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REGULAR ARTICLE

Root desiccation and drought stress responses of bareroot *Quercus rubra* seedlings treated with a hydrophilic polymer root dip

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Abstract Root hydrogel, a hydrophilic polymer, has been used to improve transplanting success of bareroot conifer seedlings through effects on water holding capacity. We examined mechanisms by which Terra-sorb® Fine Hydrogel reduces damage that occurs when roots of 1-year old, dormant northern red oak (*Quercus rubra* L.) were subjected to short-term (1, 3, and 5 h) pre-transplanting desiccation and long-term (45 days) drought stress following transplanting in a controlled environment chamber or greenhouse conditions. Hydrogel-treated seedlings had 80% greater root moisture content than non-root dipped control seedlings following the pre-transplanting desiccation period. Hydrogel reduced root membrane leakiness by 31% 5 h after the desiccation exposure. Hydrogel-treated seedlings did not show greater differences in shoot length, plant dry mass, root volume, net photosynthesis, and stomatal conductance compared with control seedlings following the 45-day drought stress exposure. A reduction in mean number of days to bud break in hydrogel-treated seedlings, combined with delayed tissue moisture loss (linked to higher stem water potential), suggests that hydrogel may have provided stress protection to aid survival under short-term desiccation, which may be beneficial toward alleviating initial transplanting stress.

Keywords Desiccation · Electrolyte leakage · Gas exchange · Hydrogel · Moisture content · Northern red oak · Stem water potential

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Introduction

Water deficit serves as a primary cause of transplant stress in forest tree seedlings (Burdett 1990; Haase and Rose 1993). Continuous water stress conditions inhibit plant growth, stomatal conductance, and CO_2 assimilation (Brakke and Allen 1995; Gómez-Cadenas et al. 1996). This desiccation-induced injury may delay root regeneration, which is essential for the establishment of newly-planted seedlings. Desiccation of the root system was associated with increased

mortality in conifers (Coutts 1981; Feret et al. 1985; Tabbush 1987; Grossnickle 1988) and broadleaved species (Insley and Buckley 1985). Seedlings are at risk of desiccation following lifting or unpacking bundles and at planting (Edgren 1984; McKay 1996). Short-term (i.e., several hours) exposure to desiccation may cause a significant decrease in survival (Hermann 1967; Coutts 1981; Tabbush 1987), which is exacerbated by continued exposure (Mullin 1978; Brønnum 2005). For example, Hermann (1967) reported 100%, 60%, and 50% survival of Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) seedlings exposed to 32°C for 15, 60, and 120 min, respectively. Plant exposure in a drying atmosphere can affect internal water status (Balneaves and Menzies 1988) with root exposure causing reduced shoot water potential (Dierauf and Garner 1975; Coutts 1981), thereby potentially affecting survival and long term growth of transplants.

Although shoots and roots may be inhibited by water stress, roots are more sensitive than shoots (Sucoff et al. 1985). Within the root systems, fine roots are readily injured by direct air exposure (Feret et al. 1985), with thin roots losing moisture more quickly than thick roots (Insley and Buckley 1985). In addition, biophysical and biochemical changes in plant cell membranes can be altered by a number of stress factors including desiccation, which is usually encountered during the interval between lifting and planting (McKay 1996). Previous studies showed that desiccation caused root death as a result of membrane leakiness (Sarvaš 2003), and is linked to poor seedling performance (Coutts 1981; McKay 1992; Davis and Jacobs 2005). Therefore, it is possible that the physiological dysfunctions may occur rapidly, perhaps even before seedlings are outplanted.

The root dipping technique, a process of coating the root system of bareroot seedlings with soil slurries, sphagnum moss, and hydrophilic polymer called hydrogel, has been practiced for more than 30 years primarily to improve transplanting success of conifer seedlings (Dierauf and Garner 1975; Magussen 1985; Echols et al. 1990; Sloan 2004). While an abundance of information concerning the beneficial effects of root dips to help alleviate desiccation is available, some evidence indicates that using clay slurry may have detrimental effects on seedlings (Mullin and Bunting 1979). Furthermore, it has been suggested that the chemical composition of hydrogel is phytotoxic, thereby contributing to cell membrane damage (Alm and Stanton 1993; Sloan 2004; Sarvaš 2003).

The electrolyte leakage test has been used as an important factor in determining the ability of seedlings to survive desiccation (McKay and White 1997). In addition, use of root respiration as a measure of seedling status could be beneficial in assessing desiccation injury. Desiccation has been shown to decrease root respiration of Douglas-fir seedlings (McCreary and Zaerr 1987).

Roots must efficiently absorb and transport water from soil to meet shoot transpirational demands. However, if water loss due to drying atmospheric conditions is greater than root water uptake, an upset in water balance could result in seedling desiccation (Sands 1984; Burdett 1990; Girard et al. 1997; Grossnickle 2000). In addition, a delay in outplanting may aggravate pre-planting desiccation stress, thereby potentially affecting seedling performance following transplanting. Therefore, protecting the root system from excessive moisture loss may help ensure survival and seedling establishment success. Transplanting stress may be aggravated when seedlings are exposed to drying conditions, as may occur during lifting, cold storage, transport to and storage on the planting site, and outplanting.

Although the use of hydrogels shows promise for conifer reforestation (Sloan 2004), limited information is available about use of hydrogels in hardwood afforestation plantings in the Central Hardwood Forest Region of the USA. Additionally, little is known about the mechanisms by which hydrogel reduces the injury that occurs when seedlings of dormant, temperate deciduous forest tree species, such as northern red oak (*Quercus rubra* L.), are exposed to drying conditions prior to planting. Hydrogels are able to protect and store many times their own weight of water (Sloan 2004), and therefore, may be effective in reducing seedling water stress, particularly in coarse-textured soils (Orzolek 1993). Specht and Harvey-Jones (2000) observed an increase in plant water uptake, stomata activity, and plant mass in northern red oak when hydrogels were incorporated into the media. On the other hand, Heiskanen (1995) reported that hydrogels provided no benefit in seedling establishment under field conditions while negative effects were reported in other studies (Wang 1989; Tripepi et al. 1991). If hydrogels were shown to be effective in alleviating

moisture loss, they could be used in planting programs to improve establishment success of temperate deciduous forest tree species. Exploring the mechanisms by which hydrogels reduce injury that occurs when roots are exposed to pre-planting desiccation, and after being subjected to drought stress following transplanting, will be useful toward understanding how this technique effectively alleviates seedling transplant stress.

