Trees (1998) 12:326–333

ORIGINAL ARTICLE

Roberto Tognetti · Gianfranco Minotta
Simone Pinzauti · Marco Michelozzi
Marco Borghetti

Acclimation to changing light conditions of long-term shade-grown beech (*Fagus sylvatica* L.) seedlings of different geographic origins

Received: 11 April 1997 / Accepted: 11 December 1997

Abstract Effects of changing light conditions on the ecophysiological condition behind survival were examined on beech from two different populations. Plants were grown in a greenhouse under simulated understorey and canopy gap light conditions. Upon exposure to high light maximum photosynthesis of shade-acclimated leaves increased followed by a reduction over several days to between high- and low-light control rates. In the reciprocal transfer, the decrease in maximum photosynthesis was rapid during the first 2–3 days and then levelled off to values comparable to low-light controls. Seedlings from Sicily (Madonie) showed generally higher maximum photosynthetic rates than those from Abetone. Leaf conductance varied in the same direction as photosynthesis in high- to low-light seedlings but to a lesser degree. Leaves grown under low light and exposed to high light experienced photoinhibition. The Abetone population was more susceptible to photoinhibitory damage than the seedlings from Sicily. Exposure to high light of shade-acclimated seedlings resulted in intermediate chlorophyll concentrations between levels of the high-light and low-light seedlings. Carotenoid concentration was unaffected by treatments. Seedlings grew more in high light, but had a lower leaf area ratio. Light-limited seedlings showed a shift in carbon allocation to foliage. Leaves formed in the new light regime maintained the same anatomy that had been developed before transfer. Seedlings from Sicily had thicker leaves than those of seedlings from Abetone. Seedlings from Abetone were found to be more susceptible to changing light conditions than seedlings from Sicily. We conclude that small forest gaps may represent a favorable environment for photosynthesis and growth of beech regeneration as a result of the limited ability of seedlings to acclimate to sudden increases in high irradiance and because of the moderate levels of light stress in small gaps.

Key words *Fagus sylvatica* · Light acclimation · Photoinhibition · Photosynthesis · Population

Introduction

In forest ecosystems acclimation to changing light conditions plays a major role in tree recruitment and competition processes (Chazdon 1988; Küppers 1994). Responses to light fluctuations in both natural and managed patches may simultaneously involve acclimation to high irradiance levels, as well as photoinhibition damage (Mulkey and Pearcy 1992). The increase in light availability over the scale of weeks can lead to differences in photosynthetic characteristics, leaf anatomy and whole-plant growth (Chazdon 1988; Kamaluddin and Grace 1993). The overall response of tree seedlings to canopy opening depends upon their ability to endure sudden exposure to high irradiances. Yet, the potential for acclimation of shade-grown forest trees to a sudden exposure to high light is still not fully understood (cf. Popma and Bongers 1991; Turnbull 1991; Kamaluddin and Grace 1992a, b, 1993; Tognetti et al. 1994, 1997; Johnson et al. 1997).

Beech (*Fagus sylvatica* L.) is a shade-tolerant tree species capable of persisting as small trees for long periods in the forest understorey; it can take advantage of a moderate and gradual increase in light availability associated with gap formation (Stickan and Zhang 1992; Küppers and Schneider 1993; Johnson et al. 1997;

R. Tognetti (✉)
Istituto per l'Agrometeorologia e l'Analisi Ambientale applicata all'Agricoltura, Consiglio Nazionale delle Ricerche,
Piazzale delle Cascine 18, I-50144-Firenze, Italy
Fax: +39-55-308910; e-mail: tognetti@sunserver.iata.fi.cnr.it

G. Minotta
Dipartimento di Colture Arboree, Università di Bologna,
via F. Re 6, I-40126-Bologna, Italy

S. Pinzauti · M. Michelozzi
Istituto Miglioramento Genetico delle Piante Forestali,
Consiglio Nazionale delle Ricerche, Via A. Vannucci 13,
I-50135-Firenze, Italy

