RESEARCH PAPER

# Constant theoretical conductance via changes in vessel diameter and number with height growth in *Moringa oleifera*

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## Abstract

**As trees grow taller, hydraulic resistance can be expected to increase, causing photosynthetic productivity to decline. Yet leaves maintain productivity over vast height increases; this maintenance of productivity suggests that leaf-specific conductance remains constant as trees grow taller. Here we test the assumption of constant leaf-specific conductance with height growth and document the stem xylem anatomical adjustments involved. We measured the scaling of total leaf area, mean vessel diameter at terminal twigs and at the stem base, and total vessel number in 139 individuals of *Moringa oleifera* of different heights, and estimated a whole-plant conductance index from these measurements. Whole-plant conductance and total leaf area scaled at the same rate with height. Congruently, whole-plant conductance and total leaf area scaled isometrically. Constant conductance is made possible by intricate adjustments in anatomy, with conduit diameters in terminal twigs becoming wider, lowering per-vessel resistance, with a concomitant decrease in vessel number per unit leaf area with height growth. Selection maintaining constant conductance per unit leaf area with height growth (or at least minimizing drops in conductance) is likely a potent selective pressure shaping plant hydraulics, and crucially involved in the maintenance of photosynthetic productivity per leaf area across the terrestrial landscape.**

**Keywords:**  Leaf area-stem scaling, metabolic scaling, *Moringa oleifera*, whole-plant conductance index, xylem conduit number, xylem conduit widening.

## Introduction

Leaves are the engines of plant primary productivity, so it is important to understand how leaves maintain productivity as plants grow in height. As a tree grows from a seedling to maturity, the distance that water must be transported increases, and longer conductive path lengths could reasonably be expected to be associated with greater hydraulic resistance, lower conductance per unit leaf area (leaf-specific conductance), and lower productivity (Ryan and Yoder, 1997; Niklas and Spatz, 2004; Koch *et al.*, 2004). Yet, because carbon fixation should directly impact growth and fitness, changes in productivity with height growth should be a major target of selection. Given variation among the members of a species, individuals whose leaf-specific productivity declines markedly with height growth will necessarily be selected against in favor of

those whose productivity declines less or even remains constant. Individuals whose per-leaf area productivity declines least with height growth will by definition have more carbon to invest in growth and reproduction per unit leaf area than individuals with greater declines. As a result, selection should always push in the direction of constant leaf-specific productivity with height growth, or minimal possible declines in productivity. If constant productivity can be achieved, it necessarily involves constant conductance per unit leaf area, given the link between conductive rate and photosynthetic productivity (Santiago *et al.*, 2004; Brodribb, 2009). Given the relationship of leaf-specific conductance with terrestrial primary productivity, testing the prediction that leaf-specific conductance should remain constant with height growth, and understanding the mechanisms that make this possible, is a priority.

The invariance of leaf-specific conductance with height growth is assumed to be possible because of a xylem vascular system made up of conduits that widen from the stem tip to the base at a rate that compensates for the resistance imposed by increasing path length (West *et al.*, 1999; Anfodillo *et al.*, 2006). Because height growth in a tree takes the leaves ever farther from the base of the trunk, the conductive path length becomes longer as trees become taller. Conduit walls impose friction, creating resistance to flow, and, all else being equal, longer conduits have greater wall area, greater friction, and thus greater resistance to flow than shorter ones (Comstock and Sperry, 2000). Therefore, if conduits remain the same diameter, conductive rates per unit leaf area will inevitably drop as path length increases. Instead, conduits widen from the stem tip to the base with height, across plant habits, such as trees, shrubs, and lianas, across monocots and dicots, as well as across biomes (Anfodillo *et al.*, 2006; Coomes *et al.*, 2007; Petit *et al.*, 2009; Olson et al., 2014, 2018; Rosell and Olson, 2014; Morris *et al.*, 2018). This widening occurs at the rate predicted to offset the resistance imposed by increasing conductive path length and keep leaf-specific conductance constant, or at least minimize the drop in conductance. In the simplest models of hydraulic architecture (West *et al.*, 1999), a unit of leaf area is assumed to have a constant number of conduits as height increases, with basipetal vessel widening leading to constant resistance regardless of conduit length, and xylem conduits are assumed to have constant terminal conduit diameters (conduit diameter at the twig apex).

