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Review

Tansley review

Causes and consequences of variation in leaf mass per area (LMA): a meta-analysis

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Key words: chemical composition, environment, functional types, growth rate, leaf anatomy, plasticity, trait spectrum.

Summary

Here, we analysed a wide range of literature data on the leaf dry mass per unit area (LMA). In nature, LMA varies more than 100-fold among species. Part of this variation (*c*. 35%) can be ascribed to differences between functional groups, with evergreen species having the highest LMA, but most of the variation is within groups or biomes. When grown in the same controlled environment, leaf succulents and woody evergreen, perennial or slow-growing species have inherently high LMA. Within most of the functional groups studied, high-LMA species show higher leaf tissue densities. However, differences between evergreen and deciduous species result from larger volumes per area (thickness). Response curves constructed from experiments under controlled conditions showed that LMA varied strongly with light, temperature and submergence, moderately with CO_2 concentration and nutrient and water stress, and marginally under most other conditions. Functional groups differed in the plasticity of LMA to these gradients. The physiological regulation is still unclear, but the consequences of variation in LMA and the suite of traits interconnected

with it are strong. This trait complex is an important factor determining the fitness of species in their environment and affects various ecosystem processes.

Abbreviations: DPI, daily photon irradiance; LD, leaf density; LMA, leaf mass per area; LVA, leaf volume per area (~thickness); RGR, relative growth rate; SLA, specific leaf area; TNC, total non-structural carbohydrates; ULR, unit leaf rate; VA, volume per area.

I. Leaf dry mass per unit area (LMA) in perspective

1. Importance for light interception and plant growth

A leaf is primarily a photosynthetic organ and as such a large number of chloroplasts are contained in a layer of mesophyll. The mesophyll is protected by an epidermis, which also can provide support for the leaf, and is intermingled with a vascular transport system. Fibres may add rigidity to a leaf and deter herbivores that are after the protein-rich mesophyll. Although this principle is quite general, there is amazing variation among the 250 000 higher plant species in how a leaf is composed from these building blocks. Consequently, there is large variation in the amount of carbon and nutrients that is invested in a certain area of light-intercepting foliage. The ratio between leaf dry mass and leaf area ('Leaf Mass per Area', LMA in g m^{-2}) can be understood as the leaf-level cost of light interception (Gutschick & Wiegel, 1988), be it that respiratory costs for construction and maintenance are not included. The LMA is a key trait in plant growth (Lambers & Poorter, 1992) and an important indicator of plant strategies (Grime, 2001; Westoby *et al.*, 2002), which has been used widely in plant ecology, agronomy and forestry, but less so in plant physiology. This review aims for a quantitative framework that structures the wealth of data available on LMA and identifies the gaps in our knowledge.

The inverse of LMA, the ratio of leaf area to leaf mass was introduced in the growth analysis literature in the middle of the last century (Coombe, 1960) and is termed SLA (specific leaf area, m^2 kg^{-1}). The SLA and the fraction of plant biomass allocated to leaves (leaf mass fraction, LMF), determine the total amount of leaf area displayed per unit plant biomass and are important parameters determining a plant's relative growth rate (RGR). By definition,

$$\mathrm{RGR} = \mathrm{SLA} \times \mathrm{LMF} \times \mathrm{ULR} \qquad \text{Eqn 1}$$

where ULR ('unit leaf rate') is the rate of increase in plant biomass per unit leaf area, a variable closely related to the daily rate of photosynthesis per unit leaf area (Poorter & van der Werf, 1998).

The merit of SLA is that it scales positively and linearly with RGR and is therefore often used in growth analyses. However, if one wishes to analyse the underlying reasons for variation in this ratio (anatomy, chemical composition), the complication arises that with each additional investment in the leaf, SLA decreases hyperbolically. In such cases, it is more straightforward to use LMA, which scales positive linearly with additional investments in the leaf. Throughout this review we will therefore use the term LMA.

2. Breakdown of LMA

Leaf dry mass per area is itself a composite attribute: for flat-leaved species LMA can be decomposed as the product of leaf density (LD, g DM cm^{-3} leaf) and leaf lamina thickness (μm; Witkowski & Lamont, 1991). Quantifying lamina thickness is less straightforward for species with complex foliage structure, such as needles (Niinemets & Kull, 1995; see Appendix A1 for a short discussion on methodological aspects). We therefore prefer a more general term and use the leaf volume to area ratio (LVA, cm^3 m^{-2}) rather than thickness. This ratio has been used previously in the anatomical literature (Shield, 1950). The LMA then is:

$$\mathrm{LMA} = \mathrm{LVA} \times \mathrm{LD} \qquad \text{Eqn 2}$$

These parameters can be relatively easily measured or estimated (Vile *et al.*, 2005). More insight into the causes of variation in these parameters can be obtained by considering LMA as the sum of the volumes of *i* different leaf tissue types per unit area – plus the volume of air spaces – each multiplied by their specific density (Roderick *et al.*, 1999):

$$\mathrm{LMA} = \sum_i (\mathrm{LVA}_i \times \mathrm{LD}_i) = \sum_i \left(\frac{V_i}{A} \times \mathrm{LD}_i \right) \qquad \text{Eqn 3}$$

In principle, for flat leaves with parallel tissue layers, the thickness of each tissue layer could be used instead of its volume : area ratio. However, for tissue types such as sclerenchyma, which are diffusely distributed throughout the leaf, thickness has no clear physical meaning. This is the second reason that we consider LVA to be a more meaningful term than thickness to characterize a leaf's anatomy.

The analysis according to Eqn 3 can be taken further by determining the number of cells in each tissue type (N_i) and the average size of the cells (S_i) of that tissue:

$$\frac{V_i}{A} = \frac{N_i \times S_i}{A} \qquad \text{Eqn 4}$$

The last factorization links in directly with the approach of the Russian school, where size of mesophyll and epidermal

cells are related to number and volume of chloroplasts (Pyankov *et al.*, 1998; Ronzhina & Pyankov, 2001). In this way, we can fully explore the anatomical traits that underlie differences in LMA, and have a framework that can quantitatively link variation over three levels of integration, from chloroplast volume to cell size and LMA to the RGR.

An alternative way to break down LMA is to consider leaf biomass as a composite of the mass of j different classes of compounds, such as proteins, lignin, total nonstructural carbohydrates (TNC) and minerals (Poorter & Villar, 1997). In this way LMA is decomposed as:

$$\mathrm{LMA} = \sum_{\mathrm{j}} \frac{M_{\mathrm{j}}}{A} \qquad \text{Eqn 5}$$

Equations 3 and 5 are interrelated, as a high volume of, say, sclerenchymatic tissue per unit area also implies a higher lignin content, while a high LVA taken up by mesophyll will also entail a greater protein content.

3. Topics of this review

In this review we first consider variation in LMA across species, as observed in the field for a wide range of plant species belonging to various functional groups and from various habitats (Section II). We then analyse to what extent inherent variation in LMA can explain the observed differences between functional groups (Section III) and decompose inherent variation in LMA in terms of the underlying anatomical traits and chemical composition (Section IV). Third, we consider how plastic LMA is to environmental differences. To this end we construct – in a second meta-analysis – response curves of LMA for a range of environmental factors, based on experiments under controlled conditions (Section V). We subsequently pay attention to variation in LMA in time and space (see Section VI), and the limited information that is known about the physiological and molecular regulation (see Section VII). Finally, we will return to the ecology and discuss the importance of LMA for the performance of plants in the field (Section VIII).

II. LMA in the field

The first reported field measurements for LMA date back at least as far as Hanson (1917). It has now become one of the leaf characteristics that is routinely measured in many field studies. Here we compiled data on 6100 average LMA values for a total of 3800 species (Appendix A2), in an aim to understand the pattern of variation across species and biomes. For that reason, species were categorised into 10 functional groups and 11 habitats. Because most researchers did not aim for a systematic sampling of the vegetation, these data will be an approximation of how functional groups and habitats differ in the average and range of LMA.

1. Functional groups

Plant species growing in the field show a wide variation in LMA, with values ranging from less than 3 g m^{-2} for freshwater *Myriophyllum* species (Gerber & Les, 1994) to > 2000 g m^{-2} for *Agave deserti* (Nobel, 1980). These are extreme values. From the 5th and 95th percentile of the overall distribution of data in the literature we derive that LMA of most terrestrial species in the field lies between 30 g m^{-2} and 330 g m^{-2} (Fig. 1a).

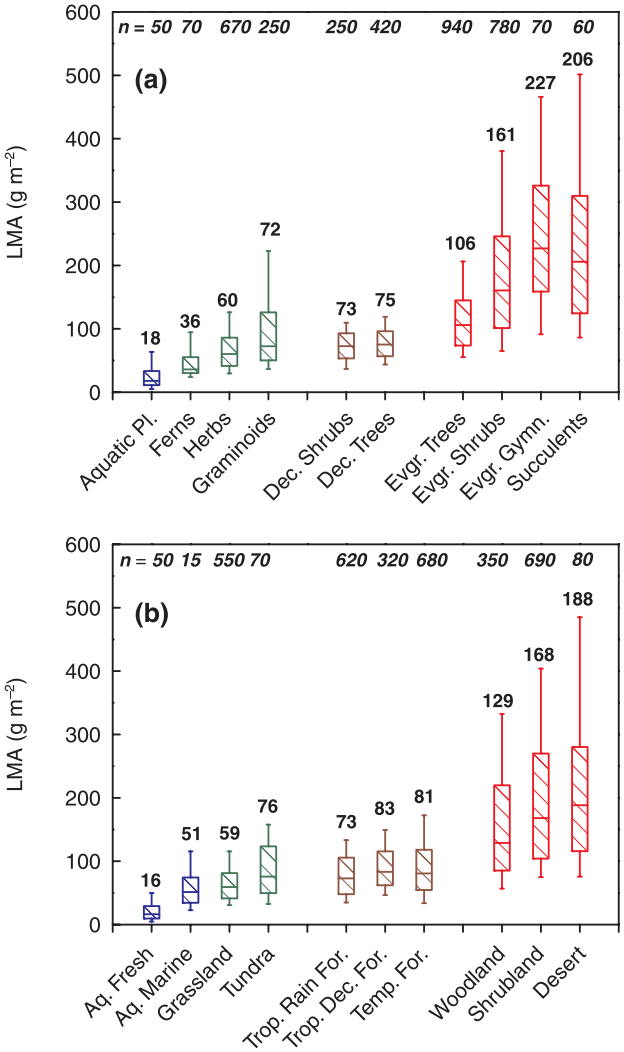

Fig. 1 Distribution of leaf dry mass per unit area (LMA) values in the field, as observed for a wide range of species from (a) different functional groups and (b) different habitats. Box plots characterize this distribution, with the bottom and top part of the box indicating the 25th and 75th percentile, respectively, the two whiskers the 10th and the 90th percentile, respectively, and the horizontal line within the box the median value. The median value is also printed right above the box plots. The total number of species present in each functional group or habitat is indicated at the top of the graphs. More information is given in Appendix A2. *Post-hoc* tests showed that most functional groups were significantly different from at least the groups beyond the adjacent groups in the graphs.

How well does a categorization into 'functional groups' of species explain the observed variation across species? Analysis of variance after ln-transformation indicates that 36% of the variation in LMA can be attributed to the groups shown in Fig. 1a, with a general trend for LMA to increase from aquatic plants < ferns < herbs/grasses < deciduous shrubs and trees < evergreen shrubs and trees and succulents. There were hardly any differences in LMA between annual and perennial herbaceous dicots, but perennial herbaceous graminoids showed, on average, a 60% higher LMA than annual grasses ($P < 0.001$). The extreme high values observed in the perennial graminoids are from species with evergreen leaves, such as *Lygenia* and *Lepidosperma*, which are found in nutrient- and water-limited ecosystems in Australia and the southern part of Africa. The cluster of very high LMA species seen in the shrub species-group are also from these regions (e.g. *Banksia* and *Hakea* species from Australia, and *Protea* from South Africa). Evergreen gymnosperms and succulents show the highest mean LMA. This is true even for fern species such as *Pyrrosia*, which are succulent and may show LMA values over 200 g m^{-2} (Winter *et al.*, 1986), much higher than most other ferns.

