

Ecophysiological traits of plant functional groups in forest and pasture ecosystems from eastern Amazônia, Brazil

Tomas F. Domingues · Luiz A. Martinelli ·
James R. Ehleringer

Received: 1 December 2005 / Accepted: 5 December 2006
© Springer Science+Business Media B.V. 2007

Abstract The plant functional group approach has the potential to clarify ecological patterns and is of particular importance in simplifying the application of ecological models in high biodiversity ecosystems. Six functional groups (pasture grass, pasture sapling, top-canopy tree, top-canopy liana, mid canopy tree, and understory tree) were established a priori based on ecosystem inhabited, life form, and position within the forest canopy profile on eastern Amazonian region. Ecophysiological traits related to photosynthetic gas exchange were then used to characterize such groups. The ecophysiological traits evaluated showed considerable variations among groups. The pasture grass functional group (a C_4 photosynthetic pathway species) showed high instantaneous water use

efficiency ($A_{\max}/g_s@A_{\max}$), high photosynthetic nitrogen use efficiency (A_{\max}/N_{area}), and high ratio of A_{\max} to dark respiration (A_{\max}/R_d). Among the species with the C_3 photosynthetic pathway, the top-canopy liana group showed the highest mean of $A_{\max}/g_s@A_{\max}$, statistically distinct from the lowest average presented by the understory tree group. Furthermore, the pasture sapling group showed the lowest average of A_{\max}/R_d , statistically distinct from the high average observed for the understory tree group. Welch-ANOVAs followed by Games–Howell post hoc tests applied to ecophysiological traits produced reasonable distinctions among functional groups, although no significant distinction was detected between the groups top-canopy tree and pasture sapling. Species distribution within the functional groups was accurately reproduced by discriminant analyses based on species averages of ecophysiological traits. The present work convincingly shows that the functional groups identified have distinct ecophysiological characteristics, with the potential to respond differently to environmental factors. Such information is of great importance in modeling efforts that evaluate the effects of dynamic changes in tropical plant communities over ecosystem primary productivity.

T. F. Domingues · J. R. Ehleringer
University of Utah, 1400 East 257 South,
Salt Lake City, UT 84112-0840, USA

L. A. Martinelli
Centro de Energia Nuclear na Agricultura,
Universidade de São Paulo - Avenida Centenário,
303 Piracicaba, Sao Paulo, SP 13416-000, Brazil

T. F. Domingues (✉)
Institute of Geography, School of GeoSciences,
The University of Edinburgh, Drummond Street,
Edinburgh EH8 9XP, Scotland, UK
e-mail: tdomingu@staffmail.ed.ac.uk

Keywords $A-c_i$ · Carbon · Nitrogen ·
Stable isotope · Stomatal conductance

Introduction

Historically, research in plant ecology has acknowledged that different plant species inhabiting the same environment often displayed similarities in life form and ecophysiological traits supporting the artificial arrangements of species into functional groups or types (Raunkiaer 1934; Holdridge 1947; Solbrig 1993; Weiher et al. 1999; Grime 2001; Cornelissen et al. 2003; Díaz et al. 2004). Aggregating species into functional groups is a common methodology useful for reducing complexity of high diversity ecosystems (e.g., tropical rain forest plant communities) (Hubbell 2005) and for linking different scales such as species to region (Dawson and Chapin 1993; Körner 1993; Smith et al. 1997). Furthermore, the study of plant functional groups is highly relevant for ecosystem process models that allow plant communities to change over time (Peng 2000) being therefore relevant for global change research (Hobbs 1997; Loreau et al. 2001). For example, the increasing concentration of carbon dioxide gas in the atmosphere might be causing changes in recruitment rates in tropical rain forest ecosystems (Phillips et al. 2004). Since such changes might affect functional groups differently (e.g., light demanding versus shade-tolerant species, C₃ vs. C₄, top-canopy trees versus top-canopy lianas), atmospheric CO₂ alone is most likely a key driver for species composition change (Cerling et al. 1998; Würth et al. 1998; Phillips et al. 2002, 2004).

Our current ecophysiological understanding of how plants interact with their environment allows plant performance in terms of carbon, nutrients and water exchange to be predicted based on a few parameters (Ehleringer and Björkman 1977; Farquhar et al. 1980; Marshall and Biscoe 1980; Farquhar and Sharkey 1982; Field and Mooney 1986; Evans 1989; von Caemmerer 2000). Through adaptation and acclimation, environmental factors have a strong influence over parameter ranges allowing simple relationships to be established (Reich et al. 1997; Ackerly et al. 2000; Domingues et al. 2005). Since such parameters exhibit natural variations and are central in plant-to-plant interactions, they should therefore be used to establish differences among plant functional groups.

Although the available literature concerning plant functional groups is substantial, studies focussed on tropical forest vegetation and its own challenges are still scarce, despite the global importance of this biome. In this work, we evaluate six a priori plant functional groups composed of species inhabiting eastern Amazonian tropical rain forest and cattle ranch pasture ecosystems. The criteria for group composition were based on where species occurred (e.g., pasture vs. forest, top-canopy vs. understory) and based on life forms (e.g., lianas vs. trees). We then used traits related to photosynthetic gas exchange to test the hypothesis of functional equivalence, where species within the same functional groups would display similar characteristics regarding carbon acquisition, water use, and nutrient utilization.

Methods

Study site

Between November 1999 and December 2003, we measured leaf gas exchange and other leaf parameters at both undisturbed eastern Amazonian terra-firme tropical rain forest and cattle ranch pasture ecosystems. Nine field campaigns from November 1999 through December 2003 covered both wet and dry seasons. The forest ecosystem was situated within the Flona-Tapajós, a 600,000 ha national forest reserve in Brazil, 50 km south of Santarém, PA (Lat. 2° 51' 22" S, Long. 54° 57' 29" W). Access to leaves of trees and lianas was from four towers ranging from ca. 25 to 45-m in height. Edaphic, micrometeorological and biological information about this region are presented elsewhere (Silver et al. 2000; Keller et al. 2001; Nepstad et al. 2002; Telles et al. 2003; da Rocha et al. 2004; Goulden et al. 2004; Vieira et al. 2004; Domingues et al. 2005). In summary, the annual precipitation of the region averages 2,000 mm yr⁻¹, being lower during El-Niño years. A well-characterized dry season extends from July through November when precipitation is usually below 100 mm month⁻¹. Air temperature above the canopy varies little throughout the year, with maximum daily temperatures ranging between 24

and 32°C and minimum daily temperatures ranging between 20 and 25°C.

