High Root-zone Temperatures, Mycorrhizal Fungi, Water Relations, and Root Hydraulic Conductivity of Container-grown Woody Plants

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Abstract. High root-zone temperatures can stress container-grown plants and ultimately reduce nursery productivity in the southern United States. Water relations of glasshouse-grown Berberis thunbergii DC 'Atoropurpurea', Buxus microphylla 'Seibold' and Zucc japonica and Pittosporum tobira, (Thunb.) Ait. 'Wheeler' were studied under high-temperature root-stress conditions using container-grown plants that were either colonized with vesicular arbuscular mycorrhizal fungi (VAM) or noncolonized. Predawn xylem water potential in stems (ψstem) increased initially (more positive) in response to high root-zone temperatures (40° to 45°C), and then decreased over a 5-day period. Stomatal conductance (gs) and evapotranspiration (ET) were reduced incrementally over time in response to high root-zone temperatures. Root damage occurred, as indicated by reductions in root quality and g, at 35° and 40°C for B. thunbergii and P. tobira, and at 40° and 45°C for the more high-temperature-resistant B. microphylla. Colonization increased g, and ET of B. microphylla at ambient (25°C) and high temperatures (45°C) and increased ET of B. thunbergii at 25°C. Colonized plants had lower (more negative) ψstem with initial exposure to increased root-zone temperatures; however, throughout the remainder of the study period there was little reduction in plant stress in the mycorrhizal isolates used. Root hydraulic conductivity (Lp) increased markedly in B. thunbergii compared to B. microphylla at 40° and 45°C, indicating less high-temperature resistance in B. thunbergii roots. Mycorrhizal colonization did not moderate hydraulic conductivity at high root-zone temperatures of 40° and 45°C. Of the two species, mycorrhizal B. thunbergii had lower Lp at 25° and B. microphylla had lower Lp at 35°C.

In southern nursery production systems, high root-zone temperatures reduce crop quality and productivity. Growth medium temperature influences the uptake of water by root systems (17, 20). Little research has been devoted to water uptake at high media temperatures. Kramer (17), using heat-killed root systems, demonstrated that plants remained alive and unwilted for several days after root death. Transpiration decreased after root death due to leaf injury and gum deposits resulting from substances released from dead cells.

Mycorrhizal fungi have been reported to improve plant water uptake under drought conditions (6, 19, 21, 33, 41). This improvement in plant water relations may be due to mycorrhizal fungi exploiting larger soil volumes, avoiding drought by maintaining a soil–root continuum (31), enhancing plant nutrition (6, 19, 33), or increasing stomatal conductance through regulation of abscisic (ABA) acid/cytokinin levels or osmoregulation (1–3, 27). Marx and Bryan (22) postulated that increased plant survival at high root-zone temperatures was due to metabolites released by the fungal symbiont.

Hydraulic conductivity (Lp) of root systems is influenced by root-zone temperatures (20, 23, 42). Low medium temperatures reduced the Lp of chilling-intolerant species, as illustrated by reduced transpiration and loss of stomatal control (20, 23). Little research has been reported on high root-zone temperatures and hydraulic root conductivity.

Mycorrhizal fungi have been reported to influence hydraulic conductivity of root systems. Under reduced nutrient levels, mycorrhizal plants increased Lp compared to noncolonized plants (10, 11, 24, 27). However, when noncolonized and colonized plants were grown at optimal nutritional levels, Lp rates were comparable (10, 24, 27). Marx and Bryan (22) reported that ectomycorrhizal Pinus taeda seedlings, colonized with Pisolithus tinctorius, survived 5 weeks exposure to medium temperatures of 40°C. Glycine max colonized with Glomus mosseae had greater shoot fresh weight at root-zone temperatures of 36° and 41°C (34) than noninoculated plants.

The objective of this study was to determine the influence of vesicular arbuscular mycorrhizal (VAM) fungi on the relative heat tolerance of selected container-grown woody plants. To achieve this objective, the effects of high root-zone temperatures on water relations, root hydraulic conductivity, and growth were studied using a high-temperature-susceptible, moderately resistant, and resistant container-grown woody plant species either colonized or noncolonized with VAM fungi.