2. Habitats

Different environmental conditions may impose different selectional forces on plants, and drive traits to a certain degree of divergence. We therefore considered how LMA of species differs between habitats. Submerged plants from fresh-water habitats have the lowest LMA (Fig. 1b), an attribute that will concur with the low light values found in these environments, a need for facilitation of gas exchange and the reduced requirement for investments in support. Seagrasses have higher LMA than freshwater species, which is likely a consequence of the higher drag by waves in this habitat that probably requires sturdier leaves (Cambridge & Lambers, 1998). Alpine grasslands show somewhat higher LMA than lowland grasslands (64 g m^{-2} vs 50 g m^{-2}), but are pooled for this graph. Tundra species have a larger LMA range than grasslands. Differences between different types of forest are remarkably small, with, as yet, too few data for boreal forest to allow for a good estimate. Habitats that stand out because of a large proportion of high-LMA species are the desert, shrublands (which we define as rather open vegetations with shrubs) and woodlands (which we define as open vegetation with trees), where either drought, nutrient limitation or both strongly hamper growth. These are the ecosystems that support evergreen shrubs, small trees or succulents, which show the highest LMA values.

Analysis of variance showed that the current separation into 11 habitats explains 36% of the total variation. Within each habitat, there is at least a two- to six-fold range in LMA. In multistorey communities, this will partly be the consequence of plants being sampled from different strata of the vegetation, which differ in light availability (Eliáš, 1979; see section V.1). However, also in larger scale comparisons across grasslands or shrublands, where only sun-exposed leaves are sampled, approx. 40–60% of the total variation in LMA is between species within habitats (Körner *et al.*, 1989; Poorter & De Jong, 1999; Wright *et al.*, 2004). The ecological aspects of LMA variation among species will be discussed in Section VIII.

III. Inherent differences

We now analyse to what extent variation in LMA is inherent, and whether functional groups of species can explain part of the observed variation. Given the strong environmental control of LMA (Section V), this requires analysis of plants grown under the same environmental conditions. Thus, we considered multispecies experiments in growth chambers, glasshouses or common gardens where all species were treated similarly and expressed the average LMA of species belonging to a given group of interest, say woody species, relative to the average of a control group, say herbaceous species. A more extended description of this approach and the database is given in Appendix A3.

1. Woody species

Not many experiments have included woody and herbaceous species under standardized conditions. However, almost all of the available evidence shows that tree species (generally seedlings) have a higher LMA than the herbaceous species they were compared with. The median difference is 37%, which is smaller than the 83% differences in the compilation of field data (Fig. 1a). The large increases in LMA of tree species with age, which are discussed in Section VI.2, may explain some of the differences between field and laboratory observations.

Evergreen species generally have higher LMA values than deciduous ones (Sobrado, 1991; Villar & Merino, 2001), both in the field and under controlled conditions (Fig. 2). However, there is generally considerable overlap between the two groups of species (Castro-Díez *et al.*, 2000; Wright *et al.*, 2005). Using phylogenetically independent contrasts, R. Villar (unpublished) showed that a large part of the overlap was caused by phylogeny. That is, evergreen species from a given family generally have a higher LMA than the deciduous species, but differences between families are as large as the differences between evergreen and deciduous species.

2. Herbaceous species

In growth chamber experiments, large-scale comparisons among herbaceous or among woody plant species have recurrently found that slow-growing species have a much higher LMA than fast-growing species (reviewed in Poorter & van der Werf, 1998; Veneklaas & Poorter, 1998), and indicate that variation in LMA is much stronger and better correlated with RGR than variation in ULR (cf. Eqn 1). However, results are

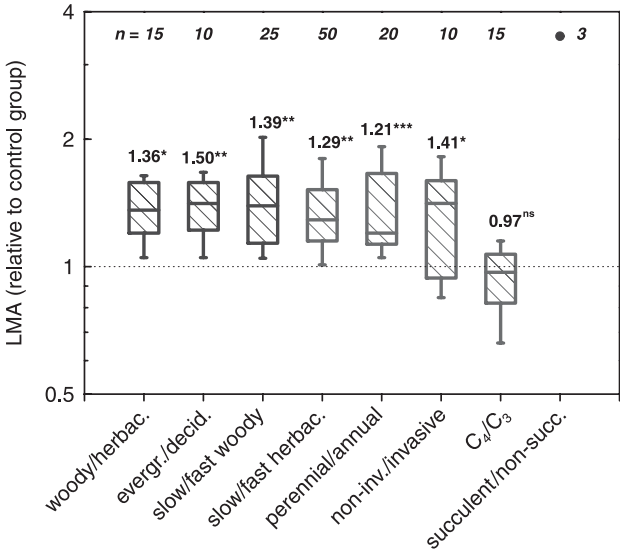

Fig. 2 Inherent differences in leaf dry mass per unit area (LMA) between different functional groups of species. The box plots give the distribution of ratios, where each ratio represents an experiment with the average value of group A (for example woody species in the first contrast) relative to the average value of group B (herbaceous species in the first contrast). Because of restricted information on the comparison of succulents with nonsucculents, we only give an average value. The number of experiments on which the various contrasts is based is indicated at the top of the graph. More information on the box plots can be found in the legend of Fig. 1. Information on the methods may be found in Appendix A3. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

not equivocal, as some other reports show ULR to be much better correlated with RGR (Shipley, 2002). Light intensity may play a role in shifting the balance between the importance of the different growth parameters, with variation in LMA becoming less important in explaining variation in RGR when daily irradiance increases (Poorter, 1999; Shipley, 2006). However, other studies show LMA to be equally important in explaining variation in RGR at low and at high light intensities (for review see Poorter & van der Werf, 1998). Although the debate about the relative importance of both growth parameters is not settled, it is nonetheless clear that in almost all experiments with herbaceous or with woody plants, the species with a slower RGR do have a higher LMA (Fig. 2).

Perennial species generally show higher LMAs than annual species. Higher LMAs for perennials have also been found in co-occurring grasses in the field (Garnier *et al.*, 1997; Li *et al.*, 2005). The difference partly coincides with a difference in RGR, and as such links with the differences between slow-growing and fast-growing species (Garnier *et al.*, 1992).

Although more an ecological than an inherent characteristic, invading species grown under controlled conditions have been reported more often than not to have a lower LMA than the native species of the habitat being invaded (Leishman *et al.*, 2007). Also in this case, the differences concur partly with those of the fast- and slow-growing syndrome. However, it is clearly not one single factor that makes a plant species an invader, and overall, there is wider variation in responses than for other comparisons.

3. Other comparisons

There are a number of small-scale comparisons between species with C_3 and C_4 pathways under controlled or at least similar conditions, in which the C_4 species have been reported to have a lower LMA than the C_3 species. However, in the few larger-scale experiments (DaMatta *et al.*, 2001; Reich *et al.*, 2003) the differences were less clear. Succulent plants, which include many CAM species, have a considerable higher LMA than nonsucculent C_3 species (Fig. 2), which is in agreement with the field data (Fig. 1a). It is unclear to what extent CAM metabolism may affect LMA independently from succulence, although it has been observed in the genus of succulent *Clusia* that CAM species had higher LMA than C_3 species (V. Barrera, unpublished).

Variation in LMA at a given level of environmental resources is also present among genotypes of a given species. In most cases the variation is 10–50% (Rebetzke *et al.*, 2004; van Rijn *et al.*, 2005), but alfalfa (*Medicago sativa*) genotypes vary more than a factor of two in LMA (Pearce *et al.*, 1969). Intraspecific variation in LMA allows estimates of broad-sense heritabilities. Results depend on species and genotypes considered, but estimated heritability may be up to 60–90% (Rebetzke *et al.*, 2004; Songsri *et al.*, 2008).

4. Laboratory versus field

Part of the species comparisons under controlled conditions are inspired by ecological questions aimed at determining the extent to which species traits are able to explain the distribution of species in the field. Given the potentially large effects of the environment, the question arises whether species-specific variation in the laboratory, generally measured for young plants, remains present under field conditions. Species in the field generally have much higher LMA, which will be caused by the higher daily photon irradiance (DPI) they experience in conjunction with lower temperatures (Section V; Garnier & Freijsen, 1994). Indeed, differences between laboratory and field were largest for tree seedlings grown at very low DPI compared with adult trees in the field (Cornelissen *et al.*, 2003a), suggesting that use of higher light intensities in controlled experiments is required to realistically scale from the laboratory to the field. Ontogeny may also play a role in explaining the absolute difference, especially in the case of trees (Section VI). Nonetheless, in most comparisons to date a positive correlation holds between species ranking in the laboratory and the field (Poorter & De Jong, 1999; Cavender-Bares *et al.*, 2004; Mokany & Ash, 2008), with an r^2 between 0.5 and 0.9.

Functional group	growth	LD (g cm^{-3})			LVA (cm^3 m^{-2})		
		P10	P50	P90	P10	P50	P90
Herbaceous	Controlled (n = 290)	0.08	0.18	0.31	125	215	380
	Field (n = 330)	0.09	0.19	0.36	130	275	555
Woody deciduous	Controlled (n = 90)	0.14	0.28	0.53	100	140	210
	Field (n = 460)	0.25	0.37	0.51	95	160	235
Woody evergreen	Controlled (n = 160)	0.16	0.28	0.53	130	200	335
	Field (n = 860)	0.27	0.42	0.58	190	395	855
CAM	Garden & Field (n = 50)	0.05	0.14	0.28	720	1710	4200

Data from plants grown under controlled conditions come from the data compiled in section III. Data from woody plants in the field are from Niinemets (1999). P10, P50 and P90 indicate the 10th, the 50th and 90th percentile.

Table 1 Estimate of the range in leaf density (LD) and leaf volume per area (LVA) for plants from different functional groups, either grown under controlled conditions (growth room, glasshouse) or in the field

IV. Relation with anatomy and chemical composition

Having analysed the overall differences in LMA between species, the question now arises how interspecific variation in LMA can be explained by variation at the anatomical and chemical level.

1. LVA versus LD

From the data we compiled here for laboratory- and field-grown plants (Appendix A2 and A5), we infer that in most C_3 and C_4 species LVA will lie between 100 cm^3 m^{-2} and 700 cm^3 m^{-2} (equivalent to μm), and dry mass density between 0.1 g cm^{-3} and 0.6 g cm^{-3}, with generally higher densities in field-grown plants (Table 1). Here we discuss a few larger-scale experiments that focused on LMA comparisons across species, and the relationship of LVA and LD with LMA. These experiments comprised analyses of grasses (Garnier & Laurent, 1994; van Arendonk & Poorter, 1994), herbaceous dicots (H. Poorter, unpublished) and deciduous as well as evergreen woody species (Castro-Díez *et al.*, 2000; R. Villar, unpublished). Within each group of species there was a weak relationship between LMA and LVA, but in none of the cases was this relationship significant (Fig. 3a). By contrast, strongly positive correlations were found with LD in all experiments but one (Fig. 3b). Considered over all species and using a scaling-slope analysis, as described in Appendix A4, LD explained 80% and LVA 20% of the differences in LMA. The LVA differences are vital, though, to explain the overall difference in LMA between deciduous and evergreen species ($P < 0.001$).