M. Borghetti
Dipartimento di Produzione Vegetale, Università della Basilicata,
via N. Sauro 85, I-85100-Potenza, Italy

Minotta and Pinzauti 1996). On the other hand, like many late successional species, it may show limited plasticity in leaf characteristics and thus increased susceptibility to photoinhibition when suddenly exposed to high irradiances (Björkman 1981; Anderson and Osmond 1987). In beech stands natural recruitment is considered a good and inexpensive method of regeneration; unsuccessful cases of natural regeneration are often attributed to unfavourable light conditions (Madsen 1995).

In a recent experiment (Tognetti et al. 1994), shade-grown leaves of beech seedlings, which all derived from sun-leaf primordia, were found to acclimate rather effectively to full light conditions.

In the present experiment, the reaction to increasing irradiances of long-term shade acclimated beech plants, in which all leaves derived from shade-leaf primordia, were studied. The main objective was to test the hypothesis that seedling performance under changing light conditions, which follow overstorey removal and gaps opening in forests, may involve acclimation of pre-existing leaves. To this end 2-year-old seedlings, grown for 2 consecutive years under either shade and moderately high light, were exposed to contrasting light regimes. After the transfer, plants were characterized by measuring gas exchange, fluorescence emission parameters, chlorophyll and carotenoid contents, leaf anatomy, and biomass allocation.

Beech populations are known to differentiate by isoenzyme markers, late winter xylem embolism, growth parameters (Borghetti et al. 1993; Leonardi and Menozzi 1995), morphology and phenology (Borghetti and Giannini 1982), and susceptibility to drought stress (Tognetti et al. 1995). The capability to acclimate to a particular light intensity may be under genetic control and results from light environment prevailing in the native habitat (Boardman 1977). Thus, the hypothesis that acclimation to changing light conditions may depend upon the geographic origin of plant material was evaluated in this experiment by studying beech populations from different geographic origins growing at ecologically contrasting sites.

Materials and methods

Plant material and experimental design

Seedlings of *F. sylvatica* were produced from seeds collected in natural populations growing in Tuscany (Abetone, northern Italy, 44°08′ N and 10°42′ E) and Sicily (Madonie, southern Italy, 37°57′ N and 14°51′ E). After 2 years in a nursery, seedlings were transplanted to 3-l plastic pots, filled with a mixture of fine gravel and local soil, and brought to a greenhouse at the University of Bologna (northern Italy).

In March 1994, before bud break, 144 plants were selected for dimensional uniformity from each population, and divided in two groups of 72 plants each; one group was maintained under a photosynthetic active photon flux density (PPFD) of 500–600 μmol m^{-2} s^{-1} (high light plants, H); the other group was shaded by suspending, 2 m above the plant canopy, a neutral density shade-cloth which reduced PPFD to 20–30 μmol m^{-2} s^{-1} (low light plants, L); PPFD was monitored with a Li-190-S1 quantum sensor (Li-Cor, Lincoln, Neb., USA). Halogen lamps (Osram 41870 WFL) were suspended 50 cm above the plant canopy to eliminate differences in the red/far-red ratio; the lamps were automatically turned on/off 30 min after sunrise and 30 min before sunset; the red/far ratio was repeatedly measured at midday with a Li-1800 portable spectroradiometer (Li-Cor); it fluctuated between 0.85 and 0.95, without significant differences between the low light and the high light treatment.

On 13 June 1995, half of H-plants, of each population, were transferred to low light conditions (HL, i.e. from high to low light), and half of L-plants were transferred to high light conditions (LH, i.e. from low to high light). Half of plants remained as controls: HH, (always under high light) and LL (always under low light). Plants were kept well watered by a drip irrigation system (leaf predawn water potential ranged between –0.08 and –0.28 MPa), fertilized with a 20/20/20 N/P/K commercial slow-release fertilizer, and shifted frequently to minimise positional effects.

