

# Differential seedling growth response to soil resource availability among nine neotropical tree species

Christopher Baraloto\*<sup>†1</sup>, Damien Bonal<sup>†</sup> and Deborah E. Goldberg\*

\* Department of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, Michigan, USA

<sup>†</sup> UMR "Ecologie des Forêts de Guyane", INRA Kourou, French Guiana

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**Abstract:** Although the potential contribution to tropical tree species coexistence of niche differentiation along light gradients has received much attention, the degree to which species perform differentially along soil resource gradients remains unclear. To examine differential growth response to soil resources, we grew seedlings of nine tropical tree species at 6.0% of full sun for 12 mo in a factorial design of two soil types (clay and white sand), two phosphate fertilization treatments (control and addition of 100 mg P kg<sup>-1</sup>) and two watering treatments (field capacity and water limitation to one-third field capacity). Species differed markedly in biomass growth rate, but this hierarchy was almost completely conserved across all eight treatments. All species grew more slowly in sand than clay soils, and no species grew faster with phosphate additions. Only *Eperua grandiflora* and *E. falcata* showed significant growth increases in the absence of water limitation. Faster-growing species were characterized by high specific leaf area, high leaf allocation and high net assimilation rate but not lower root allocation. Slower-growing species exhibited greater plasticity in net assimilation rate, suggesting that tolerance of edaphic stress in these species is related more to stomatal control than to whole-plant carbon allocation. Although relative growth rate for biomass was correlated with both its physiological and morphological components, interspecific differences were best explained by differences in net assimilation rate across six of the eight treatments. A suite of traits including high assimilation and high specific leaf area maintains rapid growth rate of faster-growing species across a wide gradient of soil resources, but the lack of plasticity they exhibit may compromise their survival in the poorest soil environments.

**Key Words:** biomass allocation, *Dicorynia*, drought stress, *Eperua*, French Guiana, *Goupia*, *Jacaranda*, plasticity, *Qualea*, *Recordoxylon*, relative growth rate, *Sextonia*, soil phosphorus, specific leaf area, *Virola*

## INTRODUCTION

Relative growth rate (RGR) is one of the most widely used measurements of plant performance (Grime & Hunt 1974, Poorter & Lambers 1991), and differences among species are frequently described based on intrinsic growth potential, or maximum RGR for biomass accumulation (RGR<sub>max</sub>; Grime *et al.* 1997, Lambers & Poorter 1992, Reich *et al.* 1997). Species with high RGR<sub>max</sub> are often associated with early successional stages of terrestrial plant communities where light is not limiting (Grime *et al.* 1997, Reich *et al.* 1992, 1997), and some regional studies have also reported associations of faster-growing species with areas of higher soil nutrients (Meziane & Shipley

1999, Wright & Westoby 1999) or increased precipitation (Wright & Westoby 1999). Accordingly, RGR<sub>max</sub> has been suggested as a useful attribute for predicting species distributions and abundances across resource gradients (Grime *et al.* 1997) and for understanding the mechanisms maintaining coexistence in terrestrial plant communities (Huston 1994, Tilman 1988).

Two different sets of ideas have been proposed to explain how species differing in RGR<sub>max</sub> might coexist, and they make very different predictions for how species will differ in observed RGR (hereafter RGR<sub>obs</sub>) across environmental conditions. Some authors present data suggesting that species hierarchies for RGR<sub>obs</sub> shift across resource gradients due to trade-offs between traits leading to rapid growth at high resources (i.e. high RGR<sub>max</sub>) versus traits maintaining relatively fast growth even when resources are low (Meziane & Shipley 1999, Sack & Grubb 2001, 2003). Sack & Grubb (2001) propose that such differences in realized growth would permit

<sup>1</sup> Corresponding author. UMR Ecofog, BP 709, 97387 Kourou Cedex, French Guiana. Email: baraloto.c@kourou.cirad.fr

coexistence in spatially heterogeneous environments encompassing these gradients. A second group of hypotheses is based on an alternative trade-off between demographic parameters: traits leading to high growth rate (high  $RGR_{max}$ ) versus traits leading to high survival (Kitajima 1994, Kitajima & Bolker 2003, Latham 1992). The 'growth rate trade-offs' hypothesis predicts that the hierarchy of observed RGR at low resource availability differs from that observed at high resource availability (operationally defined as  $RGR_{max}$ ). However, the 'growth rate–mortality trade-off' hypothesis allows that the species hierarchy for  $RGR_{max}$  may be identical to that for  $RGR_{obs}$ , regardless of the resource level at which the plants are grown. To date most studies examining differential growth response among tropical woody species have focused on light gradients (Bloor & Grubb 2003, Poorter 1999). However, much of the evidence for significant habitat associations for tropical trees suggests specialization to soil resource conditions in a heterogeneous edaphic environment (Harms *et al.* 2001, Webb & Peart 2000). Only a few studies have examined soil nutrients or water availability (Burslem 1996, Gunatilleke *et al.* 1997) and to our knowledge the interaction between nutrients and water has been studied only in the dry forests of Mexico (Huante *et al.* 1995, 1998).

In addition to understanding what trade-offs exist for RGR in different environments, it is important to understand the physiological and morphological basis of those trade-offs and differences in ecological breadth of these parameters across resource gradients so that the generality of different kinds of trade-offs can be assessed. Interspecific differences in both  $RGR_{max}$  and  $RGR_{obs}$  can be explained in terms of differences in morphological and physiological components of growth rate (cf. Lambers & Poorter 1992, Poorter & Nagel 2000). In several communities, species with high  $RGR_{max}$  have been associated with high specific leaf area (SLA), high leaf to total biomass ratio (LMR), low root to total biomass ratio (RMR), high estimated net assimilation rate (NAR) and high photosynthetic rates ( $A_{max}$ ) (Reich *et al.* 1997, Wright & Westoby 1999).

