling height increment and photochemical efficiency were analyzed using the SAS general linear models procedure (SAS Institute Inc. 1993) for a randomized complete block design. There were four seedlings per treatment. Because seedling pots were weighed 20 times, a repeated measurements analysis of variance for a randomized complete block design (Gumpertz and Brownie 1993) was

performed to evaluate water loss.

Because there were only 28 pots measured over 20 time periods, there were not enough degrees of freedom to carry out the multivariate test for time effects. Therefore, orthogonal polynomials were computed to test for linear and quadratic trends over time. This allowed the independent computation of the linear and quadratic contribution of the independent variable, time. Also, regression equations were developed to estimate the predicted amount of water loss of seedlings treated with antidesiccants at each point of time (Steel and Torrie 1960).

A logarithmic transformation was used because the residual plot from the analysis of the original data indicated non-constant variance. A log transformation resulted in a satisfactory scatter of the residuals. The Least Significant Difference test was used to determine significant differences among treatment means at the α=0.05 level.

Results

Seedling survival and height increment

During the 40 day study period there was no seedling mortality. For all treatments, seedlings flushed nine to ten days after being placed in the growth room. Analysis of variance (Table 6-1) showed no antidesiccant effects on the height increment of ponderosa pine seedlings (p=0.5579).

Table 6-1. Analysis of variance of antidesiccant treatment effects on height increment of 2+0 ponderosa pine seedlings.

Source	df	MS	F	Prob>F
Block	3	0.342	0.25	0.8570
AT[1]	6	1.122	0.84	0.5579
Error	18	1.342		

AT[1] = Antidesiccant treatments.

Table 6-2. Analysis of variance of antidesiccant treatment effects on photochemical efficiency of 2+0 ponderosa pine seedlings.

Source of variation	df	MS	F	Prob>F
(a) One week after potting				
Block	3	0.0024	1.68	0.2071
AT[1]	6	0.0023	1.62	0.1977
Error	18	0.0014		
(b) At the end of the experiment				
Block	3	0.0027	1.30	0.3042
AT[1]	6	0.0046	2.23	0.0879
Error	18	0.0375		

AT[1] = Antidesiccant treatments.

Photochemical efficiency

Analysis of variance (Table 6-2) showed no significant differences in photochemical efficiency either one week after potting ($p=0.1977$) or at the end of the experiment ($p=0.0879$).

Water loss

The repeated measures analysis of variance showed that averaged over 40 days, antidesiccant treatments had a significant effect on water loss ($p=0.0028$)(Table 6-3). Moreover, there were significant time effects ($p=0.0001$). There was also a significant interaction of time and antidesiccant treatments ($p=0.0001$).

From the first day of measurement until the end of measurement period, two concentrations of Moisturin (1:3 and 1:5) and AntiStress 1:20 reduced water loss significantly in relation to the untreated control (Table 6-4). The only exception were AntiStress 1:20 on day 38 and Moisturin 1:5 on day 2 (Table 6-4).

Using estimates of the time effects, regression equations for the untreated control and six antidesiccant treatments were estimated by the following equations.

$$LOG(WUT1) = 1.123 + 0.096 * D - 0.0027 * D^2$$

$$LOG(WUT2) = 0.649 + 0.110 * D - 0.0031 * D^2$$

$$LOG(WUT3) = 0.936 + 0.087 * D - 0.0024 * D^2$$

$$LOG(WUT4) = 1.107 + 0.116 * D - 0.0032 * D^2$$

$$LOG(WUT5) = 0.904 + 0.106 * D - 0.0029 * D^2$$

$$LOG(WUT6) = 1.073 + 0.094 * D - 0.0026 * D^2$$

$$LOG(WUT7) = 1.147 + 0.096 * D - 0.0023 * D^2$$

Where:

LOG(WUT1) is the logarithm of water loss of the untreated control.

D is the day of measurement. D^2 is the square day of the day of measurement.

LOG(WUT2) is the logarithm of water loss of Moisturin® 1:3.

LOG(WUT3) is the logarithm of water loss of Moisturin® 1:5

LOG(WUT4) is the logarithm of water loss of and AntiStress® 1:20, and so on.

The r^2 from the repeated model indicated that the model accounted for 81% of the total variation in the data.

Predicted values from the regression equations for water loss of ponderosa pine seedlings treated with antidesiccants showed that water loss by seedlings treated with Moisturin 1:3 (treatment 2) was lower than the rest of the antidesiccant treatments and the untreated control (Figure 6-1). The polynomial analysis (Table 6-5) showed that the rate of water loss slowed over the 40 days for all antidesiccant treatments and the untreated control ($p=0.0314$).

Water loss as a function of days after potting showed that the highest concentrations of both Moisturin and AntiStress were effective in reducing water loss on day two. There were no significant differences among the antidesiccants treatments and the untreated control. On day four, seedlings that were treated with Moisturin 1:5 (Trmt 3) showed a reduction in water loss. From day four until day 36, seedlings subjected to Moisturin (1:3 and 1:5) and AntiStress (1:20) had the same response of diminishing water loss. The rest of the antidesiccant treatments and the untreated control were statistically the same.

Table 6-3. Repeated measurements analysis of variance on water loss of 2+0 ponderosa pine seedlings by antidesiccant and time treatment.

Source of variation	df	MS	F	Prob>F
Block	3	0.438	0.86	0.4804
AT[1]	6	2.672	5.23	0.0028
Error (a)	18	0.510		
Time (T)	19	86.456	4322.50	0.0001
T x AT	114	0.1338	55.81	0.0001
Error (b)	342	0.000056		

AT[1] = Antidesiccant treatment.

