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American Journal of Botany, Vol. 78, No. 7. (Jul., 1991), pp. 906-915.

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CHANGES IN HYDRAULIC CONDUCTIVITY AND ANATOMY CAUSED BY DRYING AND REWETTING ROOTS OF *AGAVE DESERTI* (AGAVACEAE)¹

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Concurrent determinations of changes in hydraulic conductivity and tissue anatomy were made for roots of *Agave deserti* excised during drying and following rewetting in soil. At 30 d of drought, hydraulic conductivity had declined less than twofold for older nodal roots, tenfold for young nodal roots, and more than 20-fold for lateral roots ("rain roots" occurring as branches on the nodal roots). These decreases were consistent with increases in cortical lacunae caused by cell shrinkage and collapse. Similarly, reduction of lacunae in response to rewetting after 7 d of drought corresponded to levels of recovery in hydraulic conductivity, with young nodal roots showing full recovery, lateral roots returning to only 21% of initial conductivity, and older nodal roots changing only slightly. Increases in suberization in the exodermis, endodermis, and cortex adjacent to the endodermis in response to drying coincided with decreases in hydraulic conductivity. Measurements of axial hydraulic conductance per unit length before and after pressurization indicated that embolism caused reductions in axial conductance of 98% for lateral roots, 35% for young nodal roots, and 20% for older nodal roots at 7 d of drought. Embolism, cortical lacunae, and increasing suberization caused hydraulic conductivity to decline during drought in the three root types, thereby helping limit water loss to dry soil; the recovery in hydraulic conductivity for young nodal roots after rewetting would allow them to take up water readily once soil moisture is replenished.

Plants can lose water when their root systems have a higher water potential than the soil (Mooney et al., 1980; Dirksen and Raats, 1985; Richards and Caldwell, 1987). As the soil dries, hydraulic conductances within the soil, from the soil to the root, and within the root may decrease, thereby limiting plant water loss (Passioura, 1988). For instance, decreases with soil drying in the root hydraulic conductivity (L_p), a coefficient relating the volume of water flow per unit time to a driving force such as the difference in water potential (Nobel, 1991), have been observed in lemon (Ramos and Kaufmann, 1979), soybean (Blizzard and Boyer, 1980), barley (Shone and Flood, 1980), and the desert Crassulacean acid metabolism (CAM) plants *Agave deserti* and *Ferocactus acanthodes* (Nobel and Sanderson, 1984; Nobel, Schulte, and North, 1990). Variable L_p is particularly important for the two desert species, because prevention of water flow from a succulent shoot to dry soil must be counter-

balanced by the ability to absorb water rapidly following sporadic rainfall events (Drew, 1979). In the extreme case, roots can act as rectifiers, having a high L_p in wet soil, favoring water uptake, and a low L_p in dry soil, limiting water loss.

The tendency of root L_p to change in response to water availability, i.e., for roots to behave as rectifiers, has been investigated with respect to the structural characteristics of hypodermal sleeves in *Zea mays* (Shone and Clarkson, 1988). Other anatomical studies, though not specifically addressing root rectification, have similarly focused on the role of the hypodermis (exodermis) in regulating radial water flow (Oleson, 1978; Clarkson et al., 1987). The endodermis as a barrier to water movement has been extensively studied (Clarkson, Sanderson, and Russell, 1968; Clarkson and Robards, 1975), though how it might be involved in rectification has not been addressed. The developmental state and dimensions of vessels in the root xylem can vary with root age, location within the root system, and environmental conditions, thereby possibly contributing to variability in root L_p (McCully and Canny, 1988; Sanderson, Whitbread, and Clarkson, 1988; Luxová, 1989).

The present study examined the anatomical basis of variations in root L_p for *Agave deserti*

¹ Received for publication 25 August 1990; revision accepted 8 March 1991.

The authors thank David Alm and Francis Lopez for rewarding discussions. Financial support was provided by NSF grants BSR-88-02237 and DCB-90-02333 and the Ecological Research Division of the Office of Health and Environmental Research, Department of Energy contract DE-FC03-87-ER60615.

in response to differences in the availability of soil water. *Agave deserti*, a perennial leaf succulent of the Sonoran Desert, was used because its roots have been shown to act as rectifiers (Nobel and Sanderson, 1984). After plants were subjected to drying and in some cases rewetting in soil, whole roots and root segments were used for concurrent determinations of changes in tissue anatomy and L_p ; L_p was measured by the technique of vacuum-induced flow (Shone and Clarkson, 1988). Based on morphological criteria (Palta and Nobel, 1989), roots were classified as old or young nodal roots, previously designated "established roots," arising from nodes at the base of the stem; or as lateral roots, previously designated "rain roots," arising as branches from nodal roots. These three classes of root exhibit differences in L_p (Nobel, Schulte, and North, 1990) that may be functionally related to differences in anatomy.

MATERIALS AND METHODS

Plant material—Plants of *Agave deserti* Engelm. (Agavaceae), each approximately 25–30 cm tall with 9–15 unfolded leaves, were collected from "Agave Hill" at the University of California Philip L. Boyd Deep Canyon Desert Research Center (33°38'N, 116°24'W, 850 m elevation) near Palm Desert, California. They were placed in pots 20 cm in diameter containing soil from Agave Hill and maintained in a glasshouse at the University of California, Los Angeles. Plants alternately received 3 mo of biweekly watering with 0.1-strength Hoagland's solution and 3 mo of drought to simulate soil moisture conditions at their collection site. At the start of experiments, plants with about 30 nodal roots each were transferred to 36-cm × 28-cm × 12-cm tubs containing soil from Agave Hill in which three Wescor PCT-55-15 soil thermocouple psychrometers were placed at a depth of 6 cm in the root zone to monitor soil water potential (Ψ_{soil}). Agreement between psychrometers, which were calibrated at the beginning and end of the experiments with standard salt solutions, was ± 0.1 MPa, except at a Ψ_{soil} of -3.2 MPa, where the agreement was ± 0.3 MPa. The tubs allowed individual roots to be excised with minimal disturbance to the remaining roots and permitted the soil to dry at rates comparable to those observed in the field (Young and Nobel, 1986).