The data presented herein is from four experiments investigating the effects of hydrogel on pre-transplanting desiccation stress and post-transplanting drought effects. In the first two experiments, we examined the influence of root hydrogel dipping on tissue moisture content, root respiration, tissue electrolyte leakage, and stem water potential of northern red oak seedlings subjected to a period of drying conditions prior to transplanting. In the second two experiments, we examined the effects of hydrogel amendment on substrate water content and growth and physiology of northern red oak seedlings subjected to post-planting drought stress for 45 days. We tested the hypothesis that hydrogels reduce the injury that occurs when roots are subjected to desiccation by delaying water loss and maintaining membrane function. Northern red oak was used due to its relative sensitivity to drought stress (Weber and Gates 1990) and its importance and increased use in planting programs in the Central Hardwood Forest Region, USA (Jacobs et al. 2004).

Materials and methods

Plant materials

One-year-old bareroot northern red oak (*Quercus rubra* L.) seedlings of bulk seed origin from a southern Indiana seed source were grown using standard nursery practices for production of hardwood seedlings in this region (Jacobs 2003) at Vallonia State Nursery, Indiana DNR Division of Forestry (38°48′N, 86°06′W) near Vallonia, IN, USA. In fall 2005, seedlings were lifted from nursery beds, bundled with moistened sphagnum moss, placed in kraft-polyethylene bags, and cooler stored at 2°C. In June 2006, seedlings were transported to Purdue University in West Lafayette, IN, USA (40°25′N, 86°55′W).

Seedlings were washed free of soil and measured for height, root-collar diameter (rcd), and root volume by water displacement (Burdett 1979). The seedlings, with mean (± SE) height of 64.9±1.7 cm, rcd of 8.31± 0.2 mm, and root volume of 32.12±2.4 cm^3, were numbered, tagged, and placed in cooler storage at 2°C until desiccation treatments began.

Experiment 1. Pre-transplanting desiccation stress: effects of hydrogel on stem water potential, tissue moisture content, and root electrolyte leakage

Following cooler storage, 20 seedlings were randomly divided into two equal treatment groups: control (no root dip) and hydrogel (roots dipped in hydrogel). The experiment was a completely randomized design. Root dip (0.4%) material was prepared by mixing Terra-sorb® Fine Hydrogel (Plants Health Care, Inc., Pittsburgh, PA, USA) in deionized H_2O as recommended by the manufacturer. The root systems were dipped in the hydrogel slurry for at least 5 min to ensure complete coverage. Roots of control seedlings were wrapped in moist paper towels. When all seedlings were prepared, paper towels were removed as the control and hydrogel seedlings were placed on a mesh screen (to attain uniform air distribution) in a controlled environment chamber. The chamber provided a constant temperature of 20°C, relative humidity of 65%, and fluorescent lamps and incandescent bulbs yielded a photosynthetic photon flux density (PPFD) of 300 μmol m^{-2} s^{-1} at the mesh level. Seedlings were desiccated for 1 h, and then measured (described in detail in the measurement section) for stem water potential, tissue moisture content, and root electrolyte leakage.

Experiment 2. Pre-transplanting desiccation stress: effects of hydrogel on root electrolyte leakage, root respiration, and tissue moisture content at three desiccation periods

Following cooler storage, 54 seedlings were randomly divided into three treatment groups: control (no root dip), hydrogel (roots dipped in hydrogel), and rinsed (roots rinsed free of hydrogel in deionized H_2O following desiccation). Rinsing after the desiccation period was done to examine the contribution of hydrogel to root membrane injury.

Root systems were prepared and seedlings were desiccated as described above, but at three time intervals (1, 3, and 5 h). Following each desiccation

duration, roots in the rinsed treatment were washed free of hydrogel using deionized H_2O. The experiment was a completely randomized design with three levels of hydrogel (control, hydrogel, and rinsed), three exposure times, and six seedlings per hydrogel—exposure time combination. After desiccation, seedlings were measured (described in detail in the measurement section) for tissue moisture content and tap and lateral root electrolyte leakage.

Experiment 3. Effect of hydrogel amendment on substrate water content

Containers, 12-l Treepots™ (Stuewe and Sons, Corvallis, OR, USA), were assigned to two treatments: control (medium not amended) and hydrogel (medium amended with hydrogel). The medium was Pro-Mix 'BX' (Sphagnum peat; Premier Horticulture Inc., Quakertown, PA, USA). For the hydrogel treatment, dry hydrogel was added to the medium to achieve a final concentration of 0.2% (*w*:*w*). The amount of hydrogel that was added into the substrate was calculated based on the initial mass of each container. Six containers of each treatment (12 containers total) were arranged on a greenhouse bench following a completely randomized design. Greenhouse conditions are described in Experiment 4. Containers were watered to field capacity and substrate water content was measured by weight loss at 7, 14, 21, 28, and 35 days after potting.

Experiment 4. Post-planting desiccation stress: combined effects of watering regime and hydrogel

Following cooler storage, 48 seedlings were randomly divided into two treatment groups: control (no root dip) and hydrogel (roots dipped in hydrogel). Root systems were prepared as described above. Immediately after treatment, seedlings were transplanted into 12-l Treepots™ filled with Pro-Mix 'BX' and each container was irrigated to field capacity determined gravimetrically as described by Timmer and Armstrong (1989). Seedlings of each treatment group were further randomly divided into two soil moisture regimes (50% and 100% container capacity). The experiment was a completely randomized design with two levels of hydrogel (control and hydrogel), two soil moisture regimes, and 12 seedlings per hydrogel—soil moisture combination.

Seedlings were randomly distributed on a greenhouse bench in the Department of Horticulture and Landscape Architecture Plant Growth Facility (40°25′N, 86°55′W) at Purdue University. The greenhouse was set for 24/20°C day/night air temperatures, relative humidity of 60% to 70%, and 16 h photoperiod with PPFD of 350 to 400 μmol m^{-2} s^{-1} measured at seedling top height. To maintain the soil water content close to the target value per moisture treatment, containers were weighed daily at midday and watered as required to maintain the appropriate container capacity (Royo et al. 2001). During one irrigation each week, seedlings were fertilized with 15N-5P_2O_5-15K_2O water soluble fertilizer (Miracle-Gro® Excel® Cal-Mag; The Scotts Co., Marysville, OH, USA). The nutrient solution contained (in mg l^{-1}) 150 N, 22 P, 125 K, 50 Ca, 20 Mg, and micronutrients. Seedlings were re-arranged once per week to avoid potential environmental variation within the greenhouse bench.

After 45 days, seedlings were measured for shoot length, root volume using water displacement (Burdett 1979), days to bud break, and gas exchange (described in detail in the measurement section).

Measurements

Tissue (shoots and roots) moisture content was calculated as the difference between tissue fresh mass (FM) and dry mass (DM) after oven-drying at 70°C for 72 h.

Stem water potential (Ψ_w) was measured in excised stems with a Scholander pressure chamber (PMS Instruments, Corvallis, OR, USA).

