Effect of restricted watering on sap flow and growth in corn (*Zea mays* L.)

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Gavloski, J. E., Whitfield, G. H. and Ellis, C. R. 1992. **Effect of restricted watering on sap flow and growth in corn (*Zea mays* L.).** Can. J. Plant Sci. **72**: 361–368. The heat balance technique for measuring sap flow was used to determine how plant stress from watering various proportions of the root system in corn (*Zea mays* L.) affects sap flow and root and shoot growth. Sectional root-boxes were used to divide the root system into four equal compartments so that known proportions of the root system could be subjected to water stress. Results indicated that the root-box technique is useful in studying the effects of adverse growing conditions in corn. Treatments consisted of no watering and watering one, two, three, or four sections of the box. Sap flow was measured using gauges that worked on a heat balance principle, and aspects of root and shoot growth were also measured. Withholding water from two or more sections of the box for 26 d resulted in decreased sap flow and fresh and dry weight of stalks compared with plants where all four sections were watered (control). Plant height was lower in boxes where one or more sections were deprived of water compared with the control. Dry weight of roots was less when water was withheld from three or all sections of the roots, and fresh weight of roots was less when water was withheld from all four sections. Corn plants with even half the roots growing under stressed conditions resulted in decreased sap flow and shoot growth.

Key words: Root-box, moisture stress

Gavloski, J. E., Whitfield, G. H. et Ellis, C. R. 1992. **Effet du rationnement de l'eau sur la circulation de la sève et sur la croissance du maïs (*Zea mays* L.).** Can. J. Plant Sci. **72**: 361–368. On a eu recours à la technique du bilan thermique pour déterminer dans quelle mesure un retrait de l'approvisionnement d'eau dans diverses fractions du système racinaire peut nuire à la circulation de la sève et à la croissance des racines et des parties aériennes du maïs. Des enceintes compartimentées permettaient de diviser le système racinaire en quatre quartiers égaux et ainsi d'infliger un stress hydrique à un ou plusieurs quartiers de la racine. Les résultats montrent que cette technique est utile pour étudier les effets des conditions de végétation adverses sur le maïs. Il s'agissait de priver d'arrosage un, deux, trois ou les quatre segments des racines. La circulation de la sève était mesurée par des sondes fonctionnant selon le principe du bilan thermique et on suivait aussi divers aspects de la croissance des racines et des parties aériennes. L'absence d'arrosage pendant 26 jours dans deux quartiers du système racinaire ou d'avantage a entraîné une diminution de la circulation de la sève ainsi que du poids frais et sec des tiges, par rapport aux plantes témoins (arrosage intégral). Les plantes étaient moins hautes dans les enceintes dont un ou plusieurs quartiers étaient privés d'eau. L'absence d'approvisionnement hydrique dans trois ou quatre quartiers abaissait le poids sec de la racine, tandis que le retrait total des arrosages abaissait également le poids frais. La privation d'eau dans seulement la moitié du système racinaire était suffisante pour réduire la circulation de la sève, ainsi que la croissance des parties aériennes.

Mots clés: Enceintes racinaires, stress hydrique

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Until recently, most measurements of sap flow in the xylem of plants were based on a heat pulse technique. This technique involves measuring the time required for a discrete heat input to travel from its source to a sensor further up the stem (Bloodworth et al. 1955). A technique for measuring sap flow based on a stem heat balance method has been described by Sakuratani (1981, 1984) and further developed by Baker and van Bavel (1987) and Steinberg et al. (1990). This technique involves heating an insulated section of stem at a constant rate and measuring the heat fluxes in the radial and vertical directions. The heating and measurements are done with a gauge attached to the outside of the stem. The gauges have been shown to be useful for studying sap flow in stems of herbaceous plants (Baker and van Bavel 1987) and therefore may also be applicable for research on the effects of water stress in corn.

Tan et al. (1981) studied water relations in tomato (*Lycopersicon esculentum* Mill.) by transplanting plants into sectional root-boxes that divided the root system into four compartments. This technique has wider applications for studying the relationship between the proportion of roots growing under stressed conditions and rates of water uptake. For instance, it could be used to estimate the effect of damage or loss of a certain proportion of the root system due to insects or diseases. The decision to apply an insecticide to control corn rootworms (*Diabrotica* spp.) is frequently based on a root rating system that takes into account the amount of roots damaged or destroyed (Hills and Peters 1971). Rootworm damage to corn roots is thought to affect water and nutrient uptake (Kahler et al. 1985). It is thus important to know how plant growth and physiology of corn are affected when various proportions of its root system are damaged or stressed.