Our recent work suggests that, at least in angiosperms, terminal twig vessel diameter is not constant and instead increases predictably with height growth, suggesting that vessel number per unit leaf area might also change with height. In surveys across angiosperm species, we found that vessel diameter (*VD*) increased predictably with height (*H*) not only at the stem base, where it scaled as approximately $VD_{base} \propto H^{0.4}$, but also at the tip, where it scaled as $VD_{apex} \propto H^{0.2}$ (Olson *et al.*, 2014; Rosell and Olson, 2014). Wider conduits at the stem tip in taller trees would imply that flow within a single conduit also increases. Given two conduits with identical lengths and basal vessel diameters, the one with the wider apical diameter will offer less resistance to flow. If the number of conduits does not change, then increasing the terminal conduit diameters

would imply increasing the flow per unit leaf area. Such 'over-supply' seems unlikely, given that natural selection should favor fluid distribution networks that minimize network-level fluid volume and therefore construction costs (Banavar *et al.*, 2002). Assuming that conductance remains constant per unit leaf area, if conductance per unit of pipeline increases with tree size due to wider conduits at the stem tips in taller trees, then the number of conduits per unit leaf area must decrease accordingly. Although not conforming with the exact predictions of existing hydraulic scaling hypotheses (West et al., 1997, 1999; Hölttä et al., 2011, 2013; Drake *et al.*, 2015; Couvreur *et al.*, 2018), coordinated variation between tip conduit diameter, widening rate, and conduit number might still occur in such a way that a constant water supply per unit of leaf area is maintained as predicted.

Although the results of comparative studies to date suggest that the maintenance of leaf-specific conductance is plausible, studies of universal vessel scaling at the stem base and apex have been largely based on species-level mean values across many species, and have not included leaf area (Anfodillo *et al.*, 2006; Olson et al., 2014, 2018; Rosell and Olson, 2014; Morris *et al.*, 2018). As a result, whether conductive capacity does indeed scale isometrically with leaf area with height growth as predicted has not been tested. Moreover, because the patterns studied to date have been mostly comparative, direct documentation is missing to test whether the increases in terminal and basal vessel diameter occur within species and in such a way that they keep pace with leaf area.

We tested the prediction that the xylem network should supply a unit of leaf area with a constant flow of water during height growth in a plantation experiment with the tropical tree *Moringa oleifera* Lam. Focusing on a single species allowed us to factor out much of the variation that is introduced in comparative studies covering wide spans of habit, wood density, leaf size, leaf mass per unit area, growth ring types, and other factors that could potentially influence vessel dimensions. *Moringa* is useful for such studies because it grows quickly (8 m in the first year from seed is common, and plants fruit within 6 months) and saplings are monopodial, making accurate measurement of conductive path length feasible. We grew plants at a relatively high density for several months, and competition between plants resulted in a wide range of heights and leaf areas for the same age, ideal for estimating the rates of change with height in leaf area, vessel-widening rate, vessel diameter, total vessel number, and a whole-plant conductance index estimated from these measurements.

## Materials and methods

To provide the necessary variation in plant height and total leaf area in an acceptable time frame, we carried out an experiment using the tropical tree *Moringa oleifera* Lam. (Moringaceae, Brassicales). *Moringa oleifera* is a highly tractable woody plant study system because of its simple anatomy—wide vessels with simple perforation plates in a background of libriform fibers with axial parenchyma limited to 1–2 layers adjacent to vessels (Olson, 2001; Olson and Carlquist, 2001)—and its very fast growth. *Moringa oleifera* trees easily reach 8 m in their first year from seed, making it possible to produce plants of a wide range of heights under identical growing conditions and of the same age. The saplings grow

tall and straight, meaning that conductive path length, as approximated by the stem-tip-to-base distance, can be measured with precision (see Supplementary Fig. S1 at *JXB* online). To maximize genetic diversity and thus the likelihood of finding differing relationships between leaf area, vessel diameter, and stem length, we gathered seeds from 13 individuals of *M. oleifera* with cultivated provenances from Africa, Asia, Madagascar, and the Americas, from the International Moringa Germplasm Collection (http://www.moringaceae.org), on the lowland tropical Pacific coast of Mexico in Jalisco State.

### Experimental design

We planted 200 seeds directly in the ground at the International Moringa Germplasm Collection near the Chamela Biological Station, located at 19°29′54.34″ N, 105°2′40.46″ W. The region experiences a tropical monsoonal climate with marked dry and wet seasons, with most of the precipitation falling between June and October. The study area is characterized by an annual low temperature of 14.9 °C, a mean annual temperature of 24.9 °C, and an annual average rainfall of 752 mm (Bullock, 1986; Bullock and Solis-Magallanes, 1990; García-Oliva *et al.*, 2002). The elevation of the plantation site was ~100 m a.s.l. We planted the trees 1 m apart to encourage straight growth and competition to ensure a wide range of heights in plants of the same age. We watered the plants weekly throughout the prolonged dry season to encourage rapid growth.

