Oecologia (2008) 155:405–415
DOI 10.1007/s00442-007-0918-5

PHYSIOLOGICAL ECOLOGY - ORIGINAL PAPER

Stem and leaf hydraulics of congeneric tree species from adjacent tropical savanna and forest ecosystems

Guang-You Hao · William A. Hoffmann · Fabian G. Scholz · Sandra J. Bucci · Frederick C. Meinzer · Augusto C. Franco · Kun-Fang Cao · Guillermo Goldstein

Received: 17 June 2007 / Accepted: 4 November 2007 / Published online: 30 November 2007

Abstract Leaf and stem functional traits related to plant water relations were studied for six congeneric species pairs, each composed of one tree species typical of savanna habitats and another typical of adjacent forest habitats, to determine whether there were intrinsic differences in plant hydraulics between these two functional types. Only individuals growing in savanna habitats were studied. Most stem traits, including wood density, the xylem water potential at 50% loss of hydraulic conductivity, sapwood area specific conductivity, and leaf area specific conductivity did not differ significantly between savanna and forest species. However, maximum leaf hydraulic conductance (K_{leaf}) and leaf capacitance tended to be higher in savanna species. Predawn leaf water potential and leaf mass per area were also higher in savanna species in all congeneric pairs. Hydraulic vulnerability curves of stems and leaves indicated that leaves were more vulnerable to drought-induced cavitation than terminal branches regardless of genus. The midday K_{leaf} values estimated from leaf vulnerability curves were very low implying that daily embolism repair may occur in leaves. An electric circuit analog model predicted that, compared to forest species, savanna species took longer for their leaf water potentials to drop from predawn values to values corresponding to 50% loss of K_{leaf} or to the turgor loss points, suggesting that savanna species were more buffered from changes in leaf water potential. The results of this study suggest that the relative success of savanna over forest species in savanna is related in part to their ability to cope with drought, which is determined more by leaf than by stem hydraulic traits. Variation among genera accounted for a large proportion of the total variance in most traits, which indicates that, despite different selective pressures in savanna and forest habitats, phylogeny has a stronger effect than habitat in determining most hydraulic traits.

Keywords Plant water relations · Embolism · Vulnerability · Phylogenetic inertia

Communicated by Ram Oren.

G.-Y. Hao · K.-F. Cao
Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences, Menglun, 666303 Mengla, Yunnan Province, China

G.-Y. Hao · G. Goldstein (✉)
Department of Biology, University of Miami, Coral Gables, FL 33124, USA
e-mail: goldstein@bio.miami.edu

W. A. Hoffmann
Department of Plant Biology, North Carolina State University, Raleigh, NC 27695-7612, USA

F. G. Scholz · S. J. Bucci
Consejo Nacional de Investigaciones Cientificas y Técnicas (CONICET), Departamento de Biología, Universidad Nacional de la Patagonia San Juan Bosco, 9000 Comodoro Rivadavia, Argentina

F. C. Meinzer
USDA Forest Service, Forestry Sciences Laboratory, 3200 SW Jefferson Way, Corvallis, OR 97331, USA

A. C. Franco
Departamento de Botanica, Universidade de Brasilia, Caixa Postal 04457, Brasilia, DF 70904970, Brazil

G. Goldstein
Laboratorio de Ecología Funcional, Departamento de Ciencias Biológicas, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Ciudad Universitaria Nuñez, Buenos Aires, Argentina

Introduction

Central Brazil is dominated by vast savannas (Cerrado) subjected to a tropical climate with distinct dry and wet seasons. The savannas are dissected by rivulets, along which gallery forests are distributed. Adjacent Cerrado and forest differ not only in their structure but also in species composition, with very few species being common to both habitats (Felfili and Junior 1992). Frequent fire is one of the main factors that exclude forest species at the savanna–forest boundaries (Hopkins 1992; Swaine et al. 1992). In the absence of fire, however, forest tree species can establish and grow in adjacent savannas, but this process is quite slow (Bowman and Fensham 1991; Ratter 1992). Thus, while fire may impose an absolute constraint on the advance of forest species into the savanna, in its absence other factors may also limit the establishment and success of forest species in savanna environments.

Relative to forests, savanna environments are characterized by high irradiance, high temperature, low soil nutrient availability, large seasonal changes in water availability in the upper soil layers, and high evaporative demand throughout the year (Furley and Ratter 1988; Pivello and Coutinho 1992; Meinzer et al. 1999). We expect that savanna and forest species should exhibit different physiological and ecological traits that permit them to succeed in their respective environments. Thus, even though members of both groups of species can establish in some savanna habitats when fire is excluded, differences in their ecophysiological traits should be large enough to lead to the predominance of savanna over forest species at the savanna–forest boundary.

Specifically, we hypothesize that there are substantial differences between savanna and forest functional types with respect to their water relations. Cerrado trees have a particularly high capacity for maintaining homeostasis of water relations during seasonal drought (Meinzer et al. 1999; Bucci et al. 2005), but it is unknown whether forest species also exhibit this behavior when growing in savanna environments. Traits contributing to homeostasis of water relations include deep root systems to gain access to soil water at depth during the dry season (Jackson et al. 1999), seasonal adjustment in hydraulic architecture (Bucci et al. 2005), high resistance to xylem cavitation, and effective repair of cavitation (Bucci et al. 2003). Relative to Cerrado tree species, forest trees allocate less biomass to roots (Hoffmann and Franco 2003) and have less extensive root systems (Sternberg et al. 2004) but little is known about how other hydraulic traits differ between tree species of these two functional types.

Most studies on hydraulic architecture of savanna trees have focused on characteristics of roots and stems, but relatively few have examined leaf hydraulics. Recent work has shown that resistance to water transport in leaves accounts for 30–80% of the total hydraulic resistance of the whole-plant water transport pathway (Becker et al. 1999; Nardini 2001; Sack et al. 2002), even though the water transport pathway in leaves represents a very small fraction of that in the whole plant. Furthermore, leaves tend to be more vulnerable to embolism than the stems to which they are attached, often losing a substantial fraction of their hydraulic conductance under non-extreme conditions (Brodribb and Holbrook 2004; Woodruff et al. 2007). These findings suggest that leaves can be a major hydraulic bottleneck for plant water transport and thus traits related to water movement in leaves may be critical to the success of plants in water-limited environments.

In the present study, we characterized hydraulic properties and other functional traits for both stems and leaves of six species pairs, each consisting of one savanna species and one forest species of the same genus. Using congeneric species pairs can improve the statistical power of comparisons between two groups of species (Garnier 1992; Ackerly 1999) while ensuring phylogenetic independence, an important condition for comparative studies (Felsenstein 1985). No two genera were from the same family further guaranteeing phylogenetic independence. Moreover, the individuals studied co-occurred in savanna habitats, thus ruling out environmental effects. We predicted that when both savanna and forest species are growing in Cerrado habitats, savanna species have traits that are better adapted to drought-prone environments compared to their forest congeners. Potential adaptations of savanna species include higher resistance to drought-induced loss of hydraulic conductance, maintenance of higher conductance to water flow under field conditions, and larger water storage capacitances in plant tissues to buffer the effect of soil and atmospheric drought.

