Moisture Stress Acclimation Reduces Sensitivity of Containerized Eldarica Pine to Harsh Handling[1]

John T. Harrington, James T. Fisher, and John G. Mexal[2]

Abstract.--Improper handling of seedlings can cause increased mortality and reduce growth following outplanting. Container growing systems reduce physical handling impacts on tree seedlings; however, containerization does not alleviate the need for careful handling. To determine if moisture-stress conditioning (MSC) can improve seedling tolerance to harsh handling, containerized Pinus eldarica seedlings were subjected for 8 weeks to two growing regimes, well watered and watered only when wilted at dawn (moisture-stress conditioned, MSC). Handling treatments consisted of a factorial combination of days without water (0, 2 and 4 days) and a 90-minute incubation at either 20, 25 or 30^0 C. Shoot water potential and new root production over 14 days was used to evaluate the effects of poor handling. MSC treatments bufferred the effects of handling treatments on shoot water potential. MSC seedlings also had an average 33% greater new root production than well-watered controls. As handling treatments became more severe, new root production was reduced in control seedlings, with little change in MSC seedlings.

INTRODUCTION

Handling is often overlooked as a factor in plantation establishment success. The handling process begins when seedlings are lifted, removed from refrigerated storage or shipped from the greenhouse. The handling process ends when the seedlings are planted and many factors in the process can impact seedling performance. These factors include temperature stress, moisture stress, root desiccation, physical damage to the seedling and duration of stress imposition. In most pre-plant and planting situations, handling stresses are often caused by a combination of these factors.

Roots exposed for prolonged periods of time become so dry they no longer function in water and mineral uptake, and as loci for new root development. Rough seedling handling can cause similar reductions of seedling quality (Marx and Hatchell 1986, South and Stumpff 1990).

Root exposure and physical damage to the seedling can reduce survival (Figure 1). More specifically, exposure to elevated temperatures, low humidity and rough handling can severely reduce transplant success. Switzer (1960)

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[2]John T. Harrington is Superintendent Mora Research Center, Mora N.M., James T. Fisher & John G. Mexal both Professors of Agronomy and Horticulture, New Mexico State University, Las Cruces, N.M.

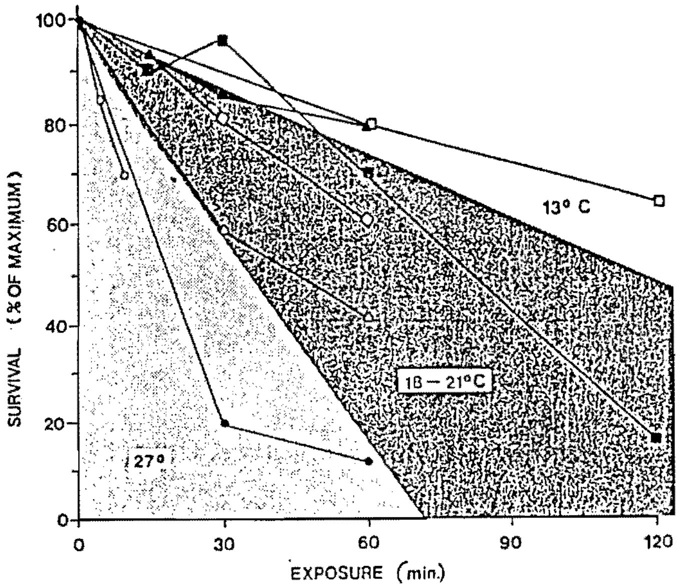

Figure 1.--Effects of exposure temperatures and duration on survival of conifer seedlings (Mexal and South 1991).

found exposures as brief as 5 minutes at 18°C reduced bareroot loblolly pine survival. However, the impact of exposure time is strongly influenced by temperature. Dierauf and Marler (1971) found 30-and 60-minute exposures at 13°C reduced survival by 14% and 20%, respectively. The exposure effect was more pronounced at 18°C, with reductions of 38% and 50%.

Rough handling practices can also influence bareroot seedling survival. Dropping bags of bareroot Sitka spruce (Picea sitchensis) seedlings once, from a height of 3 meters, reduced root growth potential (RGP) (Tabbush 1986). As the number of drops increased, RGP and survival decreased concomitantly.

