

Alfredo O. Saldaña · Carolina Hernández
Rafael E. Coopman · León A. Bravo
Luis J. Corcuera

Differences in light usage among three fern species of genus *Blechnum* of contrasting ecological breadth in a forest light gradient

Received: 17 February 2009 / Accepted: 16 September 2009 / Published online: 6 November 2009
© The Ecological Society of Japan 2009

Abstract In Chilean evergreen temperate forest, fern species of the genus *Blechnum* occur in diverse microhabitats in a light gradient. We hypothesized that differences in the habitat preferences of three co-occurring *Blechnum* species would be associated with differences in the magnitude of responses of light capture [chlorophyll (Chl) content] and use (photosynthetic capacity and performance) to light availability. We measured the abundance, chlorophyll content, photosynthetic capacity (A), and photosynthetic performance (chlorophyll fluorescence of photosystems I and II) of juvenile individuals of each species growing in different light levels in the field. While *Blechnum magellanicum* covers a broad light environments range, *B. mochaenum* is restricted to shade, and *B. penna-marina* occupies full sun sites. Despite significant interspecific differences in average total chlorophyll content, this trait did not differ among species along the light gradient. There was significant interspecific variation in both the mean value and the plasticity of Chl a :Chl b ratio and A to light availability. While *B. penna-marina* showed a flatter reaction norm (lower response) of Chl a :Chl b ratio to light availability than its two congeners, *B. mochaenum* showed a lower response of A to light availability. *B. penna-marina* and *B. magellanicum* individuals from open sites had higher light saturation points of the electron transport rate (ETR) of both photosystems (ETR_{LSP} I and II) and photochemical quenching (qL and NA) than the shade restricted *B. mochaenum*. Additionally, non-photochemical quenching values for both photosystems

(NPQ and ND) were higher in ferns species occurring in shaded sites. The adjustment of the photosynthetic capacity and performance to light availability appears to be an important mechanism of acclimation in these three *Blechnum* species that differ in their habitat preferences across a light gradient.

Keywords Ecological breadth · Forest light gradient · *Blechnum* · Chlorophyll content and fluorescence · Photosynthetic capacity

Introduction

Most pteridophytes live in moist and shady environments (Page 2002; Aldasoro et al. 2004; Karst et al. 2005), mainly because fern species are dependent upon a moist environment for growth of their nonvascular gametophyte generation. In addition, hydraulic characteristics of fern foliage result in poor control of water conduction and loss (Woodhouse and Nobel 1982; Robinson 1994; Brodribb and Holbrook 2004). Nonetheless, some fern species are able to recruit in high-irradiance sites (Saldaña et al. 2005, 2007), thus having a wide ecological breadth despite the above constraints. Moreover, there are fern species that occupy almost exclusively open sites (Banister and Fagan 1989). It is widely known that plant species growing across different light environments should be able to produce leaves that are morphologically and physiologically suited to a wide range of light environments (Björkman 1981; Walters and Field 1987; Lambers et al. 1998). Heterogeneity of light environments can influence photosynthetic performance of ferns by means of a direct effect on the photosynthetic apparatus and its damage under stress conditions (Valladares and Pearcy 1997; Lambers et al. 1998; Walters 2005). The balance between the light energy absorbed and that used for metabolism is critical in photosynthetic organisms (Hüner et al. 2002), hence for the maintenance of a positive carbon balance in a wide range of environments and for successfully occupying diverse habitats (Chazdon 1992; Spencer et al. 1994).

A. O. Saldaña (✉) · C. Hernández · L. A. Bravo ·
L. J. Corcuera
Departamento de Botánica,
Universidad de Concepción,
Casilla 160-C, Concepción, Chile
E-mail: asaldana@udec.cl
Tel.: +56-41-2204418
Fax: +56-41-2246005

R. E. Coopman
Instituto de Silvicultura, Facultad de Ciencias Forestales,
Universidad Austral de Chile, Valdivia, Chile

From an ecophysiological perspective, interspecific differences in leaf chlorophyll (Chl) content along a light gradient are important to our understanding of the ecological performance of fern species in occupying different light environments. The amount of solar radiation absorbed by a leaf is largely a function of foliar concentrations of photosynthetic pigments. Therefore, low concentrations of chlorophyll can directly limit the photosynthetic potential and primary productivity (Curran et al. 1990; Filella et al. 1995). Since the relative concentrations of chlorophylls are known to change with light environments (e.g., sun leaves have a higher Chl *a/b* ratio; Lambers et al. 1998; Larcher 2003), quantifying this ratio in plants across a light gradient may provide important information about relationships between plants and their light environment. Additionally, the maintenance of a positive carbon balance in a wide range of environments is a key feature of species that successfully occupy diverse habitats (Chazdon 1992; Spencer et al. 1994). Leaf traits relevant to net carbon gain, and hence to components of plant fitness, include photosynthetic rate (*A*) (Givnish 1988; Chazdon 1992; Reich et al. 1998). Given the influence of photosynthetic capacity on plant growth and reproduction, plasticity in this ecophysiological trait could be associated with variation in ecological breadth (Sultan et al. 1998).

The use of chlorophyll *a* fluorescence as a tool for measuring photosynthetic response to light has shown a considerable potential for assaying both the rate of energy transfer through photosystem II (PSII) and characteristics of the energy-dissipating processes (Demmig-Adams et al. 1995), which modulate the photochemical response to light. Measuring the yield of chlorophyll fluorescence provides information about changes in the efficiency of photochemistry and heat dissipation (Maxwell and Johnson 2000). In this context, the response of the apparent electron transport rate (ETR) to photosynthetic photon flux density (PPFD) expresses the relative rate of electron transport through PSII, which would potentially reduce NADP⁺, and thus, it is closely related to the rate of carboxylation (Edwards and Baker 1993; Brodribb and Hill 1997). The measurement of ETR provides a means for probing the primary photochemical response and photochemical capacity of the leaf without masking effects such as dark and light respiration rates (Brodribb and Hill 1997). Skillman and Winter (1997) argued that a relatively greater increase in Φ PSII could be observed during sunflecks; hence, this parameter is particularly relevant to fern species established in the forest understorey, where spatial and temporal variability in light availability occurs (Chiariello 1984; Walters and Field 1987). Furthermore, the extent and composition of non-photochemical quenching (NPQ) has been successfully used to explore differences in photoprotection and photoinhibition between sun and shade leaves within species (Demmig-Adams et al. 1995). Additionally, there is paucity of research on photosynthetic performance in relation to ecological breadth of ferns species.

