Seeing the forest with the leaves – clues to canopy placement from leaf fossil size and venation characteristics

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ABSTRACT

Although a variety of leaf characteristics appear to be induced by light environment during development, analysis of ontogenetic changes in living broad leaved trees has suggested that a number of other traits also lumped into the classic 'sun' versus 'shade' morphological distinctions, including leaf size, shape, and vein density, are instead controlled largely by local hydraulic environment within the tree canopy. The regularity in how these traits vary with canopy placement suggests a method for addressing a classic paleobotanical quandary: the stature of the source plant – from herb or shrub to canopy tree – is typically unknown for leaf fossils. The study of *Ginkgo* here complements previous work on *Quercus* that indicated that leaves throughout the crown are identical in size and venation at the time of bud break and that morphological adaptation to the local microenvironment takes place largely during the expansion phase after the determination of the vascular architecture is complete. Hence, variation in vein density does not reflect differential vein production so much as the distortion of similar vein networks over different final surface areas driven by variation in local hydraulic supply during expansion. Unlike the diffusely growing leaves of the angiosperm, *Quercus*, the marginally growing leaves of *Ginkgo* do show some potential for differential vein production, but expansion effects still dominate. The approach suggested here may prove useful for assessing the likelihood that two distinct fossil morphospecies actually represent leaves of the same plant and to gather information concerning canopy structure from disarticulated leaves.

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INTRODUCTION

Whole plants and the fossil record

Plants fall apart. The paleobotanical record overwhelmingly consists of isolated leaves, wood, and reproductive structures rather than complete shoot systems, a problem further compounded by the tendency for organ types to accumulate under different circumstances with different degrees of time averaging (Kidwell, 2001). Much paleobotanical research is focused on the reassembly of the whole plants that are essential for systematics, but this practice is only applicable to sites with the highest quality preservation. As a result, Carboniferous coal swamps and a few other time intervals and locations loom disproportionately large in our understanding of whole plants, while in other large geographic areas, such as the Permian of the southwestern USA (White, 1929; DiMichele *et al.*, 2004; Chaney *et al.*, 2006; DiMichele *et al.*, 2007), paleoecology has progressed, but many endemic taxa are known only from leaves. In fact, many higher-level lineages, such as Caytoniales, Peltaspermales, and Czekanowskiales, are known almost exclusively from deciduous leaves and reproductive organs for which ties to a stem and overall habit are tenuous at best (Taylor & Taylor, 1993; Doyle, 2006; Hilton & Bateman, 2006).

The use of most fossil localities for evolutionary or ecological studies depends on information concerning whole plant architecture compiled from other sites where preservation is more complete, but, even when available, such external information can be difficult to apply. Paleozoic *Cordaites* can be a tall tree (Falcon-Lang & Bashforth, 2004) or a small shrub (Rothwell & Warner, 1984); which habit should be inferred for a locality where only *Cordaites* leaves are known? Inference from living relatives can be equally problematic: for angiosperms, a complex diversity of habits from herbs to trees are often encompassed by individual families (Ricklefs & Renner, 1994; Mabberley, 1997) and, for non-angiosperms, the architectural ranges of living exemplars are suspect as predictors for fossil relatives because much broader ecologies likely existed (Wing
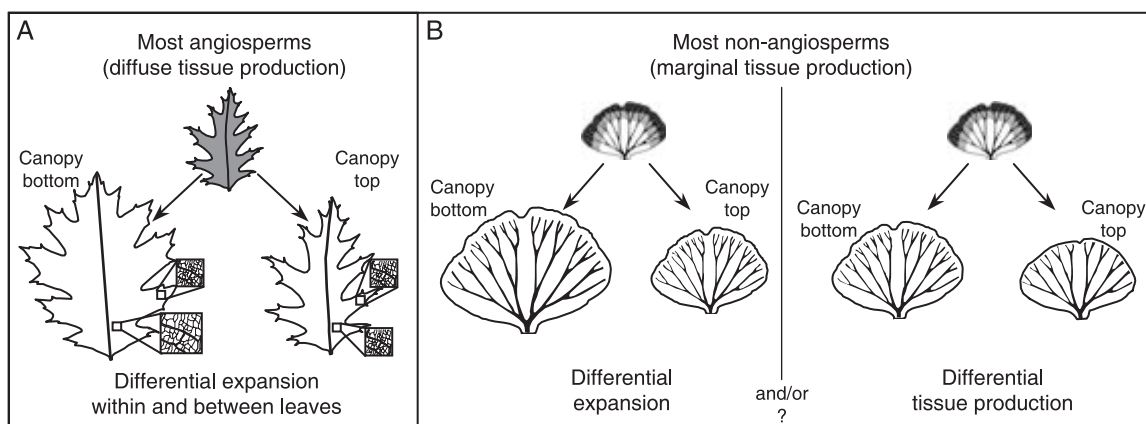
Fig. 1 Mechanisms of morphological differentiation in growing leaves.
(A) The morphologies of diffusely growing angiosperm leaves, such as *Quercus*, diverge based on greater expansion of leaves at the bottom of the canopy, particularly in proximal areas near major veins (accounting for the differences in leaf shape). (B) The morphologies of marginally growing leaves, such as *Ginkgo*, may diverge based on greater expansion of leaves at the bottom of the canopy and/or based on early cessation of marginal tissue production in leaves from the top of the canopy. Shading indicates location of tissue production in growing leaves. For simplicity, overall vein number was reduced relative to actual leaves.