### Sampling

We sampled from 5 to 13 April 2016. We harvested individuals by cutting them at soil level, and measured their heights and basal diameters. We fixed samples of the basal and terminal stem xylem from 139 individuals in 50% aqueous ethanol. All the slope estimations were performed including samples from all 139 individuals. We measured the fresh leaf area and dry leaf mass of 50 individuals to generate an equation to extrapolate leaf area from the dry leaf mass of each individual in our population. We scanned the leaves using a digital scanner (Seiko Epson, Tokyo, Japan), and used WinFolia (Regent Instruments Inc., Canada) to measure leaf area from digital images, including petioles and rachises of *M. oleifera*'s large, pinnately compound leaves. We dried the leaves and weighed them using an analytical balance (Sartorius Corporation, Gottingen, Germany).

### Anatomical characterization

We cut thin sections for light microscopy from the basal and terminal stem of each individual to measure vessel diameters and estimate the total number of vessels at the stem base. We used a sliding microtome (Gärtner *et al.*, 2014) to make transverse sections from the basal and terminal wood of each individual. We made temporary and permanent slides, and measured vessel diameter with a light microscope (Zeiss, Oberkochen, Germany). We measured vessel radial diameter, following radial transects from the pith to the vascular cambium, to capture intra-individual variation in vessel diameter. We measured 35 vessels per section to obtain per-individual mean basal and apical vessel diameters. We estimated the total number of vessels by measuring vessel density (vessels mm$^{-2}$) and multiplying this value by the total xylem area. We measured vessel density by counting the number of vessels in 35 optical fields spanning pith to cambium. We calculated the xylem area by subtracting the pith area from the xylem+pith area. Our analyses of xylem respiratory activity using triphenyl-tetrazolium chloride (Shain and Mackay, 1973; Spicer and Holbrook, 2007) suggest that for the range of sizes studied all the xylem is active sapwood (data not shown). Xylem conductive area was calculated by multiplying the mean vessel area by the total number of vessels.

### Statistical analyses

To estimate the slopes of all bivariate relationships, we used standardized major axis (SMA). In many of the relationships we examined, it seems likely that instead of one variable causing variation in the other, the variables affect each other mutually. SMA is appropriate in these cases (Smith, 2009). To reduce the influence of outliers on slope estimation, we used so-called robust SMA methods for estimating slopes of all bivariate relationships and for slope tests (Warton *et al.*, 2006; Taskinen and Warton, 2011, 2013). Moreover, robust SMA is less sensitive to outliers. In all models, we used log$_{10}$-transformed variables, with all analyses carried out in R (R Core Team, 2018) and graphs being generated using the R package ggplot2 (Wickham, 2016). Standard major axis line fitting and slope tests were performed using the R package smatr3 (Warton *et al.*, 2012).

### Testing the prediction of constant conductive capacity per unit leaf area

We used two complementary approaches to test the prediction that conductance should remain constant per unit leaf area as plants grow taller. Using the empirical scaling exponents given in Table 1, we estimated whole-plant conductance in the following way. We first estimated mean apical vessel diameter given plant height from the relationship between apical vessel diameter and height given by $VD_{apex} \propto H^a$. Subsequently, we used the relationship between basal vessel diameter and height given by $VD_{basal} \propto H^b$ to estimate the tip-to-base widening rate (*DistTip*) of vessels, assuming a power law relationship given by $VD_{basal} \propto H^a / VD_{apex} \propto H^b = VD \propto DistTip^c$. Given this tip-to-base widening rate, we calculated the total resistance of a widened tube of length $H$, made up of segments of 1 cm length, with initial diameter given by $VD_{apex} \propto H^a$. We estimated the theoretical flow within single widened tube segments ($1/R$)