Materials and methods

Study site and species selection

The study was carried out at the ecological reserve of the Instituto Brasileiro de Geografia e Estatística (IBGE; 15°56′S, 47°53′W, altitude 1,100 m) located 35 km south of Brasilia, Brazil. The site is a seasonal savanna (locally known as Cerrado) with an open to semi-closed canopy composed of evergreen and brevi-deciduous trees and herbaceous understory. Mean annual precipitation is approximately 1,500 mm with a pronounced dry season from May to September, during which less than 100 mm of precipitation occurs in most years. Mean monthly temperature ranges from 19 to 23°C with diurnal temperature ranges of 20°C common during the dry season. Soils are deep, extremely well drained and strongly acid dystrophic oxisols with high aluminum content. The study site had been protected from fire for a sufficiently long period to permit the establishment of forest species in savanna environments.

The study was carried out during the late wet season and dry season of 2006 (May–July). We chose six savanna–forest species pairs based on availability at the study site (Table 1). Each pair consisted of one savanna species and one forest species of the same genus, and no two genera were selected from the same family. Furthermore, we only selected pairs for which both species could be found in savanna, with similar soil, and topographic position. Sampling of branches and leaves was limited to outer portions of tree crowns that were fully exposed to sunlight during the daytime. This sampling design ensured phylogenetic independence while minimizing local site effects.

Wood density, leaf mass per area and leaf water content

Terminal branches with similar diameters from six individuals per species were used for measuring wood density. Sapwood with both bark and pith removed was immersed in tap water overnight to saturate the samples. After the surface was wiped dry, the volume of sapwood was measured immediately using the water-displacement method. Then the sapwood was oven dried at 60°C for 72 h before determining dry mass. For measuring leaf mass per area (LMA) and leaf water content at full turgor (WM/DM; where WM is mass of water, and DM is dry mass), several branches from six individuals of each species were cut and immediately re-cut underwater with the cut end kept under water to allow rehydration for about 2 h. Several leaves of each individual were then weighed and scanned for leaf area and then oven dried for about 48 h before determining their dry mass. The WM/DM at full turgor was expressed on a unit leaf dry mass basis (Brodribb and Holbrook 2003).

Leaf water potential

In mid-July, predawn leaf water potential (Ψ_{pd}) and midday leaf water potential (Ψ_{md}) were measured with a pressure chamber (PMS1000; Corvallis, Oreg.). Leaf samples were taken between 0400 and 0600 hours and 1200 and 1400 hours, respectively. For each of the 12 species, six leaves or terminal branches from six different individuals were cut with a sharp razor blade and sealed immediately in small plastic bags with moist paper towels in them and kept briefly in a cooler until balancing pressures were determined in the laboratory. When taking samples for measuring Ψ_{md}, only the sun-exposed leaves were selected.

Hydraulic conductivity

Stem hydraulic conductivity (K_h) was measured on ten to 16 branches per species. For most species it was possible to obtain each branch from a separate individual, but for *S. pohlii* and *H. martiana*, we were able to locate only five and seven individuals, respectively, that met our site criteria. For these species, two branches were collected from each of these individuals. We used unbranched segments of ca. 20–25 cm (mean = 23 cm). Samples were collected early in the morning, during the transition from the wet to dry seasons (May–June). These were re-cut immediately under water to avoid embolism and were transported to the laboratory with the cut end immersed in water and the free end tightly covered with opaque plastic bags. Distilled and degassed water was used as the perfusion fluid. Relatively low hydrostatic pressure generated by a constant hydraulic head of 50 cm was applied to avoid refilling of seasonally embolized vessels. Because some species have strong wound reactions that cause clogging of vessels, both ends of a segment were shaved with a sharp razor blade immediately before each measurement of flow rate. Then methyl blue dye was perfused into each end of the branch segment under a pressure head of 50 cm. Sapwood area was determined at approximately 1 cm from each end by measuring the stained cross-sectional area. The geometric mean of these two values was used to represent sapwood area in the following calculations. Hydraulic conductivity (kg m s^{-1} MPa^{-1}) was calculated as:

$$K_h = J_v/(\Delta P/\Delta L)$$

where J_v is flow rate through the segment (kg s^{-1}) and $\Delta P/\Delta L$ is the pressure gradient across the segment (MPa m^{-1}). Sapwood conducting area (A_{sw}) and distal leaf area (A_l) were measured to calculate Huber values (HV) (Tyree and Ewers 1991):

$$\mathrm{HV} = A_{sw}/A_l.$$

Specific hydraulic conductivity (K_s; kg m^{-1} s^{-1} MPa^{-1}) was calculated as the ratio of K_h to A_{sw} and leaf-specific hydraulic conductivity (K_l; kg m^{-1} s^{-1} MPa^{-1}) was calculated as the ratio of K_h to A_l.

Stem xylem vulnerability curve

Stem vulnerability curves were determined by measuring percentage loss of hydraulic conductivity (PLC) due to embolism over a range of water potential reached during dehydration by the bench drying method (Sperry et al. 1988). Because leaves of the two *Aegiphila* spp. tended to disconnect from stems when slightly dehydrated, it was not possible to determine the stem vulnerability curves for this

Table 1 Savanna and forest congeneric species investigated in this study

Savanna species	Forest species	Family
Styrax ferrugineus Nees & Mart.	*Styrax pohlii* A.DC	Styracaceae
Hymenaea stignocarpa Mart. Ex Hayne	*Hymenaea martiana* Hayne.	Leguminosae
Myrsine guianensis (Aubl.) Kuntze	*Myrsine ferruginea* Ruiz & Pav.	Myrsinanceae
Symplocos lanceolata (Mart.) A.DC	*Symplocos mosenii* Brand.	Symplocaceae
Miconia pohliana Cogn.	*Miconia cuspidata* Naud.	Melastomataceae
Aegiphila lhotzkiana Cham.	*Aegiphila sellowiana* Cham.	Verbenaceae

genus. Before dawn, long branches were cut from four to six individuals of each species. These branches were wrapped in dark plastic bags containing wet paper towels to prevent further desiccation. In the laboratory these branches were allowed to dry for different periods of time to reach a large range of water potential. The branches were then sealed into double layers of bags with wet paper towels for at least 2 h to equilibrate. Leaf water potentials for two to three leaves were then measured using a pressure chamber (Scholander et al. 1965) and one stem segment was cut under water and connected to tubing apparatus (Tyree and Sperry 1989). Water used in this system was filtered (0.2 μm) and degassed using a partial vacuum shortly before use. For a fixed pressure head, K_h is proportional to volumetric flow rate (J_v) of water through stem segments, so calculations of PLC were based on J_v rather than K_h. After the initial measurement of flow rate (J_i) the segments were flushed with 0.1 MPa pressure for 20–60 min to remove embolisms until stable readings of flow rate were reached. Then the maximum flow rate (J_{max}) was measured, using the same pressure head as before. PLC was then calculated as:

$$\mathrm{PLC} = 100(J_{\max} - J_i)/J_{\max}.$$

Leaf vulnerability curve

Leaf hydraulic vulnerability curves were determined by measuring leaf hydraulic conductance (K_{leaf}) using the partial rehydration method described by Brodribb and Holbrook (2003). The measurement was based on the analogy between rehydration of desiccated leaves and charging of a capacitor through a resistor as follows:

$$K_{\mathrm{leaf}} = C \ln(\Psi_0/\Psi_f)/t$$

where C is leaf capacitance, Ψ_0 leaf water potential before rehydration, and Ψ_f is leaf water potential after rehydration for t seconds. Capacitance values both before and after turgor loss point (π^0) were calculated from leaf pressure–volume relations (Tyree and Hammel 1972) and are expressed in absolute terms and normalized by leaf area using the following equation:

$$C = \Delta\mathrm{RWC}/\Delta\Psi_{\mathrm{L}} \times (\mathrm{DM/LA}) \times (\mathrm{WM/DM})/M$$

where RWC is leaf relative water content, DM leaf dry mass (g), LA leaf area (m^2), WM (g) mass of leaf water at 100% RWC (WM = fresh mass − dry mass), and M is molar mass of water (g mol^{-1}). The two species of *Miconia* were not included because their petioles were too small and not strong enough for measuring water potential with single leaves.

Data analysis

An electric circuit analog model was used to predict how leaves of different species differed in terms of buffering leaf water potential (Ψ_L) from dropping to a critical value at which strong stomatal control of water loss must be initiated. Briefly, under a given rate of transpiration the time lag (Δt) for Ψ_L to drop from a value equal to Ψ_{pd} to a critical value (Ψ_c), was calculated as:

$$\Delta t = C(\Psi_{\mathrm{pd}} - \Psi_{\mathrm{c}})/E$$

where E is an assumed transpiration rate (3 mmol m^{-2} s^{-1}) that is typical for Cerrado species (Meinzer et al. 1999; Bucci et al. 2005). The Ψ_c values used were the points at which K_{leaf} had fallen by 50% ($P50_{leaf}$) for species in the genera *Styrax*, *Hymenaea*, *Myrsine*, and *Symplocos*, and turgor loss point osmotic potentials (π^0) for the genera *Miconia* and *Aegiphila*. As indicated above, $P50_{leaf}$ was not determined for the genus *Miconia*, and in the genus *Aegiphila* it was deemed inappropriate to use $P50_{leaf}$ because it was more negative than π^0 in *Aegiphila lhotzkiana*. Larger Δt-values indicated that the leaves of a given species were more buffered from dropping to a water potential level where stomata closure is likely to strongly limit transpiration and consequently CO_2 assimilation.

Differences between the two species within each genus were assessed with *t*-tests. Paired *t*-tests were used to assess differences between the two functional types across all the genera using the species mean values of each trait. To compare how much of the interspecific variance of a trait can be attributed to the differences among genera or between the two functional types, fractions of total interspecific variance (r^2) that is explained by phylogeny (genus) and by functional type (savanna vs. forest) were calculated for each trait (Hoffmann and Franco 2003). The values were calculated from factorial ANOVAs, following Rosenthal and Rosnow (1985) as:

$$r_X^2 = \mathrm{SS}_X/\mathrm{SS}_{\mathrm{total}}$$

where SS_X and $\mathrm{SS}_{\mathrm{total}}$ are the sum of squares for factor X (functional type or genus) and the total sum of squares, respectively.

Results

Wood density, K_s, K_l and xylem water potential at 50% loss of hydraulic conductivity ($P50$) were not significantly different between savanna and forest species (Table 2). Wood density spanned a relatively large range from 0.38 g cm^{-3} (*Symplocos rhamnifolia*) to 0.74 g cm^{-3} (*Miconia cuspidata*) as did $P50$, which ranged from −1.5 MPa (*S. rhamnifolia*) to −3.4 MPa (*M. cuspidata*). A negative linear relationship was observed between $P50$ and wood density (ρ_{wood}) across the five pairs of species measured, indicating that higher ρ_{wood} was associated with higher resistance to embolism (Fig. 1). HVs tended to be higher in savanna species although differences between the two functional types were marginally significant by paired *t*-test ($P = 0.054$) (Table 2).

LMA was significantly higher in savanna species (*t*-test, $P < 0.05$) in all congeneric pairs and the overall difference between savanna and forest functional types was significant ($P < 0.05$; Table 3). C calculated from pressure–volume curves in the region above the turgor loss point, was significantly greater in savanna species ($P < 0.05$) and maximum K_{leaf} estimated from leaf vulnerability curves was marginally greater in savanna species ($P = 0.09$). No significant differences were found between functional types in WM/DM and the leaf water potential corresponding to $P50_{\mathrm{leaf}}$ (Table 3).

The Ψ_{pd} was significantly higher in savanna species compared to forest species in all the congeneric pairs (Table 4). However, Ψ_{md} did not differ significantly between the two functional types. The osmotic potential at full turgor (π^{100}) and at turgor lost point (π^0) calculated from pressure–volume relations were not significantly different between the two functional types. The calculated time lag (Δt) for leaf water potential to drop from predawn values to critical values close to $P50_{\mathrm{leaf}}$ or π^0 was significantly longer for savanna species (Table 4).

When the leaf and stem trait data were analyzed in a way that allowed the total interspecific variances to be partitioned between genus and functional type, the interspecific variances attributable to phylogeny (genus) were considerably high (Table 5) for all traits, even in those where significant (marginally significant) differences were detected between the two functional types (e.g., in HV, LMA, and C). This result indicated the conservatism of functional traits within closely related species and showed the importance of phylogenetic independence in comparative studies.

