HYDRAULIC ARCHITECTURE OF WHOLE PLANTS AND SINGLE LEAVES

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Abstract

This paper presents a brief account of hydraulic architecture models of whole plants and of single leaves. The model for whole plants, T-Plant, is intended to investigate the consequences of reducing the complex resistance network of large plants to single resistance values. It is argued that the single resistance values depend on the method used to measure them, i.e., the evaporative flux method versus the high-pressure flowmeter method, and the resistance can depend on time of day in the evaporative flux method.

The single leaf models explore the use of "modular resistance models" in which a complex network of hundreds or thousands of leaf resistance elements is represented by repeating modules of just two to three resistance values. These models are quite robust, i.e., capable of predicting how leaf resistances change while leaves are cut back by repeated cuts from the leaf tip to leaf base, and can predict the impact of localized interruption in water transport by blockage in the midrib.

Various methods of measuring leaf resistance to water flow are compared. Leaf manipulations involving cuts to leaves and freezing to destroy cell membranes were used to indicate the relative magnitude of vascular versus non-vascular resistances.

Résumé

Le présent article présente un bref rapport sur des modèles d'architecture hydraulique de plantes entières et de feuilles individuelles. Le modèle pour les plantes entières, T-Plant, vise à étudier les conséquences de la réduction du complexe réseau de résistance des grandes plantes à des valeurs de résistance uniques. D'aucuns disent que les valeurs de résistance uniques, dépendent de la méthode utilisée pour les mesurer, soit la méthode du flux évaporant ou la méthode du débitmètre à haute pression, et que la résistance peut dépendre de l'heure du jour dans la méthode du flux évaporant

Les modèles de la feuille individuelle étudient l'utilisation de « modèles de résistance modulaires » dans lesquels un réseau complexe de centaines ou de milliers d'éléments de résistance foliaire est représenté en répétant des modules de deux ou trois valeurs de résistance seulement. Ces modèles sont assez solides, c.-à-d. capables de prédire la façon dont les résistances foliaires changent lorsque les feuilles sont coupées à plusieurs reprises de la pointe à la base et ils peuvent prédire l'impact de l'interruption locale du transport de l'eau par obstruction de la nervure médiane.

Nous avons comparé diverses méthodes de mesure de la résistance foliaire au mouvement de l'eau. Les manipulations, comprenant les coupures et la congélation pour détruire les membranes cellulaires, ont été utilisées pour indiquer l'importance relative des résistances vasculaire et non vasculaire.

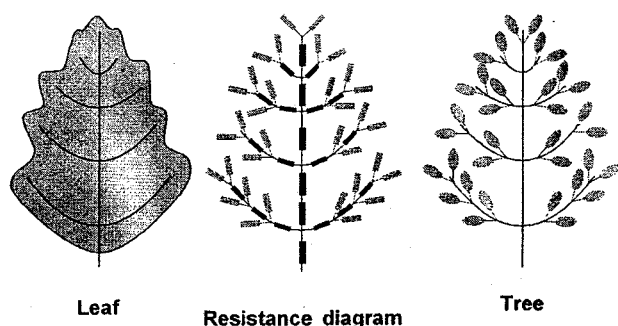**Leaf**   **Resistance diagram**   **Tree**

Figure 1: A single leaf or a whole tree can be represented by similar resistance diagrams. Resistance elements are represented by black or gray rectangles.

Whole plant models

The hydraulic architecture of a whole shoot or of a single leaf can be represented by a resistance diagram. Each hydraulic resistance element (R) equals the pressure difference driving flow through the element (ΔP) divided by the resulting flow (F), i.e., $R = \Delta P/F$. Resistance diagrams could describe an entire tree or a single leaf equally well. For the tree the black rectangles model for stem segment resistance and the white rectangles model for leaf resistance. For the leaf the black rectangles model for the leaf vein resistance and the gray rectangles for the resistance of the adjacent leaf blade (Figure 1).

Resistance diagrams can be at any level of complexity depending on the level of detail needed to answer a specific physiological question. We try to measure the values of the individual hydraulic resistances and then use the model to compute the pattern of water flow and water potentials in the resistance network. The problem comes when we try to compare different plants with hundreds or thousands of different resistance values. How do we evaluate the physiological importance of these many different resistance values?