**Table 1.** *Bivariate SMA models for allometric relationships between leaf area, plant height, vessel traits, and theoretical conductance*

| Model | Intercept | Slope | $R^2$ | 95% CI | |
|---|---|---|---|---|---|
| | | | | Intercept | Slope |
| log $LA$ ~ log $H$ | −0.694 | 1.859 | 0.84 | −0.996 to −0.391 | 1.725 to 2.00 |
| log $VD_{apex}$ ~ log $H$ | 1.219 | 0.268 | 0.48 | 1.134 to 1.304 | 0.232 to 0.309 |
| log $VD_{basal}$ ~ log $H$ | 1.291 | 0.341 | 0.84 | 1.236 to 1.346 | 0.316 to 0.367 |
| log $NV$ ~ log $H$ | 0.172 | 1.493 | 0.90 | −0.011 to 0.354 | 1.412 to 1.579 |
| log $VD_{basal}$ ~ log $LA$ | 1.427 | 0.180 | 0.73 | 1.371 to 1.482 | 0.165 to 0.198 |
| log $NV$ ~ log $LA$ | 0.786 | 0.789 | 0.89 | 0.637 to 0.936 | 0.746 to 0.835 |
| log $CA$ ~ log $XA$ | 4.931 | 1.064 | 0.98 | 4.875 to 4.987 | 1.040 to 1.088 |
| log $K_{plant}$ ~ log $H$ | −3.491 | 1.825 | 0.94 | −3.673 to -3.308 | 1.743 to 1.910 |
| log $K_{plant}$ ~ log $LA$ | −2.759 | 0.969 | 0.89 | −2.939 to -2.579 | 0.918 to 1.024 |

All relationships were derived from variables measured empirically, except for whole-plant conductance ($K_{plant}$), a trait derived from calculations based on empirically measured allometric relationships. $H$, plant height; $K_{plant}$, whole-plant conductance; $LA$, total leaf area; $NV$, total number of vessels; $VD_{apex}$, apical vessel diameter; $VD_{basal}$ basal vessel diameter; $XA$, xylem area. $n$=139 and $P<0.0001$ in all cases. Normality was tested through the Shapiro–Wilk $W$-test on residuals.

by calculating the resistance of 1 cm long segments using the Hagen–Poiseuille equation (Sperry *et al.*, 2005):

$$R = \frac{128\eta}{\pi D^4}$$

where $\eta$ is the viscosity of water and $D$ is vessel diameter. We summed the resistances of the 1 cm segments to calculate the total resistance of the widened tube along the entire conductive pathway length. We then multiplied the resistance of single widened tubes by the empirical total number of vessels in each plant to estimate whole-plant resistance. The reciprocal of the value for whole-plant resistance gave the whole-plant flow per plant. Whole-plant flow was transformed into a whole-plant conductance index ($K_{plant}$), assuming a driving pressure equivalent to a 50 cm water column. First, we tested whether the slope of the whole-plant conductance index against total leaf area scaled with a slope of 1. Second, we estimated the scaling relationships of total leaf area versus tree height and whole-plant conductance versus tree height to test the prediction that the resulting scaling exponents should be statistically indistinguishable. If whole-plant conductance keeps pace with total leaf area as plants grow taller, then the whole-plant conductance index and total leaf area should scale with height with slopes that do not differ significantly from one another. These two complementary approaches to our main question tested of the prediction of an isometric increase between total stem water flow and total leaf area with height growth. All tests were performed at $\alpha$=0.05.

## Results

Our results were consistent with the prediction of constant conductance per unit leaf area with height growth. The plants sampled encompassed over four orders of magnitude of difference in total leaf area, and over one order of magnitude of difference in height. Mean plant height ($H$) was 174.69 cm ranging from 13 to 420 cm, and most of the plants flowered. Mean per-individual total leaf area ($LA$) was 8212 cm$^2$, ranging from 16.53 to 31 431 cm$^2$. Mean per-individual basal vessel diameter ($VD_{basal}$) was 108.42 μm, ranging from 39.86 to 176.90 μm. Mean per-individual terminal twig vessel diameter ($VD_{apex}$, 'apical vessel diameter') was 63.54 μm, ranging from 32.00 to 107.31 μm. Mean xylem area in the basal section ($XA$) was 407.68 mm$^2$, ranging from 1.122 to 3604.30 mm$^2$. The mean total vessel number ($NV$) per individual in the basal xylem was 3170, ranging from 232 to 28 354. We used data from these trees to derive empirical exponents for the rate of change of total leaf area with height, and the rates of change of total vessel number, mean vessel diameter at the stem base, and mean apical vessel diameter with both height and total leaf area. We then used these exponents to test the prediction that whole-plant conductance should scale isometrically with leaf area as plants grow taller.