Our knowledge about succulent species is limited. Nielsen *et al.* (1997) reported that LVA was > 10 times higher than in nonsucculent species; this higher value was at least in part because of the presence of large volumes of water-storing cells. Leaf density was two-fold lower (cf. Table 1), probably because the water-storage cells do not contain much more than water. Therefore, the higher LMA of succulents is mainly caused by their higher LVA. We do not know whether CAM succulents differ from C_3 succulents.

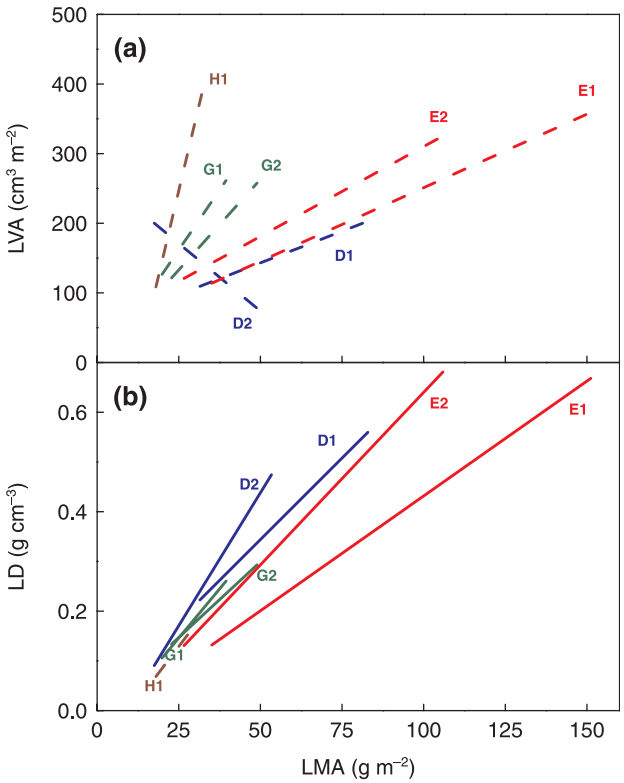

Fig. 3 Relationship between (a) leaf volume per area (LVA) with leaf dry mass per unit area (LMA) and (b) leaf density (LD) with LMA for a number of across-species comparisons. Each functional group from each study is represented by a regression line (following type II regression of Warton *et al.*, 2006). M1, monocots from Garnier & Laurent (1994); M2, monocots from van Arendonk & Poorter (1994); H1, 13 herbaceous dicots (H. Poorter, unpublished); D1 and E1, deciduous and evergreen species from R. Villar (unpublished); D2 and E2, deciduous and evergreen species from Castro-Díez *et al.* (2000). Broken lines indicate nonsignificant relationships and continuous lines a significant relationship ($P < 0.05$).

2. Anatomy

One may ask which tissues will explain the variation in LVA between deciduous and evergreen species, mentioned earlier.

In most anatomical literature, results of leaf cross-section analysis have been presented as volumetric fractions of different tissues. However, in order to break down LMA into its anatomical building blocks, one needs to calculate the volume of each tissue per unit leaf area (see Section I.2). Analysis of the deciduous–evergreen contrast showed that evergreen species had considerably greater mesophyll tissue volume per unit leaf area (R. Villar, unpublished), which is intriguing because these species did not have higher rates of photosynthesis. Evergreens do have thicker cell walls (Terashima *et al.*, 2006) and possibly they compensate their lower cell wall conductance for CO_2 with a larger volume of mesophyll. The larger volume of mesophyll resulted predominantly from evergreens having larger cells, and far less because of an increased number of cells (Castro-Díez *et al.*, 2000; R. Villar, unpublished). Within each group (deciduous or evergreen), high-LMA species had a similar total LVA compared with low-LMA species, but a relatively lower volume of epidermal tissue. Leaves with a higher volume of epidermis had a lower LD. Within the grasses, high-LMA species showed a higher volume of sclerenchymatic tissue and vascular bundles.

A low leaf density could be caused by the presence of a large volume of air spaces. This enhances conductivity within the leaf, which may facilitate photosynthesis (Niinemets, 1999). A high density can be caused by a large fraction of mesophyll, or a high proportion of lignified tissue, that may be important for leaf toughness, and thereby leaf and plant survival (Alvarez-Clare & Kitajima, 2007). To make our investigations in Eqn 3 complete, one not only needs to know the volumes taken up by the various tissues, but also their density. Unfortunately, we have little insight into the density of different tissues, let alone variation between species. Determination of densities of individual leaf fractions is not straightforward, as it is difficult to separate the various tissues from each other. Winter *et al.* (1993) reported that the volume of epidermal cells in barley consisted of 99% vacuole. In mesophyll cells, vacuoles formed 73% of the volume, with chloroplasts and mitochondria occupying 20%. As these plastids are much denser than the vacuole, one could expect mesophyll to have a higher density than the epidermis. Alternatively, one could reason that the density of epidermal cells is higher, as their cell walls are thicker (U. Niinemets, unpublished). A few studies have determined simultaneously the volume per area (VA) of the different tissues as well as LD and LMA (Van Arendonk & Poorter, 1994; Castro-Díez *et al.*, 2000; R. Villar & H. Poorter, unpublished). Using a multiple regression, we regressed the VA of epidermis, mesophyll and vascular tissue against LMA. In principle, the regression coefficients should then give an estimate of the average density of these tissues over all species. Averaged over all groups of species epidermal tissue was shown to be low in density, mesophyll densities were closest to that of the overall leaf and vascular tissue plus sclerenchyma showed the highest values (Table 2).

Table 2 Estimates of the density of different leaf tissues: epidermis (including the cuticle), mesophyll (without air spaces) and vascular tissue, derived from multiple regression analysis

Tissue	Density (g cm^{-3})
Epidermis	0.08
Mesophyll	0.31
Vascular tissue	1.40

Data are the average values from the experiments listed in the legend of Fig. 3.

These are rather rough estimates, owing partly to the fact that these cross-sections only represent part of the leaf and partly to the fact that the densities of the different tissues vary with species. They are, nonetheless, in accord with the observation that the area around the main vein has a higher LMA value than the rest of the leaf (Section V.1). By publishing these first approximations, we hope to stimulate the scientific community to come up with experimentally derived estimates.

3. Chemical composition

The dry mass of a leaf consists of a wide range of constituents that can be grouped into eight different classes of compounds: minerals, organic acids, total nonstructural carbohydrates (TNC; starch, soluble sugars, fructans), total structural carbohydrates (TSC), soluble phenolics, protein, lignin and lipids. In general, these compounds comprise *c.* 90–95% of the plant's biomass (Poorter & Villar, 1997). When LMA doubles because all leaf components double, then the concentration of these compounds per unit dry mass would remain constant, whereas the content per unit leaf area would be simply twice as high for all constituents. However, this is seldom the case. For 24 herbaceous species grown in a growth chamber, we regressed the content per unit area of the eight classes of compounds against LMA, which varied twofold between the most extreme species (Poorter & Bergkotte, 1992; Van Arendonk & Poorter, 1994). All classes of compounds (shown stacked on each other in Fig. 4a to visualize the total amount of biomass per unit leaf area) increased in content with increasing LMA of the species. However, compounds such as minerals and organic acids only increased marginally, whereas constituents such as TSC and lignin increased more than twofold. This implies that the high-LMA species have higher concentrations of these compounds than low-LMA species. For this experiment, as well as for adult tree leaves (Poorter & Bongers, 2006) and deciduous and evergreen seedlings (Villar *et al.*, 2006) we calculated the change in each compound relative to the change in LMA (Fig. 4b). For organic acids, minerals and protein, this ratio was consistently < 1, indicating that high-LMA leaves had relatively lower concentrations of these compounds.

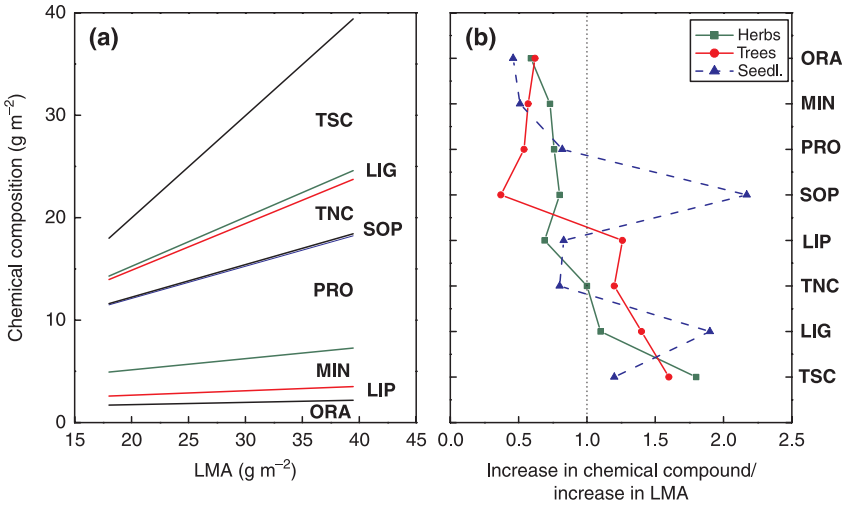

Fig. 4 (a) Leaf chemical composition of low-leaf dry mass per unit area (LMA) and high-LMA leaves in a comparison of 24 herbaceous species, described in Poorter & Bergkotte (1992). For each species the content per unit leaf area was calculated for all eight classes of compounds measured. A regression line was calculated and subsequently the estimated value for a low-LMA and a high-LMA leaf calculated. Following Eqn 5 (see text), these values were stacked on each other to add up to the full biomass of the leaf. (b) Subsequently, the ratio between the estimates of the content of each constituent of the low-LMA and the high-LMA species was divided by the ratio of the high-LMA and low-LMA value for three experiments: the 24 species described above, 10 adult temperate tree species from Poorter *et al*. (2006) and 16 deciduous and evergreen saplings (Villar *et al*., 2006). A value higher than 1 indicates that the concentration of that compound is higher in species with high-LMA leaves. ORA, organic acids; MIN, minerals; PRO, protein; SOP, soluble phenolics; LIP, lipids; TNC, total nonstructural carbohydrates; LIG, lignin; TSC, total structural carbohydrates.

Lignin and TSC had ratios consistently > 1, which implies that high-LMA leaves always have higher concentrations of these compounds than low-LMA leaves. Soluble phenolics, which we would expect to be a class of compounds that was relatively more represented in slow-growing high-LMA leaves was indeed higher in the (evergreen) seedlings, but not in the herbaceous plants or adult trees.

These data agree well with the overall picture of high-LMA leaves having lower concentrations of cytoplasmic compounds and higher concentrations of cell wall compounds than leaves of low-LMA species (Mediavilla *et al*., 2008). This can be explained by a higher fraction of sclerenchymatic tissue (van Arendonk & Poorter, 1997), rather than by smaller cell sizes in high-LMA species (Castro-Díez *et al*., 2000; R. Villar, unpublished).

V. Environmental effects

Inherent variation (Section III) explains part of the LMA variation in the field, but environmental conditions also have a major impact. We have a fairly good idea of how LMA responds qualitatively to some environmental factors (light, CO_2), but less knowledge of how LMA responds to others (waterlogging, salinity), let alone how the magnitude of response differs among environmental factors and why. We therefore compiled a third body of literature data for plants that were grown under controlled conditions (glasshouses, growth chambers) with at least two different levels of the environmental factor of interest. This was done for 12 environmental variables in total. A detailed explanation of the approach and database description is given in Appendix A5. In short, all average LMA values reported for a given species in a given experiment were scaled to the LMA value obtained at a preselected reference level. This reference level was independently defined for each environmental factor and is indicated by arrows in each panel of Fig. 5. In this way, we obtained 30–900 observations per environmental factor, depending on the literature available. From this range of data we constructed average response curves of LMA for each environmental factor and considered possible differences in plasticity between functional groups of species.