This study addressed the interspecific differences of functional responses to light of traits related to light capture and use in three sympatric fern species of the genus *Blechnum* (Pteridophyta, Blechnaceae) that differ in ecological breadth in the Chilean evergreen temperate rain forest. *Blechnum magellanicum* (Desv.) Mett. covers a broad environmental range, from shaded understoreys to open sites; *Blechnum mochaenum* Kunkel is restricted to shady and moist sites; and *Blechnum penna-marina* (Poiret) Kuhn occupies mainly full sun sites. Variation in leaf functional traits appears to be an important mechanism of acclimation of *Blechnum* species to this wide range of light environments (Saldaña et al. 2005, 2007). We evaluated the hypothesis that interspecific differences in the magnitude of responses of chlorophyll content, photosynthetic capacity and photosynthetic performance to light availability of the three *Blechnum* species reflect their ecological breadth. This was considered likely because the life cycle of these *Blechnum* species is short, relative to the life-spans of tree species whose growth and mortality control most of the variation in light environments associated with patch dynamics. First, we assessed the ecological breadth by measuring the abundance of the juvenile individuals in the field light gradient. Then, we compared the values of chlorophyll content, photosynthetic capacity, and chlorophyll fluorescence of juvenile individuals of each species growing in different light levels in situ.

Methods

Species distributions in relation to canopy openness

This study was performed in a forest stand at Katalapi Park in Pichiquillaiepe, southcentral Chile (41°31'S, 72°45'W). The climate at Pichiquillaiepe is considered temperate and strongly humid with an oceanic tendency (Di Castri and Hajek 1976), although winter frosts occur. Annual precipitation is around 2,200 mm (mean annual rainfall) (Reyes-Díaz et al. 2005). The old-growth rainforest of the lower western slopes of the Andes is composed exclusively of broad-leaved evergreen species (Saldaña and Lusk 2003).

Sampling was carried out on ten transects run through old-growth stands including tree-fall gaps of varied sizes and open sites. Sets of parallel transects were run through accessible stands, spaced at least 20 m apart, the angle and number of transects depending on terrain, access considerations, and proximity to forest margins. At 150 sample points (15 per transect) spaced at random intervals (2–10 m apart) along transects, canopy openness (percentage of diffuse light) measurements were made at 100-cm height with the LAI-2000 (Li-Cor, Lincoln, NE, USA). A pair of LAI 2000 canopy analyzers was used to quantify light environments under uniform overcast sky conditions. One instrument was used to take measurements 50 cm above each sampling

point, while the other, placed at the center of a 2-ha clearing, was programmed to take readings at 30-s intervals. Fifty meters was the minimal distance among both instruments, to ensure the independence of light measurements. Integration of data from the two instruments enabled estimation of diffuse non-intercepted irradiance (%) at each sampling point within the forest, equivalent to percentage canopy openness over the quasi-hemispherical (148°) view field of the LAI-2000 sensors. LAI-2000 measurements are a good surrogate of spatial variation in mean daily photosynthetic photon flux density within a stand (Machado and Reich 1999).

To determine the ecological preferences of the three *Blechnum* species, the presence of each fern species was recorded in a plot of 1 m² centered on each sample point. Given that in most forests the frequency distribution of light environments is highly skewed towards microsites at <ca 5% canopy openness (Montgomery and Chazdon 2002), one way to incorporate the relative availability of different light environments is to work with rank order of canopy openness at microsites, instead of actual canopy openness values (Lusk et al. 2006). Equal intervals on a rank-transformed gradient represent equal areas of microsite availability. Accordingly, we ordered our 150 samples by light environment (from 0.1 to 70% canopy openness) at 10% intervals. Thus the abundance of each fern species across the light gradient was estimated as the frequency of occurrence in the number of plots resulting within each canopy openness interval.

Chlorophyll content and photosynthetic capacity measurements

We measured chlorophyll content in 20–22 juvenile individuals of each fern species, distributed across the light gradient. For each sampled individual three circular disks of fresh tissue were punched from one frond for posterior chlorophyll extractions. The percentage of canopy openness was determined above each sampled individual. Given that some of these fern species can develop red-colored fronds from the accumulation of anthocyanins, we selected only green fully expanded fronds. We used standard extraction techniques and a spectrophotometer to measure Chlorophyll *a* (Chl *a*), Chlorophyll *b* (Chl *b*), and total Chl content (Lichtenthaler and Wellburn 1983).

Area-based photosynthetic capacity (*A*) was measured in situ in 16 individuals of each fern species, distributed across the light gradient. The percentage of canopy openness was determined above each sampled individual. *A* measurements were made using a portable infrared gas analyzer and leaf chamber (PP Systems, Hitchin, England). Photosynthetic capacity was measured at PAR 800 μmol m⁻² s⁻¹ (assumed to be a saturating level), at 20°C. These measurements were carried out in mid-growing season (October) on two fully

expanded leaves per plant. The average of these measurements was used as an individual plant value.

