

# Water relations of baobab trees (*Adansonia* spp. L.) during the rainy season: does stem water buffer daily water deficits?

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## ABSTRACT

**Baobab trees are often cited in the literature as water-storing trees, yet few studies have examined this assumption. We assessed the role of stored water in buffering daily water deficits in two species of baobabs (*Adansonia rubrostipa* Jum. and H. Perrier and *Adansonia za* Baill.) in a tropical dry forest in Madagascar. We found no lag in the daily onset of sap flow between the base and the crown of the tree. Some night-time sap flow occurred, but this was more consistent with a pattern of seasonal stem water replenishment than with diurnal usage. Intrinsic capacitance of both leaf and stem tissue (0.07–0.08 and 1.1–1.43 MPa<sup>-1</sup>, respectively) was high, yet the amount of water that could be withdrawn before turgor loss was small because midday leaf and stem water potentials (WPs) were near the turgor-loss points. Stomatal conductance was high in the daytime but then declined rapidly, suggesting an embolism-avoidance strategy. Although the xylem of distal branches was relatively vulnerable to cavitation (P50: 1.1–1.7 MPa), tight stomatal control and minimum WPs near –1.0 MPa maintained native embolism levels at 30–65%. Stem morphology and anatomy restrict water movement between storage tissues and the conductive pathway, making stored-water usage more appropriate to longer-term water deficits than as a buffer against daily water deficits.**

*Key-words:* Bombacaceae; dry forest; sap flow; stem-succulent; stomatal conductance; vulnerability.

## INTRODUCTION

Stem-succulent trees are abundant in seasonally dry tropical environments around the world and are characterized by large, water-filled stems, relatively small crowns and low-density wood (Fenner 1980; Medina 1995; Borchert & Rivera 2001). Because of their unusual morphology, it has long been assumed that they depend upon stored water to

survive (Newton 1974; Owen 1974; Wickens 1983; Schulze *et al.* 1988; Nilsen *et al.* 1990). Because stem-succulent trees are deciduous, there is little demand for water during the dry season, and stored water may therefore be important during the rainy season. Plants growing in tropical climates are often subjected to elevated temperatures, intense solar radiation and large vapour pressure deficits (Eamus 1999). Even in a well-watered tree, the rate of transpiration from the leaves can exceed the transport capacity of the stem, leading to stomatal closure during the day to conserve water and to prevent xylem cavitation (Goldstein *et al.* 1998; Eamus 1999; Brodribb *et al.* 2003). This midday depression in stomatal conductance in response to daily water deficits represents a limitation to photosynthetic productivity in many tropical trees (Eamus & Prior 2001). In stem-succulent trees, such stomatal limitation could be particularly important, because they typically maintain leaves for less than half of the year.

We hypothesized that the water in the stems and branches of stem-succulent trees is used to buffer daily water deficits during the rainy season, and thus is important to their ability to compete with trees having longer growing seasons. Specifically, stem-succulent trees may draw upon stored water during periods of high demand, thereby avoiding extreme negative pressures in the xylem that lead to high levels of cavitation, but still allowing them to keep stomata open for gas exchange (Eamus & Prior 2001). To test this hypothesis, we initiated a study in a tropical deciduous forest in Madagascar, focusing on two species of baobab trees (*Adansonia* spp. L.). Previously, only one study has examined the role of stored water in baobab trees (Fenner 1980), despite their unusual growth habit and the fact that they are widely cited in the literature as trees that use stored water to survive drought. In a related study on three species of baobab trees, we report that baobabs shed their leaves at the end of the rainy season when soil water is still available, and use very little stored water during the dry season (Chapotin, Razanameharizaka & Holbrook 2006). Furthermore, although stored water is used to flush new leaves before the onset of the rainy season, the stomata do not open until after the rainy season has begun. Because stored water does not allow baobab trees to extend their

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growing season by delaying leaf shedding or by supporting stomatal opening in newly expanded leaves, we wanted to learn whether stored water was important in maximizing gas exchange during the rainy season. The use of stored water to buffer diurnal water deficits could result in increased photosynthetic productivity and/or decreased investment in root production (Tyree *et al.* 1991).

All trees have water in their stems that may be drawn upon when water uptake from the soil becomes limiting. The timescale during which any given tree will use stored water and the degree to which it is relied upon, however, are likely to be determined by a combination of factors including soil water availability, leaf-specific conductivity, xylem vulnerability to cavitation and stem and leaf capacitances. Tyree *et al.* (1991), working in a seasonally dry forest in Panama, determined that the withdrawal of significant amounts of stem water from the evergreen tree *Schefflera morototoni* occurred only during the dry season when soil water potentials (WPs) are low. During the rainy season, when water-use rates are significantly higher, *S. morototoni* is able to supply water to the leaves at high rates, because of its high leaf-specific conductivity, without withdrawing stem water or reaching cavitation thresholds in its highly vulnerable wood. Some trees, however, do draw upon water in the stem and branches to buffer daily water deficits even when soil water is abundant (Holbrook 1995). For example, Goldstein *et al.* (1998) found that stored water contributes from 9 to 15% of total daily water use in five canopy tree species in a seasonally dry tropical forest, and temperate evergreen trees can draw upon stored water to meet up to 25% of their daily transpirational requirements (Waring & Running 1978; Waring, Whitehead & Jarvis 1979; Phillips *et al.* 2003).

A few studies have attempted to understand the role of stored water in stem-succulent trees, but it has not been determined whether they use stored water only during drought or on a daily basis. Stem-succulent trees usually have very large xylem vessels, which should result in high xylem conductivity; and high vulnerability to embolism is also to be expected as a result of their low-density wood (Hacke *et al.* 2001). Therefore, although the large volume of water present in the stems and branches suggests a dependence on stored water, the anatomical features of stem-succulent trees might lead to a pattern similar to that found in *Schefflera*. Nilsen *et al.* (1990) suggested that the high capacitance of stem-succulent trees growing in the Baja California Desert would buffer daily and seasonal environmental variations in water availability, although they made no direct measures of water uptake from storage tissues. They also concluded that water use must be low to prevent leaf water deficits from occurring, and that low rates of stomatal conductance are limiting. Fenner (1980) measured girth changes in baobab trees (*Adansonia digitata*) on both seasonal and diurnal scales and hypothesized that these changes represented the use and replenishment of stored water. He additionally reported extremely low water loss rates in cut baobab shoots, and attributed this to very effective stomatal control.