Measurements of photosynthesis, leaf conductance and chlorophyll fluorescence

Measurements were performed on seven dates, between June 1 and July 25, on nine plants, each population, per treatment (HH, LL, HL, LH) and one fully expanded leaf per plant; the same leaf (third from the top) was sampled each time.

Maximum photosynthetic rate (A_{max}) was measured under steady-state conditions between 11–14 h with a portable gas exchange open system (LCA-2, ADC, Hoddesdon, UK). Leaf conductance (g_s) was measured with a Li-1600 steady-state null-balance porometer (Li-Cor).

Chlorophyll fluorescence was measured using a PAM 2000 Modulated Fluorimeter (H. Walz, Effeltrich, Germany) on both leaves kept illuminated according to the experimental treatment (‘light-adapted’ leaves), and on leaves which were wrapped for 20 min in aluminium foil (‘dark-adapted’ leaves). All measurements were performed with the fibre optic probe at an angle of about 45° to the leaf.

The following parameters were measured: minimal (F_0), maximal (F_m) and variable ($F_v = F_m – F_0$) fluorescence of ‘dark-adapted’ leaves; steady state (F), minimal (F_0') and maximal (F_m') fluorescence for ‘light-adapted’ leaves; the fluorescence nomenclature follows van Kooten and Snel (1990).

‘Dark-adapted’ leaves were exposed to 15-min stepwise sequences of red light (from 0 to 600 μmol m^{-2} s^{-1}, and then to 0 again to estimate lasting effects); a black sheet covered the leaf and the probe during the measurement. F_0 was determined after an exposure to 3 s of far-red light (600 Hz) to open PSII reaction centres fully. The modulated light source was weak enough (0.1 μmol m^{-2} s^{-1} at 655 nm) to prevent any induction of variable fluorescence. Maximal fluorescence was measured after saturating flushes at 5000 μmol m^{-2} s^{-1} for 0.8 s generated by the internal halogen lamp.

The maximal photochemical efficiency of PSII was calculated as F_v/F_m for ‘dark-adapted’ leaves and as $(F_m'–F_0')/F_m'$ (i.e. F_v'/F_m') for ‘light-adapted’ leaves (Butler 1978); the quantum efficiency of PSII as $\Delta F/F_m'$, where $\Delta F = F_m'–F$ (Genty et al. 1989) and the apparent relative electron transport rate as ETR = $(\Delta F/F_m')$ xPPFD (Bilger et al. 1995). Photochemical quenching, which is used as an estimate of the fraction of PSII open centres, was computed as $q_p = (F_m'–F)/(F_m'–F_0')$, and non-photochemical quenching as NPQ = $(F_m/F_m')–1$ (Bilger and Björkman 1990).

Measurement of chlorophyll and caretonoid pigments

Chlorophyll (Chl *a* and Chl *b*) and total carotenoid concentrations were measured on five plants from each population, per treatment. About 0.5 g (fresh weight, fw) of leaf tissue was macerated in 10 ml *N,N*-dimethylformamide in the dark for 48 h at 4°C; pigment concentrations were determined in the extracts spectrophotometrically (DU-65, Beckman, Fullerton, Calif., USA), and expressed on a leaf dry weight (dw) basis (Moran 1982).

Measurement of biomass, leaf area and leaf anatomy

In November 1995, ten plants of each population per treatment were harvested. Leaf area ratio (LAR) was calculated as the ratio between total plant leaf area and plant dry weight; specific leaf area (SLA) as the ratio between total plant leaf area and total plant leaf weight; leaf weight ratio (LWR) was calculated as the ratio between total leaf weight and plant weight and root/shoot ratio (RSR) as the ratio between below ground and above ground biomass. All weights were measured to the nearest 0.1 mg after 48 h in an oven at 70°C. Leaf area was measured with a Li-3000 meter (Li-Cor).

