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Temperate rain forest species partition fine-scale gradients in light availability based on their leaf mass per area (LMA)

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● **Background and Aims** Ecologists are increasingly using plant functional traits to predict community assembly, but few studies have linked functional traits to species' responses to fine-scale resource gradients. In this study, it was tested whether saplings of woody species partition fine-scale gradients in light availability based on their leaf mass per area (LMA) in three temperate rain forests and one Mediterranean forest in southern Chile.
● **Methods** LMA was measured under field conditions of all woody species contained in approx. 60 plots of 2 m^2 in each site, and light availability, computed as the gap light index (GLI), was determined. For each site, species' pairwise differences in mean LMA (Δ LMA) and abundance-weighted mean GLI (Δ light response) of 2 m^2 plots were calculated and it was tested whether they were positively related using Mantel tests, i.e. if species with different LMA values differed in their response to light availability. Additionally linear models were fitted to the relationship between plot-level mean LMA and GLI across plots for each site.
● **Key Results** A positive and significant relationship was found between species' pairwise differences in mean LMA and differences in light response across species for all temperate rain forests, but not for the Mediterranean forest. The results also indicated a significant positive interspecific link between LMA and light availability for all forests. This is in contrast to what is traditionally reported and to expectations from the leaf economics spectrum.
● **Conclusions** In environments subjected to light limitation, interspecific differences in a leaf trait (LMA) can explain the fine-scale partitioning of light availability gradients by woody plant species. This niche partitioning potentially facilitates species coexistence at the within-community level. The high frequency of evergreen shade-intolerant species in these forests may explain the positive correlation between light availability and LMA.

Key words: Community assembly, fine-scale environmental gradients, Mediterranean forest, Patagonia, plant functional traits, species coexistence, temperate rain forest, trait-based approach.

INTRODUCTION

Plant community ecology has witnessed a major transition in recent years, with much effort placed on gaining a deeper understanding of how ecophysiological processes influence the assembly of ecological communities (e.g. Westoby *et al*., 2002; McGill *et al*., 2006; Adler *et al*., 2013). In particular, the quantification of ecological strategies through the measurement of plant functional traits, which capture key aspects of ecophysiology, has gained momentum (e.g. Grime, 1977; Lavorel *et al*., 2007), mainly because the use of traits represents a more synthetic approach to explain and predict ecological processes than the species-based approach (Adler *et al*., 2013; Shipley *et al*., 2016). The premise of the trait-based approach is that by considering the distributions of trait values within and among communities we will gain insights into the ecological processes governing their assembly. The trait-based approach in plant ecology has proven useful for classifying organisms into functional groups (e.g. Lavorel *et al*., 1997; Díaz and Cabido, 2001; Violle *et al*., 2007), quantifying the functional diversity of communities (e.g. Shipley *et al*., 2006; Cornwell and Ackerly, 2009) and understanding how vegetation properties and

composition change along geographical (e.g. Díaz and Cabido, 2001; McGill *et al*., 2006; Westoby and Wright, 2006; Reich *et al*., 2014) and temporal gradients (e.g. Shipley *et al*., 2006; Fajardo and Siefert, 2016). Although there is extensive literature linking functional traits to species performance and community assembly (e.g. compiled in Reich, 2014), there is still limited direct evidence of how traits mediate community assembly and species coexistence at the within-community level in natural field settings (HilleRisLambers *et al*., 2012; Adler *et al*., 2013; Reich, 2014; Sterck *et al*., 2014; Kraft *et al*., 2015).

The shift of focus in the analysis of community assembly from species identity to functional traits can lead to a more predictive community ecology and to a greater ability to generalize results across organisms and systems (McGill *et al*., 2006; HilleRisLambers *et al*., 2012; Adler *et al*., 2013; Shipley *et al*., 2016). Existing trait-based approaches seek to infer the effects of ecological processes by analysing distributions of functional traits of plant communities (Kraft *et al*., 2008; Swenson and Enquist, 2009; Uriarte *et al*., 2010). However, this phenomenological approach offers limited insights into the processes that maintain diversity (Adler *et al*., 2013), chiefly because these

trait-dispersion analyses do not identify the mechanisms generating niche partitioning. Adler *et al.* (2013), alternatively, proposed trait-based tests of coexistence built on recognized coexistence mechanisms – spatial and temporal heterogeneity, resource partitioning and frequency-dependent predation (Chesson, 2000) – to assess more rigorously whether functional traits are indeed mediating species coexistence. The idea is to find out whether the stabilizing niche differences necessary for coexistence are ultimately linked to species' functional traits. Only when traits are linked to coexistence mechanisms will we be able to make the trait-based approach for community assembly truly predictive.