Chlorophyll fluorescence and P700 measurements

Light-response curves of chlorophyll *a* fluorescence and P700 assessment were performed simultaneously with a Dual-PAM-100 measuring system (Walz-Effeltrich, Germany) in detached new fully expanded fronds. These were immediately dark adapted for 30 min (to obtain open reaction centers). A saturating pulse was applied to obtain F_m; then, leaves were exposed 5 min at each PPFD in order to obtain steady-state lectures. All measurements were performed at 15°C. Temperature was controlled with a water jacket system placed around the Dual-D leaf holder and connected to a thermoregulated water bath. Frond temperature was monitored with a fine K type thermocouple (Sper Scientific, Ltd. USA). During these measurements, the environmental temperature ranged from 15 to 18°C. Recordings and calculations were performed with the Dual-PAM 1.7 data analyses and control software (Walz-Effeltrich, Germany). The saturation pulse method originally developed for chlorophyll fluorescence quenching analysis (Schreiber et al. 1986) was used for assessment of PSI quantum yield (Klughammer and Schreiber 1994). Electron transport rate of PSII (ETR_{II}), yield of energy dissipation by downregulation Φ(NPQ), and the fraction of PSII centers in open state (qL) were calculated according to Kramer et al. (2004). Analysis of PSI was based on a routine for assessment of the maximal P700 change (P_m determination), which involves preillumination with far-red light (FR) for 2 s and a saturation pulse that induces maximal P700 oxidation followed by full reduction. The P_m determination is analogous to F_o and F_m determinations. The concept of excitation energy partitioning originally conceived for PSII by Kramer et al. (2004) was adopted for PSI. Hence, the relative electron transport rate for PSI (ETR_I) and the non-photochemical quenching donor Φ(ND) and acceptor Φ(NA) side limitation were obtained. The non-photochemical quantum yields of PSI, ND, and NA, represent the fraction of overall P700 that is oxidized in a given state and the fraction of overall P700 that cannot be oxidized by a saturation pulse in a given state due to a lack of acceptors, respectively. Both centers with oxidized P700 and acceptor side limited centers transform absorbed excitation energy quantitatively into heat. ND is a measure of donor side limitation, which is enhanced by a transthylakoid proton gradient (photosynthetic control at cyt b/f complex as well as down-regulation of PSII) and damage at the level of PSII. Additionally, NA is a measure of acceptor-side limitation, which is enhanced by dark adaptation (deactivation of key enzymes of Calvin–Benson cycle) and damage at the level of CO₂ fixation. The light saturation points of ETR_{II} (ETR_{II}_{LSP}) and ETR_I (ETR_I_{LSP}) were obtained homologous to the methodology for light curve of CO₂

assimilation proposed by Farquhar et al. (1980). Briefly, ETR_{LSP} was the PPFD corresponding to the intersection of the horizontal line described by the average of non-significant different maximal ETR values. The projection of the initial linear portion with maximal slope of the light curve corresponding to the ETR maximum quantum yields.

Data analysis

An ANCOVA was applied (species as main effect and light as covariate) in order to test interspecific differences in total chlorophyll content, Chl *a*:Chl *b* ratio, and photosynthetic capacity across the light gradient in the field. Values for chlorophyll fluorescence and P700 measurements correspond to the mean of five measurements performed in five different plants. Data were subjected to a one-way ANOVA (where the factor was species in each light environment). Before ANOVA, data were checked for normality and homogeneity of variances. In order to simplify the post hoc analysis interpretation of the light curves, the whole light rank was not considered. The analysis was performed at two independent fixed irradiances by each photosystem. These irradiances correspond to $ETR_{II,LSP}$ and $ETR_{I,LSP}$ of the shade species *B. mochaenum* (27 and 221 $\mu\text{mol photons m}^{-2}\text{s}^{-1}$) and the sun species *B. penna-marina* (58 and 435 $\mu\text{mol photons m}^{-2}\text{s}^{-1}$). A Holm-Sidak method was used for post hoc analyses. Differences were considered significant at $P \leq 0.05$. All the statistical analyses were performed with SigmaStat 3.1 software (2004, Systat Software, Inc. Richmond CA, USA).

Results

Breadth in the light gradient

The three *Blechnum* species coexisted around 30% canopy openness (Fig. 1). *B. mochaenum* was the most abundant species at low light availability; *B. magellanicum* occurred continuously across most of the light gradient (Fig. 1). *B. penna-marina* was restricted mainly to full sun sites, being the most abundant *Blechnum* species in open habitats (Fig. 1).

Chlorophyll contents and photosynthetic capacity

Whereas the mean value (elevation of reaction norms) of total Chl, Chl *a*:Chl *b* ratio, and *A* of the *Blechnum* species was different, plasticity (slope of reaction norms) of total Chl to light availability did not differ among species (Table 1). In any given light environment, Chl *a*:Chl *b* was greater on average in *B. penna-marina* than in *B. magellanicum* and *B. mochaenum* (Fig. 2b). While total Chl decreased and Chl *a*:Chl *b* ratio increased with

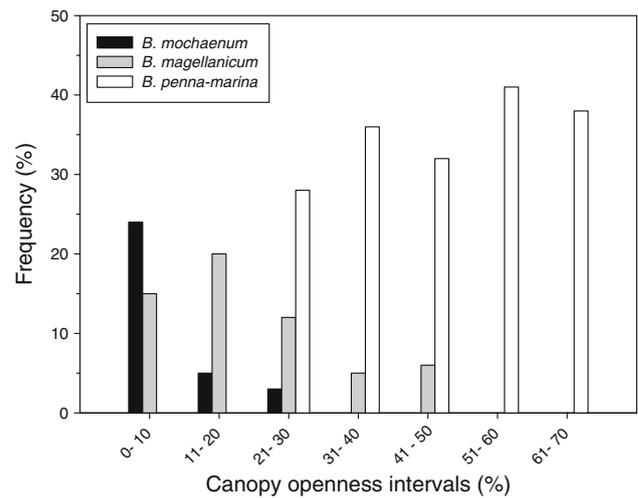


Fig. 1 Distribution (% frequency) of *Blechnum* fern species in plots across the light gradient (canopy openness intervals)