However, many species exhibit shifts in at least some of these attributes when grown under reduced resource availability (Meziane & Shipley 1999). The specialization hypothesis (Taylor & Aarssen 1988) predicts that such plasticity in performance-related traits should be higher for genotypes or species that are specialists in habitats with higher resource availability (Chapin *et al.* 1993, Lortie & Aarssen 1996). High plasticity in morphological and physiological components of growth rate along light gradients appears to be characteristic of fast-growing tropical woody species (Poorter 1999, Valladares *et al.* 2000). However, the relationship between  $RGR_{max}$  and plasticity across soil resource gradients remains to be examined.

In this paper we present results of a shadehouse experiment using different forest soils to examine the growth response to soil resource availability of nine tropical tree species representing a large proportion of community-wide variation in seed and seedling attributes associated with  $RGR_{max}$  (Table 1). To explore the degree to which observed growth hierarchies among species differ from potential growth hierarchies ( $RGR_{max}$ ), as well as to separate the roles of particular soil resources from light resources in determining growth response, the experiment was conducted in a shadehouse where we could control resource levels and estimate growth potential under highly favourable conditions (high resource availability, no competition or natural enemies). We investigated differential response to two markedly contrasting soil types characteristic of lowland forests in the Guiana Shield, a brown ferrallitic clay soil characteristic of well-drained sites, and a white sand of podzolic origin. Of the many soil properties that might differentially influence growth on these soils, we chose to manipulate two factors that have been found to influence tropical tree seedling growth – soil moisture (Burslem 1996) and soil phosphate availability (Lawrence 2003, Raaimakers & Lambers 1996).

Specifically, we address four hypotheses related to interspecific differences in  $RGR_{max}$ ,  $RGR_{obs}$  and their underlying mechanisms: (1) Faster-growing species always grow faster; that is, species hierarchies for RGR do not shift across soil resource gradients; (2) Species with higher  $RGR_{max}$  are characterized by traits favouring growth, including higher SLA, lower RMR and higher NAR; (3) Species with higher  $RGR_{max}$  exhibit greater plasticity in morphological (SLA, LMR) and/or physiological (NAR) growth components; (4) The relative contributions of NAR, LAR and SLA to  $RGR_{obs}$  change along soil resource gradients. We discuss the consequences of these results for the distribution and abundance of these species across soil resource gradients in the field.

## MATERIALS AND METHODS

The experiment was conducted in a shadehouse at the INRA research facilities in Kourou, French Guiana, approximately 60 km from permanent plots at Paracou, French Guiana (5°18'N, 52°55'W). The nine focal species were chosen to test for differences among species not exhibiting any strong edaphic preferences at the seedling stage at Paracou (Baraloto & Goldberg 2004) but representing the broad range of seed size and specific leaf area found among the tree species occurring in lowland tropical forest in French Guiana (reported in Baraloto 2001; see Table 1). Seeds were collected from within a 10-m radius of each of a minimum of five adult trees per

species at Paracou and mixed with respect to maternal source.

The experiment consisted of a factorial design of two soil types, two water treatments and two phosphorus fertilization treatments. Soil from 5–30 cm depth was collected from areas within the Paracou forest representing the brown clay and white sand soil types commonly found in the lowland tropical forests of French Guiana. Soil was sieved to 2 mm to ensure homogeneity. Seeds of all species except those of *Eperua* were germinated in boxes filled with a 1:1 mixture of these soil types and 2–4-wk-old seedlings were planted in May 1999 into 6-litre black polyethylene containers filled with one of the two soil types. Seeds of *Eperua* were weighed and planted directly into the pots. A subset of 20–30 germinating seedlings of each species was harvested at the initiation of the experiment for use in growth analyses.

A transparent plastic cover was used to prevent rainfall from entering the shadehouse and two layers of neutral shade cloth were used to reduce light levels to a moderate light level where growth would not be limited (cf. Bloor & Grubb 2003). Quantum sensors (LICOR) recording data each minute to a data logger (Campbell Scientific) received maximum instantaneous energy levels of  $101.2 \mu\text{mol m}^{-2} \text{s}^{-1}$ , on a cloudless day, with daily integrated level of  $1.65 \text{ mol m}^{-2} \text{d}^{-1}$ . These values correspond to  $6.0\% \pm 1.2\%$  of energy transmitted to sensors placed in adjacent sites open to full sun.

For 12 wk from the planting of each species, all seedlings were kept at field capacity ( $0.25 \text{ m}^3 \text{ m}^{-3}$  in clay;  $0.14 \text{ m}^3 \text{ m}^{-3}$  in sand) with water added every 2–3 d. Subsequently, seedlings in the 'no drought' treatment continued to be watered in this manner, while seedlings in the water-limitation treatment received one-third of the water necessary to keep pots at saturation. The volume and frequency of watering was based on trials of ten pots filled with each soil type filled to saturation and weighed over a 2-wk period. This watering treatment simulated the drought stress experienced near the end of the dry season, when gravimetric moisture at 0–10 cm can reach as low as 7% in the white-sand sites, and 13% in the clay sites (Baraloto *et al.* 2005a). Volumetric moisture in each pot was measured using a Theta Probe (ML1-108, Delta-T Devices, Cambridge, UK) at the final harvest. The clay soil lost almost twice as much water after drought treatments than did the sand soil ( $F_{\text{tmt-soil}} = 185$ ;  $df = 1, 1076$ ;  $P < 0.001$ ).