Experiments were begun near the end of a 3-mo watering period, so older nodal roots (6–12 mo in age) had experienced at least one droughting episode, whereas young nodal roots (4–6 wk) and lateral roots (also 4–6 wk) had

grown only under well-watered conditions. Nodal roots were generally 20–40 cm long; the distal 10-cm portion was considered young based on its white color and immature stele, whereas older segments were brown and were excised from near midroot. Lateral roots averaged 10 cm in length.

Root hydraulic conductivity—Individual root segments about 10 cm long were excised, submerged in distilled water, recut, and all tissues external to the stele were dissected away for approximately 15 mm at the proximal end. The exposed stele was then trimmed under water and inserted into a 10-mm length of Tygon tubing affixed to a 115-mm-long glass capillary (internal diameter of 1 mm) that was half-filled with filtered, distilled water. The capillary plus tubing was inserted through a compression fitting (McCown and Wall, 1979) consisting of a brass nut with a silicone gasket that formed a tight seal around the Tygon tubing when compressed by a threaded brass adapter. For measurements on root segments with excised tips, the distal end of the root was dried, sealed with a hydrophilic dental impression material (polysiloxane), and then covered with two layers of acrylic co-polymer. The same type of seal was applied to the bases of excised lateral roots and to lateral abscission scars on nodal roots. The root segment was then suspended in a 500-cm³ tank containing aerated 0.1-strength Hoagland's solution maintained at 25 C by circulation through a coil in a water bath.

A partial vacuum (monitored with a Validyne PS309 digital manometer) was applied to the open end of the capillary, and the flow thereby induced was measured by following the progress of the capillary meniscus with a traveling microscope capable of resolving to 0.01 mm. When the volumetric flow rate of water (Q_v , m³ sec⁻¹) became constant at a given pressure (P , MPa; generally within 30 min), L_p (m sec⁻¹ MPa⁻¹) was calculated as follows (Nobel, Schulte, and North, 1990);

$$L_p = \frac{\Delta Q_v \cdot 1}{\Delta P \cdot A}$$

where A is the lateral surface area of the root segment. The projected area of the root segment was measured with a Delta-T area meter and multiplied by Π to obtain A . For five young nodal roots from the same plant under wet conditions, the standard error for the variation of L_p about its mean was 11%, similar to the variation of 12% for such measurements of L_p among six plants.

Root axial conductance—To measure root axial conductance, both ends of the root segment were trimmed under distilled water, the proximal end was sealed to the water-filled capillary, and the distal end was immersed in 10 mM oxalic acid in filtered, degassed, distilled water to reduce blockage of the vessels during measurements (Sperry, Donnelly, and Tyree, 1988). Root segments were longer than the longest vessel, as determined by forcing compressed air through comparable segments (Zimmermann and Jeje, 1981). Q_v was determined as in the L_p measurements and used to calculate the root axial conductance per unit length (K_h , $m^4 \text{ sec}^{-1} \text{ MPa}^{-1}$) as follows:

$$K_h = \frac{Q_v}{\Delta P/l}$$

where l is the length of the root segment across which the pressure drop ΔP was applied (Calkin, Gibson, and Nobel, 1986).

After K_h was measured, the root segment was placed in a 250-cm³ tank containing 10 mM oxalic acid and pressurized in a pressure chamber at 0.18 MPa for 20 min to remove embolisms (Sperry, 1986). The pressurization was repeated until K_h reached a stable maximal value (usually after two treatments), and then 0.1% crystal violet was drawn through the segment to determine which vessels were conducting. Diameters of stained and unstained vessels were measured under the light microscope and used in the Hagen-Poiseuille equation to predict theoretical maximal values of K_h (Gibson et al., 1985). At any point along a root, the volume flux density ($m^3 \text{ m}^{-2} \text{ sec}^{-1}$) into it equals the drop in water potential from the root surface to the root xylem times the local radial conductivity. To calculate root radial conductivity, L_R ($m \text{ sec}^{-1} \text{ MPa}^{-1}$), averaged over an entire root segment, values of K_h , L_p , and length and diameter of root segments were incorporated into a model of Landsberg and Fowkes (1978).

Anatomical procedures—To test for the presence of a dimorphic exodermis composed of two cell types with different structural and cytochemical properties (Shishkoff, 1987), root segments were cleared overnight in 2 M KOH, rinsed in distilled water, stained for 24 hr in the cytoplasmic stain trypan blue to distinguish between suberized and unsuberized cells (Phillips and Hayman, 1970; Shishkoff, 1987), transferred to 50% ethanol for 5 min to improve resolution, mounted in glycerine, and observed with a Zeiss microscope at $\times 600$ – $\times 1,500$, using bright-field and phase-contrast optics. Freehand sections of fresh tissue stained

for suberin with Sudan III and Sudan IV (Jensen, 1962) or for lignin and general anatomical features with 0.05% toluidine blue O were also examined microscopically.

Root segments were prepared for scanning electron microscopy (SEM) by fixation in 2.5% glutaraldehyde in 25 mM phosphate buffer at pH 7.0 (O'Brien and McCully, 1981), dehydrated in an ethanol series, critical-point dried, and sputter-coated with gold-palladium. The segments were viewed with an ETEC Autoscan microscope at $\times 30$ – $\times 3,000$ at 10 kV. Paper tracings from SEM photographs of root transverse sections were measured with a Delta-T area meter to determine the cortical area occupied by lacunae. In fresh, freehand sections stained with toluidine blue O and examined by light microscopy, the percentage of the cortical area occupied by lacunae for four young nodal roots at 7 d of drought agreed within 5% with measurements made using SEM.

RESULTS

Measurements of L_p —Root hydraulic conductivity was measured for three categories of roots from plants in drying soil (Fig. 1). Initial values of L_p were highest for young nodal roots, slightly lower for lateral roots, and about fivefold lower for older nodal roots. After 1 d of drying, L_p declined about 50% for young nodal roots and lateral roots. By 7 d, L_p had declined 87% for young nodal roots and 97% for lateral roots. During the 30 d of drying, changes in L_p for older nodal roots were not significant (Fig. 1).