The objectives of this research were to use the heat balance technique of Baker and van Bavel (1987) to determine how plant stress from watering various proportions of the root system of corn affects sap flow and root and shoot growth.

MATERIALS AND METHODS

All experiments were conducted in a greenhouse at the Agriculture Canada Research Station in Harrow, Ontario, in the spring of 1990. The greenhouse was maintained at 25–28°C during the day and not less than 17°C during night hours. One corn seed ('Pioneer' corn hybrid 3707) was planted in each of twenty 12.7-cm diam (5-inch STD) pots containing a sandy soil. Plants were fertilized 14 d after planting with 1 g of Peters general purpose (20-20-20)N-P-K water-soluble fertilizer (J. R. Johnson, St. Paul, MN).

Root-boxes consisting of four compartments, each 20.3 × 15.2 × 40.6 cm deep, were prepared as described by Tan et al. (1981). Plastic bags (82.7 × 36 cm), used to line the root-boxes, were cut to a length of 60 cm, and five holes (1.0-cm diam) were made with a cork borer about 2 cm from the bottom and 2–3 cm apart on each side of the bag. One bag was placed in each section of the box, and the holes were aligned along the bottom of the box to allow for drainage. Plastic flaps above the top of each section were folded over the top edges of the wood, and bags were stapled in place. The purpose of the plastic bags was to prevent warping of the wood by water and to allow soil and roots to be removed easily from each section for assessment at the completion of the experiment.

About 2.5 cm of coarse gravel was put in each section of the root-boxes, and they were filled to within 5 cm of the top with a steam-sterilized Brookston clay soil (15.1% sand, 46.0% silt, and 38.8% clay) 3 d before transplanting. Soil was watered each day for 3 d to allow for a slightly moist clay for transplanting.

A single corn plant was transplanted into each of the sectional root-boxes at 29 d of growth (3–4 leaf stage). All sections of the root-boxes were watered immediately after transplanting, and plants were fertilized with 1.5 g plant^{-1} of Peters general purpose (20-20-20) N-P-K fertilizer. Plants that appeared weak due to transplanting were staked for additional support for 2 d.

Soil moisture was maintained at or near field capacity by adding water daily to the drip point at the drainage holes. Sixteen days after transplanting, plants were divided into five treatments with two replications per treatment. Treatments consisted of no watering and watering one, two, three, or four sections of the box. Those sections still receiving water were maintained at or near field capacity by watering daily. Sections deprived of water were not watered for the remainder of the experiment. The experiment was repeated to obtain four replicates of each treatment.

Plant height was measured 1 d before treatment and 7, 14, and 25 d post-treatment by selecting the leaf that could be extended to the greatest height and measuring from soil level to the tip of this leaf. To measure sap flow, 19-mm diam sap-flow gauges (SGB19-WS by Dynagage™, Dynamax Inc., Houston, TX) were attached to a portion of the lower stalk of eight of the plants 11 d post-treatment. Lower leaves (V4 and V5) were removed from the plants so that the gauges would be in direct contact with the stalk. An electrical insulating compound (Dow-Corning compound 4) was applied to the area of the lower stem which would be covered by the gauges and to the inside of the gauges. As recommended by the manufacturer, the insulating compound was also used to seal any gaps between the plant and upper and lower edges of the gauges. Wire leads attached to the gauges were connected to an AM16 multiplexer (Campbell Scientific Corp., Logan, UT), which was connected to a CR21X (Campbell Scientific Corp., Logan, UT) datalogger. Programming software for the multiplexer was supplied by Dynamax Inc. (MU21XF). The gauges were supplied with power by an external power source (Elanco Electronics Inc., Wheeling, IL, model XP750) calibrated to deliver 3 V.

Constant values for heater resistance, stem diameter, and thermocouple gap were entered into the datalogger as required for correct operation of the gauges. Heater resistance (ohms) and stem diameter constants were specific to each gauge and were supplied by the manufacturer. Gauges were left to operate overnight, and the minimum predawn K_{sh} value (zero sap flow where radial heat transfer is related to signal from themopile; see Baker and van Bavel 1987) was entered for each gauge. The rate of sap flow was recorded every 30 min until completion of the experiment. Since only eight gauges could be operated at one time, sap flow was measured in only one plant from treatments with two or three sections of the roots watered each time the experiment was repeated.