& Sues, 1992) before the restriction of these lineages by the global rise of angiosperm dominance. External information can be particularly misleading during times of rapid evolutionary or environmental change. For example, timing the ecological spread of angiosperms to the forest canopy by inference of habit from the closest living relatives of leaf fossils would be dependent on dangerous assumptions concerning the ordering of trait acquisition. Hence, diverse applications would be available if information could be gleaned concerning the whole plant from individual organs, such as if the total leaf area of a tree crown could be estimated from hydraulic parameters of the wood or if plant stature could be estimated from the leaves.

Variability of leaves within a tree

A number of leaf morphological and anatomical traits that vary over a tree canopy have traditionally been thought of as 'sun' versus 'shade' characteristics for which differences are induced by the local light environment (Dickison, 2000), however, recent work suggests an alternative explanation for variance in a subset of these traits – and specifically for differences in leaf size, shape, and venation that can be preserved in fossils – based on differential hydraulic limitations at the top and bottom of the canopy. Plant primary growth is of limited duration and consists of successive stages, beginning with an interval of tissue production by cell divisions and followed by a second discrete interval of dramatic volumetric growth of the tissue without further cell division due only to the expansion of existing cells through water uptake by the cell vacuole (Sachs, 1991). In oak, leaves at the top and bottom of the canopy were found to be identical at the time of bud break and to possess the full complement of veins, differentiation only came later during leaf expansion (Zwieniecki *et al.*, 2004a). With the

greater hydraulic supply available to leaves at the bottom of the canopy relative to leaves at the top, the same leaf tissue and vein network is stretched during expansion over a greater area. There is even differentiation within individual leaves (Zwieniecki *et al.*, 2004a), so that there is greater expansion in the proximal areas near the water supply of major veins than in the distal areas near the leaf margin (Fig. 1A). The importance of hydraulics as a determinant of leaf morphology has been further supported by theoretical work that could reproduce the range of linear leaf morphologies exhibited by plants with only the systematic variation of hydraulic parameter values over a biologically reasonable range and then determining the largest leaf that could be adequately supplied with water (Zwieniecki *et al.*, 2004b).

Due to differential expansion, larger leaves with low vein density are produced that are well adapted to the low light and low transpirational demand they experience in the lower canopy, and smaller leaves with high vein density are produced that are well adapted to the high light and high transpirational demand they experience in the upper canopy – and these differences are produced before the irradiance of the two environments diverge (Zwieniecki *et al.*, 2004a). Light induction is important for differences that are established at the end of leaf development, such as in chlorophyll content and cuticle thickness (Kürschner, 1997; Barbacka & van Konijnenburg-van Cittert, 1998; Dickison, 2000; Guignard *et al.*, 2001), but a serious hurdle exists to light playing a major role in determining leaf size: leaves typically are the very source of self-shading that determines light levels within a canopy. Thus, size and vein density differences are established during leaf expansion, but all leaves are equally 'sun' leaves until after that expansion occurs.