The thickness of leaves, and of palisade and spongy parenchyma, were measured on 0.5 µm-thin leaf cross-sections under a light microscope, following the procedure described by Bussotti et al. (1995).

Results

Gas exchange, fluorescence and chlorophyll concentration

High light (HH) plants showed a much higher A_{max} than low light (LL) plants, with values generally higher in the Sicily population. In LH plants, A_{max} increased by 50–70% within the first hour of exposure to high light, declined the day after the transfer and stabilized at values higher than LL control plants, but still 40–50% lower than those measured in HH plants (Fig. 1). In the reciprocal transfer (HL plants), A_{max} decreased by 50–60% during the first 2–3 days and levelled off on values equivalent to LL controls. In most cases, HL and LH plants from Sicily showed higher A_{max}. Leaf conductance (g_s) varied in the same direction as A_{max} in HL plants; in LH plants, g_s started to increase four days after the transfer to high light, with a more evident trend in the Sicily population. As for A_{max}, g_s values were generally higher in plants from Sicily (Fig. 1).

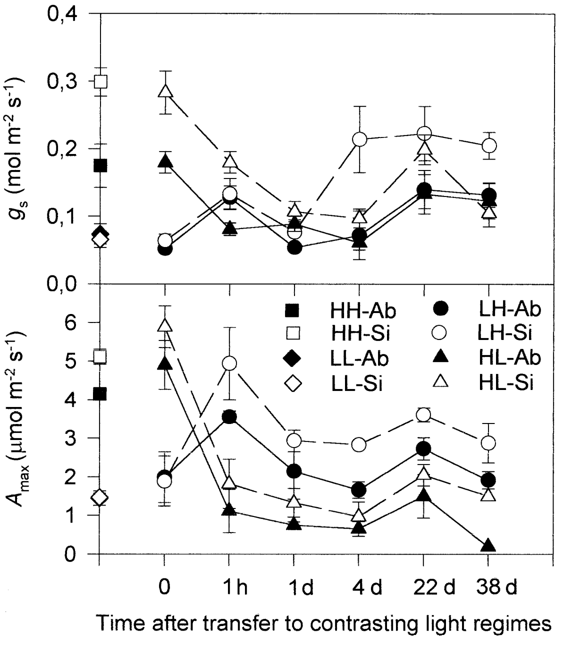

Fig. 1 Time course of leaf conductance and maximum photosynthesis (g_s, upper panel and A_{max}, lower panel) in leaves of beech seedlings which were allowed to develop fully under a high light regime (*HH*), then transferred to a low light regime (*HL*), or, allowed to develop fully under a low light regime (*LL*), then transferred to a high light regime (*LH*). Values are means ± SE for Abetone (*Ab*) and Sicily (*Si*) populations. Overall means instead of time course for HH and LL seedlings are reported on *Y*-axis. On the *X*-axis time is reported as hours (*h*) and days (*d*), and *0* indicates the day of transfer (measurements taken just before)