The published studies on stem-succulent trees do not directly assess the dynamics of water movement into or out of the storage tissue, nor do they determine how the water relations of the parenchymatous wood affect the ability of the tree to draw upon stored water. In this study, we describe patterns of water usage in baobab trees during the rainy season to determine whether these stem-succulent trees use stored water to buffer short-term water deficits. We compare rates of water uptake from internal water stores and from the soil, and we assess the extent that the availability of stored water affects the stomatal behavior of the leaves. We also examine the capacitance, vulnerability to cavitation and turgor-maintenance properties of the wood to learn the extent to which water stored in the tree is accessible to the water transport system, and the time scale of its use.

## MATERIALS AND METHODS

### Field site

This study was conducted at the Kirindy Forestry Reserve, one of the largest remaining tracts of tropical dry forest in Western Madagascar (long 44°49'E, lat 20°27'S) (Rakotonirina 1996). The mean annual rainfall is 770 mm, but can be highly variable and ranges from 300 to 1400 mm year<sup>-1</sup> (Ganzhorn *et al.* 1990). Most of the rain falls from December to March, and rainfall is generally non-existent from April to October. Soils are sandy with low water retention, except near seasonal watercourses where increased clay content provides a higher water-retaining capacity (Sorg & Rohner 1996).

### Study species

The eight species in the genus *Adansonia*, commonly referred to as baobabs, have a disjunct distribution across Africa, Madagascar and Australia (Armstrong 1983). Two species of baobab were included in this study, *Adansonia za* Baill. and *Adansonia rubrostipa* Jum. and H. Perrier. Both species are found over a large range of habitat types in Western and Southern Madagascar, and exhibit substantial variation in overall morphology (Baum 1996). In the Kirindy Forest, these species are emergent above a canopy composed mainly of dry season-deciduous trees. Study trees ranged from 1 to 2.3 m in diameter, and from 15 to 25 m in height. The crown of the tree was accessed for installation of sap flow probes, stomatal conductance measurements, and leaf and branch sampling by using the single rope technique (Laman 1995). Once the main branches of the tree were reached, access to distal branches and leaves was facilitated by the compact nature of the crown and by the large size of the branches relative to their length.

### Sap flow measurements

Rates of sap flow were measured using the thermal dissipation technique (Granier 1985), with probes similar in design

to those described by James *et al.* (2002). Underlying temperature gradients in the stem, which were revealed by using unheated reference probes, led to large errors when the traditional Granier calibration was applied to continuously heated probes (Lu, Urban & Zhao 2004). The probes were therefore heated discontinuously, following Do & Rocheteau (2002a, b), and the rates of sap flow were calculated by taking the difference between heated and unheated measurements and then by applying calibration formulas provided by Do & Rocheteau (2002b). Probes were inserted to a depth of 1 cm beneath the bark, because initial tests with probes inserted at greater depths (3, 5, 10, 15 and 20 cm) indicated that sap flow could not be detected beyond 1–2 cm. Probes were placed at the base of the trunk (0.5–1.0 m above the ground) to measure sap flow from the roots into the stem, at the top of the trunk (within 1 m of the point of main branch divergence) to measure sap flow from the main stem to the branches, and on distal branches (8–13 cm in diameter) to measure sap flow from the branches to the leaves. Two to four probes were installed at each location, and calculated sap flow rates from probes at the same location were averaged. Probes were installed in two trees of each species, and each tree was measured for 5–9 d.

### Stomatal conductance measurements

Stomatal conductance of fully illuminated canopy leaves was measured with a Li-Cor 1600 steady-state porometer (Li-Cor, Lincoln, NE, USA). Measurements were taken every 30–60 min from dawn to dusk to quantify the diurnal pattern of stomatal conductance for 4 d during the 2002 rainy season (January and February) in three trees of *A. rubrostipa* and one tree of *A. za*. Ten mature, sunlit leaves per tree were measured at each point in time, and the average value was calculated. Two of these diurnal time courses were paired with concurrent measurements of leaf WP. Measured leaves were distributed, to the extent possible given the inherent difficulties with access, throughout the outer crown of the tree. Air saturation deficit (ASD) at each measurement time was calculated from the relative humidity and temperature of the air as measured by the porometer, according to the formula

$$\text{ASD} = e_s - e_a, \quad (1)$$

where  $e_s$  is the saturated water vapour pressure, and  $e_a$  is the ambient water vapour pressure of the air. Leaf-temperature measurements were not available for all sampling times, but because leaf and air temperatures, measured by the porometer, differed only by a very small amount, we therefore report ASD rather than leaf-to-air vapour pressure deficit.

### Leaf WP

Leaf WPs were measured with a Plant Moisture Stress Instrument Company pressure chamber (PMS Instrument Company, Albany, OR, USA) the compressed air for which

was generated with an AxSOR FX-2000 high-pressure hand pump (AxSOR, Hova, Sweden). Several leaves per tree ( $n = 3$  trees each for *A. rubrostipa* and *A. za*) were collected at dawn and between 1300 and 1500 h to obtain minimum and maximum WP values on several occasions during the 2002 and 2003 rainy seasons (January to March). To obtain a measure of branch WP, leaves were covered and allowed to equilibrate before the measurement was taken. Measurements of WP were also made concurrently with diurnal courses of leaf stomatal conductance. Five leaves per tree in two trees of *A. rubrostipa* were excised and measured in the pressure chamber every 60–75 min from dawn to dusk.

### Stem WP

Wood cores (0–3 cm beneath the bark; 12 mm in diameter) were extracted from stems of *A. rubrostipa* and *A. za*, and their WP were measured using the Shardakov method (Slavik 1974). One core was extracted from each of 5 trees per species at dawn, and between 1300 and 1400 h during the 2002 rainy season. A razor blade was used to shave off the outer surface of each section and then to further divide it into five equal pieces. The pieces were placed in a series of sugar solutions with osmotic potentials ranging from 0.05 to 0.8 MPa for 30 min. The WP of the wood was estimated to be equal to the osmotic potential of the solution with which the wood came into equilibrium, as determined by comparing the density of a series of safranin-dyed solutions having the same osmotic potentials.