Materials and Methods

Plant material and mycorrhizal fungi colonization. High-temperature root stress studies were conducted with Berberis thunbergii 'Atoropurpurea', Pittosporum tobira 'Wheeler', and Buxus microphylla 'Seibold', which were selected as high root-zone temperature-susceptible, moderately resistant, and resistant, respectively, as previously determined (25).

Rooted liners of the above species were obtained from a local commercial source and planted in 3.8-liter polyethylene containers. The growth medium consisted of 3 composted pine barks: 1 sand (v/w) amended with 2.95 kg m⁻³ dolomitic limestone, 2.95 kg m⁻³ gypsum, and 75 g m⁻³ fritted trac elements (W.R. Grace & Co., Fogelsville, Pa.). Each plant was colonized with either 100 g of a Sorghum sudanense root and soil blend colonized

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nized with *Glomeris etunicatus* Baker and Gerd. and *Glomeris fasciculatum* (Thax. sensu Gerd.) Gerd. and Trappe, or 100 g of noncolonized root and soil blend as previously described (41). Plants were grown in a glasshouse for 12 months, watered daily with distilled water, and fertilized bi-weekly with 20N–8.6P–16.6K soluble fertilizer at 100 ppm N.

*B. thunbergii* and *Buxus microphylla* were used for root hydraulic conductivity studies. Rooted liners were planted into 350-ml aluminum cans with slits cut in the bottom for drainage. Plants were colonized with *Glomeris etunicatus* and *Glomeris fasciculatum* using 25 g of inoculum and grown in a glasshouse for 10 months.

**Water relations.** Colonized and noncolonized glasshouse-grown plants were subjected to four root-zone temperatures over a 5-day period in temperature controlled chambers (26). Root-zone temperatures were held constant as a stress parameter. The four temperatures were 25°C ± 0.4°C (ambient glasshouse media temperature), 35°C ± 0.2°C, 40°C ± 0.2°C, and 45°C ± 0.3°C. Container medium surfaces were uninsulated to prevent anoxia in the medium, which potentially could influence stomatal conductance (4). Plant measurements were recorded over a 6-day period, with the initial daily measurements recorded prior to placement into the chambers. During the treatment period, all plants were watered daily with distilled water at sunset after all measurements were recorded.

Parameters recorded were predawn shoot xylem water potential (Ψ<sub>Ψ</sub> <sub>p</sub>), stomatal conductance (g<sub>st</sub>), and plant evapotranspiration (ET). These parameters were measured for each of the four root-zone temperatures concurrently. The 6-day treatments were replicated three times with a different set of six plants for each colonization and temperature level. For determining Ψ<sub>p</sub>, four of the six plants of each colonization and temperature level and species were used. For determining g<sub>st</sub> and ET, two plants were used. This process was repeated for each species as a separate experiment.

Predawn Ψ<sub>p</sub> (32, 37) was measured with a pressure chamber (Plant Water Status Console, Model 3000, Soilmoisture Equipment Corp., Santa Barbara, Calif.) according to Schoffland et al. (35). Terminal stem sections, 8 to 10 cm long, were cut and placed immediately into the pressure chamber. The predawn period begins at 0400 HR and was completed prior to sunrise.

Morning and afternoon g<sub>st</sub> were measured with a steady-state porometer (LI-COR). The morning period began at 0900 HR and was completed by 1100 HR. The afternoon period began at 1300 HR and was completed by 1500 HR. Measurements were taken from three leaves of each plant and treatment combination. Each leaf was measured with the steady-state porometer using 0.65 cm<sup>2</sup> of abaxial surface. The abaxial surface was measured since the species treated were hypostomatos (12).

After allowing excess water from the previous day to drain, evapotranspiration rates were determined gravimetrically by weighing the plants four times throughout the daylight hours, subtracting the weights, dividing the elapsed time, and dividing by the total leaf area yielding three ET rates: morning, midday, and afternoon. Blank containers with growth medium and no plants were used as a correction factor, accounting for medium surface evaporation at different temperatures. Leaf area was determined at the conclusion of each 6-day treatment period with a leaf area meter (LI-COR). Shriveled leaves for *B. thunbergii* at high media temperature treatments were gathered and leaf area was estimated by leaf dry weight. The linear regression model for estimating leaf area of *B. thunbergii* was: A<sub>L</sub> = 14.41 + 213.30W<sub>L</sub>; where A<sub>L</sub> = the leaf area (cm<sup>2</sup>) and W<sub>L</sub> = the leaf dry weight (g); r<sup>2</sup> = 0.91.