A phosphate addition treatment was implemented to release half of the plants from any P limitation in the two soils. Based on Raaimakers & Lambers (1996), we made four additions totalling  $100 \text{ mg P kg}^{-1}$  soil, added as  $\text{KH}_2\text{PO}_4$  solutions into the pots after they had been watered. Both the white sand and the clay pots that received fertilizer contained more than  $13 \text{ mg kg}^{-1}$  of Olsen-extractable phosphate-P at the end of the experi-

ment, among the highest levels observed in surface cores in lowland forest in French Guiana (Baraloto & Goldberg 2004, Baraloto *et al.* 2005a).

Fifteen replicates of each species  $\times$  soil  $\times$  watering  $\times$  phosphate addition combination were arranged in a randomized block design, with five replicates within each of three blocks arranged perpendicular to daily solar movement ( $n = 120$  per species). Pots were placed at least 2.5 m from the shadehouse edge to avoid the potential effects of edge on light and microclimate (Brown & Jennings 1998).

The experiment was completely harvested after 12 mo. For each seedling, stem diameter at soil surface, height and leaf number were recorded, and seedlings were partitioned into leaf, stem and petiole, root and remaining cotyledons. Leaf surface area was measured immediately with a LICOR 3000 leaf area meter (LI-COR Inc, Lincoln, Nebraska, USA). All plant parts were then dried to constant mass at  $50^\circ\text{C}$  and weighed to a precision of 1 mg.

Based on these measurements, we calculated mass ratios of each individual for leaves (LMR), roots (RMR) and stems (SMR) as biomass fractions of each component proportional to total biomass (Hunt 1978). We also described leaf morphology for each individual by calculating specific leaf area (SLA) as the ratio of leaf area to leaf dry biomass, and leaf area ratio (LAR) as the ratio of leaf area to total dry biomass (Hunt 1978). We then determined RGR using the equation:

$$\text{RGR} = (\ln m_2 - \ln m_1) / (t_2 - t_1),$$

in which  $m$  is the total dry biomass (g), and the denominator represents the number of days between the initial planting date and harvest dates for each individual. For the smaller-seeded species ( $< 2 \text{ g}$  dry mass; all except *Eperua* spp.), we calculated initial biomass from 10–30 seedlings harvested at the time of planting. For these species, RGR was calculated using a randomized pairing (with replacement) of individuals at the final harvest with one of the 30 seedlings sampled at the time of planting; this approach is conservative as it maximizes the variance in the initial seedling size. However, for large-seeded species exhibiting high variability in seed mass (*Eperua* spp.), this approach is not feasible because initial seedling size can be highly positively correlated with seed mass (Baraloto *et al.* 2005b). For these species, we estimated initial seedling biomass on an individual basis using relationships between fresh seed mass and initial seedling biomass calculated for 20 individuals at the beginning of the experiment. We estimated  $\text{RGR}_{\text{max}}$  as the highest treatment mean of  $\text{RGR}_{\text{obs}}$  among the eight treatments.

We also estimated NAR using the equations described by Hunt (1978), after verifying the assumed linear relationships between leaf area and plant dry mass (see Poorter 1999). We used initial leaf surface areas

**Table 1.** Summary of seed and seedling morphological attributes for the nine focal species. Species were chosen to represent a gradient of seed mass, seedling morphology and specific leaf area among common species found on both clay and white sand soils at Paracou, French Guiana.

Species	Family	Seed dry mass (g) <sup>1</sup>	Cotyledon function	Seedling biomass (g) <sup>2</sup>	Seedling height (cm)	Seedling leaf area (cm <sup>2</sup> )	Specific leaf area (m <sup>2</sup> kg <sup>-1</sup> )
<i>Goupia glabra</i> Aubl.	Goupiaceae	0.0015	Foliar	1.34	20.6	395	59.9
<i>Jacaranda copaia</i> (Aubl.) D. Don	Bignoniaceae	0.0047	Foliar	2.40	16.4	331	29.3
<i>Qualea rosea</i> Aubl.	Vochysiaceae	0.096	Foliar	0.60	15.1	58.1	22.5
<i>Recordoxylon speciosum</i> (Benoist) Gazel ex Barneby	Caesalpiniaceae	0.21	Foliar	1.12	15.9	216	38.6
<i>Dicorynia guianensis</i> Amshoff	Caesalpiniaceae	0.35	Reserve	2.76	24.3	447	33.3
<i>Sextonia rubra</i> (Mez) van der Werff	Lauraceae	1.18	Reserve	2.67	22.8	264	20.2
<i>Virola michelii</i> Heckel	Myristicaceae	1.28	Reserve	2.82	25.7	242	20.9
<i>Eperua falcata</i> Aubl.	Caesalpiniaceae	4.53	Reserve	7.69	38.0	468	19.0
<i>Eperua grandiflora</i> (Aubl.) Benth.	Caesalpiniaceae	31.3	Reserve	20.43	49.5	635	13.0

<sup>1</sup> Dry seed mass corresponds to the mean of 30–50 groups of seeds for small-seeded species (<0.1 g), 30–50 seeds for species of intermediate size (<3 g), and the estimated dry weight for larger seeds planted in the experiments, for which wet mass was measured.

<sup>2</sup> Seedling data are the mean of eight seedlings at 1-y age in the reference treatment, i.e. clay with drought and no phosphate additions.

calculated from the same seedlings as those used in the RGR calculations at the beginning of the experiment (see Table 1).