At days 38 and 40 only two concentrations of Moisturin (1:3 and 1:5) were effective in controlling water loss relative to the untreated control. AntiStress 1:20 was efficient in controlling water loss for the first 36 days but was not significantly different from the untreated control on days 38 and 40 (Table 6-4).

Discussion

The survival and height increment of ponderosa pine seedlings was not affected by the six antidesiccants treatments tested in the growth room. These results differ from with Roy's (1966) findings that Douglas-fir seedlings treated with Foli-gard antidesiccant had a greater height increment than the untreated control. In contrast, Ranney et al.

Table 6-4. Mean log water loss for each day of measurements and antidesiccant treatments of 2+0 ponderosa pine seedlings.

D / AT[1]	Untreated Control	Moisturin			AntiStress 2000		
		1:3	1:5	1:7	1:20	1:30	1:40
Day 2	1.06a	0.64b	0.87a	1.02a	0.84b	1.01a	1.07a
Day 4	1.31a	0.87b	1.09b	1.34a	1.10b	1.27a	1.35a
Day 6	1.44a	0.99b	1.22b	1.50a	1.26b	1.39a	1.49a
Day 8	1.53a	1.07b	1.33b	1.61a	1.36b	1.44a	1.58a
Day 10	1.59a	1.14b	1.37b	1.67a	1.43b	1.54a	1.64a
Day 12	1.65a	1.25b	1.42b	1.74a	1.49b	1.59a	1.69a
Day 14	1.69a	1.30b	1.46b	1.79a	1.53b	1.63a	1.72a
Day 16	1.73a	1.35b	1.49b	1.84a	1.58b	1.67a	1.75a
Day 18	1.77a	1.38b	1.52b	1.88a	1.62b	1.70a	1.80a
Day 20	1.80a	1.42b	1.55b	1.92a	1.66b	1.74a	1.85a
Day 22	1.83a	1.46b	1.56b	1.96a	1.69b	1.77a	1.90a
Day 24	1.86a	1.49b	1.57b	1.99a	1.72b	1.80a	1.95a
Day 26	1.88a	1.51b	1.63b	2.02a	1.75b	1.82a	1.98a
Day 28	1.90a	1.53b	1.65b	2.04a	1.77b	1.87a	2.01a
Day 30	1.92a	1.55b	1.67b	2.07a	1.80b	1.87a	2.04a
Day 32	1.94a	1.57b	1.68b	2.09a	1.82b	1.88a	2.07a
Day 34	1.96a	1.58b	1.70b	2.12a	1.85b	1.90a	2.10a
Day 36	1.98a	1.60b	1.72b	2.14a	1.87b	1.92a	2.13a
Day 38	1.99a	1.61b	1.74b	2.16a	1.89a	1.94a	2.15a
Day 40	2.03a	1.63b	1.75b	2.18a	1.91a	1.97a	2.17a

Antidesiccant treatments means in the same row followed by the same letter do not differ significantly according to LDS (α=0.5).
D= day of measurement. AT[1] = Antidesiccant treatment.

Table 6-5. Analysis of variance for the linear and quadratic model of water loss of 2+0 ponderosa pine seedlings treated with antidesiccants.

Source of variation	df	MS	F	Prob>F
Linear				
Block	3	0.0149	0.46	0.7122
AT[1]	6	0.0504	1.56	0.2155
Error	18	0.5817		
Quadratic				
Block	3	0.0066	2.24	0.1182
AT[1]	6	0.0090	3.03	0.0314
Error	18	0.0029		

AT[1] = Antidesiccant treatment.

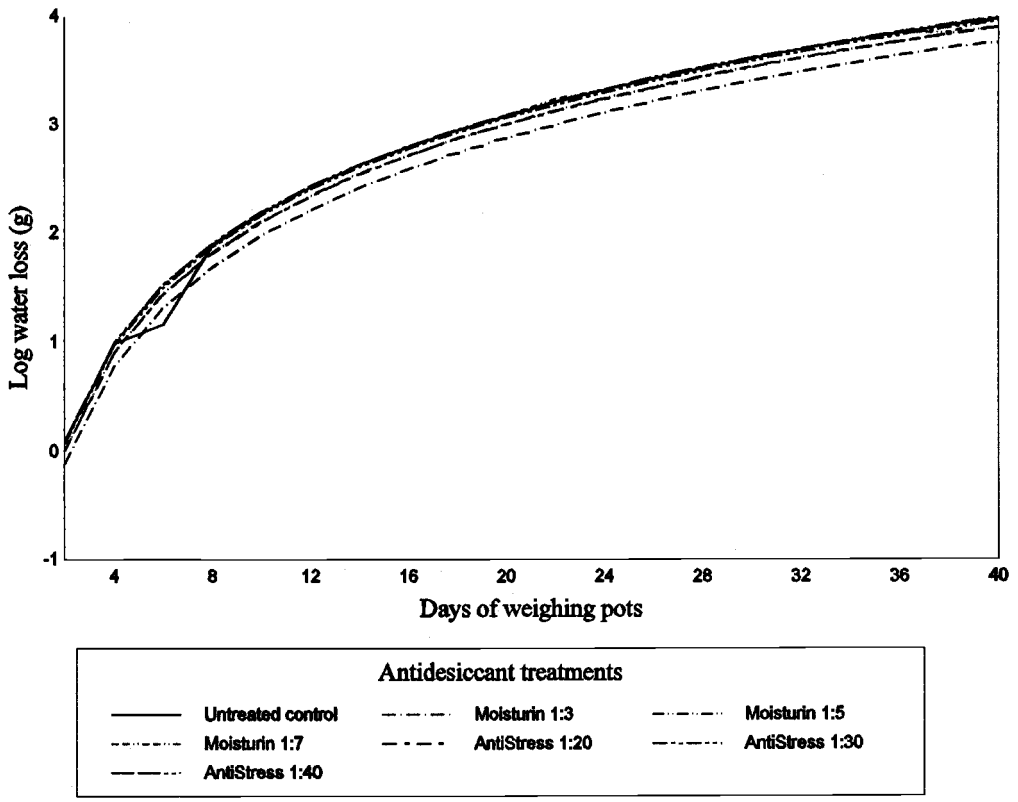

Figure 6-1. Log water loss per day of weighing pots of 2+0 ponderosa pine seedlings treated with antidesiccants.

