A better understanding of the mechanistic basis of stomatal control is necessary to understand why modes of stomatal response differ among individual trees, and to improve the theoretical foundation for predictive models and manipulative experiments. Current understanding of the mechanistic basis of stomatal control is reviewed here and discussed in relation to the plant hydraulic system. Analysis focused on:

1. The relative role of hydraulic conductance in the vicinity of the stomatal apparatus versus whole-plant hydraulic conductance;
2. The influence of guard cell inflation characteristics and the mechanical interaction between guard cells and epidermal cells; and
3. The system requirements for moderate versus dramatic reductions in stomatal conductance with increasing evaporation potential.

Special consideration was given to the potential effect of changes in hydraulic properties as trees grow taller. Stomatal control of leaf gas exchange is coupled to the entire plant hydraulic system and the basis of this coupling is the interdependence of guard cell water potential and transpiration rate. This hydraulic feedback loop is always present, but its dynamic properties may be altered by growth or cavitation-induced changes in hydraulic conductance, and may vary with genetically related differences in hydraulic conductances. Mechanistic models should include this feedback loop. Plants vary in their ability to control transpiration rate sufficiently to maintain constant leaf water potential. Limited control may be achieved through the hydraulic feedback loop alone, but for tighter control, an additional element linking transpiration rate to guard cell osmotic pressure may be needed.

**Keywords:** hydraulic conductivity, stomatal conductance, stomatal models, transpiration control, transpiration rate, tree water use.

**Introduction**

Although the link between stomatal and hydraulic conductance has long been realized, its mechanistic basis has been difficult to unravel. With increasing transpiration rate, the sensitivity of the stomatal response and the extent of the decline in leaf water potential are both determined by hydraulic conductance (Raschke 1970). The characterization of this relationship is confounded by several factors, including: (1) the many other signals to which stomata respond; (2) the lack of a clearly defined set-point for the stomatal control system; and (3) an unresolved mechanism for stomatal sensing of transpiration rate. Much progress has been made in the pursuit of understanding each of these factors (see reviews by Schroeder et al. 2001, Buckley and Mott 2002). Although empirical models of stomatal function have been developed (e.g., Jarvis 1976, Farquhar and Wong 1984, Ball et al. 1987, Tardieu and Davies 1993, Leuning 1995, Dewar 2002), improvements in the mechanistic modeling of stomatal function are necessary to explain observed differences among individual trees and to provide a theoretical basis for predictive models or manipulative experiments. Because the stomatal apparatus is ultimately a hydraulically operated mechanical device, mechanistic models of stomatal function require an understanding of both the hydraulic fluxes and the mechanical forces that govern the movement of guard cells. This review focuses on these key elements and evaluates the influence of hydraulic conductances at both the stomatal apparatus and the whole-plant scale. Special attention is given to the effect of potential changes in the hydraulic system of growing trees, and to the extreme conditions that prevail in tall trees.

**Theory**

**Definition of terms for hydraulic conductance and conductivity**

The steady-state flow of fluids through wood is described by Darcy’s Law (Scheidegger 1974), which, substituting molar for mass units, equates molar flux ($J_s$; flow rate/sapwood area) with the product of sapwood-area-specific hydraulic conductivity ($K_s$; mol m$^{-1}$ s$^{-1}$ MPa$^{-1}$) and the hydraulic pressure gradient ($\Delta P/\Delta L$; MPa m$^{-1}$):

$$J_s = -K_s \left( \frac{\Delta P}{\Delta L} \right)$$  (1)

A complete list of symbols and units is provided in Table 1. When viewing flow through the tree as a whole, the hydraulic pressure gradient may be approximated as $\Delta P_{a.}/\Delta L_{a.-}$, where
\( \Delta P_{s-l} \) is the hydraulic pressure difference between soil and leaf (pressure in leaves minus pressure in soil) and \( \Delta L_{s-l} \) is the length of the flow path between soil and leaf. The hydraulic pressure difference between soil and leaf that is due to the vertical column of xylem water hanging from the plant’s internal surfaces, \( \rho gh \), must be added to \( \Delta P_{s-l} \), giving:

\[
J_s = -K_s \left( \frac{\Delta P_{s-l} + \rho gh}{\Delta L_{s-l}} \right)
\]

(2)

where \( \rho \) is the density of water, \( g \) is acceleration due to gravity, and \( h \) is the height of the water column (e.g., vertical distance from soil to sites of evaporation in canopy leaves). In the case of water flowing up a tree, i.e., against gravity, \( \rho gh \) acts to oppose \( J_s \), although in many gravity-fed hydrological systems it assists flow.

To maintain compatibility with the leaf-area-specific units for leaf transpiration rate (\( E \); mol m\(^{-2}\) leaf s\(^{-1}\)), liquid-phase flux of water through the xylem is conveniently expressed as molar flux of water relative to leaf area fed (\( J_L \); mol m\(^{-2}\) leaf s\(^{-1}\)):

\[
J_L = -K_L \left( \frac{\Delta P_{s-l} + \rho gh}{\Delta L_{s-l}} \right)
\]

(3)

where \( K_s \) is the leaf-area-specific hydraulic conductivity (mol m\(^{-1}\) s\(^{-1}\) MPa\(^{-1}\)) and \( K_L \) is obtained from \( K_s \) as:

\[
K_L = K_s \left( \frac{A_L}{A_s} \right)
\]

(4)

where \( A_s \) is sapwood cross-sectional area and \( A_L \) is leaf area fed. For a given hydraulic path length, e.g., from soil to tree canopy, the leaf-area-specific whole-plant hydraulic conductance from soil to leaf, \( k_{L(s-l)} \), is given by:

\[
k_{L(s-l)} = \frac{K_L}{\Delta L_{s-l}}
\]

(5)