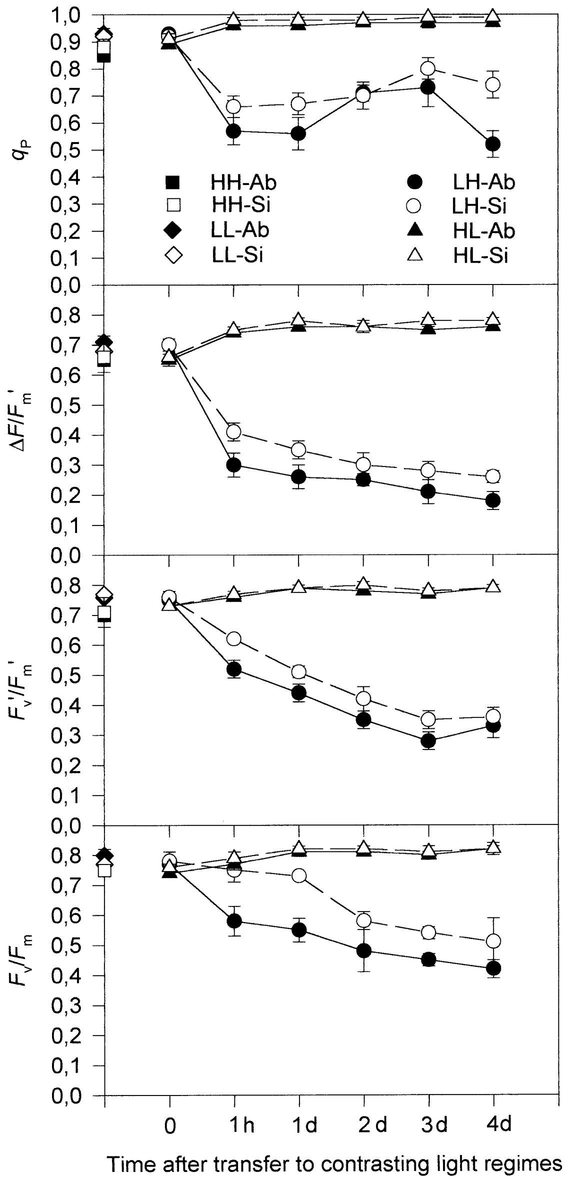

Fig. 2 Time course of chlorophyll fluorescence parameters (q_p, $\Delta F/F_m'$, F_v'/F_m' and F_v/F_m from upper to lower panel) in leaves of beech seedlings which were allowed to develop fully under a high light regime (*HH*), then transferred to a low light regime (*HL*), or, allowed to develop fully under a low light regime (*LL*), then transferred to a high light regime (*LH*). Values are means ± SE for Abetone (*Ab*) and Sicily (*Si*) populations. Overall means instead of time course for HH and LL seedlings are reported on *Y*-axis. On the *X*-axis time is reported as hours (*h*) and days (*d*), and *0* indicates the day of transfer (measurements taken just before)

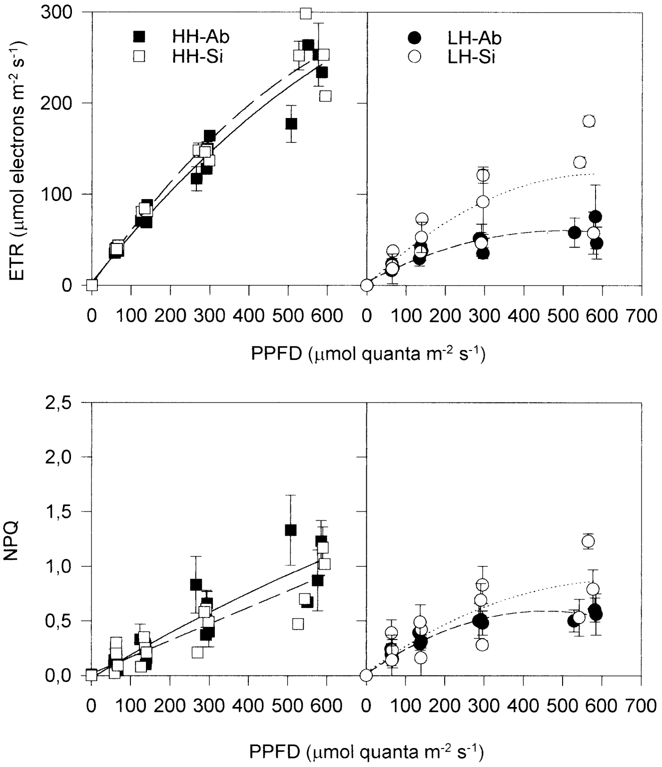

Fig. 3 The relationship of apparent relative electron transport rates (ETR, $\Delta F/F_m'\times$PPFD) to incident PPFD, and non-photochemical quenching (NPQ) in relation to incident PPFD in leaves of beech seedlings which were allowed to develop fully under a high light regime (*HH*), or allowed to develop fully under a low light regime and then transferred to a high light regime (*LH*). Values are means ± SE for Abetone (*Ab*) and Sicily (*Si*) populations. Each line represent a second order polynomial equation fit to the data ($P < 0.001$, for ETR $R^2 = 0.97$, 0.96, 0.87 and 0.72, while for NPQ $R^2 = 0.83$, 0.82, 0.96 and 0.71, respectively, for HH-Ab, HH-Si, LH-Ab and LH-Si). Measurements were taken before transfer, *left panels*, and during 3 days after transfer, *right panels*