All traits analyzed scaled closely with plant height (Table 1). Total leaf area was predicted well by plant height, and scaled with height with a slope of 1.859 (Fig. 1A). Apical vessel diameter increased predictably with height with a slope of 0.268 (Fig. 1B). Basal vessel diameter was also closely predicted by height, and increased with a slope of 0.341 (Fig. 1C). The resulting rate of widening of the vessels in the plants of our population was $VD_{basal} \propto H^{0.341}/VD_{apex} \propto H^{0.268}$=0.073. Total vessel number increased with total leaf area with a slope of 0.789 (Fig. 1D), and its confidence interval did not include isometry (Table 1). Congruently, the total number of vessels also increased with height at a lower rate than total leaf area, with a slope of 1.493 (Fig. 1E). As a result of the increase in
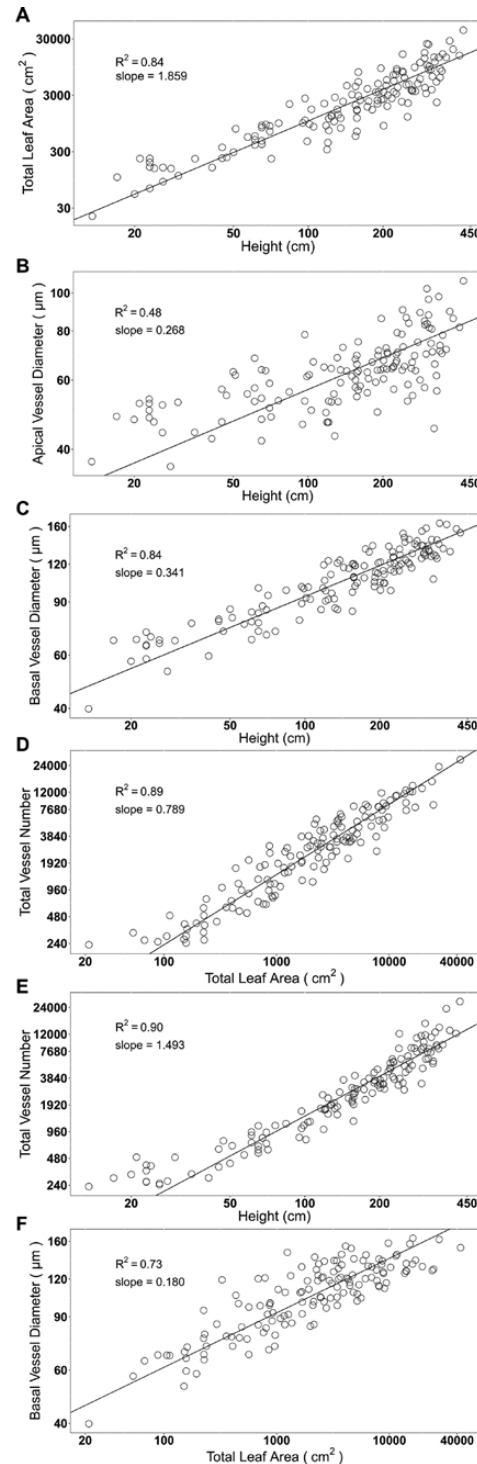


**Fig. 1.** Scaling relationships of total leaf area with height, and vessel traits with height and total leaf area, in *Moringa oleifera*. (A) Total leaf area closely scales with height as $LA \propto H^{1.859}$. (B) Vessel diameter varies predictably with height at the stem apex. (C) Given a constant widening rate, vessel diameter at the stem base also varies predictably with height. (D) Total vessel number increases at a lower rate than total leaf area, presumably because wider apical and basal vessel diameters with increasing height cause an increase in individual vessel conductance with height, allowing a similar area of leaf to be supplied by fewer vessels. (E) Accordingly, total vessel number increases with height at a lower rate than leaf area. (F) Average vessel diameter at the stem base becomes wider in taller plants, leading to an increase in basal vessel diameter with total leaf area. The confidence intervals for the slopes and intercepts are shown in Table 1. $P$<0.001 for all $R^2$. $n$=139 in all cases.

basal vessel diameter with height, basal vessel diameter also scaled predictably with total leaf area (Fig. 1F). Conductive area (*CA*), understood as the total vessel lumen area, increased at a higher rate than xylem area, with a slope of 1.063, and its confidence interval did not include isometry (Table 1). These scaling exponents were used to estimate a whole-plant conductance index and test whether it increased at the same rate as total leaf area with height growth.

The rate of increase in the whole-plant conductance index was statistically indistinguishable from the rate of increase in total leaf area with height growth, and congruently the whole-plant conductance index increased isometrically with total leaf area as trees increased in height. Calculated based on the empirical exponents in Table 1, whole-plant conductance increased with height at a rate of $K_{plant} \propto H^{1.825}$ (Fig. 2A), which did not differ significantly from the rate of increase in total leaf area with height ($F_{1,137}=0.644$, $P=0.424$), which was $LA \propto H^{1.859}$ (Fig. 1A). Also in agreement with predictions, the whole-plant conductance index scaled with leaf area with an exponent of 0.969 (Fig. 2B), and did not differ significantly from isometry ($F_{1,137}=1.249$, $P=0.266$). Thus, both methods of calculation are consistent with the prediction that whole-plant conductance should scale isometrically with leaf area with height growth.