**Table 1: Symbols, description and units for terms used.**

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>( A_L )</td>
<td>Leaf area</td>
<td>m(^2)</td>
</tr>
<tr>
<td>( A_s )</td>
<td>Sapwood cross-sectional area</td>
<td>m(^2)</td>
</tr>
<tr>
<td>( a )</td>
<td>Stomatal pore width</td>
<td>( \mu )m</td>
</tr>
<tr>
<td>( c )</td>
<td>Constant that is proportional to diffusive conductance to the direct component of the ( D )-response stimulus</td>
<td>MPa</td>
</tr>
<tr>
<td>( D )</td>
<td>Leaf-to-air vapor pressure difference</td>
<td>kPa</td>
</tr>
<tr>
<td>( E )</td>
<td>Transpiration rate</td>
<td>mol m(^{-2}) s(^{-1})</td>
</tr>
<tr>
<td>( g^* )</td>
<td>Representative of bulk stomatal elastic modulus</td>
<td>MPa</td>
</tr>
<tr>
<td>( g_{sw} )</td>
<td>Stomatal conductance to water vapor</td>
<td>mol m(^{-2}) s(^{-1})</td>
</tr>
<tr>
<td>( g )</td>
<td>Acceleration due to gravity</td>
<td>m s(^{-2})</td>
</tr>
<tr>
<td>( H )</td>
<td>Open loop gain of the stomatal hydraulic negative feedback system</td>
<td>–</td>
</tr>
<tr>
<td>( h )</td>
<td>Tree height</td>
<td>m</td>
</tr>
<tr>
<td>( J_L )</td>
<td>Molar flow rate of water per unit leaf area</td>
<td>mol m(^{-2}) s(^{-1})</td>
</tr>
<tr>
<td>( J_s )</td>
<td>Molar flow rate of water per unit sapwood area</td>
<td>mol m(^{-2}) s(^{-1})</td>
</tr>
<tr>
<td>( K_s )</td>
<td>Leaf-area-specific hydraulic conductivity</td>
<td>mol m(^{-1}) s(^{-1}) MPa(^{-1})</td>
</tr>
<tr>
<td>( K_L )</td>
<td>Sapwood-area-specific hydraulic conductivity</td>
<td>mol m(^{-1}) s(^{-1}) MPa(^{-1})</td>
</tr>
<tr>
<td>( k_{L(s-l)} )</td>
<td>Leaf-area-specific hydraulic conductance from soil to leaf</td>
<td>mol m(^{-2}) s(^{-1}) MPa(^{-1})</td>
</tr>
<tr>
<td>( k_{L(e-g)} )</td>
<td>Leaf-area-specific hydraulic conductance from soil to epidermis</td>
<td>mol m(^{-2}) s(^{-1}) MPa(^{-1})</td>
</tr>
<tr>
<td>( k_{L(g)} )</td>
<td>Leaf-area-specific hydraulic conductance from soil to guard cell</td>
<td>mol m(^{-2}) s(^{-1}) MPa(^{-1})</td>
</tr>
<tr>
<td>( k_{L(g-e)} )</td>
<td>Leaf-area-specific hydraulic conductance from epidermis to guard cell</td>
<td>mol m(^{-2}) s(^{-1}) MPa(^{-1})</td>
</tr>
<tr>
<td>( \Delta L_{s-l} )</td>
<td>Hydraulic path length from soil to leaf</td>
<td>m</td>
</tr>
<tr>
<td>( m )</td>
<td>Ratio of sensitivities of ( a ) to ( P_e ) and ( a ) to ( P_g )</td>
<td>–</td>
</tr>
<tr>
<td>( \Delta P )</td>
<td>Difference in hydraulic pressure over path length (( \Delta L ))</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Delta P_{s-l} )</td>
<td>Difference in hydraulic pressure between soil and leaf</td>
<td>MPa</td>
</tr>
<tr>
<td>( P_e )</td>
<td>Epidermal cell turgor pressure</td>
<td>MPa</td>
</tr>
<tr>
<td>( P_g )</td>
<td>Guard cell turgor pressure</td>
<td>MPa</td>
</tr>
<tr>
<td>( P_a )</td>
<td>Atmospheric pressure</td>
<td>kPa</td>
</tr>
<tr>
<td>( \Pi_L )</td>
<td>Epidermal cell osmotic pressure</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Pi_g )</td>
<td>Guard cell osmotic pressure</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Pi_{g(max)} )</td>
<td>Maximum guard cell osmotic pressure</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Psi_l )</td>
<td>Leaf water potential</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Psi_s )</td>
<td>Soil water potential</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Psi_g )</td>
<td>Guard cell water potential</td>
<td>MPa</td>
</tr>
<tr>
<td>( \Psi_e )</td>
<td>Epidermal cell water potential</td>
<td>MPa</td>
</tr>
<tr>
<td>( \rho )</td>
<td>Density of water</td>
<td>kg m(^{-3})</td>
</tr>
</tbody>
</table>
Both $K_i$ and $K_l$ are independent of the length of the hydraulic pathway, whereas $k_{L(i-t)}$ decreases with increasing $\Delta L_{s-l}$ (tree height) if all else remains constant. Below (see Discussion), the effect of increasing hydraulic path length on the stomatal control mechanism is considered for the case of a growing tree where, although all else may not remain constant, it has been shown that $k_{L(i-t)}$ sometimes declines with height.

If $J_L$ is known, and $\Delta P_{s-l}$ is measured as the difference between soil and leaf water potential, $\Psi_i - \Psi_l$, then $k_{L(i-t)}$ may be calculated as:

$$k_{L(i-t)} = \frac{J_L}{\Psi_i - \Psi_l - \rho gh}$$

Equation 6 can be adapted to represent the leaf-area-specific hydraulic conductance between any two points along the plant hydraulic pathway. The analysis here uses leaf-area-specific whole-plant hydraulic conductance from soil to guard cell ($k_{L(i-g)}$), and leaf-area-specific whole-plant hydraulic conductance from soil to epidermal cells ($k_{L(i-e)}$):

$$k_{L(i-g)} = \frac{E}{\Psi_i - \Psi_g - \rho gh}$$

and

$$k_{L(i-e)} = \frac{E}{\Psi_i - \Psi_e - \rho gh}$$

where $\Psi_g$ and $\Psi_e$ are guard cell and epidermal cell water potential, respectively.