**Plant growth and recovery analysis.** At the conclusion of the 6-day temperature treatment period, plants used for the predawn measurements were returned to the glasshouse, watered daily, and allowed to recover for 10 weeks. They then were graded for survival, regrowth, and plant quality. The visual grading system for quality of shoots and roots ranged from 1 to 5, with 1 being poorest and 5 being best.

Shoot and root dry weights, leaf tissue N and P levels, and leaf area were determined for plants used in the morning and afternoon experiments. Root and shoot dry weights were determined after drying at 60°C for 48 hr in a convection oven. Total leaf N and P were analyzed using a Technicon Autoanalyzer II following procedures described by Stein (40).

**Root hydraulic conductivity.** Colonized and noncolonized glasshouse-grown plants were subjected to root-zone temperatures of 25°C ± 0.4°C (SE), 35°C ± 0.2°C, 40°C ± 0.2°C, and 45°C ± 0.3°C for a 48-hr period using a high temperature root chamber (26). Container surfaces were uninsulated to prevent anoxia in the media.

Effect of increasing root-zone temperatures of L<sub>p</sub>, was determined by placing plants grown in aluminum cans into the temperature chambers and bringing the can media to temperature by flushing with distilled water heated to the temperature. Media temperatures reached experimental levels within 5 min using this technique. Prior to placement in the chamber, plants were maintained in the laboratory under low light levels (2 µmol·s<sup>−1</sup>·m<sup>−2</sup>) for 12 hr to decrease transpiration and xylem sap tension (19, 30).

After 30 min of initial exposure to treatment temperatures, plants were de-topped, placed into a pressure chamber, and scaled with 2 cm of stem exposed (8, 19, 41). Pressure chamber temperatures within ±0.5°C were maintained with a recirculating water bath through a 25-cm round by 20-cm high polyvinylchloride enclosure around the pressure vessel.

A modified 0.5-ml pipette (lower constriction of the pipette removed) was attached with Tygon tubing (41) to the exposed stem and flow rates were recorded as 10 µl of exudate produced per second. Two exudate flow rates were recorded sequentially. Hyperbaric pressure was applied gradually with compressed air to the intact root systems at a rate of 0.1 MPa·min<sup>−1</sup> (42) and was maintained for 10 min. The pressure ranged from 0.3 to 0.6 MPa and was released between readings. The pressures used were selected from the linear portion of flux vs. pressure curves and were at a high enough level not to be influenced by osmotic conditions (8, 24, 36). Flow rates were recorded intermittently for 48 hr. There were two colonization levels and two species for each treatment combination. Root hydraulic conductivity L<sub>p</sub> (cm·s<sup>−1</sup>·MPa<sup>−1</sup>) was calculated from the total flux divided by the total surface area of the roots and the applied hydraulic pressures.

The total surface area (SA<sub>R</sub>) was estimated by root dry weight (DWTR) using the linear model SA<sub>R</sub> = 32.25 + 740.60DWTR<sup>0.7</sup>, where r<sup>2</sup> = 0.86 for *B. thunbergii*; and SA<sub>R</sub> = −14.44 + 524.19DWTR, where r<sup>2</sup> = 0.96 for *B. microphylla*.

**Mycorrhizal fungi colonization analysis.** Mycorrhizal fungi colonization levels were determined from representative root.
samples at the conclusion of the experiment. Root samples were fixed in 1 formalin (10% aqueous): 1 glacial acetic acid: 18 70% ethyl alcohol, by volume (FAA) and later cleared and stained with trypan blue as described by Phillips and Hayman (28). Colonization levels were estimated by the grid intercept technique (41). Colonization levels of colonized and noncolonized B. thunbergii, B. microphylla, and P. tobrig roots were relatively low (Table 1). These levels may have been due to relatively high fertility regimes or may have been due to the difficulty in observing fungal structures due to root morphology.