Spatial heterogeneity in resource availability has been proposed as an important coexistence mechanism that may be strongly linked with functional traits (Adler *et al.*, 2013). For functional trait differences to promote coexistence via the spatial heterogeneity mechanism, species with different traits need to be favoured in different local environments. Previous studies have shown that species' leaf traits in a given community vary significantly across light environments (e.g. Lusk and Reich, 2000; Baltzer and Thomas, 2007; Sterck *et al.*, 2014). In this study, we show how a widely used functional trait, leaf mass per unit area (LMA), relates to spatial variation in one resource: light. If partitioning of spatial gradients in resource availability can effectively reduce niche overlap and promote coexistence, then species with different LMA values should be favoured in different local light availability conditions. We therefore predicted a positive relationship between species' pairwise differences in LMA and differences in their response to fine-scale gradients in light availability. To test this prediction, we took advantage of the natural variation in light availability found in the understorey of temperate rain forests of southern Chile. We concentrated on early life stages (i.e. saplings), because it is known that adaptations to the regeneration niche, encompassing the environmental conditions encountered in the early phases of a plant's life cycle, play a central role in coexistence of woody species (Grubb, 1977; Poorter, 2007), and also because regeneration traits in general have been much overlooked in the trait-based approach for community assembly models (Larson and Funk, 2016). We chose LMA – the leaf dry investment per unit of light-intercepting leaf area deployed (Poorter *et al.*, 2009) – because it is a trait intimately related to light availability (and therefore carbon investment), reflecting different light acquisition strategies (Wright *et al.*, 2004; Pérez-Harguindeguy *et al.*, 2013). Leaf functional traits including LMA have also been shown to be closely linked to species' regeneration niches (Grubb, 1977; Poorter, 2007). In temperate rain forests, species with resource-conservative leaf economics trait values, such as high LMA, long leaf life span and low leaf nitrogen and phosphorus concentrations, are generally favoured in sites with lower light availability. In contrast, species with resource-acquisitive leaf economics traits (low LMA, short leaf life span, etc.) tend to be favoured in sites with higher light availability (e.g. Walters and Reich, 1999; Lusk, 2002; Wright *et al.*, 2004). Thus, according to previous studies (e.g. Walters and Reich, 1999; Lusk, 2002; Wright *et al.*, 2004; Poorter and Bongers, 2006) and the leaf economics spectrum (Wright *et al.*, 2004; Reich, 2014), we expected that LMA should decrease across microsites with increasing light availability within forest communities.

MATERIALS AND METHODS

Research sites

We selected four second-growth forests – three temperate rain forests and one Mediterranean forest – in southern Chile. The first temperate rain forest site, Exploradores (EXP), was located in the Región de Aysén (46°29′S, 73°12′W, 75 m a.s.l.). The forest is on moraine deposits of glacial erosion originating from the Exploradores Glacier (Aniya *et al.*, 2011). The dominant tree species are *Drimys winteri* (Winteraceae), *Embothrium coccineum* (Proteaceae), *Amomyrtus luma* (Myrtaceae) and *Raukaua laetevirens* (Araliaceae) (Fajardo and Piper, 2015). The second site was located in Aiken Private Park (AIK) in Región de Aysén (45°27′S, 72°45′W, 40 m a.s.l) (Fig. 1). Soils at the site are typical Andisols, with acid pH, and low nitrogen and phosphorus availability. The dominant tree species are *Laureliopsis philippiana* (Atherospermaceae), *Myrceugenia planipes* (Myrtaceae), *Fuchsia magellanica* (Onagraceae) and *Azara lanceolata* (Flacourtiaceae). The third site was located in Katalapi Private Park (KAT), Región de Los Lagos (41°31′S, 72°45′W, 59 m a.s.l), near the Cordillera de Quillaipe in the Andean foothills. The dominant tree species are *L. philippiana*, *Aextoxicon punctatum* (Aextoxicaceae), *Eucryphia cordifolia* (Cunoniaceae) and *A. luma*. The climate of the temperate rain forest sites is cold-temperate and super-humid (Luebert and Pliscoff, 2006), with mean annual temperature of approx. $10\,°C$ and annual precipitation of $\geq 3000\,mm$ ($2322\,mm$ at Katalapi), regularly distributed throughout the year. The Mediterranean forest site was located in Los Ruiles National Reserve (RUI) in Región del Maule (35°50′S, 72°30′W, 350 m a.s.l.). The mean temperatures for the year and the growing season are $15{\cdot}2$ and $20{\cdot}3\,°C$, respectively; mean annual precipitation is $642\,mm$ and falls mostly in winter, with summer growing season precipitation of approx. $58\,mm$ (Luebert and Pliscoff, 2006). The study area belongs to the Coastal Range, and the soils at the study site are Alfisols, originated from granitic parental material, with neutral pH and poor nutrient availability. The dominant tree species are the winter deciduous *Nothofagus glauca* (Nothofagaceae), *Gevuina avellana* (Proteaceae), *Lithraea caustica* (Anacardiaceae) and *Cryptocarya alba* (Lauraceae) (Table 1). While light is expected to be a limiting factor controlling assembly of tree communities in the temperate rain forest sites, it is probably much less limiting than drought in Mediterranean forests. We therefore included a Mediterranean forest site to serve as a counterexample (a negative control) for testing our hypotheses relating to light limitation. All the sites were either remote in location or under conservation (National or privates reserves) and thus had a minimum degree of human disturbance. All temperate rain forest sites are very similar in composition and are composed of only two deciduous species, *Ribes magellanicum* (Saxifragaceae) and *Nothofagus antarctica* (Nothofagaceae). These temperate rain forests are also called cold-temperate rain forests (Veblen and Schlegel, 1982) because of the low temperatures throughout the year; mean