**Stomatal hydraulics**

Stomata are coupled hydraulically to each other and to the rest of the leaf (Raschke 1970, Mott et al. 1997, Buckley and Mott 2000, Mott and Franks 2001). Hydraulic coupling of stomata may play a role in their sensitivity to perturbations in evaporative demand (see below). A consequence of this linkage is that stomatal regulation of leaf gas exchange is directly influenced by the cuticular transpiration from stomatal guard cells and epidermis, however small, the question is not so much whether this mechanism exists, but what contribution it makes to the overall stomatal response to evaporative demand.

For example, Meinzer (1982) showed that partially removing the cuticle of Douglas-fir needles with a hexane wash increased stomatal sensitivity to $D$, proving the potential for cuticle-mediated stomatal response to changes in $D$. However, Frensch and Schulze (1988), using simultaneous pressure probe and gas exchange measurements on *Trädescantia virginiana* L., a plant with a well-documented but typically mild stomatal response to $D$, observed no significant change in epidermal turgor or transpiration rate in response to a step change in $D$ when stomata were closed, suggesting that in *T. virginiana* the stomatal response to $D$ could not be mediated via the cuticle. It therefore seems that cuticular transpiration may in some cases play a role, but is not always an essential component of the mechanism regulating stomatal conductance in response to $D$. For these reasons, most of the analysis here focuses on the case where guard cell or epidermal cuticular transpiration contrib-
utes only minimally to stomatal control. Later, this is contrasted with the response characteristics of a stomatal control system that is sensitive to cuticular transpiration.

All mechanistic models of stomatal regulation of transpiration rate require assumptions to be made about the pattern of hydraulic fluxes in the vicinity of stomatal guard cells. Theoretical analyses of Meidner (1976), Cowan (1977), Tyree and Yianoulis (1980) and Pickard (1982) all show that for a uniformly wet sub-stomatal chamber, most of the transpirational flux will emanate from wet surfaces closest to the stomatal pore. However, it is not clear how far the exterior waxy cuticle of the leaf surface extends into the throat of the stomatal pore (or even further, into the substomatal cavity), and so it is difficult to generalize about the extent of wet cell surfaces in the vicinity of the stomatal pore. There are two conceptual models that divide a broad spectrum of possibilities regarding the pathways taken by the transpiration stream once it leaves the xylem and flows toward the stomatal pores: the first, shown in Figure 1A(i), is based on the assumption that the cuticular covering of stomatal guard cells is extensive, and that wet surfaces of the leaf interior extend only as far as the adjacent subsidiary or epidermal cells. This means that these cells support the bulk of the transpirational water flux, and that because there is negligible evaporative loss from guard cells, \( \Psi_g = \Psi_e \) in the steady state. The second model, shown in Figure 1A(ii), assumes that cuticular covering of guard cells is not extensive, and that the surfaces of the guard cells that face the leaf interior, being devoid of a substantially impermeable cuticular covering, are wet and support the bulk of the transpirational water flux. In this case, \( \Psi_g \) and \( \Psi_e \) are not equal in the steady state, and may be expressed by rearrangement of Equations 8 and 9, as:

\[
\Psi_g = \Psi_e - \left( \frac{E}{k_{L(s-g)}} \right) - \rho gh
\]  

(10)

and

\[
\Psi_e = \Psi_g - \left( \frac{E}{k_{L(s-e)}} \right) - \rho gh
\]  

(11)

In this second model, the hydraulic conductance between subsidiary cell and guard cell, \( k_{L(s-g)} \), has a strong influence on the water potential of the guard cell for any given steady state \( E \). If \( k_{L(s-g)} \) is relatively low, then \( \Psi_g \) will be much lower than \( \Psi_e \). If \( k_{L(s-g)} \) is relatively high, then \( \Psi_g = \Psi_e \), \( k_{L(s-g)} \) is the sum of \( k_{L(s-e)} \) and \( k_{L(e-g)} \):

\[
k_{L(s-g)} = \left( \frac{1}{k_{L(s-e)}} + \frac{1}{k_{L(e-g)}} \right)^{-1}
\]  

(12)

Stomatal mechanics

The opening and closing of stomatal pores results primarily from increasing or decreasing guard cell turgor, \( P_g \). In addition, stomatal pore size \( a \) may be influenced by changes in the turgor of surrounding epidermal cells, \( P_e \): an increase in \( P_e \) can push guard cells closer together to reduce pore size, and decreasing \( P_e \) can act in the reverse (Figure 1B). At any point in time, \( P_g \) and \( P_e \) are defined by:

\[
P_e = \Psi_e + \Pi_e
\]  

(13)

and

\[
P_g = \Psi_g + \Pi_g
\]  

(14)

where \( \Pi_g \) and \( \Pi_e \) are guard cell and epidermal cell osmotic pressures (MPa), respectively. In the short term, \( \Pi_e \) is kept relatively constant, and \( P_e \) changes passively with \( \Psi_e \). In contrast,
Π_g is actively regulated by guard cells in direct response to both internal (within-leaf) and external (environmental) stimuli. Guard cell turgor pressure, and hence stomatal conductance, may change relatively quickly in response to stimuli that alter Π_g or Ψ_e (Grantz and Zeiger 1986).

The characteristics of guard cell inflation have been measured and described quantitatively for several species (Meidner and Edwards 1975, Franks et al. 1998, 2001, Franks and Farquhar 2001). Figure 2 is typical of the form of the relationship between a, P_g and P_e, and shows how P_e can reduce stomatal pore size over the entire range of P_g. In this example, the reduction in a due to P_e ranges from 100% at low P_g to about 50% at high P_g, but across species, the effect may not always be this great. Increasing a as a result of decreasing P_e means that elaborate countermeasures need to be built into the stomatal control mechanism to limit transpirational water loss under conditions that tend to decrease P_e, such as increasing evaporative demand or decreasing soil water potential.