Leaves were more vulnerable to loss of hydraulic conductance than stems (cf. $P50_{\mathrm{leaf}}$ and $P50$; Tables 2, 3). Values of K_{leaf} at midday, inferred from the leaf hydraulic vulnerability curves, were substantially lower than the maximum K_{leaf} or even close to zero (Fig. 2). The Ψ_{md} was higher than π^0 in all species, with the exception of *Aegiphila sellowiana*, indicating that leaves of these species still maintained turgor at midday during the middle of the

Table 2 Stem functional traits of savanna (*S*)–forest (*F*) congeneric species (values are mean ± 1 SE). Values followed by *different letters* were significantly different between the two congeneric species at $P < 0.05$. ρ_{wood} Wood density, *HV* Huber values, K_s specific hydraulic conductivity, K_l leaf area specific hydraulic conductivity, *P50* xylem water potential at 50% loss of stem hydraulic conductivity

Species		ρ_{wood} (g cm^{-3})	HV[a] ($\times 10^{-4}$)	K_s (kg m^{-1} s^{-1} MPa^{-1})	K_l ($\times 10^{-4}$ kg m^{-1} s^{-1} MPa^{-1})	$P50$ (MPa)
Styrax	S	0.49 ± 0.009 a	1.89 ± 0.29	2.59 ± 0.43	4.87 ± 0.97	−3.35
	F	0.54 ± 0.006 b	2.02 ± 0.35	3.45 ± 0.47	6.90 ± 1.38	−2.00
Hymenaea	S	0.72 ± 0.007 a	1.80 ± 0.13	3.59 ± 0.82	6.56 ± 1.74	−3.17
	F	0.56 ± 0.010 b	1.46 ± 0.21	2.94 ± 0.80	4.12 ± 1.44	−2.80
Myrsine	S	0.52 ± 0.015 a	1.81 ± 0.23	2.00 ± 0.13 a	3.84 ± 0.69	−2.12
	F	0.62 ± 0.013 b	1.47 ± 0.16	3.25 ± 0.54 b	4.34 ± 1.14	−3.08
Symplocos	S	0.38 ± 0.008 a	2.78 ± 0.59	1.62 ± 0.19 a	4.17 ± 0.85 a	−1.50
	F	0.48 ± 0.012 b	1.88 ± 0.32	4.78 ± 0.66 b	8.56 ± 1.69 b	−1.60
Miconia	S	0.53 ± 0.007 a	2.89 ± 0.91 a	4.73 ± 0.88	10.92 ± 2.63 a	−3.10
	F	0.74 ± 0.004 b	0.75 ± 0.08 b	5.37 ± 0.74	3.44 ± 0.60 b	−3.40
Aegiphila	S	0.49 ± 0.017 a	1.50 ± 0.17	6.17 ± 1.31 a	8.46 ± 1.49 a	–
	F	0.41 ± 0.007 b	1.26 ± 0.13	13.78 ± 2.25 b	16.83 ± 2.80 b	–

[a] When tested across all genera, only HV was marginally different between S and F functional types ($P = 0.054$)

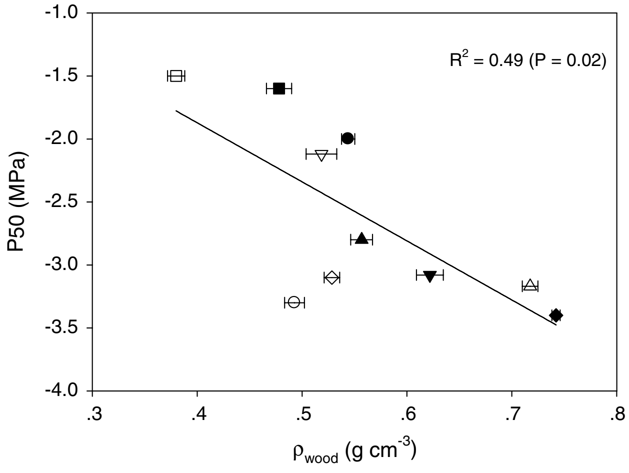

Fig. 1 Relationship between wood density (ρ_{wood}) and the stem xylem water potential causing 50% loss of hydraulic conductivity (*P50*). Each point represents one species (*open symbols* savanna species, *filled symbols* forest species) of the genera *Styrax* (*open circle*, *filled circle*), *Hymenaea* (*open triangle*, *filled triangle*), *Myrsine* (*open inverted triangle*, *filled inverted triangle*), *Symplocos* (*open square*, *filled square*), and *Miconia* (*open diamond*, *filled diamond*). *Error bars* show ±1 SE (n = 6)

dry season (Table 4). In *Styrax*, *Symplocos* and *Miconia*, Ψ_{md} was well above π^0, whereas in the remaining genera Ψ_{md} was quite close to π^0 (Table 4, Fig. 2), suggesting that different genera had different "safety margins" for avoiding turgor loss. The inferred K_{leaf} at π^0 ranged from about twofold lower than maximum K_{leaf} (Fig. 2j) to almost total loss (Fig. 2d, h, i).

Discussion

Leaf versus stem properties and functional coordination across tissues and species

Most stem traits including ρ_{wood}, $P50$, K_{s} and K_{l} did not differ significantly between savanna and forest species (Tables 2, 3). Wood density in many tree species from different ecosystems is correlated with a suite of hydraulic-related characteristics such as stem water storage capacity, the efficiency of xylem water transport, regulation of leaf water status, and avoidance of turgor loss (Meinzer 2003; Bucci et al. 2004a; Gartner and Meinzer 2005). Sapwood water storage capacity and resistance to drought-induced xylem cavitation exhibit opposite trends with variation in ρ_{wood}; sapwood water storage capacity decreases as ρ_{wood} increases (Stratton et al. 2000; Scholz et al. 2007), but xylem becomes more resistant to drought-induced cavitation with increasing ρ_{wood} (Hacke et al. 2001). Due to these trade-offs, either high or low ρ_{wood} can be adaptive in a given environment.

In contrast to stem hydraulic traits, leaf properties such as maximum K_{leaf} and C were significantly higher in savanna trees compared to their forest counterparts, suggesting that the greater leaf hydraulic efficiency may be more critical than stem hydraulics in adapting to the drought-prone Cerrado environment. Leaves are a major bottleneck in the whole-plant hydraulic continuum and K_{leaf} is functionally correlated with leaf water flux-related structural traits and leaf gas exchange (Sack and Holbrook 2006). Maximum K_{leaf} is highly variable among species

Table 3 Leaf functional traits of S–F congeneric species. Values followed by *different letters* were significantly different between the two congeneric species at $P < 0.05$. *LMA* Leaf mass per area, *WM/DM* leaf water content at full turgor, *C* leaf capacitance, K_{leaf} maximum leaf hydraulic conductance, $P50_{leaf}$ leaf water potential at 50% loss of K_{leaf}; for other abbreviations, see Table 2