Understanding of differential expansion of similar leaves provides previously unavailable opportunities for the interpretation

of the whole plant from leaf fossils. Since leaf size and vein density are both measurable in fossils, whole plant size – at least at a binary level of larger tree versus shrub or other smaller understory element – may be distinguishable by the variability of these traits among a sample of fossil leaves. A few examples of sun/shade pronouncements in fossils have been made primarily based on epidermal and cuticular characteristics (e.g. Kürschner, 1997; Barbacka & van Konijnenburg-van Cittert, 1998; Guignard et al., 2001), but a mechanism of light induction provides no clear expectations of how variance within other individual sun/shade traits should be correlated or how to reliably distinguish the phenotypic range within a plant from that of multiple distinct species. Differential leaf expansion after similar tissue production, however, provides explicit requirements for how leaf size, vein density, and the overall venation network should be related.

Expectation of consistent leaf trait correlations across a canopy is complicated by alternative modes of leaf growth. While tissue production in angiosperms occurs diffusely throughout the leaf (Foster, 1952; Pray, 1955; Poethig & Sussex, 1985; Dolan & Poethig, 1998), most other plants have cell divisions adding new tissue limited to a band of activity along the distal margin (Pray, 1960, 1962; Zurakowski & Gifford, 1988; Boyce, 2007). These leaves with marginal growth, which dominate the first 250 million years of the leaf fossil record prior to the radiation of the angiosperms (Boyce, 2005; Boyce, 2008), might be expected to show different patterns of variance within a canopy since marginal growth places all tissue production directly at the distal site of maximum hydraulic stress. Thus, while oak showed reduced expansion in the distal, marginal areas that are both the most exposed to the environment and the farthest from the proximal water source, a marginally growing leaf may also exhibit less distal tissue production if activity of the marginal meristem is truncated by water stress (Fig. 1B).

This issue is here investigated with study of the marginally growing leaves of Ginkgo, including ontogenetic analyses in order to compare the role played by expansion in the morphological differentiation of marginally growing leaves to that previously established for expansion in diffusely growing leaves and measurement of mature leaves from trees of different heights in order to begin to assess the possibility of gleaning canopy information from the dispersed leaf record. Building on previous investigation (Zwieniecki et al., 2004a), this work provides a case study from which explicit hypotheses are developed concerning how leaf size, vein density, and absolute vein number should covary. Once tested across a broader array of living plants, these hypotheses may provide a method for assessing the stature of the parent plant from the morphological variance found in a fossil leaf bed.

METHODS

Leaves were sampled at heights of 2 and 10 m from the base of a female Ginkgo tree approximately 15 m tall every 2 or 3 days throughout leaf growth from bud break to completion of leaf expansion, and were then sampled one more time in late summer to verify that no part of the spring growth period was missed. Measurements were limited to short shoot leaves because long shoot leaves are both morphologically distinct and produced continually over the growing season. Only the three largest and most peripheral leaves of each short shoot were sampled because including the younger, central leaves would obscure patterns by averaging across an ontogenetic progression. Leaves that were grossly misshapen or damaged were excluded. Two or three short shoots were collected at each height per collection date with a minimum of six leaves analyzed per sample.

Ginkgo leaves are tightly enrolled early in ontogeny (Fig. 2), preventing exact and complete measurements of the area and
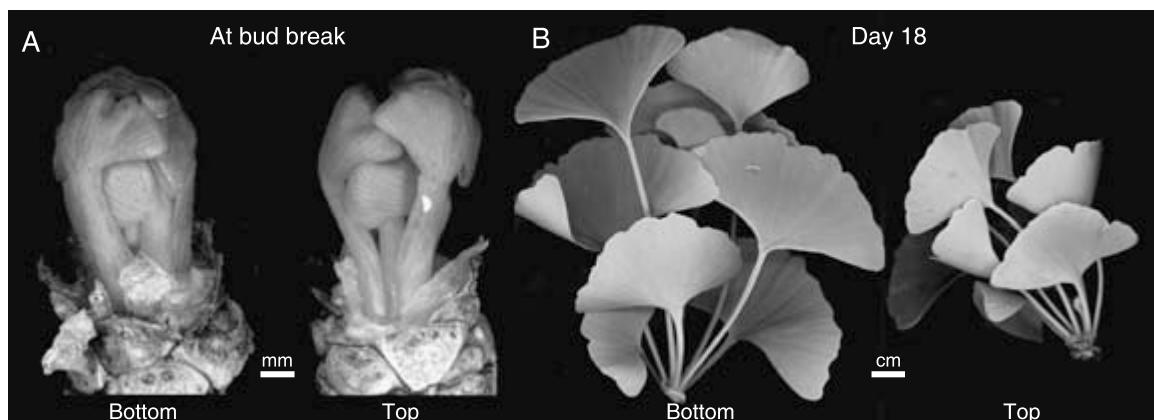