We examined effects of treatments on these dependent variables using analyses of covariance (ANCOVAs) with soil type, watering regime, and phosphate fertilization (two levels each) as fixed effects. Species was also treated as a fixed effect so that statistical interactions between species and the soil treatments could be analysed. A block term was included in the analyses to account for error associated with different light levels in the shadehouse. In addition, the soil moisture value for each individual at harvest date was entered as a covariate in the models to account for finer-scale variation in climate conditions related to edge effects in the shadehouse. We also included the total biomass at the time of harvest as a covariate to account for the effects of size on response to treatments.

A major problem that occurs when measuring allocation patterns and growth rates concerns allometric changes that occur with ontogeny. Most woody plant species undergo changes in biomass allocation during transitions between juvenile stages (Coomes & Grubb 1998, Rincon & Huante 1997, Walters *et al.* 1993). As a result, estimates of plasticity in allocation across treatments may be inflated if they do not account for plant size. To account for this potential effect, we first looked for evidence of size-dependent biomass allocation by comparing linear and exponential regressions between two size components. For example, we determined allometric relationships between root and shoot biomass, using linear models fitted to log-transformed data ( $\ln(x+1)$ ) and between leaf area and total biomass (McConnaughay & Coleman 1999). We did not include a constant in these models so that the relationship was forced through the origin, thus accounting for allometric changes across even the smallest plant sizes. When size-dependent allocation was present, we accounted for plant size in our estimates of plasticity for that parameter using

a novel index calculated as the difference between the maximum and minimum slopes of the allometric models among the eight treatments.

To examine the contribution of interspecific changes in NAR, LMR and SLA to changes in RGR in response to resource supply, we calculated Pearson correlation coefficients between RGR and its components across the eight treatments, with Bonferroni-corrected probability values.

## RESULTS

### Differential growth response

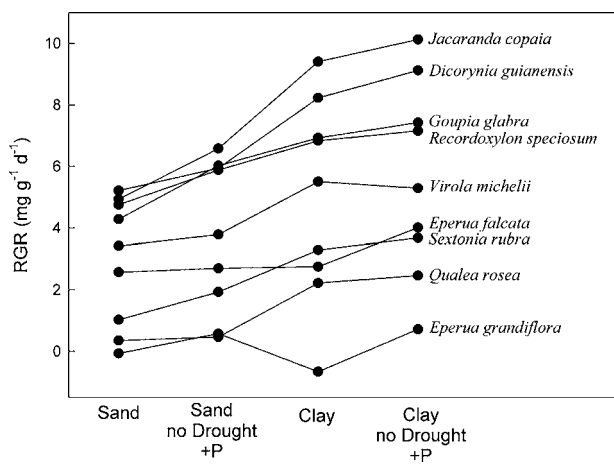
Biomass accumulation ( $RGR_{obs}$ ) differed strongly among species, and species also differed strongly in the magnitude of response of  $RGR_{obs}$  to treatments. In concert these factors explained about 90% of the variance for  $RGR_{obs}$  (Table 2). Similar results were found for growth in terms of seedling height and leaf area (data not shown). Despite the highly significant interactions between species and treatments (Table 2), in no case did the direction of the response differ among species. In fact, in only five of 64 pairwise comparisons did shifts occur in the species RGR hierarchy among treatments. Thus, the strong hierarchy for  $RGR_{max}$  (estimated from the clay soil with no drought and added P; Friedman's test = 51.3,  $P < 0.001$ ,  $n = 9$ ) was almost completely conserved across all eight treatments (Kendall's concordance = 0.92; Figure 1).

All species except *E. grandiflora* grew more slowly in the sand than clay soils, although the magnitude of this growth reduction was greatest for the faster-growing *Jacaranda* and *Dicorynia* (Figure 1). No species grew faster with phosphate-P additions in either soil type, and only *E. grandiflora* and *E. falcata* showed significantly faster growth in the absence of drought. *Eperua falcata* in particular responded to the complete soil

**Table 2.** Summary of analyses of covariance (ANCOVAs) for seedling growth and morphological parameters in the shadehouse experiment. Separate ANCOVAs were performed on each seedling parameter, with F-values and the corresponding probability levels reported. Data are from the final harvest of 873 individuals of nine species. Soil moisture, measured for each individual pot, and the total biomass of each individual at harvest, were entered into the models as covariates.  $RGR_{obs}$  = Relative growth rate for biomass, RMR and LMR represent total biomass proportion of roots and leaves, respectively. SLA is specific leaf area, or the surface area per unit mass of leaf blades, and was log-transformed for the analysis.

Factor (df)	$RGR_{obs}$	RMR	LMR	SLA
Total biomass at harvest (1 as covariate)	183***	0.80	7.12**	18.1***
Soil moisture (1 as covariate)	0.41	0.01	0.87	0.16
Block (2)	1.22	0.26	0.22	0.04
Species (8)	269***	47.1***	34.7***	227***
Soil type (1)	46.9***	0.99	351.3***	1.16
Watering regime (1)	4.09*	0.52	0.07	3.41
Phosphate addition (1)	0.01	0.01	16.9***	0.03
Species × Soil type (8)	23.1***	8.30***	17.5***	5.16***
Species × Water (8)	4.67***	5.86***	5.86***	2.86**
Species × Phosphate (8)	2.14*	2.26	4.17***	0.90
Soil type × Water (1)	2.24	0.14	7.83***	0.63
Soil type × Phosphate (1)	0.33	0.01	19.3***	0.59
Water × Phosphate (1)	0.01	0.04	0.03	0.01
Species × Soil type × Water (8)	2.45*	2.02	2.70**	5.75***
Species × Soil type × Phosphate (8)	1.41	1.87	2.92**	1.04
Species × Water × Phosphate (8)	0.49	1.85	2.24*	2.66*
Soil type × Water × Phosphate (1)	0.19	0.03	4.77*	0.23
Species × Soil type × Water × Phosphate (8)	1.53	2.48*	2.74**	1.62
Model $r^2$	0.89	0.73	0.77	0.93

\*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001.