The pasture site used in this study (Lat. 3° 0' 44" S, Long. 54° 32' 14" W) was located approximately 10 km from the primary forest site, and was therefore under the same regional climatic regime of the forest site. The pasture was characteristic of an old (ca. 13 years) cattle ranch farm planted with the C₄ grass *Brachiaria brizantha* Stapf. (Sakai et al. 2004), and moderately colonized by saplings of forest species.

Plant species and functional groups

Leaves from 72 individual plants belonging to 63 species were analyzed for this study (Table 1). Such species and respective plant functional groups were chosen due to their relevance to ecosystem level carbon, water and energy cycles. Photosynthetic gas exchange measurements were performed on a subgroup of 28 species. Plant voucher materials of species obtained from one of the forest towers were sent to the IAN-EMBRA-PA herbarium (Belém, Br) for taxonomic identification and subsequent incorporation into its collection. When possible, species identification for the remainder of the forest species was obtained from available inventories (Keller et al. 2001; Miller et al. 2004).

Based on life form and species distribution within the canopy profile, we grouped species from the forest site into four functional groups: top-canopy tree, top-canopy liana, mid canopy tree, and understory tree (Table 1). Top-canopy species were characterized as having fully sun-lit leaves in the upper 30% of the forest canopy, while understory species occupied the lower 10% of the canopy profile. We grouped plants from the pasture site into two functional groups: pasture grass (one C₄ species) and pasture sapling (four C₃ species).

Leaf properties

Leaf mass to leaf area ratio (LMA g m⁻²) was obtained by dividing fresh leaf area by its dry weight. Leaf area was obtained by tracing fresh leaves onto paper and posterior area determination of digitalized images. Leaf weight was obtained after drying leaves at 65°C for 48 h.

Leaf δ¹³C, δ¹⁵N and nitrogen content (N_{leaf}) were determined on sub-samples (1–2 mg) of the dried leaf samples by continuous-flow isotope ratio mass spectrometry (IRMS Delta Plus Finnigan, San Jose, CA, USA) coupled with an elemental analyzer (Carlo Erba, Milan, Italy) at the Laboratório de Ecologia Isotópica, CENA-USP, Piracicaba, Brazil. Isotope ratios are expressed in delta notation relative to PDB standard as:

$$\delta X = \left[\left(\frac{R_{\text{sample}}}{R_{\text{standard}}} \right) - 1 \right] \times 1000$$

δX is either δ¹³C or δ¹⁵N, while R_{sample} and R_{standard} are ratios of heavy to light isotopes of the sample and standard, respectively. The precision of the isotopic measurements was ±0.2‰.

Foliar total nitrogen contents were obtained with the elemental analyzer on a mass basis (N_{mass}) and multiplied by the corresponding LMA in order to express on an area basis (N_{area}) (g N m⁻²).

Gas exchange measurements

Leaf level gas exchange measurements were collected with an infrared gas analyzer coupled to a leaf chamber equipped with a red–blue light source and an external CO₂ source (model LI-6400, Li-Cor, Nebraska, USA). Photosynthetic carbon assimilation rate (A), dark respiration rates (R_d), stomatal conductance to water vapor (g_s), and the ratio between intercellular to atmospheric carbon dioxide concentration (c_i/c_a) were calculated by the LI-6400 software (OPEN versions 3.3, 4.04, and 5.1) based on equations presented by von Caemmerer and Farquhar (1981). On all occasions, the leaf area used was 6 cm² and the stomatal ratio used was 0.5. The environment inside chamber was controlled to maintain leaf temperature at 30°C, relative humidity ca. 80%, CO₂ partial pressure at the sample cell at 36 Pa, and saturating levels of photosynthetically active photon flux density (PPFD) (800 μmol m⁻² s⁻¹ for understory plants and 1800 μmol m⁻² s⁻¹ for other species). All measurements were performed on fully expanded leaves. Field measurements were usually limited to morning hours (8:00–13:00 h local time).

Table 1 The distribution within functional groups of plant species evaluated from both primary forest and pasture ecosystems from Santarém, Brazil