The effect of rewetting on root L_p after 7 d of drying was examined for the three types of roots (Fig. 2). For young nodal roots, L_p increased to 50% of the maximal value at 2 d in well-watered soil, to 81% of maximal at 4 d, and reattained maximal value at 7 d (Fig. 2). New apical growth on the young nodal roots was observed at 3 d of rewetting and thereafter, but the rate of water uptake for four roots with the new growth excised did not differ significantly from that measured on the entire segments. For lateral roots dried in soil for 7 d, L_p was 18% of the maximum at 4 d and 21% at 7 d of rewetting (Fig. 2). For older nodal roots after 7 d of drought, rewetting had no significant effect on L_p (Fig. 2). At 7 d of rewetting after 30 d of drought ($\Psi_{\text{soil}} = -3.2$ MPa), L_p for young nodal roots increased to $21\% \pm 5\%$ ($N = 5$) of the maximal value, whereas L_p for lateral roots did not respond to rewetting, remaining at 0% of initial L_p . For older nodal roots, L_p was basically unaffected by 30 d of drying and the subsequent rewetting.

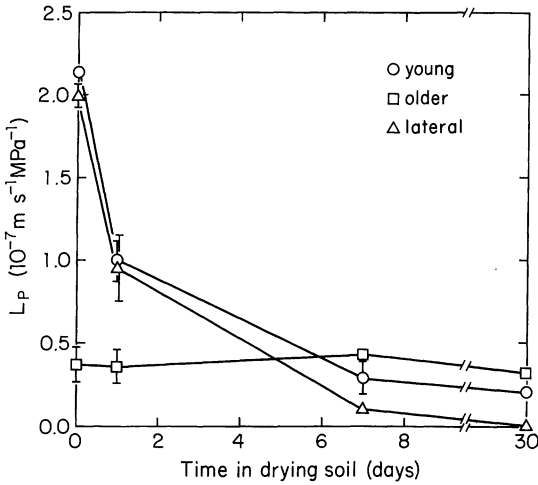


Fig. 1. Effects of duration of drying on root hydraulic conductivity (L_P) for young (4–6 wk) and older (6–12 mo) nodal roots and for lateral roots (4–6 wk) of *Agave deserti*. Ψ_{soil} was -0.5 , -1.0 , -1.3 , and -3.2 MPa at 0, 1, 7, and 30 d, respectively. Data are means \pm SE (represented by vertical bars except when smaller than the symbol) for duplicate measurements on three plants ($N = 4$ to 6).

Determination of K_h and L_R —Axial conductance per unit length was measured before (K_h^{init}) and after removal of embolisms by pressurization (K_h^{final}) for root segments in drying soil at 0, 7, and 30 d (Table 1). Under all conditions, K_h for older nodal roots was higher than for young nodal roots and lateral roots by at least two orders of magnitude, and K_h in well-watered plants was not increased by pressurization (Table 1). During drought, older nodal roots showed less difference between K_h^{init} and K_h^{final} than did young nodal roots or lateral roots. After 7 d of drought, K_h^{init}/K_h^{final} was 34% lower than under well-watered conditions for young nodal roots, 20% lower for older nodal roots, and 98% lower for lateral roots. K_h^{init}/K_h^{final} for the two types of nodal roots was not significantly different between 7 and 30 d of drought (Table 1; K_h could not be measured on laterals at 30 d of drought).

Stain drawn through root segments after pressurization to remove embolisms indicated that virtually all vessels in older nodal roots were then conducting. For these roots, the mean vessel diameter was $67 \pm 2 \mu\text{m}$ (SE for $N = 72$) and K_h^{final} (Table 1) was $97\% \pm 5\%$ ($N = 4$) of the value calculated using the diameters of stained vessels in the Hagen-Poiseuille equation. Late metaxylem vessels were consistently unstained in young nodal roots; with these vessels (mean diameter of $80 \pm 4 \mu\text{m}$; $N = 55$) excluded from calculations, K_h^{final} was $94\% \pm 9\%$ of calculated K_h based on the protoxylem

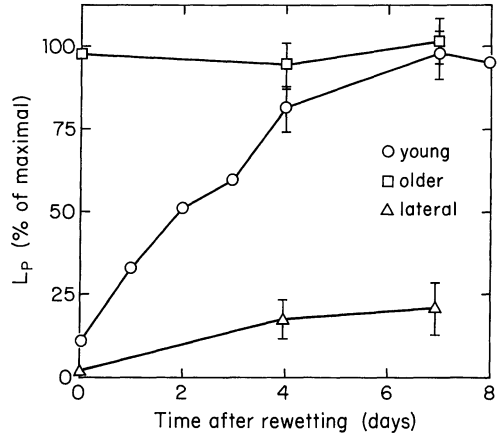


Fig. 2. Responses of L_P to rewetting after 7 d of drying. On the last day of drought, Ψ_{soil} was -1.4 MPa, after which the plants were watered and Ψ_{soil} was subsequently maintained at 0.0 MPa. At 4 and 7 d of rewetting, data are means \pm SE (two roots from each of two plants; $N = 4$). Data for remaining days are for single comparable roots from two plants. Maximal L_P was the average for two roots from each plant before drying.

and early metaxylem vessels (mean diameter of $12 \pm 1 \mu\text{m}$; $N = 90$). For lateral roots, the late metaxylem (mean diameter of $52 \pm 3 \mu\text{m}$; $N = 24$) was usually stained, and K_h^{final} was $92\% \pm 9\%$ of calculated K_h .

Root radial conductivity was calculated using K_h^{init} (to give L_R^{init}) and K_h^{final} (to give L_R^{final}) for the three root types under three conditions of soil moisture (Table 1). In all cases, L_R^{final} was not significantly different from L_R^{init} despite large differences between K_h^{init} and K_h^{final} . Similarly, at 7 d of drought, L_P^{init}/L_P^{final} indicated that embolisms decreased hydraulic conductivity by only 7% for nodal roots and 9% for lateral roots. For older nodal roots, no changes in L_R and L_P due to pressurization were observed (Table 1).