The wide variation in overall structure and physical arrangement of guard cells and adjacent epidermal cells in different species suggests that the magnitude of the mechanical interaction between the two is likely to vary considerably across species. This is now confirmed by a growing number of direct measurements of a at different P_g and P_e, with the cell pressure probe (Franks 2003). What has also emerged from these pressure probe studies is that the mechanical interaction varies considerably over the range of P_g and P_e for a given stomatal complex. Therefore, the mechanical relationship, m, between guard cells and epidermis is specific to a given combination of P_g and P_e, and for any such combination is best defined by the ratio of the sensitivities of a to P_e at constant P_g, and a to P_g at constant P_e (Cooke et al. 1976):

\[
m = -\frac{\frac{\partial a}{\partial P_e}}{\frac{\partial a}{\partial P_g}}\bigg|_{P_e}
\]

When m > 1 (the usual case), there will be a net reduction in stomatal aperture for the same increase in P_g and P_e. Under these conditions, the epidermal cells are said to have a “mechanical advantage” over the guard cells (m is sometimes referred to as the mechanical advantage). The larger the value of m, the more difficult it is for guard cells to open against the opposing force of the adjacent epidermal cells. If there is no mechanical interaction between guard cells and epidermis, then m = 0.

If the dependencies of a on P_g with zero epidermal turgor and maximum epidermal turgor, P_e(max), are defined by functions f_1(P_g) and f_2(P_g), respectively, and the reduction in a due to P_e is assumed to be proportional to P_e, then a may be expressed as (Franks et al. 1998):

\[
a = f_1(P_g) - \frac{P_g(f_1(P_g) - f_2(P_g))}{P_e(max)}
\]

If stomatal pore size is unaffected by P_e, then Equation 16 reduces to:

\[
a = f_1(P_g)
\]

The functions f_1(P_g) and f_2(P_g) are as yet unknown for any forest tree species, but it could be assumed that the aperture versus guard cell turgor relationship for the stomata of trees is qualitatively similar to that for the stomata in herbaceous plants. These qualitative features, as illustrated in Figure 2, are: (1) in the absence of any mechanical interaction with epidermal cells, the relationship between a and P_g approximates an exponential decay function; (2) in the presence of mechanical interaction with epidermal cells, the relationship between a and P_g is sigmoidal; and (3) maximum stomatal aperture for any given epidermal turgor pressure is achieved with guard cell turgor pressures of about 4.0 MPa. For simplicity, the simulations presented here use the parameters for f_1(P_g) and f_2(P_g) given in Franks et al. (1998), and for illustrative purposes, an arbitrary multiplier of 0.04 is used in all simulations to scale from stomatal aperture to stomatal conductance, g_sw (g_sw = 0.04 x a). The magnitude of the scaling factor, 0.04, which accounts for differences in stomatal size and density, was chosen because it yields stomatal conductances in the range typical of forest trees.

Interaction between leaf gas exchange and leaf hydraulics

When the leaf boundary layer conductance is high (leaves well coupled to the atmosphere), the stomatal conductance to water vapor on a leaf area basis, g_sw, is given by:
where $D$ is the leaf-to-air vapor pressure difference (kPa) and $P_a$ is atmospheric pressure (kPa). When Equations 7 and 18 hold, and if transpiration is sustained mainly from the moist surfaces of guard cells just inside the stomatal pore (Figure 1A (ii); see Stomatal hydraulics), then:

$$g_{sw} = \frac{E}{D/P_a}$$  \hspace{1cm} (18)$$

Although Equation 19 accurately describes the relationship between $g_{sw}$, $E$, $D$, $k_{L(s–g)}$, $\Psi_e$, $\Psi_g$ and $h$ in the steady state, it cannot be used as a predictive tool for studying the response of $g_{sw}$ to $D$ or $k_{L(s–g)}$ because the physical structure of the system makes $g_{sw}$, $E$ and $\Psi_g$ interdependent. For example, an instantaneous change in $D$ will change $E$, which will change $\Psi_g$, which will change $P_g$, which will change $g_{sw}$, which will change $E$. This circular flow of information forms part of the elaborate feedback system that controls stomatal movement, and is illustrated in Figure 3. A more formal mathematical description of this system is summarized in the block diagram of Figure 4, which is adapted from Franks and Farquhar (1999) and built on the general principles of feedback in the stomatal mechanism as outlined by Cowan (1977). This model captures the basic biophysical linkages between stomatal guard cells and the attached plant hydraulic system. It is coupled to the other basic biophysical linkages between stomatal guard cells and the transpiration stream. It is this role that is at the core of stomatal action, and it is best studied through examination of stomatal response to changes in $D$.

## Discussion

The mode of stomatal response to increasing evaporative demand

As air becomes drier, the leaf-to-air vapor pressure difference ($D$) increases, and so does the potential rate of transpiration. Stomata cannot protect the plant from declining water potentials associated with drying soil, or from the static negative water potentials imposed by the water columns hanging vertically in the xylem, but by regulating their aperture, and therefore, the transpiration rate, they can minimize the additional drop in tissue water potentials associated with the hydraulic drag on the transpiration stream. It is this role that is at the core of stomatal action, and it is best studied through examination of stomatal response to changes in $D$.

