Water Use and Growth of Honey Locust and Tree-of-Heaven at High Root-zone Temperature

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Abstract. Honey locust (*Gleditsia triacanthos* **var.** *inermis* **Wind.) and tree-of-heaven** *Ailanthus altissima* **(Mill.) Swingle] sometimes are exposed to high root-zone temperatures in urban microclimates. The objective of this study was to test the hypothesis that seedlings of these species differ in how elevated root-zone temperature affects growth, leaf water relations, and root hydraulic properties. Shoot extension, leaf area, root : shoot ratio, and root and shoot dry weights were less for tree-of-heaven grown with the root zone at 34C than for those with root zones at 24C. Tree-of-heaven with roots at 34C had a lower mean transpiration rate (E) than those grown at 24C, but leaf water potential (ψ_l) was similar at both temperatures. In contrast, shoot extension of seedlings of honey locust grown with roots at 34C was greater than honey locust at 24C, E was similar at both temperatures, and ψ_l was reduced at 34C. Hydraulic properties of root systems grown at both temperatures were determined during exposure to pressure in solution held at 24 or 34C. For each species at both solution temperatures, water flux through root systems (J_v) grown at 34C was less than for roots grown at 24C. Roots of tree-of-heaven grown at 34C had lower hydraulic conductivity coefficients (L_p) than those grown at 24C, but L_p of roots of honey locust grown at the two temperatures was similar.**

Elevated root-zone temperatures may contribute to premature senescence and high mortality rates of trees in urban microclimates. Graves and Dana (1987a) found that root-zone temperatures at urban tree planting sites averaged more than 7C higher than those in a nearby temperate forest, and temperatures > 30C were observed frequently in soils at street tree-planting sites.

Data on the responses of temperate trees found in urban landscapes to elevated root-zone temperature are limited, but the growth and water status of red maple (*Acer rubrum* L.) (Graves et al., 1989b) and tree-of-heaven (Graves et al., 1989a) are sensitive to high root-zone temperatures in the range documented at urban sites. In both species, stem elongation, leaf area, and root and shoot biomass were reduced with the root zone > 30C. Leaves of plants with roots at 36C had high diffusive resistance, indicating that water transport was limited by stomatal closure or the capacity of the root system to extract water. Preliminary trials, however, indicated that seedling growth in honey locust was maintained at the temperatures that reduced growth in red maple and tree-of-heaven.

Therefore, the objective of this study was to test the hypothesis that seedlings of honey locust and tree-of-heaven differ in how elevated root-zone temperature affects growth and water transport in leaves and roots.

Growth and shoot water relations. Seeds of tree-of-heaven and honey locust (F.W. Schumacher, Sandwich, Mass.) were sown 12 and 26 Jan. 1988, respectively, in flats of Jiffy Mix (W.R. Grace, Fogelsville, Pa.). Flats were placed in a greenhouse in which they were exposed to outdoor photoperiods and day/night cycles of 20 to 30C. Thirty seedlings of each species were planted singly on 2 Feb. in 100% ground, calcined clay (Waverly Mineral Products, Bala Cynwyd, Pa.) in l-liter plastic containers. Seedlings were held in the greenhouse until 7 Mar. and irrigated once daily with tap water. Irradi-

ance and relative humidity in the greenhouse were not measured.

On 7 Mar., seedlings were transferred to a plant growth chamber with 16-h photoperiods. Irradiance, measured with a quantum radiometer (LI-COR, Lincoln, Neb.) at shoot apicies, was 410 ($\pm$40) $\mu mol \cdot m^{-2} \cdot s^{-1}$ (400 to 700 nm). Cool-white fluorescent and incandescent lamps emitted 90% and 10% of the irradiance, respectively. Air temperature during light and dark hours was 24 and 21C, respectively, with 49% ($\pm$5%) relative humidity. Every 3 days, through 21 Mar., and every day between 21 and 28 Mar. plants were irrigated to container capacity with Hoagland solution no. 1 (Hoagland and Arnon, 1950). Plants were irrigated to container capacity all other days through 21 Mar. with deionized water.