Cortical lacunae—In young nodal and lateral roots, the cortex was the tissue most visibly affected by drought (Figs. 3, 4). Lacunae caused by cellular dehydration and collapse were fairly evenly distributed throughout the cortex in all transections examined from roots that had been droughted for 7 or 30 d, with laterals showing more discontinuities than nodal roots. At 7 d of drought, lacunae represented $24\% \pm 1\%$ (SE for $N = 4$) of the transectional cortical area of young nodal roots (Fig. 3) and $60\% \pm 11\%$ ($N = 3$) of that for lateral roots (Fig. 4). By 30 d, lacunae occupied $31\% \pm 4\%$ ($N = 3$) of the cortical area of young nodal roots and $89\% \pm 4\%$ ($N = 4$) of that for lateral roots. Also, the reduction in transectional area affording an uninterrupted radial pathway across the cortex

TABLE 1. Summary of root conductance properties for *A. deserti* during drought. Axial conductance was measured before (K_h^{init}) and after pressurization (K_h^{final}) for 8-cm segments of established roots (young and older, 3 mm diameter) and lateral roots (1 mm diameter). Root radial conductivity (L_R) was calculated using mean L_p (Fig. 1), K_h^{init} , K_h^{final} , and root dimensions. Values of L_p^{init} were as in Fig. 1; L_p^{final} was calculated using K_h^{final} , L_R^{init} , and root dimensions. Ψ_{soil} was -0.5 , -1.4 , and -3.2 MPa on 0, 7, and 30 days, respectively. Data are presented as mean \pm SE ($N = 4-6$)

Duration of drought Root type	K_h^{init}	K_h^{final}	L_R^{init}	L_R^{final}	$\frac{L_p^{init}}{L_p^{final}}$
	(m ⁴ sec ⁻¹ MPa ⁻¹)		(m sec ⁻¹ MPa ⁻¹ $\times 10^{-7}$)		
0 d					
Young	$1.82 \pm 0.33 \times 10^{-12}$	$1.87 \pm 0.15 \times 10^{-12}$	18.3	17.8	0.99
Older	$7.59 \pm 1.81 \times 10^{-9}$	$7.51 \pm 1.77 \times 10^{-9}$	0.382	0.382	1.00
Lateral	$5.00 \pm 1.30 \times 10^{-12}$	$4.83 \pm 0.98 \times 10^{-12}$	2.40	2.43	1.01
7 d					
Young	$2.63 \pm 0.76 \times 10^{-12}$	$4.07 \pm 1.07 \times 10^{-12}$	0.371	0.336	0.93
Older	$7.47 \pm 1.59 \times 10^{-9}$	$9.21 \pm 1.87 \times 10^{-9}$	0.427	0.427	1.00
Lateral	$0.83 \pm 0.31 \times 10^{-12}$	$46.5 \pm 11.4 \times 10^{-12}$	0.117	0.107	0.91
30 d					
Young	$6.01 \pm 0.21 \times 10^{-12}$	$11.00 \pm 0.5 \times 10^{-12}$	0.232	0.223	0.96
Older	$10.66 \pm 2.30 \times 10^{-9}$	$12.66 \pm 2.8 \times 10^{-9}$	0.322	0.322	1.00

was calculated by adding the fractions of the cortical circumference represented by the widths of the lacunae (taking overlap of lacunae into consideration). At 7 d of drought, this fractional region unavailable for radial conduction was $49\% \pm 2\%$ ($N = 4$) of the cortical area for young nodal roots and $88\% \pm 3\%$ ($N = 4$) of that for lateral roots; at 30 d of drought, the unavailable region was $64\% \pm 5\%$ ($N = 3$) for young nodal roots and $98\% \pm 9\%$ ($N = 4$) for lateral roots.

In young nodal roots rehydrated after 7 d in drying soil, lacunae decreased appreciably in 1 d (Figs. 5, 6). Cortical lacunae in lateral roots droughted for 7 d were not markedly diminished by rewetting. In older nodal roots, the cortical area occupied by lacunae did not change in response to drying or rewetting.

Exodermis—As indicated by Sudan staining, suberization increased in the exodermis of young nodal roots and lateral roots as drying progressed. At 30 d of drought, lateral roots had one to three layers of suberized exodermal cells, whereas young nodal roots had six to ten layers, as did older nodal roots.