Gauges were removed 26 d post-treatment (V14-V15 stage of growth), and plants were cut at ground level to separate roots and shoot growth. Two leaves immediately below the top ear, and one leaf above the top ear, were removed to record leaf area using a leaf area meter (model 3100, LI-COR, Inc., Lincoln, NE). The stalks were split lengthwise, and the length of each internode (up to 14th node) was measured. Stalks were cut into 10- to 15-cm long sections and weighed to obtain fresh weight.

Soil was carefully searched by hand to recover all roots. Roots were soaked overnight in water, washed, and weighed. Roots and stalks were dried at 60°C, and dry weight was recorded after 6 d.

Data Analysis

The two periods of time the experiment was performed were treated as blocks, and data were combined to provide a total of four replications.

Sap flow was analyzed as a completely randomized block design (Statistical Analysis System Institute, Inc. (SAS) 1987) for each of the days post-treatment. Intervals of 30 min, from 12:00 to 15:00 h, were used as the time component because this interval represented the time when sap flow was greatest and when the maximum differences between treatments could be expected. Treatment means for sap flow (g h^{-1}) were compared using Tukey's HSD test.

Plant height, leaf area, and fresh and dry stalk and root weight were analyzed by analysis of variance (SAS 1987). Treatment means were compared using Tukey's HSD test.

RESULTS

A representative example of sap flow during the day is presented for a plant where all root-box sections were watered and for a plant where no sections were watered for 25 days after restricted watering treatments were initiated (Fig. 1). A comparison of the plants reveals that sap flow was greatly reduced when no watering took place compared with when all the sections were watered. The diurnal pattern of sap flow, however, was similar, beginning ca. 06:00 h and increasing to a first peak for both plants ca. 10:00 h and later to a second peak for the watered plant at 15:00 h. For both plants, sap flow decreased after 15:00 h and ceased after 20:00 h. Sap flow in general fluctuated during the day, with a maximum oscillation of ca. 50 g h^{-1}.

For all treatments, greater sap flow occurred after 22 d than after 18 d (Fig. 2). Factors other than length of time without water, such as light intensity, relative humidity, and increased plant size, are likely responsible for this increase in sap flow. However, the effect of restricted watering on sap flow observed after 18 d was still evident after 22 d.

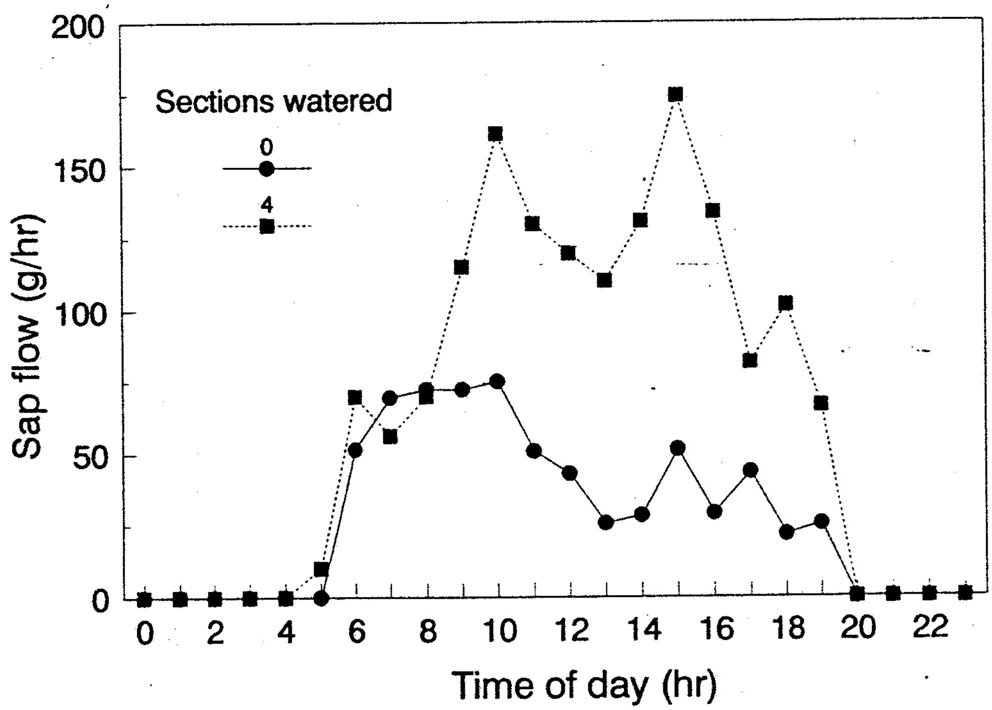

Fig. 1. Hourly sap flow for corn grown in sectional root-boxes 25 days after restricted watering treatments were initiated.