Containerized seedlings have increased resistance to mishandling. This is attributed to the root ball (or plug; media, water and roots) of containerized stock (Fancher et al. 1989). The root ball and container protect root systems and cushion rough handling. Containerized stock is not completely protected from these factors in the case of extremely harsh handling. Containerized seedlings can be susceptible to the temperature and moisture stresses associated with the handling process.

Tissue-water status is a critical factor in a seedling's ability to withstand handling stress. Moisture-stress conditioning (MSC) improves drought tolerance in plants (Seiler 1985, Zwiazek and Blake 1989). However, the benefits of short, sublethal exposure of plants to droughts is relatively short-term (Morgan 1984), but the effects can be long enough to increase tolerance to harsh handling.

The purpose of this study was to examine the effects of MSC on container-grown eldarica pine (Pinus eldarica) seedling tolerance to water stress and root exposure, as measured by new root production.

METHODS AND MATERIALS

Plant Material

Seed used in this study was obtained from a stand of open-pollinated eldarica pine at the Fabian Garcia Research Center (FGRC) in Las Cruces, NM, and sown on January 29, 1990. The seed was germinated and grown in 175-cc R-L Tubes (Ray-Leach Corp., Oregon, USA) containing a 2:1:1 (v/v/v) peat moss:perlite:vermiculite mixture. Seedlings were grown under ambient light conditions, supplemented with the light from 100-watt incandescent bulbs to maintain a 16-h photoperiod. Greenhouse temperatures ranged from 18.5 - 24°C during the day to 13 -24°C during the night. Relative humidity in the greenhouse ranged from 40 - 70% during the day to 65 - 95% during the night. Seedlings were fertilized weekly with 15 ml of a Peter's Conifer Grower nutrient solution (150 ppm N, 22.5 ppm P, 118.5 ppm K). Seedlings were kept in these growing regimes until July 25, 1990, when 400 seedlings were selected to be treated. Seedling selection was based on morphological uniformity.

Moisture Availability Treatments

Seedlings were randomly placed in one of two moisture availability treatment levels (200 trees per level). One level was a well-watered control, and the second level consisted of watering seedlings only when the shoot was wilted at dawn. Moisture availability treatment was applied for 8 weeks. Seedlings remained in the greenhouse throughout treatment imposition. Seedlings were fertilized every 2 weeks with 15 ml of a Peter's Conifer Hardener nutrient solution

(50.00 ppm N, 109.10 ppm P, 69.17 ppm K).

Handling Treatments

A factorial arrangement of handling treatments consisted of three levels of last watering date and three levels of exposure temperature. Last watering date levels consisted of 1) watered immediately before exposure treatment, 2) watered 2 days before exposure treatment and 3) watered 4 days before exposure treatment. Treatments involved removing the seedlings from their tubes and placing them in a temperature controlled incubator at either 20, 25 or 30°C for 90 minutes.

Seedling Measurements

Root collar diameter and shoot length were measured for each seedling before and after moisture availability treatment imposition. Root collar diameter was measured to the nearest 0.1 mm. Shoot length, from the cotyledon scar to the tip of the growing apex, was measured to the nearest 1.0 mm.

Shoot water potential, osmotic potential at full turgor, and osmotic potential at incipient plasmolysis were measured during moisture availability treatment imposition. Measurements were taken on the upper 2 cm of the shoot of three seedlings at approximately 2-week intervals on two seedlings per moisture availability treatment level. This was done with a Scholander Pressure Bomb (PMS Corp. Corvallis, OR) with a graphic pressure volume analysis procedure described by Schulte and Hinkley (1985).

Following the 90-minute exposure, and again after new root production incubation, shoot water potential was measured at the root collar of two seedlings from each water*handling*MSC treatment.

New root production was used to test seedling response to handling treatments. New root growth was evaluated using a 14-d aeroponics system (Harrington 1991). New root growth was expressed by an increase in root volume. The technique involves rinsing the root system free of the growing medium, measuring the initial volume of the root system gravimetrically and measuring the volume again after incubation in the root growth tanks (Harrington 1991).