Table 1 ANCOVA of total chlorophyll content (mg cm^{-2}), Chl *a*:Chl *b* ratio, and photosynthetic capacity (*A*, $\mu\text{mol m}^{-2}\text{s}^{-1}$) of *Blechnum mochaenum*, *B. magellanicum* and *B. penna-marina* growing across a light gradient

	<i>df</i>	SS	<i>F</i>	<i>P</i>
Total Chl				
Species	2	0.003	8.06	0.001
Light	1	0.001	2.27	0.137
Species \times light	2	0.001	1.28	0.286
Chl <i>a</i> :Chl <i>b</i>				
Species	2	0.002	18.46	0.001
Light	1	0.001	10.24	0.002
Species \times light	2	0.001	3.22	0.051
<i>A</i>				
Species	2	21.853	28.14	0.001
Light	1	8.261	21.27	0.001
Species \times light	2	4.489	5.78	0.006

Main factor: species; covariate: light

light availability in *B. mochaenum* and *B. magellanicum*, these traits showed a relatively flat response to light in *B. penna-marina* (Fig. 2a, b). Photosynthetic capacity increased with light availability in all three species (Fig. 2c). ANCOVA showed differences in slope among species for Chl *a*:Chl *b* and *A* (Table 1). Thus, although all three species displayed fronds of higher Chl *a*:Chl *b* ratio and greater photosynthetic capacity in high light, these increase were more marked in *B. mochaenum* and *B. magellanicum* than in *B. penna-marina* (Fig. 2b, c).

Effect of light environment on chlorophyll fluorescence and P700 light response curves of the three species

Changes in the use of absorbed light energy were performed by comparing $ETR_{II,LSP}$ and $ETR_{I,LSP}$ of the strict shade and sun fern species. At 27 $\mu\text{mol photons m}^{-2}\text{s}^{-1}$ all species showed a similar ETR_{II} (about

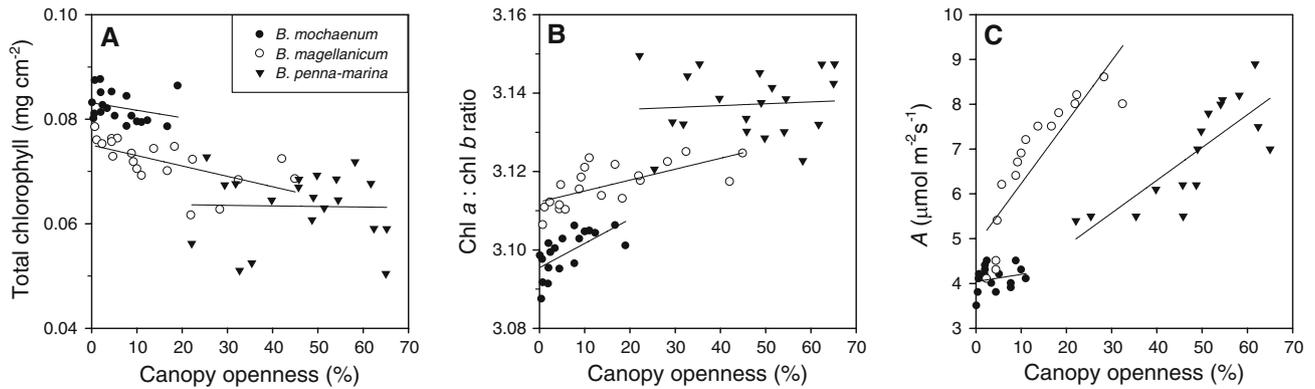


Fig. 2 Variation in chlorophyll content (mg cm^{-2}) (a), Chl *a*:Chl *b* ratio (b), and photosynthetic capacity (A , $\mu\text{mol m}^{-2}\text{s}^{-1}$) (c) in *Blechnum mochaenum* (filled circles), *B. magellanicum* (open circles),

and *B. penna-marina* (filled triangles) juvenile individuals across the light gradient in an evergreen temperate forest

$7.5 \mu\text{mol electrons m}^{-2}\text{s}^{-1}$). *B. penna-marina* exhibited a slightly higher but significant ETRII than the understorey species *B. mochaenum* and *B. magellanicum* shade ferns (Fig. 3a). At the ETRII_{LSP} ($221 \mu\text{mol photons m}^{-2}\text{s}^{-1}$) for *B. penna-marina*, this fern species exhibited a 51% higher ETRII than *B. magellanicum* sun individuals, 77% than *B. magellanicum* shade individuals, and 83% than the umbrophile fern *B. mochaenum*. Additionally, *B. penna-marina* and *B. magellanicum* (both sun and shade individuals) were able to increase their ETRII in 33, 180, and 473% relative to the *B. mochaenum* ETRII_{LSP}, respectively (Fig. 3a). At *B. mochaenum* ETRII_{LSP} *B. penna-marina* exhibited an 80% lower $\Phi(\text{NPQ})$ than the other species (Fig. 3c). At the *B. penna-marina* ETRII_{LSP}, this species exhibited a 54% lower $\Phi(\text{NPQ})$ than *B. magellanicum* sun and shade ferns, and 22% than *B. mochaenum*. Additionally, *B. penna-marina*, *B. magellanicum*, and *B. mochaenum* were able to increase their $\Phi(\text{NPQ})$ in 500, 132, and 82% relative to the *B. mochaenum* ETRII_{LSP}, respectively (Fig. 3c). At $27 \mu\text{mol photons m}^{-2}\text{s}^{-1}$, *B. penna-marina* exhibited significantly higher qL (38%) than *B. magellanicum* sun individuals and 80% than understorey ferns *B. mochaenum* and *B. magellanicum* shade individuals (Fig. 3e). At the *B. penna-marina* ETRII_{LSP}, this fern species exhibited a 73% higher qL than *B. magellanicum* sun ferns, 420% than *B. magellanicum* shade, and 940% than the umbrophile fern *B. mochaenum* (Fig. 3e). Consistently at *B. penna-marina* ETRII_{LSP}, *B. penna-marina*, *B. magellanicum* sun and shade ferns and *B. mochaenum* were able to maintain different fractions of PSII reaction centers open relative to initial dark conditions (59, 34, 11, and 6%), respectively (Fig. 3e). At *B. mochaenum* ETRII_{LSP} ($58 \mu\text{mol photons m}^{-2}\text{s}^{-1}$) sun species showed equal ETRI. Understorey ferns *B. mochaenum* and *B. magellanicum* shade individuals presented a 56 and 44% significantly lower ETRI than sun ferns, respectively (Fig. 3b). At the *B. penna-marina* ETRII_{LSP} ($435 \mu\text{mol photons m}^{-2}\text{s}^{-1}$) this fern species exhibited a 106% higher ETRI than *B. magellanicum* sun individuals, 300% than *B. magellanicum* shade individuals, and