The electrolyte leakage test was conducted as described by Apostol and Zwiazek (2003). Briefly, tap (3-cm segment excised from middle part of the root system) and lateral (six sections with 1-cm length and ≥2 mm diameter) root samples were placed in separate vials containing 15 ml of deionized H_2O. [Note: all control roots were washed three times with deionized H_2O for 5 min each time.] After 6-h incubation on an orbital shaker, electrical conductivities of the solutions were measured with an electrical conductivity meter (HI 8033, Hanna Instruments Inc., Woonsocket, RI, USA). Total electrolytes were obtained by autoclaving the samples at 120°C for 20 min. The total electrolyte content of the solutions was measured and electrolyte leakage was expressed as a percentage of the total electrolytes.

Root respiration was determined as follows: root samples (0.5–0.8 g) were allowed to equilibrate in the dark at 27°C for 15 min, blotted dry with paper towels, and placed inside the cuvette of a LI-6400 infrared gas analyzer equipped with a CO_2 mixer control unit (LI-COR, Lincoln, NE, USA). Measurements were made at the same temperature, reference CO_2 concentration of 400 μmol m^{-2} s^{-1}, and flow rate at 500 μmol s^{-1}. Following enclosure in the cuvette, data was logged when root samples reached a steady-state value. Root respiration (measured of CO_2 efflux) was expressed as μmol of CO_2 released per g^{-1} root FM s^{-1}.

Days to budbreak was defined as the interval between planting and when the terminal bud scales parted to expose new (green) foliage.

Gas exchange measurements (net photosynthesis, A, and stomatal conductance, g_s) were conducted at midday using a LI-6400 portable infrared gas analyzer equipped with a red LED light source (LI6400-02) and a CO_2 mixer control unit (LI-COR, Lincoln, NE, USA). Measurements were taken on the second leaf basipetal from the top of the first flush. All measurements were made at PPFD of 350 μmol m^{-2} s^{-1}, reference CO_2 concentration of 400 μmol m^{-2} s^{-1}, leaf temperature of 28°C, RH of 55–60%, and flow rate at 500 μmol s^{-1}. Following enclosure in the leaf cuvette, data was logged when a leaf reached a steady-state value (coefficient of variations of CO_2 and H_2O within the chamber was <0.25%) as described by Apostol et al. (2007). Leaf area was determined with an LI-6200 leaf area meter (LI-COR, Lincoln, NE, USA) and tissue dry mass was obtained after oven-drying at 70°C for 72 h.

Statistical analysis

Tests for normality and constant variance, to ensure validity of the assumptions of analysis of variance (ANOVA), indicated no transformations were necessary. Because the results of an earlier ANCOVA analysis (SAS 9.1 Institute Inc., Cary, NC) showed that effects of initial shoot height and root volume (covariates) were not significant for measured variables, it was appropriate to use a general linear model (SAS Institute Inc.) for data analysis.

For Experiment 1 (n=10) Student's t-test was used to determine significant differences between the control and hydrogel treatments (α=0.05) on stem water potential (Ψ_w), tissue moisture content (MC), and tap and lateral root electrolyte leakage (EL).

For Experiment 2 (n=6), ANOVA was used to determine treatment effects by two-way interactions between hydrogel treatment and exposure time on tissue MC, EL, and root respiration.

For Experiment 3 (n=6), ANOVA was used to determine treatment effects by two-way interactions between hydrogel and desiccation period on substrate water content.

For Experiment 4 (n=12), ANOVA was used to determine treatment effects by two-way interactions between root dip and soil moisture regime on days to bud break, shoot length, root volume, tissue dry mass (DM), total leaf area, net photosynthesis (A), and stomatal conductance (g_s).

Results

Experiments 1 and 2: pre-transplanting desiccation stress

Hydrogel did not significantly affect lateral or tap root EL (Fig. 1). Although mean lateral root MC was significantly (P=0.0002) increased in hydrogel-treated seedlings, tap root MC was not (Fig. 1). Control seedlings had an 80% reduction in lateral root MC compared with seedlings treated with hydrogel. Control seedlings had significantly (P=0.0295) lower Ψ_w than hydrogel-treated seedlings (Fig. 2).

Root MC in control seedlings was significantly (P=0.0001) lower than in hydrogel-treated seedlings (Fig. 3a). For root MC, the hydrogel × exposure time interaction was significant, but exposure time was not (Table 1). At 1 h, hydrogel-treated seedlings had 80% higher root MC than control seedlings, but after 5 h, root MC of hydrogel-treated seedlings had decreased and was only 40% higher than the control seedlings. A similar pattern was observed in stem MC with seedlings treated with hydrogel showing higher values than control seedlings (Fig. 3b; Table 1).

Hydrogel treatment (P=0.0410) and its interaction with time (P=0.0127) significantly affected root EL (Table 1). Root EL values were similar in both control and hydrogel treatments at 1 and 3 h after desiccation (Fig. 4). However, at the end of the exposure time (5 h), EL values of hydrogel-treated seedlings were significantly (31%) lower than the control seedlings, which were similar to seedlings in the rinsed treatments.