Statistical Analysis

The conditioning (moisture availability) portion of the study was a completely randomized design. There were 200 seedlings used per moisture availability treatment level to define shoot height and root collar growth. Shoot height and root collar growth were analyzed using analysis of variance. Analysis of variance was used to examine the effect of moisture availability treatment levels on plant water relations during treatment imposition.

The handling treatment design consisted of a factorial arrangement of three levels of last watering date and three levels of incubation temperature. The experimental design was a randomized complete block design with blocking by root growth analysis tank. All analyses were performed using SAS Version 5.0 (SAS Institute, Cary, NC).

RESULTS

Moisture availability significantly impacted seedling growth. Shoot elongation and root collar diameter growth were reduced 45% and 57%, respectively, in MSC seedlings (Figure 2). Root system volume was also reduced 32% in MSC seedlings. While not tested, MSC seedlings appeared to have a more pronounced blue foliage color and more dead needles than controls.

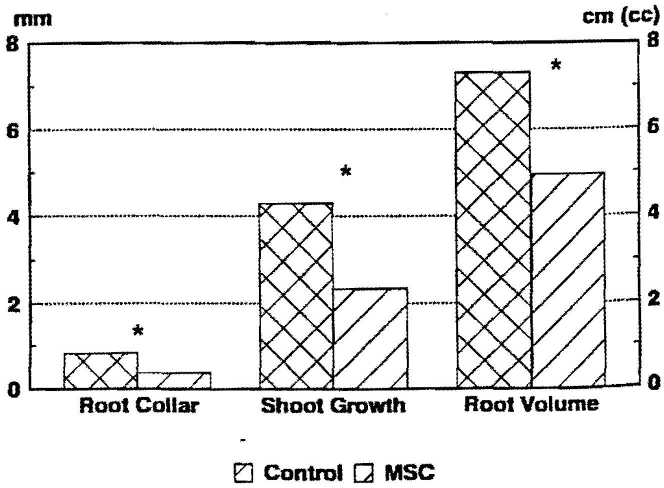

Figure 2.--Effects of moisture stress conditioning on root collar growth (mm), shoot growth (cm) and, final root volume (cc) following 8 weeks of moisture availability treatment. * denotes significant difference.

Predawn water potentials and osmotic potentials at full turgor and incipient plasmolysis were significantly influenced by moisture availability treatments following MSC treatment imposition (Figure 3). MSC seedlings had lower water and osmotic potentials following treatment imposition.

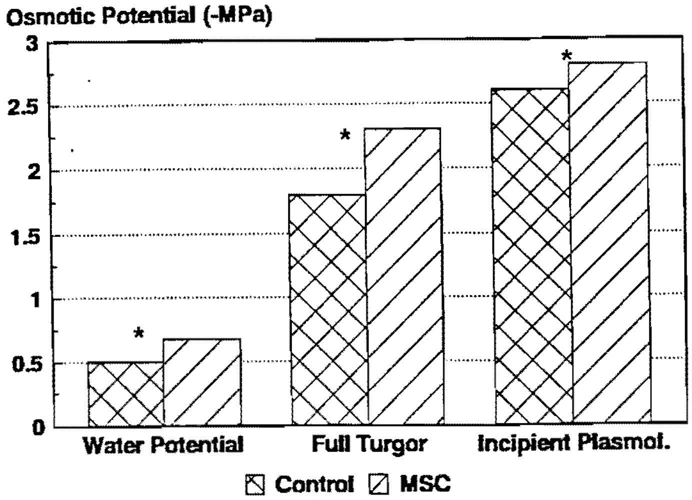

Figure 3.--Effects of moisture stress conditioning on shoot water potential. osmotic potential at full turgor and, osmotic potential at incipient plasmolysis following 8 weeks of moisture availability treatment. * denotes significant difference.

MSC seedlings tended to have higher shoot water potentials than control seedlings during the dry-down exposure (Figure 4a). However, the only significant difference occurred at 2 d without water. After 14 d in the root growth chamber, all treatments had recovered to about -0.64 MPa, except the control treatment which went 4 d without water (Figure 4b).

MSC seedlings also had higher water potentials following exposure to high temperature (Figure 5a). Again, after 14 d in the root growth chamber, water potentials increased for both treatments (Figure 5b). However, controls averaged 0.5 to 1.5 Mpa lower than MSC seedlings. MSC appeared to prevent the imposition of severe stress and to speed the recovery to pretreatment water potentials.