Species		LMA$^{\mathrm{a}}$ (g m^{-2})	WM/DM (g g^{-1})	C^{a} (mmol m^{-2} MPa^{-1})	Maximum $K_{\mathrm{leaf}}^{\mathrm{b}}$ (mmol m^{-2} s^{-1} MPa^{-1})	$P50_{\mathrm{leaf}}$ (MPa)
Styrax	S	176.1 ± 8.8 a	1.14 ± 0.03	561.8 ± 58.4 a	48.7	−1.2
	F	122.2 ± 2.0 b	1.10 ± 0.02	443.3 ± 55.4 b	57.5	−1.4
Hymenaea	S	145.2 ± 4.0 a	1.16 ± 0.05	499.9 ± 71.1 a	55.5	−1.6
	F	103.6 ± 2.3 b	0.90 ± 0.02	209.9 ± 32.6 b	28.5	−1.4
Myrsine	S	194.1 ± 5.1 a	1.57 ± 0.04 a	1,023.8 ± 120.3 a	75.5	−1.1
	F	86.9 ± 7.9b	1.75 ± 0.22 b	489.9 ± 86.2 b	36	−1.0
Symplocos	S	145.2 ± 6.4 a	1.56 ± 0.05 a	470.0 ± 7.9 a	15.4	−1.3
	F	122.9 ± 4.8 b	1.95 ± 0.07 b	415.7 ± 28.4 b	14.7	−1.3
Miconia	S	184.2 ± 6.0 a	1.06 ± 0.02 a	767.2 ± 89.9 a	–	–
	F	96.6 ± 2.0 b	1.23 ± 0.02 b	361.5 ± 57.0 b	–	–
Aegiphila	S	95.6 ± 3.4 a	2.28 ± 0.07 a	1,169.3 ± 113.3 a	34.4	−0.8
	F	78.2 ± 3.0 b	2.71 ± 0.11 b	1,683.6 ± 131.4 b	21.7	−1.7

[a] For comparisons across all genera, significant differences between the two functional types ($P < 0.05$)

[b] For comparisons across all genera, difference was marginal ($P = 0.09$)

Table 4 Predawn leaf water potential (*Ψ_{pd}*) and midday leaf water potential (*Ψ_{md}*), leaf osmotic potential at full turgor (*π^{100}*), turgor loss point (*π^{0}*), and calculated time lag (*Δt*) for leaf water potential to drop from the value equal to Ψ_{pd} to a critical value under a given value of transpiration rate. Values followed by *different letters* were significantly different between the two congeneric species at $P < 0.05$. For other abbreviations, see Table 2

Species		Ψ^{a}_{pd} (MPa)	Ψ_{md} (MPa)	π^{100} (MPa)	π^{0} (MPa)	Δt^{a} (s)
Styrax	S	−0.44 ± 0.03 a	−1.41 ± 0.08 a	−2.12 ± 0.07	−2.49 ± 0.06	143
	F	−0.70 ± 0.04 b	−1.73 ± 0.13 b	−2.07 ± 0.07	−2.46 ± 0.15	103
Hymenaea	S	−0.17 ± 0.05 a	−2.13 ± 0.12 a	−2.35 ± 0.20	−2.64 ± 0.17	233
	F	−0.66 ± 0.09 b	−2.31 ± 0.13 b	−2.03 ± 0.14	−2.32 ± 0.13	52
Myrsine	S	−0.36 ± 0.02 a	−1.54 ± 0.04 a	−1.53 ± 0.10	−1.76 ± 0.11	253
	F	−0.60 ± 0.05 b	−1.22 ± 0.04 b	−1.63 ± 0.08	−1.79 ± 0.08	66
Symplocos	S	−0.25 ± 0.03 a	−1.18 ± 0.07 a	−1.29 ± 0.07 a	−1.45 ± 0.11 a	164
	F	−0.27 ± 0.03 a	−1.18 ± 0.05 a	−1.68 ± 0.09 b	−1.95 ± 0.10 b	143
Miconia	S	−0.44 ± 0.03 a	−1.42 ± 0.07 a	−1.42 ± 0.08 a	−1.75 ± 0.08 a	568
	F	−0.53 ± 0.04 b	−1.14 ± 0.04 b	−2.23 ± 0.15 b	−2.66 ± 0.18 b	146
Aegiphila	S	−0.45 ± 0.04 a	−1.04 ± 0.08 a	−1.05 ± 0.07	−1.25 ± 0.08	292
	F	−0.76 ± 0.05 b	−1.33 ± 0.06 b	−1.08 ± 0.05	−1.20 ± 0.06	274

[a] For comparisons across all genera, significant differences between the two functional types ($P < 0.05$)

Table 5 Fraction of total interspecific variance (r^2) that is explained by phylogeny (*Genus*) and by functional type (S vs. F). For other abbreviations and superscripts see Tables 2, 3 and 4

Stem and leaf traits	Factor	
	Functional type	Genus
ρ_{wood}	0.03	0.62
HV^{b}	0.31	0.27
K_s	0.12	0.69
K_l	0.02	0.54
$P50$	0.003	0.69
LMA^{a}	0.53	0.29
WM/DM	0.02	0.93
C^{a}	0.03	0.79
Maximum K^{b}_{leaf}	0.14	0.65
$P50_{leaf}$	0.10	0.31
Ψ^{a}_{pd}	0.43	0.39
Ψ_{md}	0.001	0.93
π^{100}	0.03	0.79
π^{0}	0.03	0.80
Δt^{a}	0.37	0.42

(Brodribb and Holbrook 2005) and responds to environmental factors such as irradiance (Sack and Frole 2006). Due to its ecological importance to plant water relations and its high variability, maximum K_{leaf} might be one of the most important traits that explain the differential adaptation of savanna and forest functional types to drought.

Leaf capacitance is positively correlated with K_{leaf} in northern temperate tree and climber species (Sack et al. 2003; Sack and Tyree 2005). Although the water stored in leaves accounts for only a small fraction of daily transpiration, it may play an important role in buffering the change of leaf water potential as transpiration rate and root water supply fluctuate (Sack and Tyree 2005). Atmospheric evaporative demand, rather than soil water availability, tends to dominate patterns of water use in adult trees in the Cerrado (Meinzer et al. 1999). Thus the significantly higher *C* values in savanna species (Table 3) compared to forest species could be an adaptive trait for plants growing in Cerrado environments. The significantly higher calculated time lag (Δt) in savanna species (Table 4) further suggests that savanna species are more buffered from rapid changes in leaf water potential, mainly due to their larger *C* values. Similar to *C*, LMA was also higher in savanna species (Table 3); however, this trait is probably independent of K_{leaf} ($r^2 = 0.35$, $P = 0.07$) as found in temperate deciduous, Mediterranean, and tropical rainforest species (Tyree et al. 1999; Nardini 2001; Sack et al. 2003, 2005), and probably it is more relevant to plant carbon economy, growth, and nutrients (Hoffmann et al. 2005).

Some traits related to drought resistance, such as those derived from pressure–volume relations (e.g., π^{100} and π^{0}), were shown to be independent of K_{leaf} (Sack et al. 2003). Thus, it is not surprising that WM/DM, π^{100} and π^{0} were not significantly different between the two functional types in this study, while K_{leaf} was still higher in savanna species. When growing in savanna environments, forest species may adjust their osmotic characteristics more readily, but adjustments in K_{leaf} are likely to involve changes in the leaf vasculature that are probably under more rigid genetic control or have less acclimation capacity, thus imposing limits on the success of forest species in savanna–forest boundary environments.