620% than the umbrophile fern *B. mochaenum*. Additionally, *B. penna-marina* and *B. magellanicum* sun and shade individuals were able to increase their ETRI in 300, 94, and 50% relative to the *B. mochaenum* ETRII_{LSP}, respectively (Fig. 3b). At *B. mochaenum* ETRII_{LSP} *B. penna-marina* and *B. magellanicum* sun ferns exhibited a 0.74% lower $\Phi(\text{ND})$ than *B. magellanicum* shade ferns and an 81% than *B. mochaenum* (Fig. 3d). At the *B. penna-marina* ETRII_{LSP} this species exhibited a 25% lower $\Phi(\text{ND})$ than *B. magellanicum* sun ferns, 36% than *B. magellanicum* shade individuals, and *B. mochaenum*. Additionally, *B. penna-marina* and *B. magellanicum* sun and shade ferns, and *B. mochaenum* were able to increase their $\Phi(\text{ND})$ in 620, 440, 118, and 57% relative to *B. mochaenum* ETRII_{LSP}, respectively (Fig. 3d). At *B. mochaenum* ETRII_{LSP} *B. penna-marina* and *B. magellanicum* sun ferns exhibited a 460 and 260% higher $\Phi(\text{NA})$ than shade species. (Fig. 3f). At the *B. penna-marina* ETRII_{LSP} there were no differences between fern species reaching a value of 0.08 (Fig. 3f).

Discussion

Blechnum fern species that coexist in the Chilean evergreen temperate forest differed in ecological breadth in relation to light availability. Their differences in photosynthetic responses to light variation are consistent with their ability to occupy different light environments. Light is the most important resource limiting plant survival and growth in the understorey of humid forests (Pacala et al. 1994; Finzi and Canham 2000; Saldaña and Lusk 2003). Therefore, the ability of fern species to capture and use this resource may determine to a large extent their ecological breadth in this forest ecosystem (Saldaña et al. 2005, 2007).

Both *Blechnum* species that coexist in forest shaded environments (*B. magellanicum* and *B. mochaenum*) showed greater chlorophyll content than the open-site species *B. penna-marina*. This interspecific difference in chlorophyll content reflects that their distribution across