Fig. 1. Depiction of the temperate rain forest understorey light availability in Aiken Park (45°27′S, 72°45′W, 40 m a.s.l), Aysén Region, Chile.

temperature differences between gap and shade conditions in Aiken and Exploradores forest sites do not surpass 2 °C (F. Piper, unpubl. res.).

Sampling, tissue collection and processing

Sampling was conducted in February and March 2014 and 2015. At each forest site, we located at least 60 sampling points at 10–30 m intervals using a random walk design (Underwood, 1997). In more detail, we detoured from hiking trials and established a first sampling point at least 10 m away from the trial. Then a list of 10–15 sampling point co-ordinates was created using a random walk design with a random start point, where bearing directions (constrained to 180°) and distances (constrained from 10 to 30 m) were randomly generated. In this way, we took advantage of natural variation in light availability, and we were able to cover a significant range of light environments, including deep shade understoreys and large canopy gaps. We set a 2 m^2 circular plot at the centre of each sampling point and counted all saplings contained in the plot. Saplings were considered all woody plants with a height of 30–100 cm. Although most woody species were tree species, some shrub species also occurred, particularly at the Los Ruiles site. We note that by covering a relatively wide range of sapling sizes (i.e. approx. 30–100 cm) we combined data across many years, thus integrating annual variation in seed production, dispersal, herbivory and other ecological processes. Thus, our sampling – although a snapshot – integrates final survival (relative abundance at a specific light availability microsite) as a representative outcome of all the ecological processes mentioned before. Species cover was estimated by two observers through visual estimation of the percentage of area covered by each species in the plot. The classification was as follows: species cover class 1, <1 % of the plot area; class 2, 1–5 %; class 3, >5–15 %; class 4, >15–25 %; class 5, >25–50 %; class 6, >50–75 %; and class 6, >75 % of the plot area. Given that in some species LMA will show ontogenetic variation within a given light environment, we collected only leaves from the upper portion (no self-shading leaves) of the crown of one or two sapling individuals of each species at each 2 m^2 plot. We restricted the tissue collection to foliage without browsing or other damage. We note here that we did not fully follow the sampling protocol recommended by Pérez-Harguindeguy *et al.* (2013), because we worked with saplings and intentionally considered leaves under shade. Leaves of each species were placed in plastic bags, and stored in a cooler for transportation. In the laboratory, leaves were laid flat and photographed with a reference square of known area, and total projected leaf area was calculated using SIGMAPROC image-processing software (Systat Software Inc., Richmond, CA, USA). All leaves were then dried at 70 °C for 72 h and subsequently weighed. We computed leaf mass per area (LMA, g m^{-2}) as the dry mass of a leaf divided by its one-sided surface area. Because LMA values were log-normally distributed, we conducted all analyses using log-transformed values.

To characterize canopy openness and the light environment, we took digital hemispherical photographs of the canopy from a height of approx. 50 cm above the centre of each plot. We used a 7 mm Nikon f 7·4 fisheye lens (the lens has an orthographic projection of 180° angle of view), mounted on a Nikon Coolpix 5000 digital camera (Nikon Corporation, Tokyo, Japan). Photographs were taken under cloudy sky conditions or

TABLE 1. *List of sapling woody species and their respective plot-level weighted gap light index (GLI) and mean values of leaf mass per area (LMA, g m^{-2}) across three temperate rain forests (Aiken, AK; Exploradores, EX; and Katalapi, KA) and one Mediterranean forest (Los Ruiles, RU) in southern Chile*