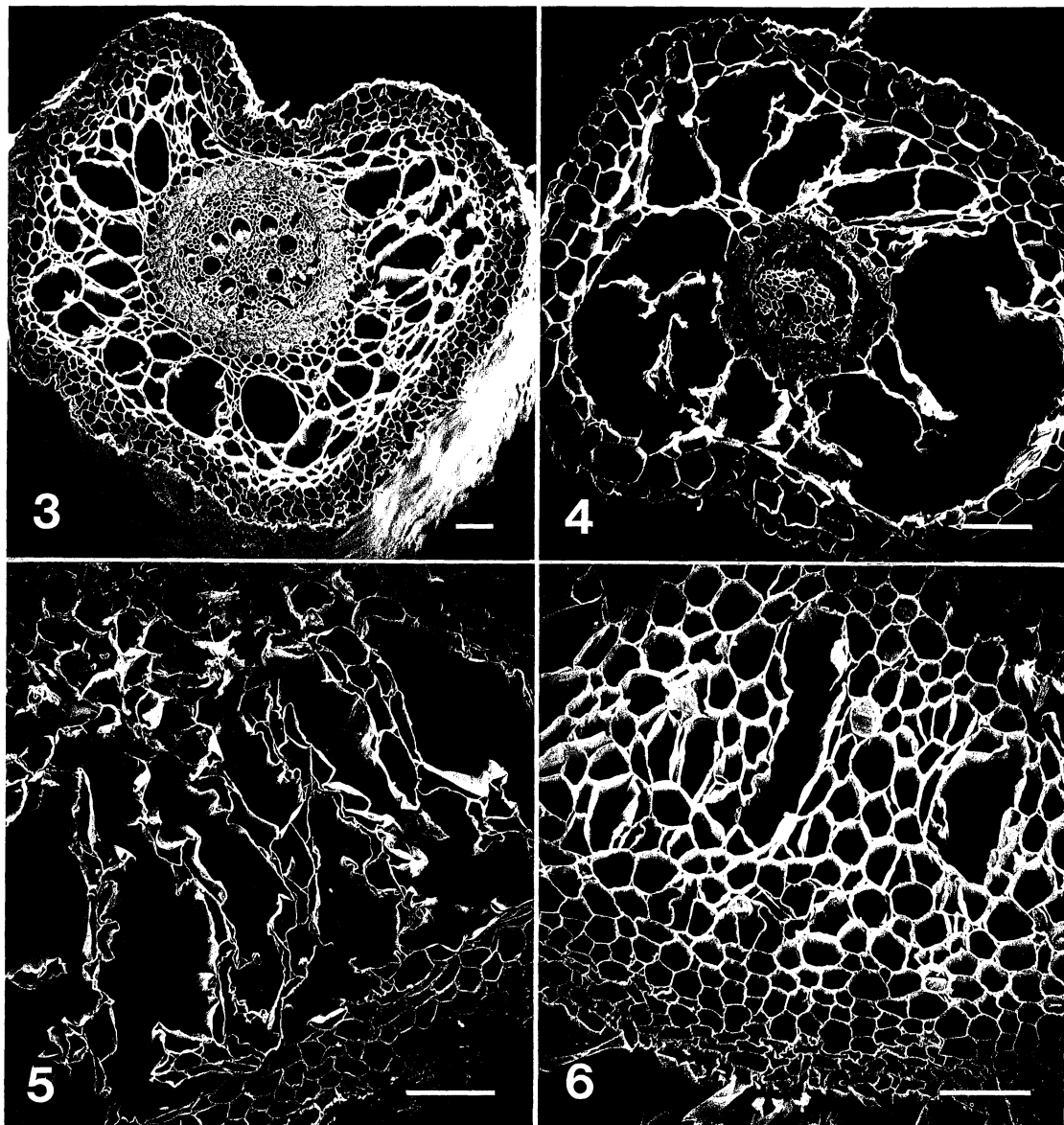
In young nodal roots and lateral roots under all soil moisture conditions, differential uptake of the cytoplasmic stain trypan blue revealed a dimorphic exodermis in a 10-mm region beginning about 20 mm from the tip. Interspersed among the more frequent, suberized, unstained cells were regularly occurring, densely cytoplasmic cells that lacked suberin lamellae in their walls and hence took up the stain (Fig. 7). In contrast, all cells in the exodermis of older roots were suberized and lignified.

Endodermis and adjacent cells—At 0 d of drought, suberin was present only in the Casparian bands of endodermal cells in the distal 15 mm of young nodal roots and lateral roots; proximal to this region, most endodermal cells developed suberized outer tangential walls. In roots of *A. deserti* subjected to drought (Fig. 8), such walls were observed in the endodermis much closer to the tip. Under all soil moisture conditions, regularly occurring passage cells (Fig. 8) lacking suberin in their tangential walls (though possessing Casparian bands) were observed in the endodermis of young nodal roots and lateral roots but not in the endodermis of older nodal roots.

Thick, suberized, lignified walls in the cortical cells adjacent to the endodermis (Fig. 8) developed closer to the tip in roots of *A. deserti* subjected to drought than in roots from well-watered plants. As was the case for the exodermis, these thickened cortical layers numbered two to three in lateral roots at 30 d of drought, whereas young nodal roots were similar to older nodal roots in having six or more such layers.

DISCUSSION

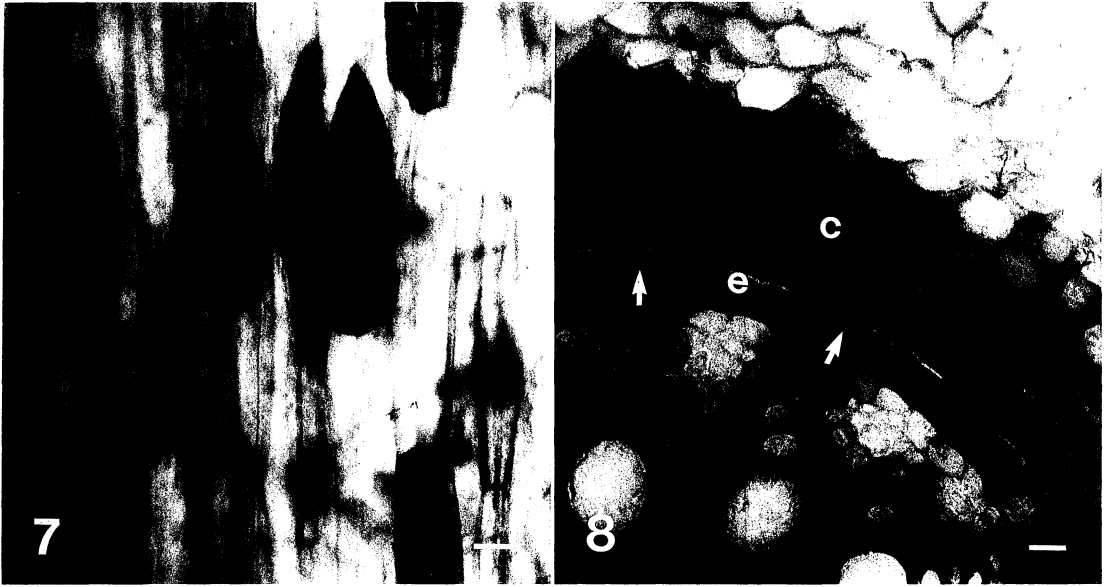
The tissue structure and hydraulic conductivity of older nodal roots of *Agave deserti* changed very little in response to 30 d of drought. For young nodal roots and lateral roots, on the other hand, drying caused marked physical and physiological changes. The decline in L_p for the latter two root types was apparently related to anatomical characteristics in which they differed from older roots, at least initially. The changes in conductivity pro-



Figs. 3–6. Scanning electron microscope photographs of transections of roots of *A. deserti*. Bars = 100 μm . 3. Young nodal root droughted for 7 d. 4. Lateral root droughted for 7 d. 5. Cortical cells in young nodal root droughted for 7 d. 6. Cortical cells in proximal segment of root depicted in Fig. 5, rehydrated for 1 d.

duced by 7 d of drought were largely permanent for lateral roots but reversible by 7 d of rewetting for young nodal roots, implying that the latter were capable of moderate and reversible rectification; that is, they restricted water flow to dry soil but readily took up water when it again became available. By 30 d of drought, the convergence of L_p values for young nodal roots and older nodal roots was accompanied by their increasing similarity with respect to both cortical lacunae and number of suberized layers.