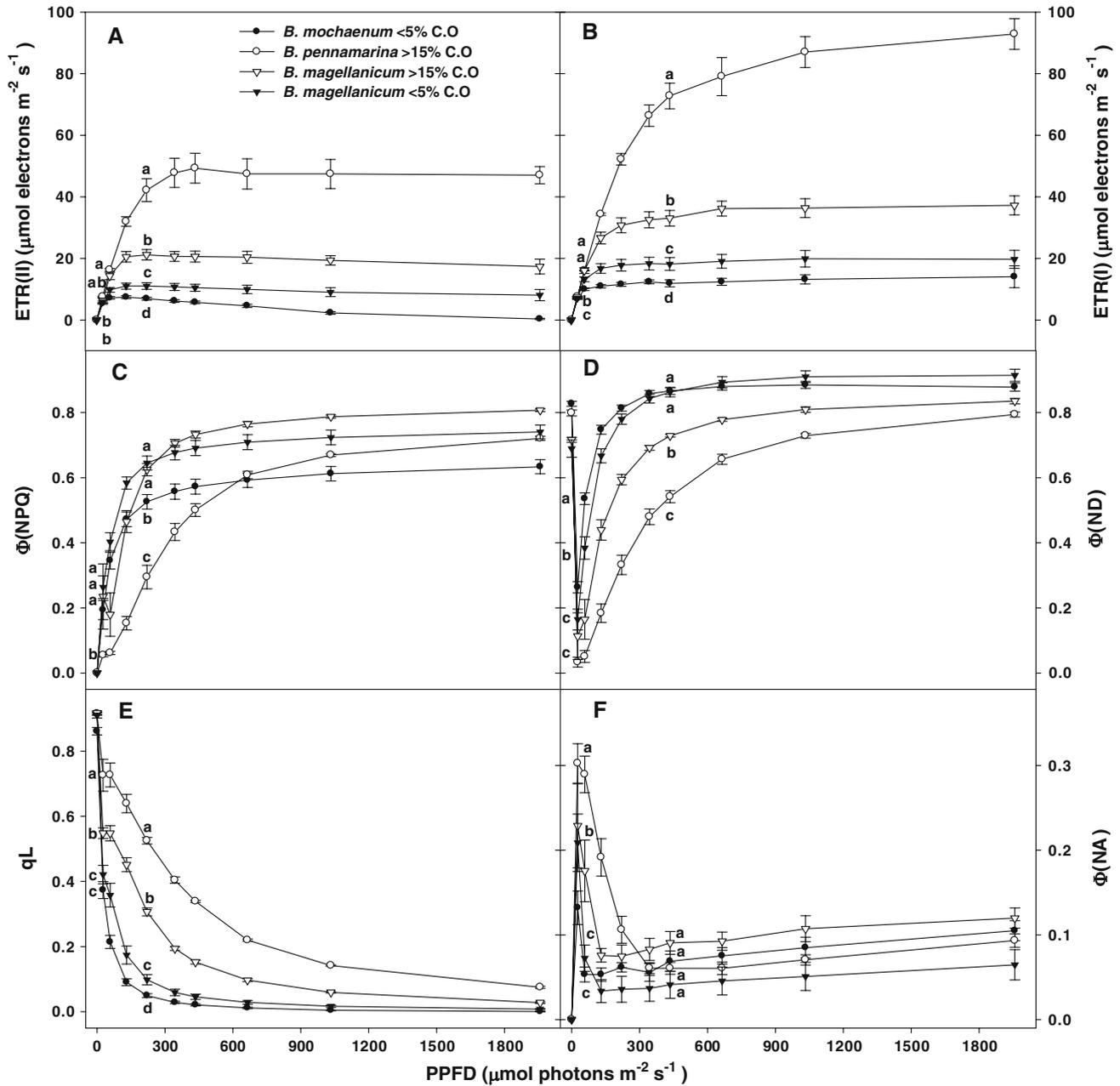


Fig. 3 Chlorophyll fluorescence and P700 light response curves of three *Bhechmum* fern species growing in two contrasting light environments (<5 and >15% of canopy openness, % C.O) in an evergreen temperate forest. **a** Electron transport rate through PSII and **b** PSI. **c** Quantum yield of regulated non-photochemical energy dissipation. **d** Non-photochemical quenching $\Phi(\text{ND})$ is a measure of donor side limitation, which is enhanced by a transthylakoid proton gradient and damage at the level of PSII. **f** Non-

photochemical quenching $\Phi(\text{NA})$ is a measure of acceptor side limitation, which is enhanced by dark adaptation and damage at the level of CO_2 fixation. **e** Relative redox state of PSII in response to increased actinic light intensity. Different letters on data point within PPFD and between fern species and growth light conditions indicate significant differences evaluated with the Holm-Sidak method ($n = 5$; $P \leq 0.05$)

the light gradient is concomitant with the fact that light harvesting complexes (LHC) are greater in plants growing in shade environments. The interspecific differences in response of Chl *a*:Chl *b* ratio could be related to the more restricted ecological breadth of *B. mochaenum* with respect to the other two species.

Additionally, the more plastic Chl *a*:Chl *b* ratios of *B. magellanicum* may allow this species to occupy a wider range of light environments, reflecting the ability of this fern species to adjust its light-capture mechanism. A change in Chl *a*:Chl *b* ratio alters the amount of light that can be intercepted by LHC. This is

one of the most important biochemical mechanisms in maximizing carbon gain in response to light availability variation (Lambers et al. 1998; Larcher 2003). Hence, the apparent lack of biochemical adjustment in response to prolonged exposure to high light may partly explain why *B. mochaenum*, a shade-tolerant species (Saldaña et al. 2005), does not occur on open sites.

When these three *Blechnum* species were exposed to a range of irradiances, the interspecific differences in photosynthetic capacity and performance could explain their habitat preferences and specificity. The relatively low photosynthetic capacity and performance of *B. mochaenum* at higher light levels may therefore limit its distribution to shaded sites by means of several mechanisms. Low *A* determines low growth rates, with the result that *B. mochaenum* is outcompeted by faster-growing species in high light (Lambers and Poorter 1992). Alternatively, low photosynthetic capacity and performance could render *B. mochaenum* susceptible to photoinhibition in high light (Long et al. 1994). *B. magellanicum* individuals may cope with the heterogeneity of light availability changing the structure of its LHCs. At any given light level, ETRI and ETRII were generally higher in ferns from open sites (*B. penna-marina* and *B. magellanicum* open-site individuals) than in ferns from shaded sites (*B. mochaenum* and *B. magellanicum* shade individuals) (Fig. 2a, b). The higher ETR(I) and ETR(II) of *B. penna-marina* in fronds from open sites at high light intensity may reflect a higher capacity of this species for cyclic electron transport or reduction of P700 by stromal components as discussed elsewhere (Asada et al. 1992). Simultaneous measurements of light-response curves of PSI and PSII provide information on electron transport capacity and limitations considering the interplay of the consecutive light reactions of PSII and PSI, which are connected via the intersystem electron transport chain. The fates of light energy in exposed and shaded *Blechnum* species reflect a previously described mechanism for flowering plants (Roberts et al. 1998): a combination of both high photosynthetic light use and thermal dissipation, which diminishes the excess excitation energy and photodamage.

The interspecific differences in the increase of thermal dissipation of excess energy (NPQ) with light intensity in these *Blechnum* species (Fig. 2c), reflect that *B. magellanicum* individuals are able to acclimate to different light environments. *B. penna-marina* showed lower NPQ than the other two fern species, however, this species has the greater photochemical quenching as well, reflecting its preference by high-irradiance habitat. On the other hand, the shade species *B. mochaenum* and *B. magellanicum* open-site individuals showed greater NPQs than *B. penna-marina* and *B. magellanicum* shade individuals. The process of non-photochemical quenching is known to be induced in plants under conditions of stress to dissipate excess light energy, protecting the plant from light damage (Horton et al. 2000). It has been reported that plants growing in the shade typically show lower photosynthetic capacity, high levels of NPQ, and mini-

mal light use (Griffiths and Maxwell 1999). However, *B. magellanicum* individuals from open sites showed a slightly higher NPQ response than shade species. Therefore, it seems that *B. magellanicum* is also able to withstand high light and sustain a primary photochemical response under such condition. This probably reflects the capacity of *B. magellanicum* shade fronds to adjust the photochemical capacity of individuals from shade to use high-energy inputs coming from sunflecks, as observed in many understory plants (Leakey et al. 2004).