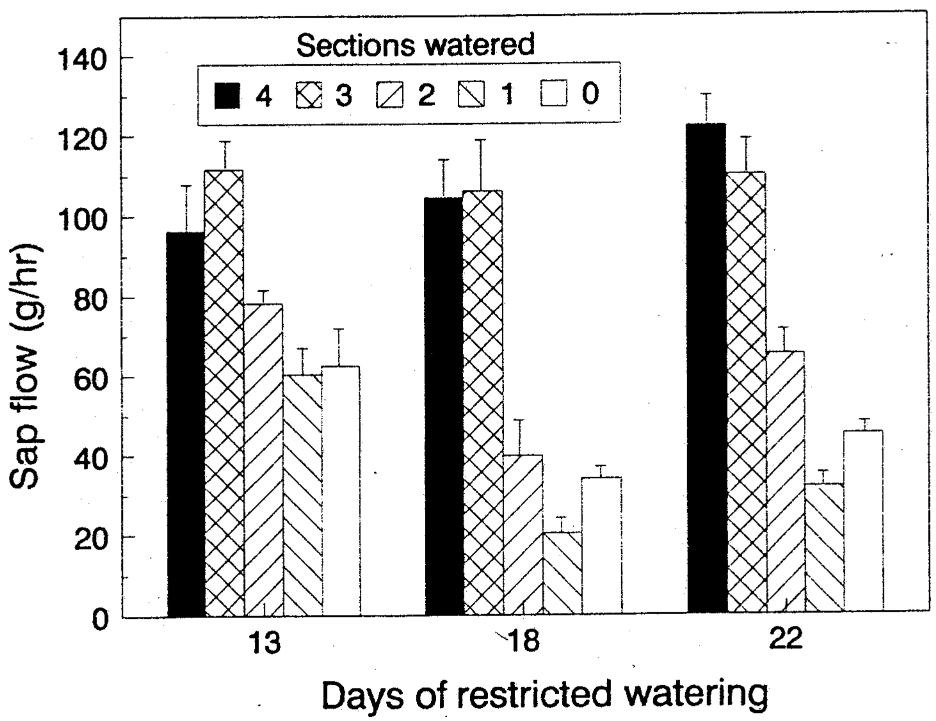

Fig. 2. Sap flow (mean ± SE) in corn grown in sectional root-boxes where various proportions of the roots received water. For 2 and 3 sections watered, $n = 2$, and for 0, 1 and 4 sections watered, $n = 4$.

Withholding moisture from one section of the box had no effect on sap flow in corn. When water was withheld from two or more sections of the box, sap flow decreased ($P > 0.001$) after 18 d (Table 1). After 22 d sap flow was less $P < 0.003$) for those plants

Table 1. Effect of supplying water to various proportions of the root system of corn on sap flow, dry and fresh weight of stalks, percent water in stalks, leaf area, and the ratio of dry weight of stalks to dry weight of roots (S_T/R)

Sections watered[z]	Sap flow (g/h) various days post-treatment			Dry stalk weight (g)	Fresh stalk weight (g)	% water in stalks	Leaf area[y] (cm^2)	S_T/R[x]
	13	18	22					
4	96*a*	105*a*	122*a*	91*a*	646*a*	86	2295*ab*	4.1
3[w]	112*a*	106*a*	110*ab*	82*a*	563*a*	85	2397*a*	3.5
2[w]	78*a*	40*b*	65*abc*	66*b*	441*b*	85	2278*ab*	3.0
1	60*a*	20*b*	32*c*	49*c*	377*b*	87	2236*ab*	2.9
0	63*a*	34*b*	45*bc*	22*d*	165*c*	87	1776*b*	2.5

a–d Means within a column followed by the same letter are not significantly different at $P>0.05$, using Tukey's studentized range (HSD) test (Statistical Analysis System Institute, Inc. 1987).
[z] Corn seedlings were transplanted into boxes divided into four sections. Treatments consisted of only watering a certain number of sections after 45 d of growth.
[y] Two leaves immediately below the top ear and one leaf above the top ear were used for leaf area measurements.
[x] Dry weight of roots shown in Table 2.
[w] Sap flow was measured on only two replications; all other measurements and treatments had four replications.

where 0 or 1 section was watered compared with plants where two or more sections were watered.