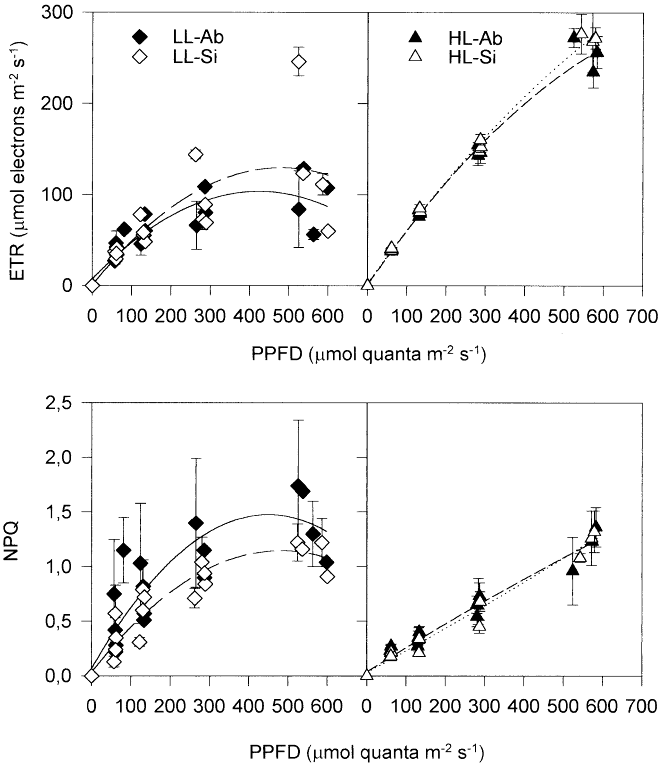

Fig. 4 The relationship of apparent relative electron transport rates (ETR, $\Delta F/F_m'\times$PPFD) to incident PPFD, and non-photochemical quenching (NPQ) in relation to incident PPFD in leaves of beech seedlings which were allowed to develop fully under a high light regime and then transferred to a low light regime (*HL*), or allowed to develop fully under a low light regime (*LL*). Values are means ± SE for Abetone (*Ab*) and Sicily (*Si*) populations. Each line represent a second order polynomial equation fit to the data ($P < 0.001$, for ETR $R^2 = 0.80$, 0.65, 0.99 and 1, while for NPQ $R^2 = 0.79$, 0.90, 0.97 and 0.97, respectively, for LL-Ab, LL-Si, HL-Ab and HL-Si). Measurements were taken before transfer, *left panels*, and during 3 days after transfer, *right panels*

Modulated fluorescence was utilized for evaluating the functioning of the photosynthetic machinery soon after the exposure to contrasting light regimes (Fig. 2). In LH plants, $\Delta F/F_m'$, F_v'/F_m', F_v/F_m and q_p sharply decreased soon after the transfer to high light; 4 days after the transfer, these parameters were 50–80% of those measured on LL control plants; in most cases lower values were found in the Abetone plants. As measured as 1-q_p, 30–40% of the PSII reaction centres remained open in Abetone plants, and 20–30% in those from Sicily. In HL plants, the same fluorescence parameters showed a slight increase after transfer to shade that was similar in the two populations, and remained slightly above HH control plants. In both HH and LL control plants, fluorescence parameters remained relatively constant over the entire study, (Fig. 2); only bulk-averaged values are reported for clarity.