1. Radiation

Generally, evidence shows that it is not instantaneous peak irradiance, but rather the total photon irradiance integrated over the day (DPI) that determines the LMA of a plant (Chabot *et al*., 1979; Niinemets *et al*., 2004). That is, a short period of high light intensity has the same effect on LMA as a period twice as long with half the irradiance. For all experiments we therefore estimated the average DPI during the growth of the plants. An increase in DPI causes a strong increase in LMA in almost all experiments (Fig. 5a). The response is nonlinear ($P < 0.001$; Table 3; see Appendix A5 for more information): it is particularly strong at low light, and increases more slowly above a DPI of 20 mol m^{-2} d^{-1}.

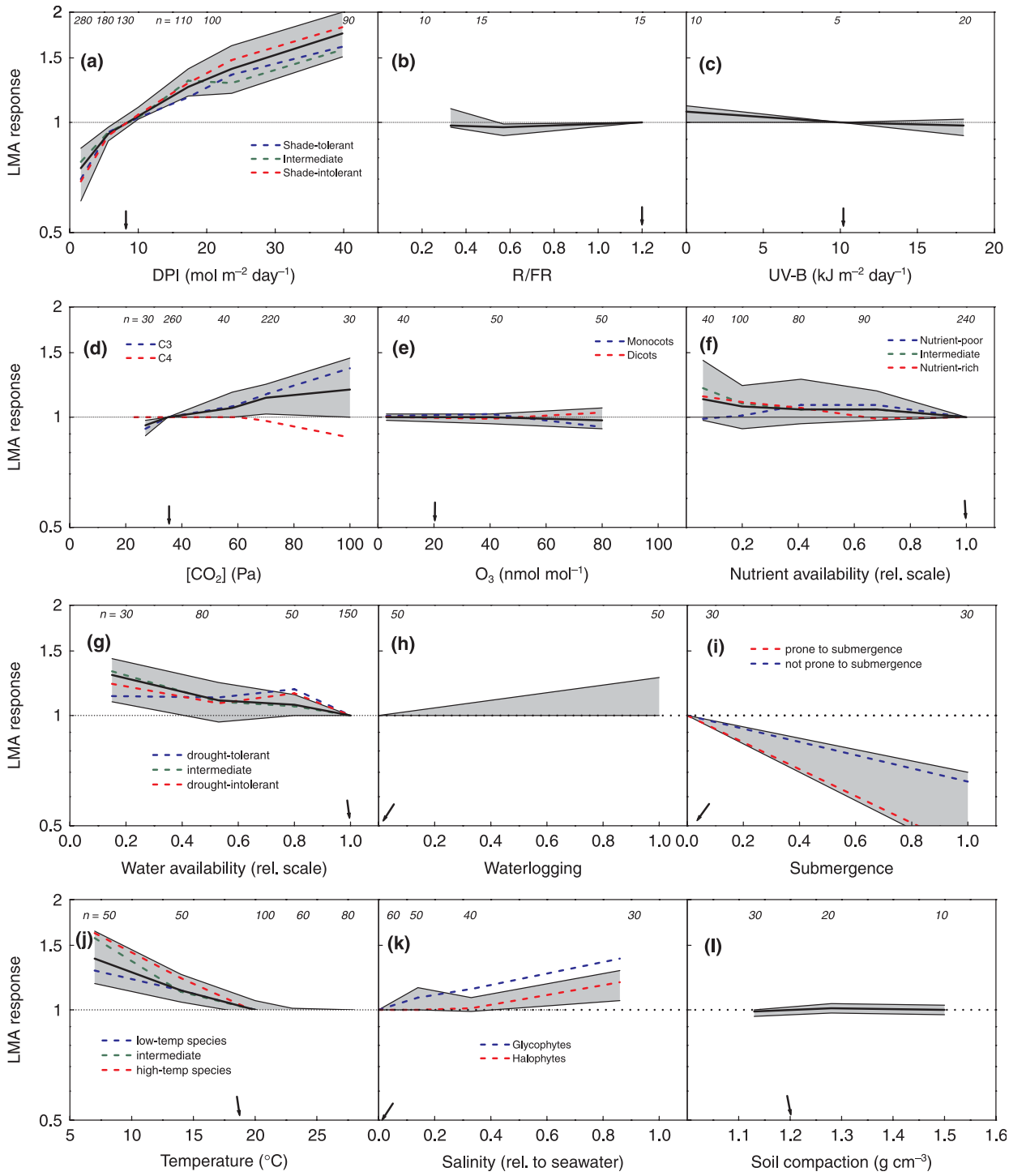

Fig. 5 Characterization of the response in leaf dry mass per unit area (LMA) of plants grown in a range of environmental conditions: (a) daily photon irradiance (DPI); (b) red : far-red ratio (R/FR); (c) UV-B; (d) CO_2 concentration; (e) ozone; (f) nutrient availability; (g) water availability (drought stress); (h) waterlogging; (i) submergence; (j) temperature; (k) salinity; (l) soil compaction. Data are a compilation of the literature. For each environmental factor a reference condition was chosen (indicated by arrows), and all data from different species were normalized to that condition. For more information see Appendix A5. The shaded area indicates the interquartile range (between the 25th and the 75th percentile) of the observed ratios in that part of the response curve. The bold continuous line within the shaded area indicates the median value. Dashed lines indicate the median value for a specific subgroup of species and are labelled in the graph. Generally, species were classified with respect to their original habitat to grow at high, intermediate or low levels of the environmental factor studied. The total number of observations present in each part of the response curve is indicated at the top of each graph. The *y*-axis is logarithmic to correct for the fact that ratios are logarithmic by nature.

Table 3 Response curves of scaled leaf dry mass per unit area (LMA) values as affected by a range of environmental factors

Environmental factor	Type of species	Constant × 10^3	Linear component × 10^3	Quadratic component × 10^3	df	r^2	Response ratio
Irradiance	All	–375	43.6	–0.458	890	0.72***	**2.56**
	Shade	–363	42.5	–0.576	150	0.63***	**2.04**
	Sun	–408	47.9	–0.510	420	0.74***	**2.78**
Red : far red ratio	All				40	0.00^{ns}	1.00
UV-B	All	30.2	–2.96		30	0.11^{+}	1.05
CO_2	All	–182	5.85	–0.020	590	0.26***	1.27
	C3	–251	8.00	–0.029	470	0.40***	1.37
	C4	–65.1	3.38	–0.039	120	0.14***	1.12
O_3	All	–24.5	1.73	–0.023	140	0.09*	1.26
	Monocots	–27.0	3.65	–0.051	30	0.46**	1.13
	Dicots				100	0.00^{ns}	1.00
Nutrients	All	128	–116		550	0.05***	1.12
Water availability	All	239	–231		310	0.18***	1.22
Waterlogging	All	0	121		90	0.19***	1.13
Submergence	All	0	–628		70	0.40***	**1.87**
	Subm.-prone	0	–756		40	0.68***	**2.13**
	Other	0	–466		30	0.66***	**1.58**
Temperature	All	694	–49.9	0.719	320	0.40***	**1.68**
	Boreal	633	–51.3	0.956	160	0.33***	1.45
	Tropical	1273	–98.6	1.563	70	0.56***	**2.51**
Salinity	All	26.9	141		180	0.10***	1.13
	Glycophytes	25.4	423		80	0.23***	1.44
	Halophytes	–6.50	152		100	0.15***	1.14
Compaction	All	–153	114		70	0.07*	1.04

Scaling was done relative to a predefined value of each environmental factor, as explained in Appendix A5. For each factor is indicated whether there is a linear relationship (only linear component significant), a nonlinear relationship (also the quadratic component significant), or no relationship at all. To allow for the logarithmic nature of ratios, all scaled LMA values were ln-transformed before the linear regression analysis. If there was a significant difference in plasticity between predefined subgroups of species, regression estimates for each subgroup are given as well. For each relationship the degrees of freedom (df) and the r^2 are indicated, and the response ratio, which is defined as the ratio between the highest and the lowest fitted LMA value along the regression line. This indicates how plastic LMA is for that parameter within the ranges considered here. Response ratios larger than 1.5 are printed in bold type. In the case of submergence, submergence-prone species were taken as those terrestrial plants growing in wet habitats or close to the river. For clarity, all estimates of regression constants were multiplied by 1000. +, $0.05 < P < 0.10$; *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

The response is ecologically adaptive: by increasing the area of a given unit of leaf biomass, the interception of light is increased under low-light conditions, while more photosynthetic biomass per unit leaf area enhances photosynthetic capacity in high light. Interestingly, species that were a priori classified by us as being shade-intolerant showed a higher plasticity in their response to light than shade-tolerant species (Table 3). This corroborates the result of Valladares *et al.* (2000), who found the same difference in plasticity for LMA between 16 *Psychotria* species that were characteristic of gaps and understory. A scaling-slope analysis of LMA with its two underlying components (see Appendix A4 for an explanation) showed that LD and LVA were equally important in explaining the increased LMA with high light intensity (Fig. 6). These differences relate strongly to the leaf morphology of the plants. Generally, it is the VA of (palisade) parenchyma that increases with light, whereas the VA of the epidermis remains remarkably constant (Hanson, 1917; Onoda *et al.*, 2008). Enhanced density at higher irradiance is partly associated with a greater fraction of palisade parenchyma with more tightly packed cells, and partly with a higher TNC concentration (Niinemets *et al.*, 1998).

Although most experiments have studied the effect of shade by changing the total amount of light, plants grown in the lower part of a vegetation will not only experience a lower DPI, but also a change in light quality because of a decrease in the red to far-red (R : FR) ratio. Only a few experiments have considered the effect of light quality separately from light quantity. Both increases and decreases have been reported, but on average, the R : FR ratio seems to alter LMA to a surprisingly minor degree (Fig. 5b).

In the field, a higher irradiance will coincide with an increase in the amount of UV-B, which is supposed to be harmful for the plant. Plants can protect themselves by the accumulation of UV-screening compounds, such as soluble phenolics, in the vacuoles. We therefore would have expected higher LMA values at higher UV levels, but the values were marginally lower (Fig. 5c). However, this claim awaits further confirmation, as it is based on only a small amount of data.

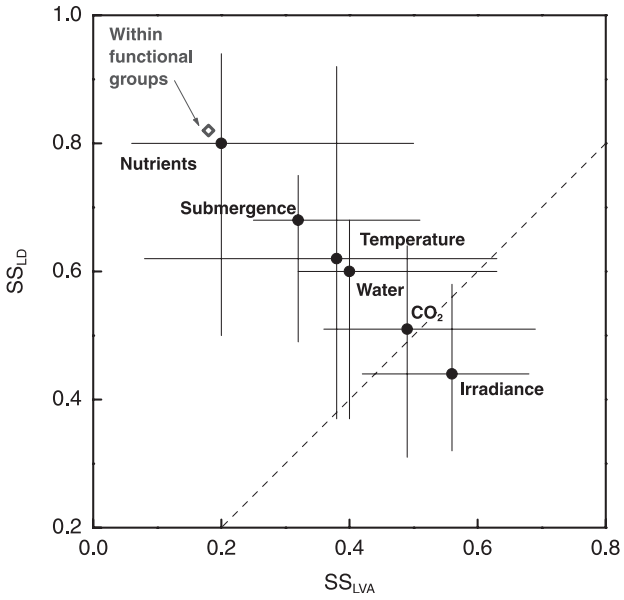

Fig. 6 Median scaling slopes (SS) of leaf density (LD) and leaf volume per area (LVA) indicating how a relative change in leaf dry mass per unit area (LMA) accompanies a relative change in these factors. All slopes are positive, indicating that LMA is positively affected by LD and LVA. The lines indicate the interquartile range for each factor. An explanation of the scaling slopes is given in Appendix A4. Data pertain to LMA variation caused by different environmental variables (light, CO_2, water, temperature, submergence, nutrients), compiled in Appendix A5. The median scaling slope across a number of species comparisons within grasses, within herbaceous dicots, and within deciduous and evergreen species is also given ('within functional groups', diamond). References for these data are given in the legend of Fig. 3.