### Leaf pressure–volume curves

Intrinsic capacitances and turgor-loss points were obtained by subjecting previously rehydrated leaves to a series of paired measurements of WP and relative water content (RWC) as they were allowed to air-dry. After the measurements were completed, the leaves were dried at 65 °C for 24 h to determine dry weights (DWs). Pressure–volume curves were generated for 3 leaves per tree, for each of four trees, and intrinsic capacitances and turgor-loss points were calculated from the pressure–volume curves according to the method described by Koide *et al.* (1989).

### Stem pressure–volume curves

Six wood cores (0–3 cm beneath the bark; 12 mm in diameter) were extracted from each of 5 trees per species. The surface of each core was shaved clean with a razor blade and divided into two sections which were used for paired measurements of RWC and WP. One pair of wood sections was measured fresh, one was allowed to rehydrate in a moist paper towel for several hours, and the other four pairs were air dried to varying degrees. WP was measured on one section from each pair (with the Shardakov method, as described above) and RWC on the other. DWs for RWC calculations were obtained by drying wood sections at 75 °C for 24 h. Intrinsic capacitance and turgor-loss points

for the stems were calculated as for the leaves, except that the limited number of samples required that all data for a species be combined to generate a single pressure–volume curve for each species.

### Per cent loss conductivity (PLC)

PLC was measured on branch segments from distal branches three times during the year to determine levels of native embolism. Branch segments were perfused with a 10 mmol solution of KCl at pressures lower than that calculated to dislodge embolisms in the largest vessels (2.4 kPa), and the water flow rate through the stem ( $F_i$ ) was measured using a steady-state flowmeter (Feild & Holbrook 2000; Feild, Brodribb & Holbrook 2002). Segments were then flushed with the KCl solution for 1 min at high pressure to expel any embolisms, after which the flow rate was measured again ( $F_a$ ). The high-pressure flush was generated by fitting a caulking gun to a KCl solution-filled syringe and by connecting this to the branch segment. Branch segments were 10–27 cm in length and 0.5–1 cm in diameter (without bark). Initial measurements using air perfusion indicated that branch segments of this length contained very few vessel endings, such that embolisms should be easily removed once the pressure threshold set by the capillary equation for the smallest vessels has been exceeded. PLC was calculated as  $(F_a - F_i)/F_a$ . Because of the open vessels in these short segments, these measurements were not used to calculate specific conductivity. Three to five branch segments from each of 3 trees per species were measured. Branches were collected in the early afternoon during the dry season (2003), the leaf-flushing period (2001, additional measurements were also made at dawn during this time) and the rainy season (2002).

### Vulnerability curves

Vulnerability curves were generated on distal branch segments (0.75–1.0 cm in diameter, bark excluded, and 10–15 cm in length) of *A. rubrostipa* ( $n = 7$ ) and *A. za* ( $n = 4$ ) collected at dawn during the 2002 rainy season. Initial conductivity was measured on each branch segment. Segments were then subjected to increasing levels of compressed air to induce radial air seeding (Cochard, Cruiziat & Tyree 1992). Compressed air was generated with an Axsor FX-2000 high-pressure hand pump (Axsor, Hova, Sweden). Conductivity was measured after each pressurization event, and PLC was calculated as  $(F_a - F_i)/F_a$ . PLC was plotted versus air-seeding pressure to generate the vulnerability curves, which were fit to a 5-parameter sigmoid curve.

### Stem water content

Cores 12 mm in diameter were extracted from the stem between 0.5 and 1.5 m from the ground using an increment borer. Cores were divided into sections 5 cm in length from 0 to 30 cm beneath the bark and were immediately sealed

into pre-weighed plastic bags. Fresh weight (FW) and volume were determined before drying the cores at 75 °C for 24 h to determine DW. Water content was calculated on a per cent of fresh volume basis (Domec & Gartner 2002). Cores were extracted from five trees of each species five times between October 2001 and March 2002. Diameter at breast height was also measured to gauge stem water loss and uptake through stem shrinkage and swelling.