Almost without exception, studies of the stomatal response to an increase in $D$ report a decline in stomatal conductance (Darwin 1898, Lange et al. 1971, Schulze et al. 1972, Hall et al. 1976, Whitehead et al. 1981, Monteith 1995, Oren et al. 1999, Buckley and Mott 2002). The mode of this response is often described as resembling an exponential decay or negative log function of $D$ (e.g., Lohammar et al. 1980, Granier and Bédréa 1996, Oren et al. 1999), and this often yields functions accounting for a significant proportion of the variance in large data sets. However, the true nature of the relationship between $g_{sw}$ and $D$, revealed under more carefully controlled conditions, may deviate somewhat from this simplification, and the underlying mechanism has remained a puzzle for some time. One of the first studies to illustrate both the generality of the overall response (declining $g_{sw}$ with $D$) and the variability of the form of this response across woody species was that of Waring and Franklin (1979). Their results, reproduced in Figure 5, highlight two characteristics that vary across different species. The first relates to a region of the curve at low values
of $D$ where $g_{sw}$ is relatively insensitive to $D$, or sometimes actually increases (Farquhar 1978, Monteith 1995). This characteristic may be virtually absent in some species, while in others, it may occur for values of $D$ approaching or even exceeding 1.0 kPa. It could be argued that if the primary function of stomata is to protect against dangerously low tissue water potentials, then this region may represent the conditions where water potential is above the danger threshold (threshold for onset of cavitation-induced air embolisms in the xylem) and therefore no stomatal closure is required. However, the greater the range of $D$ for which stomatal closure is unnecessary, the greater the under-utilization of the conductive potential of the hydraulic system, because higher $g_{sw}$ and potentially higher CO$_2$ assimilation rates could have been sustained.

The second, and more important, characteristic of the $g_{sw}$ versus $D$ curve that varies across species is the rate of decline of $g_{sw}$ with $D$, or $\partial g_{sw}/\partial D$. Due to the nonlinear dependence of $g_{sw}$ on $P_s$, and the interaction between guard and epidermal cells (see Theory section), this sensitivity changes with $D$. In most observations, $\partial g_{sw}/\partial D$ is negative. The steeper the slope of the tangent at any point on the $g_{sw}$ versus $D$ curve, the more negative $\partial g_{sw}/\partial D$, and the smaller the increase in $E$ for any increment in $D$. Under certain conditions, $\partial g_{sw}/\partial D$ is sufficient to result in no change in $E$ with an increase in $D$ (i.e., constant $E$) and, in some cases is so steep that $E$ actually decreases with an increase in $D$. Therefore, the sensitivity of $\partial g_{sw}/\partial D$ will determine the additional water potential draw down that a plant experiences in association with an increase in evaporative demand. With increasing $E$, leaf water potential will decrease; with decreasing $E$, leaf water potential will increase; when $E$ and $J_l$ are both constant, leaf water potential will remain constant. The magnitude of the change in leaf water potential for any change in $E$ will depend on the hydraulic conductance. The lower the value of $k_{l(e-l)}$, the greater the change in leaf water potential. However, if $k_{l(e-l)}$ is very high, the change in bulk leaf water potential with changing $E$ may be too small to detect, and may therefore appear to be zero. This effect is evident in the results of Nonami et al. (1990), where the water potential of epidermal, subsidiary and mesophyll cells of *Tradescantia virginiana* declined significantly with increasing $E$, even though xylem water potential appeared to remain constant.

Stomatal sensitivity to $D$ falls into three general categories: (1) highly sensitive, to the point of maintaining constant $E$ with increasing $D$; (2) moderately sensitive, allowing some increase in $E$ with increasing $D$; and (3) oversensitive, whereby $E$ decreases with an increase in $D$. Examples of these categories are shown in Figure 6 for leaves of three different rainforest canopy tree species. Although it would seem ideal that stomatal control kept $E$, and hence $\Psi_l$, constant and close to the dew point, it may be too small to de-
threshold of cavitation in the xylem so as to gain the most from a given investment in xylem (Tyree and Sperry 1988, Sperry et al. 1993), this degree of control does not appear to be the general case. Why some species appear capable of very tight control of \( E \) (e.g., \( A. \) peralatum, Figures 6A and 6B) whereas others do not (e.g., \( S. \) sayeri, Figures 6C and 6D), has yet to be fully explained.

The requirements for a decrease in \( g_{sw} \) with increasing \( D \)

If there were no mechanical interaction between guard and epidermal cells, then according to the system outlined in Figure 4, \( P_g \) would always decline with increasing \( D \), and therefore so would \( g_{sw} \) (Figure 7, line 1). The mechanical interaction between epidermal and guard cells presents a problem; in many instances, the mechanical relationship \( m \) is such that an equal reduction in \( P_g \) and \( P_e \) will lead to a net increase in stomatal aperture because of the strong influence of \( P_e \) (see Figure 2). An equal reduction in steady state \( P_g \) and \( P_e \) with increasing \( D \) would occur if guard cells supported no transpiration (Figure 1A(i)), or if the guard cells did support transpiration and the hydraulic conductances \( k_{L(s-e)} \) and \( k_{L(s-g)} \) were similar. This effect is simulated in Figure 7, line 2, where \( g_{sw} \) increases with \( D \), until the draw down in \( \Psi_e \) is sufficient to reduce \( P_e \) to zero, after which further increases in \( D \) result in decreasing \( g_{sw} \). This effect can be induced artificially when \( P_g \) (not \( D \)) is manipulated directly in vitro with epidermal strips bathed in different osmotic solutions (Glinka 1971). The mechanical interaction between guard and epidermal cells has not been quantified for many species, but, for example, it is known to be substantial for \( T. \) virginiana (Figure 2), which always appears to exhibit a reduction in \( g_{sw} \) with increasing \( D \) (Shackel and Brinckmann 1985, Nonami et al. 1990, Franks and Farquhar 2001). In a purely hydraulic feedback model where, as a first approximation, \( \Pi_e \) is maintained constant, it is possible to overcome the tendency for stomata to open with declining epidermal water potential if: (1) guard cells sustain a substantial portion of the evaporative flux, as per Figure 1A(ii); and (2) \( k_{L(s-g)} \) is much less than \( k_{L(s-e)} \). Line 3 in Figure 7 simulates the effect of these conditions, using a value for \( k_{L(s-e)} \) that is ten times higher than that for \( k_{L(s-g)} \). It is therefore possible to explain moderate reductions in \( g_{sw} \) with \( D \) in terms of this latter set of conditions. However, more dramatic reductions in \( g_{sw} \) with \( D \) may be achieved with this feedback system if \( \Pi_e \) does not remain constant, but is sensitive, even slightly, to some component of the hydraulic feedback loop, such as \( P_e \).