Fig. 2 Examples of Ginkgo leaves early and later in growth.
Short shoots from the bottom and top of the canopy shown (A) on the day of bud break and (B) 18 days later. Bud scales removed from the younger short shoots to expose the leaves.

venation of a leaf for the first 10 days after bud break. However, the distal margin along which growth occurs and along which all veins end serves as an indication of total leaf size that can be measured consistently in all leaves regardless of age. This distal perimeter is measured as the minimum convex path, rather than the complete marginal length, in order to exclude the irregular lobing and crenulation that is typical of *Ginkgo* leaves but is not relevant to their overall size. Vein density was measured in two ways. First, either the total number of vein endings was tallied in leaves sufficiently flat, providing veins/mm when coupled with perimeter measurements above, or the distance spanned by as many consecutive vein endings as visible was measured in younger leaves (five to 14 endings in the youngest and most tightly enrolled). Second, vein density was also measured as the length of vein per area of lamina (mm/mm^2) at six discrete points across the leaf in leaves 12 days past bud break and again in fully mature leaves.

Additionally, measurements of distal perimeter, vein density, and vein number were made on mature leaves from a smaller *Ginkgo* tree for which 20 leaves were sampled from each of the canopy bottom (2 m) and top (6 m) following the same sampling strategy outlined above for the ontogenetic analysis.

Perimeters and vein densities measured with IMAGE J (National Institute of Health, Bethesda, MD, USA). Statistical analyses performed using PAST (Hammer, Harper, and Ryan, 2001).

RESULTS

Ontogeny of leaf growth in *Ginkgo*

At the time of bud break, *Ginkgo* leaves are similar in both size and venation density at the top and bottom of the canopy, differentiation comes only with later growth (Fig. 3A,B). By the time leaves are relatively flat, at day 10, the total number of vein endings is established and no change is seen over later ontogeny (Fig. 3C), indicating that all tissue production at the marginal meristem has ceased. Since differences in perimeter length are largely established after day 10, this morphological divergence is primarily the result of differential expansion, as was previously seen in oak. However, a small but significant difference (*t*-test: $P_{(equal\ means)}$ << 0.001) in the total number of vein endings – for all leaves measured for the ontogenetic analysis, a cumulative mean of 129.1 (17.1 standard deviation (SD)) vein endings in leaves from the top of the canopy and 156.9 (20.5 SD) at the bottom – suggests that marginal meristematic production of tissue was truncated earlier in the more stressed leaves from the top of the canopy. Thus, leaves from the top of the canopy have a higher vein density despite actually having fewer veins overall because that vein network is stretched less over a smaller area during expansion.

While previous work in oak showed substantially more expansion proximally near the major veins than near the distal margin of the leaf, *Ginkgo* shows a much more even pattern
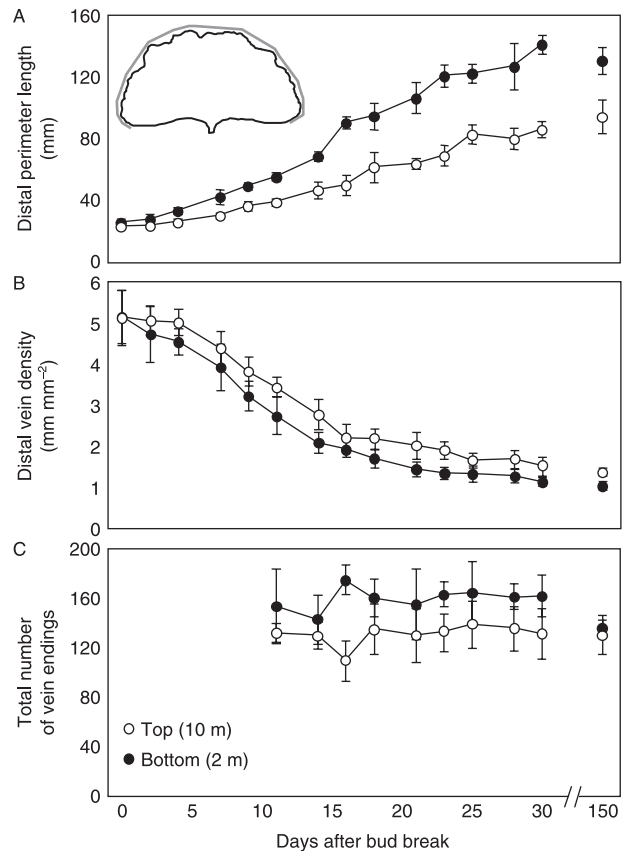