(1989) found that Folicote antidesiccants (wax emulsion film) did not improve height increment of colt cherry trees (*Prunus avium* x *pseudocerasus*).

The Moisturin and AntiStress 2000 treatments tested did not affect the photochemical efficiency of ponderosa pine seedlings as revealed by the Fv / Fm ratio. These results may be due to the environmental conditions the seedlings were exposed to. The seedlings were probably not under stress. Some studies have demonstrated that environmental stresses affect photosystem II efficiency and lead to a characteristic decrease in the Fv / Fm ratio (Krause and Weis 1991). However, in this study the antidesiccants and concentrations used did not affect light absorption or the photochemical efficiency of photosystem II as shown by the Fv / Fm ratio. This result was consistent with Huner et al. (1993) but disagrees with work done by Kozlowski and Constantinidou (1986) who reported that antidesiccants alter the optical properties of leaves.

Moisturin 1:3 and 1:5 and AntiStress 1:20 were effective in reducing the amount of water loss by ponderosa pine seedlings. The results obtained in this study are consistent with earlier findings (Gale and Hagan 1966; Tracy and Lewis 1981; Kramer 1983; Ranney et al. 1989). Undoubtedly, the reduction in water loss was the result of an increased resistance to water movement created by the antidesiccant film (Gale and Hagan 1966). It should be noted, however, that at the conclusion of the 40 day experiment, some mortality of roots was observed. Depending upon when root mortality occurred, the likely increased resistance to flow may have contributed to decreased water loss as the experiment progressed.

Conclusions

This experiment demonstrated that the antidesiccants applied to the whole seedling did not affect seedling survival or height growth. The results also showed that the photochemical efficiency of the seedlings, as measured by the Fv / Fm ratio, was not affected. However, several of the antidesiccant treatments were effective in significantly reducing water loss. Moisturin 1:3 and 1:5 had the lowest mean values over the 40 day study period followed by AntiStress 1:20 which was effective over the first 36 days. Moisturin 1:3 was the most effective treatment in reducing water loss but this did not increased height growth over the forty day period.

Several of the antidesiccants tested in this experiment showed that they were able to reduce water loss after seedlings were placed in the growth room. If these results could be replicated in the field, antidesiccants might be helpful to maintaining favorable seedling water potential after outplanting.

References

Burdett, A.N. 1990. Physiological processes in plantation establishment and the development of specifications for forest planting stock. Can. J. For. Res. 20:415-427.

Burdett, A. N. and D.G. Simpson. 1984. Lifting, grading, packaging, and storing. In Duryea, M.L. and T.L. Landis (eds). Forestry Nursery Manual: production of bareroot seedlings. Martinus Nijhoff / Dr. W. Junk Publishiers, The Hague. pp. 227-234.

Davenport, D.C., M.A. Fisher, and R.M. Hagan. 1972. Some counteractive effects of antitranspirants. Plant Physiol. 49:722-724.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects on antitranspirants on water relations and photosynthesis of woody plants. J. Amer. Soc. Hort. Sci. 99(4):297-304.

Duryea, M.L. and D.P. Lavender. 1982. Water relations, growth, and survival of root-wrenching Douglas-fir seedlings. Can. J. For. Res. 12:545-555.

Gale, J. and R.M. Hagan. 1966. Plant antitranspirants. Ann. Rev. Plant Physiol. 17:269-282.

Gale, J. and A. Poljakoff-Mayber. 1965. Effect of antitranspirant treatment on leaf temperatures. Plant and Cell Physiol. 6:111-115.

Gumpertz, M.L. and C. Brownie. 1993. Repeated measures in randomized block and split-plot experiments. Can. J. For. Res. 23:635-639.

Helgerson, O.T., M. Newton and D.H. McNabb. 1992. Site preparation. In Hobbs, S.D., S.D. Tesch., P.W. Owston., R.E. Steward., J.C. Tappeiner II and G.E. Wells (eds). Reforestation practices in southwestern Oregon and northern California. Forest Research Laboratory. Oregon State University. Corvallis, Oregon. pp. 232-256.

Huner, N.P.A., G. Öquist, V.M. Hurry, M. Krol, S. Falk and M. Griffith. 1993. Photosynthesis, photoinhibition and low temperatures acclimation in cold tolerant plants. Photosynthesis Research. 37:19-39.

Kozlowski, T.T. and H.E. Constantinidou. 1986. Response of woody plants to environmental pollution. Forestry Abstracts. 47:5-51.

Kramer, P.J. 1983. Water relations of plants. Academic Press. San Diego, California. 489 p.

Krause, G.H. and E. Weis. 1991. Chlorophyll fluorescence and photosynthesis: the basic. Ann. Rev. Plant. Physiol. Plant Mol. Biol. 42:313-349.

Martin, J.D. 1974. Antitranspirants, possible uses in floriculture. Flor. Rev. 155(4051):19-21, 130-138.

Meyer, B.S. and D. B. Anderson. 1952. Plant physiology. D. Van Nostrand Co, Inc. Canada. 748 p.

Öquist, G. and E. Ögren. 1985. Effects of winter stress on photosynthetic electron transport and energy distribution between the two photosystems of pine essayed by chlorophyll fluorescence kinetics. Photosynthesis Research. 7:19-30.