After 21 days in the growth chamber, 16 plants of both species were selected for study based on uniformity of stem length. Eight plants of a species were assigned randomly in a completely randomized design to a 20-day root-zone temperature treatment of 24 or 34C ($\pm$ lC) (Graves and Dana, 1987b) in the growth chamber such that the shoot environment was not altered. Daily irrigations continued with Hoagland solution no. 1 that was heated or cooled to maintain root-zone temperatures during application. Distance between the shoot apex and the medium surface in each container was measured when treatments began (day 0) and on days 5, 10, 15, and 20.

Xylem water potential (ψ_l) of each plant was estimated using a pressure chamber (Scholander et al., 1965) before the photoperiod and 6 h after the photoperiod began on day 20. ψ_l was measured on a fully expanded leaf from the basipetal half of each shoot in random order. Transpiration rate (E) was measured gravimetrically during the middle hour of the photoperiod on day 20. Total leaf area of each plant then was determined with a LI-COR 3000 leaf area meter, and the mean transpiration rate (E) was assessed as mmol H_2O/m^2 per sec. Root and shoot weights were determined after drying samples in a forced-air oven at 65C for 48 h and allowing them to cool at room temperature for 1 h.

Table 1. Growth and shoot water relations of seedlings of honey locust and tree-of-heaven grown with root zones at 24 or 34C. Values are means of eight replicates. SE of means are shown where least significant difference (LSD, $\alpha = 0.05$) values are not applicable.

Dependent variable	Honey locust		Tree-of-heaven		LSD
	Root-zone temp (°C)				
	24	34	24	34	
Root dry wt (g)	0.83	0.80	3.65	2.00	1.01
Shoot dry wt (g)	1.12	1.30	6.20	5.18	0.58
Root : shoot biomass ratio	0.74 (0.04)	0.67 (0.09)	0.59 (0.03)	0.39 (0.02)	
Leaf area (cm^2)	105.5	125.9	929.2	616.0	78.6
ψ_l before photoperiod (MPa)	−0.40 (0.03)	−0.43 (0.02)	−0.38 (0.03)	−0.48 (0.02)	
ψ_l during photoperiod (MPa)	−1.44 (0.10)	−1.62 (0.04)	−0.64 (0.04)	−0.68 (0.04)	
Transpiration rate (mmol·m^{-2}·s^{-1})	1.31	1.54	1.56	0.84	0.32

Table 2. Water flux (J_v) and hydraulic conductivity coefficients (L_p) with root systems of honey locust and tree-of-heaven grown with root zones at 24 or 34C. Hydraulic properties were measured in solution at 24 and 34C. Values are means of six to eight (honey locust) and three or four (tree-of-heaven) replicates. (Table 3 shows the significance of sources of variation in these data.)

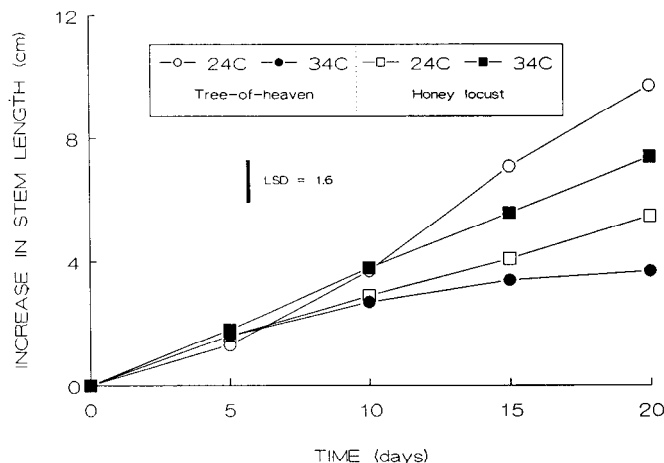| Root hydraulic property	Pressure (MPa)	Honey locust				Tree-of-heaven			
		Growth temp (°C)							
		24	24	34	34	24	24	34	34
		Measurement temp (°C)							
		24	34	24	34	24	34	24	34
J_v (cm^3·mg^{-1}·s^{-1} × 10^{-7})	0.345	2.53	4.79	1.35	2.94	10.16	13.89	1.14	3.21
	0.414	3.16	5.91	1.82	3.95	11.78	15.81	1.17	3.92
	0.483	4.02	6.77	2.54	4.85	13.97	17.73	1.55	4.77
	0.552	5.10	7.59	3.67	5.57	17.13	20.61	2.01	5.84
L_p (cm^3·mg^{-1}·s^{-1}·MPa^{-1} × 10^{-6})		1.24	1.34	1.11	1.27	3.35	3.20	0.43	1.27