Fig. 3 Ontogenetic divergence of *Ginkgo* leaves from the canopy top and bottom. (A) Length of distal leaf perimeter through ontogeny. Perimeter measured is only that along which veins end and measurement smoothes over the irregular marginal crenulations, as indicated by the gray line on the inset leaf diagram. (B) Vein density through ontogeny, as measured at the vein endings along the margin. (C) Total number of vein endings through later ontogeny (a measurement not possible in younger leaves that are enrolled). In all graphs error bars indicate ± one standard deviation. Measurements made every 2 or 3 days for the first 30 days after bud break and then once again 4 months later.

(Fig. 4). There is indeed a substantially lower density near the proximal attachment of the leaf earlier in ontogeny (Fig. 4A) (ANOVA: $P_{(equal\ means)}$ << 0.001 for both canopy top and bottom); however, this likely reflects the tendency in marginally growing leaves for expansion to begin proximally while cell divisions and tissue production are still occurring distally (Boyce, 2007), and this pattern is lost by the time the leaves are fully expanded (Fig. 4B) (ANOVA: $P_{(equal\ means)}$ > 0.30 for both canopy top and bottom). In mature leaves, vein densities are relatively flat and there is no conspicuous increase in vein density near the distal margin. Whether this lack of an overall pattern, contrary to what was seen in an angiosperm, reflects the different vein anatomy and hydraulics of equitable water delivery in leaves with a single order of long, nonreticulating veins (Zwieniecki *et al.*, 2005) or reflects the preemptive loss of potentially water stressed tissue due to truncation of marginal growth before expansion occurs, patterns of differential expansion within individual leaves may be unlikely to be prevalent in
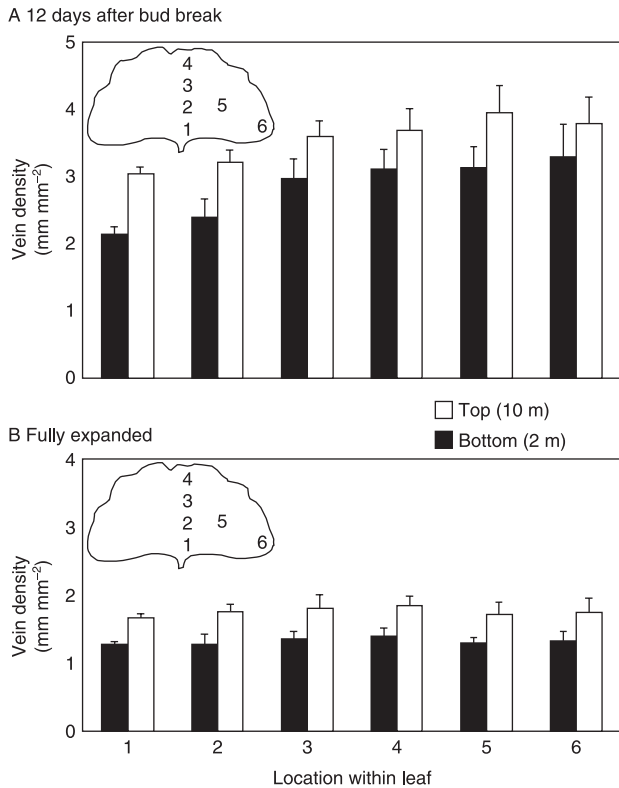
A 12 days after bud break

B Fully expanded

Fig. 4 Distribution of vein densities in growing and mature leaves. Vein density (mm/mm) at six points indicated on the inset diagram leaf measured at the top (white bars) and bottom (black bars) of the canopy 12 days after bud break and again when leaves were fully expanded. Error bars indicate ± one standard deviation.

other plants that also have marginal growth, at least those with venation as simple as the single order of open veins found in *Ginkgo*.