Species	Family	n	Mean GLI (range)	Mean LMA (s.d.)	Forest
Ovidia andina	Thymelaeaceae	2	11·34 (10·31–12·36)	35·09 (12·49)	EX
Myrceugenia ovata	Myrtaceae	4	11·37 (4·50–19·68)	134·65 (50·91)	RU
Laureliopsis philippiana	Atherospermaceae	52	11·88 (7·13–62·14)	49·66 (26·67)	AK, EX, KA
Myrceugenia planiples	Myrtaceae	74	12·26 (2·53–62·14)	73·46 (22·37)	AK, EX, KA
Rhaphithamnus spinosus	Verbenaceae	52	12·59 (2·53–53·75)	46·40 (23·20)	AK, EX, KA, RU
Podocarpus nubigenus	Podocarpaceae	37	12·76 (5·73–68·13)	90·55 (35·30)	AK, EX, KA
Azara lanceolata	Flacourtiaceae	40	13·08 (7·15–29·70)	38·70 (20·94)	AK, EX, KA
Amomyrtus meli	Myrtaceae	21	14·54 (8·16–62·14)	58·58 (28·25)	KA
Raukaua laetevirens	Araliaceae	64	14·99 (5·73–86·94)	58·77 (23·85)	AK, EX, KA
Sophora macrocarpa	Fabaceae	6	15·41 (11·56–19·68)	45·23 (12·20)	RU
Aextoxicon punctatum	Aextoxicaceae	29	15·79 (10·66–45·36)	93·33 (18·07)	KA, RU
Myrceugenia correifolia	Myrtaceae	6	15·83 (14·47–19·07)	122·05 (15·13)	RU
Persea lingue	Lauraceae	12	15·87 (11·42–20·19)	88·86 (28·26)	RU
Maytenus magellanica	Celastraceae	24	16·01 (7·92–84·32)	72·84 (48·95)	EX
Lomatia ferruginea	Proteaceae	93	16·17 (6·75–86·94)	70·98 (33·93)	AK, EX, KA
Azara integrifolia	Flacourtiaceae	19	16·98 (4·50–33·81)	106·04 (32·79)	RU
Lomatia dentata	Proteaceae	3	17·11 (12·29–22·57)	120·51 (21·97)	RU
Cryptocarya alba	Lauraceae	19	17·19 (12·71–22·57)	100·13 (23·26)	RU
Peumus boldus	Monimiaceae	5	17·51 (15·49–21·77)	84·08 (8·39)	RU
Lithraea caustica	Anacardiaceae	26	17·65 (11·77–36·71)	117·55 (44·06)	RU
Ribes punctatum	Saxifragaceae	9	17·99 (12·29–20·77)	99·72 (29·23)	RU
Ribes magellanicum	Saxifragaceae	10	18·14 (10·33–44·27)	44·86 (24·31)	AK, EX
Ugni molinae	Myrtaceae	26	18·58 (11·42–36·71)	166·06 (58·76)	RU
Aristotelia chilensis	Elaeocarpaceae	33	18·62 (5·73–62·13)	45·65 (29·31)	AK, KA, RU
Quillaja saponaria	Rosaceae	4	18·74 (14·90–22·57)	100·50 (15·43)	RU
Fuchsia magellanica	Onagraceae	32	18·88 (6·35–84·32)	39·01 (38·68)	AK, EX, KA, RU
Myrceugenia exsucca	Myrtaceae	12	18·99 (4·5–52·48)	113·62 (48·48)	RU
Drimys winteri	Winteraceae	65	19·03 (6·75–86·94)	92·31 (44·41)	AK, EX, KA, RU
Amomyrtus luma	Myrtaceae	91	19·11 (6·75–82·94)	82·64 (35·99)	AK, EX, KA
Crinodendron hookerianum	Elaeocarpaceae	6	19·40 (10·45–45·36)	56·64 (22·17)	KA
Gevuina avellana	Proteaceae	36	20·47 (9·50–68·21)	94·16 (41·65)	KA, RU
Nothofagus glauca	Nothofagaceae	28	21·24 (11·42–52·48)	60·54 (15·95)	RU
Luma apiculata	Myrtaceae	53	21·68 (6·75–85·48)	94·17 (47·96)	AK, EX, KA, RU
Caldcluvia paniculata	Cunoniaceae	35	21·69 (6·75–85·48)	58·25 (32·31)	AK, KA
Gaultheria insana	Ericaceae	26	22·75 (11·42–52·48)	201·23 (47·74)	RU
Escallonia pulverulenta	Escalloniaceae	7	25·05 (12·71–52·48)	124·82 (56·05)	RU
Eucryphia cordifolia	Cunoniaceae	24	28·37 (8·72–68·21)	69·82 (34·56)	KA
Embothrium coccineum	Proteaceae	87	33·76 (7·92–92·55)	77·61 (50·47)	AK, EX, KA
Weinmannia trichosperma	Cunoniaceae	20	40·02 (8·16–86·94)	105·16 (48·85)	AK, EX, KA
Gaultheria mucronata	Ericaceae	30	42·87 (9·50–92·55)	122·90 (40·35)	AK, EX, KA, RU
Lomatia hirsuta	Proteaceae	12	48·51 (18·97–82·94)	159·42 (29·87)	KA, RU
Nothofagus betuloides	Nothofagaceae	10	51·06 (10·31–86·94)	128·80 (22·19)	EX
Escallonia rubra	Escalloniaceae	10	64·09 (10·96–90·17)	82·52 (27·11)	EX
Berberis darwinii	Berberidaceae	3	69·20 (18·19–85·35)	217·68 (44·88)	EX
Nothofagus nitida	Nothofagaceae	6	64·69 (38·15–85·48)	188·60 (25·48)	AK, EX
Tepualia stipularis	Myrtaceae	3	77·33 (70·26–85·48)	143·84 (31·71)	AK
Berberis microphylla	Berberidaceae	14	77·91 (10·31–93·39)	153·17 (31·11)	AK, EX
Nothofagus antarctica	Nothofagaceae	11	82·58 (18·19–93·39)	101·68 (15·37)	AK, EX
Baccharis patagonica	Asteraceae	12	82·69 (44·27–93·39)	112·78 (25·02)	AK, EX
Escallonia virgata	Escalloniaceae	3	83·06 (78·59–92·55)	91·98 (6·34)	EX