Ranney, T.G., N.L. Bassuk, and T.H. Whitlow. 1989. Effects of transplanting practices on water relations of 'Colt' cherry trees during reestablishment. J. Environ. Hort. 7(1):41-45.

Rietveld, W.J. 1989. Transplanting stress in bareroot conifer seedlings: its development and progression to establishment. North. J. Appl. For. 6:99-107.

Roy, D.F. 1966. Effects of a transpiration retardant and root coating on survival of Douglas-fir planting stock. Tree Planters' Notes. 79(1):10-13.

Sands, R. 1984. Transplanting stress in radiata pine. Aust. For. Res. 14:67-72.

SAS Institute Inc., 1993. SAS companion for the microsoft windows environment. Version 6. First edition. Cary, NC. 356 p.

Steel, R.G.D. and J.H. Torrie. 1960. Principles and procedures of statistics. McGraw-Hill Book Company, Inc. New York. 481 p.

Teskey, R.O. 1991. Measuring water uptake by roots. In Lassoie, J.P. and T.H. Hinckley (eds). Techniques and approaches in forest tree ecophysiology. CRC Press Inc. Boca Raton, FL. pp. 77-90.

Tracy, T.E. and A.J. Lewis. 1981. Effects of antitranspirants on hydrangea. HortScience. 16(1):87-89.

CHAPTER 7

GENERAL CONCLUSIONS

This thesis research examined the effects of three concentrations of two antidesiccants on survival, growth and physiology of 2+0 ponderosa pine seedlings. Four experiments were established and four hypotheses tested. One experiment was performed in the field, two in a greenhouse, and one in a walk-in growth room.

There were five physiological responses analyzed, chlorophyll fluorescence, stomatal conductance, electrolyte leakage, water loss and root growth potential. There were six phenological and morphological variables studied, budbreak and budset, seedling survival, height and diameter increments, and foliar damage.

The antidesiccants tested affected root growth potential (experiment 3, Chapter 5). Seedlings treated with Moisturin 1:3 had reduced growth of new roots. New root growth is known to enhance photosynthesis in newly planted seedlings by alleviating water stress and therefore allowing increased opening of the stomatal aperture and additional carbon dioxide assimilation. Nevertheless, seedling shoot growth was not affected in this research. Possibly, the potential negative effects of reduced root growth due to antidesiccants may have been negated by the optimal growing conditions that were maintained.

Antidesiccants reduced stomatal conductance of ponderosa pine seedlings (experiment 1, Chapter 3). But the reduction in stomatal conductance did not translate into an improvement in seedling survival or height growth. The high survival of ponderosa pine may in part be a function of the adaptation mechanisms that regulates

water uptake and water loss which conveys cellular resistance and permits survival of seedlings in dry environments with or without antidesiccants. However, in this field experiment post-planting moisture conditions were unusually favorable. Had they been more typical, the outcome of this experiment might have been different.

Overall, with one exception, when antidesiccants were applied to actively growing seedlings and then subjected to different periods of wind exposure, water loss increased or remained unchanged from the untreated control (experiment 2, Chapter 4). However, in another experiment several antidesiccants decreased water loss (experiment 4, Chapter 6). As in other experiments though, the growth of these seedlings was not affected.

In experiment 3 (Chapter 5), the antidesiccants tested on ponderosa pine seedlings had a negative effect on the following variables: budbreak, budset, and foliar damage. But height and diameter increments as well as survival were not affected. It is known that ponderosa pine exhibits fixed growth which means that the number of cells are determined the previous growing season. Consequently, antidesiccant effects on height increment might not have been detectable until the second growing season after antidesiccant application. When seedlings were stored dry in the cold room, height and diameter increments were improved regardless of antidesiccants. Days to budbreak and budset, and foliar damage were increased when stored wet. Moreover, antidesiccants did not affect chlorophyll fluorescence but this variable was affected by the storage length. Seedlings that were stored 40 days had better photochemical efficiency than seedlings stored 20 days.

It is speculated that the disparities between physiological, morphological, and phenological variables were generated by stress associated with the storage treatments in the cold room. It is hypothesized that seedlings met chilling requirements when they were stored more than 20 days in the cold room. As a result the photochemical efficiency of these seedlings was higher than those stored 20 days. Moreover, it was found that antidesiccants applied to dormant seedlings did not affect light absorption or chlorophyll fluorescence.

In this thesis research the proposition was that antidesiccants would have a positive effect on ponderosa pine seedlings. Based on the preponderance of evidence from the four experiments conducted, it is concluded that under the conditions of this study, antidesiccants had little overall effect on ponderosa pine seedlings.

Even though this thesis research proceeded as planned, there are several limitations worth mentioning. In the first experiment (Chapter 3) newly ouplanted seedlings in the field were probably never subjected to severe water stress. In the second experiment (Chapter 4) the wind treatments were not truly replicated from a statistical perspective. Operationally this was not achievable. Under ideal circumstances different sets of seedlings should have been used for each wind exposure treatment replication. Seedlings grown in the greenhouse in experiment 3 (Chapter 5) did not experience drought conditions. Had a series of increasing moisture stress conditions been imposed, the potential effects due to antidesiccants could have been more thoroughly tested. The water uptake experiment (Chapter 6) did not have an adequate number experimental units to carry out the multivariate analysis. Finally, plant water potential was not

measured but probably should have been in most of the experiments.

The results of this research imply that antidesiccants applied to ponderosa pine seedlings might not be of much benefit. However, the use of antidesiccants needs to be further investigated in the Pacific Northwest. The use of these chemicals to prevent winter desiccation could lead to new applications especially in higher elevation areas where seedlings can simultaneously be exposed to cold soils and warm upslope winds. It would also be important to know whether several applications in a single growing season would benefit seedling survival and growth in water-limiting environments. Finally, research on the use of antidesiccants in semi-arid regions of the world should continue to be pursued.