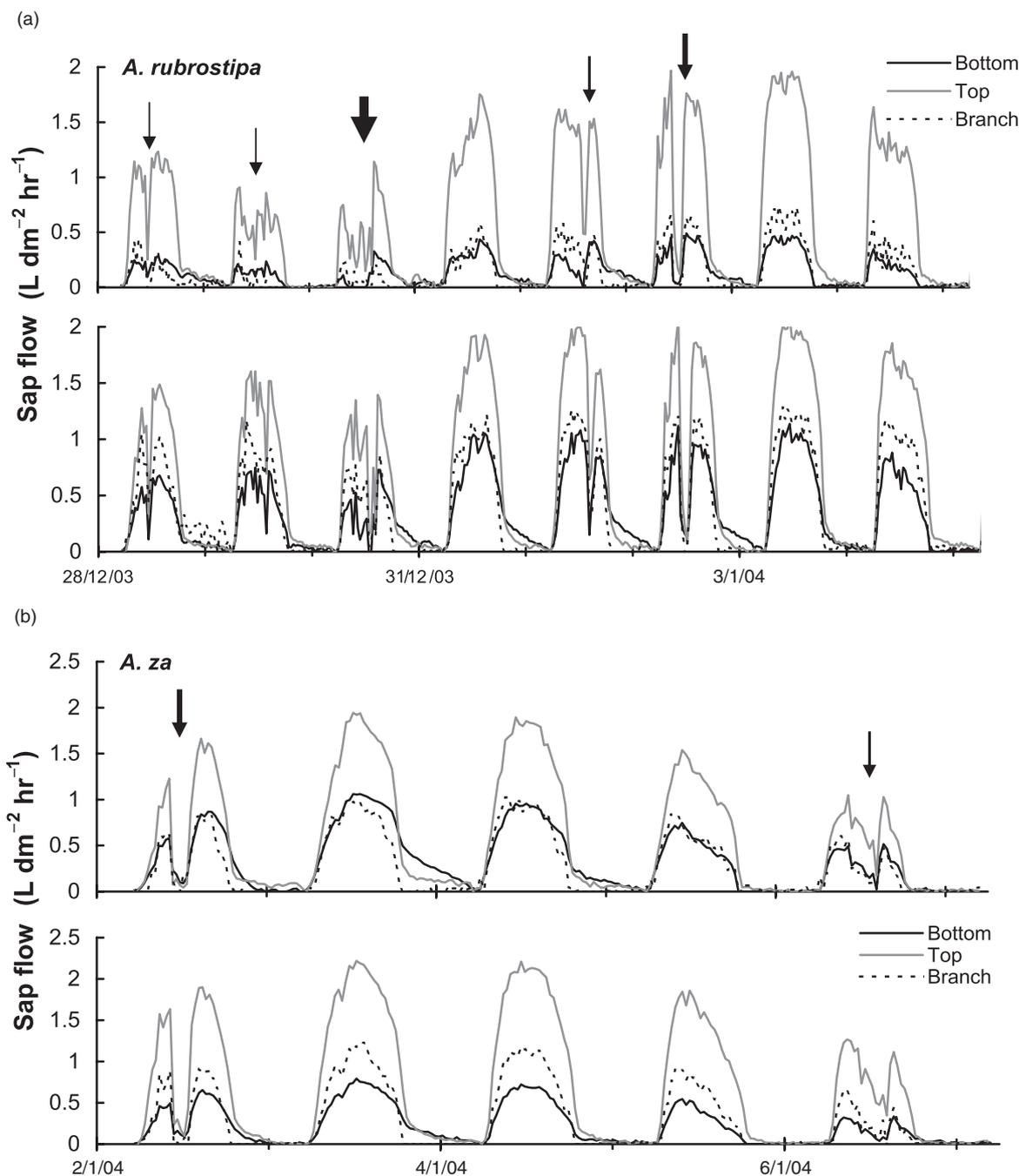
## RESULTS

### Sap flow measurements

Sap flow methods can be used to assess the use of stored water from tree stems in two ways: (1) Rates of sap flow can be used to calculate the total volume of water flowing through each point in the tree, or (2) the temporal pattern of sap flow at different points in the tree can be compared for evidence of time lags. Although the first method allows for the quantification of the daily amount of water withdrawn from storage, it requires highly accurate determination of sapwood area at each point in the tree and is sensitive to differences in sap flow rates around the circumference of the tree. The second method does not allow for a volumetric quantification of daily stored water usage, but avoids the errors that often result when scaling up from point measurements in large trees. This approach is based on the fact that withdrawing water from storage leads to a phase shift between the sap flow traces at the top and at the bottom of the tree. Conversely, when the traces are largely in phase with each other, then the amount of water withdrawn will be very small. Because of the difficulties in completing an accurate water budget in these large trees, we used the second method in this study.

Daily onset and cessation of sap flow were consistent between all points in the tree, and no phase shift between the traces at the bottom and top of the tree was detectable, at least within the 30 min resolution period of our data (Fig. 1). When the data were scaled so that daily sap flow maxima at different points in the tree were roughly equal (not shown), the phase shift was completely absent on most days; and in some instances, the shift was in the opposite direction (onset of flow at the bottom of the tree before onset at the top). Although the resolution of our data does not preclude a very small phase shift, this would correspond to only a very small amount of water relative to total water use, and to an even smaller fraction of the total water available in the tree.

The rates of sap flow measured at the base of the stem were generally lower than at the top of the stem. This suggests that the region of conductive sapwood area at the base of the stem is larger than at the top, or that sap flow rates at the base were underestimated. Although baobab stems do exhibit some taper, it was not sufficient to cause these differences, and the conductive sapwood depth is likely to differ at these two positions. Vessel diameter and density at the two positions were similar (unpublished results); therefore, although the calibration formula has not