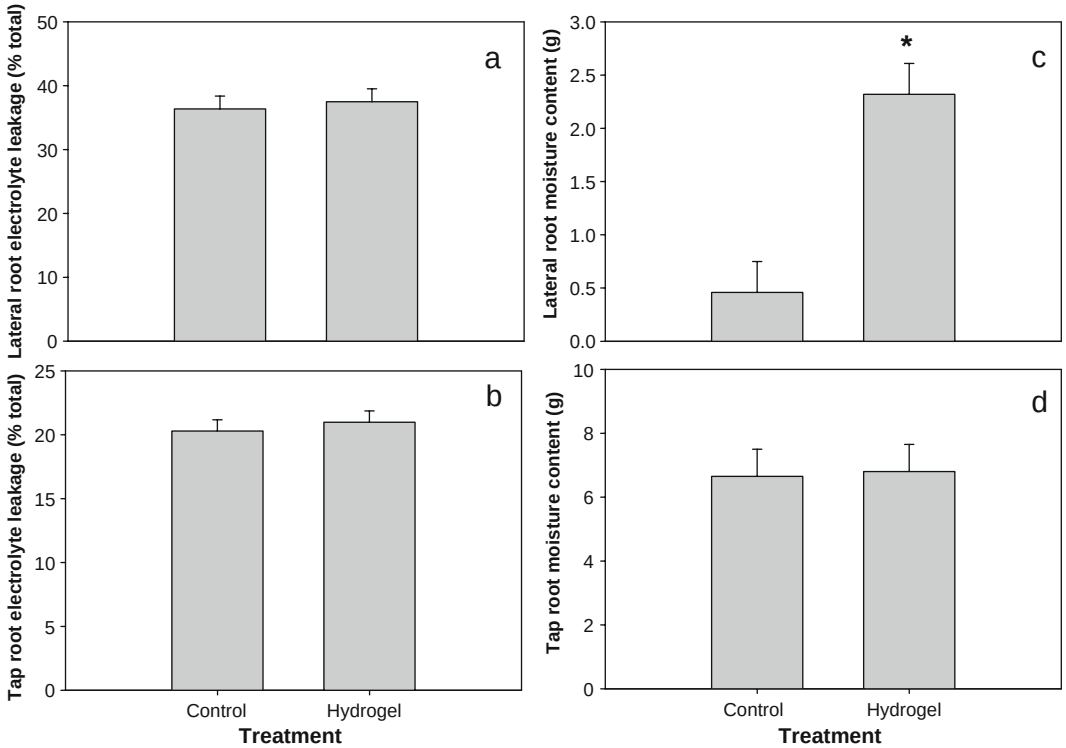

Fig. 1 Root moisture content and electrolyte leakage of control (no root dip) and hydrogel-treated (roots dipped in hydrogel) northern red oak seedlings desiccated for 1 h in a controlled growth chamber. Least square means ± SE (n=10) are shown. *Bars* marked with an *asterisk* indicate a significant difference from the control at α=0.05

Neither the main effects (treatment and time) nor the treatment × time interaction altered root respiration (data not shown). Mean values of root respiration rates in control and hydrogel-treated seedlings remained constant during the desiccation period. Mean root respiration rate in control seedlings was 0.025 μmol CO_2 released g^{-1} root FM s^{-1} compared with 0.015 μmol CO_2 released g^{-1} root FM s^{-1} in hydrogel-treated seedlings.

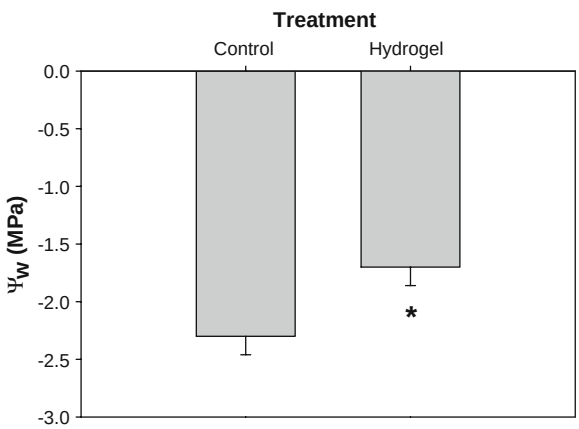

Fig. 2 Stem water potential (Ψ_w) of control (no root dip) and hydrogel-treated (roots dipped in hydrogel) northern red oak seedlings desiccated for 1 h in a controlled growth chamber. Least square means ± SE (n=10) are shown. *Bars* marked with an *asterisk* indicate a significant difference from the control at α=0.05

Experiment 3. Effect of hydrogel amendment on substrate water content

Both desiccation period (P=0.0001) and hydrogel (P=0.0001) and their interactions (P=0.0108) significantly affected substrate water content. Hydrogel addition to the substrate showed higher water retention compared with the control substrate (Fig. 5). Evaporative water loss (difference between day 35 and day 7) in control was 712 g of water compared with 585 g of water measured in medium amended with hydrogel.

Experiment 4: post-planting desiccation stress

Soil moisture regime (P=0.0075) and hydrogel (P=0.0036) had significant effects on days to budbreak (Fig. 6). For days to budbreak, the moisture regime ×

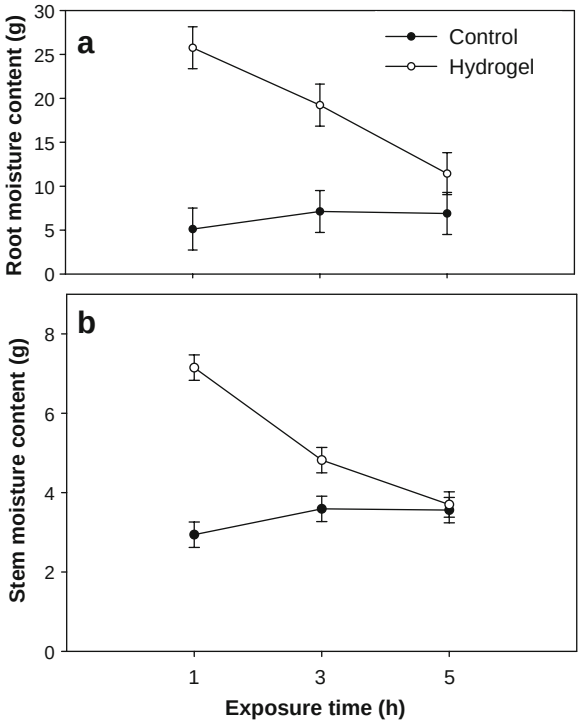

Fig. 3 Root moisture content (**a**) and stem moisture content of control (no root dip) and hydrogel (roots dipped in hydrogel), northern red oak seedlings desiccated at three time intervals (1, 3, and 5 h). Each data point represents least square means ± SE (n=6)

hydrogel interaction was significant (P=0.0041; Table 2). Among the measured growth parameters, shoot length (P=0.0025), leaf area (P=0.0001), and shoot DM (P=0.0055) were significantly reduced in water-stressed seedlings compared with the control seedlings (Fig. 6) while root volume (P=0.0010) was significantly increased in response to water stress after 45 days of treatments. Total leaf area (P=0.0142), shoot DM (P=0.0311), and root volume (P=0.0174) were significantly higher in seedlings treated with hydrogel than for control seedlings.

Both A (P=0.0010) and g_s (P=0.0001) were significantly reduced in water-stressed plants compared with well-watered plants (Fig. 7). However, hydrogel and its interaction with moisture regime did not significantly affect A or g_s (Table 2).

Discussion

Nearly all forest tree seedlings planted in the Central Hardwood Forest Region of the USA originate from bareroot nurseries (Jacobs 2003; Dey et al. 2008). Although root coatings and *Sphagnum* moss may limit root desiccation of nursery seedlings, our earlier findings (unpublished data) showed that the hydrogel effectively coated the root systems and retained moisture longer throughout the drying time than did *Sphagnum* moss. Compared to container seedlings where roots are enclosed within media, unprotected roots in bareroot seedlings are highly prone to desiccation (McKay 1996; Sarvaš 2003). The process of removing seedlings from their protective bundles at planting sites increases risk of root desiccation (McKay 1996). The observed initial high level of moisture content in roots of hydrogel-treated seedlings compared with control seedlings (Fig. 3a) suggests that water bound in the hydrogel protects the roots from moisture loss. It appears that hydrogel acts as a reservoir of moisture, gradually releasing loosely bound water into the roots as desiccation occurs. Similar to our results, initial moisture content of roots was higher than that of stems (McKay et al. 1999) and the largest change in MC during exposure occurred in fine roots (Coutts 1981).