Fig. 1. Increase in stem length of seedlings of honey locust and tree-of-heaven grown with root zones at a constant 24 or 34C. Each value is the mean of eight replicates. The vertical bar represents the LSD (α = 0.05).

Table 3. Significance of sources of variation in water flux (J_v) of root systems of honey locust and tree-of-heaven at 24 and 34C.

Source of variation	Significance of variation in J_v	
	Honey locust	Tree-of-heaven
Measurement temp (MT)	**	NS
Growth temp (GT)	**	*
Pressure	***	***
Pressure (linear) × MT	NS	NS
Pressure (linear) × GT	NS	**
Pressure (quadratic)	NS	NS
Pressure (quadratic) × MT	NS	NS
Pressure (quadratic) × GT	NS	NS
Pressure × MT × GT	NS	NS

NS,*,**,***Nonsignificant or significant at α = 0.05, 0.01, and 0.001, respectively.

Dependent variables were analyzed by analysis of variance (ANOVA). A least significant difference (Steel and Torrie, 1980) was determined for variables with significant (α = 0.05) interactions between time, species, or treatment.

Root conductance to water flow. Water conductance properties of root systems of honey locust and tree-of-heaven were analyzed in separate experiments using methods similar to those of Fiscus (1977).

Seeds of honey locust were germinated as described for the previous experiment. Seedlings were planted in ground, calcined clay in l-liter plastic containers 21 days after seeds were sown. Plants were transferred to the growth chamber and grown with the environmental conditions described for the growth and shoot water relations study. Seven days after seedlings were moved to the growth chamber, eight uniform plants were assigned randomly to root zones at 24 or 34C. Plants were irrigated once daily to container capacity with temperature-controlled Hoagland solution no. 1 such that root-zone temperatures were maintained during application. Two plants were selected at random from each treatment on day 56 of treatment. They were moved to a laboratory and irrigated to container capacity with deionized water. Root systems were gently separated from the growing medium, and stems were severed immediately below the scars where the cotyledons had been attached. Weight of leaves from each plant was determined after they were dehydrated as described for the previous experiment.

Tissues peripheral to the vascular cambium were peeled from the upper 5 cm of the stump of each root system to prevent axial water flow through cortical cells. Each stem was mounted singly in the lid of a stainless steel chamber filled with 50% Hoagland solution (no. 1, pH 5.7 to 5.9). Air was bubbled in the solution of the sealed chamber until pressure reached 0.345 MPa. Solution aeration was maintained by venting gas through the lid at the same rate it entered the solution, 0.8 cm^3·s^{-1}. One hour elapsed between severing shoots and attaining a vessel pressure of 0.345 MPa.

Root exudate was collected in tygon tubing fitted to each stump and was weighed on a balance every 3 min. Steady-state root water flux (J_v) was achieved within 3½ h and then was recorded for 1 h at each of four pressures (0.345, 0.414, 0.483, and 0.553 MPa) applied in ascending order, allowing five observations per system at each pressure. Root systems were held for 30 min at each new pressure to regain steady state before data were collected. J_v was expressed on the basis of leaf dry weight. Compared with several other traits, using leaf dry weight to express data resulted in the least variation in J_v among plants within treatments during a preliminary experiment.

A replication of this experiment consisted of J_v determinations on two consecutive days. Four plants were analyzed each day, two from each growth temperature treatment. On the first day, temperature of the chamber solution was selected at random as 24 or 34C (± lC). Procedures were repeated with different plants during the same hours the 2nd day, but the alternate solution temperature was used. Four replications were performed. All data were combined for analysis.