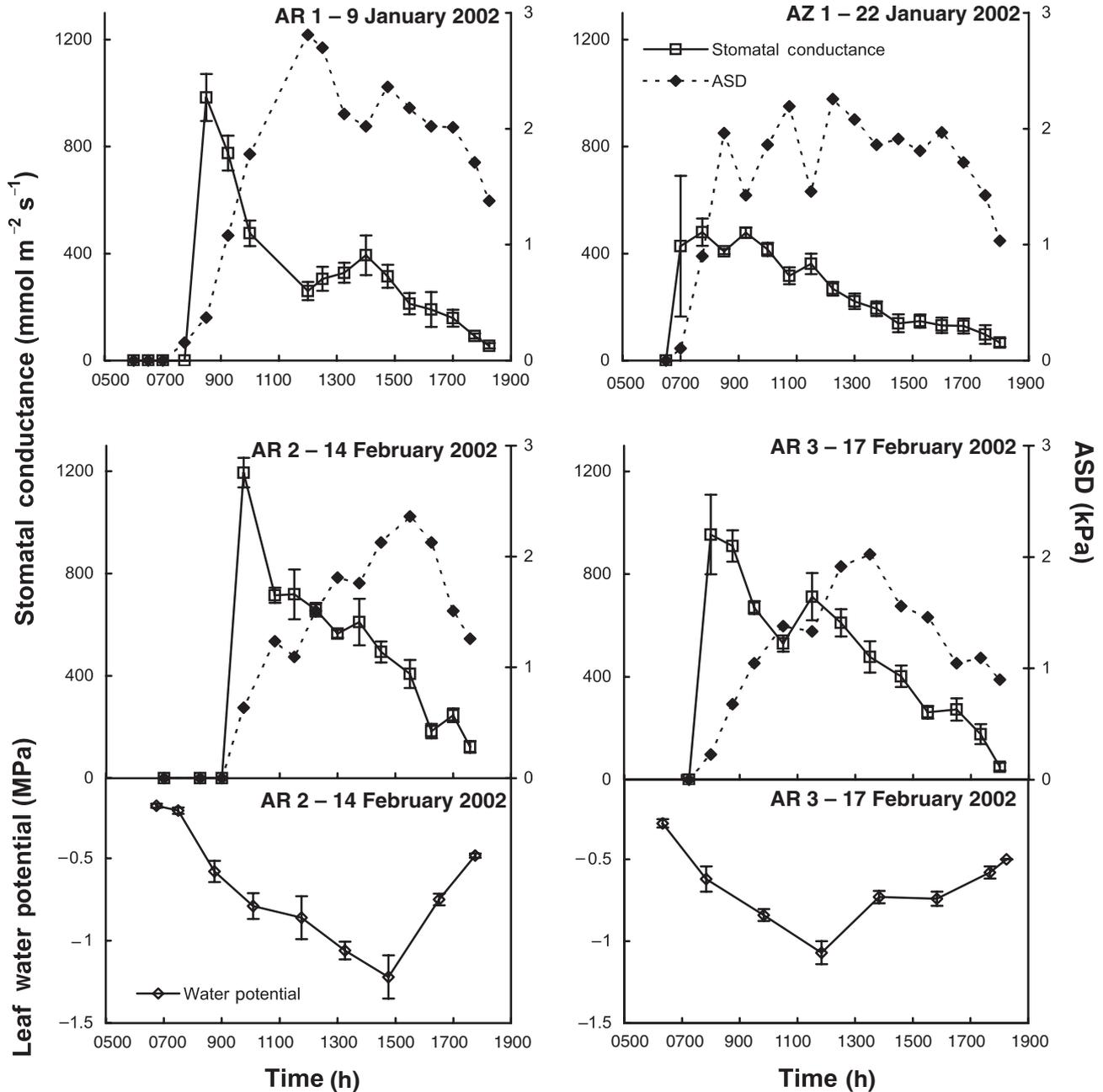


**Figure 1.** Sap flow rates during the early rainy season for two trees each of *Adansonia rubrostipa* (a) and *Adansonia za* (b). Each line is the average of values from two to four probes placed in equivalent locations on the same tree. Rainfall events are marked by arrows, the size of which represents the magnitude of the rainfall event.

been explicitly tested in stem-succulent trees, it is unlikely to differ significantly between the two positions. Furthermore, sap flow rates at the base of the stem do not appear to reach zero on certain nights, indicating that maximum night-time temperatures were not reached, and sap flow rates for that day were underestimated.

Sap flow rates at all points in the measured trees increased significantly after the onset of the rainy season.

The sap flow rates were highest after a rainfall event and continued into the night at the bottom of the tree and to a lesser extent at the top of the tree, although sap flow rates in the branches consistently dropped to zero at dusk (Fig. 1). Night-time sap flow occurred only during the first few nights after a large rainfall event and gradually decreased as the upper soil levels dried out. Sap flow rates were high throughout the day, in contrast to the pattern



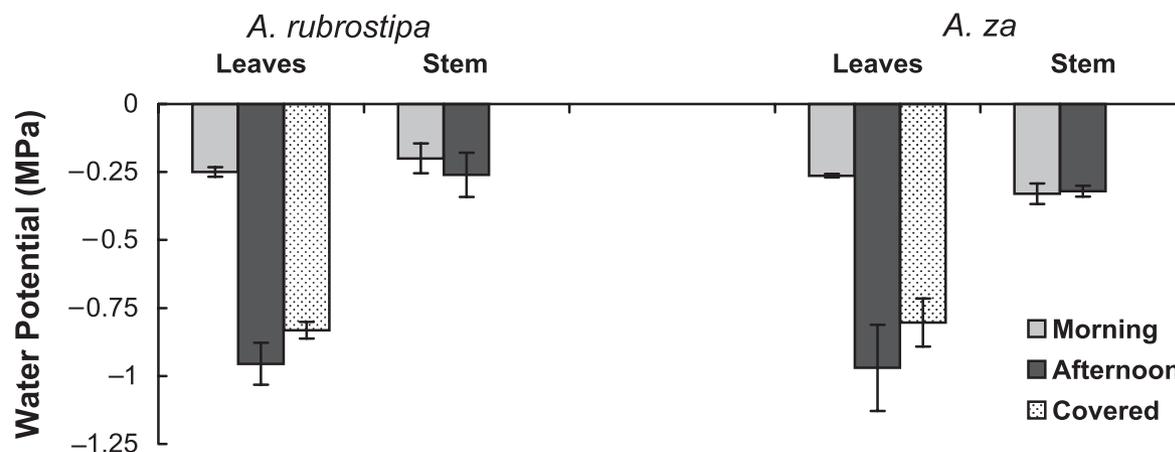
**Figure 2.** Daily time courses of stomatal conductance ( $\text{mmol m}^{-2} \text{s}^{-1}$ ) and air saturation deficit (ASD, kPa) of three trees of *Adansonia rubrostipa* (AR1, AR2 and AR3) and one tree of *Adansonia za* (AZ1) (upper four graphs). Daily time courses of leaf water potential (WP) (MPa, bottom two graphs) for two trees of *A. rubrostipa* (AR2 and AR3), made on the same days as the stomatal conductance and ASD time courses. Error bars are  $\pm 1$  SE; 10 leaves per tree for stomatal conductance and 5 leaves per tree for WP.

observed at the end of the dry season after scattered rainfall events, when sap flow occurred only in the early morning hours when ASD was low (Chapotin *et al.* 2006). The data presented in this study were collected during the first 2 weeks of the 2003–2004 rainy season when cumulative rainfall totals were still quite low. Preliminary sap flow data collected during the latter part of the 2002 rainy season (mid-February to March) indicated no night-time sap flow. Although methodological problems prevented these data

from being adequately analysed, the overall daytime versus night-time pattern in sap flow is still evident.

#### Stomatal conductance measurements and leaf and stem WP

Stomatal conductance during the rainy season was highest ( $800\text{--}1100 \text{ mmol m}^{-2} \text{ s}^{-1}$ ) in the morning and quickly decreased with rising ASD (Fig. 2). The rate of decline



**Figure 3.** Water potential (WP) (MPa) in stems and leaves of *Adansonia rubrostipa* and *Adansonia za* at dawn and during the early afternoon. Additional measurements of covered leaves were made during the afternoon to estimate branch WP. All measurements were made during the rainy season. Values are the mean  $\pm$  1 SE;  $n = 3$  trees, 5 leaves per tree for leaf WP;  $n = 5$  trees, 1 core per tree for stem WP.

became slower by late morning, but stomatal conductance continued to decrease over the course of the day. Leaf WP declined steadily throughout the morning, reached a minimum value ranging from  $-1.1$  to  $-1.2$  MPa between 1200 and 1500 h, and then recovered in the afternoon, although only to about  $-0.5$  MPa by sundown (Fig. 2). Average minimum (early afternoon) leaf WPs remained above  $-1.0$  MPa, and values from covered leaves were greater by about  $0.15$  MPa, but not significantly different from uncovered leaves (Student's  $t$ -test,  $P > 0.05$ ) (Fig. 3). Pre-dawn values were near  $-0.25$  MPa. Stem WP values ranged from  $-0.2$  to  $-0.3$  MPa, and there was no significant difference between pre-dawn and afternoon values (Fig. 3), which was consistent with a pattern of no stored-water usage.