Table 1 Results of ANOVA (P-values) testing for the main treatment effects (hydrogel and exposure time) and interaction effects (hydrogel × exposure time) on root moisture content, stem moisture content, root electrolyte leakage, root respiration, and substrate water content

Source	Measured variable				
	RMC	SMC	REL	RR	SWC
Hydrogel (H)	0.0001	0.0152	0.0410	0.5271	0.001
Exposure time (ET)	0.1463	0.6599	0.2231	0.0951	0.001
H × ET	0.0118	0.0440	0.0127	0.3554	0.011

RMC Root moisture content, *SMC* stem moisture content, *REL* root electrolyte leakage, *RR* root respiration, *SWC* substrate water content

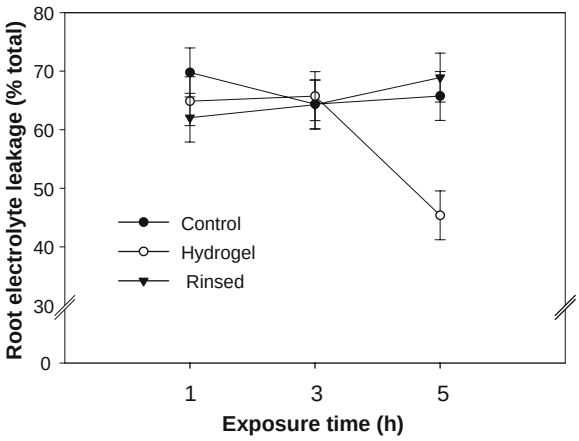

Fig. 4 Root electrolyte leakage of control (no root dip), hydrogel (roots dipped in hydrogel), and rinsed (roots rinsed free of hydrogel in deionized H_2O after a period of desiccation) northern red oak seedlings desiccated at three time intervals (1, 3, and 5 h). Each data point represents least square means ± SE (n=6)

Seedling moisture status has been identified as a factor affecting survival of newly-planted seedlings (McKay and White 1997). We found that desiccation in control seedlings resulted in drying of lateral roots, causing a decline in root MC over time (Fig. 3a) that, in turn, could affect root growth and root water uptake. It has been reported that reduction in root water uptake is related to restrictions in new root growth (Wan et al. 1999). Losses in lateral root MC were also consistent with the low (more negative) Ψ_w in control seedlings and this was alleviated by dipping of roots in the hydrogel slurry (Figs. 1 and 2a), which may have distributed moisture across the roots resulting in stem hydration (Fig. 2b).

Although desiccation tolerance involves many different facets, cell membrane stability is a basic requirement for the maintenance of physiological functions in plants (Bewley 1979). Symeonidou and Buckley (1999) observed an increase in root EL of desiccated seedlings that had a higher proportion of fine and lateral roots. EL increased with duration of desiccation (McKay and White 1997) indicating a loss of membrane integrity and more ion leakage (Crowe et al. 1987) that consequently leads to failure in root functions (Apostol and Zwiazek 2003; Huang et al. 2005) and root death (McKay and White 1997). Concern about potential phytotoxic properties of the hydrogel on northern red oak root functions was tested by comparing root EL between control and hydrogel-treated seedlings. At 5 h, hydrogel showed significantly lower EL values (Fig. 4) than rinsed seedlings (no surface ions), suggesting that the chemical composition of hydrogel was not a factor contributing to root membrane injury. Because Terra-Sorb® is a polyacrylamide cation absorbing polymer (Martin et al. 1993), it is possible that it could alter ion uptake resulting in nutrient imbalance in plants. This is worthy of further study.

The loss of fine roots (lateral roots in the case of our experiment) is related to poor seedling performance (Symeonidou and Buckley 1999). In our study, we found that hydrogel significantly reduced root EL of seedlings exposed to 21°C for 5 h (Fig. 4), but no statistical evidence of differences was observed in control and hydrogel-treated seedlings exposed to similar conditions for 1 h (Fig. 3). We have found that the exposure period for Experiment 1 was not sufficient to detect injury, suggesting that the hydrogel may have alleviated injury when seedlings were severely desiccated. Sloan (2004) showed that root dips did not increase survival of non-stressed and properly handled planting stock and of seedlings planted shortly after lifting. We propose that the ability of measured parameters (i.e., root EL) to detect mechanisms of seedling injury depends on the nature and duration of the stress before planting and the quality of planting stock (McKay 1996; Siemens and Zwiazek 2003; Brønnum 2005).

Root respiration rate has been shown to be correlated with carbohydrate content (Williams and

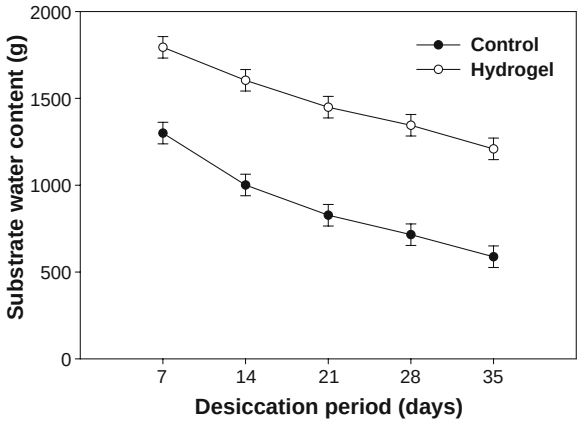

Fig. 5 Substrate water content (g) with and without hydrogel (control) amendment at five, 7-day intervals. Each data point represents least square means ± SE (n=6)