Withholding moisture from two or more sections of the box resulted in reduced fresh ($P < 0.001$) and dry ($P < 0.001$) weights of stalks. Leaf area, however, was not affected until moisture was withheld from the entire root system ($P < 0.05$). The percent water in the stalk was the same for all treatments (Table 1).

A ratio of dry weight of stalks to dry weight of roots (S_T/R) was calculated instead of a ratio of shoot weight to root weight because the bottom leaves of the corn plants were removed to attach the sap-flow gauges. S_T/R decreased as the percentage of roots receiving moisture decreased (Table 1).

Depriving the roots of water for 7 d resulted in decreased growth of plants as shown by lower plant height ($P < 0.05$) for plants with all four sections deprived of water (Fig. 3). After 14 d of restricted watering, plant height was less ($P < 0.001$) in boxes where three or four sections were deprived of water compared with plants where all four sections were watered (control). Depriving one or more sections of water for 25 d resulted in lower ($P < 0.001$) plant height compared with the control. Length of the upper internodes (7–13) increased as the number of root-box sections receiving moisture increased (Fig. 4). Thus the smaller length of the upper internodes in plants with restricted watering treatments appears to be responsible for the lower height of the plants compared with the controls.

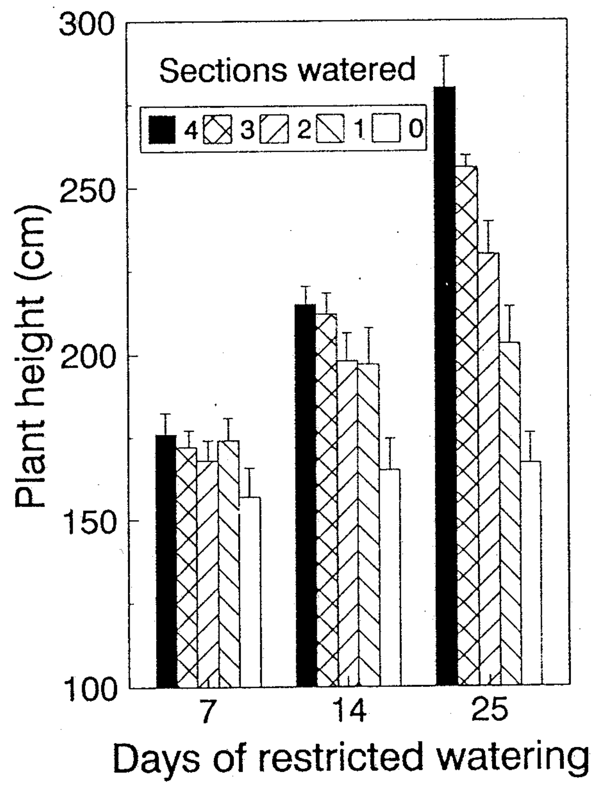

Fig. 3. Height of corn plants grown in sectional root-boxes with various proportions of the root system receiving water over a 26-d period. $n = 4$.

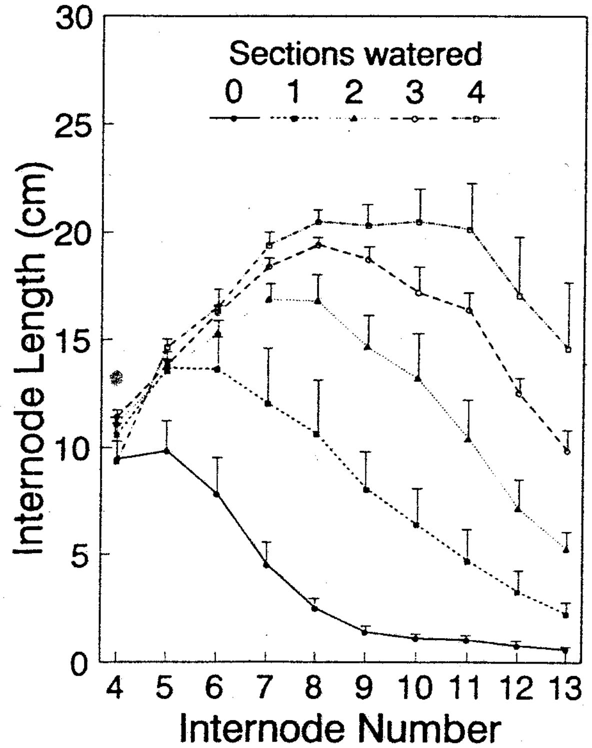

Fig. 4. Internode length of corn grown in sectional root-boxes with various proportions of the roots receiving water over a 26-d period. $n = 4$.