Species	Family	Functional group	Height (m)	Relative height (%)
<i>Abuta rufescens</i> *	Menispermaceae	Upper liana	33	86
<i>Arrabidaea prancei</i> *	Bignoniaceae	Upper liana	26	85
<i>Combretum</i> sp.*	Combretaceae	Upper liana	34	83
<i>Memora tanaecicarpa</i> *	Bignoniaceae	Upper liana	26	85
<i>Prionostemma aspera</i> *	Hippocrateaceae	Upper liana	38	93
<i>Tetrapteryx</i> sp.*	Malpighiaceae	Upper liana	38	93
<i>Copaifera duckei</i> *	Caesalpiniaceae	Upper tree	36	89
<i>Cordia bicolor</i> *	Boraginaceae	Upper tree	27	86
<i>Goupia glabra</i>	Celastraceae	Upper tree	26	85
<i>Lecythis lurida</i>	Lecythidaceae	Upper tree	29	71
<i>Lecythis lurida</i> *	Lecythidaceae	Upper tree	32	91
<i>Licania michelli</i>	Chrysobalanaceae	Upper tree	24	77
<i>Manilkara huberi</i> *	Sapotaceae	Upper tree	30	72
<i>Micropholis</i> sp.*	Sapotaceae	Upper tree	32	91
<i>Ocotea rubra</i>	Lauraceae	Upper tree	32	91
<i>Perebea molis</i>	Moraceae	Upper tree	27	87
<i>Tachigali myrmecophila</i> *	Caesalpiniaceae	Upper tree	27	86
<i>Amphirrhox surinamensis</i>	Violaceae	Mid tree	4	13
<i>Anomalocalyx uleanus</i> *	Euphorbiaceae	Mid tree	21	60
<i>Bactris elegans</i>	Arecaceae	Mid tree	4	13
<i>Chimarrhis turbinata</i>	Rubiaceae	Mid tree	11	28
<i>Copaifera duckei</i>	Caesalpiniaceae	Mid tree	26	63
<i>Coussarea macrophylla</i>	Rubiaceae	Mid tree	4	13
<i>Coussarea racemosa</i>	Rubiaceae	Mid tree	6	19
<i>Derris amazonica</i>	Fabaceae	Mid tree	14	34
<i>Derris</i> sp.	Fabaceae	Mid tree	8	26
<i>Erismia uncinatum</i>	Vochysiaceae	Mid tree	10	32
<i>Eschweilera coriacea</i>	Lecythidaceae	Mid tree	26	73
<i>Faramea capillipes</i>	Rubiaceae	Mid tree	6	15
<i>Faramea platyneura</i> *	Rubiaceae	Mid tree	9	22
<i>Faramea stenopetala</i>	Rubiaceae	Mid tree	6	19
<i>Forsteronia mollis</i>	Apocynaceae	Mid tree	14	45
<i>Iryanthera sagotiana</i>	Myristicaceae	Mid tree	18	58
<i>Lecythis lurida</i>	Lecythidaceae	Mid tree	28	68
<i>Lecythis lurida</i>	Lecythidaceae	Mid tree	28	80
<i>Lecythis lurida</i>	Lecythidaceae	Mid tree	16	52
<i>Lecythis</i> sp.*	Lecythidaceae	Mid tree	14	35
<i>Licania macrophylla</i>	Chrysobalanaceae	Mid tree	4	13
<i>Licania michelli</i>	Chrysobalanaceae	Mid tree	21	69
<i>Licaria brasiliensis</i>	Lauraceae	Mid tree	23	73
<i>Licaria brasiliensis</i>	Lauraceae	Mid tree	14	45
<i>Machaerium amazonense</i>	Fabaceae	Mid tree	16	39
<i>Manilkara huberi</i>	Sapotaceae	Mid tree	16	52
<i>Memora tanaecicarpa</i>	Bignoniaceae	Mid tree	20	65
<i>Mouriri brachyanthera</i>	Melastomataceae	Mid tree	9	23
<i>Moutabea guianensis</i>	Polygalaceae	Mid tree	16	52
<i>Perebea mollis</i>	Moraceae	Mid tree	21	68
<i>Poecilanthus effusa</i>	Fabaceae	Mid tree	4	13
<i>Pouteria macrophylla</i>	Sapotaceae	Mid tree	16	40
<i>Pouteria macrophylla</i>	Sapotaceae	Mid tree	10	31
<i>Pouteria venosa</i>	Sapotaceae	Mid tree	6	19
<i>Protium puncticulatum</i> *	Burseraceae	Mid tree	18	53
<i>Protium</i> sp.	Burseraceae	Mid tree	8	19
<i>Protium spruceanum</i>	Burseraceae	Mid tree	20	65
<i>Sclerolobium melinonii</i>	Caesalpiniaceae	Mid tree	11	31
<i>Sclerolobium paraense</i> *	Caesalpiniaceae	Mid tree	19	46

Table 1 continued

Species	Family	Functional group	Height (m)	Relative height (%)
<i>Tabebuia serratifolia</i>	Bignoniaceae	Mid tree	13	37
<i>Tachigali myrmecophila</i> *	Caesalpiniaceae	Mid tree	19	61
<i>Virola elongata</i>	Myristicaceae	Mid tree	4	13
Not identified	Not identified	Mid tree	7	21
Not identified	Not identified	Mid tree	16	39
<i>Amphirrhox surinamensis</i>	Violaceae	Understory	1	3
<i>Coussarea micrococca</i>	Rubiaceae	Understory	1	3
<i>Cydista aequinoetialis</i>	Bignoniaceae	Understory	2	6
<i>Duguetia cadaverica</i> *	Annonaceae	Understory	1	3
<i>Faramea capillipes</i>	Rubiaceae	Understory	2	5
<i>Miconia acinodendron</i> *	Melastomataceae	Understory	1	3
<i>Palicourea</i> sp.	Rubiaceae	Understory	2	5
<i>Rinorea neglecta</i> *	Violaceae	Understory	1	2
<i>Sclerolobium paraense</i> *	Caesalpiniaceae	Understory	1	2
Not identified	Not identified	Understory	1	3
Not identified*	Not identified	Understory	1	2
Not identified*	Not identified	Understory	1	2
<i>Desmodium</i> sp.*	Fabaceae	Understory	1	100
<i>Dichapetalum sprucanum</i> *	Dichapetalaceae	Understory	1	100
<i>Vismia</i> sp.*	Guttiferae	Understory	1	100
Not identified*	Not identified	Understory	1	100
<i>Brachiaria brizantha</i> *	Poaceae	Pasture grass	1	100

* Plants associated with photosynthetic gas exchange measurements

Maximum carboxylation capacity values (V_{cmax}) were calculated for species used for gas exchange measurements (Table 1). For that, 117 $A-c_1$ response curves were produced by varying carbon dioxide concentration while under saturating light levels ($800 \mu\text{mol m}^{-2} \text{s}^{-1}$ for understory species and $1800 \mu\text{mol m}^{-2} \text{s}^{-1}$ for all other species) and 124 A -PPFD response curves were produced by varying PPFD level under constant CO_2 concentrations (36 Pa). During all response curves, the leaf temperature and relative humidity were held constant at 30°C and 80%, respectively. Maximum rate of electron transport values (J_{max}) were obtained only from $A-c_1$ curves. A biochemical photosynthesis model (Farquhar et al. 1980; von Caemmerer 2000) was fitted to the field data by nonlinear least square method using Microsoft Excel solver routine (Microsoft Corporation, Excel v. X for Mac) assuming non-negative values and using automatic scaling, quadratic estimates, central derivative, and conjugate search. By iteration, this routine returned V_{cmax} and J_{max} values associated with the smallest error. The V_{cmax} and J_{max} values reported here are scaled to the reference leaf temperature of 30°C .

Statistical analyses

Averages of parameters for individual species were used for statistical analysis. A Levene's test was applied to each parameter to determine homogeneity of variances. For parameters that presented homogeneous variances, one-way analyses of variance (ANOVA) were employed to test if ecophysiological parameter means differed among functional groups. For parameters that presented heterogeneous variances, a Welch-ANOVA was employed to test differences among functional groups means. In the cases that Welch-ANOVA tests indicated significant differences among parameter means, Games-Howell post hoc tests were applied in order to establish pairwise comparisons among functional groups. Linear discriminant analyses based on ecophysiological parameters were used to classify species into the a priori established plant functional groups. We used JMP statistical software (JMP 5.1.2 for MacIntosh, SAS Institute Inc., Cary, NC) for all statistical analyses, with the exception of the Games-Howell post hoc tests where SPSS statistical software was used (SPSS version 13.0 for windows, SPSS Inc. Chicago, IL).