The list of species is ordered according to the mean GLI where they are most abundant; the hierarchy gives an idea of how shade tolerant they are.

late in the day to avoid direct exposure to sun. Photographs were also underexposed by two stops, to increase contrast between the sky and foliage (Zhang *et al.*, 2005). From these digital photographs, we computed the gap light index (GLI; Canham, 1988), which is an estimation of the percentage of photosynthetically active radiation reaching the point where the photograph is taken. The GLI was computed for each 2 m^2 plot using the Gap Light Analyzer (GLA ver. 2) software (Frazer *et al.*, 2000).

Statistical analysis

Species resource partitioning based on their LMA. Testing whether resource partitioning reduces niche overlap and promote coexistence in nature is not trivial (Adler *et al.*, 2013; Reich, 2014; Kraft *et al.*, 2015), particularly because one should ideally measure the effect of the resource level (light availability) on the plant's overall fitness (i.e. per capita growth rate) (Chesson, 2000) in the absence of competition. Although discarding the effect of competition is almost impossible in any

observational study, the forest understoreys we sampled had a low density of saplings (AIK, 2.1; EXP, 3.2; KAT, 2.8; and RUI, 3.3 saplings m^{-2}); thus we can assume relatively low competition among saplings. We note that adult trees also compete with saplings for light and below-ground resources, but, due to the highly asymmetric nature of these interactions, we view effects of adult trees on resource availability to saplings as a source of spatial environmental heterogeneity rather than competition *per se*. We used species abundance (cover) in a given plot as a proxy for fitness, recognizing that abundance probably reflects the effects of competition to some degree (Fox, 2012). We stress that species can only have high relative abundance if individuals have high probabilities of surviving, reproducing and growing, i.e. high fitness (Shipley *et al*., 2016). With this in mind, we quantified each species' response to light availability as the abundance-weighted mean GLI of plots in which the species occurred, using the formula:

$$LR_i = \frac{\sum_{s=1}^{n} GLI_s a_{i,s}}{\sum_{s=1}^{n} a_{i,s}},$$

where LR_i is the light response of species i, GLI_s is the GLI of plot s, $a_{i,s}$ is the cover of species i in plot s, and n is the total number of plots in a site. LR represents a species' optimum position along the light gradient. We assessed the role of traits in the light partitioning by testing whether species with different LMA values differed in their responses to light availability. For each site, we calculated species' pairwise differences in mean LMA (Δ LMA) and abundance-weighted mean GLI (Δ LR) and tested whether they were positively related using Mantel tests.

Relationship between LMA and light availability. To test whether LMA varies with light availability (GLI), we fitted linear models to the relationship between plot-level mean (PLM) LMA and GLI across plots for each site. We computed the PLM LMA values for each plot as the abundance-weighted average of species mean trait values, using species trait values measured within a particular plot. Visual inspection of the data showed that PLM LMA and GLI had a non-linear relationship with non-normal error distribution. We therefore log-transformed both variables and modelled the relationship using linear regression (Xiao *et al*., 2011).