The "holy grail" for hydraulic architecture is to reduce the resistance diagram to a single equivalent resistor so we only have one value to compare. But how do we define the equivalent resistance? One way is to define the equivalent resistance value operationally, i.e., by the methods we use to measure it. Two methods in common use are the evaporative flux (EF) method (Tsuda and Tyree, 2000) and the high-pressure flowmeter method (Tyree *et al.*, 1995). In the EF method the average evaporative flux is estimated by some means (e.g., gas exchange cuvette) and the average leaf water potential is estimated by another means (e.g., pressure bomb) and the whole plant equivalent resistance is equal to R = (average water potential drop)/(average evaporative flux). In the HPFM method the resistance of the root and shoot are measured separately by pressure perfusion and added together.

These two methods in theory should not produce the same resistance value. This is because the pattern of water flow in a shoot is not the same during pressure perfusion and free evaporation. During pressure perfusion with the HPFM the perfusion rate exceeds the rate of evaporation in every leaf. Hence the pressure of water at the evaporative surface in each leaf is zero. During free evaporation the rate of evaporation differs from leaf to leaf; leaves in sun evaporate faster than leaves in shade and in sun-exposed leaves the rate of evaporation differs with the angle of the leaf surface relative to the sun's rays. Hence the pressure potential drop from the base of the plant to each leaf differs.

The T-Plant model was used to investigate the probable effect of method used to measure equivalent resistance on the value of the resistance. T-Plant is an offshoot of Y-Plant (Pearcy and Yang, 1996), which is a three-dimensional crown architecture model for assessment of light capture and carbon gain in plants. The model loads a file that describes the three-dimensional architecture of a plant measured in the field. The measured data set provides stem-segment diameters, lengths, azimuth angle (compass direction the stem points), and angle of the stem with respect to the ground. Similar parameters are used to define the 3-D location of leaves. Computational methods are used to compute diffuse light and direct incident light interception of every sunny and shaded portion of every leaf based on any position of the sun in the sky. Net light absorption is used to compute net carbon assimilation based on photosynthetic light response curves or A-C$_i$ curves. T-Plant is a rewrite of Y-Plant starting from basic principles and using a computational method that is 3 times faster, otherwise both models produce similar results. T-Plant has additional computational routines to estimate stomatal conductance (g$_s$) based on the Berry-Ball equation. Transpiration from each leaf is computed from energy budget equations based on g$_s$ and information on air temperature, humidity and wind using equations suggested by Pearcy. Computational modules were also added to solve for the hydraulic architecture of the plant, based on measured hydraulic conductances of single leaves, stem segments and root systems from which leaf and stem water potentials can be calculated for any time of the day.

Space does not permit the full development of the quantitative differences between the EF and HPFM methods, but analysis that we have done with the aid of T-Plant indicated that resistances estimated by the EF method do change with the time of day because this affects the sun exposure of the shoot (Figure 2). Hence the EF method might give the false impression that the whole plant hydraulic resistance changes with time even though the resistances of the individual leaves and stem segments are constant. In contrast, the HPFM method produces a value independent of the time of day.

Single leaf models (maize)

Single leaf models of leaf hydraulic architecture are potentially complex because the vascular and non-vascular resistances of leaves must be viewed as a very large network of resistors in parallel and in series. Wei et al. (1999) have demonstrated that maize leaves can be described by two resistance elements repeated > 100