## Discussion

In *Moringa oleifera,* whole-plant conductance increased at the same rate as leaf area with height, providing clues regarding a major vector of natural selection on plant hydraulic systems (Fig. 2). This coincidence with predictions is remarkable since it emerges from the concatenation of a series of empirical scaling exponents (Table 1), namely, leaf area scaling with height $LA \propto H^{1.859}$, stem tip vessel diameter with height $VD_{apex} \propto H^{0.268}$, the rate of widening of vessel diameter with distance from the stem tip $VD \propto DistTip^{0.073}$, and the total number of vessels with leaf area $NV \propto LA^{0.789}$. Even relatively small variation, due to error or biological reality, in any of the scaling exponents involved would lead to substantial deviations in the whole-plant conductance–leaf area scaling relationship, making the coincidence we observed between results and predictions striking.

Constant whole-stem conductance per unit leaf area would have important implications at both individual and community scales. Predictable tip-to-base vessel widening within leaves accounts for vessel diameter scaling predictably with individual leaf area across species (Sack *et al.*, 2012; Gleason *et al.*, 2018). Standardizing measurements of vessel diameter for leaf area should reveal that leaves within a tree are similar from the point of view of vessel diameter and widening rate. This expectation
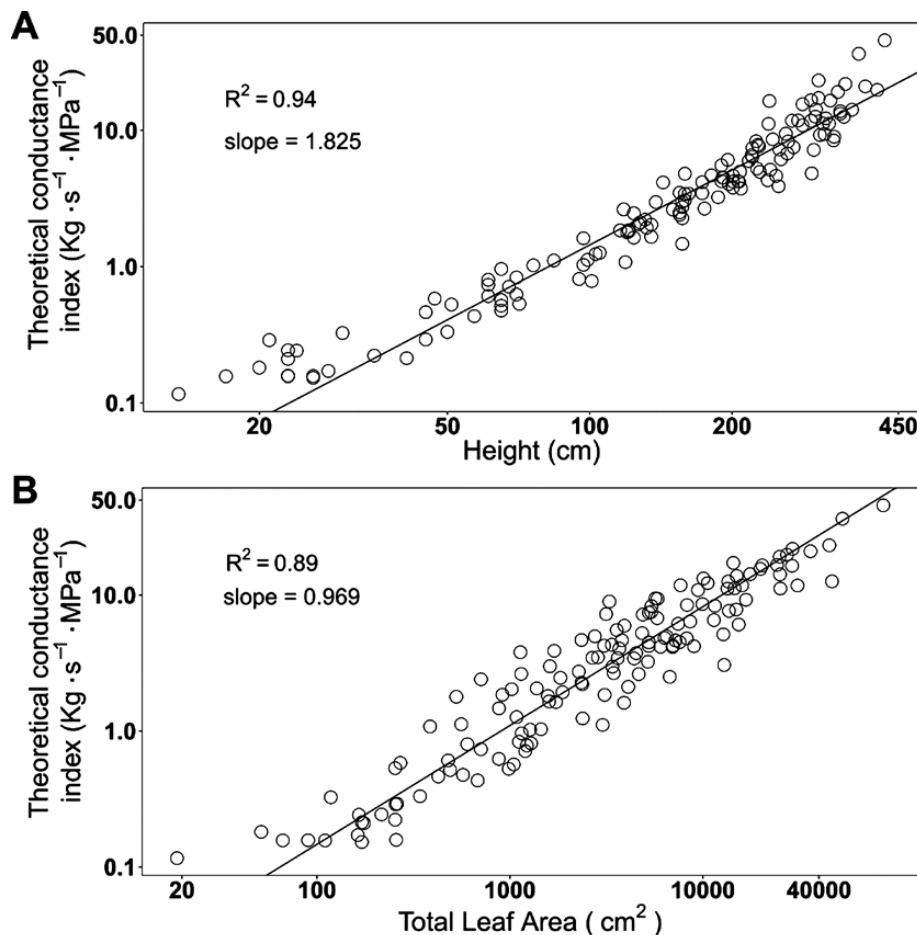