Canopy variation in Ginkgo trees of different height

In contrast to the substantial differences seen between leaves sampled at 2 m and 10 m height in the larger tree discussed above (Figs 3 and 4), leaves sampled at 2 m and 6 m from the bottom and top of a smaller *Ginkgo* tree exhibit little morphological distinction. No significant difference is seen between the bottom and top of this smaller canopy as far as distal perimeter, means of 152.3 mm (19.2 SD) and 147.8 mm (11.7 SD), respectively (*t*-test: $P_{(equal\ means)}$ = 0.36), or vein number, means of 179.2 (17.0 SD) and 185.9 (12.8 SD), respectively (*t*-test: $P_{(equal\ means)}$ = 0.16). The difference between bottom and top vein density is significant, means of 1.18 veins/mm (0.07 SD) and 1.27 veins/mm (0.06 SD), respectively (*t*-test: $P_{(equal\ means)}$ << 0.001); however, this difference of 8% is small in comparison to the 33% difference in vein density seen across 8 m of canopy height in the mature leaves of the larger *Ginkgo*, means of 1.09 veins/mm (0.12 SD) and 1.45 veins/mm (0.19 SD) for the two last ontogenetic sampling dates (*t*-test: $P_{(equal\ means)}$ << 0.001).

ASSESSING PLANT HABIT FROM FOSSIL LEAVES

As previously demonstrated in an angiosperm with diffuse leaf growth, divergence in overall leaf morphology between the top and bottom of a *Ginkgo* tree crown is primarily due to differential leaf expansion. Differential production of tissue at the marginal meristem is distinguishable, but its role is limited in *Ginkgo* and its relative importance may be even less in many other marginally growing leaves. In the distinct fan-shaped leaves of *Ginkgo* with a single order of veins that dichotomize all along their length, any loss of marginal growth will result in fewer vein dichotomies, but many other plants with marginal leaf growth have vein dichotomies concentrated proximally. In such leaves, a truncation of marginal tissue production may lead to a shorter leaf, but is unlikely to alter the total number of veins (Fig. 5A). The outcome of an early termination of
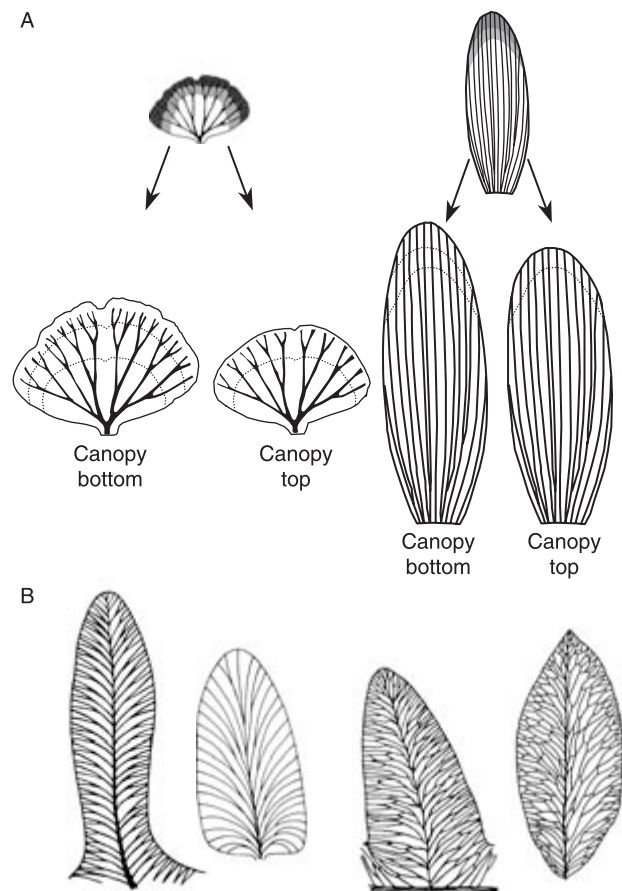

Fig. 5 Effect of marginal growth truncation on different leaf morphologies. (A) Early cessation of meristem activity will result in fewer vein endings in leaves with distributed vein dichotomies, such as *Ginkgo*, but will not alter the number of veins in leaves with proximally concentrated dichotomies, such as *Cordaites*. Shading indicates location of tissue production in growing leaves. For simplicity, overall vein number was reduced relative to actual leaves. (B) Examples of marginally growing leaves with more complex venation and meristem growth for which predicting effects of truncating marginal growth is more difficult.

meristematic growth cannot be so easily predicted for a third class of marginally growing leaves, those with a more complex architecture of a primary midvein and branching secondaries (Fig. 5B). There, the interplay between the different hydraulic conductivities of the midvein versus secondary veins and the different growth rates along distal and lateral margins will determine whether a truncation of marginal growth results in fewer dichotomies within the secondary veins, in production of fewer secondary veins off of the midvein, or a combination of both. However, the higher vein densities resulting from less expansion at the top of the canopy should still be associated only with either a decrease or no change in vein production regardless of the exact venation architecture that results.