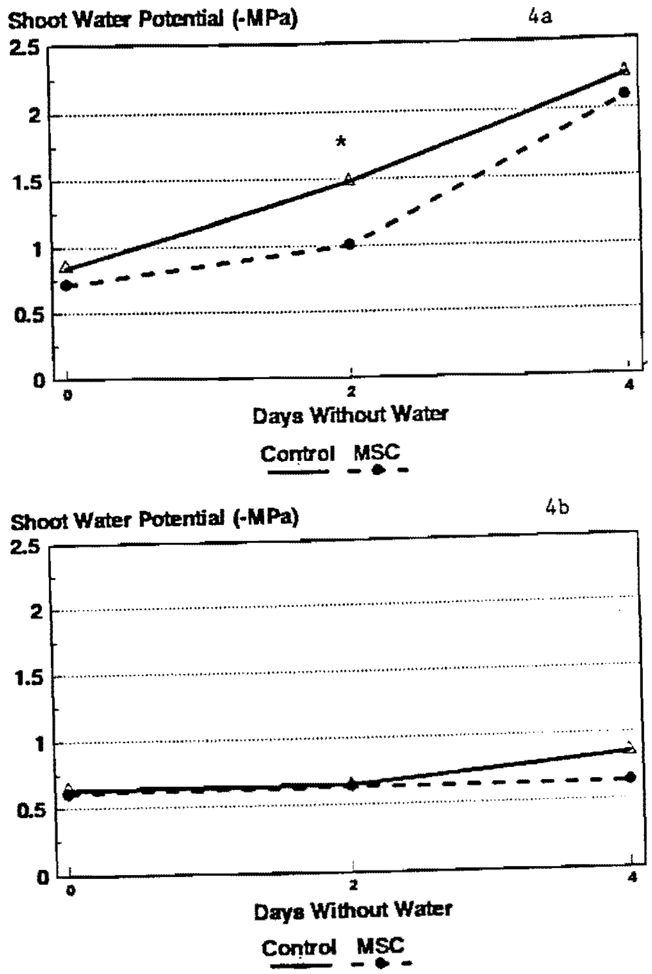

Figure 4.--a. Shoot water potentials of control and MSC seedlings by days without water prior to new root incubation. b. Shoot water potentials of control and MSC seedlings by days without water following new root incubation. * denotes significant difference.

Days without water had a negative impact on new root production of control seedlings (Figure 6). RGP decreased linearly with increasing stress. Furthermore, RGP was related to shoot water potential at the beginning of the RGP test. Following 4 d without water, RGP was reduced 56% compared to control seedlings at the 0 d level. Conversely, MSC seedling new root production was not affected by days without water. New root production averaged 0.58 cc, regardless of treatment.

As exposure temperature increased, new root production in MSC seedlings increased, while the opposite was true for control seedlings (Figure 7). RGP of MSC seedlings at 30°C was 30% greater