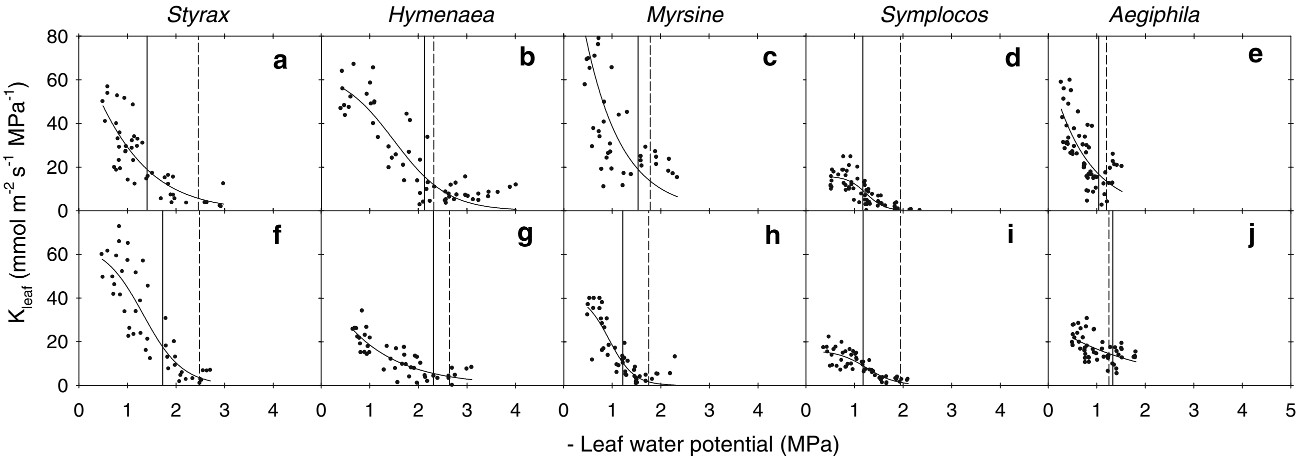

Fig. 2a–j Leaf hydraulic conductance (K_{leaf}) as a function of leaf water potential (Ψ_L) for five pairs of congeneric species. **a–e** Savanna species; **f–j** forest species. Each point represents the average K_{leaf} from two leaves of a single branch. A sigmoid function was fitted to the data. *Vertical solid lines* indicate midday leaf water potential, and *dashed lines* show Ψ_L at the turgor loss point

In both savanna and forest species, leaves were more vulnerable to drought-induced loss of hydraulic function than stems, with $P50_{leaf}$ being substantially higher than $P50$ of stems in all species (Tables 2, 3). The greater hydraulic vulnerability of leaves compared to stems is consistent with results from other studies, which showed that K_{leaf} can be depressed by moderate environmental stresses, such as high irradiance and low relative humidity at midday (Brodribb and Holbrook 2004). Based on the leaf vulnerability curves, predicted midday K_{leaf} values were much lower than the maximum values in each species (Fig. 2). Although other mechanisms may be involved, the decrease in K_{leaf} at midday was probably caused by embolism within the leaf xylem. In that case, embolism repair should occur in leaves of both savanna and forest species in order for them to recover from midday minimum K_{leaf} values, perhaps by a mechanism similar to the one suggested by Bucci et al. (2003) involving the production of osmotically active solutes, which would result in a transient pressure imbalance inside leaf petioles, leading to radial water movement and refilling of embolized vessels. However, direct measurement of the daily course of K_{leaf} is needed to further demonstrate the diurnal dynamic of embolism formation and repair processes in leaves.

To test whether the various leaf and stem hydraulic traits are functionally correlated as found in previous studies (Bucci et al. 2004a), we did all pairwise correlation analysis between functional traits and some were found to be significantly correlated. The negative correlation between $P50$ and wood density (Fig. 1) that has also been found in plants of other ecosystems may indicate a trade-off between hydraulic safety and efficiency (Hacke et al. 2001; Jacobsen et al. 2005). The coordination between π^0 and leaf water content (Fig. 3) suggests that leaves with lower water content had an increased ability to tolerate lower water potentials without losing turgor, while leaves with higher water content had greater ability to maintain higher water potentials.

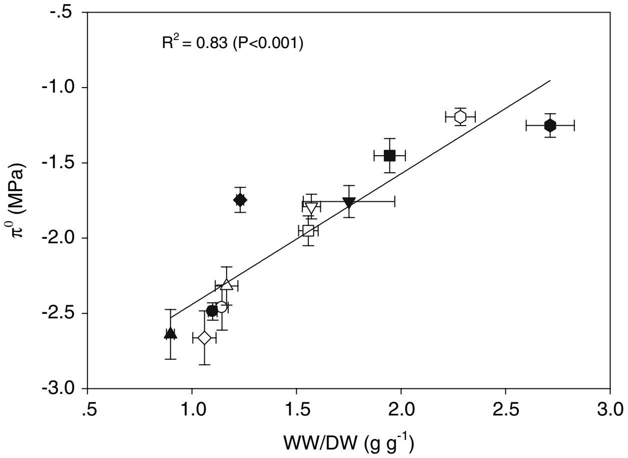

Fig. 3 Relationship between turgor loss point (π^0) and leaf water content per unit leaf dry mass at full turgor (*WM/DM*). *Open hexagon* Savanna species of *Aegiphila*, *filled hexagons* forest species of *Aegiphila*; *other symbols* are the same as in Fig. 1. *Error bars* show ±1 SE (n = 5–6)

Most interestingly, the positive correlation found between K_l of stems and C probably reflects the coordination between stem and leaf hydraulics (Fig. 4). Recent studies have shown that there is a strong positive correlation between C and K_{leaf} (Sack and Tyree 2005), and leaves with high K_{leaf} tend to close stomata relatively slowly after excision (Aasamaa et al. 2001). Thus, under the savanna environment, leaves with high C should be more likely to have higher stomatal conductance, transpiration rate and K_{leaf} than congeneric forest species. Correspondingly, higher rates of transpiration in species of higher leaf

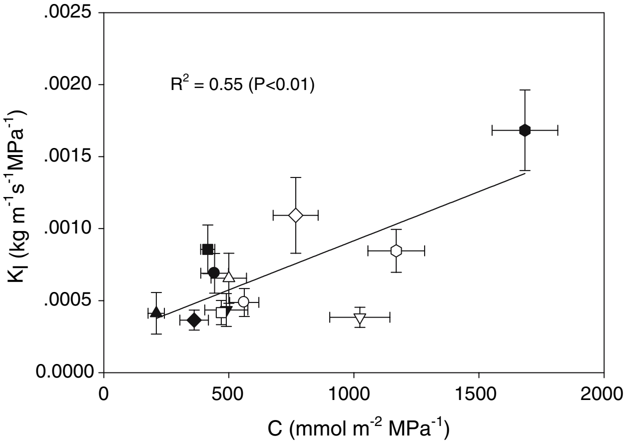

Fig. 4 Relationship between leaf area specific hydraulic conductivity (K_l) of terminal branches and area weighted leaf absolute capacitance (*C*). *Symbols* as in Figs. 1 and 3. *Error bars* show ±1 SE (n = 10–16 for K_l and n = 6 for *C*)

capacitance require terminal branches with higher K_l to support the higher rate of water use. These results suggest a strong selective pressure for hydraulic coordination between different plant organs.