Methods used for tree-of-heaven were identical to those for honey locust except for the following. Seedlings were potted in 2.5-liter containers 56 days after seeds were sown. Treatments were applied 14 days after seedlings were introduced to chamber conditions. Six plants were assigned to each of the two root-zone temperatures, and treatments lasted 28 days. J_v was determined each minute for 40 min at each of the four pressures, allowing 10 observations per sample at each pressure. Root systems were held 60 min at each new pressure to allow J_v to regain steady state before collecting data.

The root conductance experiments were analyzed as a split split-plot design with solution temperature as whole plots, growth temperature as subplots, and pressure as the sub-subplot treatment. ANOVA was performed for J_v of each species using a generalized linear model procedure (SAS Institute, 1986). F-max tests showed the assumption of homogeneity of variances was violated for both species, and this could not be corrected by transforming data. Plots of predicted vs. residual values revealed irregular data points that exceeded predicted values by more than three standard deviations

of the mean. J_v failed to attain steady state in root systems from which such data were generated. We assumed this was caused by physical damage to roots during preparation. These data, < 10% of the observations, were excluded as outliers, and the assumption of homogeneity of variances was no longer violated during ANOVA.

Within species and for all measurement and growth temperature combinations, J_v was fitted to regression models of the form $J_v = b_0 + b_1$ (pressure). The hydraulic efficiency coefficient (L_p) was determined as the slope of the linear regression of J_v on pressure (Fiscus, 1977).

Growth and shoot water relations. Stems elongated more in seedlings of honey locust with roots at 34C than in those at 24C, but the reverse was true for tree-of-heaven (Fig. 1). Seedlings of tree-of-heaven grown with roots at 34C also exhibited reduced root and shoot dry weights, smaller root : shoot biomass ratios and less leaf area than plants grown at 24C (Table 1). Root-zone temperature did not affect these traits in seedlings of honey locust.

Before the beginning of the photoperiod on day 20, ψ_l ranged from –0.48 to –0.38 MPa for both species (Table 1). All plants had lower ψ_l during the photoperiod, but the decrease was most evident among honey locust with roots at 34C. Seedlings of tree-of-heaven grown at 34C had a mean E $\approx 46\%$ less than that at 24C (Table 1). E of honey locust was similar at the two temperatures.

Root conductance to water flow. Roots of honey locust measured in solution at 34C showed higher J_v than those measured at 24C, and at both solution measurement temperatures, J_v was higher for systems grown with roots at 24C than for those grown at 34C (Tables 2 and 3). The pressure **x** measurement temperature and pressure **x** growth temperature interaction terms were not significant for honey locust (Table 3), indicating that L_p at the two temperatures was not different (Table 2).

Measurement temperature did not affect the J_v through root systems of tree-of-heaven (Tables 2 and 3). Roots of tree-of-heaven grown at 24C had higher J_v than those of root systems grown at 34C (Tables 2 and 3). The interaction between pressure and growth temperature was significant (Table 3), indicating L_p of systems grown at 24C was higher than L_p of roots grown at 34C (Table 2).

These experiments support the hypothesis that high root-zone temperature affects root hydraulic properties, leaf water relations, and growth of seedlings of honey locust and tree-of-heaven differently. The capacity for seedlings of honey locust to collect, transport, and transpire water was not affected by continuous exposure of roots to 34C. In contrast, the L_p of roots of tree-of-heaven grown at 34C was lower than the L_p of roots of tree-of-heaven formed at 24C. Assuming that the hydraulic properties of a whole root system are analogous to those of a single membrane (Fiscus, 1977), the L_p of a root system defines the gradient in water potential within the hydraulic pathway ($\Delta\psi$) between root and

shoot tissues at any given flow rate. Thus, the $\Delta\psi$ required to sustain a given rate of water flow in tree-of-heaven grown with roots at 34C likely was larger than that required for plants with root zones at 24C. The relatively high ψ_l and relatively low E during the photoperiod suggest that seedlings of tree-of-heaven did not develop a $\Delta\psi$ sufficient to override the increased resistance to flow in the roots, probably because of a transient decline in ψ_l that induced stomatal closure. Because the driving force for water movement depends largely on E, the magnitude of $\Delta\psi$ would have decreased as stomata closed. Therefore, E may have decreased to a level governed principally by the rate of water absorption.