**Fig. 2.** Leaf-specific theoretical conductance remains constant with height growth in *Moringa oleifera.* (A) Theoretical whole-plant conductance ($K_{plant}$) scales statistically identically with height, as does leaf area, as $K_{plant} \propto H^{1.825}$ (see Fig. 1A). (B) $K_{plant}$ scales isometrically with total leaf area, congruent with expectations regarding the way in which selection should favor individuals with constant photosynthetic productivity per unit leaf area with height growth. Confidence intervals for slopes and intercepts are shown in Table 1. $P<0.001$ for all $R^2$. $n=139$ in all cases.

implies that leaves can in principle be functionally equivalent in terms of their vessel dimensions on a per-area basis, regardless of their position in the crown and distance from the stem base. At the community scale, constant leaf-specific conductance allows estimation of whole-community productivity as proportional to the sum of the leaf area of trees within the community (West *et al.*, 2009). Therefore, knowing how leaf area (and thus productivity) scales as individual trees increase in size, together with the assumption that forest resource use is always maximal (meaning that a community is saturated with individuals, whether it is composed of many small or few large individuals), allows prediction of the slope of tree size distributions in any community (Simini *et al.*, 2010; Anfodillo *et al.*, 2012). Deviation from the predicted distribution can even be diagnostic of forest disturbance (Coomes *et al.*, 2003; Sellan *et al.*, 2017). This energy equivalence expectation, supported here by constant conductance per unit leaf area, represents a potential link between hydraulics, stand productivity, and the shaping of optimal canopy height and density by microsite (Eagleson, 1982; Cabon *et al.*, 2018). The finding of constant conductance per unit leaf area with height growth is therefore important for understanding forest function.

With regard to understanding plant hydraulic architecture, our results point to an intricate coordination between plant height, a constant tip-to-base vessel-widening rate, and variation in vessel diameter at the stem base and at the stem apex—the latter a feature not predicted or included in any model of plant hydraulic function to date (Figs 1 and 3). As an individual becomes taller, the vessels at the stem apex become wider, with $VD_{\mathrm{apex}} \propto H^{0.268}$ (Fig. 1B). Given a constant tip-to-base vessel-widening rate, wider vessels at the stem apex will lead to even wider vessels at the stem base with height growth (Fig. 1C) than would be produced with uniform terminal stem vessel diameters. Given two conduits of identical length and basal
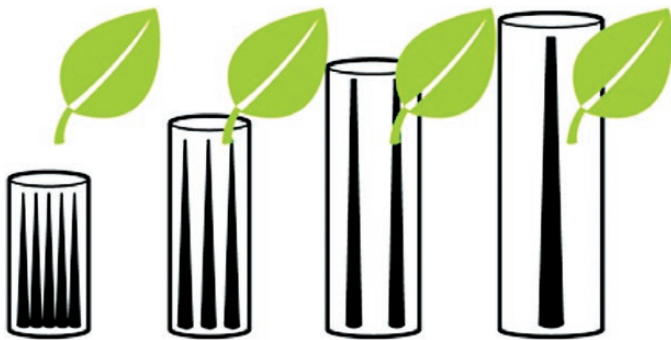


**Fig. 3.** The intricate covariation of plant height, tip-to-base vessel widening rate, stem tip vessel diameter, stem base vessel diameter, and number of vessels per unit leaf area. The white cylinders represent plants of different heights; the black cones within them represent vessels. In our experiment, for a given unit of leaf area, represented by the constant-sized leaf above each stem, the number of vessels decreased with increasing plant height. At the same time, terminal vessel diameter increased, represented by the wider apices of the vessels. Given tip-to-base vessel widening, the rate of which is constant over height increases, vessel diameter also becomes predictably wider at the stem base with height growth, represented by the wider bases of the vessels. This delicate synchronization of multiple factors led to a similar whole-plant conductance per leaf area as individuals become taller. (This figure is available in colour at *JXB* online.)

diameter, the one with the wider terminal diameter will have lower resistance with only a minimally greater investment in conduit wall material, given that flow is expected to increase as $VD^4$ while conduit wall material would increase proportionally to vessel perimeter and therefore only linearly with the increase in vessel diameter. As a result, terminal twig conduit widening with increasing height seems likely to be a major aspect of plant economy, minimizing resource investment per unit leaf area with height growth (Bettiati *et al.*, 2012; Olson *et al.*, 2014). It is also likely what allowed the total number of vessels in our study plants to increase with leaf area with an exponent of less than 1, $NV \propto LA^{0.789}$ (Fig. 1D). This means that taller plants supply the same leaf area with proportionally fewer vessels (Fig. 1A, E), and again the widening of terminal conduit diameter and the reduction in resistance it brings is surely involved in this proportional reduction in vessel number (Fig. 3). In turn, this reduction is likely the result of selection favoring minimized increases in sapwood carbon cost per unit leaf area with height increase (Olson *et al.*, 2014; Rosell *et al.*, 2017); indeed, it is hard to imagine what other process would lead to terminal conduit widening with height. Our results in *Moringa*, together with patterns of vessel scaling with stem length across vessel-bearing angiosperms (Anfodillo *et al.*, 2006; Olson et al., 2014, 2018; Rosell and Olson, 2014), show that as plants grow taller, not only stem basal diameter but also terminal twig vessel diameter becomes wider, with a concomitant reduction in the number of vessels per unit leaf area, giving clues to significant vectors of selection on plant hydraulic systems.