BIBLIOGRAPHY

Alm, A. and J.Stanton. 1990. Field trials of root dipping treatments for red pine, jack pine, and white spruce nursery stock in Minnesota. Tree Planters' Notes. 41(3):18-20.

Badertscher, D. 1991. Moisturin. Burke's protective coatings. Technical Bulletin. Washougal, WA. 5 p.

Balneaves, J.M. 1988. Packaging and cool-storage effects on growth of *Cupressus macrocarpa* seedlings. N. Z. For. Sci. 18(3):297-303.

Balneaves, J.M. and M.I. Menzies. 1990. Water potential and subsequent growth of *Pinus radiata* seedlings: influence of lifting, packaging, and storage conditions. N. Z. For. Sci. 20(3):257-267.

Binder, W.D. and P. Fielder. 1991. A standardized environmental preconditioning protocol for making variable chlorophyll fluorescence measurements. British Columbia. Minister of Forestry. FRDA. Project 2.19. Extension. 9 p.

Blum, A. and A. Ebercon. 1981. Cell membrane stability as a measure of drought and heat tolerance in wheat. Crop Science. 21:43-47.

Bolhar-Nordenkampf, H.R., M. Hofer and E.G. Lechner. 1991. Analysis of light-induced reductions of the photochemical capacity in field-grown plants. Evidence for photoinhibition?. Photosynthesis Research. 27:31-39.

Burdett, A.N. 1990. Physiological processes in plantation establishment and the development of specifications for forest planting stock. Can. J. For. Res. 20:415-427.

Burdett, A.N. and D.G. Simpson. 1984. Lifting, grading, packaging, and storing. In Duryea, M.L. and T.L. Landis (eds). Forestry Nursery Manual: production of bareroot seedlings. Martinus Nijhoff / Dr. W. Junk Publishiers, The hague. pp. 227-234.

Ceulemans, R., R.Gabriels., and I. Impens. 1983. Antitranspirant effects on transpiration, net CO_2 exchange rate and water-use efficiency of azalea. Scientia Horticulturae. 19:125-131.

Chen, T.H.H., P. Murakami., P. Lombard, and L.H. Fuchigami. 1991. Desiccation tolerance in bare-rooted apple trees prior to transplanting. J. Environ. Hort. 9(1):13-17.

Cleary, B.D., R.D. Greaves, and P.W. Owston. 1988. Seedlings. In Cleary, B.D., R.D. Greaves, and R.K. Hermann (eds). Regenerating Oregon's Forests. A guide for the regeneration forester. Oregon State University. Extension Service. Fifth printing. Corvallis, OR. pp. 63-97.

Colombo, S.J. and K.D. Odlum. 1987. Efficacy of six antitranspirants on black spruce container seedlings. New Forests. 3:239-244.

Colombo, S.J., D.P. Webb and C. Glerum. 1984. Frost hardiness testing: An operational manual for the use with extended greenhouse culture. Ontario Ministry of Natural Resources, Forest Research Report No. 110. 14 p.

Coutts, M.P. 1981. Effect of root or shoot exposure before planting on the water relation, growth, and survival of sitka spruce. Can. J. For. Res. 11(3):703-709.

Davenport, D.C., M.A. Fisher and R.M. Hagan. 1972. Some counteractive effects of antitranspirants. Plant Physiol. 49:722-724.

Davenport, D.C., P.E. Martin, and R.M. Hagan. 1972. Antitranspirants for conservation of leaf water potential of transplanted citrus trees. HortScience. 7(5):511-512.

Davies, W.J. and T.T. Kozlowski. 1975a. Effects of applied abscisic acid and plant water stress on transpiration of woody angiosperms. For. Sci. 21(2):191-195.

Davies, W.J. and T.T. Kozlowski. 1975b. Effect of applied abscisic acid and silicone on water relations and photosynthesis of woody plants. Can. J. For. Res. 5:90-96.

Davies, W.J. and T.T. Kozlowski. 1974. Short-and long-term effects of antitranspirants on water relations and photosynthesis of woody plants. J. Amer. Soc. Hort. Sci. 99(4):297-304.

Duryea, M.L. and D.P. Lavender. 1982. Water relations, growth, and survival of root-wrenching Douglas-fir seedlings. Can. J. For. Res. 12:545-555.

Earth Info, Inc. 1994. U.S. Weather data to 1993-Wester states on CD-Rom.

Englert, J.M. 1992. Physiological and cultural conditions affecting postharvest handling of bare-root nursery plants. M.S. Thesis. Oregon State University. 79 p.

Evans, J. 1982. Plantation forestry in the tropics. Clarendon Press. Oxford. 472 p.

Gale, J. 1961. Studies of plant antitranspirants. Physiol. Plant. 14:777-786.

Gale, J. and R.M. Hagan. 1966. Plant antitranspirants. Ann. Rev. Plant Physiol. 17:269-282.

Gale, J. and A. Poljakoff-Mayber. 1965. Effects of antitranspirant treatment on leaf temperatures. Plant and Physiol. 6:111-115.

Gale, J. and A. Poljakoff-Mayber. 1967. Plastic films on plants as antitraspirants. Science. 156:650-652.

Garber, M.P. and J. G. Mexal. 1980. Lift and storage practices: their impact on successful establishment of southern pine plantations. N. Z. J. For. Sci. 10(1):72-78.

Grossnickle, S.C., J.E. Major., J.T. Arnott, and V.M. Lemay. 1991. Stock quality assessment through integrated approach. New Forests. 5:77-91.