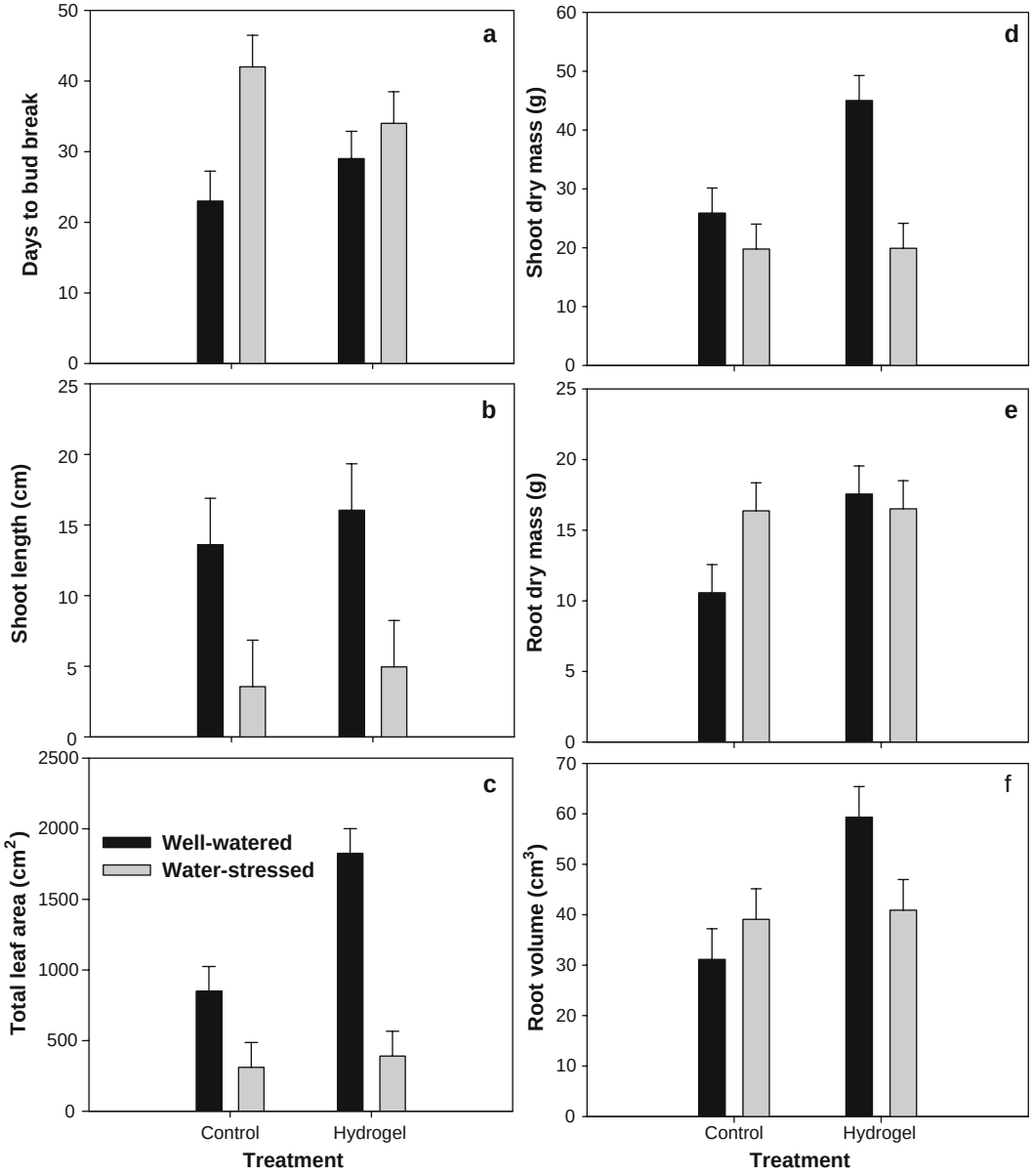

Fig. 6 Effects of hydrogel on days to budbreak and growth of northern red oak seedlings subjected to drought stress 45 days after the initiation of treatments. Each data point represents least square means ± SE (n=12)

Farar 1990). Several researchers working with woody plants have suggested that root death is related to carbohydrate availability (Marshall 1986; Kosola and Eissenstat 1994). Roots exposed to drying conditions have shown reduced root respiration rates (Bryla et al. 1997). In our study, however, desiccation did not significantly alter root respiration, which suggests that desiccation had no apparent effect on the short-term carbohydrate status during the conditions we described. The lack of significant difference between control and hydrogel-treated seedlings may be partly explained by the large proportion of stored carbohydrates in the roots of northern red oak seedlings during dormancy (Farmer 1975). Although there are indications that hydrogel-treated seedlings showed slightly lower root respiration rates than those in controls, the exact role of hydrogel in maintaining root metabolic activity remains unclear.

Table 2 Results of ANOVA (*P*-values) testing for the main treatment effects (hydrogel and soil moisture regime) and interaction effects (hydrogel × soil moisture regime) on days to budbreak, shoot length, total leaf area, shoot dry mass, root dry mass, root volume, net photosynthesis, and stomatal conductance

Source	Measured variable							
	DBB	SL	TLA	SDM	RDM	RV	*A*	g_s
Hydrogel (H)	0.0036	0.8745	0.0142	0.0311	0.0414	0.0174	0.1317	0.1060
Soil moisture (SM)	0.0075	0.0025	0.0001	0.0055	0.2417	0.3918	0.0010	0.0001
H × SM	0.0041	0.5634	0.0044	0.0460	0.0940	0.0355	0.8313	0.7180

DBB Days to budbreak, *SL* shoot length, *TLA* total leaf area, *SDM* shoot dry mass, *RDM* root dry mass, *RV* root volume, *A* net photosynthesis, g_s stomatal conductance

Similar to other studies (Hüttermann et al. 1994; Arbona et al. 2005), we found that substrate amended with hydrogel retained more moisture than those with substrate alone (Fig. 5). Hydrogel amendments may improve seedling growth and establishment by increasing substrate water retention capacity and regulating water supply available for plants (Arbona et al. 2005), which may be of value to seedlings particularly during dry periods and in soils with low water holding capacity.

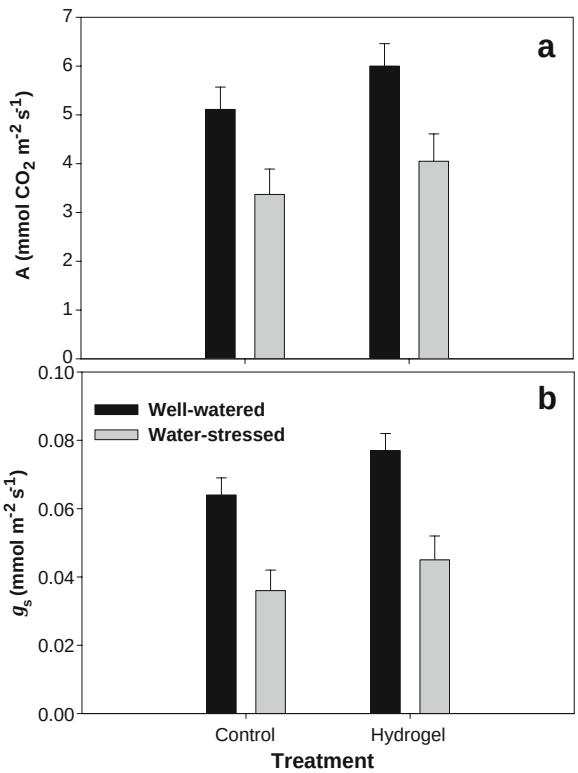

Fig. 7 Effects of hydrogel on net photosynthesis (*A*) and stomatal conductance (g_s) of northern red oak seedlings subjected to drought stress 45 days after the initiation of treatments. Each data point represents least square means ± SE (n=12)