Results

Gas exchange parameters

Due to the occurrence of heterogeneous variances detected for most of the parameters (Table 2), a Welch-ANOVA was applied to detect inter-group differences in parameter means. The ecophysiological data evaluated provided substantial distinctions among functional groups. Yet, no among-groups differences were detected based on $g_s@A_{max}$, N_{mass} or C/N ratio. Following the ANOVA tests, post hoc tests (Games–Howell) were applied in order to determine which traits contributed to the definition of the functional groups. The pasture grass functional group was excluded from the inter-group statistical comparisons because it was composed by a single species (*B. brizantha*) and because it displayed a very distinct pattern of ecophysiological traits due to the C_4 photosynthetic pathway used by that species. As expected from its physiological characteristics, the pasture grass functional group presented low c_i/c_a mean associated with high $\delta^{13}C$ average, while achieving high photosynthetic rates (Table 2).

Among the groups with the C_3 photosynthetic pathway, the understory tree group was the most distinct. This group was statistically distinct from all other groups based on LMA and $\delta^{13}C$ (Table 2). Furthermore, this group differed from the mid canopy tree group based on J_{max} and N_{area} , and from both the top-canopy tree and the top-canopy liana groups based on A_{max} , V_{cmax} , J_{max} , N_{area} , and c_i/c_a (Table 2). The understory tree group could be further separated from the top-canopy liana group based on R_d , and $\delta^{15}N$ (Table 2).

The mid canopy group was less sharply distinct from other groups when compared to the patterns presented by the understory tree group. Nonetheless, the mid canopy group was statistically different from pasture sapling group based on R_d , different from the top-canopy liana group based on $\delta^{13}C$, and different from the top-canopy tree group based also on $\delta^{13}C$ (Table 2).

The pasture sapling, top-canopy tree and top-canopy liana groups were similar to each other, although the top-canopy tree and the top-canopy

Table 2 Averages of gas exchange parameters for plant functional groups occurring at pasture and primary forest ecosystems

	P-value	Pasture grass	Pasture sapling	Upper canopy tree	Upper canopy liana	Mid canopy tree	Understory tree
LMA, g m ⁻²	0.0001*	69.0 ± 1.1 (35)	97.4 ± 21.4 (4) ab	129.5 ± 21.7 (10) a	122.9 ± 42.1 (6) ab	89.5 ± 36.8 (31) b	55.8 ± 13.6 (9) c
A_{max} , $\mu\text{mol m}^{-2} \text{s}^{-1}$	0.03*	24.0 ± 1.0 (38)	11.5 ± 5.7 (4) ab	10.7 ± 4.3 (6) a	7.7 ± 1.1 (6) a	7.8 ± 2.4 (6) ab	4.6 ± 1.8 (6) b
V_{cmax} , $\mu\text{mol m}^{-2} \text{s}^{-1}$	0.02*	34.4 ± 2.6 (16)	86.3 ± 42.1 (4) ab	81.0 ± 29.5 (6) b	59.7 ± 14.3 (6) b	57.5 ± 16.4 (6) ab	31.6 ± 15.2 (6) a
J_{max} , $\mu\text{mol m}^{-2} \text{s}^{-1}$	0.0001*	158.5 ± 16.4 (8)	122.0 ± 57.0 (4) ab	112.0 ± 34.1 (6) b	87.5 ± 11.3 (6) b	81.5 ± 28.6 (6) b	37.9 ± 8.9 (6) a
R_d , $\mu\text{mol m}^{-2} \text{s}^{-1}$	0.0001*	0.94 ± 0.13 (11)	1.57 ± 0.34 (4) a	1.36 ± 0.90 (6) abc	1.25 ± 0.50 (6) ac	0.58 ± 0.25 (6) bc	0.30 ± 0.06 (6) b
$\delta^{13}C$, ‰	0.0001*	-12.6 ± 0.1 (40)	-30.4 ± 1.1 (4) abc	-29.9 ± 1.0 (11) b	-28.6 ± 0.9 (6) a	-32.2 ± 1.8 (46) c	-35.2 ± 0.7 (12) d
$\delta^{15}N$, ‰	0.06*	4.4 ± 0.5 (40)	5.5 ± 4.5 (4) ab	6.4 ± 1.5 (11) ab	7.8 ± 1.6 (6) a	6.1 ± 1.3 (46) ab	5.3 ± 1.0 (12) b
N_{area} , g m ⁻²	0.001	0.86 ± 0.02 (35)	2.11 ± 0.83 (4) abc	2.61 ± 0.68 (10) a	2.48 ± 0.66 (6) ab	1.84 ± 0.69 (31) b	1.33 ± 0.33 (9) c
c_i/c_a	0.003	0.30 ± 0.01 (37)	0.69 ± 0.03 (4) ab	0.67 ± 0.05 (6) b	0.61 ± 0.08 (6) b	0.67 ± 0.04 (6) ab	0.77 ± 0.07 (6) a
$g_s@A_{max}$, mol m ⁻² s ⁻¹	0.24	0.17 ± 0.01 (37)	0.23 ± 0.10 (4)	0.18 ± 0.10 (6)	0.11 ± 0.03 (6)	0.13 ± 0.05 (6)	0.11 ± 0.02 (6)
N_{mass} , mg g ⁻¹	0.76	12.7 ± 0.5 (40)	24.6 ± 14.8 (4)	21.2 ± 6.4 (11)	20.8 ± 3.2 (6)	22.6 ± 6.5 (46)	23.6 ± 7.4 (12)
C/N	0.71	33.7 ± 1.2 (40)	26.1 ± 14.4 (4)	25.2 ± 7.2 (11)	23.5 ± 3.8 (6)	23.1 ± 7.4 (46)	20.7 ± 7.2 (12)

Numbers are mean ± SD except for pasture grass group (mean ± SEM). Numbers within parentheses are sample size for the grass functional groups and the number of species involved for the other functional groups. Different letters in the same row indicate that means are statistically different (Games–Howell post hoc test; alpha = 0.1)

* indicates that a Welch-ANOVA was applied due to unequal variances

liana groups were different from each other based on $\delta^{13}\text{C}$ (Table 2). The establishment of more conspicuous differences among groups was hampered by large within group variances. For example, despite the 2-fold variation in mean $g_s@A_{\max}$ between the pasture sapling and the understory tree functional groups (Table 2), no statistical difference was detected for this parameter between these two groups.