The divergence of leaf size and venation characteristics within a tree canopy, thus, is likely to result only from differential leaf expansion except in the case of an easily recognizable subset of the plants with marginal leaf growth and, even within this subset, higher vein density leaves from the canopy top are expected to have an equal number or fewer veins relative to the canopy bottom, rather than more. The presence at a fossil locality of larger, lower vein density leaves and smaller high vein density leaves that are otherwise similar may suggest the sun and shade leave of a single tree species, but how to evaluate that supposition is not obvious if the pattern results from the induction of increased vein production by high light microenvironments. However, the alternative mechanism of differential leaf expansion now seen in both marginally (Fig. 3) and diffusely growing leaves (Zwieniecki *et al.*, 2004a) suggests further expectations that should be met (Table 1). These hypotheses must first be subjected to testing against a broader sampling of living plants than the two species on which they are based, but eventually may provide a tool for paleontological investigation.

As an example, Late Paleozoic *Cordaites* leaves were borne on plants ranging from low shrubs to tall trees. The leaves are long and strap-shaped with a single order of veins running to the distal margin with proximally concentrated vein dichotomies.

Table 1 Expectations of morphological variance within a canopy based upon mode of leaf growth.

Leaf type	Expectations for canopy variance
Diffusely growing	Greater expansion at canopy bottom (including more near main veins)
	Higher vein density at canopy top
	Equal vein number throughout canopy
Marginally growing:	
Single vein order (vein branching evenly spread)	Greater expansion at canopy bottom
	Higher vein density at canopy top
	Fewer veins at canopy top
Single vein order (vein branching only proximal)	Greater expansion at canopy bottom
	Higher vein density at canopy top
	Equal vein number throughout canopy
Multiple vein orders	Greater expansion at canopy bottom
	Higher vein density at canopy top
	Equal or fewer veins at canopy top

The true veins of the leaf as well as the sclerenchymatous ribbing parallel to the true veins that is typical of *Cordaites* (Harms & Leisman, 1961; Crookall, 1970) might equally serve as landmarks established in early development for the tracking of later expansion as long as they are included or excluded consistently. However, in those species with a different number of intervein ribs on the abaxial and adaxial leaf surfaces, analysis of the species would require that the ribbing could be clearly distinguished from the veins and excluded unless cuticular information was to be available that could verify that all measurements were being taken from the same leaf surface. Individual morphospecies have vein densities that can vary by a factor of two or more, such as 1.5 to 3 veins/mm in *C. angulostriatus* or 1.6 to 3.6 veins/mm in *C. principalis* (Crookall, 1970). Such species ranges that lump all known localities and may span different environments and time periods are not likely to be relevant to the range of individual plants, but within-locality ranges, such as 1.6 to 2.6, and 2.0 to 3.4 veins/mm for *C. principalis*, can still be proportionally larger than the differentiation between canopy bottom and top seen here in *Ginkgo* and are suggestive of large trees. Conversely, the narrow ranges seen in other *Cordaites* species are suggestive of smaller plants as long as a sufficiently large number of leaves were measured in order to ensure that the narrow range is not simply an inadequate sampling of a larger vein density distribution – particularly given that leaves from the top of the canopy are likely to be overrepresented in fossil deposits (Ferguson, 1985). Before any interpretation, however, a similar total number of veins must be verified in leaves at either end of the density range to establish that the range is consistent with differentiation within the canopy of a large tree as opposed to the artificial lumping of distinct species. This example highlights the importance of characteristics that often go unreported: the total number of vein endings in marginally growing leaves or some alternative metric in diffusely growing leaves (such as average number of tertiary veins between secondary veins in a particular area of the leaf), covariance of vein density with leaf size, and variance in vein density and leaf size within individual localities across a known number of samples as opposed to species averages pooled across localities.