RESULTS

In total, we measured LMA on 1304 saplings representing 50 woody species from 25 different families (Table 1) in four forests – three temperate rain forests and one Mediterranean forest – covering 11° of latitude in southern Chile. Although in most forests large canopy gaps were rather scarce, we were able to cover a wide range of GLI values (a surrogate of light availability reaching the forest floor, Table 1). Canopy cover values ranged from 2·69 to 62·33 % for AIK (mean = 16·38 %, s.d. = 16·30), 6·58 to 74·98 % for EXP (mean = 19·93 %, s.d. = 10·36), 2·01 to 57·68 % for KAT (mean = 9·20 %, s.d. = 9·65) and from 3·20 to 39·69 % for RUI (mean = 11·23 %, s.d. = 6·50).

The woody species surveyed in these forests strongly differed in their LMA values (Table 1). Species showing the lowest LMA values were *Fuchsia magellanica* and *Azara lanceolata* (Table 1), which are shade-intolerant species and are commonly found in the temperate rain forest sites. The highest LMA values were represented by *Berberis darwinii*, which is found in one of the temperate rain forests, and by *Gaultheria insana* and *Ugni molinae*, which are only found in Los Ruiles, the Mediterranean forest (Table 1). The woody species representing the extreme values of LMA are shrubs or treelets, and not tree species.

In all the temperate rain forests we sampled, we found a positive and significant ($P < 0·05$) correlation between species' pairwise differences in LMA (Δ LMA) and differences in their response to light availability, measured as absolute difference in abundance-weighted mean GLI (Δ light response, LR) of plots in which species occur (Fig. 2). However, we did not find a significant relationship between Δ LMA and Δ light response in the Mediterranean forest, Los Ruiles ($r = 0·164$, $P = 0·133$, Fig. 2). Although we recognize that the positive relationship between Δ LMA and Δ light response found in the temperate rain forests may also be due to a positive relationship between leaf lamina and LMA, i.e. species with larger leaves have high LMA while species with smaller leaves may have low LMA, we found no correlation between leaf area and LMA ($R^2 = -0·12$, $P = 0·47$) for the species under study.

Contrary to our original expectation, plot-level mean LMA increased significantly with increasing GLI in all forests (Fig. 3). This positive relationship was stronger in the three temperate rain forests, with R^2 ranging from 0·54 (Exploradores) to 0·73 (Katalapi), than in the Mediterranean forest, Los Ruiles ($R^2 = 0·38$).

DISCUSSION

Temperate rain forest species partition fine-scale gradients in light availability based on LMA

We demonstrate here that woody species in temperate rain forests in southern Chile respond differently to light gradients, and that these responses are linked to differences in a functional trait, LMA. This result constitutes a prerequisite for trait-based species coexistence via the spatial heterogeneity mechanism (Adler *et al*., 2013). Few studies so far have incorporated functional traits of early-stage development plant organisms (e.g. saplings) into community assembly models (Larson and Funk, 2016). Specifically, we found that woody species partitioned fine-scale gradients in light availability based on their LMA, and this niche partitioning potentially facilitates species coexistence at the within-community level. In the temperate rain forests we investigated, shade-intolerant species with high LMA were predominant in high-light microsites (i.e. canopy gaps) while shade-tolerant species with low LMA prevailed in shade environments (see below). We propose that the relationship between resource-use strategy and regeneration niche may ultimately drive differences in foliar traits (i.e. LMA) between co-occurring species. In contrast to the temperate rain forest sites, we found that woody species in the Mediterranean forest we surveyed did not partition the fine-scale gradient in light availability. This is most probably because, unlike temperate
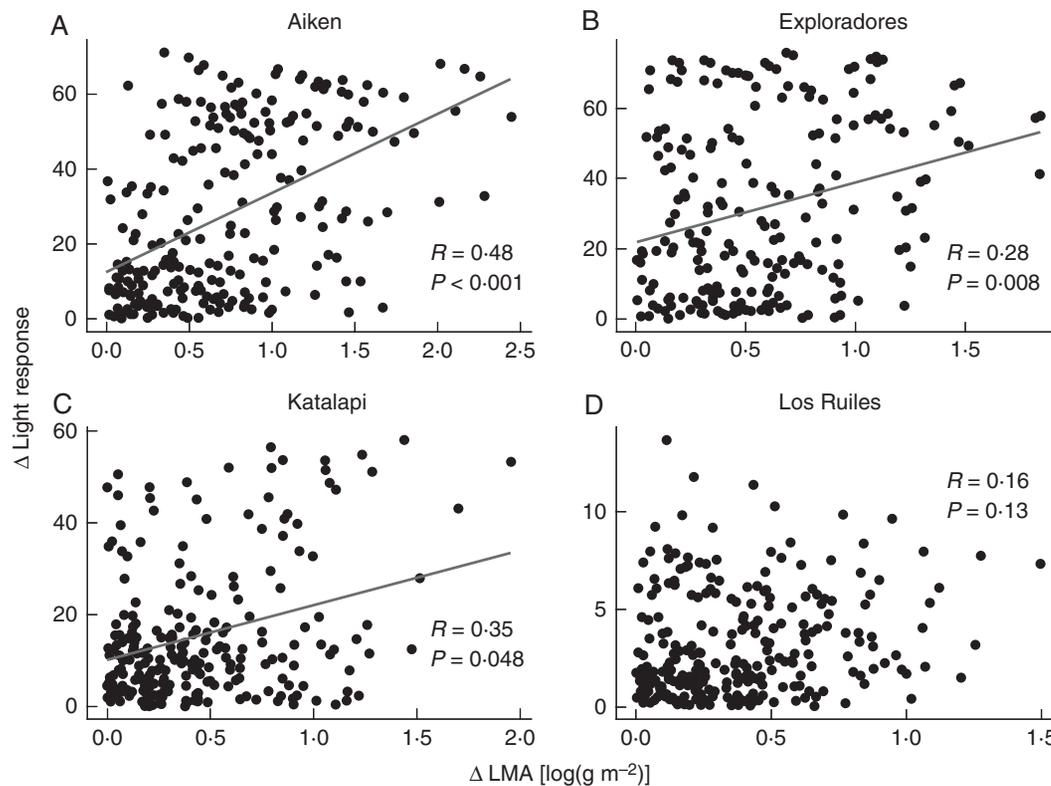
FIG. 2. Relationship between species' pairwise difference in mean leaf mass per area (Δ LMA, g m^{-2}) and difference in response to gradient of light availability (measured as difference in abundance-weighted mean GLI, Δ light response) for woody species in three temperate rain forests (Aiken, Exploradores and Katalapi) and one Mediterranean forest (Los Ruiles) in southern Chile.