Despite the lack of statistically significant distinctions between the pasture sapling group and both the top-canopy tree and the top-canopy liana groups, relationships among parameters suggest divergent physiological set points among them (Fig. 1). Based on N_{area} , LMA, and R_d , these groups were similar to each other (Fig. 1A and B), indicating that full sun-light leaves were acclimated to similar environmental conditions. When A_{\max} or $g_s@A_{\max}$ were considered the top-canopy liana group tended to resemble the mid canopy tree group (Fig. 1C). The fact that the top-canopy liana group tended to show higher $\delta^{13}\text{C}$ values and lower c_i/c_a values (Fig. 1D) suggests that this group might be more conservative in its water use, in agreement with relatively low stomatal conductances when compared to top-canopy trees.

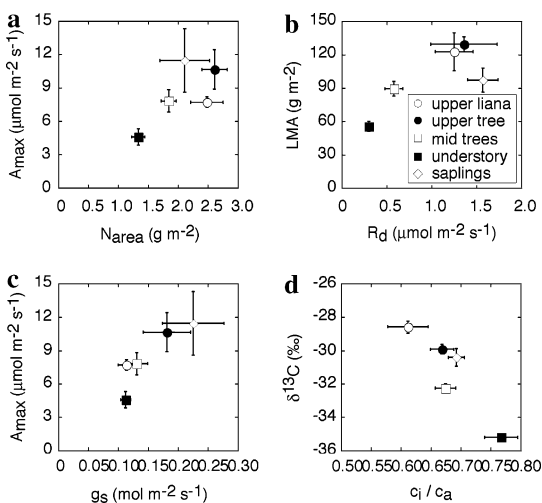


Fig. 1 The interrelationships among parameters associate with photosynthetic gas exchange for plant functional groups occurring at a primary tropical rain forest site and at a pasture site (Santarém, Brazil)

Photosynthetic efficiency

The trade-offs between carbon gain, transpirational water loss, nutrient use, and metabolic rates were expected to vary among the proposed functional groups. Trade-off settings of plant functional groups were compared in Table 2. The C_4 photosynthetic pathway of the pasture grass set this group apart as being highly efficient group in acquiring carbon while employing relatively low amounts of resources such as N and water. Further analyses were based on comparisons among groups with the C_3 photosynthetic pathway. The instantaneous water use efficiency ($A_{\max}/g_s@A_{\max}$) suggests homogeneity among functional groups, although the top-canopy liana group was significantly different from the understory tree group (Fig. 2). Despite large intergroup variations in both A_{\max} and N_{area} values (Table 2), the constancy observed for nitrogen use efficiency (A_{\max}/N_{area}) among the C_3 functional groups points to a strong interdependence between these two parameters, suggesting that N was allocated to match potential photosynthetic rates. The carbon assimilation efficiency (A_{\max}/R_d) tended to be similar among the groups evaluated indicating that changes in R_d were proportional to changes in A_{\max} , although the understory tree group showed higher efficiency when compared to the pasture sapling group.

Discriminant analyses

Based on species averages, linear discriminant analyses were applied to the dataset in order to test the ability of sets of ecophysiological parameters to reproduce the a priori plant groupings. The application of the discriminant analysis resulted in no misclassifications when LMA, $\delta^{13}\text{C}$, N_{area} , $g_s@A_{\max}$, A_{\max} , R_d , V_{cmax} , and J_{max} were considered. That was the smallest set of parameters needed for a 100% correct classification. The inclusion of other parameters (c_i/c_a , $\delta^{15}\text{N}$, N_{mass} , or C/N) did not improve or even decrease the classification efficiency. When the discriminant analysis was based solely on gas exchange parameters (hard traits according to Hodgson et al. 1999) (A_{\max} , V_{cmax} , J_{max} , $g_s@A_{\max}$, R_d , and c_i/c_a), the accuracy of species classification

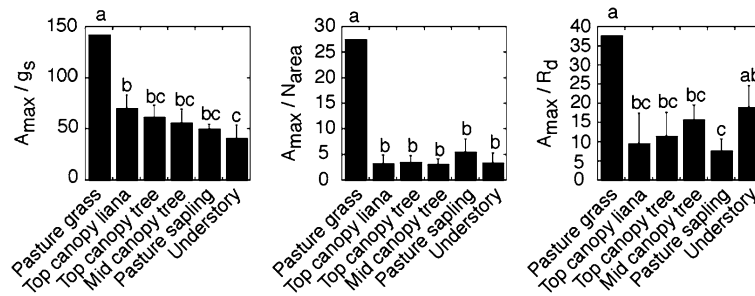


Fig. 2 Plant functional groups averages and standard error of the mean of instantaneous water use efficiency ($A_{max}/g_s@A_{max}$), photosynthetic nitrogen use efficiency

(A_{max}/N_{area}), and carbon use efficiency (A_{max}/R_d). Distinct letters indicates statistical significant differences in functional groups averages ($\alpha = 0.1$)

within the functional groups decreased to 76%. On the other hand, when the discriminant analysis was based on leaf anatomical and chemical properties (soft traits) (LMA, $\delta^{15}N$, $\delta^{13}C$, C/N, N_{mass} , and N_{area}), the precision of species classification within the functional groups decreased to 61%, although the precision improved slightly when only $\delta^{15}N$, $\delta^{13}C$, and N_{area} were used (67%). Such patterns suggests that both hard and soft traits are necessary in correctly characterizing plant functional types based on ecophysiological traits related to gas exchange.

Discussion

The averages and ranges of photosynthetic gas exchange parameters (A_{max} , V_{cmax} , J_{max} , $g_s@A_{max}$, R_d , and c_i/c_a) presented in this study were within values published for the Amazon region (Roberts et al. 1990; McWilliam et al. 1996; Sá et al. 1996; Carswell et al. 2000; Meir et al. 2001). In addition, averages and ranges of leaf chemical and anatomical parameters (LMA, $\delta^{13}C$, and N_{area}) were also in agreement with other Amazon tropical forest studies (Lloyd et al. 1995; Reich et al. 1991; Martinelli et al. 1998; Carswell et al. 2000; Ometto et al. 2002).