Grantz and Zeiger (1986) have proposed that because of the similarity in many cases between the kinetics of stomatal responses to light and \( D \), it is highly probable that the response to \( D \) involves a metabolic component. Line 4 in Figure 7 shows how, in a stomatal control system incorporating hydraulic negative feedback, where guard cells sustain a large portion of the

![Figure 6](image_url) Response of stomatal conductance \( (g_{sw}; A, C, E) \) and transpiration rate \( (E; B, D, F) \) in three tropical rainforest canopy trees to a step change in leaf-to-air vapor pressure difference \( (D) \) from 1.0 to 2.0 kPa. Arrows indicate the time at which the step change was applied. The results illustrate three categories of response, as indicated by the position of the new steady state transpiration rate \( (E) \) relative to \( E \) at 1.0 kPa: (A and B), the reduction in \( g_{sw} \) is such that \( E \) is held almost constant; (C and D), the reduction in \( g_{sw} \) is insufficient to maintain \( E \) constant and \( E \) increases with the increase in \( D \) (most typical type of response); (E and F), the reduction in stomatal conductance to water vapor \( (g_{sw}) \) is so pronounced that \( E \) declines to a value below what it was before the step change in \( D \) (less common response). Measurements were carried out in situ with an open-flow leaf gas exchange analyzer (LI-6400, Li-Cor, Lincoln, NE). Before the step change, leaves were brought to steady state at 1.0 kPa vapor pressure deficit, ambient CO2 concentration 350 µmol mol\(^{-1}\), 30 °C and 1000 µmol m\(^{-2}\) s\(^{-1}\) PAR. Soil was wet from recent sustained heavy rains. Data were collected from sun leaves at the top of the canopy on 35–40 m tall trees. (A and B) = \( A. \) peralatum (Bailey) Edlin; (C and D) = \( S. \) sayeri (F. muell.) B. Hyland; and (E and F) = \( D. \) pettigrewianum Bailey. Data were collected at the Australian Rainforest Canopy Crane facility, Cape Tribulation, Australia.
transpirational flux, the combined effect of low \( k_{L(s–g)} \) and \( \Pi_g \) decreasing from its set point at \( D = 0 \) \((\Pi_{g(max)})\) in proportion to \( P_e \), results in a more dramatic reduction in \( g_w \) with increasing \( D \). Although \( k_{L(s–g)} \) has yet to be measured, there is much indirect evidence to suggest that this last set of conditions is met in many cases in plants that exhibit a substantial reduction in \( g_w \) with increasing \( D \). Figure 8 is one typical example: despite a smooth decline in \( g_w \) following a step increase in \( D \), the presence of a mechanical influence of \( P_e \) on \( g_w \) is seen when \( D \) is returned to its original low value and the sudden restoration in \( P_e \) causes a momentary reduction in \( g_w \) (marked as *). The subsequent rise in \( g_w \) is only slight, suggesting not only that \( \Pi_g \) must have declined during the period of higher \( D \), but that after \( D \) was returned to its former value, \( \Pi_g \) could not do so in the short to medium term.

Combined feedback and feedforward

Through the inherent coupling of \( E \) and \( P_e \) the hydraulic negative feedback loop will always be active in transpiring leaves. Though it may not always be the only mechanism acting to down-regulate \( E \) in the face of increasing evaporative demand (see below), it can produce the more moderate and fully reversible patterns of stomatal response to \( D \). Within limits, it is the hydraulic conductance \( k_{L(s–g)} \) that will account for much of the sensitivity of \( g_w \) to \( D \) in this feedback loop. Franks and Farquhar (1999) derived an expression for this sensitivity in terms of the open loop gain \( H \):

\[
H = \frac{Dg_w}{\epsilon^* k_{L(s–g)}}
\]

where \( \epsilon^* \) is a measure of bulk guard cell elasticity (smaller \( \epsilon^* \) means greater elasticity). The higher the magnitude of \( H \), the higher (in magnitude) the stomatal sensitivity to \( D \). Based on this principle, a plant with lower \( k_{L(s–g)} \) would show greater sensitivity to \( D \), all else being equal. Equation 20 also predicts that plants with higher stomatal conductances will show greater stomatal sensitivity to \( D \), provided their hydraulic properties are not too dissimilar. In accordance with this, Oren et al. (1999) found, from a survey of published data for a range of trees and shrubs, that plants with higher \( g_w \) at low \( D \) did tend to have greater stomatal sensitivity to \( D \).

The simple hydraulic negative feedback mechanism described above can, at best, exhibit sufficient stomatal sensitivity to \( D \) to hold \( E \) constant, although in reality it will fall far short of this because of the likely instabilities associated with the necessarily high loop gain required for nearly constant \( E \). Furthermore, reliance on low hydraulic conductance for greater stomatal sensitivity could place additional stress on the hydraulic system in the form of potentially damaging low xylem water potentials. Alternatively, a reduction in \( k_{L(s–g)} \) as a result of cavitation-induced air embolisms in the xylem may temporarily serve to reduce stomatal conductance and curb the increase in transpiration rate (Figure 9A), but the associated fall in leaf water potential (Figure 9B) will increase the probability of further cavitation events, perhaps leading to the runaway embolism pattern described by Tyree and Sperry (1988). However, it was the emergence of data of the form in Figure 6E and 6F, showing that the reduction in \( g_w \) in response to increasing