rain forests, regeneration in Mediterranean forests is most strongly limited by water rather than light availability (Parada and Lusk, 2011), or perhaps regeneration here is mediated by traits other than LMA. In fact, the range of GLI values found in this Mediterranean forest (50 %) was considerably lower than in the temperate rain forests (80 %). The different patterns observed in the two forest types in this study suggest that functional traits may mediate species coexistence according to the limiting factors of a given site: light in temperate rain forests and probably soil moisture in Mediterranean forests.

Our findings are in accordance with the results of other studies showing significant relationships between LMA and other leaf traits and patterns of species abundance or occurrence at much coarser spatial scales (Katabuchi *et al.*, 2012; Laurans *et al.*, 2012; Mason *et al.*, 2012). Previous studies have also shown that species' leaf traits in a given community vary significantly across light environments (e.g. Lusk and Reich, 2000; Baltzer and Thomas, 2007; Sterck *et al.*, 2014), including a study in the temperate rain forest of southern Chile (Lusk, 2002). Our results demonstrate that species partition light gradients based on differences in leaf traits at within-community scales relevant to maintaining local species coexistence. Although we did not measure species per capita growth rates or the increase of species abundances when they are rare – the necessary evidence to test for stable species coexistence – our results suggest that differences in LMA may contribute to

stabilizing niche differences among species, satisfying a prerequisite for species coexistence (Adler *et al.*, 2013).