In the present study, no beneficial effects of hydrogel on growth and gas exchange in drought-stressed seedlings were found, which was contrary to the studies reported for citrus plants where hydrogel contributed to improved seedling growth, survival, and photosynthesis (Arbona et al. 2005). It is plausible that hydrogel may have alleviated the drought effects before measurements were made and this was partly explained by the ameliorating effects of hydrogel on tissue moisture content and membrane leakiness on root desiccation (Experiments 1 and 2) and on the number of days to budbreak in seedlings exposed to post-transplanting drought (Experiments 3 and 4). Days to budbreak appeared to be the most sensitive and early indicator of desiccation at the time of transplanting and is reported to be an early measure of physiological activity in temperate deciduous forest tree species (Englert et al. 1993; Jacobs et al. 2008). Also, we propose that hydrogel may have helped seedlings survive through a short-term drought. Once seedlings establish in substrate, new roots, which are the most efficient and functional roots available for water transport, emerge and facilitate water uptake. It is important to note, however, that hydrogel may be unable to protect new roots that elongate beyond the protective covering. Seedling responses to treatments could also be affected by factors such as seedling age, drought intensity, treatment duration, and concentrations of hydrogel and substrate used.

In conclusion, our short-term, pre-transplanting desiccation study showed that hydrogel alleviated seedling injury by reducing root membrane leakiness 5 h after the desiccation exposure and alleviating water loss compared with control seedlings. The contribution of delayed tissue moisture loss and

maintenance of cell membrane integrity in hydrogel-dipped roots exposed to drying conditions prior to transplanting may prove advantageous for seedling establishment success. Although we observed no evidence of differential effects of hydrogel-treated roots exposed to drought stress compared with control seedlings on early seedling growth and gas exchange 45 days after treatment, the ameliorative effects of hydrogel on days to budbreak may have helped seedlings to survive short-term drought. To better understand the importance of hydrogel in alleviating stress, future research should address the mechanical and physiological effects of hydrogel over time immediately following treatment exposure. While root dipping increases reforestation costs, this practice may be justified when planting into soils with low water holding capacity and during periods of high vapor pressure deficit. Further testing is required, however, to help confirm the utility of hydrogel in the field.

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References

Alm A, Stanton J (1993) Polymer root dip increases survival of stressed bareroot seedlings. Northern J Appl For 10:90–92

Apostol KG, Zwiazek JJ (2003) Hypoxia affects root sodium and chloride concentrations and alters water conductance in salt-treated jack pine (*Pinus banksiana*) seedlings. Trees (Berl) 17:251–257

Apostol KG, Jacobs DF, Wilson BC, Salifu KF, Dumroese RK (2007) Growth, gas exchange, and root respiration of *Quercus rubra* seedlings exposed to low root zone temperatures. For Ecol Manag 253:89–96

Arbona V, Iglesia DJ, Jacas J, Primo-Millo E, Talon M, Gómez-Cadenas A (2005) Hydrogel substrate amendment alleviates drought effects on young citrus plants. Plant Soil 270:73–82, doi:10.1007/s11104-004-1160-0

Balneaves JM, Menzies MI (1988) Lifting and handling procedures at Edendale Nursery—effects on survival and growth of 1/0 *Pinus radiata* seedlings. N Z J Sci 18:132–134

Bewley JD (1979) Physiological aspects of desiccation tolerance. Annu Rev Plant Physiol 130:195–238, doi:10.1146/annurev.pp.30.060179.001211

Brakke M, Allen LH (1995) Gas exchange of citrus seedlings at different temperatures, vapor-pressure deficits, and soil-water contents. J Am Soc Hortic Sci 120:497–504

Brønnum P (2005) Preplanting indicators of survival and growth of desiccated *Abies procera* bareroot planting stock. Scand J For Res 20:36–46, doi:10.1080/02827580510008257

Bryla D, Bouma TJ, Eissenstat DM (1997) Root respiration in citrus acclimates to temperature and slows during drought. Plant Cell Environ 20:1411–1420, doi:10.1046/j.1365-3040.1997.d01-36.x

Burdett AN (1979) A nondestructive method for measuring the volume of intact plant parts. Can J For Res 9:120–122

Burdett AN (1990) Physiological processes in plantation establishment and the development of specifications for forest planting stock. Can J Res 20:415–427, doi:10.1139/x90-059

Coutts MP (1981) Effects of root or shoot exposure before planting on the water relations, growth, and survival of Sitka spruce. Can J For Res 11:703–709, doi:10.1139/x81-097

Crowe JH, Crowe LM, Carpenter JF, Winstrom AC (1987) Stabilization of dry phospholipid bilayers and proteins by sugars. Biochem J 242:1–10

Davis AS, Jacobs DF (2005) Quantifying root system quality of nursery seedlings and relationship to outplanting performance. New For 30:295–311, doi:10.1007/s11056-005-7480-y

Dey DC, Jacobs DF, McNabb K, Miller G, Baldwin V, Foster G (2008) Artificial regeneration of major oak (*Quercus*) species in the eastern United States—a review of the literature. For Sci 54:77–106

Dierauf TA, Garner JW (1975) A test of Agricol as a root dip 1975, occasional report 47. Virginia Division of Forestry, Glen Allen, VA, 5 pp

Echols RJ, Meier CE, Ezell AW, McKinley CR (1990) Dry site survival of bareroot and container seedlings of southern pines from different genetic sources given root dip and ectomycorrhizal treatments. Tree Plant Notes 41:13–21

Edgren JM (1984) Nursery storage to planting hole: a seedling's hazardous journey. In: Duryea ML, Landis TD (eds) Forest nursery manual: production of bareroot seedlings. Forest Research Laboratory, Oregon State University, Corvallis, OR, pp 235–242, Martinus Nijhoff/Dr. W. Junk

Englert JM, Fuchigami LH, Chen THH (1993) Effects of storage temperatures and duration on the performance of bare-root deciduous hardwood trees. J Arboric 19:106–112

Farmer REJ (1975) Dormancy and root regeneration of northern red oak. Can J For Res 5:176–185

Feret PF, Kreh RE, Mulligan C (1985) Effects of air drying on survival, height, and root growth potential of loblolly pine seedlings. S J Appl For 9:125–128

Girard S, Clément A, Boulet-Gercout B, Guehl JM (1997) Effects of exposure to air on planting stress in red oak seedlings. Ann Sci For 54:395–401, doi:10.1051/forest:19970407

Gómez-Cadenas A, Tadeo DR, Talon M, Primo-Millo E (1996) Leaf abscissions induced by ethylene in water stressed intact seedlings of *Cleopatra mandarin* requires previous abscissic acid accumulation in roots. Plant Physiol 112:401–408