Statistical analyses. Water relations experiments were analyzed as a repeated measures design (16) with three replications. Experimental variability attributable to photosynthetic photon flux (PPF) was considered and entered into the analysis of variance as a covariant for $g_r$ and ET (39). Each species was analyzed as a separate experiment. Data were analyzed with SAS General Linear Models using Type II sums of squares (38) to account for variability attributable to nondetectable instrumentation responses. Shoot and root quality means of the individual plants within each replication were averaged, ranked, and analyzed using SAS General Linear Models on the main effects only (39). Significant effects are presented as the original unranked data. Root hydraulic conductivity experiments were analyzed as a repeated measures design with two replications, with two plants per replicate and two flux readings per observation. Each species was analyzed as a separate experiment. The data were analyzed with SAS General Linear Models using Type III sums of squares to account for variability attributable to nondetectable instrumentation responses.

Results

Preadawn water relations

Berberis thunbergii 'Atropurpurea'. Root-zone temperatures of 40° and 45°C increased $\psi_{solv}$, compared to all other root-zone temperatures at day 1 for both colonization levels (Fig. 1A). During day 1 at 45° this increase was greatest for noncolonized plants. Compared to day 0, $\psi_{solv}$ at 40° and 45° was lower at 3 to 5 days for colonized and noncolonized plants. Noncolonized plants had lower $\psi_{solv}$ than colonized plants at 45° on day 5. Colonized plants had lower $\psi_{solv}$ than noncolonized plants at 40° on day 5.

Buxus microphylla japonica. Root-zone temperatures of 40° and 45°C increased $\psi_{solv}$ at day 1 for both colonization levels (Fig. 1B). The increase in $\psi_{solv}$ was accentuated by the occurrence of free water (water-soaking) in the leaves (Fig. 2). At 40°, this increase was maintained through day 5. At 45°, however, the increase (greater than at 40°) was maintained through day 4, followed by a decrease to near day 0 levels.

Pittosporum tobira 'Wheeler'. $\psi_{solv}$ was highest at day 1 in root-zone temperatures of 40° and 45°C (Fig. 1C). From day 3 to 5, $\psi_{solv}$ of plants at 40° and 45° decreased, whereas there was little $\psi_{solv}$ change at 25° and 35°.

Stomatal conductance

Berberis thunbergii 'Atropurpurea'. Root-zone temperatures of 40° and 45°C decreased $g_r$ throughout the treatment period regardless of colonization levels (Fig. 3A). Stomatal conductance of plants at 25° and 35° remained similar throughout the treatment period and was higher than plants with media temperatures of 40° and 45°.

Buxus microphylla japonica. Root-zone temperature of 45°C decreased $g_r$ throughout the treatment period regardless of colonization levels (Fig. 3B). Stomatal conductance remained high throughout the treatment period of 25° and 35°, and declined at 40° after day 3. Generally, afternoon $g_r$ exceeded morning $g_r$ for unstressed plants. At day 1 for 45°, $g_r$ during the morning $0.332 \pm 0.014$ cm$^2$ s$^{-1}$ exceeded the afternoon $0.220 \pm 0.024$ cm$^2$ s$^{-1}$ and continued on this trend throughout the treatment period. Colonization increased $g_r$ during the afternoon through- out the 6-day period at 25° $0.436 \pm 0.013$ vs. $0.397 \pm 0.014$ cm$^2$ s$^{-1}$ and 35° $0.435 \pm 0.013$ vs. $0.405 \pm 0.013$ cm$^2$ s$^{-1}$ (25).

Pittosporum tobira 'Wheeler'. Root-zone temperatures of 35° to 45°C decreased $g_r$ from days 0 to 5 (Fig. 3C). Colonization accentuated the $g_r$ decrease at 35° only. Stomatal conductance of plants at 25° increased through day 4. At 25°, afternoon $g_r$ was higher than morning $g_r$; however, with increasing temperature stress at 40° and 45°C, $g_r$ was consistently reduced in both morning and afternoon readings (25).