No significant correlations were found either between stem hydraulic parameters (K_s or K_l) and K_{leaf} or between $P50$ of terminal branches and $P50_{leaf}$. Although this is surprising at first sight, these results are consistent with our finding that leaves are much more vulnerable to loss of hydraulic function than stems. The $P50_{leaf}$ in each species is relatively high regardless of the stem $P50$ values, which may have resulted in the weak correlation between these two parameters. It is suggested in the literature that the resistance to water transport in leaves is “disproportionately” high compared to other plant organs (Sack et al. 2003). In other words, leaves are the bottleneck in the plant water transport pathway, which may be responsible for the independence of K_{leaf} and K_h.

Comparison between savanna and forest species

It seems that the savanna and forest species growing in the Cerrado environment differ mainly in leaf hydraulic properties rather than stem traits. Furthermore, differences in rooting depth and hydraulic architecture are likely to be important in determining the relative success of savanna over forest species in the Cerrado environments.

Cerrado woody plants have the ability to access deeper water resources relative to forest species (Hoffmann et al. 2004). The less negative Ψ_{pd}s found in savanna species compared to their congeneric forest counterparts (Table 4) were consistent with the prediction that savanna species have deeper roots, enabling them to access moister soil at depth. It is possible that Ψ_{pd} may not reflect the weighted average soil water potentials due to soil–plant water potential disequilibrium resulting from nocturnal transpiration (Bucci et al. 2004b). However, as the differences in Ψ_{pd} were relatively large in most genera, it is reasonable to expect that differences in Ψ_{pd} among congeneric pairs largely reflect differences in their patterns of water uptake and rooting depth. It has been hypothesized that root:shoot ratio and LMA can evolve relatively quickly apparently requiring few genetic changes (Chapin et al. 1993). The observation that these traits are evolutionarily labile is consistent with our findings that LMA and Ψ_{pd} were two of the traits that were least conserved within genera (Table 5).

The higher HV of savanna compared to forest species (P = 0.054) indicates that savanna species have more sapwood to transport water per unit leaf area. In Cerrado environments with potentially high transpirational demand, higher HV can help in maintaining water balance during the dry season (Bucci et al. 2005). The high HV of savanna species is consistent with the low leaf area index observed in Cerrado ecosystems (Hoffmann et al. 2005). The difference in HV between the two groups of species growing in similar environmental conditions suggests that the divergence in hydraulic architecture is determined by genetic differences, rather than by environment.

Phylogenetic inertia

Contrary to our predictions, most of the stem traits and many leaf traits were not significantly different between savanna and forest tree species. Most of the interspecific variances in these functional traits can be attributable to phylogeny (genus) rather than to functional type (savanna or forest trees) (Table 5), despite the large differences in the physical environments where savanna and forest species typically occur, indicating high levels of phylogenetic inertia in closely related savanna and forest species. These findings are consistent with a previous study by Hoffmann and Franco (2003) examining seedling characteristics such as seed mass, relative growth rate, and leaf area ratio.

Conclusion

The main mechanistic findings of this comparative study were that leaves across all genera are more vulnerable to drought-induced loss of hydraulic function compared to stems, and that leaf hydraulic characteristics differ more consistently between savanna and forest species than stem hydraulic characteristics. We suggest that in the Cerrado environment with high evaporative demand and potentially low soil water availability, leaf hydraulic traits are under stronger selective pressure than are stem hydraulic traits. Higher K_{leaf} and leaf capacitance in savanna species may thus be the most important traits that enable these species to succeed in the Cerrado environment. An electric circuit analog model predicted that compared to forest species, savanna species took longer for their leaf water potentials to drop from predawn values to critical values, suggesting that savanna species were more buffered from changes in leaf water potentials. Besides stems and leaves, roots are also likely to play an important role in the hydraulics of the whole plant and need to be addressed in future investigations. Although the differences between savanna and forest species were significant with regard to some important hydraulic traits, variation among genera still accounted for a large proportion of the total variance, which suggests the prevalence of phylogenetic conservatism in many plant hydraulic traits.

Acknowledgements This work was supported by grants from the National Science Foundation (USA) under grant number DEB-0542912, the A. W. Mellon Foundation, and CNPq (Brazil). We are grateful to personnel of the IBGE ecological reserve for logistic support. We thank Ryan Adasme and Eric Manzané for field assistance. We thank Yong-Jiang Zhang and Juan Pablo Giraljo for suggestions on laboratory methods. This work complies with Brazilian law.

References

Aasamaa K, Sober A, Rahi M (2001) Leaf anatomical characteristics associated with shoot hydraulic conductance, stomatal conductance and stomatal sensitivity to changes of leaf water status in temperate deciduous trees. Aust J Plant Physiol 28:765–774

Ackerly DD (1999) Comparative plant ecology and the role of phylogenetic information. In: Press MC, Scholes JD, Barker MG (eds) Physiological plant ecology. Cambridge University Press, Cambridge, pp 391–413

Becker P, Tyree MT, Tsuda M (1999) Hydraulic conductances of angiosperms of versus conifers: transport sufficiency at the whole-plant level. Tree Physiol 19:445–452

Bowman DM, Fensham RJ (1991) Response of a monsoon forest–savanna boundary to fire protection, Weipa, northern Australia. Aust J Ecol 16:111–118

Brodribb TJ, Holbrook NM (2003) Stomatal closure during leaf dehydration, correlation with other leaf physiological traits. Plant Physiol 132:2166–2173

Brodribb TJ, Holbrook NM (2004) Diurnal depression of leaf hydraulic conductance in a tropical tree species. Plant Cell Environ 27:820–827

Brodribb TJ, Holbrook NM (2005) Leaf hydraulic capacity in ferns, conifers and angiosperms: impacts on photosynthetic maxima. New Phytol 165:839–846

Bucci SJ, Scholz F, Goldstein G, Meinzer F, Sternberg L (2003) Dynamic changes in hydraulic conductivity in petioles of two savanna tree species: factors and mechanisms contributing to the refilling of embolized vessels. Plant Cell Environ 26:1633–1645

Bucci SJ, Goldstein G, Meinzer FC, Scholz FG, Franco AC, Bustamante M (2004a) Functional convergence in hydraulic architecture and water relations of tropical savanna trees: from leaf to whole plant. Tree Physiol 24:891–899