Withholding water from 75 or 100% of the roots resulted in less dry weight of roots ($P < 0.001$) compared with that of the controls (Table 2). Fresh weight of roots was less ($P < 0.001$) when all the roots (four sections) were deprived of water. The percent water in the roots was lower in boxes with three or four sections deprived of water than in the controls.

DISCUSSION

The hourly fluctuations in sap flow in corn observed in this study were also observed by Cohen et al. (1987), who reported that sap flux in corn was strongly fluctuating. In contrast with their results, however, we did not observe that fluctuations became significantly larger as the soil dried. Cohen et al. (1987) suggested that because their technique for measuring sap flow required the insertion of a thermocouple into the plant, sap flow may have been detected only in a limited number of conducting bundles and that variation in sap velocity in specific bundles may not have been accounted for. The gauge design we used to measure sap flow does not require penetration of the stem.

Tan et al. (1981) found that watering only 50 or 75% of the root system of tomato plants, grown in a sandy-loam soil, over a 14-d period did not reduce transpiration compared with a fully watered plant. When only 25% of the root system was watered, they reported a 20% reduction in transpiration rate as measured by weighing the boxes each day and accounting for the amount of water added to each watered section. In our experiment, transpiration was measured by recording sap flow, which was not affected until water was withheld for over 13 d (Table 1). A clay soil was used, which may explain why significant differences between the treatments took longer than expected to occur.

Blackman and Davies (1985) divided the root system of corn equally between two

Table 2. Effect of supplying water to various proportions of the root system of corn on fresh and dry root weights and the percent water in roots ($n = 4$)

Sections watered[z]	Dry root weight (g): Per section, watered	Per section, dry	Total	% in watered sections	Fresh root weight (g)	% water in roots
4	5.5	—	22.0*ab*	100	130.0*a*	83
3	6.0	5.4	23.2*a*	77	120.8*a*	81
2	5.5	5.6	22.1*a*	49	113.9*a*	81
1	5.8	3.6	16.6*b*	35	75.6*ab*	78
0	—	2.2	8.9*c*	—	30.9*b*	71

a–c Means within a column followed by the same letter are not significantly different at $P > 0.05$, using Tukey's studentized range (HSD) test (Statistical Analysis System Institute, Inc. 1987).

[z] Corn seedlings were transplanted into boxes divided into 4 equal sections. Treatments consisted of providing water to only a certain number of sections after 45 d of growth.

13-cm diam pots containing a compost–sand mix and found that drying part of the root system resulted in partial closure of stomata, even though leaf water potential and turgor remained unaffected. They suggested that a continuous supply of cytokinin from roots may be necessary to sustain maximal stomatal opening and an interruption of this supply due to soil drying may act as an indicator of inhibited root activity, resulting in restricted stomatal opening and thereby restricted water loss.

Relative water content, bulk water, xylem potential, stomatal conductance, and leaf turgor potential have been measured in various studies to characterize water relations in plants subject to stress. In the present experiment, sap-flow measurements using the heat balance technique provided a means to monitor the effects of water stress. This technique has been estimated to provide within 10% a true measure of transpiration (Sakuratani 1981). Use of the sectional root-boxes also allowed for watering a portion of the root system and thus the restriction of stress to known quantities of root mass. Although changes in sap flow were not immediate for various watering regimes, the continuous monitoring capability, ease of operation, and nonintrusive design of the sap-flow gauges make this technique desirable for studying water relations in corn.