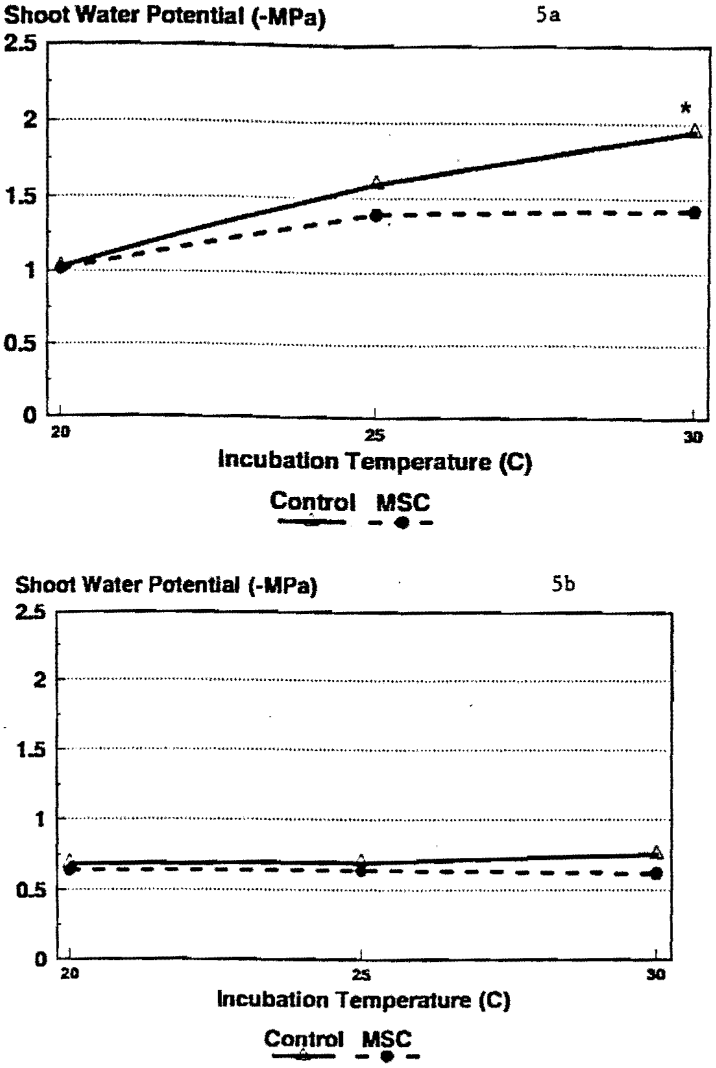

Figure 5.--a. Shoot water potentials of control and MSC immediately following exposure to high temperatures. b. Shoot water potentials of control and MSC exposed to high temperatures following new root growth incubation. * denotes significant difference.

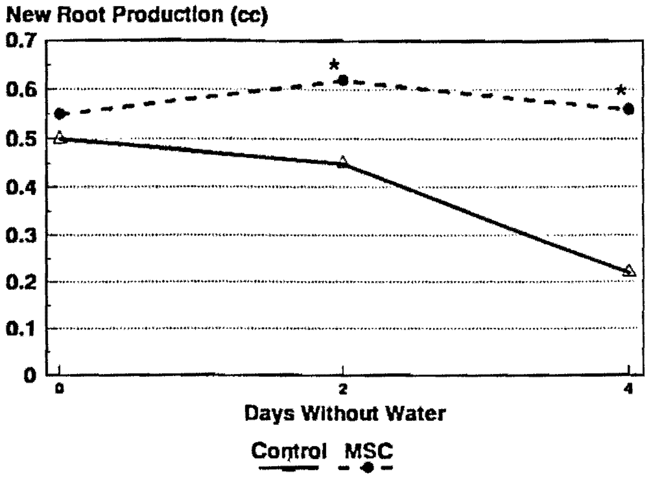

Figure 6.--Effect of moisture stress conditioning on new root production by the number of days without water prior to new root growth incubation. * denotes significant difference.

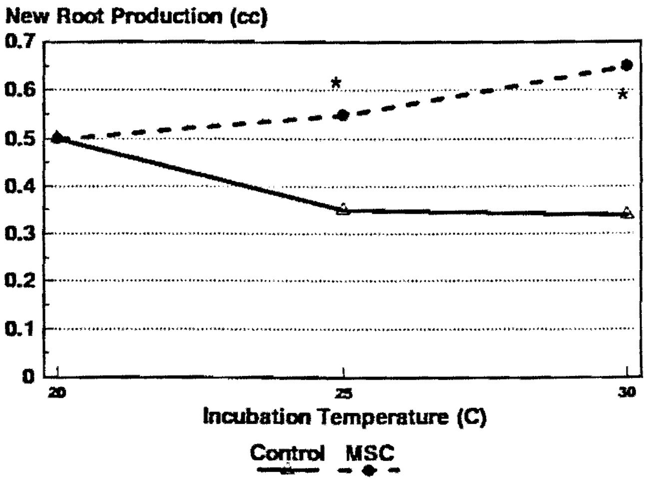

Figure 7.--Effect of moisture stress conditioning on new root production following seedling exposure to either 20, 25 or 30^{0}C for 90 minutes. * denotes significant difference.

than at 20^{0}C. Conversely, RGP of control seedlings at 30^{0}C was 30% lower than RGP at 20^{0}C. MSC seedlings appear better adapted to harsh conditions of both water stress and heat.