The cortex was the tissue most affected by drought for roots of *A. deserti*, as has also been observed for barley (Clarkson, Sanderson, and Russell, 1968) and perennial rye grass (Jupp and Newman, 1987). Air-filled lacunae developed in the cortex of young nodal roots and lateral roots during drought and were present in the cortex of the previously droughted older nodal roots. Whether water flows across the cortex predominantly apoplastically or symplastically, its radial movement would be impeded by these cortical lacunae, thereby caus-



Figs. 7, 8. Light microscope photographs of young nodal roots of *A. deserti* subjected to 1 d of drought. Bars = 10 μm . 7. Longitudinal section of exodermis, stained with trypan blue; dark cells are unsuberized. 8. Fresh, free-hand transection stained with toluidine blue O; e = endodermal cells, with suberized (unstained) outer tangential walls; c = adjacent cortical cells with thickened, suberized, lignified walls; arrows point to endodermal passage cells.

ing L_p to decline. The decreases in L_p in response to drought corresponded to increases in cortical area occupied by lacunae. The cortex of lateral roots was affected more severely by drought than that of young nodal roots, and that of older nodal roots was essentially unaffected. After 8 d of drought, Ψ_{soil} was -1.8 MPa, close to the value of -2.0 MPa that caused 50% of the cortical cells to die in roots of rye grass (Jupp and Newman, 1987). Cell death for *A. deserti* apparently occurred in the cortex of lateral roots after 7 d; the limited increase in L_p after rewetting (to 21% of maximal L_p , as opposed to nearly 100% for nodal roots) is consistent with the observation that more than half of the cortical area was taken up by lacunae.

The limited recovery of lateral roots in response to rewetting, the full recovery of L_p values accompanying the reduction of lacunae in rehydrated young nodal roots, and the relative constancy of L_p and cortical area in older nodal roots all suggest that cortical discontinuities are involved in the variability of L_p . However, the effect of cortical lacunae on L_p may be relatively small, particularly for young nodal roots. If decreasing the cortical area by 24% (the area occupied by lacunae in young nodal roots at 7 d of drought) were to decrease radial conductivity (L_R) by the same amount, L_p for young nodal roots would decline by only 12% instead of the 87% decrease measured after 7 d of drought. Even diminishing L_R by

49%, the amount of cortical area unavailable for water conduction along uninterrupted radial paths, would diminish L_p by only 29%, one-third of the measured decrease in L_p . Moreover, the 50-fold decrease in L_R calculated for young nodal roots after 7 d of drought is inconsistent with either a 24% reduction in overall cortical area or a 49% decrease in the amount of cortex affording a direct radial pathway for the movement of water.

The exodermis and endodermis are more likely than the undifferentiated cortex to restrict water flow, at least after extensive suberization has occurred in these two regions (Perumalla and Peterson, 1985; Clarkson et al., 1987). Although the thick-walled, suberized cells in these regions may permit some apoplastic flow of water (Sanderson, 1983), the thin-walled cells of the cortex (from which the suberized cells differentiate) would offer much less resistance to apoplastic flow (Jones et al., 1988). In addition, suberin lamellae and lignin in the thickened walls of the exodermis, endodermis, and adjacent cortical layers may seal off plasmodesmata (Robards, Clarkson, and Sanderson, 1979; Walker et al., 1984), thereby directly limiting symplastic flow.

In young nodal roots and lateral roots of *A. deserti* subjected to drought, suberization close to the tip and the proliferation of suberized, lignified cell layers in the exodermis and in the cortex adjacent to the endodermis coincided with decreases in L_p and L_R . Young nodal roots

had two to three times as many thickened layers as did lateral roots at 7 d of drought; consistent with this, L_R decreased 50-fold for young nodal roots and 20-fold for lateral roots over 7 d of drought, suggesting a link between diminished radial conductivity and additional suberized layers. Furthermore, the low L_P for older nodal roots compared with values for young nodal roots and lateral roots under well-watered conditions may reflect the extensive suberization in older roots that had occurred during previous drought episodes.

The properties of suberin lamellae do not explain the rectifierlike behavior of roots, because the reduction in conductivity caused by their presence in a tissue is not reversible by rewetting. Suberized tissue that has been kept continuously moist may be ten to 50 times more permeable to water than after it has been exposed to air, but its permeability is not restored when the tissue is remoistened (Vogt, Schönherr, and Schmidt, 1983; Clarkson et al., 1987). This is consistent with the behavior of older nodal roots of *A. deserti* that, having previously experienced one or more drought episodes, showed no increase in L_P after rewetting.

The recovery in L_P for droughted young nodal roots in response to rewetting was similar to that for roots of *A. deserti* rehydrated in solution after drying in air (Nobel and Sanderson, 1984). Some of this recovery could have been due to new apical growth, but measurable increase in root length was not seen before the third day after rewetting. Moreover, excising the new root tips had no effect on water uptake, presumably because of the immaturity of the vascular tissue. Before new growth occurred, the restoration of conductivity may have been partially due to the properties of the unsuberized cells that alternate with suberized cells to produce a dimorphic exodermis (Shishkoff, 1987). Regularly occurring unsuberized cells were seen near the tips of young roots in a 10-mm region where the exodermis is one to two cell layers thick. These unsuberized cells could function as do the passage cells described for barley and other species (Esau, 1977) in facilitating water movement across the otherwise fairly impermeable exodermis.