**Figure 1.** Species hierarchies for relative growth rate (RGR) in contrasting soil resource conditions. Shown are the mean RGR ( $n = 7-15$ ) for each species in four treatments simulating natural field conditions versus non-limiting water (no drought) and increased phosphate availability (+P) in white sand and clay soils. Interactions between species and treatment are summarized in Table 2.

moisture gradient, growing fastest in the clay soil that was maintained at field capacity (Figure 1).

**Plasticity in components of growth rate**

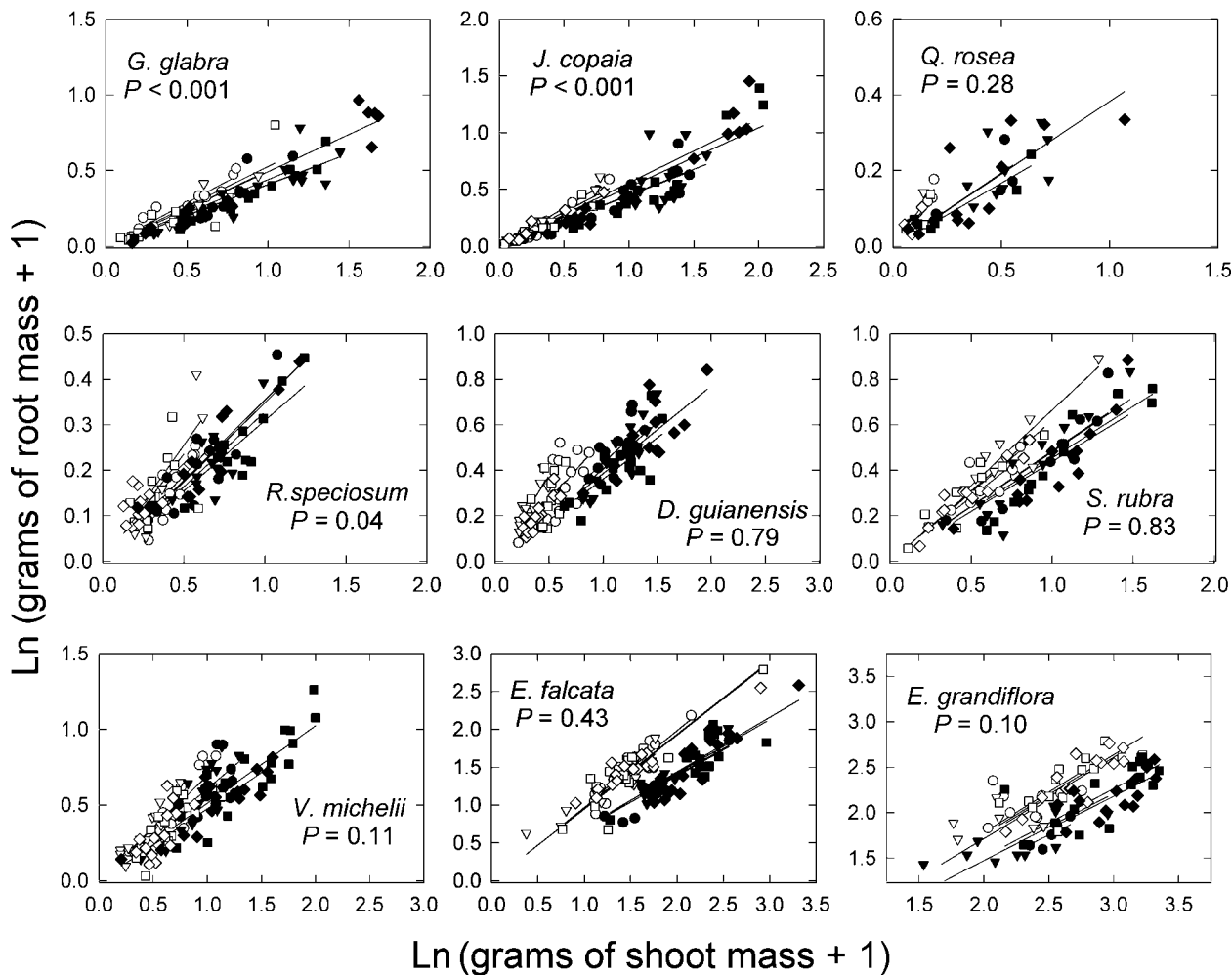
In general, larger seedlings allocated more to stems and less to leaves, with no significant trend regarding

**Table 3.** Pearson correlation coefficients, with Bonferroni probability values, between maximum relative growth rate ( $RGR_{max}$ ) and seed and seedling traits among nine species. Absolute values for traits were estimated using species means within the soil treatment that best represents average field conditions, the clay soil with no phosphate added but with simulated drought. Plasticity values were determined across all soil treatments as described in the text.