Gumpertz, M.L. and C. Brownie. 1993. Repeated measures in randomized block and split-plot experiments. Can. J. For. Res. 23:625-639.

Hagiladi, A. and O. Ziv. 1986. Use of antitranspirants for control of powdery mildew on field grown roses. J. Environ. Hort. 4(2):65-68.

Hale, M.G. and D.M. Orcutt. 1987. The physiology of plants under stress. John Wiley and Sons New York. 206 p.

Hales, S. 1961. Vegetable staticks. First published 1727. Oldbourne Book Co. Ltd. Great Britain. 241 p.

Helgerson, O.T., M. Newton and D.H. McNabb. 1992. Site preparation. In Hobbs, S.D., S.D. Tesch., P.W. Owston., R.E. Steward., J.C. Tappeiner II and G.E. Wells (eds). Reforestation practices in southwestern Oregon and northern California. Forest Research Laboratory. Oregon State University. Corvallis, Oregon. pp. 232-256.

Henckel, P.A. 1964. Physiology of plants under drought. Ann. Rev. Plant Physiol. 15:363-386.

Huner, N.P.A., G. Öquist, V.M. Hurry, M. Krol, S. Falk and M. Griffith. 1993. Photosynthesis, photoinhibition and low temperatures acclimation in cold tolerant plants. Photosynthesis Research. 37:19-39.

Ingram, D.L. and D. Buchanan. 1981. Measurement of direct heat injury of roots of three woody plants. HortScience. 16(6):769-771.

Ingram, D.L. and T.H. Yeager. 1987. Effects of irrigation frequency and a water-absorbing polymer amendment on ligustrum growth and moisture retention by a container medium. J. Environ.Hort. 5(1):19-21.

Jarvis, P.G. 1975. Water transfer in plants. In de Vries, D.A. and N. Algan (eds). Heat transfer in the biosphere. I Transfer processesin plant environment. John Wiley and Sons. New York. pp. 369-394.

Johnson, J.D. 1991. Measuring stress-related plant growth regulators. In Lassoie, J.P. and T.M. Hinckley (eds). Techniques and approaches in forest tree ecophysiology. CRC Press. Boca Raton, Florida. pp. 275-292.

Johnson, J.D. and M.L. Cline. 1991. Seedling quality of southernpines. In Duryea, M.L. and P.M. Dougherty (eds). Forest Regeneration Manual. Klumer Academic Publishers. The Netherlands. pp. 143-159.

Kastori, R., N. Petrovic., and Z. Stankovic. 1991. Possibilities ofusing antitranspirants in plant production. Bioloski-Vestnik. 39(1-2):109-113.

Kozlowski, T.T. and H.A. Constantinidou. 1986. Responses of woody plants to environmental pollution. Part I. Sources and types of pollutants and plant responses. Forestry Abstracts. 47:5-51.

Kozlowski, T.T. and J.B. Mudd. 1975. Responses of plants to air pollution. In Kozlowski, T.T. (eds). Responses of plants to air pollution. Academic Press. New York. pp. 1-8.

Kramer, P.J. 1983. Water relations of plants. Academic Press. New York. 489 p.

Krause, G.H. and E.Weis. 1984. Chlorophyll fluorescence as a tool in plant physiology. II. Interpretation of fluorescence signals. Photosynthesis Res. 5:139-157.

Krause, G.H. and E.Weis. 1991. Chlorophyll fluorescence and photosynthesis: the basic. Annu.Rev. Plant Physiol. Plant Mol. Biol. 42:313-349.

Krugman, S.L. and E.C.Stone. 1966. The effects of cold nights on the root-regenerating potential of ponderosa pine. Forest Science. 12:451-459.

Kyaw, W., G.A. Berkowitz, and M. Henninger. 1991. Antitranspirants-induced increases in leaf water potential tuber calcium and decrease tuber necrosis in water-stressed potato plants. Plant Physiology. 96:116-120.

Lebovits, A. 1966. Permeability of polymers to gases, vapors, and liquids. Modern Plastics. 43(7):139-213.

Lee, K.J. and T.T. Kozlowski. 1974. Effects of silicone antitranspirant on woody plants. Plant and Soil. 40:493-510.

Lefevre, R.E., A.C. Cameron, and N.C. Peterson. 1991. Influence of moisture loss during storage and new growth of conifer seedlings. J. Environ. Hort. 9(2): 92-96.

Leopold, A.C., M.E. Musgrave, and K.M. Williams. 1981. Solute leakage resulting from leaf desiccation. Plant Physiol. 68:1222-1225.

Levitt, J. 1972. Responses of plants to environmental stresses. Academic Press. New York. 697 p.

Lipe, W.N. and J.A. Skinner. 1979. Effect of an antitranspirant on water use, growth, and yield of greenhouse-grown potatoes. HortScience. 14(3):239-241.

Magnussen, S. 1985. Effects of root-coating with the polymer Waterlock on survival and growth of drought-stressed bareroot seedlings of white spruce (*Picea glauca* (Moench) Voss) and red pine (*Pinus resinosa* Ait.). Tree Planters' Notes. 37(1):15-19.

Marshall, J.D. 1985. Carbohydrates status as a measure of seedling quality. In Duryea, M.L. (ed). Proceedings: evaluating seedling quality: principles, procedures, and predictive abilities of a major test. Forest Research Laboratory. Oregon State University. pp. 49-58.

Marshall, J.G. D.R. Cyr. and E.B. Dumbroff. 1991. Drought tolerance and the physiological mechanisms of resistance in northern coniferous seedlings. For. Can., Ont. Region, Sault Ste. Marine, Ont. COFRDA. rep 3314. 17 p.

Martin, J.D. 1974. Antitranspirants, possible uses in floriculture. Flor. Rev. 115(4051):19-21, 130-138.