Unlike tree-of-heaven, root L_p of honey locust was not reduced in plants grown with roots at 34C relative to those grown at 24C (Tables 2 and 3). Furthermore, the low ψ_l observed during the photoperiod indicates that a large $\Delta\psi$ was maintained at high root-zone temperature (Table 1). E was unaffected by the higher growth temperature despite the decline in ψ_l (Table 1). Solute accumulation in leaves during exposure to high root-zone temperature may have enabled turgor and E to be maintained at low ψ_l. Whether or not osmotic adjustment or some other mechanism to maintain turgor was involved, water transport to expanding tissues in seedlings of honey locust was not impaired at high root-zone temperature, and shoot and root growth were similar to that observed for plants with 24C root zones (Table 1).

J_v was reduced among plants of both species grown with roots at 34C (Tables 2 and 3). Others have found that roots formed at high temperatures are thinner than those produced at temperatures optimal for growth (Hellmers, 1963; Nielsen, 1974). Thus, high growth temperature may have reduced J_v by decreasing xylem vessel diameter, thereby increasing the axial resistance to water transport (Oosterhuis, 1983). Increased suberization or the deposition of secondary cell wall materials behind the zone of elongation where root water uptake is most rapid (Boyer, 1985) also may have reduced J_v at high temperature. Elevated root-zone temperature also might have influenced resistance by altering membrane structure and fatty acid composition. High temperature acclimation appears to involve changes in the degree of fatty acid unsaturation and fluidity of polar lipids (Berry and Bjorkman, 1980). Markhart et al. (1980) found that increased unsaturation in membrane fatty acids during temperature acclimation in soybean *(Glycine max* L.) was associated with increased resistance to root water uptake. Roots of honey locust grown at both temperatures showed greater J_v when measured at 34C than when solution at 24C was used (Tables 2 and 3). Although not significant at the $\alpha = 0.05$ level, the same trend was evident in tree-of-heaven, probably from the reduced viscosity of aqueous solutions at high temperature (Dainty et al., 1963).

Changes in ψ_l, E, J_v, and L_p reported herein are consistent with numerous reports dem-

onstrating that the root-zone environment can modify water transport characteristic of roots and alter ψ_l during transpiration. Water stress (Levy et al., 1983; Oosterhuis and Wiebe, 1986; Ramos and Kaufmann, 1979), salinity (Shalhevet et al., 1976), and hypoxia (Kuiper, 1964; Newman, 1976) are among the conditions affecting the capacity of roots to supply water to leaves. Each of these factors likely interacts with high temperature to influence growth-limiting hydraulic resistances of trees in urban microclimates.

Both species used in this study survived root zones constantly at 34C, but they differed dramatically in the control over water use at high temperature. Our findings are consistent with a previous report that stomata of honey locust are relatively insensitive to low ψ_l (Potts and Herrington, 1982). The capacity for this species to maintain growth with roots at 34C is unique when compared with tree-of-heaven and other temperate tree species subjected continuously to root-zone heat (Barney, 1951; Graves et al., 1989a, 1989b; Gur et al., 1976; Hellmers, 1963; Nightingale, 1935). Thus, honey locust appears unusually well adapted to microclimates with high soil temperature. How high temperature interacts with other environmental factors, such as drought, that may stress trees of this species is not clear. Graves and Wilkins (1991) found that growth of honey locust in solution culture was reduced when solution osmotic potential was reduced from –0.05 to –0.10 MPa by polyethylene glycol 8000 (PEG). In contrast to those at –0.05 MPa, growth of seedlings stressed with PEG was not reduced by 35C root-zone temperature. Further studies are needed to determine whether these responses are consistent with those of honey locust grown in urban landscapes.

Literature Cited

Barney, C. W. 1951. Effects of soil temperature and light intensity on root growth of loblolly pine seedlings. Plant Physiol. 26:146-163.

Berry, J. and O. Bjorkrnan. 1980. Photosynthetic response and adaptation to temperature in higher plants. Annu. Rev. Plant Physiol. 31:491-543.