All three *Blechnum* species displayed increases in photosynthetic capacity with greater available light. However, in any given light environment, *A* was greater on the average in *B. penna-marina* and *B. magellanicum* than in *B. mochaenum*. The latter species behaves photosynthetically like a typical “shade” plant (sensu Björkman 1981), with low rates of photosynthesis. The relatively low plasticity of *B. mochaenum* to higher light levels may therefore limit its distribution to shaded sites by means of several mechanisms (Saldaña et al. 2005). There was evidence of differential plasticity in photosynthetic capacity, with *B. magellanicum* and *B. penna-marina* showing a steeply increase in *A* with light availability.

The differences observed in these three *Blechnum* species in their natural habitat preferences across a light gradient probably reflect intrinsic or structural differences between shade and light ferns photosynthetic machineries. The occurrence of spatial and temporal environmental variability product of forest gap dynamics (Denslow 1987), determines habitats that differ in light availability (Chiariello 1984; Walters and Field 1987). In this context, the wide light gradient used by *B. magellanicum* in this temperate forest may reflect its ability to maintain the photosynthetic performance and display physiological responses that appears to be important mechanisms of acclimation to light availability. This is suggested by the fact that this fern can adjust its photosynthetic capacity to forest-changing environments such as when gaps are created or closed or even to shorter-term light variations, such as sunflecks. On the contrary, our results suggest that *B. mochaenum* and *B. penna-marina* lack the ability to adjust their photosynthetic machineries to maintain a high photosynthetic capacity and performance in wide range of light environments, being restricted to shaded understories and full sun sites, respectively. Further research on the ecophysiological basis of fern ecological breadth should also include the relationship between photosynthetic capacity and plant growth. Finally, we cannot rule out that the photosynthetic differences between the two extreme species could be the result of phenotypic acclimation rather than being inherent. Experimental exposure to a common growth irradiance is thus required to understand better the contribution of this functional responses to the segregation of the species along the light gradient.

Acknowledgments We wish to thank Veronica Briceño for her help with the fluorescence measurements.