times, i.e., a modular resistance model with repeating modules of two resistors. In maize the resistance elements for each 1 cm length of leaf consist of a vascular resistance (R_x, representing all the parallel veins in 1 cm of leaf) and a non-vascular resistance (R_m, representing the resistance from parallel veins to the mesophyll air spaces). Values of R_x and R_m were estimated by measuring the hydraulic resistance of an intact maize leaf and the resistance as the leaf was progressively cut back from the apex to the base. The vascular (R_x) and non-vascular (R_m) resistances for the 1-cm sections can then be used to calculate xylem pressures in leaves versus distance under two water flow regimes, i.e., (1) when perfused by the HPFM and (2) during free evaporation. The pressure profiles differ, hence the flow pattern and equivalent resistances will differ (Figure 3). From this we conclude that the hydraulic resistance measured by the HPFM should be approximately 10% less than that measured by the EF method.
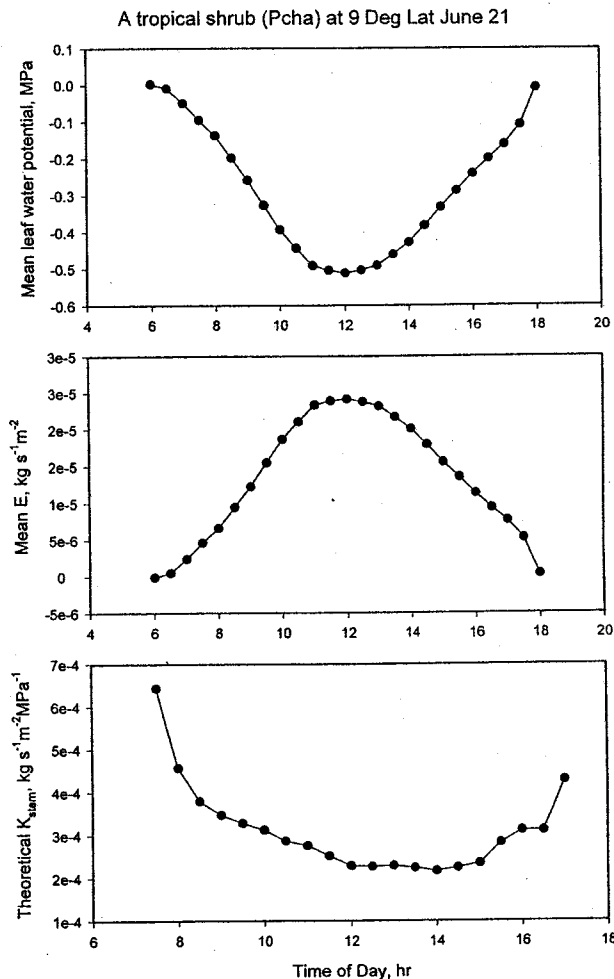

Figure 2: Theoretical output of T-Plant, which is a hydraulic architecture model: T-Plant inputs information on the three-dimensional structure of a plant and then computes the light interception, photosynthesis, leaf energy budget, and evaporation of 2 mm-wide rectangular grid areas on every leaf. This information is used to compute water potential profiles in the stems, petioles and leaves of the plant at 30-minute intervals based on the position of the sun with respect to the location of each leaf in the canopy and shading by leaves within the canopy. The model has been used to estimate the average evaporation rate from each leaf and the theoretical conductance of the plant. Plants with symmetrical crowns, with leaves approximately equidistant from the base of the plant and with little mutual shading have theoretical conductances independent of the time of day (data not shown). Plants with asymmetrical crowns and with leaves of unequal distance from the base have theoretical, whole-plant conductances that vary with time of day.
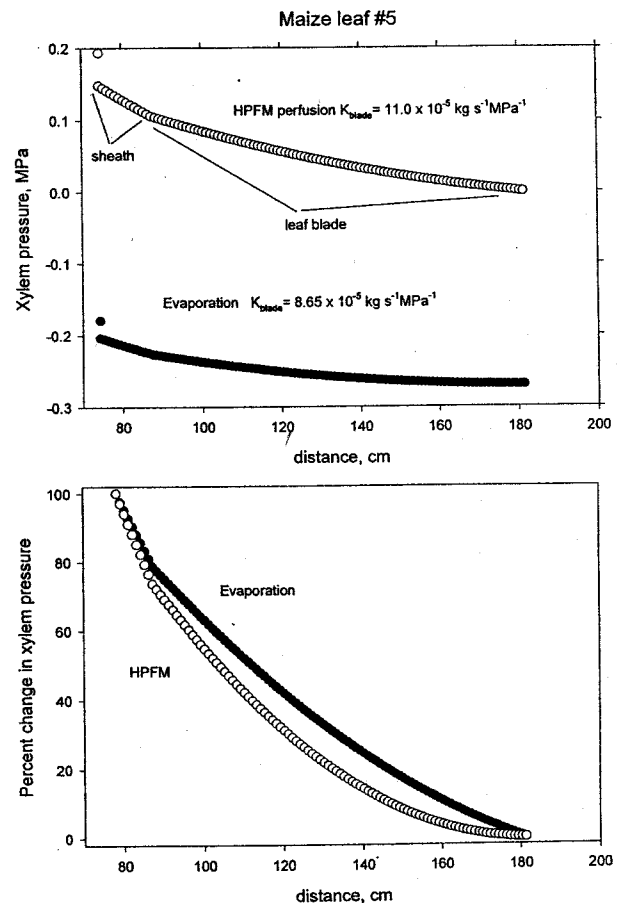

Figure 3: Hydraulic architecture model of a whole maize plant was used to compute pressure profiles in the 5th leaf from the base of large maize plant under two conditions: (1) during perfusion with a HPFM and (2) during free evaporation from the leaf surfaces. The upper figure shows the absolute pressure profiles in the xylem of the leaves. The lower curve shows the percent change in pressure from the base to the tip of the 5th leaf.