For ‘dark-adapted’ leaves exposed to 15-min stepwise sequences of red light, the response of ETR and NPQ to incident PPFD, before and after transfer to contrasting light conditions, is depicted in Figs. 3 and 4. Up to a PPFD of 500–600 μmol m^{-2} s^{-1} an almost linear increase of ETR was observed in HH plants (data were more scattered at higher PPFD). ETR of LL plants saturated at a considerably lower value, 250–300 μmol m^{-2} s^{-1}, despite LL plants from Sicily showed a high point at 500 μmol m^{-2} s^{-1}. LH and HL plants displayed trends similar to LL and HH plants, respectively. NPQ increased with PPFD, but the relationship was scattered, particularly in LL plants. A more pronounced response of NPQ to increasing PPFD was observed in HL as compared to LH plants. Lasting effects were negligible (data not shown). Differences between population were relatively evident only for LH plants, those from Sicily showing higher values of ETR and NPQ at increasing PPFD values.

Chlorophyll concentration (on a dry weight basis) was much higher in LL than in HH plants, and in the Sicily as compared to the Abetone population. In response to shading, chlorophyll concentration increased in HL plants from Sicily, but not in those from Abetone. In LH plants the chlorophyll concentration decreased after exposure to light, without differences between populations

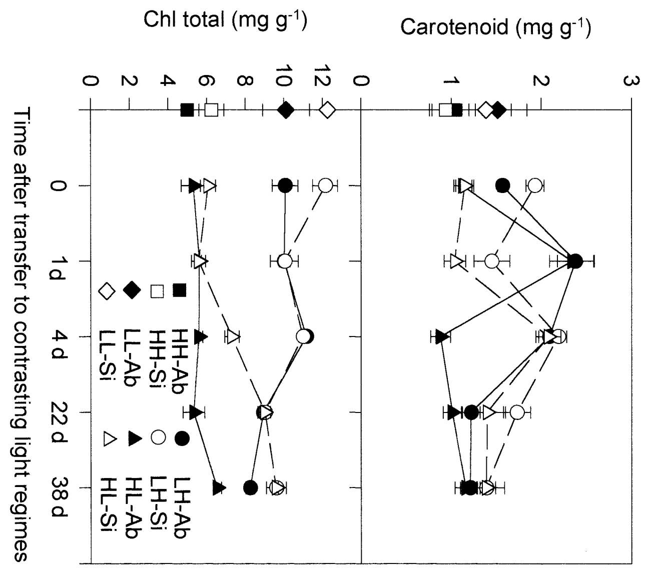

Fig. 5 Time course of pigment concentrations (Carotenoid, *upper panel* and Chl total, *lower panel*) in leaves of beech seedlings which were allowed to develop fully under a high light regime (*HH*), then transferred to a low light regime (*HL*), or, allowed to develop fully under a low light regime (*LL*), then transferred to a high light regime (*LH*). Values are means ± SE for Abetone (*Ab*) and Sicily (*Si*) populations. Overall means instead of time course for HH and LL seedlings are reported on *Y*-axis. On the *X*-axis time is reported as hours (*h*) and days (*d*), and *0* indicates the day of transfer (measurements taken just before)

(Fig. 5). The ratio between chlorophyll *a* and *b* (Chl *a*/*b*) was higher ($P < 0.01$) in HH than in LL plants (4.40 ± 0.45 vs 3.35 ± 0.11), and in Abetone with respect to the Sicily population (3.63 ± 0.12 vs 2.72 ± 0.10). Chl *a*/*b* decreased after transfer to low light in HL plants, by 30% and 10% for the Abetone and Sicily population. In contrast, no change was observed in LH plants after transfer to high light.

No regular trend of carotenoid concentration was observed during the experiment after the transfer to contrasting light levels (Fig. 5); in HL plants the value after 38 days was not different from the initial one; in contrast, a slight decrease was observed in LH plants.