In *Hoya carnosa*, unsuberized exodermal cells show radial channels bordered by lignified thickenings in their tangential walls, implying potential valvelike activity if the channel configurations change with hydration (Öleson, 1978). The frequency of the unsuberized cells in the exodermis of younger roots of *A. deserti* is such that changes in their permeability to water would contribute substantially to vari-

ability in L_P . In addition, the presence of these cells in the exodermis of young nodal roots that increase in L_P after rewetting and their absence in older roots that show no such recovery suggest a possible role of the dimorphic exodermis in rectification. Although no such valvelike activity has been attributed to the passage cells in the endodermis of other species, their restriction to young roots in *A. deserti* suggests that they also may facilitate water uptake when the soil is rewetted.

Lateral roots of *A. deserti*, which have fewer cell layers outside the stele than do nodal roots, were also more susceptible to embolism than were nodal roots. The almost total loss of axial conductance ($K_h^{\text{init}}/K_h^{\text{final}} = 0.02$) for lateral roots at 7 d of drought indicated a level of embolism comparable to that measured in sugar maple twigs after winter freezing (Sperry, Donnelly, and Tyree, 1988). The 20% loss of axial conductance for older nodal roots at 30 d of drought is similar to values obtained for stems of several woody species, and the 45% loss of conductance for young nodal roots suggests that they were more vulnerable to embolisms than are most woody stems subjected to comparable water stress (Tyree and Sperry, 1989). Embolism may be less serious for roots than for stems, because rewetting of the soil and the subsequent reestablishment of root pressure could quickly restore axial flow (Milburn, 1979), particularly for the comparatively small vertical distances involved.

Changes in K_h attributable to embolism might be involved in root rectification; however, reductions in K_h had a relatively small effect on L_P for roots of *A. deserti*. For undroughted young nodal roots, immature vessels led to a low K_h that limited L_P , as L_R^{init} then exceeded L_P by ninefold. However, L_P for young nodal roots at 7 d of drought was decreased by only 7% when calculated using embolized as opposed to unembolized values of K_h ; even though K_h was reduced 98% by embolism for lateral roots droughted for 7 d, the calculated decrease in L_P was only 9%, indicating that L_R , not K_h , was limiting overall conductivity.

Increases in cortical lacunae, in exodermal, endodermal, and cortical suberization, and in embolism all contributed to the tenfold decline in L_P for young nodal roots of *A. deserti*. At 30 d of drought, young nodal roots differed from older nodal roots chiefly in having fewer functional late metaxylem vessels; with respect to the radial pathway for water uptake, the two types of root converged in both structure and conductivity. Young nodal roots droughted for 7 d and subsequently rewatered behaved as

rectifiers, although less so than documented earlier for *A. deserti* (Nobel and Sanderson, 1984) with regard to both decline in L_p and rate of recovery after rewetting.

A large discrepancy exists between the decreases in L_p reported here and the predicted 10^5 -fold decrease in root system conductivity for *A. deserti* that prevents plant water loss to the soil during drought (Schulte and Nobel, 1989). Anatomical changes within individual roots, at least at the tissue level, could not account for such a large decline in L_p . Conductivity of the root system as a whole could decrease by as much as 30% with the abscission of lateral roots, as occurs in *A. deserti* in response to drought (Franco and Nobel, 1990). In addition, though embolism in the individual roots may not greatly reduce L_p , embolism in the root-shoot transition zone may be more important in terms of hydraulic architecture (Luxová, 1989). Furthermore, decreases in L_p in very dry soils may owe less to changes in the roots than to reduced contact between roots and soil, such as might result from root shrinkage (Faiz and Weatherly, 1982). Information on drought-induced structural changes in roots of *A. deserti*, when combined with measurements of L_p on roots in soil, should help assess this last possibility.