Hydraulic resistance patterns in leaves of Quercus, Viburnum and Prunus

The HPFM has been used to evaluate semi-quantitatively the relative magnitude of vascular and non-vascular pathways by measuring hydraulic resistance of Quercus and Viburnum leaves before and after altering water flow patterns by making cuts in leaf blades or by altering non-vascular resistances by freezing and thawing. The pattern of cuts is shown in Figure 4. The cuts modified vascular pathways by allowing water to flow through vessels from the base of the petiole to vessels cut open to the atmosphere at various distances from the petiole. However, not all water entering the petiole travels to the cut vessels; some water will still travel via the non-vascular pathway. Hence the cuts give only a semi-quantitative picture of relative resistances of the pathways. Freezing and thawing of leaf blades eliminates the membrane component of the non-vascular pathways, because electron microscope pictures of leaf tissue revealed complete destruction of the membranes after freezing and thawing. However, freezing does not eliminate all the non-vascular resistance components because resistance to water flow in cell walls (in vessels and formerly living cells) remains.

The pattern of resistances in whole leaves and after the cuts and/or freezing is shown in Figure 5. Leaf resistances of whole leaves fall to 26% and 43% (see Figure 5) of

the whole leaves (controls) after freezing in Quercus and Viburnum, respectively. Hence we conclude that both species have substantial non-vascular resistances. This conclusion is confirmed by the effect of the first cut in unfrozen samples. The first cut in unfrozen tissue results in a resistance significantly below that of frozen and thawed whole leaves in Quercus; this suggests that the non-vascular, cell-wall pathway offers more resistance than the vascular pathway to the first cut. In Quercus, the resistance of previously frozen blades is not significantly different from the unfrozen leaves after the first through fourth cuts; this is also consistent with the non-vascular, cell-wall resistance being more than the vascular resistance after every cut. Approximately the same can be said for Viburnum but the non-vascular, cell-wall resistances are not as large relative to the vascular resistances since freezing and thawing consistently lowers the resistance in the first to third cuts. However, these conclusions have to be accepted with caution because the vascular and non-vascular pathways are a complex array of resistances simultaneously in series and in parallel, thus making interpretation difficult.
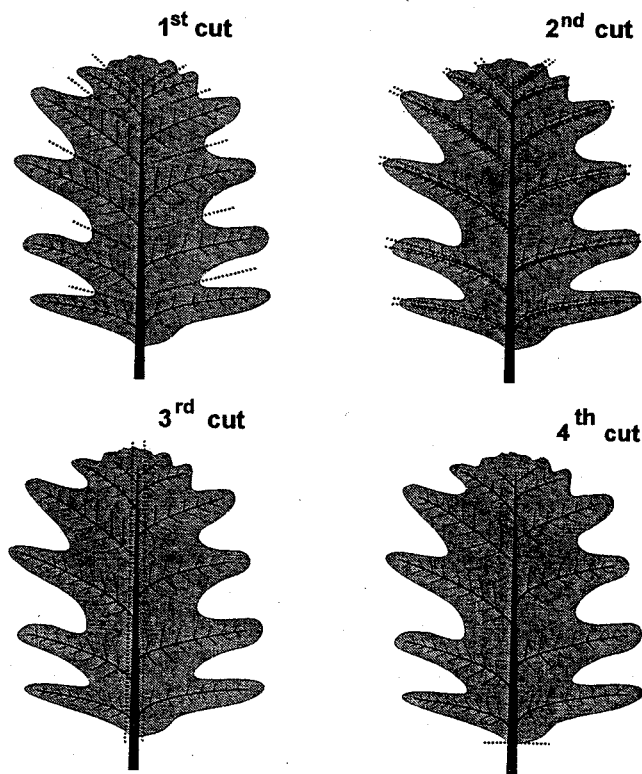