Plant evapotranspiration

Berberis thunbergii 'Atropurpurea'. There was decreased ET throughout the treatment period at 40° and 45°C, with the greatest reduction in colonized plants (Fig. 4A). Evapotranspiration of colonized plants at 25° increased except for day 2. No differences at any temperature or colonization level were detected among the morning, mid-day, and afternoon ET rates (25).

Buxus microphylla japonica. Greatest reduction of ET occurred at root-zone temperatures of 45°C (Fig. 4B). Evapotranspiration increased on day 1 at 40° and, from days 3 to 5, was comparable to day 0. Evapotranspiration rates were similar for 25° and 35°; however, mycorrhizal colonization increased ET. Evapotranspiration was higher during the afternoon than morning (25).

Pittosporum tobira 'Wheeler'. Compared to plants at 25°C, medium temperatures from 35° to 45° reduced ET at 1 to 5 days (Fig. 4C). At 45° and 40°, no differences were attributable to mycorrhizal fungi colonization. At 35°, colonized plants had

Table 1. The effects of mycorrhizal fungi on total plant, shoot, and root weights and leaf area and colonization levels for Berberis thunbergii 'Atropurpurea', Buxus microphylla japonica, and Pittosporum tobira 'Wheeler'.

<table>
<thead>
<tr>
<th>Colonization</th>
<th>Total plant dry wt (g)</th>
<th>Shoot dry wt (g)</th>
<th>Root dry wt (g)</th>
<th>Leaf area (cm$^2$)</th>
<th>Colonization (%)</th>
<th>A$^*$</th>
<th>B$^*$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>B. thunbergii</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncolonized</td>
<td>9.26</td>
<td>5.85</td>
<td>3.41</td>
<td>455.37</td>
<td>3.30</td>
<td>3.90</td>
<td></td>
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<tr>
<td>Colonized</td>
<td>13.67</td>
<td>8.63</td>
<td>5.04</td>
<td>625.81</td>
<td>28.30</td>
<td>12.40</td>
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<tr>
<td>Pr &gt; F</td>
<td>0.0265</td>
<td>0.0196</td>
<td>0.0695</td>
<td>0.0489</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>B. microphylla</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncolonized</td>
<td>59.03</td>
<td>30.71</td>
<td>28.33</td>
<td>951.20</td>
<td>3.70</td>
<td>6.70</td>
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</tr>
<tr>
<td>Colonized</td>
<td>68.69</td>
<td>36.30</td>
<td>32.39</td>
<td>1077.30</td>
<td>27.80</td>
<td>19.10</td>
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<tr>
<td>Pr &gt; F</td>
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<td>0.0079</td>
<td>0.0286</td>
<td>0.0934</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>P. tobira</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncolonized</td>
<td>104.52</td>
<td>91.68</td>
<td>12.84</td>
<td>3265.30</td>
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<td></td>
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<tr>
<td>Colonized</td>
<td>89.37</td>
<td>78.26</td>
<td>11.11</td>
<td>2633.10</td>
<td>35.60</td>
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<tr>
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<td>0.1271</td>
<td>0.3179</td>
<td>0.0379</td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

*Means between columns were significantly different where $F < 0.05$.

*Means of 24 observations.

Colonization levels from plant roots used in water relations studies.

Colonization levels from plant roots used in root hydraulic conductivity studies.
Fig. 1. Effects of root-zone temperature, mycorrhizal fungi, and time on predawn xylem water potential (ψ_{p}) for (A) Berberis thunbergii ‘Aropurpurea’, LSD_{0.05} = 0.064; (B) Buxus microphylla japonica, LSD_{0.05} = 0.008; and (C) Pittosporum tobira ‘Wheeler’, LSD_{0.05} = 0.016.

higher ET than those at 25°, whereas at 25°C noncolonized plants had higher ET. Evapotranspiration was lowest during the morning throughout the treatment period (25).

Growth and nutritional analysis

Mycorrhizal fungi increased total plant dry weight, shoot dry weight, and leaf area of *B. thunbergii* (Table 1). Shoot dry weight of *B. microphylla* was increased by mycorrhizal fungi, whereas leaf area of *P. tobiara* was reduced (Table 1). Mycorrhizal colonization had no effect on N and P tissue levels among all plant species (data not shown).