References

- Aldasoro JJ, Cabezas F, Aedo C (2004) Diversity and distribution of ferns in sub-Saharan Africa, Madagascar and some islands of the South Atlantic. *J Biogeogr* 31:1579–1604
- Asada K, Heber U, Schreiber U (1992) Pool Size of electrons that can be donated to P700+, as determined in intact leaves: donation to P700+ from stromal components via the inter-system chain. *Plant Cell Physiol* 33:927–932
- Banister P, Fagan B (1989) The frost resistance of fronds of *Blechnum penna-marina* in relation to season, altitude, and short-term hardening and dehardening. *New Zeal J Bot* 27:471–476
- Björkman O (1981) Responses to different quantum flux densities. In: Lange O, Nobel P, Osmond C, Ziegler H (eds) *Physiological plant ecology 1*, responses to the physical environment. Springer, Berlin Heidelberg New York, pp 57–107
- Brodribb TJ, Hill R (1997) Light response characteristics of a morphologically diverse group of southern hemisphere conifers as measured by chlorophyll fluorescence. *Oecologia* 110:10–17
- Brodribb TJ, Holbrook NM (2004) Stomatal protection against hydraulic failure: a comparison of coexisting ferns and angiosperms. *New Phytol* 162:663–670
- Chazdon R (1992) Photosynthetic plasticity of two rain forest shrubs across natural gaps transects. *Oecologia* 92:586–595
- Chiariello N (1984) Leaf energy balance in the wet lowland tropics. In: Medina E, Mooney H, Vázquez-Yanes C (eds) *Physiological ecology of plants of the wet tropics*. Dr. W. Junk, The Hague, Netherlands, pp 85–98
- Curran PJ, Dungan JL, Gholz HL (1990) Exploring the relationship between reflectance red edge and chlorophyll content in splash pine. *Tree Physiol* 7:33–48
- Demmig-Adams B, Adams W, Logan B, Verhoeven A (1995) Xanthophyll cycle dependant energy dissipation and flexible photosystem II efficiency in 380 plants acclimated to light stress. *Aust J Plant Physiol* 22:249–260
- Denslow JS (1987) Tropical rainforest gaps and tree species diversity. *Annu Rev Ecol Syst* 18:431–451
- Di Castri F, Hajek E (1976) *Bioclimatología de Chile*. Vicerrectoría Académica. Universidad Católica de Chile, Santiago
- Edwards GE, Baker NR (1993) Can assimilation in maize leaves be predicted accurately from chlorophyll fluorescence analysis? *Photosynth Res* 37:89–102
- Farquhar GD, von Caemmerer S, Berry JA (1980) A biochemical model of photosynthetic CO₂ assimilation in leaves of C3 species. *Planta* 149:78–90
- Filella I, Serrano L, Serra J, Peñuelas J (1995) Evaluating wheat nitrogen status with canopy reflectance indexes and discriminant-analysis. *Crop Sci* 35:1400–1405
- Finzi A, Canham C (2000) Sapling growth in response to light and availability in a southern New England forest. *For Ecol Manag* 131:153–165
- Givnish TJ (1988) Adaptation to sun and shade: a whole-plant perspective. *Aust J Plant Physiol* 15:63–92
- Griffiths H, Maxwell K (1999) Does exposure in forest canopies relate to photoprotective strategies in epiphytic bromeliads? *Funct Ecol* 13:15–23
- Horton P, Ruban AV, Wentworth M (2000) Allosteric regulation of the light harvesting system of photosystem II. *Philos Trans R Soc Lond B Biol* 355:1361–1370
- Hüner NPA, Ivanov AG, Wilson KE, Miskiewicz E, Krol M, Öquist G (2002) Energy sensing and photostasis in photoautotrophs. In: Storey KB, Storey JM (eds) *Sensing signaling and cell adaptation*. Elsevier, Amsterdam, pp 243–255
- Karst J, Gilbert B, Lechowicz MJ (2005) Fern community assembly: the roles of chance and the environment at local and intermediate scales. *Ecology* 86:2473–2486
- Klughammer C, Schreiber U (1994) An improved method, using saturating light pulses, for the determination of photosystem-I quantum yield via P700+ -absorbency at 830 nm. *Planta* 192:261–268
- Kramer DM, Johnson G, Kiirats O, Edwards GE (2004) New fluorescence parameters for the determination of Q(A) redox state and excitation energy fluxes. *Photosynth Res* 79:209–218
- Lambers H, Poorter H (1992) Inherent variation in growth rate between higher plant: a search for physiological causes and ecological consequences. *Adv Ecol Res* 22:187–261
- Lambers H, Chapin FS, Pons T (1998) *Plant physiological ecology*. Springer, Berlin Heidelberg New York
- Larcher W (2003) *Physiological plant ecology*, 4th edn. Springer, Berlin Heidelberg New York
- Leakey ADB, Scholes JD, Press MC (2004) Physiological and ecological significance of sunflecks for dipterocarp seedlings. *J Exp Bot* 56:469–482
- Lichtenthaler H, Wellburn A (1983) Determination of total carotenoids and chlorophyll *a* and *b* of leaf extract in different solvents. *Biochem Soc Trans* 603:591–592
- Long SP, Humphries S, Falkowski PG (1994) Photoinhibition of photosynthesis in nature. *Annu Rev Plant Physiol Plant Mol Biol* 45:633–662
- Lusk CH, Chazdon RL, Hofmann G (2006) A bounded null model explains juvenile tree community structure along light availability gradients in a temperate rain forest. *Oikos* 112:131–137
- Machado JL, Reich PB (1999) Evaluation of several measures of canopy openness as predictors of photosynthetic photon flux density in a forest understory. *Can J For Res* 29:1438–1444
- Maxwell K, Johnson GN (2000) Chlorophyll fluorescence—a practical guide. *J Exp Bot* 51:659–668
- Montgomery RA, Chazdon RL (2002) Light gradient partitioning by tropical tree seedlings in the absence of canopy gaps. *Oecologia* 131:165–174
- Pacala S, Canham C, Silander J, Kobe R (1994) Sapling growth as a function of resources in a northern temperate forest. *Can J For Res* 24:2172–2183
- Page CN (2002) Ecological strategies in fern evolution: a neopterozoological overview. *Rev Palaeobot Palynol* 119:1–33
- Reich P, Walters M, Ellsworth D, Vose J, Volin J, Gresham C, Bowman W (1998) Relationship of leaf dark respiration to leaf nitrogen, specific leaf area and leaf life span: a test across biomes and functional groups. *Oecologia* 114:471–482
- Reyes-Díaz M, Alberdi M, Piper F, Bravo LA, Corcuera LJ (2005) Low temperature responses of *Nothofagus dombeyi* and *Nothofagus nitida*, two evergreen species from south central Chile. *Tree Physiol* 25:1389–1398
- Roberts A, Borland AM, Maxwell K, Griffiths H (1998) Ecophysiology of the C-3 CAM intermediate *Clusia minor* L. in Trinidad: seasonal and short-term photosynthetic characteristics of sun and shade leaves. *J Exp Bot* 49:1563–1573
- Robinson J (1994) Speculations on carbon dioxide starvation, Late Tertiary evolution of stomatal regulation and floristic modernization. *Plant Cell Environ* 17:345–354
- Saldaña A, Lusk CH (2003) Influencia de las especies del dosel en la disponibilidad de recursos y regeneración avanzada en un bosque templado lluvioso del sur de Chile. *Rev Chil Hist Nat* 76:639–650
- Saldaña A, Gianoli E, Lusk CH (2005) Physiological and morphological responses to light availability in three *Blechnum* species (Pteridophyta, Blechnaceae) of different ecological breadth. *Oecologia* 145:252–257
- Saldaña A, Lusk CH, González WL, Gianoli E (2007) Natural selection on ecophysiological traits of a fern species in a temperate rainforest. *Evol Ecol* 21:651–662
- Schreiber U, Schliwa U, Bilger W (1986) Continuous recording of photochemical and non-photochemical chlorophyll fluorescence quenching with a new type of modulation fluorometer. *Photosynth Res* 10:51–62
- Skillman JB, Winter K (1997) High photosynthetic capacity in a shade-tolerant Crassulacean acid metabolism plant. *Plant Physiol* 113:441–450
- Spencer W, Teeri J, Wetzel R (1994) Acclimation of photosynthetic phenotype to environmental heterogeneity. *Ecology* 75:301–314

- Sultan S, Wilczek A, Bell D, Hand G (1998) Physiological response to complex environments in annual *Polygonum* species of contrasting ecological breadth. *Oecologia* 115:564–578
- Valladares F, Pearcy RW (1997) Interactions between water stress, sun-shade acclimation, heat tolerance and photoinhibition in the sclerophyll *Heteromeles arbutifolia*. *Plant Cell Environ* 20:25–36
- Walters RG (2005) Towards an understanding of photosynthetic 460 acclimation. *J Exp Bot* 56:435–447
- Walters M, Field C (1987) Photosynthetic light acclimation in two rainforest *Piper* species with different ecological amplitudes. *Oecologia* 72:449–456
- Woodhouse R, Nobel P (1982) Stipe anatomy, water potentials and xylem conductances in seven species of ferns (Filicopsida). *Am J Bot* 69:135–142