Martin, U., S.G. Pallardy and Z.A. Bahari. 1987. Dehydration tolerance of leaf tissues of six woody angiosperm species. Physiol. Plantarum. 69:182-186.

Martineau, J.R., J.E. Specht., H.Williams and C.Y. Sullivan. 1979. Temperature tolerance in soybeans. I. Evaluation of a technique for assessing cellular membrane thermostability. Crop Science. 19:75-78.

McKay, H.M. 1992. Electrolyte leakage from fine roots of conifer seedlings: a rapid index of plant vitality following cold storage. Can. J. For. Res. 22:1371-1377.

Mexal, J.G. and T.D. Landis. 1990. Target seedling concepts: height and diameter. In Rose, R., S. Campbell., T. Landis (eds). Target Seedling Symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report R-M 200. pp. 17-35.

Meyer, B.S. and D.B. Anderson. 1952. Plan physiology. D.Van Nostrand Co, Inc. Canada. 748 p.

Milburn, J.A. 1979. Water flow in plants. Longman. London. 225 p.

Mitchell, W.K., G. Dunsworth., D.G. Simpson, and A. Vyse. 1990. Planting and seeding. In Lavender, D.P., R. Parish., C.M. Johnson., G. Montgomery., A. Vyse., R.A. Willis, and D. Winston (eds). Regenerating British Columbia's Forest. University of British Columbia. Vancouver. pp. 235-253.

Molina, R. and J.M. Trappe. 1984. Mycorrhiza management in bareroot nurseries. In Duryea, M.L. and T.L. Landis (eds). Forestry Nursery Manual: production of bareroot seedlings. Martinus Nijhoff / Dr. W. Junk Publishiers, The Hague. pp. 211-223.

Nauth, R. 1947. The chemestry and technology of plastics. Reinhold Publishing Co. New York. 522 p.

Nobel, P.S. 1991. Physicochemical and environmental plant physiology. Academic Press, Inc. San Diego, Cal. 635 p.

Noggle, G.R., and G.J. Fritz. 1983. Introductory plant physiology. Prentice-Hall, New Jersey. 627 p.

Odlum, K.D. and S.J. Colombo. 1987. The effect of three film-forming antitranspirants on moisture stress of outplanted black spruce seedlings. Tree Planters' Notes. 38(4):23-26.

Olofinbona, M.O., T.T. Kozlowski. and P.E. Marshall. 1974. Effects of antitranspirants on distribution and utilization of photosynthate in *Pinus resinosa*. Plant and Soil. 40:619-635.

Omi, S.K. 1991. Effects of fall lifting and long term storage on ponderosa pine seedlings and quality. Ph.D. Thesis, Oregon State University. 205 p.

Öquist, G. and G. Malmberg. 1989. Light and temperature dependent inhibition of photosynthesis in frost-hardened and un-hardened seedlings of pine. Photosynthesis Res. 20:261-277.

Öquist, G. and E. Ögren. 1985. Effects of winter stress on photosynthetic electron transport and energy distribution between the two photosystems of pine essayed by chlorophyll fluorescence kinetics. Photosynthesis Research. 7:19-39.

Owston, P.W. and W.I. Stein. 1972. Coating material protect douglas-fir, nobel fir seedlings against drying conditions...not recommended for extended storage-more test needed. Tree Planter's Notes. 23(3):21-23.

Pallardy, S., J.S. Pereira, and W.C. Paerker. 1991. Measuring the state of water in tree systems. In Lassoie, J.P. and T.M. Hinckley (eds). Techniques and approaches in forest tree ecophysiology. CRN. Boca Raton, FL. pp. 27-74.

Peck, K.M. and S. J. Wallner. 1982. Ecotypic differences in heat resistance of aspen leaves. HortScience. 17(1):52-53.

Perkins, T.G., G.T. Adams, and R.M. Klein. 1991. Desiccation or frezing?. Mechanisms of winter injury to red spruce foliage. Amer. J. Bot. 78(9):1207-1217.

Poljakoff-Mayber, A., J. Gale., H. Aluma., I. Kahane., H. Ginsburg., Z. Goldschmidt., I. Nir., and E. Shabi. 1967. Final technical report on effect of plant antitranspirants on certain physiological processes of forest seedlings and other plant material. Project No. A10-FS-10. The Hebrew University of Jerusalem. Jerusalem. 107 p.

Ranney, T.G., N.L Bassuk, and T.H. Whitlow. 1989. Effect of transplanting practices on growth and water relations of 'Colt' cherry trees during reestablishment. J. Environ.Hort. 7(1):41-45.

Rietveld, W.J. 1989. Transplanting stress in bareroot conifer seedlings: its development and progression to establishment. North. J. Appl. For. 6(3):99-107.

Ritchie, G.A. 1985. Root growth potential: principles, procedures and predictive ability. In Duryea, M.L. (ed). Proceedings: Evaluating seedlings quality: principles, procedures, and predictive abilities of a major test. Forest Research Laboratory, Oregon State University. Corvallis, OR. pp. 93-106.

Ritchie, G.A. and Y. Takana. 1990. Root growth potential and the target seedling. In Rose, R., S. Campbell., T. Landis (eds). Target Seedling Symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report R-M 200. pp. 37-51.

Rose, R., W.C. Carlson, and P. Morgan. 1990. The target seedling concept. In Rose, R., S. Campbell., T. Landis (eds). Target Seedling Symposium: proceedings, combined meeting of the Western Forest Nursery Association. USDA Forest Service, Rocky Mountains Forest and Range Experimental Station. General Technical Report R-M 200. pp. 2-8

Rowan, S.J. 1983. Loss of feeder roots lower seedlings survival more than sever black root rot. Tree Planters' Notes. 34:18-20.