Figure 4: A Quercus petraea leaf illustrating the position of cuts (indicated by dashed lines) made to the leaf blade. Similar cuts were made on a Viburnum tinus leaf.
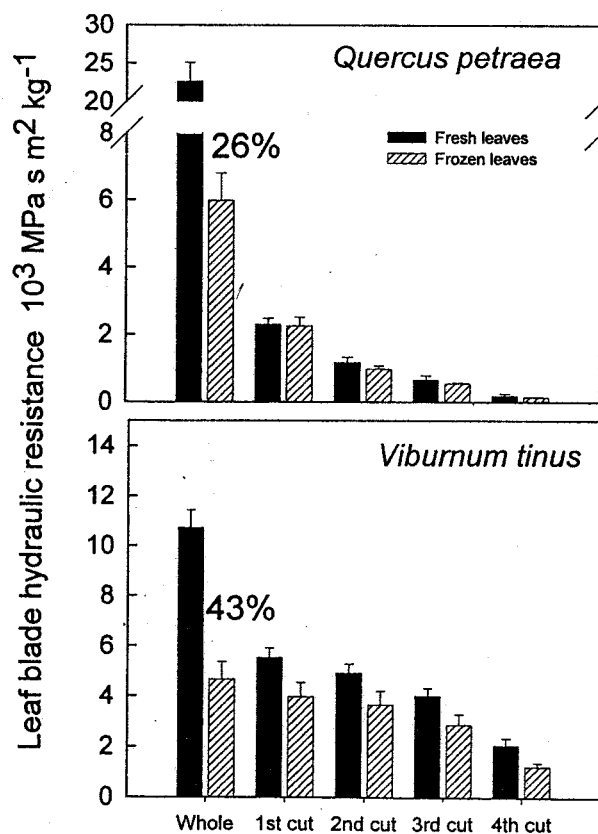

Figure 5: Bar diagram showing the influence of cuts without freezing (solid bars) and after freezing and thawing (hatched bars) on leaf hydraulic resistance. "Whole" indicates leaves without cuts. Cuts are made as shown in Figure 4.
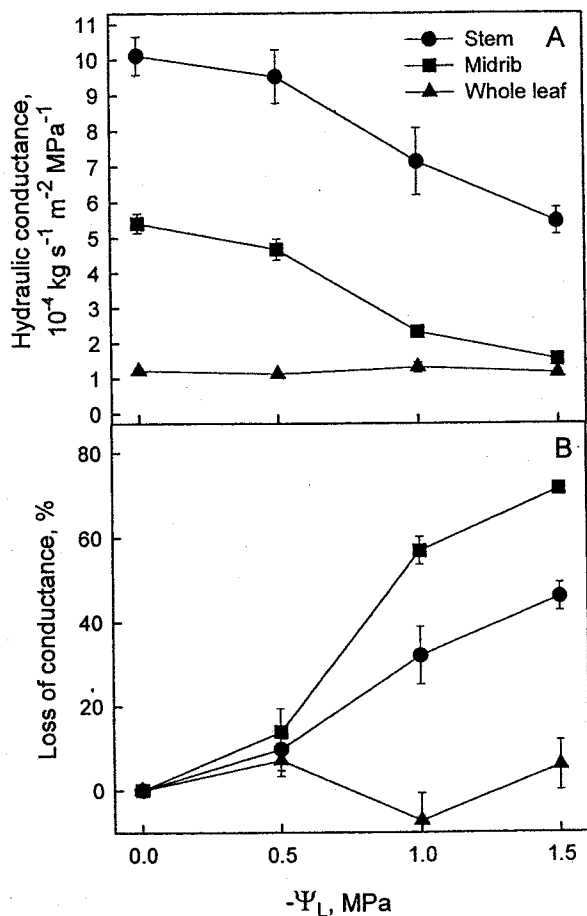
Figure 6: Change in hydraulic conductance of stems, midribs and whole leaves of Prunus laurocerasus L. shoots during dehydration to leaf water potentials shown on x-axis. Upper graph shows absolute conductance values and lower graph loss of conductance as percent of the maximum conductance.
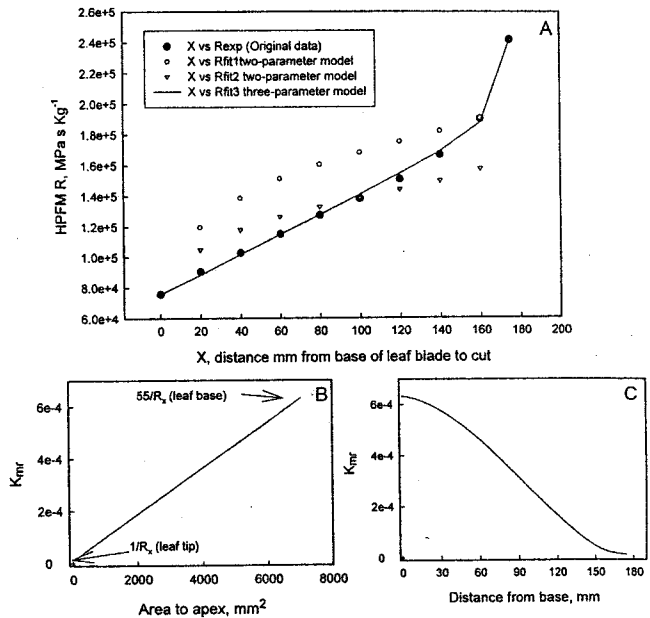