Trait	r	P
Dry seed mass	-0.68	0.04
Specific leaf area (SLA)	0.64	0.06
Leaf mass ratio (LMR)	0.73	0.04
Leaf area ratio (LAR)	0.65	0.06
Root-shoot ratio (RS)	-0.35	0.36
Net Assimilation Rate (NAR)	0.78	0.01
Plasticity in SLA	-0.15	0.70
Plasticity in LMR	-0.12	0.77
Plasticity in LAR	-0.16	0.68
Plasticity in RS	0.28	0.46
Plasticity in NAR	-0.77	0.02

root allocation (Table 2). In addition, larger seedlings had denser leaves (reduced SLA) and higher  $RGR_{obs}$ . Faster-growing species (those with higher  $RGR_{max}$ ) were generally characterized by high SLA, high LMR and high NAR but smaller seeds. Interestingly, however, faster-growing species did not have lower root allocation, as has often been found in other studies (Table 3).

Given these strong effects of size on morphology, we adjusted for variation in plant size to compare allocation



**Figure 2.** Allometric relationships between root and shoot biomass for individual seedlings of nine tropical tree species planted in eight soil resource treatments. Treatments on clay (filled) and sand (open) soils included a control simulating field conditions with drought (circles), phosphate addition (triangle), no drought (squares) and phosphate addition with no drought (diamonds). Differences among treatments for the slopes of linear regressions (i.e. root–shoot ratio) were tested with ANCOVA models, for which the probability values are shown. Note that the lines of regressions from multiple treatments are superimposed for some species.

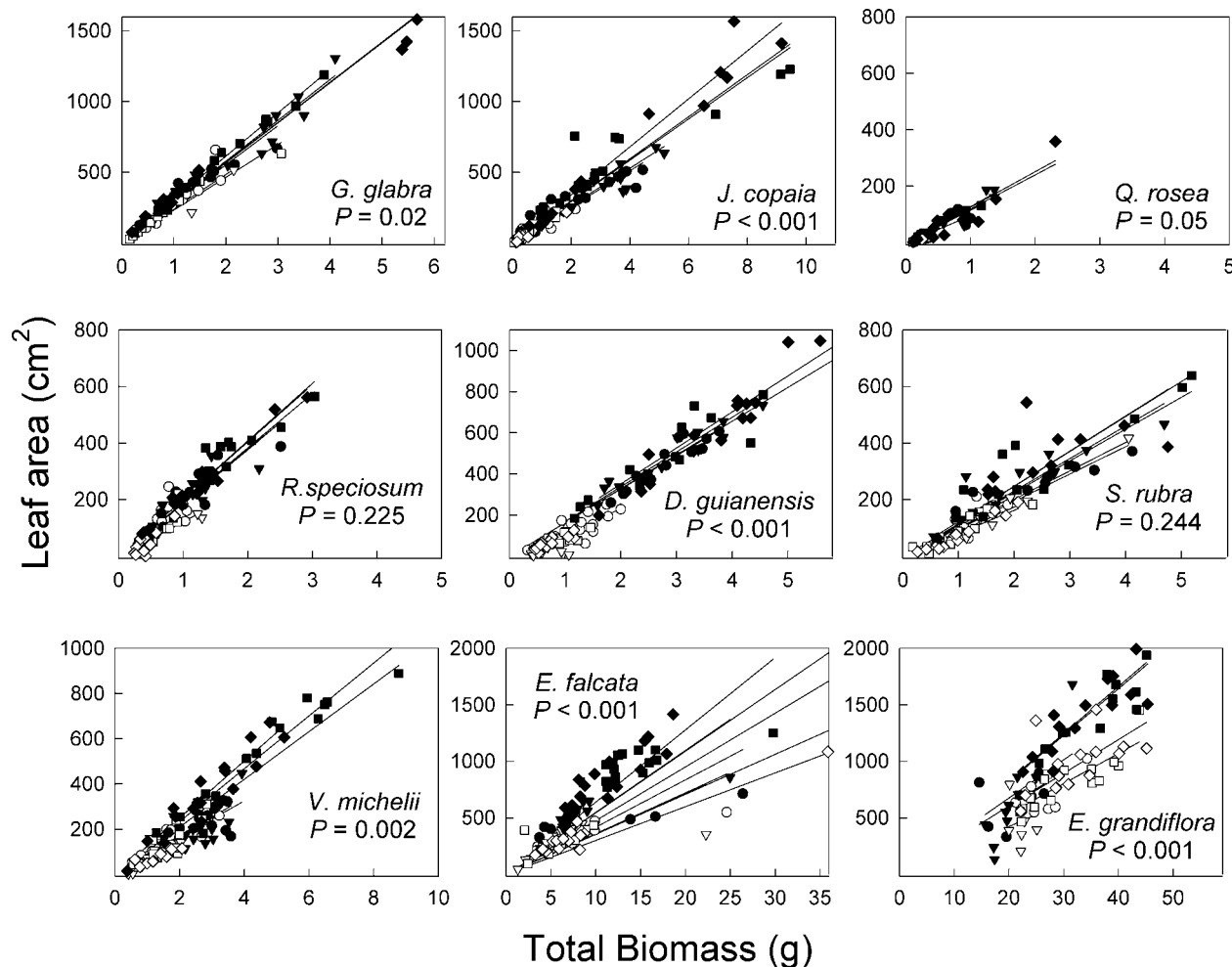
response to the treatments among species (Figures 2 and 3). Most species responded to sand soils by increasing root allocation and decreasing leaf area, irrespective of seedling size. However, the magnitude of shifts in allocation patterns, i.e. plasticity in allocation, differed among the nine species. We observed no clear pattern between plasticity in biomass allocation patterns and growth capacity. However, the slower-growing species appeared to be more plastic in physiological response to stress; plasticity in NAR was significantly negatively correlated with  $RGR_{max}$  (Table 3).