**Figure 7.** Simulation of four variations in the properties of the stomatal control mechanism that result in decreasing stomatal conductance \( (g_w) \) with increasing leaf-to-air vapor pressure difference \( (D) \): (1) no mechanical interaction between guard cells and epidermal cells \( (m = 0) \), constant guard cell osmotic pressure \( (\Pi_g; 3.5 \text{ MPa}) \), and no difference between the leaf-area-specific hydraulic conductance from soil to leaf \( (k_{L(s–g)}) \), and from soil to epidermis \( (k_{L(s–e)}) \); (2) with mechanical interaction between guard cells and epidermal cells, constant \( \Pi_g \) \((3.5 \text{ MPa}) \), and no difference between \( k_{L(s–g)} \) \( k_{L(s–e)} \); (3) as for 2, but with \( k_{L(s–g)} \) ten times that of \( k_{L(s–e)} \); and (4) as for 3, but with \( \Pi_g \) decreased from a maximum of 3.5 MPa, in direct proportion to \( P_e \). The conditions in 4, which combine low epidermal-to-guard cell hydraulic conductance with decreasing \( \Pi_g \) produce a response that most closely resembles that typified in Figure 5 for forest trees.

**Figure 8.** Response of *Normanbya normanbyi* stomatal conductance to water vapor \( (g_w) \) to a step change in leaf-to-air vapor pressure difference \( (D) \) from 1.0 to 2.0 kPa, and back to 1.0 kPa. Application of the change is indicated by arrows. Immediately following the return of \( D \) to 1.0 kPa, \( g_w \) showed the common transient “wrong-way” response (marked by *), in this case, a slight decrease before increasing, indicating a mechanical interaction between guard cells and epidermis. Note also that \( g_w \) did not return to its original value, suggesting \( \Pi_g \) changed, and does not recover in the short term. Conditions as for Figure 6.
$D$ could be large enough to result in a decrease in $E$ (Schulze et al. 1972; see also references in Farquhar 1978) that meant hydraulic negative feedback could no longer be regarded universally as the mechanism by which $g_{sw}$ responded to $D$. Several alternative hypotheses have been proposed, but the simplest is that stomata are sensitive to a portion of the transpiration flux that is independent of the stomatal pores. This hypothesis was first proposed by Cowan (1977), who referred to it as feedforward control because, instead of $g_{sw}$ responding to $E$, it responds in this case directly to the environmental factors that promote changes in $E$. For this reason, Cowan has also referred to this mechanism as the direct response of $g_{sw}$ to $D$ (Cowan 1994).

Since it was first proposed as a mechanism to explain the more dramatic reductions in $g_{sw}$ with increasing $D$, the elements of feedforward control of $E$, if present, have been difficult to identify. Furthermore, the phenomenon of decreasing $E$ with increasing $D$ is not easy to induce or reproduce under controlled conditions, free from the complications of other changing conditions such as light, temperature or soil moisture, so identifying the feedforward mechanism is all the more difficult. This unpredictability, and the often sluggish and irreversible nature of the response, is uncharacteristic of the dynamic behavior of most true control systems, and it is perhaps worth questioning whether in some cases of feedforward-like behavior the plant is actually exhibiting symptoms more characteristic of a breakdown in control. These irregularities have been noted elsewhere (Franks et al. 1997, Mencuccini et al. 2000), prompting caution in the interpretation of feedforward-like behavior.

If a feedforward mechanism is indeed present in some plants, then data with which to characterize it in detail are limited. However, the mechanism may prove to be relatively simple. If it can be assumed that: (1) the basis of the stimulus for all modes of response to $D$ is the rate of water loss from the leaf (Mott and Parkhurst 1991); (2) feedforward control comprises the sensing of this stimulus directly, i.e., independent of the component of $E$ that passes through the stomatal pores and therefore independent of bulk leaf water status; and (3) the response includes change in $\Pi_g$, then these components may be linked by the following expression for $\Pi_g$:

$$\Pi_g = \Pi_{g\text{max}} - \frac{cD}{P_a} \tag{21}$$

where $c$ is a constant directly proportional to a diffusive conductance that is supplying the "direct" portion of the stimulus to $D$ (e.g., guard cell or epidermal cuticular conductance). The component $cD/P_a$, in Equation 21, which is the result of translation of the direct stimulus (an evaporative flux) into an osmotic flux, is a feedforward element that can work together with the ever present hydraulic negative feedback system. This dual feedback-feedforward system may be more representative of the true nature of the mechanism of the $g_{sw}$ response to $D$. The effect of incorporating this element into the feedback model is shown in Figure 10, using different values of $c$. These simulations closely resemble the modes of response observed in many species, but the validation of this simple mechanism depends on experimental proof of a linkage between a portion of the cuticular transpiration and $\Pi_g$.

**The potential effects of increasing tree height and the role of stomatal mechanics**

From Equation 5, it can be seen that a potential effect of increasing hydraulic path length is to decrease hydraulic conductance. Although this may be offset by an increase in $K_L$ with height (Pothier et al. 1989), several studies have reported a decline in whole-plant hydraulic conductance with height (Mencuccini and Grace 1996, Ryan et al. 2000, McDowell et al. 2002b). These studies were concerned primarily with hydraulic conductances representative of $k_L(L-G)$, but it could be assumed that because this is included as part of the series constituting $k_L(L-G)$ and $K_{L(L-G)}$, then these too may have decreased in a proportional manner. In addition to this, there is the effect of the downward pull of the vertical head of water in the xylem ($\rho gh$), which appears at the leaf as an additional drop in water...
increasing ΔLs–l on kL(s–l–l). Schafer et al. (2000) concluded that increased A/L partially compensated for the negative effect of increased hydraulic path length on gsw in *Fagus sylvatica* L. McDowell et al. (2002a) reached a similar conclusion in their study of *Pseudotsuga menziesii* and a meta-analysis of data from 13 whole-tree studies. However, if these compensating mechanisms do not completely offset the effects of increasing hydraulic path length, then the decline in kL(s–l–l) with increasing tree height is likely to affect gsw.