Figure 7: Upper figure: Solid circles indicate measured leaf resistance versus distance from base of leaf to a cut made perpendicular to the midrib. The point at the far right is for the intact leaf. Small symbols indicate theoretical fit in which grid elements containing the midrib are assumed to have the same resistance as the leaf blade. Any two points can be fit, but not the entire data set. The solid line is the theoretical fit for the three-parameter model where Rx (vascular leaf-blade resistance of a grid element), Rm (non-vascular leaf-blade resistance of the grid element), and conductance at the base of the midrib are specified as N/Rx where N = 55 in this fit. For all other midrib grids the conductance is given as a linear function of leaf area (lower left graph) that results in the midrib conductance versus distance given in the lower right graph.

Embolism and leaf hydraulic conductance measured by the vacuum method

A few studies have used acoustic methods to detect cavitation events in leaves (Milburn, 1973; West and Gaff, 1976; Kikuta et al., 1997; Salleo et al., 2000), but no work has been done on single leaves to evaluate the effect of embolisms resulting from cavitation events on whole leaf hydraulic conductance. The HPFM and EF methods are likely to produce different values of leaf resistance in unembolized leaves because of differences in flow pattern as in maize (see above). In embolized leaves the resistance by the EF method is likely to be more than by the HPFM method because perfusion of leaves by the HPFM is likely to dissolve embolisms very rapidly, hence the HPFM tends to measure the maximum conductance (minimum resistance). Another method known to work in highly branched shrubs without dissolving embolisms is the vacuum chamber method (Kolb et al., 1996). In preliminary

studies we confirmed that the EF and vacuum chamber methods gave mean resistances in Prunus leaves that did not differ significantly (R_{LL} = 8200 MPa s m^2 kg^{-1}, N = 7) whereas the HPFM measurement on similar field- collected leaves had resistance values that were significantly lower (R_{LL} = 5800, N = 7). At this point we cannot say how much of the difference was due to native embolism versus differences in water flow pathway in the three methods.

Vulnerability curves of Prunus stems, midribs and whole leaves were generated by dehydrating whole branches to leaf water potentials of −0.5, −1.0 and −1.5 MPa and comparing them unstressed branches. These studies revealed that by −1.5 MPa, leaf midribs had lost 70% of their conductivity compared to 45% in stems, but whole leaves had < 10% loss of conductance (Figure 6). These results are consistent with leaf resistance being dominated by non-vascular resistance of the leaf blade and because of vascular redundancy, i.e., the multitude of alternative water pathways in the leaf blade afforded by the network of minor veins.