2. Atmospheric composition

Concentrations of CO_2 higher than ambient generally result in increased LMA, whereas the scarce observations made at preindustrial CO_2 levels show a decreased LMA (Fig. 5d). Over the range of concentrations considered, the response is somewhat saturating ($P < 0.001$; Table 3), also when only C_3 species are considered. The increase in LMA for C_3 species coincides with both an increase in LVA and in LD (Fig. 6). Contrary to the effect of light, no increase in the number of mesophyll layers is observed. Nevertheless, thickness does increase (Sims *et al.*, 1998a), reflecting larger mesophyll cells (Radoglou & Jarvis, 1990). The positive effects of CO_2 on leaf density are strongly connected to the accumulation of starch, with concentrations that sometimes surpass 40% of the leaf biomass. The response of LMA does not appear to be very adaptive: on a TNC-free basis, the effect of CO_2 on LMA is generally small or absent (Allen *et al.*, 1998; Roumet *et al.*, 1999).

There is an intriguing contrast between C_3 and C_4 species in their response, with C_4 species even showing a decrease in LMA at the highest CO_2 concentration. This is based on only seven observations, but the average deviates significantly from unity ($P < 0.001$). As far as we know, the effect of high [CO_2] on the leaf anatomy of C_4 species has not been investigated. Generally, no accumulation of TNC is found, even though the rate of photosynthesis is stimulated (Wand *et al.*, 1999). It is possible that the indirect effect of an improved water status because of reduced stomatal conductance, in conjunction with a higher leaf temperature owing to reduced transpiration can explain the decrease in LMA in C_4 species (cf. Fig. 5g,j; Ghannoum *et al.*, 2000).

Ozone increases leaf starch concentrations (Schmitt *et al.*, 1999), thereby altering leaf density but not leaf thickness. The net effect of ozone on LMA is small, with a tendency towards increased LMA at high ozone levels in dicots and towards decreased LMA in monocots (Fig. 5e). The number of observations is limited, especially for monocots, but the difference between groups is significant ($P < 0.05$). Interestingly, ozone sensitivity is likely to be higher for species with an inherently low LMA, possibly because this trait is connected with faster growth and a greater stomatal conductance that results in greater ozone uptake at a given ambient ozone concentration (Bassin *et al.*, 2007).

3. Nutrients

Lack of nutrients, most notably N, will decrease growth more than photosynthesis, leading to accumulation of TNC. This in itself may increase LMA, as in the case of elevated CO_2. However, at low nutrient availabilities, leaf protein concentration will be much lower, sometimes decreasing quantitatively even more strongly than TNC increases (Fig. 5b in Poorter & Villar, 1997). The net consequence can be that the sum of both compounds, which together form 30–60% of the leaf biomass, remains constant. The effect of low nutrient availability on leaf anatomy is much smaller than the effect of light (Shields, 1950) and, consequently, the overall effect of nutrients on LMA is moderate, and (on average) only shows when plants are severely limited in growth (Fig. 5f). The impact of nutrient limitation on whole-plant leaf area is much stronger and, consequently, reduced leaf area may result in higher irradiances in lower leaves, thereby increasing total-plant LMA as well. The variation in response is wide across experiments and cannot be explained by plasticity differences between species from nutrient-poor and nutrient-rich habitats (Table 3; Fig. 5f). As far as changes in LMA are driven by nutrient stress, they are due more to alteration in LD than in LVA (Fig. 6). Of all environmental factors considered, nutrient stress is the one that affects LMA proportionally most through a change in density.

4. Water

The degree to which plants experience water limitation depends on species-specific alterations in stomatal conductance after

withholding water, which complicates assessment of drought effects on foliage structure. Furthermore, drought treatments are diverse and range from suddenly withholding water after a life-long period of optimal supply, repeated dry periods with intermediate watering or more or less constant low water conditions throughout the experiment. The first approach does not yield data that are relevant to answer our questions as leaf structure and chemistry hardly had time to acclimatize. The last method approaches steady-state conditions and is most relevant for the current analysis, but is only rarely used. Therefore, we also included the repeated dry–wet cycle experiments in the analysis. The experimental evidence points towards an increase in LMA with decreasing water availability (Fig. 5g). Starch levels are reported to be variable, either reducing as the result of sugar hydrolysis to decrease leaf osmotic potential or staying constant under drought stress (Chaves *et al.*, 2003; Schurr *et al.*, 2000), and so cannot explain the increase in LMA. Leaves developed under low water availability do have lower expansion rates. The cells are therefore smaller and more tightly packed, with a lower fraction of air spaces (Maximov, 1929; Shields, 1950). The thickness of cell walls is also greater, collectively resulting in a greater LD. By contrast, LVA is reported to decrease in drought-developed leaves owing to limited cell expansion growth (Lambers *et al.*, 2008), although this is not supported by our analysis in Fig. 6. These responses result in more rigid leaves that wilt less easily under dry conditions, which may be advantageous when plants are also competing for light. A smaller transpiring leaf surface at the leaf (LMA) and plant level will reduce the water requirements under dry conditions. We did not find a difference in plasticity for species from wet and dry environments.

Waterlogging is a peculiar condition where only the root system is inundated. The anaerobic environment around the roots hinders uptake of water of most species because of reduced root conductance. As the shoot stays in air, evaporative demand remains constant, with the consequence that the shoot suffers from drought stress. This is likely the reason why the LMA response is rather similar to that of drought (i.e. waterlogging results in smaller leaves with greater LMA; Fig. 5h). Some species are able to escape these problems through internal aeration of the roots by developing aerenchyma in existing roots or new, adventitious roots with extensive aerenchyma (Kozlowski, 1997).

Plants that are entirely submerged face partly different problems compared with those that are waterlogged. Apart from slow diffusion of gases in the roots, there is also only limited ability for gas exchange in the shoot (Voesenek *et al.*, 2006). Without special adaptations for CO_2 and bicarbonate uptake, such as those present in aquatic plants, the rate of photosynthesis strongly decreases. Moreover, light intensity also decreases upon flooding, especially when water turbidity increases the concentration of soil particles and humic substances. From this perspective, one would expect LMA to decrease under flooding and this is indeed what happens (Fig. 5i). Interestingly, continuous wetting of leaves of nonsubmerged *Phaseolus* plants also decreases LMA (Ishibashi & Terashima, 1995). The lower LMA in submerged leaves coincides with a decrease in cell wall and cuticle thickness, which reduce the overall CO_2 liquid-phase diffusion pathway to chloroplasts (Mommer *et al.*, 2005). Apart from structural changes in leaf anatomy, the decrease is also caused by strongly reduced TNC levels, reflecting reduced photosynthesis and increased glycolysis. Amphibious plants and other species characteristic of wetter habitats show a greater decrease in LMA than other species ($P < 0.05$). This is intriguing, as tolerant species have thinner cell walls anyway (Mommer *et al.*, 2007). The fact that submerged leaves change more in LD than in LVA (Fig. 6) may be because, in most experiments, quite some leaves were already present before the submergence started.

5. Temperature

At low temperatures, limited cell expansion leads to a large number of small cells per unit area and, accordingly, more cell wall material per unit leaf volume (greater LD) and more cell layers (greater LVA; Atkin *et al.*, 2006). More cell layers also imply a higher protein content per unit leaf area. In addition, the content of secondary compounds such as proline can increase (Usadel *et al.*, 2008). Consequently, LMA of plants grown at low temperature is higher (Fig. 5j). Ball *et al.* (2002) showed that a higher LVA could reduce the incidence and severity of freezing stress by slowing down the rate of freezing. At higher temperatures, LMA decreases, but not at the same rate, making the response nonlinear ($P < 0.01$). There is a clear difference in the response of boreal and tropical species, with the tropical species changing LMA more for a given change in temperature ($P < 0.05$). The LD contributes more strongly to the change in LMA with temperature than does LVA (Fig. 6).

6. Other factors

Salt stress typically induces physiological drought and the LMA response is qualitatively the same as under low water stress. However, there is an additional problem for most species, which is that they have to isolate the accumulated NaCl in the vacuole. Salinity therefore increases the LVA, mainly owing to an increase in mesophyll cell size or number of layers (Longstreth & Nobel, 1979; Kozlowski, 1997). Little is known about changes in LD, but reduction in intercellular spaces has been observed. The overall effect is a small increase in LMA (Fig. 5k). Halophytes are better than glycophytes in excluding salts, which may be the reason that halophytes increase LMA only at higher salinities and relatively less strongly than halophytes.

Soil compaction increased LMA to some extent, but the changes are generally very modest (Fig. 5j). A similar picture emerges for plants under mechanical stress, where increases

are small (Anten *et al.*, 2006) or absent (Kobayashi *et al.*, 1999). Wind combines mechanical stress with increased transpiration, but here also the effect is small or nonexistent (Retuerto & Woodward, 1982 and references therein).

The bigger picture that emerges from this analysis is that LMA is high in cases where carbon availability is high, either as a result of the faster rate of photosynthesis at high supplies of light and CO_2 or in conditions where the rate of photosynthesis is decreased, but the demand for carbohydrates for growth is hampered even more strongly (low nutrients, low temperature). This may also come about by arresting cell expansion through reduced turgor, as in the case of drought stress. For the environmental variables considered here, the ranges used in the wide variety of experiments analysed span most of what plants are likely to experience in the field. On average, LMA varies strongest with light, followed by temperature and submergence (see the response ratios in Table 3). The effects of CO_2, nutrients and drought are more modest than the first three, and changes are marginal for most of the other factors considered. This agrees well with field observations on naturally-grown *Quercus ilex* trees from a wide range of locations, where LMA was found to change relatively strongly with light and temperature, but less so with nutrients (Ogaya & Peñuelas, 2007). The shift in LMA is generally caused more by changes in LD than LVA, with the most extreme case being nutrient stress (Fig. 6). Irradiance, in fact, is the only factor where LVA is a stronger determinant of LMA (Fig. 3).

VI. Differences in space and time

In the analyses described in Sections III and V, most of the LMA data were determined at the end of the experiment, and calculated as an average for the whole plant. As whole-plant averages are partly driven by within-plant shading as well as by within-plant differences in leaf age, we consider here LMA variation within a plant and with time.

1. Within-plant and within-leaf variation

Shipley (1995) found in a study with 30 herbaceous species that although most of the variation in LMA was caused by species (70%), considerable variation in LMA was present among the leaves of the same individual (26%). Little variability was found between individuals of the same species (4%). Differences within an individual can be large, a spectacular example is that of the 100-m tall *Sequoia* trees, where LMA increases with the height of the canopy from 150 g m^{-2} to 800 g m^{-2} (Koch *et al.*, 2004; Fig. 7a). However, that is an extreme example, and the difference is generally twofold, both for tree species and for herbaceous plants (Fig. 7a,b). Differences can be partly species-specific (Lichtenthaler *et al.*, 2007), but the light-gradient always plays a crucial role in determining the LMA (Anten & Hirose, 1999; Niinemets, 2007). Other factors covary with light in canopies, most notably water pressure deficit (drier air), air temperature and wind speed (Baldocchi *et al.*, 2002). Moreover, for larger trees it may become exceedingly difficult to transport water against the gravity gradient (Niinemets, 1997). This may be another reason for leaves at the top to become smaller and more xeromorphic (Maximov, 1929). A good example is the adult *Sequoia* mentioned earlier. Although leaves at the top show very high LMA values, a seedling that germinated almost at the top of the tree in some detritus and thus experienced exactly the same radiation environment had much larger leaves, with presumably a lower LMA. Koch *et al.* (2007) could show that reduced water availability in taller trees was a prime factor for the differences in LMA.