Grossnickle SC (1988) Planting stress in newly planted jack pine and white spruce. 1. Factors influencing water uptake. Tree Physiol 4:71–83

Grossnickle SC (2000) Ecophysiology of northern spruce species: the performance of planted seedlings. NRC Research, Ottawa, Ontario, Canada

Haase DL, Rose R (1993) Soil moisture stress induces transplant shock in stored and unstored 2+0 Douglas-fir seedlings of varying root volume. For Sci 39:275–294

Heiskanen J (1995) Water status of sphagnum peat and a peat–perlite mixture in containers subjected to irrigation regimes. HortSci 30:281–284

Hermann RK (1967) Seasonal variation in the sensitivity of Douglas-fir seedlings to exposure of roots. For Sci 13:140–149

Huang X, Lakson AN, Eissenstat DM (2005) Interactive effects of soil temperature and moisture on Concord grape root respiration. J Exp Bot 56:2651–2660, doi:10.1093/jxb/eri258

Hüttermann A, Zommordi M, Reise K (1999) Addition of hydrogels to soil for prolonging the survival of *Pinus halepensis* seedlings subjected to drought. Soil Tillage Res 50:295–304, doi:10.1016/S0167-1987(99)00023-9

Insley H, Buckley GP (1985) The influence of desiccation and root pruning on the survival and growth of broadleaved seedlings. J Hortic Sci 60:377–387

Jacobs DF (2003) Nursery production of hardwood seedlings. Purdue University Cooperative Extension Service, West Lafayette, IN, FNR-212, 8 pp

Jacobs DF, Ross-Davis AL, Davis AS (2004) Establishment success of conservation tree plantations in relation to silvicultural practices in Indiana, USA. New For 28:23–26, doi:10.1023/B:NEFO.0000031329.70631.d0

Jacobs DF, Wilson BC, Ross-Davis AL, Davis AS (2008) Cold hardiness and transplant response of *Juglans nigra* seedlings subjected to alternative storage regimes. Ann For Sci 65:606

Kosola KR, Eissenstat DM (1994) The fate of surface roots of citrus seedlings in dry soil. J Exp Bot 45:1639–1645, doi:10.1093/jxb/45.11.1639

Magussen S (1985) Effects of root-coating with polymer WaterLock on survival and growth of drought-stressed bareroot seedlings of white spruce. USDA Tree Plant Notes 37:15–19

Marshall JD (1986) Drought and shade interact to cause fine-root mortality in Douglas-fir seedlings. Plant Soil 91:51–60, doi:10.1007/BF02181818

Martin CA, Ruter JM, Roberson RW, Sharp WP (1993) Element absorption and hydration potential of polyacrylamide gels. Commun Soil Sci Plant Anal 24:539–548

McCreary DD, Zaerr JB (1987) Root respiration has limited value for assessing Douglas fir seedling quality. Can J For Res 17:1144–1147, doi:10.1139/x87-175

McKay HM (1992) Electrolyte leakage from fine roots of cold conifer seedling: a rapid index of plant vitality following cold storage. Can J For Res 22:1371–1377

McKay HM (1996) A review of the effect of stresses between lifting and planting on nursery stock quality and performance. New For 13:363–393

McKay HM, White IMS (1997) Fine root electrolyte leakage and moisture content: indices of Sitka spruce and Douglas-fir seedling performance after desiccation. New For 13:139–162, doi:10.1023/A:1006571805960

McKay HM, Jinks RL, McEvoy C (1999) The effect of desiccation and rough-handling on the survival and early growth of ash, beech, birch and oak seedlings. Ann For Sci 56:391–402, doi:10.1051/forest:19990504

Mullin RE (1978) Root exposure, root dipping, and extended spring planting of white pine seedlings. For Chron 54:84–87

Mullin RE, Bunting WR (1979) Another look at clay dipping of bare-root nursery stock. For Chron 55:183–188

Orzolek MD (1993) Use of hydrophilic polymers in horticulture. HortTech 3:41–44

Royo A, Gil L, Pardos JL (2001) Effect of water conditioning on morphology, physiology and field performance of *Pinus halepensis* Mill. seedlings. New For 21:127–140, doi:10.1023/A:1011892732084

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

Sarvaš M (2003) Effect of desiccation on the root system of Norway spruce (*Picea abies* [L.] Karst.) seedling and a possibility of using hydrogel STOCKOSORB® for its protection. J For Sci 11:531–536

Siemens JA, Zwiazek JJ (2003) Effects of water deficit stress and recovery on the root water relations of trembling aspen (*Populus tremuloides*) seedlings. Plant Sci 165:113–120, doi:10.1016/S0168-9452(03)00149-3

Sloan JP (2004) The use of rootdips on North American conifer seedlings: a review of literature. Tree Plant Note 45:26–31

Specht A, Harvey-Jones J (2000) Improving water delivery to the roots of recently transplanted seedlings trees: the use of hydrogels to reduce leaf loss and hasten root establishment. For Res 1:117–123

Sucoff E, Buschena C, Tamte P (1985) Desiccation and water potentials in the roots, leaves, and shoots of bare-root red pine and white spruce. Can J For Res 15:989–992, doi:10.1139/x85-157

Symeonidou MV, Buckley GP (1999) The effect of pre-planting desiccation stress and root pruning on the physiological condition and subsequent field performance of one year old *Prunus avium* and *P. cerasifera* seedlings. J Hortic Sci Biotechnol 74:386–394

Tabbush PM (1987) Effect of desiccation on water status and forest performance of bare-rooted Sitka spruce and Douglas-fir transplants. Forestry 60:31–43, doi:10.1093/forestry/60.1.31

Timmer VR, Armstrong G (1989) Growth and nutrition of containerized *Pinus resinosa* seedlings at varying moisture regimes. New For 3:171–180, doi:10.1007/BF00021580

Tripepi RR, George MW, Dumroese RK, Wenny DL (1991) Birch seedling response to irrigation frequency and a hydrophilic polymer amendment in a container medium. J Environ Hortic 9:119–123

Wan X, Landhäusser SM, Zwiazek JJ, Lieffers V (1999) Root water flow and growth of aspen (*Populus tremuloides*) at low root temperatures. Tree Physiol 19:879–884

Wang YT (1989) Medium and hydrogel affect production and wilting of tropical ornamental plants. HortSci 24:941–944

Weber JA, Gates DM (1990) Gas exchange in *Quercus rubra* (northern red oak) during a drought: analysis of relations among photosynthesis, transpiration, and leaf conductance. Tree Physiol 7:215–225

Williams JHH, Farar JF (1990) Control of barley root respiration. Physiol Plant 79:259–266, doi:10.1111/j.1399-3054.1990.tb06740.x