Plant recovery

During the 10-week recovery period after root-zone temperature treatment, mycorrhizal fungi colonization did not affect shoot or root quality for any of the test species (25). Shoot quality of *B. thunbergii* and of *P. tobiara* was reduced at root-zone temperatures of 40° and 45°C, and root quality was reduced from 35° to 45° when compared to 25° (Table 2). Shoot quality of *B. microphylla* was reduced at 45° and root quality was reduced at 40° and 45°.

Root hydraulic conductivity

Both *B. thunbergii* and *B. microphylla* Lp increased with increasing temperatures (Fig. 5). There was a decrease Lp compared to initial Lp values for *B. microphylla* at all colonization levels over time at 35°, 40°, and 45°C, and Lp decreased for *B. thunbergii* at 25°, 35°, and 40° (Fig. 5). Root hydraulic conductivity of colonized *B. thunbergii* plants was reduced initially compared to noncolonized plants at 25° (Fig. 5). There was no effect of colonization levels on Lp of *B. thunbergii* from 35° to 45° (Fig. 5). Root conductivity of colonized *B. microphylla* plants was reduced initially compared to noncolonized plants at 35° but not after 24 or 48 hr (Fig. 5). Colonization had no effect on *B. microphylla* plants at 25°, 40°, and 45° (Fig. 5).

Discussion

Water uptake by the test species was not restricted at 40° or 45°C from day 0 through day 1, as illustrated by increased \( \psi_{\text{shoot}} \) (Fig. 1) and increased \( L_p \) (Fig. 5). Syvertsen (42) demonstrated that \( L_p \) of *Citrus* rootstocks increased from 15° to 30°C. The increased levels of \( \psi_{\text{shoot}} \) at 45° and day 1 compared to 25° (Fig. 1) and free water in the leaves (Fig. 2) indicated that a temperature-induced osmotic flux condition was present. Assuming that the stomata were closed and the atmospheric humidity was at or near saturation, predawn \( \psi_{\text{shoot}} \) should reflect the water potential of the media (32). The observed increase in \( \psi_{\text{shoot}} \) should reflect the water potential of the media (32). The observed increase in \( \psi_{\text{shoot}} \) from 25° to 45° can be described by estimating media water potential differences due to media temperature using Van’t Hoff's principle.

Increased \( L_p \) with temperature for both *B. thunbergii* and *B. microphylla* (Fig. 5) suggested that biologically controlled resistances to water uptake may have been reduced at higher media temperatures. Membrane lipids become more fluid at high temperatures (29). Root-zone temperatures from 40° to 45°C may have altered or disrupted membrane lipids, thus allowing greater \( L_p \). Histological analysis of high-temperature-stressed *Citrus* roots illustrated membrane disruption (15).

The linear increase in *B. microphylla* \( L_p \) from 25° to 45° and the deviation from linearity of *B. thunbergii* \( L_p \) from 35° to 45° indicated that *B. thunbergii* roots were more susceptible to high root-zone temperature membrane disruption compared to *B. microphylla* (Fig. 5). Deviations from linearity in flux rates of root membranes were reported when roots of chilling-sensitive species were subjected to low root-zone temperatures (20).

Mycorrhizal fungi colonization maintained reduced \( \psi_{\text{shoot}} \) at 40° and 45°C for *B. thunbergii*, and at 45° for *P. tobiara*, after initial exposure through day 1 (Fig. 1 A and C). Resistance to water transport at these temperatures may have increased in roots of colonized plants, slowing the increased water flux generated by temperature differences. The increased resistance to flux may have been due to increased membrane integrity of colonized roots. This increased resistance was observed for colonized *B. thunbergii* \( L_p \) compared to noncolonized \( L_p \) initially from 25° to 40° only (Fig. 5).