In herbaceous dicots, foliage develops gradually from bottom to top, and essentially all leaves may have experienced high irradiance during their formation, with the duration of the period of high light intensity depending on the rate of canopy expansion. Nevertheless, the LMA gradient within the canopy of herbaceous species may be as large as that for woody species (Fig. 7b). Apparently, they are plastic enough to acclimatize to the developing light gradient, which may in the end be as strong in herbaceous vegetation as in temperate forests.

The LMA values also differ within a leaf. Niinemets *et al.* (2007) showed that across a range of species the midrib had a substantially higher LMA (sixfold on average) than the rest of the lamina (cf. section IV.3). However, this is species dependent, and for wheat the effect of the main vein on LMA was only marginal (Rawson *et al.*, 1987). For this species and for most graminoids, LMA is higher at the basal than at the distal part of the leaf (Rawson *et al.*, 1987). This has consequences for a vegetation consisting of graminoids without stems: they do not follow the general trend for trees and dicot herbs, and have the highest LMA values down in the canopy, rather than at the top (see the insert in Fig. 7b).

2. Temporal changes

In those deciduous trees where most leaves appear at the beginning of the growing season and stay until autumn, LMA is high right after bud-burst, drops rapidly during leaf expansion, after which there is an increase again (Jurik, 1986), presumably because of a build-up of cell wall material and chloroplasts. After these first 30 d, LMA remains remarkably constant in most trees, until the onset of senescence when LMA decreases again (Fig. 7c). A similar pattern was reported for wheat leaves (Rawson *et al.*, 1987). Older leaf cohorts within evergreen shrubs or trees may have similar or higher LMA values compared with younger leaves (Fig. 7d; Wright *et al.*, 2006). Shading of older foliage by younger leaves makes the separation of age and light effects difficult (Brooks *et al.*, 1994). Age-dependent increases in LMA, when present, are associated with accumulation of carbon-rich chemicals, possibly reflecting thickening and enhanced lignification of cell walls and an overall increase in LD (Niinemets, 1997).

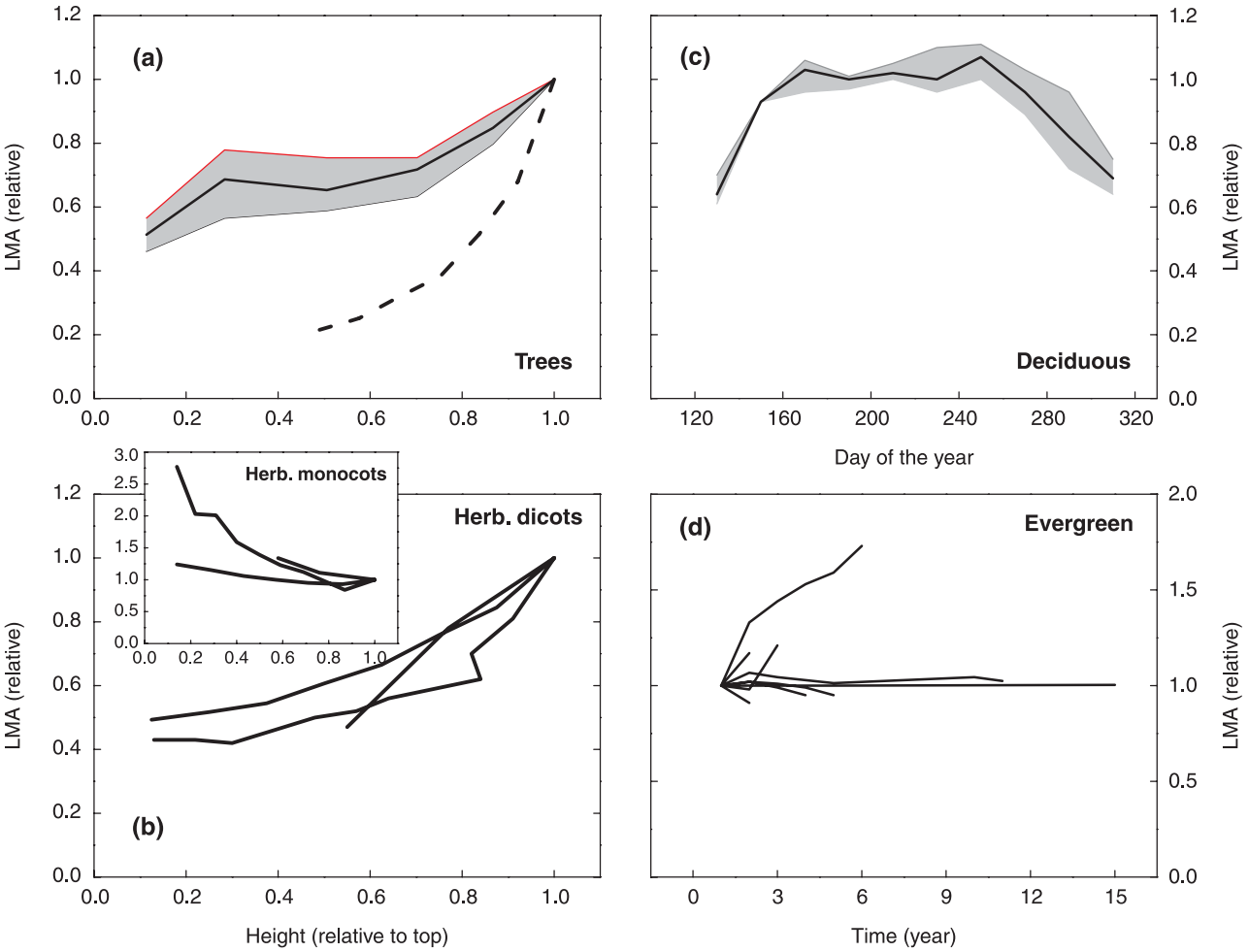

Fig. 7 (a) Distribution of leaf dry mass per unit area (LMA) over the vertical axis in (a) tree canopies; and (b) herbaceous stands, with an insert pertaining to graminoid vegetations. In (a), the shaded area gives the interquartile range (between the 25th and 75th percentile of the distribution) of the distribution of 12 different profiles along woody species. The dark line represents the median. The broken line is from the data of Koch *et al*. (2004) on *Sequoia* and is not included in the profile distribution of the other species. All LMA and height data were normalized against leaves at the top of the tree or vegetation. (c) Changes over time of the LMA of temperate deciduous woody species. All LMA data were normalized against the value at Julian day 160 of the year. All observations are from the Northern Hemisphere. (d) The LMA of different leaf cohorts in evergreen species. All LMA data were normalized against the LMA of fully expanded leaves of the youngest leaf cohort. Data for the various graphs are listed in the Supporting Information, Table S4.

For woody species, leaf longevity is always much shorter than plant longevity, and although an individual leaf will not necessarily increase in LMA with age, older plants almost invariably have leaves with higher LMA (Thomas & Winner, 2002; Niinemets, 2006). The increase can be more than twofold (Veneklaas & Poorter, 1998). Plant size may have a confounding effect here as well, as discussed above. In most herbaceous species, there is also an increase in LMA over time (Poorter & Pothmann, 1992; Villar *et al*., 2005). To what extent these differences are caused by an increase in the LMA of existing leaves, or a higher LMA of later-developing leaves is unclear.

Changes in LMA also occur on a diurnal scale, with lowest values at the end of the night and 10–50% higher values at the end of the day (Sims *et al*., 1998b; Tardieu *et al*., 1999). These fluctuations correlate strongly with the build-up of TNC during the day and breakdown followed by retranslocation of sugars during the night. However, changes in TNC alone can only explain part of the diurnal variation, implying that other components such as amino acids must also contribute to this rhythm (Plhák, 1984; Bertin *et al*., 1999).

3. Plasticity

Leaves that have been grown in a high-light climate and are switched to low light can substantially (30–50%) decrease LMA within a number of days (Sims & Pearcy, 1992; Pons & Pearcy, 1994). In fully developed leaves, this reduction can only occur as the result of breakdown or retranslocation of proteins and other compounds, such as starch and

hemicellulose (Matile, 1974), which decrease LD. Leaves that are still in the expansion phase, can adjust LVA better upon momentary shading than older leaves (Yamashita *et al.*, 2002). In fully developed leaves, inflexible LVA is the primary factor constraining LMA and leaf photosynthetic adjustment to higher irradiance (Oguchi *et al.*, 2005).

VII. Molecular regulation and physiology

Owing to the complexity of the underlying anatomical and chemical traits (Eqns 2–4), LMA can be expected to be a polygenic trait. At least eight quantitative trait loci (QTL) are found in *Arabidopsis* (H. Poorter, unpublished), and three to five QTL in some other species (Yin *et al.*, 1999; Ter Steege *et al.*, 2005). Although the process of leaf expansion is relatively well investigated, an integrated picture of the molecular regulation of LMA is currently lacking. Here we discuss possible key control points in the regulation of LMA and its role in the physiology of the plant.

It is an attractive idea that a well-adapted leaf can be of crucial importance for the functioning of a plant. Although retranslocation of leaf compounds may occur (Section VI.3), there is limited flexibility for a plant to adjust leaf morphology and anatomy after leaf expansion has stopped. Adequate and early sensing is therefore of critical importance in leaf formation. In that respect it may not be surprising that for some tree species leaf anatomy is already determined in the leaf bud and depends on the light conditions that the leaves of the previous cohort experienced (Eschrich *et al.*, 1989; Uemura *et al.*, 2000). Most herbaceous species will have to rely on a regulation mechanism that is dependent on the sensing of previously formed leaves. In an interesting experiment, Yano & Terashima (2001) shaded either the older leaves or the apex of *Chenopodium*. They found that the leaf anatomy of the newly developed leaves was determined by the light status of the older leaves, but that chloroplast differentiation depended on the local environment of the developing leaf. What are the sensing and signalling mechanisms to gauge the environment?

1. Carbohydrate status

Under most conditions where TNC accumulation occurs (high light, high CO_2, low nutrients, low temperature) LMA is increased (Section V). This is also true for tomato (*Lycopersicon esculentum*) plants where the source : sink ratio was increased experimentally by removing a number of fruits (Bertin & Gary, 1998). The result of starch accumulation may be direct: *Arabidopsis thaliana sex1* mutants lack the enzyme to break down starch, and partly as a consequence of high starch contents per leaf area have dense, thick leaves with a higher LMA than wild-type plants (Table 4). In addition to the increase of leaf dry mass as sugars accumulate, it is pertinent to ask whether there is also a possible regulatory link between the increase in LMA and availability of nonstructural carbohydrates. It has been suggested that trehalose-6-phosphate (T6P) is a sensor for the sugar status of the plants. Mutants of *Arabidopsis* and *Nicotiana* that are genetically manipulated to have artificially low T6P levels in their cells have low LMA, whereas genotypes with high T6P levels have leaves with high LMA (Schluepmann *et al.*, 2003; Pellny *et al.*, 2004). However, evidence incongruent with an important role of sugars is the finding that feeding a plant directly with sugars has little effect on LMA (Begna *et al.*, 2001). Araya *et al.* (2008), manipulating the environment of older leaves, could not find a difference in the sugar status of the younger leaves either.