Although  $RGR_{max}$  was positively correlated with both its physiological (NAR) and morphological (LAR, SLA, LMR) components (Table 3), interspecific differences in  $RGR_{obs}$  were best explained by differences in NAR across six of the eight treatments (Table 4). In the two most

**Table 4.** Pearson correlation coefficients between relative growth rate ( $RGR_{obs}$ ) and its morphological (LAR, SLA, LMR) and physiological (NAR) components within each of the eight soil resource treatments. Bonferroni probabilities are indicated as \* $P < 0.05$ ; \*\* $P < 0.01$ .

Treatment	NAR	LAR	SLA	LMR
Sand	0.75	0.80*	0.79*	0.79*
Sand + P	0.80*	0.64	0.30	0.76*
Sand, no drought	0.76*	0.70	0.63	0.88**
Sand, no drought, + P	0.72	0.70	0.63	0.71
Clay	0.79*	0.81*	0.77*	0.68
Clay + P	0.84*	0.79*	0.65	0.65
Clay, no drought	0.80*	0.67	0.61	0.72
Clay, no drought, + P	0.66	0.67	0.63	0.68

stressful treatments, morphology was best correlated with  $RGR_{obs}$ , but it was LMR rather than SLA that best explained variation in  $RGR_{obs}$ .



**Figure 3.** Allometric relationships between leaf area and total biomass for individual seedlings of nine tropical tree species planted in eight soil resource treatments. Differences among treatments for the slopes of linear regressions (i.e. leaf area ratio) were tested with ANCOVA models, for which the probability values are shown. Note that the lines of regressions from multiple treatments are superimposed for some species. Treatment symbols are the same as in Figure 2.

## DISCUSSION

### Differential seedling growth across soil resource gradients

Although we did observe differences among species in their response to the soil treatments, the strong hierarchy we observed for RGR among species was almost completely conserved across all eight treatments. These data refute the RGR crossover hypothesis for seedling niche differentiation across soil resource gradients (Sack & Grubb 2001). One possible interpretation of our results is that soil resources were still somewhat limiting under the high-resource treatment, and so no shift in species performance hierarchies would be expected (Tilman 1988). The range of RGR we observed is indeed among the lower levels reported in the literature for pot studies of young tropical tree seedlings in light levels between

2% and 8% of full sun, which ranges from about 3–15 mg g<sup>-1</sup> d<sup>-1</sup> (Bloor & Grubb 2003, Kitajima 1994, Poorter 1999). Seedlings may have become limited by other mineral nutrients even in the relatively richer clay soil, where total N was only 0.92 g kg<sup>-1</sup>. Lawrence (2003) summarizes several studies in which mineral nutrients other than phosphorus, including magnesium and calcium, have been found to limit tropical tree seedling growth. However, the conditions represented in the experiment encompass almost the entire gradient of soil resources found in the forest from which both seeds and soil were obtained (Baraloto *et al.* 2005a), so it does constitute the relevant gradient for considering the role of trade-offs in this system.

Although pot experiments in shadehouses such as ours are useful for isolating particular factors under controlled conditions, they have been criticized because

they may not be a reliable guide to processes in the field, where interactions with other organisms such as herbivores might explain differential performance on different soil types (Fine *et al.* 2004). However, our results are consistent with those from a complementary field experiment with the same species, in which species rankings for height growth did not change among four soil types representing the gradient of conditions measured in this experiment (Baraloto *et al.* 2005a). Still, we warn against the interpretation that soil resource availability exerts little influence on the distribution of these or other tropical tree species across edaphic gradients, because we manipulated soil resource treatments at only one light level, and species rankings for growth may change in response to interactions between light and soil conditions (Huante *et al.* 1998). In particular, shifts in RGR along moisture gradients may be more likely to occur below the light compensation point (about 2% of full sun PAR for many species; Poorter 1999) beyond which stomatal conductance for water vapour may limit carbon assimilation (Bloor & Grubb 2003). Nonetheless, we note that in the complementary field experiment species rankings for height growth did not change among soil types either within or between plots with contrasting light availability (understorey plots receiving 1% daylight and gap plots receiving about 9% of daylight), but species with higher survival in resource-poor microhabitats tended to grow slower in resource-rich microhabitats (Baraloto *et al.* 2005a).

We did not observe any evidence for strong interactions among soil resource factors studied in this experiment. When soil moisture levels were accounted for, only one species (*E. falcata*) grew faster in the clay soil than the sand soil in the absence of drought. Furthermore, our data suggest that the degree to which seedling performance of these species is limited by low phosphate availability must be questioned. None of the species we studied exhibited any evidence for phosphate limitation. The lack of response for the faster-growing, smaller-seeded *G. glabra* and *J. copaia* is inconsistent with studies from other tropical forests, where small-seeded, early successional species often grow faster with increasing phosphate supply rates (Burslem *et al.* 1995, Grubb *et al.* 1994, Gunatilleke *et al.* 1997, Huante *et al.* 1995, Perrejin 2002, Raaimakers & Lambers 1996). An important intervening factor may be the role of soil symbionts such as mycorrhizas. However, spores of fungi associated with arbuscular mycorrhizas were found in samples from all of the treatments (data not shown). We also assayed root colonization at the final harvest for *D. guianensis* and *E. falcata*. Unlike *D. guianensis*, *E. falcata* does not exhibit growth responses to experimental mycorrhizal inoculation, but will form mycorrhizal structures under a variety of conditions (de Grandcourt *et al.* 2003). In our assays, we found no differential pattern of

mycorrhizal colonization among treatments for either species although seedlings were colonized, suggesting that the treatment effects we observed were not influenced by mycorrhizal status.