Bucci SJ, Scholz F, Goldstein G, Meinzer FC, Hinojosa JA, Hoffmann WA, Franco AC (2004b) Processes preventing nocturnal equilibration between leaf and soil water potential in tropical savanna tree species. Tree Physiol 24:1119–1127

Bucci SJ, Goldstein G, Meinzer FC, Franco AC, Campanello P, Scholz FG (2005) Mechanisms contributing to seasonal homeostasis of minimum leaf water potential and predawn disequilibrium between soil and plant water potential in Neotropical savanna trees. Trees 19:296–304

Chapin FS, Autumn K, Pugnaire F (1993) Evolution of suites of traits in response to environmental stress. Am Nat (Suppl) 142:S78–S92

Felfili JM, Junior MCDS (1992) Floristic composition, phytosociology and comparison of cerrado and gallery forest at Fazenda Agua Limpa, Federal District, Brazil. In: Furley PA, Procter J, Ratter JA (eds) Nature and dynamics of forest-savanna boundaries. Chapman & Hall, London, pp 393–416

Felsenstein J (1985) Phylogenies and the comparative method. Am Nat 125:1–15

Furley PA, Ratter JA (1988) Soil resources and plant communities of the central Brazilian cerrado and their development. J Biogeogr 15:97–108

Garnier E (1992) Growth analysis on congeneric annual and perennial grass species. J Ecol 80:665–675

Gartner BL, Meinzer FC (2005) Structure–function relationships in sapwood water transport and storage. In: Holbrook NM, Zwieniecki MA (eds) Vascular transport in plants. Elsevier, Boston, pp 307–331

Hacke UG, Sperry JS, Pockman WT, Davis SD, McCulloh KA (2001) Trends in wood density and structure are linked to prevention of xylem implosion by negative pressure. Oecologia 126:457–461

Hoffmann WA, Franco AC (2003) Comparative growth analysis of tropical forest and savanna woody plants using phylogenetically independent contrasts. J Ecol 91:475–484

Hoffmann WA, Orthen B, Franco AC (2004) Constraints to seedling success of savanna and forest trees across the savanna–forest boundary. Oecologia 140:252–260

Hoffmann WA, Franco AC, Moreira MZ, Haridasan M (2005) Specific leaf area explains differences in leaf traits between congeneric savanna and forest trees. Funct Ecol 19:932–940

Hopkins B (1992) Ecological processes at the forest-savanna boundary. In: Furley PA, Procter F, Ratter JA (eds) Nature and dynamics of forest–savanna boundaries. Chapman & Hall, London, pp 21–23

Jackson PC, Meinzer FC, Bustamante M, Goldstein G, Franco A, Caldas L, Igler E, Causin F (1999) Partitioning of soil water among tree species in a Brazilian Cerrado ecosystem. Tree Physiol 19:717–724

Jacobsen AL, Ewers FW, Pratt RB, Paddock WA, Davis SD (2005) Do xylem fiber affect vessel cavitation resistance? Plant Physiol 149:546–556

Meinzer FC (2003) Functional convergence in plant responses to the environment. Oecologia 134:1–11

Meinzer FC, Goldstein G, Franco AC, Bustamante M, Igler E, Jackson P, Caldas L, Rundel PW (1999) Atmospheric and hydraulic limitations on transpiration in Brazilian Cerrado woody species. Funct Ecol 13:273–282

Nardini A (2001) Are sclerophylls and malacophylls hydraulically different? Biol Plant 44:239–245

Pivello VR, Coutinho LM (1992) Transfer of macronutrients to the atmosphere during experimental burnings in an open cerrado (Brazilian savanna). J Trop Ecol 8:487–497

Ratter JA (1992) Transitions between cerrado and forest vegetation in Brazil. In: Furley PA, Procter J, Ratter JA (eds) Nature and dynamics of forest-savanna boundaries. Chapman & Hall, London, pp 417–429

Rosenthal R, Rosnow RL (1985) Contrast analysis: focused comparisons in the analysis of variance. Cambridge University Press, New York

Sack L, Frole K (2006) Leaf structural diversity is related to hydraulic capacity in tropical rain forest trees. Ecology 87:483–491

Sack L, Holbrook NM (2006) Leaf hydraulics. Annu Rev Plant Biol 57:361–381

Sack L, Tyree MT (2005) Leaf hydraulics and its implications in plant structure and function. In: Holbrook NM, Zwieniecki MA (eds) Vascular transport in plants. Elsevier, Amsterdam, pp 93–114

Sack L, Melcher PJ, Zwieniecki MA, Holbrook NM (2002) The hydraulic conductance of the angiosperm leaf lamina: a comparison of three measurement methods. J Exp Bot 53:2177–2184

Sack L, Cowan PD, Jaikumar N, Holbrook NM (2003) The ‘hydrology’ of leaves: co-ordination of structure and function in temperate woody species. Plant Cell Environ 26:1343–1356

Sack L, Tyree MT, Holbrook NM (2005) Leaf hydraulic architecture correlates with regeneration irradiance in tropical rainforest trees. New Phytol 167:403–413

Scholander PF, Hammel HT, Brandstreet ED (1965) Sap pressure in vascular plants. Science 148:339–346

Scholz FG, Bucci SJ, Goldstein G, Meinzer FC, Franco AC, Miralles-Wilhelm F (2007) Biophysical properties and functional significance of stem water storage tissues in neotropical savanna trees. Plant Cell Environ 30:236–248

Sperry JS, Tyree MT (1988) Mechanism of water stress-induced xylem embolism. Plant Physiol 88:581–587

Sternberg LSL, Bucci S, Franco AC, Goldstein G, Hoffmann WA, Moreira MZ, Scholz F (2004) Long range lateral root activity by Neo-tropical savanna trees. Plant Soil 270:169–178

Stratton L, Goldstein G, Meinzer FC (2000) Stem water storage capacity and efficiency of water transport: their functional significance in a Hawaiian dry forest. Plant Cell Environ 23:99–106

Swaine MD, Hawthorne WD, Orgle TK (1992) The effects of fire exclusion on savanna vegetation at Kpong, Ghana. Biotropica 24:166–172

Tyree MT, Ewers FW (1991) The hydraulic architecture of trees and other woody plants. New Phytol 119:345–360

Tyree MT, Hammel HT (1972) The measurement of the turgor pressure and the water relations of plants by the pressure-bomb technique. J Exp Bot 23:267–282

Tyree MT, Sperry JS (1989) Vulnerability of xylem to cavitation and embolism. Annu Rev Plant Mol Biol 40:19–48

Tyree MT, Sobrado MA, Stratton LJ, Becker P (1999) Diversity of hydraulic conductance in leaves of temperate and tropical species: possible causes and consequences. J Trop For Sci 11:47–60

Woodruff DR, McCulloh KA, Warren JM, Meinzer FC, Lachenbruch B (2007) Impacts of tree height on leaf hydraulic architecture and stomatal control in Douglas-fir. Plant Cell Environ 30:559–569