Differences among functional groups

The dataset presented in this study suggests large within-group variances (Table 2), similar to what Wright et al. (2005) observed from a much larger dataset. For example, the coefficient of variation

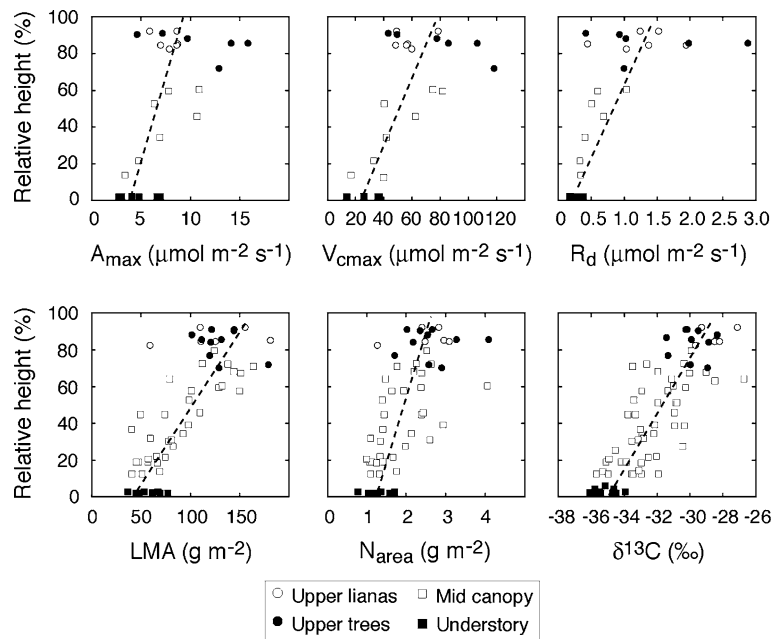
associated with A_{max} values averaged 35% among the functional groups. Nonetheless, significant differences among functional groups were detected indicating that our a priori functional group classification indeed mimic distinct ecophysiological characteristics. The exception to that was the absence of differences between the top-canopy tree and the pasture sapling groups, although the small sample size of pasture sapling ($n = 4$) associated with large variances might have impaired our ability to detect real differences between these two groups, if they indeed exist.

Differences in ecophysiological parameter means among the groups within the forest ecosystem came out to be more conspicuous. The structure of forest canopies causes strong gradients in environmental parameters such as light levels, wind, temperature, and relative humidity (Shuttleworth 1989; Domingues et al. 2005). The ecophysiological properties of forest leaves evaluated in this study were strongly associated with its relative position within the canopy profile (Fig. 3), causing understory species to be most different from upper canopy species.

The pasture grass displayed high photosynthetic efficiency (Fig. 2), resulting from the C_4 photosynthetic pathway (Percy and Ehleringer 1984). By increasing the CO_2 concentration at the sites of carboxylation, C_4 plants operate with lower c_i/c_a , lower g_s , and require lower amounts of the enzymes responsible for photosynthetic carbon fixation, therefore requiring less nitrogen.

While the pasture grass showed the highest water use efficiency, the understory group tended

Fig. 3 Relationships between leaf relative position within the canopy profile and parameters related to photosynthetic gas exchange for plant species occurring at a primary tropical rain forest site (Santarém, Brazil)



toward lower efficiency (Fig. 2). Understory plants possess low concentration of carboxylase enzyme, low light-saturated photosynthetic rates, low respiration rates, low light compensation point and generally slow growth (Denslow 1987). Such features are related to the low light levels prevailing at the forest floor (Leight 1975; Chazdon et al. 1996), and the use of sun-flecks has been shown to play an important role in daily carbon gain for species inhabiting the dark understory environment (Chazdon 1988). In order to minimize stomatal limitation of photosynthesis during ephemeral sun-flecks, understory plants usually operate with relative high stomatal conductances (Pearcy 1990), consequently resulting in low instantaneous water use efficiency. On the other hand, understory species tended to show high carbon use efficiency (Fig. 2) that is necessary to achieve a positive carbon balance at low light level that prevails at the forest floor.

The top-canopy liana group had a $\delta^{13}\text{C}$ mean significantly different from the top-canopy trees (Table 2), indicating that lianas tended to operate under lower c_i/c_a values. Although no other parameter was significantly different between these two groups, mean A_{\max} and $g_s@A_{\max}$ were lower in the top-canopy liana group when

compared to the top-canopy tree group (28 and 39% lower, respectively). Such patterns found in the top-canopy liana group were consistent with stomatal limitation of photosynthesis due to water stress. When compared to trees, lianas usually had a much greater ratio of leaf area per unit of conducting tissue (Putz 1983). During periods of high evaporative demand, such allometry might place lianas at a higher risk of xylem embolism, justifying a conservative use of water in this functional group.

Implications to global changes

Changes in atmospheric CO_2 levels over geological times have been related to major changes in fauna and flora (Cerling et al. 1998). Thus, the 33% rise in atmospheric CO_2 witnessed during the last 200 years is expected to interfere with ecological interactions (Drake et al. 1997). Increases in atmospheric CO_2 improve the quantum yield of C_3 photosynthesis (Ehleringer et al. 1997), decreasing the present-day advantage of C_4 species in hot climates, therefore decreasing Amazonian pasture productivity relative to forest plants. Furthermore, improvements in quantum yields might enhance the success rate of seedling

establishment at the very light-limited tropical forest understory, with implications for changing forest dynamics (Würth et al. 1998; Phillips et al. 2004).

Another implication of rising CO₂ levels is the consequential improvement in water use efficiency, simply because a lower g_s is required to meet the demand for photosynthetic assimilated carbon (Drake et al. 1997). The present dataset indicates that top-canopy lianas are more conservative in water use (Fig. 2). If improvements in water use efficiency translates into increased productivity for lianas, then it might help to explain the observed pan-tropical increase in liana density (Phillips et al. 2002). Due to its detrimental effects on tree growth, increasing liana density can potentially promote profound changes in ecosystem function, species composition and carbon, water and energy fluxes (Schnitzer and Bongers 2002).

Conclusions

Statistically significant differences based on ecophysiological parameters related to gas exchange were established among the plant functional groups that were identified a priori, with the exception that no differences were observed between the top-canopy tree and the pasture sapling groups. A combination of several leaf physiological (hard traits) and chemical-anatomical properties (soft traits) was necessary to reproduce the a priori grouping without misclassifications, reinforcing the relevance of collecting a broad range of traits. The ecophysiological differences reported for the plant functional groups evaluated in the present study provide important information necessary for the simplification of ecosystem scale studies in Amazônia and other tropical forest ecosystems.

Acknowledgments We are thankful to the assistance provided by our friends at the LBA-ECO Santarém regional office. We are also in debt to J. Ometto and A. Calil for a great work on sample analyses. Financial support for this work was provided partially by a research grant from NASA LBA-Ecology to J.R.E., L.B.F., and L.A.M and partially by a research grant from NASA LBA-Ecology to J.R.E., J.A.B., and L.A.M.