### Morphological and physiological plasticity in components of RGR

Our results concur with those from other plant communities regarding some of the basic allocation patterns among plants. The larger-seeded species studied here produce larger initial seedlings both in height as well as weight (Table 1), as has been described elsewhere (Baraloto *et al.* 2005b, Bloor & Grubb 2003, Osunkoya *et al.* 1994, Silman 1996). In addition, the largest-seeded species grew most slowly, as has been found in other communities (Maranon & Grubb 1993, Swanborough & Westoby 1996). As predicted, the faster-growing species exhibited many attributes favouring rapid growth, including higher SLA, LMR and NAR. However, we did not find the predicted negative correlation between fast growth and allocation to roots. Theory predicts that species better adapted to limiting soil resources should allocate more biomass to roots to better exploit the limited resource supply (Tilman 1988). Indeed, higher root allocation is positively correlated with rankings of species performance under low soil resource conditions in grasslands (Tilman & Wedin 1991, Wilson & Tilman 1991) and heathlands (Berendse *et al.* 1992). Higher root allocation did characterize the two larger-seeded *Eperua* species, both of which exhibit low RGR<sub>max</sub>, but we observed no consistent pattern among the other species. We note that this lack of differences may not reflect what actually occurs in the field because our experiment was conducted in pots in which the development of roots is not natural. Still, our results suggest that tolerance of limiting soil resources among tropical tree seedlings may not always be explained by increased root allocation.

Our data refute the hypothesis that faster-growing species are more plastic in the components of RGR. We did not find any relationship between RGR<sub>max</sub> and the plasticity of morphological components of RGR (SLA, LMF, LAR; Table 3). However, because RGR<sub>max</sub> does not appear to explain specialization to particular soil types among these species, our results do not necessarily refute the specialization hypothesis (Lortie & Aarssen 1996). Tropical woody species specializing in high-light environments (gap species) do tend to have higher RGR<sub>max</sub> and higher plasticity in both morphological and physiological parameters underlying RGR (Poorter 1999, Valladares *et al.* 2000). In contrast, we found a significant negative relationship between physiological plasticity (in NAR) across soil resource gradients and RGR<sub>max</sub>. Plasticity in NAR has been suggested to be associated



with plants adapted to stressful soil environments in which control of water loss contributes to survival during periodic drought (Grime & Mackey 2002). In a study on three tropical rain-forest tree species, Bonal & Guehl (2001) observed that differences among species in tolerance to soil drought could be explained by plant-level ecophysiological characteristics. In their study, species with high maximum rates of carbon assimilation were associated with greater sensitivity to water limitation. *Eperua falcata* in particular was found to exhibit very high WUE at the leaf level as well as the whole plant level, in addition to low sensitivity to water limitation.

Our results thus suggest that the ability to regulate water loss versus carbon gain may be of particular importance across soil type gradients. Conflicting evidence exists concerning the contribution of LAR and NAR to  $RGR_{obs}$  across resource gradients. Among species characteristic of open environments, LAR contributes more strongly to  $RGR_{obs}$  than does NAR, often because of responses in SLA (Meziane & Shipley 1999, Poorter & Remkes 1990). For tropical tree seedlings in the forest understorey, where light availability is generally less than 2% of full sun, control of stomatal conductance may be vital to maintaining a positive carbon balance (Bloor & Grubb 2003). Accordingly, several studies have found that NAR explains the most variation in  $RGR_{obs}$  under lower-light conditions (Bloor & Grubb 2003, Osunkoya *et al.* 1994, Popma & Bongers 1988). In contrast, Poorter (1999) found NAR to become important at higher light levels, whereas LAR explained most variation in  $RGR_{obs}$  at low light levels. In our experiment, NAR best explained between-species differences in  $RGR_{obs}$  in most soil treatments, suggesting the importance of leaf respiration for these species even at light levels characteristic of small canopy gaps. Still, in the most stressful conditions, LAR played a more prominent role, and not because of changes in SLA, perhaps because increased allocation to roots helped to buffer growth decreases.

### Implications for species distributions and coexistence

Several types of ecological trade-offs may contribute to differential species distributions across environmental gradients and thereby promote regional coexistence, including response to low versus high levels of a resource, different types of resources (soil vs. light) and different types of performance response (growth vs. survival) (Baraloto *et al.* 2005a). Although we found no evidence for a trade-off involving microhabitat partitioning for seedling growth along soil resource gradients, our results are consistent with a potential trade-off between growth and survival across these gradients, similar to that described for shade tolerance by Latham (1992), Kitajima

(1994) and Walters & Reich (2000). The faster-growing species in our study possess a suite of traits that appear to be rather canalized in their developmental expression; i.e. they exhibit little plasticity. While this suite of traits clearly permits them to grow faster across wide environmental gradients, it may compromise their survival, particularly in resource-poor environments. Evidence from the complementary field experiment supports this predicted trade-off between high survival when resources are limiting and fast growth when resources are less limiting (Baraloto *et al.* 2005a). The slower-growing species in our study appear to tolerate soil resource stresses because they are more plastic in assimilation rates, not because they have higher allocation to roots or are more plastic in this allocation. We suggest that future work should focus on the ecophysiological mechanisms underlying growth-survival trade-offs, such as leaf or stem hydraulic conductance versus photosynthetic capacity and/or structural or chemical defences, especially across environments that include interactions between light and soil resource gradients (Sack & Grubb 2002).

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