#### Leaf and stem pressure–volume curves

Stem tissue had a very high intrinsic capacitance, ranging from  $1.10$  to  $1.43$  MPa $^{-1}$  (Table 1). These values were similar to values obtained from *Sabal palmetto* but were considerably greater than values reported for *Ferocactus acanthodes*, *Espeletia* spp. and *Pinus sylvestris* (Holbrook & Sinclair 1992). Leaf capacitances ( $0.07$ – $0.08$  MPa $^{-1}$ ) were

much lower than stem capacitances but were within the range of leaf values reported for other tropical dry forest tree species (Chapotin *et al.* 2003). Turgor loss was estimated to occur at relatively high WPs and water contents (Table 1). The turgor-loss point in stems (from  $-0.22$  to  $-0.32$  MPa) and leaves (from  $-1.12$  to  $-1.26$  MPa) were very close to the observed minimum field WPs.

#### PLC

Native PLC values in *A. za* ranged from 36 to 40% and did not vary significantly throughout the year. *A. rubrostipa* PLC values ranged from 42 to 65%, with dry season values significantly greater than rainy season values (Student's  $t$ -test). There was no difference between early morning and afternoon values for measurements made during the leaf-flushing period (Table 2).

#### Vulnerability curves

On the basis of a single vulnerability curve for each species, calculated by combining data from 5 trees per species, *A. rubrostipa* was more vulnerable to cavitation than *A. za*.

	Leaves		Wood	
	<i>A. rubrostipa</i>	<i>A. za</i>	<i>A. rubrostipa</i>	<i>A. za</i>
Capacitance (MPa $^{-1}$ )	$0.08 \pm 0.004$	$0.07 \pm 0.003$	1.43	1.10
Turgor-loss point (MPa)	$-1.12 \pm 0.074$	$-1.26 \pm 0.076$	$-0.22$	$-0.32$
Turgor-loss point (RWC)	$0.92 \pm 0.006$	$0.91 \pm 0.008$	0.94	0.85
Osmotic potential	$-0.86 \pm 0.036$	$-1.05 \pm 0.068$	$-0.20$	$-0.25$

**Table 1.** Tissue–water relations parameters calculated from pressure–volume curves for *Adansonia rubrostipa* and *Adansonia za*

Leaf values are the mean  $\pm$  1 SE. No SE is given for the wood values because the curves for individual trees did not have enough data points to calculate the parameters; therefore, data from all five trees for each species were combined into one pressure–volume curve. RWC, relative water content.

PLC	Dry season Afternoon	Leaf-flushing period		Rainy season Afternoon
		Morning	Afternoon	
<i>A. rubrostipa</i>	65 ± 2.8	32 ± 5.2	51 ± 8.5	42 ± 7.2
<i>A. za</i>	40 ± 7.5	39 ± 3.6	40 ± 6.5	37 ± 4.2

Values are the mean ± 1 SE.

Dry season values for *A. rubrostipa* were significantly greater than rainy season values ( $P = 0.04$ , Student's *t*-test).

Measurements were made during the early afternoon, except during the leaf-flushing period when additional early morning measurements were made.

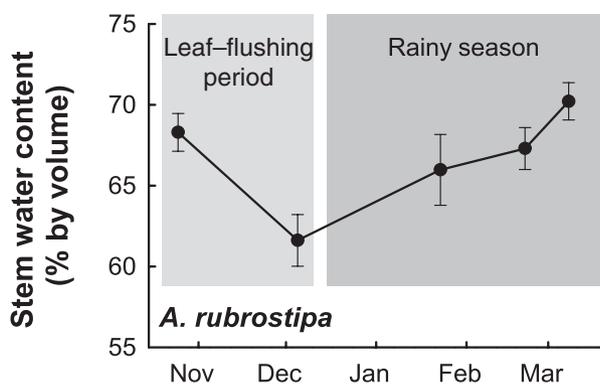
The air-seeding pressure leading to 50% loss of conductivity was 1.1 and 1.7 MPa in *A. rubrostipa* and in *A. za*, respectively. Both species had a loss of conductivity greater than 95% at a seeding pressure of 5.0 MPa.

### Stem water content

Volumetric stem water content decreased in *A. rubrostipa* before the onset of the rainy season, during the leaf-flushing period (see also Chapotin *et al.* 2006). Water content then gradually increased over the course of the rainy season, returning to pre-leaf-flushing levels by early March (Fig. 4). In *A. za*, volumetric water content did not change significantly, but stem diameter shrank an average of 2–3 cm during the leaf-flushing period in two different years and then recovered during the rainy season. A previous study showed that similar amounts of water are withdrawn in the two species, although volumetric water content remains constant in *A. za* and stems do not shrink considerably in *A. rubrostipa* (Chapotin *et al.* 2006).

### DISCUSSION

Baobab trees have high wood water content, a large stem volume and are often leafless for more than half the year. Because of the vast quantity of water present in the stems



**Figure 4.** Volumetric stem water content (0–30 cm beneath the bark) in *Adansonia rubrostipa* during the leaf-flushing period and the rainy season. Values are the mean ± 1 SE;  $n = 5$  trees.

**Table 2.** Per cent loss conductivity (PLC) for small branch segments of *Adansonia rubrostipa* and *Adansonia za*

and in the branches of baobab trees, and because of the high temperatures and low relative humidity that can be reached in the upper canopy, we expected baobabs to draw upon stored water on a diurnal basis. We hypothesized that baobab trees would use stored water to buffer daily water deficits by drawing water from the stem during the day and by replenishing the water at night when evaporative demand is lower. This pattern has been observed in trees from a wide range of environments (Holbrook 1995; Goldstein *et al.* 1998; Phillips *et al.* 2003). Our data revealed no evidence, however, that baobabs use stored water to maximize daily stomatal conductance.