Vein density between top and bottom leaves differs by just 8% and 30% in the smaller and larger *Ginkgo* individuals, but up to 60% in the proximal areas of *Quercus* leaves. Those proportional differences are likely to reflect the relative sizes of the trees and their canopy depths with bottom and top measurements being at 2 m and 6 m height in the smaller *Ginkgo*, 2 m and 10 m height in the larger *Ginkgo*, and 5–7 m and 15–18 m height in *Quercus*, although that range of effects may also reflect differences intrinsic to the plant lineages. Conceivably, the same amount of differentiation as across 10 m of canopy height could occur over 5 m of canopy height if stem hydraulic resistance was doubled, while a change in plant water stress might be expected to shift values for the canopy top and bottom together without altering the overall extent of

differentiation. Thus, continuing the earlier example, the large differences in vein density within individual localities for *Cordaites* species suggest large trees indeed, but the exact heights may not be quantifiable. However, *Ginkgo* and *Quercus* measurements involve a total of three trees and the possibility of the proportional range in density informing the size of the tree canopy cannot be assessed without sampling of more species, a larger number of discrete heights within the canopy, and more individuals of different stature within species.

Ginkgo was chosen for this study because it is the only tree widely available in North America involving marginal leaf growth and the availability of other living exemplars is patchy at best. The southern hemisphere conifers *Agathis*, *Nageia*, and some species of *Araucaria* provide examples of proximally concentrated dichotomies in a leaf with a single order of dichotomizing veins that can be used to test suppositions concerning canopy differentiation in those leaf morphologies (Fig. 5A). However, living analogues are absent among seed plants for most leaves with the more complex marginal growth and vein patterns (Fig. 5B) that are characteristic of a variety of extinct groups where the stem is unknown, like *Caytonia*, or where diversity is high and different taxa may be occupying different niches within a locality, like *Glossopteris*. Many of these morphologies are available among living ferns (Boyce, 2005), and subjecting different individual plants to varying degrees of hydraulic stress after leaf initiation may mimic the morphological variance of the top and bottom of a tree canopy.

Paleontological inference will always be complicated by the absence of many previously important leaf morphologies from living trees. The possibility of scale leaves and other forms of heteroblasty is another complication not even touched here. The reliability of canopy assessments based on leaf size and venation can be tested with independent evidence from carbon isotopic analysis of leaf fossil organic matter (Fricke *et al.*, 2007). Leaves from the top of a canopy are well documented to be isotopically heavier than leaves from the bottom of the same tree canopy because of reduced isotopic discrimination in leaves that have their stomata closed for a larger proportion of the time due to water stress and that have a lower internal CO_2 partial pressure due to higher irradiance, and because CO_2 closer to the ground is both more abundant and isotopically depleted in comparison to the well-mixed atmosphere due to the recycling of light CO_2 respired from the soil (Farquhar *et al.*, 1982; Ehleringer *et al.*, 1986; Koch, 1998; Secord *et al.*, 2008). Isotopic analysis of a sizable population of each leaf species at each locality is unlikely to become standard paleobotanical practice, but might provide verification when prudent of the more easily applied estimates based on measurement of leaf size, venation density, and total vein number.

Because leaves are the primary point of interaction between plant and atmosphere, the morphology and anatomy preserved in the leaf fossil record have been a valuable source of information concerning past climates (Wolfe, 1993; Wilf, 1997; Wilf *et al.*, 1998) and atmospheric compositions (McElwain

& Chaloner, 1995; Beerling & Woodward, 1997; Kürschner, 1997; McElwain *et al.*, 2005). While following in that tradition, the potential application outlined here emphasizes two final points. First, most paleoclimate work using plant fossils necessarily is restricted to Cenozoic applications because it is based solely on correlations between leaf characteristics and climate parameters in the living world and, thus, is applicable only to the angiosperms on which the correlations are based. Only when the physiological mechanisms underlying plant/environment correlations are understood can these approaches be applied to pre-angiosperm environments (e.g. McElwain & Chaloner, 1995; Beerling & Woodward, 1997; McElwain *et al.*, 2005). Second, the regularities of plant morphological responses to their environment have been used primarily to inform on that external environment, but these regularities may also inform on the biology of the plants themselves. From the perspective of a leaf, its location within the larger parent plant is as much a part of the environment in which it must function as local CO_2 concentrations or temperature.

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