References

- Ackerly DD, Dudley SA, Sultan SE, Schmitt J, Coleman JS, Linder CR, Sandquist DR, et al (2000) The evolution of plant ecophysiological traits: recent advances and future directions. *BioScience* 50:979–995
- Carswell FE, Meir P, Wandelli EV, Bonates LCM, Kruijt B, Barbosa EM, Nobre AD, Grace J et al (2000) Photosynthetic capacity in a central Amazonian rain forest. *Tree Physiol* 20:179–186
- Cerling TE, Ehleringer JR, Harris JM (1998) Carbon dioxide starvation, the development of C₄ ecosystems, and mammalian evolution. *Philos Trans Roy Soc London B* 353:159–171
- Chazdon RL (1988) Sunflecks and their importance to forest understorey plants. In: Begon M, Fitter AH, Ford ED, MacFadyen A (eds) *Advances in ecological research*. Academic Press, San Diego, pp 1–63
- Chazdon RL, Pearcy RW, Lee DW, Fetcher N (1996) Photosynthetic responses of tropical forest plants to contrasting light environments. In: Mulkey SS, Kitajima K, Wright SJ (eds) *Tropical forest plant ecophysiology*. Chapman & Hall, New York, pp 5–55
- Cornelissen JHC, Lavorel S, Garnier E, Díaz S, Buchmann N, Gurvich DE, Reich PB, ter Steege H, Morgan HD, van der Heijden MGA, Pausas JG, Poorter H (2003) Handbook for protocols for standardised and easy measurements of plant functional traits worldwide. *Aust J Bot* 51:335–380
- da Rocha HR, Goulden ML, Miller SD, Menton MC, Pinto LDVO, de Freitas HC, Figueira AMS (2004) Seasonality of water and heat fluxes over a tropical forest in eastern Amazonia. *Ecol Appl* 14:22–32
- Dawson TE, Chapin FS III (1993) Grouping plants by their form-function characteristics as an avenue for simplification in scaling between leaves and landscapes. In: Ehleringer JR, Field CB (eds) *Scaling physiological processes Leaf to globe*. Academic Press, San Diego, pp 313–319
- Denslow JS (1987) Tropical rainforest gaps and tree species diversity. *Annu Rev Ecol Syst* 18:431–451
- Díaz S, Hodgson J, Thompson K, Cabido M, Cornelissen J et al (2004) The plant traits that drive ecosystems: evidence from three continents. *J Veget Sci* 15:295–304
- Domingues TF, Berry JA, Martinelli LA, Ometto JPHB, Ehleringer JR (2005) Parameterization of canopy structure and leaf-level gas exchange for an eastern Amazonian tropical rain forest (Tapajos National Forest, Para, Brazil). *Earth Interact* 9:17
- Drake BG, Gonzalez-Meler MA, Long SP (1997) More efficient plants: a consequence of rising atmospheric CO₂? *Annu Rev Plant Physiol Plant Mol Biol* 48:609–639
- Ehleringer JR, Björkman O (1977) Quantum yields for CO₂ uptake in C₃ and C₄ plants: dependence on temperature, CO₂ and O₂ concentration. *Plant Physiol* 59:86–90
- Ehleringer JR, Cerling TE, Helliker BR (1997) C₄ photosynthesis, atmospheric CO₂, and climate. *Oecologia* 112:285–299