DISCUSSION

Treatment Effects

Moisture-stress conditioning treatment resulted in both decreased root collar growth and height growth. This was also found in previous studies (Harrington 1991, Seiler 1985, Seiler and Johnson 1988, O'Reilly et al. 1989). In western hemlock seedlings, degree of branching is also reduced by moisture availability, further contributing to reductions in shoot size (O'Reilly et al. 1989). Moisture-stress conditioning also resulted in smaller root systems. Other researchers have found similar reduction in MSC black alder and loblolly pine (Seiler 1985, Seiler and Johnson 1988).

Moisture-stress conditioning effects were evident by the second week of treatment, with MSC seedlings having 36% lower osmotic potential at full turgor relative to control seedlings. This constituted 78.6% of the overall adjustment for the 8-week treatment period (Harrington 1991). Hydroponically cultured black spruce (Picea mariana) seedlings showed considerable osmotic adjustment after only 1 week of moisture-stress treatment (Zwiazek and Blake 1989). This indicates some conifer seedlings can more readily adjust to stressful environments. To obtain an appreciable gain in drought tolerance, specifically osmotic adjustment, may take only 2 weeks. Such a short exposure can minimize the growth losses caused by such a treatment, while providing the physiological benefits of osmotic adjustment.

Handling Effects

Stresses imposed during handling can influence the overall magnitude of transplanting stress as defined by Rietveld (1989). He defines transplant stress as both a condition (physiological state value) and a recovery process (time function). Handling effects can influence both the state of the seedling and its ability to recover and acclimate to its new environment.

Tissue water status is a critical factor in seedling ability to withstand transplant stress. Water stress can detrimentally affect almost every physiological process in a plant. Handling can severely impact the water status of a seedling. The shift in reforestation practices to a more intensive and expensive container system was, in part, caused by problems associated with the handling process.

While containerization can minimize the physical effects of handling, poor handling can reduce the physiological quality of container-grown seedlings.

Moisture-stress conditioning effectively buffered the effects of handling treatments on seedling water relations. As days without water increased, MSC seedlings consistently maintained higher shoot water potentials relative to control seedlings. As handling incubation temperatures increased, MSC seedlings also maintained higher shoot water potentials than control seedlings. Seedlings receiving MSC, and subjected to the most severe treatment imposed (4 d without water and a 90 minute incubation at 30°C), had shoot water potentials 0.45 MPa higher than controls immediately following treatment. These data indicate MSC reduces the detrimental effects of harsh handling on seedling physiology. This is further supported by the root production data.

Across all handling treatments, MSC seedlings produced more new roots than control seedlings. Elevated exposure temperatures enhanced new root production of MSC seedlings. New root production was 10% greater at 25°C, and 30% greater at 30°C than root production at 20°C. The opposite was found in control seedlings with reductions in new root production of 30.6% at 25°C and 35.3% at 30°C. Feret et al. (1985) also demonstrated a 35-minute exposure of bareroot loblolly pine seedlings resulted in a 35% decline in root growth potential.

The exposure effects on new root production observed here demonstrate effects of exposure on seedling performance. However, reductions in survival attributable to exposure can be variable. Bareroot loblolly pine seedlings exposed for 1 h at 18°C had a 58% reduction in survival (Dierauf and Marler 1971). In another study, an exposure as brief as 10 minutes at the same temperature resulted in a 30% reduction (Switzer 1960). Summarizing data from several studies, Mexal and South (1991) found exposure temperature was correlated with survival of southern pines. Exposure to high temperatures might cause complete mortality after as little as 1 h, while 2 h exposure to lower temperatures might cause no more than 25% mortality. Research indicates this sensitivity can be ameliorated by the conditioning treatments the seedlings receive in the nursery.

CONCLUSIONS

These results demonstrate how containerized eldarica pine seedlings can acclimate quickly to reduced water levels, and that MSC enhances their ability to generate new root tissue. It appears MSC can effectively moderate the effects of harsh handling (elevated exposure temperatures and intervals without water) in containerized eldarica pine seedlings. As increasing numbers of private, noncommercial landowners begin planting trees as part of state and national reforestation programs, it will be necessary to produce seedlings more resilient to the effects of mistreatment. Moisture stress conditioning may be one practice that will help in this effort.

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