The increased \( \psi_{\text{shoot}} \) remained generally for 1 to 2 days for all species, followed by a precipitous decline (Fig. 1). Kramer (17) suggested that xylem vessels become occluded after prolonged exposure to high media temperatures, resulting in leaf death and abscission. Root cell membrane disruption generally begins at around 40°C with exposures >300 min (14). Root cell disruption may have resulted in the release of previously compartmentalized materials that may have been toxic to leaves (17), resulting in decreased \( \psi_{\text{shoot}} \), decreased \( g_s \), and decreased ET at high temperatures (Figs. 1, 3, and 4). Mycorrhizal fungi colonization did not appear to mediate any of these efforts.

Mycorrhizal fungi colonization of *B. thunbergii* appeared to cause a large decrease in \( \psi_{\text{shoot}} \) at 40°C after day 1 (Fig. 1). The cause of this response is perplexing, but can be explained by differences in biomass. Colonized *B. thunbergii* plants were larger than controls, as indicated by increased levels of plant dry weight and leaf area (Table 1). Perhaps 40°C media was high enough to occlude xylem vessels, but not high enough to release toxic materials causing leaf abscission; thus, the increased biomass reduced the \( \psi_{\text{shoot}} \) for colonized plants at a rate greater than for noncolonized plants. This difference cannot be illustrated from the \( g_s \) or ET data because they both are normalized to 1 cm² (Figs. 3 and 4).

Fig. 2. Abaxial surface of *Buxus microphylla japonica* leaves showing predawn leaf hydration at high (40° and 45°C) root-zone temperatures. Note water-soaking effects along leaf blade periphery.
Fig. 3. Effects of root-zone temperature, mycorrhizal fungi, and time on afternoon stomatal conductance ($g_a$) for (A) Berberis thunbergii ‘Atropurpurea’, LSD$_{0.05}$ = 0.006; (B) Buxus microphylla japonica, LSD$_{0.05}$ = 0.007; and (C) Pittosporum tobira ‘Wheeler’, LSD$_{0.05}$ = 0.008.
Fig. 4. Effects of root-zone temperature, mycorrhizal fungi, and time on afternoon evapotranspiration (ET) for (A) Berberis thunbergii 'Atropurpurea', LSD$_{0.05} = 0.083$; (B) Buxus microphylla japonica, LSD$_{0.05} = 0.087$; and (C) Pittosporum tobira 'Wheeler', LSD$_{0.05} = 0.368$.
Data from this study concur with Ingram (14) that B. thunbergii and P. tobiar are susceptible to root-zone stress temperatures beginning around 40° and B. microphylla is susceptible to root-zone stress temperatures beginning around 45°.

Mycorrhizal fungi colonization did not enhance high-temperature root-zone stress, as suggested by previous studies. The root-zone temperatures used in this study were markedly higher than those reported by Marx and Bryan (22) and Schenk and Smith (34). Perhaps the temperature tolerance limit of the fungal symbiont was passed. Future research should focus on selecting mycorrhizal isolates indigenous to higher-temperature soils.

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Micropropagation of Cherry Rootstocks: I.

Response to Culture

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Abstract. Establishment, shoot proliferation, root induction, and transplanting stages were accomplished with *Prunus avium × P. cerasus* cv. "Cot", *Dwarf Mahaleb* ("P. mahaleb") (both vegetatively propagated rootstock cultivars) and seedling Mazzard sweet cherry ("P. avium") '46-1 Mazzard', ("P. avium"), a clonal rootstock seed tree, could be established and multiplied by the same procedure. However, no rooting was obtained. Limited rooting was possible with seedling Mazzard trees < 3 years in age. It was concluded that the limiting factor in '46-1 Mazzard' rooting was the mature status of the source material.

Sweet cherry is an important fruit crop in California, but has been subject to decline due to such diseases as crown and root rot (*Phytophthora* spp.), western X (Buckskin mycoplasma), and stem pitting (tomato ringspot virus). All are associated with unadjustable of current rootstocks and, in particular, susceptibility to wet and heavy soil conditions (9, 11). As compared to *Prunus mahaleb* L., Mazzard seedings are more resistant to various *Phytophthora* spp. and better adapted to heavy, wet soils, but trees are large and not precocious. Mahaleb seedlings are more tolerant of western X disease than Mazzard seedlings. A clonal selection ('Dwarf Mahaleb') has been used as an interstock to produce partial dwarfing with sweet cherry cultivars.

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