Table 4 Per cent difference in leaf dry mass per unit area (LMA) of *Arabidopsis* mutants relative to their wild type

Mutant	Wild type	% Difference	*P*
Cryptochrome 1 (cry1)	Ler	–42	***
Phytochrome B (phyB)	Ler	–26	***
GA-insensitive (gai)	Ler	+33	**
Starch excess (sex1)	Col-0	+41	***

Plants were grown in pots for 35 d at a daily photon irradiance of 11 mol m^{-2} d^{-1}.

2. Effect of light

Although the R : FR ratio does not affect LMA strongly (Fig. 5b), differences in light quality may still be important. Soybean and cucumber grown under blue-light deficient conditions show lower LMA values (Britz & Sager, 1990; S. Hoogewoning, unpublished). *Arabidopsis* mutants disturbed in blue-light sensing because of a nonfunctional cryptochrome 1 do indeed have pale green leaves with lower LMA than the wild type (Table 4). This is also true for mutants disturbed in phytochrome B. Given that there was so little effect of the R : FR ratio on LMA, this could suggest that phytochrome B also acts as a sensor for light quantity.

3. Effect of hormones

Any sensing of sugars or light has to be followed by a signal transduction pathway to make the sensing effective. Hormones often play a role. There is a relatively strong effect of gibberellins (GA) on LMA: the mutants hampered in GA synthesis or perception show an increased LMA, both in *Arabidopsis* (Tab. 4) as well as in *Lycopersicon* (Nagel *et al.*, 2001), and addition of GA decreases LMA (Dijkstra *et al.*, 1990). The role of other hormones seems to be smaller. Tomato mutants deficient in abscisic acid are unable to close their stomata. They have a high LMA that is associated with their deteriorated water status (Nagel *et al.*, 1994; Fig. 5g). Mutants that lack any ethylene sensing have leaves with a lower LMA (Tholen *et al.*, 2004). Application of cytokinins

increases LMA (Pons *et al.*, 2001). The distribution of cytokinins is also a regulating factor in the retranslocation of proteins and other compounds upon shading of older leaves in the vegetation, and thereby their decreased LMA (Boonman *et al.*, 2007).

4. Relationship with physiology

Within a given species, there is often a tight relationship between photosynthetic capacity and LMA. This is well illustrated by plants grown at different light intensities: plants grown at high-light intensity have a considerably higher photosynthetic capacity per unit leaf area, but also a higher LMA with a higher volume per area of mesophyll (Section V.1). When expressed per unit leaf dry mass, however, photosynthetic capacity is similar for high-light and low-light plants, as is the protein concentration (Evans & Poorter, 2001). Thus, photosynthetic capacity scales linearly with the biomass investment in the leaf, making leaf anatomy the main driver of the light-saturated rate of photosynthesis. As in low-light plants, genetic manipulations or mutations that decrease the photosynthetic machinery often decrease LMA as well. For example, antisense Rubisco plants do have a lower photosynthetic capacity per unit leaf area. But they also have a lower LMA (Quick *et al.*, 1991), similar to low-light plants, which implies that their mass-based rates of photosynthesis are not affected as much as the area-based rates. Consequently, RGR values are rather similar to those of the wild type (Masle *et al.*, 1993).

Genotypes may show similar covariation of LMA and photosynthesis. When the first portable infrared gas analysers allowed screening of a wide range of genotypes of alfalfa, a strong positive relationship was reported between photosynthetic capacity and LMA (Pearce *et al.*, 1969). The authors therefore concluded that LMA was a good proxy to select for high-yielding genotypes. However, per unit leaf mass, all genotypes performed the same, which implies that the C-gain and growth rate of individually-grown plants could well be similar.

VIII. Ecological consequences

1. LMA is an important component of plant strategies

The LMA plays a central role in various plant strategy schemes (Westoby, 1998; Grime, 2001; Westoby *et al.*, 2002), where it can be considered an index of a species' position along a continuum between low-LMA species at one end (that realize a fast resource acquisition and growth), and high-LMA species at the other (that realize a high resource conservation and persistence). Low-LMA species therefore tend to have a fitness advantage under high-resource conditions and are typically found in productive habitats, whereas high-LMA species have a fitness advantage under adverse growing conditions and are typically found in unproductive habitats. As a consequence, LMA varies among species that partition environmental gradients, and tends to be higher for species from oligotrophic habitats (Poorter & De Jong, 1999; Wright & Westoby 1999), evergreen species from shaded (Walters & Reich, 1999; Lusk *et al.*, 2008; Poorter, 2009) or low-rainfall habitats (Wright *et al.*, 2001; Santiago *et al.*, 2004; Schulze *et al.*, 2006) and species from undisturbed habitats (Louault *et al.*, 2005). The LMA is found to be one of the main traits that determines the primary axis of specialization among British herbaceous plants (Grime *et al.*, 1997). As such, it has been used together with leaf dry matter content (Hodgson *et al.*, 1999; Caccianiga *et al.*, 2006) to assign species a position in the CSR-strategy scheme of Grime (2001).

2. LMA as part of a trait complex

The LMA is part of a whole suite of interconnected traits that together shape the performance of plants. Species with low LMA tend to have a high concentration of proteins and minerals, a high water content, a low concentration of lignin and other secondary compounds, and a fast metabolism (high rates of photosynthesis and respiration per unit leaf dry mass). Such species also show a high rate of photosynthesis per unit leaf nitrogen; they generally have leaves that require less force to tear apart or puncture and have a short life-span (Lambers & Poorter, 1992 and references therein; Wright & Westoby, 2002). As a consequence of this, such species have a high RGR under optimal growth conditions. High LMA leaves seem to be built to persist; under laboratory conditions, where herbivores are offered a choice between a range of leaves with different LMA, high-LMA leaves are generally avoided (Fig. 8a), and similar preferences can be observed in the field (Pérez-Harguindeguy *et al.*, 2003). Because of a better defence against herbivores and physical hazards, there is a positive correlation between LMA and leaf longevity in the field (Fig. 8b; Wright *et al.*, 2004). Species with an inherently high LMA not only have a greater lifespan of leaves but also of roots (Ryser, 1996), thereby more efficiently conserving the acquired nutrients and carbon. A high leaf and root longevity enhances the residence time of nutrients in plants, thus providing high-LMA species with a competitive advantage on nutrient-poor soils (Aerts & Chapin, 2000). However, this comes at the expense of a reduced inherent growth rate.

This complex of correlated traits is rather robust: when world-wide comparisons are made across species growing in their natural habitats, we see again the same trade-offs with low-LMA species having a higher nitrogen concentration, and specific assimilation and respiration rates, and high-LMA species showing a long leaf lifespan (Reich *et al.*, 1997; Díaz *et al.*, 2004; Wright *et al.*, 2004).

3. Is LMA a good predictor of plant performance?

When it comes to an ecological evaluation, the individual role of each of these traits is not easily separated and evaluated.

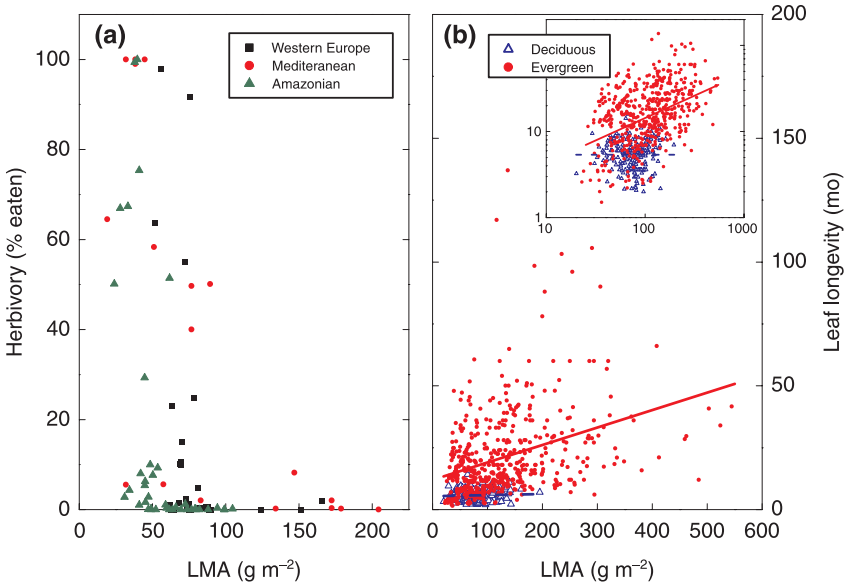

Fig. 8 (a) Herbivory (% of leaves eaten from different species) as a function of the leaf dry mass per unit area (LMA) of those species. Data were obtained in cafeteria experiments, where herbivores (snails) were allowed to choose between leaves of a variety of species. Data from Cornelissen *et al*. (1999), R. Villar (unpublished) and L. Poorter (unpublished). (b) Leaf longevity of leaves under field conditions as dependent on LMA. Separate lines are given for deciduous ($n = 230$; $r^2 = 0.00$; $P > 0.4$) and evergreen ($n = 510$; $r^2 = 0.13$; $P < 0.001$) species. Data are from the glopnet data set (Wright *et al*., 2004), supplemented with 60 observations from tropical tree species (L. Poorter, unpublished) and 100 observations from Shiodera *et al*. (2008). Results were essentially similar when data were analysed after log-transformation (see insert).

One way to get quantitative insight into the effects of a single trait on plant performance is to use a modelling approach. Spitters & Aerts (1983), for example, modelled the effect of weeds on field crop performance, and showed that a low LMA before canopy closure provided weeds with a competitive advantage over crop species. Using a game-theoretical model, Schieving & Poorter (1999) showed that a genotype in a vegetation that realizes a lower LMA than its neighbours can increase its fitness, even to the extent that the productivity of the whole stand would decrease. Sterck *et al*. (2006) combined field data with a process-based plant model to show that LMA is the best predictor of interspecific variation in sapling growth in high-light gaps, whereas leaf lifespan is the best predictor of interspecific variation in sapling survival in the shaded forest understory.

In real plants, where LMA is one of the correlated traits in the trait complex, the situation may be more complex. The LMA is often used because it is one of the easy to measure parameters, with a good foundation in growth theory. However, this does not imply that for specific aspects it is always the best predictor of plant performance. For example, in the cafeteria experiments described above, some low-LMA leaves are eaten as little as high-LMA species. In such cases protein and water content will be better predictors for a given subset of species than LMA itself (Pérez-Harguindeguy *et al*., 2003). Secondary defence compounds will also play a role in herbivory. In those cases, even small concentrations that hardly affect LMA may significantly curb the set of herbivores able to feed on given leaves (Coley, 1983). Also, the relationship between LMA and leaf longevity is more subtle than may be expected at first sight. When deciduous and evergreen species are analysed independently they do not follow the same overall relationship: the slope of the line describing this relationship is relatively steep in the case of evergreens (Fig. 8b; $P < 0.001$) but does not significantly deviate from zero for the deciduous species. This implies that evergreens can gain a higher leaf longevity per additional investment of biomass. Deciduous species, which are genetically programmed to abscise their leaves at the end of the growing season, do not show such a gain in leaf longevity with an increase in LMA. The positive relationship between LMA and leaf longevity, although general across species, may even reverse when sun and shade leaves on the same plant are considered: sun leaves have a higher rate of photosynthesis and a higher LMA, but often a lower leaf longevity (Miyaji *et al*., 1997; Poorter, 2001 and references therein; Lusk *et al*., 2008).