Shifts in the plot mean trait values along the light gradient

We found that variation in light availability was a major factor explaining variation in LMA along the entire light gradient of all our forests. For all forests, including temperate rain and the Mediterranean forests, we found a strong and positive relationship between LMA and GLI. On average, species having higher LMA were more frequently found in high-light canopy gaps, whereas species having low LMA prevailed in shade environments. This is in contrast to what has been found in moist tropical and temperate rain forests (Walters and Reich, 1999; Lusk, 2002; Poorter *et al.*, 2008). We believe this pattern may be explained because low LMA in saplings prevailing in low-light environments is more instrumental in maximizing light capture (through increased leaf lamina area) per unit of biomass investment, whereas high LMA in high-light environments maximizes photosynthetic capacity (i.e. thicker leaves with more photosynthetic machinery) per unit leaf area. The inter-specific pattern we found in the southern forests of Chile resembles what is generally found when looking at plastic responses to light or variation within individuals (e.g. sun vs. shade leaves), particularly for deciduous species (Bazzaz, 1979;
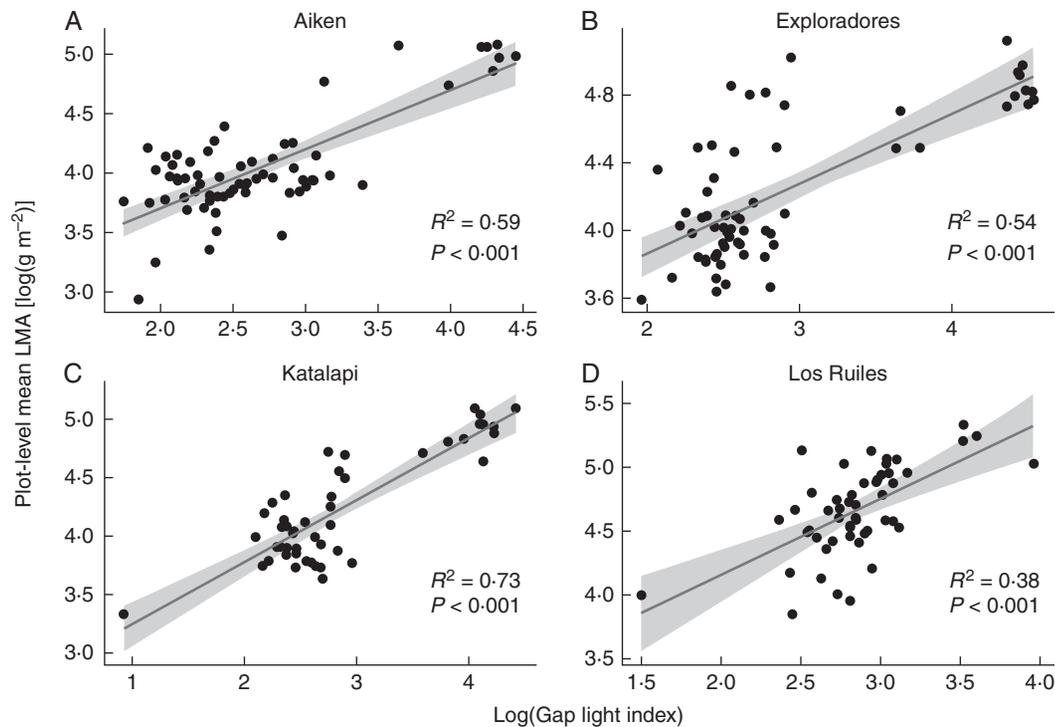
FIG. 3. Relationship between plot-level mean (PLM) leaf mass per area (LMA, g m^{-2}) and gap light index (GLI, a percentage measure of light availability) in three temperate rain forests (Aiken, Exploradores and Katalapi) and one Mediterranean forest (Los Ruiles) in southern Chile.

Givnish, 1988; Janse-Ten Klooster *et al.*, 2007; Lusk *et al.*, 2008). It is interesting to note that, in contrast to the northern hemisphere, where winter deciduous species that are also shade tolerant are very common, e.g. *Fagus sylvatica*, *Acer pseudoplatanus* and *A. saccharum* (Janse-Ten Klooster *et al.*, 2007), in the temperate rain forest of southern Chile, deciduous shade-tolerant species are non-existent (Grubb *et al.*, 2013). In evergreen species, particularly those of southern Chile, high LMA promotes shade tolerance by providing a long leaf life span (Lusk, 2002). Finally, we note that the relationship between LMA and GLI was non-linear (linearized by log transforming both variables, equivalent to a power law model), with rapid increases in PLM LMA at low GLI and little increase in PLM LMA at high GLI. These patterns indicate a saturation of LMA values with light availability beyond a specific threshold.

Limitations

A limitation of our study is that we used realized species abundance to quantify species' light responses. Species abundance has been found to be an imperfect proxy for direct responses of fitness to the environment (Fox, 2012), especially because it is also the result of biotic interactions (e.g. competition). Others have argued that relative abundance is a useful measure of how species respond to the environment; after all, where species show high relative abundance is where they have higher probabilities of surviving, reproducing and growing (Shipley *et al.*, 2016). Further, it may be difficult or impossible to measure species' responses to the environment in the absence of competition in the field, making relative abundances a pragmatic alternative, particularly for tree species.

CONCLUSIONS

Recently, it has been argued that linking species' functional trait differences with specific coexistence mechanisms is a necessary step toward improving the predictive power of trait-based approaches in community ecology. Here, we show that differences in a key functional trait, LMA, predict differences in woody sapling species' responses to light availability in temperate forests in southern Chile. This finding provides evidence of trait-based partitioning of fine-scale light availability by woody species at the regeneration level. We also propose that the concept of resource-use strategy successfully applied to adults can be used to define the regeneration niche of plant species (see Larson and Funk, 2016). This trait-based niche partitioning of fine-scale light availability was not evident in the Mediterranean forest community we studied, where light is not expected to be limiting. The identification of specific functional traits that mediate species responses to resource limitations within communities will improve our ability to understand responses of plant communities to shifts in resource availability and stress in the face of rapid environmental changes, including disturbance and global climate change.

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