Our results showed an increase in the stalk to root ratio from 2.5 to 4.1 as the number of sections watered increased from zero to four. Shank (1945) observed a similar increase in the ratio of shoot weight to root weight of corn from 2.5 to 3.4 by increasing the soil moisture level from between 7.5 and 15.5% to 21%. This increase indicates that root mass made up a smaller percentage of total plant mass as the percentage of roots receiving moisture increased. The growth of roots in drying soil is important if the rate of water uptake by plants is to be maintained (Caldwell 1976). A net increase in root growth and a decrease in the ratio of shoot weight to root weight by plants growing in conditions of low water availability therefore may be interpreted as an important adaptation of plants (Sharp and Davies 1979).

In an experiment where different proportions of the roots of peach (*Prunus persica* L.) seedlings received water, Tan and Buttery (1982) found that the reduction in root growth in dry sections was accompanied by the production of more roots in wet sections. They suggested that peach seedlings respond to withholding of water from part of the root system by a redistribution of growth in favour of roots in the wet sections. In our experiment, restricted watering appeared to have no effect on root growth in watered sections but decreased root growth in dry sections when three or four sections were deprived of water (Table 2).

This research shows that withholding water from 50% or more of the root system of corn can lead to decreased sap flow and shoot growth. Thus, corn plants with even half the roots growing under stressed or damaged conditions may be adversely affected. Plants in this experiment were grown in sectional root-boxes in close to an ideal environment in the greenhouse, and observations of growth characteristics presented here may differ under field conditions. However, results obtained in this experiment suggest that the root-box technique will be useful in studying the effects of root damage or stress in corn by biotic or abiotic sources.

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Baker, J. M. and van Bavel, C. H. M. 1987. Measurement of mass flow of water in the stems of herbaceous plants. Plant Cell Environ. **10**: 777–782.

Blackman, P. G. and Davies, W. J. 1985. Root to shoot communication in maize plants of the effects of soil drying. J. Exp. Bot. **36**: 39–48.

Bloodworth, M. E., Page, J. B. and Cowley, W. R. 1955. A thermoelectric method for determining the rate of water movement in plants. Soil Sci. Soc. Am. Proc. **19**: 411–414.

Caldwell, M. M. 1976. Root extension and water absorption. Pages 63–85 *in* O. L. Lange, L. Kappen and E. D. Schulze, eds. Water and plant life. Ecological Studies, Vol. 19, Springer-Verlag, Berlin, Germany.
Cohen, Y., Huck, M. G., Hesketh, J. D. and Frederick, J. R. 1987. Sap flow in the stem as related to water stress in corn and soybean. Pages 163–168 *in* R.J. Hanks, ed. Proc. of Int. Conf. on Measurement of Soil and Plant Water Stress, Vol. 2, Utah State University, Logan, UT.
Hills, T. M. and Peters, D. C. 1971. A method of evaluating postplanting insecticide treatments for control of western corn rootworm larvae. J. Econ. Entomol. **64**: 764–765.
Kahler, A. L., Olness, A. E., Sutter, G. R., Dybing, C. D. and Devine, O. J. 1985. Root damage by western corn rootworm and nutrient content in maize. Agron. J. **77**: 769–774.
Sakuratani, T. 1981. A heat balance method for measuring water flux in the stem of intact plants. J. Agric. Meteorol. **37**: 9–17.
Sakuratani, T. 1984. Improvement of the probe for measuring water flow rate in intact plants with the stem heat balance method. J. Agric. Meteorol. **40**: 273–277.
Shank, D. B. 1945. Effect of phosphorus, nitrogen and soil moisture on top-root ratios of inbred and hybrid maize. J. Agric. Res. (Washington, D.C.), **70**: 365–377.
Sharp, R. E. and Davies, W. J. 1979. Solute regulation and growth by roots and shoots of water-stressed maize plants. Planta **147**: 43–49.
Statistical Analysis System Institute, Inc. 1987. SAS/STAT guide for personal computers, version 6 edition. SAS Institute Inc., Cary, NC. 1028 pp.
Steinberg, S. L., van Bavel, C. H. M. and McFarland, M. J. 1990. Improved sap flow gauge for woody and herbaceous plants. Agron. J. **82**: 851–854.
Tan, C. S. and Buttery, B. R. 1982. The effect of soil moisture stress to various fractions of the root system on transpiration, photosynthesis, and internal water relations of peach seedlings. J. Am. Soc. Hortic. Sci. **107**: 845–849.
Tan, C. S., Cornelisse, A. and Buttery, B. R. 1981. Transpiration, stomatal conductance, and photosynthesis of tomato plants with various proportions of root system supplied with water. J. Am. Soc. Hortic. Sci. **106**: 147–151.