One of these vectors is carbon economy, and our results suggest an additional feature that is not incorporated into any optimality model of plant hydraulic construction, which is that vessel lumen transectional area relative to the rest of the xylem becomes slightly higher with height increase. In *Moringa*, the number of vessels per unit leaf area scales as $NV \propto LA^{0.789}$, implying that taller plants have fewer vessels per unit leaf area. Yet, even though there are fewer vessels per unit leaf area with height growth, the proportion of vessel lumen area per unit leaf area increases in taller plants. Vessel diameter scales with leaf area as $VD_{\mathrm{basal}} \propto LA^{0.180}$ (Fig. 1F). Consequently, individual vessel area scales with leaf area as $VA \propto LA^{0.360}$, and since the total number of vessels scales with leaf area as $NV \propto LA^{0.789}$, conductive area scales with leaf area as $CA \propto LA^{1.149}$. These findings coincide very closely with the predictions of West *et al.* (1999), where $NV \propto H^3$, $VA \propto H^{0.5}$, and $LA \propto H^3$, such that $CA \propto LA^{1.15}$. While this finding is remarkably congruent with the predictions of West *et al.* (1999), when expressed in terms of xylem area it points to additional details not incorporated in their model, namely that conductive area scales as $CA \propto XA^{1.06}$, meaning that the proportion of vessel lumen area relative to the rest of the xylem becomes slightly higher with height growth (Olson *et al.*, 2014). This suggests that as trees grow taller, they deviate some carbon from support tissue such as fibers to vessels. This property very likely helps trees maintain leaf-specific conductance constant with height growth while minimizing the increases in carbon costs per unit leaf area expected to occur as plants grow taller (Hölttä *et al.*, 2011).

The isometric increase between leaf area and whole-plant conductance estimated using only vessel dimensions also

suggests that other properties of plant hydraulic systems that are known to contribute to the resistance of the vascular system must also increase proportionally with vessel diameter. Our index of conductance was calculated using the Hagen–Poiseuille equation assuming continuous tubes running the entire lengths of stems. This assumption omits much biological detail, including the irregularity of internal vessel walls, the constrictions imposed by perforation plates, and the finite lengths of vessels and the concomitant passage of water through pits from vessel to vessel. Yet, this simplified set of assumptions accurately predicts vessel diameter–stem length exponents, as well as yielding results consistent with predictions of constant leaf-specific conductance with height growth. This suggests that the other sources of resistance, including the distribution of vessel lengths from the tip to the base of a tree, must scale similarly with vessel diameter. End-wall resistance and lumen resistance increase isometrically among a diverse set of tracheid- and vessel-bearing plants (Sperry *et al.*, 2005), suggesting that selection minimizing decreases in conductance has also been an important factor influencing conduit length distributions. Similarly, total pit permeable area increases isometrically with tracheid lumen area (Schulte, 2012; Lazzarin *et al.*, 2016), and since pore diameter also increases proportionally to pit membrane area, pit resistance likely also remains a constant fraction of total resistance as trees increase in size (Schulte *et al.*, 2015). These proportionalities must occur for simple Poiseuille assumptions to predict conduit diameter scaling accurately. Ultimately, proportionality between different components of the total resistance of the conductive system is expected to result from selection favoring constant conductance per leaf area as trees increase in height.

In summary, our results show that during height growth, plants maintain a constant supply of water per unit leaf area through an intricate interplay between the tip-to-base vessel-widening rate, terminal twig vessel diameter, and the total number of vessels. Our findings support the general prediction that increasing height is accompanied by compensatory changes in xylem structure that maintain leaf photosynthetic productivity constant, a finding that underwrites the scaling of carbon assimilation from leaves to trees to communities.

## Supplementary data

Supplementary data are available at *JXB* online.

Fig S1. View of the experimental *Moringa oleifera* plantation and an individual tree.

## Acknowledgements

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