From the perspective of the stomatal control mechanisms described above, it is easy to demonstrate how height-related change in kL(s–l–l) and kL(s–g) may translate into changes in gsw. Figure 11 shows one example simulation with fixed Kc over 40 m. The basis of these changes in gsw is a predicted increase in the sensitivity of gsw to D as kL(s–g) decreases (refer to Equation 20). Whether this mechanism is expressed fully, or is compensated by other whole-tree processes, may depend upon environmental and genetic variables. There is clearly much to learn about the mechanism of height-related effects on leaf gas exchange control in trees. In the pursuit of this, special attention should be given to the hydro-mechanical properties of the stomatal apparatus in trees. It can be seen in the simulations of Figure 11C that the mechanical interaction between stomata and epidermal cells potentially has a dramatic effect on the way that height-related alterations to hydraulic conductance will manifest themselves in the form of changes in gsw. With no mechanical interaction, the model predicts a steady decline in gsw with height, the slope of which is steeper the smaller Kc (Figure 11C, dotted line). With strong mechanical interaction, gsw may actually increase over the initial change in height, until P1 reduces to zero, after which it will decline with height (Figure 11C, solid line). Furthermore, detecting the effect of height-induced changes in gsw may depend on the height range and frequency at which gsw is sampled. Too few sample heights, or too small a height range, could fail to detect the true pattern of change in gsw with height, assuming fixed mechanical relationships between guard and epidermal cells. These relationships could themselves change with height. Lastly, any change that tends to reduce the water potential in the canopy, including a height-related reduction in hydraulic conductance or increase in the hydrostatic component pgL, may be counteracted by osmotic adjustment, e.g., an increase in Πe and Πg(max). Osmotic adjustment in response to reduced water potentials caused by drought and increased salinity is well documented (Morgan 1984), but little is known at present about the role of this mechanism in counteracting height-related effects on canopy water potential and productivity as trees grow taller.

Conclusions

The focus of this review has been the linkage between stomatal control and the plant hydraulic system. This relationship underlies the nature of leaf gas exchange regulation, and changes in hydraulic conductances with increasing tree height or adverse environmental conditions may significantly alter gas exchange characteristics. The approach here has been to assume a maximum guard cell osmotic pressure (constrained ulti-

![Figure 10](image-url)

**Figure 10.** Simulation of the addition of a feedforward element to the hydraulic feedback model. Instead of guard cell osmotic pressure remaining constant, it is sensitive to leaf-to-air vapor pressure difference (D). Depending on the magnitude of the sensitivity (corresponding to c in Equation 21), transpiration rate (E) may decrease with increasing D. Stomatal conductance to water vapor = gsw. Other model parameters are as for Figure 7, line 3.

Correlation between stomatal and hydraulic conductance to the transpirational flux has now been widely observed (see Stomatal hydraulics), but less is understood about how stomatal conductance is affected by a decline in hydraulic conductance as trees grow taller. It has been proposed that the stomatal control mechanism is affected directly by these height-related (and by association, age-related) changes to plant hydraulic conductances, and that this could contribute to declining productivity as trees get taller (Ryan and Yoder 1997). The basis of this argument is the possibility that a reduction in hydraulic conductance will lower the water potential in the canopy. However, others have proposed that in some cases adaptive mechanisms could compensate for these height-induced changes (reviewed by Becker et al. 2000). Several possible compensating mechanisms are evident in the combined expression for kL(s–l–l) (from Equations 4 and 5):

\[
k_{L(s–l–l)} = \frac{K_L}{\Delta L_{s–l}} = \frac{A_k K_s}{A_L \Delta L_{s–l}}
\]

For example, in addition to an increase in Kc, an increase in Ks or an increase in the ratio A/L may counteract the effects of potential of about 0.01 MPa per meter of vertical height (Scholander et al. 1965, Zimmermann 1983).

For example, in addition to an increase in Kc, an increase in Ks or an increase in the ratio A/L may counteract the effects of...
mately by genetic characteristics but set on a daily basis mainly by light flux, CO2 concentration and temperature), and to examine the mechanism by which actual stomatal conductance deviates from the corresponding theoretical maximum through the interaction of transpiration rate, hydraulic conductances, leaf tissue water potentials and the mechanical interaction between guard cells and epidermal cells. The extent of this deviation is a measure of the ability of the plant to regulate transpiration rate and hence leaf water potential, and this varies considerably across species. The hydraulic feedback loop comprising, in its simplest expression, the interdependence of transpiration rate and guard cell turgor, could be considered the foundation of this component of stomatal control, and it alone could account for perhaps the most moderate stomatal responses to D. However, for this to work at all, guard cells may have to sustain a considerable portion of the transpirational flux, with $k_{l(e-g)}$ being much lower than $k_{l(e-g)}$ in order to overcome the counteractive influence of the interaction between guard cells and epidermal cell. This requirement is diminished the more mechanically isolated the guard cells are from the epidermis. A linkage between this feedback loop and guard cell osmotic pressure, possibly via the turgor of epidermal or mesophyll cells, could allow for greater sensitivity of $g_{sw}$ to D. The effect on this system if hydraulic conductances change with tree height will depend significantly on both the alteration to the gain of the hydraulic feedback loop and the mechanical interaction between guard cells and epidermal cells. Further insights into these mechanisms will be gained from more quantitative studies of hydraulic conductances at the scale of the stomata–epidermal complex and the whole plant, as well as how these and the hydro-mechanical properties of stomata vary with increasing tree height.

Acknowledgments

I thank Rick Meinzer and Guillermo Goldstein for inviting this review. This work was supported by grants from the Australian Research Council RIEF scheme, and a Harvard University Charles Bul lard Fellowship. I also gratefully acknowledge the support of N.M. Holbrook (Harvard University), D. Foster (Harvard Forest) and R.E. Cook (Arnold Arboretum), and many valuable discussions with G.D. Farquhar.

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