Roy, D.F. 1966. Effects of a transpiration retardant and root coating on survival of Douglas-fir planting stock. Tree Planter's Notes. 79(1):10-13.

Sands, R. 1984. Transplanting stress in radiata pine. Aust. For. Res. 14:67-72.

SAS Institute Inc., 1993. SAS companion for the microsoft windows environment. Version 6. First edition. Cary, NC. 356 p.

Schildknecht, C.E. 1952. Vinyl and related polymers. John Willey and Sons, Inc. New York. 723 p.

Schönherr, J. and M.J. Bukovac. 1972. Penetration of stomata by liquids. Plant Physiol. 49:813-819.

Shanahan, J.F., I.B. Edwards., J.S. Quick and J.R. Fenwick. 1990. Membrane thermostability and heat tolerance of spring wheat. Crop Science. 30:247-251.

Shekour, G.M., C.R. McDavid, and R.A.I. Brathwaite. 1991. Responses of sweetcorn to different rates and frequencies of application of PMA and Vapor Gard. Tropical Agriculture. 68(3):225-230.

Simpson, D.G. 1984. Filmforming antitranspirants: their effects on root growth capacity, storability, moisture stress avoidance, and field performance of containerized conifer seedlings. Forestry Chronicle. 60(6):335-339.

Sinclair, T.R., G.E. Bingham., E.R. Lemon and L.H. Allen Jr. 1975. Water use efficiency of field-grown maize during moisture stress. Plant Physiol. 56:245-249.

Slatyer, R.O. and J.F. Bierhuizen. 1964. The influence of several transpiration suppressants on transpiration, photosynthesis, and water-use efficiency of cotton leaves. Aust. J. Biol. Sci. 17(1):131-146.

Stannett, V. and H. Yasuda. 1964. Permeability. In Raff, R.A.V. and K.W. Doak (eds). Crystalline olefin polymers. Part II. High Polymers. Vol. XX. Interscience Publishers. New York. pp. 131-184.

Steel, R. G.D. and J.H. Torrie. 1960. Principles and procedures of statistics. Mc Graw-Hill Book Company Inc. New York. 481 p.

Steinberg, S.L., M.J. McFarland, and J.W. Worthington. 1990. Antitranspirant reduce water use by peach trees following harvest. J. Amer. Soc. Hort. Sci. 115(1):20-24.

Sterrett, J.P. 1985. Paclobutrazol: a promising growth inhibitor for injection into woody plants. J. Amer. Soc. Hort. Sci. 110(1):4-8.

Swietlik, D. and S. Miller. 1983. The effect of paclobutrazol on growwth and response to water stress of apple seedlings. J. Amer. Soc. Hort. Sci. 108(6):1076-1080.

Teskey, R.O. 1991. Measuring water uptake by roots.In Lassoie, J.P. and Hinckley, T.M. (eds). Techniques and approaches in forest tree ecophysiology. CRC Press. Boca Raton, Fl. pp. 77-90.

Theophrastus. 300 BC. Enquiry into plants and minor works on odours and weather signs. Book IV: of the trees and plants special to particular districts and position. William Heinemann. London.

Tracy, T.E. and A.J. Lewis. 1981. Effects of antitranspirants on hydrangea. HortScience. 16(1):87-89.

van den Driessche, R. 1989. Changes in osmotic potential of Douglas-fir (*Pseudotsuga menziesii*) in relation to temperature and photopheriod. Can. J. For. Res. 19:413-421.

Vera-Castillo, J.A.G. 1986. Estudios de algunos factores que influyen en la produccion de *Pinus montezumae* Lamb., en vivero. Tesis de Maestria en Ciencias. Colegio de Postgraduados. Chapingo, Mexico. 141 p.

Vidaver, W.E., G.R. Lister., R.C. Brooke, R.C., and W.D. Binder. 1990. A manual for the use of variable chlorophyll fluorescence in the assessment of the ecophysiology of conifer seedlings. FRDA Project 2.19. Canada-British Columbia Forest Resources Development Agreement. 60 p.

Waisel, Y. G.A. Borger. and T.T. Kozlowski. 1969. Effects of phenylmercury acetate on stomatal movement and transpiration of excised *Betula papyrifera* Marsh. leaves. Plant Physiol. 44:685-690.

Wakeley, P.C. 1965. Results of southern pine planting experiments in the middle twenties. J. Forestry. 67:237-241.

Weber, F.R. and C. Stoney. 1986. Reforestation in arid lands. Volunteers in Technical Assistance. Arlington, VA. 335 p.

Weyers, J.D.B. and H. Meidner. 1990. Methods in stomatal research. Longman Scientific and Technical. Great Britain. 233 p.

Whitlow, T.H., N.L. Bassuk., T.G. Ranney, and D.L. Reichert. 1992. An improved method for using electrolyte leakage to assess membrane competence in plant tissues. Plant Physiol. 98:198-205.

Williams, P.A., A.M. Gordon, and A.W. Moeller. 1990. Effects of five antitranspirants on white spruce pine seedlings subjected to greenhouse drought. Tree Planters' Notes. 4(1):34-38.

Wilson, V. 1968. Planning rework programs on nonstocked plantations on the Umpqua National Forest. In U.S. Forest Service (ed). First Reforestation Workshop. Proceedings. 164-179.

Win, K., G.A. Berkowitz, and M. Henninger. 1991. Antitranspirant-induced increases in leaf water potential increase tuber calcium and decrease tuber necrosis in water-stressed potato plants. Plant Physiol. 96:116-120.

Wood, J.E. 1990. Black spruce and jack plantation performance in boreal Ontario: 10-year results. North. J. Appl. For. 7(4):175-179.