### Sap flow data suggest seasonal rather than diurnal use of stored water

In evaluating this hypothesis, our primary approach was to use sap flow probes placed in the tree at several different positions. Stored-water usage should lead to time lags in the onset and termination of daily sap flow between the base of the tree and the branches in the crown (Holbrook 1995; Wullschlegel, Meinzer & Vertessy 1998). If stem water is being withdrawn, daily sap flow onset in the branches, as measured by the probes nearest the leaves, should precede sap flow onset at the base of the stem. Sap flow through the branches should then cease at the end of the day when stomata are shut, while sap flow in the main stem continues into the night as the water depleted during the day is replenished by uptake from the soil. This was not the pattern we observed. Instead, the onset of sap flow in the morning during the rainy season occurred simultaneously at the base and at the top of the stem and in the branches in both species of baobab (Fig. 1). Sap flow at all three positions quickly reached daily maximum levels and remained high throughout the day. At the end of the day, sap flow also declined abruptly at all three positions, coinciding with stomatal closure in the leaves. Sap flow rates in the branches reached 0, but after the initially steep decline, sap flow rates in the main stem diminished gradually throughout the night.

We believe that the night-time sap flow observed at the base of the stem, and to a lesser extent at the top of the stem, reflects a seasonal replenishment of water withdrawn from storage rather than a daily replenishment of water

used for buffering daily water deficits. Before the onset of the rainy season, the stem water content decreased by 10–12% as a result of stored-water usage to support leaf flushing (Chapotin *et al.* 2006). The sap flow data presented in this study correspond to measurements made during the first 2 weeks of the 2003–2004 rainy season. Cumulative rainfall by the end of this study period was only 180 mm (yearly average of 770 mm), and most of the rainfall events were small in magnitude and did not always penetrate the litter layer. The return to pre-flushing water-content levels in the stem, which occurs gradually over a period of 2–3 months (Fig. 4), requires water uptake above that necessary to meet the daily transpirational demand. We propose that the water required to return stem water content to the level measured before leaf flushing is met, in part, through rainy season night-time sap flow.

Sap flow rates in this study appear fairly low, especially, because given the large leaf area in the crown, a substantial volume of water would need to move through the tree to meet daily transpirational demand. Assuming a sapwood depth of 1 cm, conductive sapwood area for the trees in this study ranged from 3.3 to 4.3 dm<sup>2</sup> at the base and from 2.2 to 2.6 dm<sup>2</sup> at the top of the stem. Using these estimates when scaling up to the whole tree level yields very low whole-tree water-use values, yet we measured almost no flow deeper into the sapwood. We believe that sapwood depth is actually restricted to a much narrower band and that sap flow rates were underestimated by using 1 cm probes. Large underestimations can occur when a portion of the probe is situated in non-conductive tissue or when there are gradients in flow across the length of the probe (Clearwater *et al.* 1999; Lu *et al.* 2004). Studies on ring-porous trees, such as various *Quercus* spp., have indicated that the conductive region of sapwood can be extremely narrow, and furthermore, that flow rates are highly asymmetric across this region, such that there exists a large peak in flow within a few millimeters of the cambium (Cermák *et al.* 1992; Granier *et al.* 1994; Phillips, Oren & Zimmermann 1996). Apparently, in contrast to many tropical trees, but similar to many ring-porous trees in temperate regions, baobabs may rely primarily on the current year of xylem growth for water transport. Tests using saffranin dye pushed through distal branches from the baobab trees in this study support this conclusion, because the dye was always restricted to the outermost vessels in the xylem just beneath the cambium.

### Stomatal control limits water use

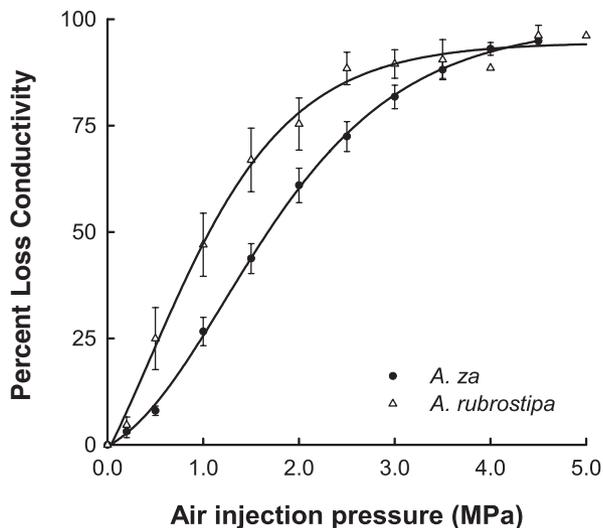
Stomatal conductance reached very high values, particularly in the morning when the ASD was low. Although values were higher than usually reported for tropical trees, they were still within range of values reported for some seasonally dry tropical forests (Meinzer *et al.* 1997; Andrade *et al.* 1998; Stratton, Goldstein & Meinzer 2000; Brodribb *et al.* 2003). These early morning rates of stomatal conductance were not sustained; stomatal conductance declined rapidly in the morning and continued to fall

throughout the rest of day. Apparently, stored water was not accessed when water transport rates from the soil to the leaves became limiting, but rather, stomata closed to prevent excessive water loss. In conjunction with strict stomatal control, afternoon leaf WPs rarely dropped below –1.2 MPa (Figs 2 & 3). Some authors have suggested that plants may fall into two groups: those that regulate stomatal opening to avoid cavitation and those that maintain high rates of stomatal conductance at the expense of xylem conduit safety (Nardini & Salleo 2000). Baobab trees are therefore aligned with the former group, which generally maintains high WPs at the expense of high photosynthetic rates.