To examine the level of vascular redundancy, water flow through the midrib was interrupted by cutting the midrib 20 mm from the leaf base, and sealing the cut (8 mm wide) by epoxy. The vacuum method revealed
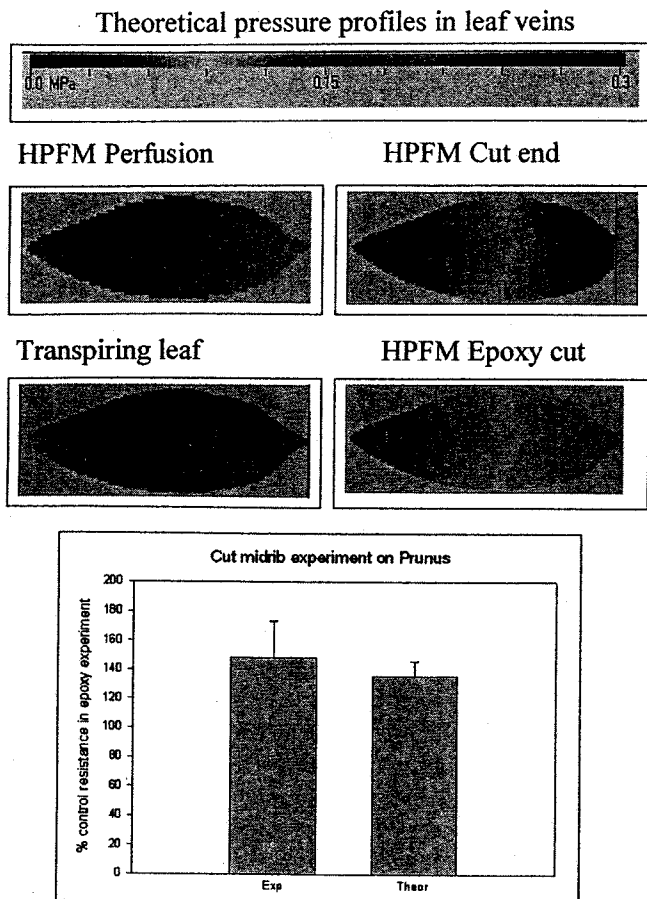
Theoretical pressure profiles in leaf veins

HPFM Perfusion HPFM Cut end

Transpiring leaf HPFM Epoxy cut

Cut midrib experiment on Prunus

Figure 8: Theoretical water potential profiles in the veins of Prunus laurocerasus leaves. For HPFM pressure profiles the color scale is positive. For transpiring leaf, interpret the color scale as negative values (transpiration flux density = 1.4 x 10-4 kg s-1 m-2). See text for details.

that the leaf resistance increased by only 40% of the intact leaf. In order to quantify vascular redundancy, we attempted to parameterize a modular resistance model for a dicot leaf.

A modular resistance model of Prunus leaves

A modular resistance model was written which could take as the input the x-y coordinates of a leaf outline (the leaf margins). The model then divided the leaf area into 2 mm-square grids. Each grid was connected to each of the four adjacent square grids by equal vascular resistances, R_x (MPa s kg^{-1}), and each grid had a single non-vascular resistance, R_m, which represented average resistance for water flow from the minor veins of the grid to the mesophyll air spaces. Grid elements containing the midrib were assigned a maximum conductance at the base of the midrib and a conductance equal to that of the leaf blade at the apex of the midrib; hence the conductances varied from N/R_x to $1/R_x$, where N is a number greater than one.

At all midrib-grids in between, the midrib conductance was assumed to be proportional to the leaf area to the apex of the midrib grid. Although the model required thousands of resistance elements to describe a *Prunus* leaf, the modular nature of the model required only three parameters: R_x, N/R_x, and R_m.

The model was parameterized much like maize leaves, e.g., the resistance of a whole *Prunus* leaf was measured, then the leaf was progressively cut back, with cuts perpendicular to the midrib, until only the petiole remained. The leaf resistance was measured after each cut, and values of R_x, N/R_x, and R_m were found that best fit the data (Figure 7). The model could then be used to predict pressure profiles in xylem in intact leaves, in leaves with tips cut off, in transpiring leaves, and in leaves with midribs blocked by epoxy (Figure 8).

Theoretical pressure profiles were qualitatively similar for transpiring leaves and for leaves perfused by the HPFM. In Figure 8 pressures are color-coded and the color-coded values should be interpreted as negative values in transpiring leaves. The v-shared color pattern in the figures shows the influence of the midrib grids that are more conductive than leaf blade grids; pressures decrease from the midrib to the leaf margins along paths perpendicular to the midrib. When the leaf tip is excised, the pressure at the cut surface is zero (atmospheric) and this disrupts the v-shaped pressure profile near the cut. The model is very robust because it accurately predicts how leaf resistance changes as the leaf is cut back from the apex. It also accurately predicts the percent change in leaf resistance when flow through the midrib is interrupted by epoxy. Near the epoxy, water flow is diverted to the leaf blade and flows around the interruption to rejoin the midrib where the characteristic v-shaped pressure profile resumes. Note the steep pressure gradients near the epoxy. The predicted change of resistance from the model is not significantly different from the experimental values.

Conclusion

More research needs to be done to see if the simplified modular model of leaf resistance will correctly predict the impact of embolism and other types of mechanical damage (e.g., due to insects and pathogens) on hydraulic resistance of leaf blades.

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