Most of the above-mentioned parameters relate to performance comparisons of key traits in different species. However, do these differences ultimately lead to increased fitness and if so, under which conditions? This question is not easily answered. There have been experiments in which an inherently high LMA, as observed in the glasshouse, could be positively related to reproductive output of these genotypes in the field under low nutrient conditions, but negatively related in high-nutrient sites (data from Biere, 1996, presented in Poorter & Garnier, 2007). Clearly, more experimental work is required.

4. The importance of LMA variation for ecosystem properties

Leaf traits also have consequences for ecosystem functioning, through the processes of primary production, trophic transfer, litter decomposition, and carbon and nutrient cycling (Lavorel & Garnier, 2002; Díaz *et al*., 2004). Low-LMA leaves are eaten preferentially by herbivores (Fig. 8a; Louault *et al*., 2005), and a productive vegetation with low-LMA leaves may

therefore enhance the carrying capacity for herbivores. Leaves also have an 'after-life' effect: species that produce low-LMA leaves with high nutrient contents also decompose much faster, leading to increased carbon and nutrient cycling, thereby speeding up ecosystem productivity in different biomes (Cornelissen *et al.*, 1999). Moreover, species-driven variation in decomposition rate (18-fold variation across species within a site) is much larger than climate-driven variation (sixfold differences for the same material across sites; Cornwell *et al.*, 2008).

How can we scale up from leaf traits to ecosystem processes and services? Ecosystem processes are determined either by the trait values of the dominant species that contribute most to vegetation biomass (the mass ratio hypothesis) or by the variation in trait values because species fulfil complementary roles (the niche complementarity hypothesis; Díaz *et al.*, 2007). Garnier *et al.* (2004) tested these hypotheses by calculating for different successional plots the community-weighted LMA (i.e. the sum of the LMA of the species in the community weighed by their relative abundance), and the coefficient of variation in species LMA. Species LMA increased markedly during old-field succession, with fast-growing early-successional species with low LMA being replaced by slow-growing late-successional species with high LMA. Community-level LMA was the best predictor of ecosystem processes, lending strong support to the mass ratio hypothesis. Community-level LMA had a strong positive impact on the soil nitrogen and carbon pools, and a strong negative impact on the primary productivity and decomposition rate of the successional communities.

IX. Conclusions and perspectives

In this paper we have shown first that considerable variation in field-measured LMA can be explained by categorizing species into functional groups or into biomes. However, a large part of the variation is still unaccounted for, presumably because within groups and biomes there is great variation in species' strategies for carbon gain and conservation (Westoby *et al.*, 2000), just as there is among co-occurring species.

Second, we demonstrated that there is inherent variation in LMA between a number of functional groups of species. Considering the volumes of the different tissues per unit area and their densities provides a consistent framework for linking variation in different anatomical tissues directly to LMA, and in this way also to variation in the growth rate of plants. Although considerable work has been done on leaf anatomy, we have still a relatively poor quantitative understanding of the importance of changes in cell number and cell volumes of the different tissues for LMA and growth.

Third, we have constructed, from a wide body of literature data, LMA response curves for 12 different environmental variables. We could rank these factors by the quantitative effect they had on LMA, and showed that ecologically different groups of species respond differently to a given environmental factor. Moreover, by presenting the interquartile range (distance between the 75th percentile and the 25th percentile) for the response at a given level of that environmental parameter, we provide a benchmark by which new results can be evaluated quantitatively against the present literature. Once having established how species respond in general to a given environmental factor, one of the challenges ahead is to evaluate how species respond to interacting environmental factors. Another interesting point is to what extent the environmental response of *Arabidopsis*, the model species of the moment, is representative of the response of different species groups.

Finally, we have discussed various mechanisms by which a high or low LMA can contribute to the success of a given plant species in the field, with important consequences for ecosystem processes. However, LMA is part of a trait complex, and our mechanistic understanding of the genetic and physiological factors determining this success is still limited. Clearly, if we want to understand the response of plants to their environment, there are many challenges ahead.

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Appendix A1: measuring LMA

In the literature, leaf mass is generally taken as the dry mass, and leaf area as the projected leaf area, which coincides with half the total area of both sides of flat leaves. However, in needle-like leaves the situation is more complicated, as projected leaf area is not necessarily similar to half the total leaf area. Different investigators have followed different solutions here using either projected or half the total leaf surface area. From the perspective of light interception by convex objects, half of the total surface area is the quantity scaling with light interception if the foliage angular distribution were uniform (Chen & Black, 1992) and this would be our preferred approach.

Another complication is that some researchers include the petiole in their measurements, whereas others only consider the leaf blade. Yet others even remove part of the midrib. Moreover, there can be an issue for leaves that are curled, epinastic, or somewhat cup-shaped. A third methodological aspect especially relevant under field conditions is the water status of the plant material collected. Plants measured directly after harvest may have an LMA 20% lower than when saturated with water overnight (Garnier *et al.*, 2001). This effect is especially strong for low-LMA species and may introduce extra variability in species comparisons.

Undoubtedly, all three types of variation are present in our compiled database. However, there is no simple way to correct for these different approaches in the current analysis, not least because they are not always well documented. Therefore, we decided to include all values as reported, except in those cases where the total leaf area of both sides was measured (e.g. Fliervoet & van de Ven, 1984). These values were recalculated on a one-sided basis. For more extended recommendations on measurement procedures see Cornelissen *et al.* (2003b).

Appendix A2: field data

Literature data from the glopnet database (Wright *et al.*, 2004) and Niinemets (2001) were supplemented by those listed in the Supporting Information, Table S1. Species were categorized into one of 10 functional groups. All species that were succulent were allocated to the functional group succulents, even ferns. Gymnosperms only belonged to the category gymnosperms. For species with more than one entry in the database, we averaged all LMA values before the analysis. For each functional group the distribution was characterized by percentiles. For the distribution of species over habitats we used the classification of Whittaker (1975), where we defined grassland as those vegetations with hardly any woody species present (steppes, European grasslands including Mediterranean ones), shrubland as low vegetation with considerable presence of woody species (Mediterranean maquis, Fijnbos) and woodland as open vegetation with trees present (such as savannas). We included temperate rain forest into the general group of temperate forests and excluded very specific vegetation types, such as mangroves.

Appendix A3: inherent differences

Inherent differences in LMA between functional groups of species were taken from the literature, listed in the Supporting Information, Table S2. For each experiment where species from group A and B were grown together, the average LMA was calculated per group of species and then the ratio between these two averages was calculated. For comparisons of fast- and slow-growing species, where differences are not dichotomous but continuous, we averaged the LMA of the 33% fastest-growing species and of the 33% slowest-growing species, and then calculated a ratio from these average values. The ratios calculated per experiment were subsequently averaged over all experiments considered. Because ratios are distributed ln-normally, we carried out a ln-transformation before statistical testing.

Appendix A4: LVA and density

Leaf thickness and leaf density are not very often measured along with LMA. For those experiments where thickness was determined microscopically, we calculated density with Eqn 2. Alternatively, density can be estimated from the dry mass–fresh mass ratio, and an estimate of leaf thickness derived from that (Vile *et al.*, 2005). The correlation between density and dry to fresh mass ratio is not always that strong, partly because different leaf tissue fractions, lamina, petiole and midrib, have different relationships between density and dry to fresh mass ratio (Niinemets *et al.*, 2007), partly because cross-sections only measure part of the leaf lamina.

We subsequently used a ln–ln scaling slope analysis to estimate the contribution of LVA (T) and LD (D) to variation in LMA (L), as explained in Appendix 1 of Poorter & van der Werf (1998). In short, if two species A and B or plants subjected to treatment A and B differ in LMA, they also differ in the product of T and D. The relative difference in LMA then becomes:

$$\frac{L_A}{L_B} = \frac{T_A \times D_A}{T_B \times D_B} \qquad \text{Eqn A1}$$

After ln-transformation Eqn A1 becomes

$$(\ln L_A - \ln L_B) = (\ln T_A - \ln T_B) + (\ln D_A - \ln D_B) \qquad \text{Eqn A2}$$

By dividing both the leaf and right side of Eqn A2 by its left side, one gets:

$$1 = \frac{(\ln T_A - \ln T_B)}{(\ln L_A - \ln L_B)} + \frac{(\ln D_A - \ln D_B)}{(\ln L_A - \ln L_B)} \qquad \text{Eqn A3}$$

The first part of the right-hand term of Eqn A3 gives the relative contribution of LVA to variation in the LMA ratio, the second part the contribution of LD, with the sum being one.

As such, they indicate the relative importance of each of these factors. This approach can easily be extended by more species or treatments by calculating the slope of the ln-transformed density or LVA against ln-transformed LMA (Poorter & van der Werf, 1998), and can be used to generalize about the importance of LD and LVA in causing LMA differences across a range of experiments. In all cases, a value of 1 for this slope indicates that the given variable is completely responsible for variation in LMA, whereas a value of 0 indicates that variation in the given variable does not correlate with variation in LMA at all.

Appendix A5: environmental effects

To be able to compare different experiments, we choose a standard level for each factor that was in the range of values for most published experiments. For example, in the case of CO_2, this value was 350 μmol mol^{-1}. For each experiment values at high or low concentrations were then expressed relative to the LMA value for plants grown at 350 μmol mol^{-1}. Where no such treatment was included, we estimated the reference LMA by linear interpolation, or very rarely by extrapolation in those cases where the closest CO_2 concentrations were at most 10% apart from the reference concentrations. A reference value for nutrient or water supply is not easily defined; in this case we scaled all LMA values relative to the LMA value of plants at the highest nutrient or water treatment and used the biomass at low supply relative to that at high supply as an indicator of the severity of the treatment. Where the highest level was supra-optimal for growth, we choose the level where plants were showing the highest biomass. In simple dichotomous treatments (such as control versus submergence) the LMA values were expressed relative to the control plants.

We subsequently calculated the distribution of the scaled LMA values over different subsections of the response curve. For example, the response to CO_2 was calculated for the observations up to a concentration of 300 μmol mol^{-1}, from 300 to 400, from 400 to 600, from 600 to 800 and above 800. In each subsection we characterized the distribution by percentiles, and a response curve was constructed from the median values of LMA response and CO_2 concentration in each subsection. Plant species were categorized as far as possible with respect to their ecological preference and the response curve analysed for each category separately. Thus, in the case of CO_2, plants were categorized as C_3 and C_4. The median values for each group are also presented in the graphs as broken lines. We then analysed, by regression, whether the LMA response was changing linearly with a given factor, or whether there was also a significant quadratic component to it. This analysis was carried out after ln-transformation of the scaled LMA data, as ratios are ln-normally distributed by nature. In the case of significant treatment by species group interaction response curves were also calculated for species subgroups. Response ratios were calculated for each environmental factor by dividing the highest fitted value across the range by the lowest one.

The literature on which the database was built is listed in the Supporting Information, Table S3. No differentiation was made between LMA data measured for whole plants or for a specific leaf. The selected reference values for the environmental conditions were: (a) daily photon irradiance, 8 mol m^{-2} d^{-1}; (b) red : far-red ratio (R : FR), 1.2; (c) UV-B, 10 kJ m^{-2} d^{-1}; (d) CO_2 concentration (35 Pa); (e) ozone, 20 nmol mol^{-1}; (f) nutrient availability, biomass at the least-limiting nutrient level; (g) water availability, biomass at the least-limiting water level; (h) waterlogging, control treatment; (i) submergence (control treatment); (j) temperature, average daily temperature of 18°C; (k) salinity, 0% seawater; (l) soil compaction, 1.2 g soil cm^{-3}.

Supporting Information

Additional supporting information may be found in the online version of this article.

Table S1 Literature used for the meta-analysis: field observations

Table S2 Literature used for the meta-analysis: inherent differences

Table S3 Literature used for the meta-analysis: effect of the environment

Table S4 Literature used for Fig. 7

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