LITERATURE CITED

- BLIZZARD, W. E., AND J. S. BOYER. 1980. Comparative resistance of the soil and the plant to water transport. *Plant Physiology* 66: 809-814.
- CALKIN, H. W., A. C. GIBSON, AND P. S. NOBEL. 1986. Biophysical model of xylem conductance in tracheids of the fern *Pteris vittata*. *Journal of Experimental Botany* 37: 1054-1064.
- CLARKSON, D. T., AND A. W. ROBARDS. 1975. The endodermis, its structural development and physiological role. In J. J. Torrey and D. T. Clarkson [eds.], *The development and function of roots*, 415-436. Academic Press, London.
- , J. E. STEPHENS, AND M. STARK. 1987. Suberin lamellae in the hypodermis of maize (*Zea mays*) roots; development and factors affecting the permeability of hypodermal layers. *Plant, Cell and Environment* 10: 83-93.
- , J. SANDERSON, AND R. S. RUSSELL. 1968. Ion uptake and root age. *Nature* 220: 805-806.
- DIRKSEN, C., AND P. A. C. RAATS. 1985. Water uptake and release by alfalfa roots. *Agronomy Journal* 77: 621-626.
- DREW, M. C. 1979. Root development and activities. In D. W. Goodall, R. A. Perry, and K. M. W. Howes [eds.], *Arid-land ecosystems: structure, functioning and management*, vol. 1, 573-606. Cambridge University Press, Cambridge.
- ESAU, K. 1977. *Anatomy of seed plants*. Wiley and Sons, New York.
- FAIZ, S. M. A., AND P. E. WEATHERLY. 1982. Root contraction in transpiring plants. *New Phytologist* 92: 333-343.
- FRANCO, A. C., AND P. S. NOBEL. 1990. Influences of root distribution and growth on predicted water uptake and interspecific competition. *Oecologia* 82: 151-157.
- GIBSON, A. C., H. W. CALKIN, D. O. RAPHAEL, AND P. S. NOBEL. 1985. Water relations and xylem anatomy of ferns. *Proceedings of the Royal Society of Edinburgh* 86B: 81-92.
- JENSEN, W. A. 1962. *Botanical histochemistry: principles and practice*. W. H. Freeman, San Francisco.
- JONES, H., R. A. LEIGH, R. G. WYN JONES, AND A. D. TOMOS. 1988. The integration of whole root and cellular hydraulic conductivities in cereal roots. *Planta* 174: 1-7.
- JUPP, A. P., AND E. I. NEWMAN. 1987. Morphological and anatomical effects of severe drought on the roots of *Lolium perenne* L. *New Phytologist* 105: 393-402.
- LANDSBERG, J. J., AND N. D. FOWKES. 1978. Water movement through plant roots. *Annals of Botany* 42:493-508.
- LUXOVÁ, M. 1989. The vascular system in the roots of barley and its hydraulic aspects. In B. C. Loughman, O. Gasparíková, and J. Kolek [eds.], *Structural and functional aspects of transport in roots*, 15-20. Kluwer Academic Publishers, Dordrecht.
- MCCOWN, R. L., AND B. H. WALL. 1979. Improvement of the pressure chamber measurements of two legumes by constriction of stems. *Plant and Soil* 51: 447-451.
- MCCULLY, M. E., AND M. J. CANNY. 1988. Pathways and processes of water and nutrient movement in roots. *Plant and Soil* 111: 159-170.
- MILBURN, J. A. 1979. *Water flow in plants*. Longman, New York.
- MOONEY, H. A., S. L. GULMON, P. W. RUNDEL, AND J. EHLERINGER. 1980. Further observations on the water relations of *Prosopis tamarugo* of the northern Atacama Desert. *Oecologia* 44: 177-180.
- NOBEL, P. S. 1991. *Physicochemical and environmental plant physiology*. Academic Press, San Diego.
- , AND J. SANDERSON. 1984. Rectifier-like activities of roots of two desert succulents. *Journal of Experimental Botany* 35: 727-737.
- , P. J. SCHULTE, AND G. B. NORTH. 1990. Water influx characteristics and hydraulic conductivity for roots of *Agave deserti* Engelm. *Journal of Experimental Botany* 41: 409-415.
- O'BRIEN, T. P., AND M. E. MCCULLY. 1981. *The study of plant structure: principles and selected methods*. Termarcaphi Pty Ltd., Melbourne.
- ÖLESON, P. 1978. Studies on the physiological sheaths in roots I. Ultrastructure of the exodermis in *Hoya carnosa* L. *Protoplasma* 94: 325-340.
- PALTA, J. A., AND P. S. NOBEL. 1989. Root respiration for *Agave deserti*: influence of temperature, water status and root age on daily patterns. *Journal of Experimental Botany* 40: 181-185.
- PASSIOURA, J. B. 1988. Water transport in and to roots. *Annual Review of Plant Physiology and Plant Molecular Biology* 39: 245-265.
- PERUMALLA, C. J., AND C. A. PETERSON. 1985. Deposition of Casparian bands and suberin lamellae in the exodermis and endodermis of young corn and onion roots. *Canadian Journal of Botany* 64: 1873-1878.
- PHILLIPS, J. M., AND D. S. HAYMAN. 1970. Improved procedures for clearing roots and staining parasites and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Transactions of the British Mycological Society* 55: 158-161.
- RAMOS, C., AND M. R. KAUFMANN. 1979. Hydraulic re-

- sistance of rough lemon roots. *Physiologia Plantarum* 45: 311–314.
- RICHARDS, J. H., AND M. M. CALDWELL. 1987. Hydraulic lift: substantial nocturnal water transport between soil layers by *Artemisia tridentata* roots. *Oecologia* 73: 486–489.
- ROBARDS, A. W., D. T. CLARKSON, AND J. SANDERSON. 1979. Structure and permeability of the epidermal, hypodermal layers of the sand sedge (*Carex arenaria*, L.). *Protoplasma* 101: 331–347.
- SANDERSON, J. 1983. Water uptake by different regions of the barley root. Pathways of radial flow in relation to the development of the endodermis. *Journal of Experimental Botany* 34: 240–253.
- , F. C. WHITBREAD, AND D. T. CLARKSON. 1988. Persistent xylem cross-walls reduce the axial hydraulic conductivity in the apical 20 cm of barley seminal root axes: implications for the driving force for water movement. *Plant, Cell and Environment* 11: 247–256.
- SCHULTE, P. J., AND P. S. NOBEL. 1989. Responses of a CAM plant to drought and rainfall: capacitance and osmotic pressure influences on water movement. *Journal of Experimental Botany* 40: 61–70.
- SHISHKOFF, N. 1987. Distribution of the dimorphic hypodermis of roots in angiosperm families. *Annals of Botany* 60: 1–15.
- SHONE, M. G. T., AND D. T. CLARKSON. 1988. Rectification of radial water flow in the hypodermis of nodal roots of *Zea mays*. *Plant and Soil* 111: 223–229.
- , AND A. V. FLOOD. 1980. Studies on uptake and loss of water by barley roots in relation to changes in root resistance. *Journal of Experimental Botany* 31: 1147–1159.
- SPERRY, J. S. 1986. Relationship of xylem embolism to xylem pressure potential, stomatal closure and shoot morphology in the palm *Rhapis excelsa*. *Plant Physiology* 80: 110–116.
- , J. R. DONNELLY, AND M. T. TYREE. 1988. Seasonal occurrence of xylem embolism in sugar maple (*Acer saccharum*). *American Journal of Botany* 75: 1212–1218.
- TYREE, M. T., AND J. S. SPERRY. 1989. Vulnerability of xylem to cavitation and embolism. *Annual Review of Plant Physiology and Plant Molecular Biology* 40: 19–38.
- VOGT, E., J. SCHÖNHERR, AND H. W. SCHMIDT. 1983. Water permeability of periderm membranes isolated enzymatically from potato tubers (*Solanum tuberosum* L.). *Planta* 158: 294–301.
- WALKER, R. R., M. SEDGLEY, M. A. BLESING, AND T. J. DOUGLAS. 1984. Anatomy, ultrastructure and assimilate concentrations of roots of citrus genotypes differing in ability for salt exclusion. *Journal of Experimental Botany* 35: 1481–1494.
- YOUNG, D. R., AND P. S. NOBEL. 1986. Predictions of soil-water potentials in the north-western Sonoran Desert. *Journal of Ecology* 74: 143–154.
- ZIMMERMANN, M. H., AND A. A. JEJE. 1981. Vessel-length distribution in stems of some American woody plants. *Canadian Journal of Botany* 59: 1882–1892.