- Evans JR (1989) Photosynthesis and nitrogen relationships in leaves of C_3 plants. *Oecologia* 78:9–19
- Farquhar GD, von Caemmerer S, Berry JA (1980) A biochemical model of photosynthetic CO_2 assimilation in leaves of C_3 species. *Planta* 149:78–90
- Farquhar GD, Sharkey TD (1982) Stomatal conductance and photosynthesis. *Annu Rev Plant Physiol Plant Mol Biol* 33:317–345
- Field CB, Mooney HA (1986) The photosynthesis-nitrogen relationship in wild plants. In: Givnish TJ (ed) *On the economy of plant form and function*. University Press, Cambridge, pp 25–55
- Goulden ML, Miller SD, da Rocha HR, Menton MC, de Freitas HC, Figueira AMES, de Souza CAD (2004) Diel and seasonal patterns of tropical forest CO_2 exchange. *Ecol Appl* 14:42–54
- Grime JP (2001) *Plant strategies, vegetation processes, and ecosystem properties*. Wiley, New York
- Hobbs RJ (1997) Can we use plant functional types to describe and predict responses to environmental changes? In: Smith TM, Shugart HH, Woodward FI (eds) *Plant functional types. Their relevance to ecosystem properties and global change*. University Press, Cambridge, pp 66–90
- Hodgson JG, Wilson PJ, Hunt R, Grime JP, Thompson K (1999) Allocating C-S-R plant functional types: a soft approach to a hard problem. *Oikos* 85:282–294
- Holdridge LR (1947) Determination of world plant formations from simple climatic data. *Science* 105:367–368
- Hubbell SP (2005) Neutral theory in community ecology and the hypothesis of functional equivalence. *Funct Ecol* 19:166–172
- Keller M, Palace M, Hurr G (2001) Biomass estimation in the Tapajos National Forest, Brazil. Examination of sampling and allometric uncertainties. *Forest Ecol Manage* 154:371–382
- Körner C (1993) Scaling from species to vegetation: the usefulness of functional groups. In: Schulze E-D, Mooney HA (eds) *Biodiversity and ecosystem function*. Springer-Verlag, Heidelberg, pp 117–140
- Leight EG Jr (1975) Structure and climate in tropical rain forest. *Annu Rev Ecol Syste* 6:67–85
- Lloyd J, Grace J, Miranda AC, Meir P, Wong SC, Miranda BS, Wright IR, Gash JHC et al (1995) A simple calibrated model of Amazon rainforest productivity based on leaf biochemical properties. *Plant Cell Environ* 18:1129–1145
- Loreau M, Naeem S, Inchausti P, Bengtsson J, Grime JP, Hector A, Hooper DU, Huston MA, Raffaelli D, Schmid B, Tilman D, Wardle DA (2001) Biodiversity and ecosystem functioning. Current knowledge and future challenges. *Science* 294:804–808
- Marshall B, Biscoe PV (1980) A model for C_3 leaves describing the dependence of net photosynthesis on irradiance. I. Derivation. *J Exp Bot* 31:29–39
- Martinelli LA, Almeida S, Brown IF, Moreira MZ, Victoria RL, Sternberg LSL, Ferreira CAC, Thomas WW (1998) Stable carbon isotope ratio of tree leaves, boles and fine litter in a tropical forest in Rondonia, Brazil. *Oecologia* 114:170–179
- McWilliam A-LC, Cabral OMR, Gomes BM, Esteves JL, Roberts JM (1996) Forest and pasture leaf-gas exchange in south-west Amazonia. In: Gash JHC, Nobre CA, Roberts JM, Victoria RL (eds) *Amazonian deforestation and climate*. John Wiley & Sons, West Sussex, pp 265–286
- Meir P, Grace J, Miranda AC (2001) Leaf respiration in two tropical rainforests: constraints on physiology by phosphorus, nitrogen and temperature. *Funct Ecol* 15:378–387
- Miller SD, Goulden ML, Menton MC, da Rocha HR, de Freitas HC, Figueira AMES, de Souza CAD (2004) Biometric and micrometeorological measurements of tropical forest carbon balance. *Ecol Appl* 14:114–126
- Nepstad DC, Moutinho P, Dias-Filho MB, Davidson E, Cardinot G, Markewitz D, Figueiredo R, Vianna N et al (2002) The effects of partial throughfall exclusion on canopy processes, aboveground production, and biogeochemistry of an Amazon forest. *J Geophys Res* 107(D20):8085, doi:10.1029/2001JD000360
- Ometto JPH, Flanagan LB, Martinelli LA, Moreira MZ, Higuchi N, Ehleringer JR (2002) Carbon isotope discrimination in forest and pasture ecosystems of the Amazon Basin, Brazil. *Global Biogeochem Cycles* 16(4):1109, doi:10.1029/2001GB001462
- Pearcy RW (1990) Sunflecks and photosynthesis in plant canopies. *Annu Rev Plant Physiol Plant Mol Biol* 41:421–453
- Pearcy RW, Ehleringer JR (1984) Comparative ecophysiology of C_3 and C_4 plants. *Plant Cell Environ* 7:1–13
- Peng C (2000) From static biogeographical model to dynamic global vegetation model: a global perspective on modelling vegetation dynamics. *Ecol Modell* 135:33–54
- Phillips OL, Vásquez Martínez R, Arroyo L, Baker TR, Killeen T, Lewis SL, Malhi Y, Mendoza et al (2002) Increasing dominance of large lianas in Amazonian forests. *Nature* 418:770–774
- Phillips OL, Baker TR, Arroyo L, Higuchi N, Killeen TJ, Laurance WF, Lewis SL, Lloyd J et al (2004) Pattern and process in Amazon tree turnover, 1976–2001. *Philos Trans Roy Soc London B* 359:381–407
- Putz FE (1983) Liana biomass and leaf area of a “Tierra Firme” forest in the Rio Negro Basin, Venezuela. *Biotropica* 15:185–189
- Raunkiaer C (1934) *The life forms of plants and statistical plant geography*. Clarendon Press, Oxford
- Reich PB, Walters MB, Ellsworth DS (1997) From tropics to tundra. Global convergence in plant functioning. *Proc Natl Acad Sci* 94:13730–13734
- Reich PB, Uhl C, Walters MB, Ellsworth DS (1991) Leaf lifespan as a determinant of leaf structure and function among 23 amazonian tree species. *Oecologia* 86:16–24
- Roberts J, Cabral OMR, de Aguiar LF (1990) Stomatal and boundary-layer conductances in an Amazonian Terra Firme Rain Forest. *J Appl Ecol* 27:336–353
- Sá TD de A, da Costa J de PR, Roberts JM (1996) Forest and pasture conductances in southern Pará, Amazonia. In: Gash JHC, Nobre CA, Roberts JM, Victoria RL (eds) *Amazonian deforestation and climate*. John Wiley & Sons, West Sussex, pp 241–264

- Sakai RK, Fitzjarrald DR, Moraes OLL, Staebler RM, Acevedo OC, Czikowsky MJ, da Silva R, Brait E et al (2004) Land-use change effects on local energy, water, and carbon balances in an Amazonian agricultural field. *Global Change Biol* 10:895–907
- Schnitzer SA, Bongers F (2002) The ecology of lianas and their role in forests. *Trends Ecol Evol* 17:223–230
- Shuttleworth WJ (1989) Micrometeorology of temperate and tropical forest. *Philos Trans Roy Soc London B* 324:299–334
- Silver WL, Neff J, McGroddy M, Veldkamp E, Keller M, Cosme R (2000) Effects of soil texture on below-ground carbon and nutrient storage in a lowland Amazonian forest ecosystem. *Ecosystems* 3:193–209
- Solbrig OT (1993) Plant traits and adaptative strategies: Their role in ecosystem function. In: Schulze E-D, Mooney HA (eds) *Biodiversity and ecosystem function*. Springer-Verlag, Heidelberg, pp 97–116
- Smith TM, Shugart HH, Woodward FI (1997) Plant functional types. Their relevance to ecosystem properties and global change. University Press, Cambridge
- Telles E de CC, de Camargo PB, Martinelli LA, Trumbore S, da Costa ES, Santos J, Higuchi N, Oliveira RC Jr (2003) Influence of soil texture on carbon dynamics and storage potential in tropical forest soils of Amazonia. *Global Biogeochem Cycles* 17(2):1040, doi:10.1029/2002GB001953
- Vieira S, de Camargo PB, Selhorst D, da Silva R, Hutyrá L, Chambers JQ, Brown IF, Higuchi N et al (2004) Forest structure and carbon dynamics in Amazonian tropical rain forests. *Oecologia* 140:468–479
- von Caemmerer S, Farquhar GD (1981) Some relationships between the biochemistry of photosynthesis and the gas exchange of leaves. *Planta* 153:376–387
- von Caemmerer S (2000) *Biochemical models of leaf photosynthesis*. CSIRO Publishing, Collingwood
- Weiher E, van der Werf A, Thompson K, Roderick M, Garnier E, Eriksson O (1999) Challenging Theophrastus: a common core list of plant traits for functional ecology. *J Veget Sci* 10:609–620
- Wright IJ, Reich PB, Cornelissen JHC, Falster DS, Garnier E, Hikosaka K, Lamont BB, Lee W et al (2005) Assessing the generality of global leaf trait relationships. *New Phytologist* 166:485–496
- Würth MKR, Winter K, Körner C (1998) In situ responses to elevated CO₂ in tropical forest understorey plants. *Funct Ecol* 12:886–895