Boyer, J.S. 1985. Water transport. Annu. Rev. Plant Physiol. 36:473-516.

Dainty, J., P.C. Croghan, and D.S. Fensom. 1963. Electro-osmosis with some applications to plant physiology. Can. J. Bot. 41:952-966.

Fiscus, E.L. 1977. Determination of hydraulic and osmotic properties of soybean root systems. Plant Physiol. 59:1013-1020.

Graves, W.R. and M.N. Dana. 1987a. Root-zone temperature monitored at urban sites. HortScience 22:613-614.

Graves, W.R. and M.N. Dana. 1987b. A system to control root-zone temperature. HortScience 22:957-958.

Graves, W.R. and L.C. Wilkins. 1991. Growth of honey locust seedlings during high root-zone temperature and osmotic stress. HortScience 26:1312-1315.

Graves, W. R., R.J. Joly, and M.N. Dana. 1989a. Influence of root-zone temperature on growth of *Ailanths altissima* (Mill.) Swingle. J. Environ. Hort. 7:79–82.

Graves, W. R., R.J. Joly, and M.N. Dana. 1989b. Root-zone temperature affects water status and

growth of red maple. J. Amer. Soc. Hort. Sci. 114:406-410.

Gur, A., J. Hepner, and Y. Mizrahi. 1976. The influence of root temperature on apple trees. II. Clonal differences in susceptibility to damage caused by supraoptimal root temperature. J. Hort. Sci. 51:195-202.

Hellmers, H. 1963. Effects of soil and air temperature on growth of redwood seedlings. Bot. Gaz. 125:172-177.

Hoagland, D.R. and D.I. Amen. 1950. The water-culture method for growing plants without soil. Calif. Agr. Expt. Sta. Circ. 347.

Kuiper, P.J.C. 1964. Water uptake of higher plants as affected by root temperature. Meded. Land-bouwhogeschool Wageningen 64:1-11.

Levy, Y., J.P. Syvertsen, and S. Nemec. 1983. Effect of drought stress and vesicular-arbuscular mycorrhiza on citrus transpiration and hydraulic conductivity of roots. New Phytol. 93:61-66.

Markbart, A.H. III, M.M. Peet, N. Sionit, and P.J. Kramer. 1980. Low temperature acclimation of root fatty acid composition, leaf water potential, gas exchange and growth of soybean seedlings. Plant Cell Env. 3:345-441.

Newman, E.I. 1976. Interaction between osmotic- and pressure-induced water flow in plant roots. Plant Physiol. 57:738-739.

Nielsen, K.F. 1974. Roots and root temperature, p. 293-333. In: E.W. Carson (ed.). The plant root and its environment. Univ. Press Virginia, Charlottesville.

Nightingale, G.L. 1935. Effects of temperature on root growth, anatomy, and metabolism of apple and peach roots. Bot. Gaz. 96:581-639.

Oosterhuis, D.M. 1983. Resistances to water flow through the soil-plant system. S. Afr. J. Sci. 79:459-465.

Oosterhuis, D.M. and H.H. Wiebe. 1986. Water stress preconditioning and cotton root pressure-flux relationships. Plant and Soil 95:69-76.

Potts, D.F. and L.P. Herrington. 1982. Drought resistance adaptations in urban honeylocust. J. Arboriculture 8:75-80.

Ramos, C. and M.R. Kaufmann. 1979. Hydraulic resistance of rough lemon roots. Physiol. Plantarum 45:311-314.

SAS Institute, Inc. 1986. SAS system for regression. SAS Institute, Inc., Cary, N.C.

Scholander, P. F., H.T. Hammel, D. Bradstreet, and E.A. Hemmingsen. 1965. Sap pressure in vascular plants. Science 148:339–346.

Shalhevet, J., E.V. Maas, G.J. Hoffman, and G. Ogata. 1976. Salinity and the hydraulic conductance of roots. Physiol. Plantarum 38:224-232.

Steel, R.G.D. and J.H. Torrie. 1980. Principles and procedures of statistics. 2nd ed. McGraw-Hill, New York.