### Stem anatomy and morphology limit access to stored water

Night-time sap flow rates were generally low, suggesting that water movement into the storage tissues occurs slowly. We found no evidence for the presence of any radially oriented tracheids or other conducting cells, and vessels beyond the conductive sapwood region were frequently occluded by tyloses. Water transport from the conductive xylem just beneath the bark to the storage tissues deep in the stem must therefore be through a high-resistance pathway consisting mostly of parenchyma tissue, which in some cases forms wide bands that completely isolate the bands of wood with vessels (Chapotin 2005). Night-time flow rates were considerably lower than those during the day, which reflects both the larger resistance of this pathway and the smaller driving gradient between the storage tissues and the soil. Furthermore, rates of night-time water uptake were greatest when soil water was abundant (i.e. after a large rainfall event), and night-time sap flow tended to decline with time after a rainfall event (Fig. 1). The opposite pattern would occur if night-time sap flow were linked to daily stored water usage, because demand for stored water would be greatest when access to soil water was most limited. We expect that night-time sap flow should gradually diminish and eventually cease, because water content in the stem becomes fully replenished over the course of the rainy season. Preliminary measurements from the latter part of the 2002 rainy season (mid-February to March), which indicated no night-time sap flow, were consistent with this expectation.

Baobab xylem vessels are few in number and large in diameter (Chapotin 2005). Loss of function in a relatively small number of vessels could therefore affect the water-transport pathway disproportionately more than in other trees. Results from the vulnerability curves generated through radial air seeding indicate that vessels are quite vulnerable to cavitation. The pressure at which 50% loss of conductivity occurred, P<sub>50</sub>, was 1.1 and 1.7 MPa in *A. robustipa* and in *A. za*, respectively (Fig. 5). These values were obtained from small branches, where mean vessel diameter is one-fifth of the mean vessel diameter in the main stem and the wood is several times denser (Chapotin 2005). Several studies have shown that within a given tree,



**Figure 5.** Vulnerability curves for small branch segments of *Adansonia rubrostipa* ( $n = 7$ ) and *Adansonia za* ( $n = 4$ ). Values are the mean  $\pm 1$  SE of per cent loss conductivity (PLC) at each air injection pressure. The curves had the function,  $f = y_0 + a/[1 + \exp[-(x - x_0)/b]]^c$ , with the constraint that  $c > 0$ .

vessels in the main stem are more vulnerable to cavitation than vessels in the small branches, and that this pattern scales with vessel diameter (Sperry & Saliendra 1994; Dunham *et al.* 2004; Sangsing *et al.* 2004) (but see Choat *et al.* 2005b). The significantly lower wood density in the main stem than in the branches should also lead to greater xylem vulnerability (Hacke *et al.* 2001; Choat *et al.* 2005a). We therefore expect vessels in baobab stems to be even more vulnerable than in branches as reported here. The water-conducting pathway of the baobab tree appears optimized for rapid transport of large volumes of water when soil water is readily available, while tight stomatal control in the leaves reflects the need to keep the xylem vessels safe from cavitation. When water limitations develop, the stomata quickly begin to close, preventing WPs in the xylem from dropping so low that excessive cavitation and consequent vessel blockage occur. There were few differences in PLC in xylem vessels from the rainy to the dry season or from early morning to afternoon, suggesting that baobab trees may not be able to easily refill embolized vessels, but instead might rely on an embolism-avoidance strategy.

With this proposed embolism-avoidance strategy, the reason why baobab trees do not use stored water to alleviate daily water deficits elicits interest. Despite the large volumes of water in the stem, this water may not be immediately accessible. Just as replenishment of stored water in the stem depleted before the onset of the rainy season is a slow process, accessing this water should also take place on a longer timescale, because water held in storage tissues is separated from the transpiration stream by a high-resistance pathway. As a source of water, stem reserves may be more suited to buffering longer-term water deficits where

demand at any given time is small. Actively transpiring trees during the rainy season require large quantities of water which stem reserves are unlikely to meet; buffering of daily water deficits may therefore not be feasible. Seemingly, baobab trees have a pattern of water use more similar to the *Schefflera* trees in the Tyree *et al.* (1991) study than to trees that use stored water on a daily basis (e.g. Goldstein *et al.* 1998). Unlike *Schefflera*, however, baobab trees are deciduous and do not maintain leaves on the tree during the prolonged dry season. There may be no regularly occurring periods during which stem water reserves are drawn upon to survive drought, except during the leaf-flushing period when stomata remain completely shut.

### Stem water withdrawal could lead to turgor loss in parenchyma tissue

In addition to the limitations posed by the physical distance between the storage site and the conducting pathway, the tissue–water relations of the stem wood are such that withdrawing large amounts of water from the wood could be detrimental. Results from the pressure–volume curves generated for sections of stem wood indicate that although capacitance is high, decreases in stem water content could quickly result in turgor loss. Afternoon WP values of the stem wood during the rainy season were  $-0.26$  and  $-0.32$  MPa for *A. rubrostipa* and for *A. za*, respectively. Similar measurements made during the morning and during the leaf-flushing period (data not shown) indicate that the WP of the woody tissue is expected to lose turgor ( $-0.22$  and  $-0.32$  MPa for *A. rubrostipa* and *A. za*, respectively) were at WPs very near the actual measured WPs. Thus, baobab stems are operating near their turgor-loss point, and the withdrawal of significant amounts of water from the stem wood would result in turgor loss in the parenchyma tissue.

The role of the parenchyma cells in woody stems, when present in such large quantities, is likely to be multifold. In addition to water storage, parenchyma tissue may be important for carbohydrate storage or structural support, and the potential impacts of turgor loss because of excessive water withdrawal include decreased tissue function and loss of mechanical integrity (Chapotin 2005). Although the water stored in the stems and branches of baobab trees is drawn upon during the leaf-flushing period and may buffer water deficits during occasional periods of extended drought, extensive use of stored water does not seem possible, and other explanations for the unusual morphology and high water content of baobab stems need to be examined.

### ACKNOWLEDGMENTS

This project was made possible through funding by the Sinclair Kennedy Traveling Fellowship, the Arnold Arboretum and the Department of Organismic and Evolu-

tionary Biology at Harvard University, the Garden Club of America and the National Science Foundation. We are thankful to Lalao Andriamahefarivo and to other Missouri Botanical Garden staff in Antananarivo for acquiring research permits and logistical support, and to the German Primate Center [Deutsches Primatenzentrum (DPZ)] for permission to work at the Kirindy Research Station. Michael Burns provided invaluable technical expertise and Randol Villalobos assisted with equipment assembly. Rodolphe Rasoloarivony, Heather Patt and Mario Ramohavelo provided field assistance. We thank the two anonymous reviewers for their comments and suggestions.

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Received 3 June 2005; accepted for publication 13 September 2005