Biomass, leaf area and leaf anatomy

Whole-plant, root, stem and foliage biomass were significantly larger in HH plants, as compared to the other treatments, and in the Abetone population; HL and LH plants did not differ, showing values which are intermediate between those found in HH and LL plants. RSR and LWR were larger in HH and in HL plants, respectively, and in the Sicily as compared to the Abetone population. Total leaf area per plant, LAR and SLA were larger in the Abetone population. Total leaf area and the number of leaves per plant were lowest in LL plants,

Table 1 Growth characteristics and anatomical features of leaves of beech seedlings from two Italian populations (Abetone and Sicily) 4 months after transfer to the contrasting light regime (*HH*, permanently under high light conditions; *LL*, permanently under low light conditions; *LH*, fully expanded under low light conditions and then transferred to high light conditions; *HL*, fully expanded under high light conditions and then transferred to low light conditions). Values are means, g DW, m^2 or mm, for individual plants in each treatment or population. Different letters within a row and population indicate statistically different values at $P \leq 0.05$ (Duncan's New Multiple Range Test). Summary of the analysis of variance (ANOVA), *F*-ratio (*$P<0.05$, ** $P<0.01$, *** $P<0.001$, **** $P<0.0001$, ns not significant), is also reported

Parameter	Abetone				Sicily				Population (P) *F*-ratio	Treatment (T) *F*-ratio	P ×T interaction *F*-ratio
	HH	LL	LH	HL	HH	LL	LH	HL			
Foliage DW (g)	4.21a	1.27b	2.98a	3.34a	2.33a	0.87b	1.52c	2.02a	31.6881****	16.5343****	1.8367ns
Stem DW (g)	11.35a	5.13b	8.78a	8.87a	7.88a	3.64b	5.49c	5.62c	31.3637****	16.8067****	0.7755ns
Roots DW (g)	20.80a	4.98b	11.40c	11.28c	15.14a	4.29b	7.65c	8.92c	14.7905***	45.7859****	1.6558ns
Total DW (g)	36.35a	11.39b	23.17c	23.49c	25.35a	8.80b	14.66c	16.55c	25.0194****	33.7011****	1.4340ns
RSR ($g\ g^{-1}$)	1.35a	0.83b	0.98b	0.93b	1.51a	0.96b	1.09bc	1.17c	12.5643***	25.0440****	0.3613ns
LWR ($g\ g^{-1}$)	0.115a	0.110a	0.124a	0.141b	0.092a	0.100a	0.106a	0.123b	19.2787****	10.7164****	0.4554ns
Leaf area (m^2)	0.089a	0.046b	0.087a	0.091a	0.039ab	0.030b	0.042ab	0.044a	69.7823****	8.0266****[a]	2.6683*[b]
Number of leaves per plant	110.5a	45.9b	84.9a	93.9a	85.2a	57.7b	73.7ab	82.2ab	2.0518ns	9.3325****	1.4090ns
LAR ($m^2\ g^{-1}$)	0.0025a	0.0040b	0.0037b	0.0039b	0.0015a	0.0034b	0.0029c	0.0027c	63.3021****	45.6503****	1.6430ns
SLA ($m^2\ g^{-1}$)	0.0212a	0.0368b	0.0306c	0.0281c	0.0167a	0.0349b	0.0276c	0.0218d	27.6373****	90.0040****	1.6677ns
Leaf thickness (μm)	81.88a	60.72b	55.27b	83.56a	111.96a	71.67b	70.56b	109.55a	55.9131****	49.9858****	3.4904*
Palisade thickness (μm)	29.71a	18.57b	14.84b	27.11a	47.91a	21.54b	25.62b	44.93a	89.5506****	69.1905****	9.9404****[c]
Spongy thickness (μm)	35.65ab	26.37a	27.89ab	37.14b	45.31a	33.05b	27.48b	47.53a	8.4790**	10.9238****	0.9859ns
Palisade/spongy (μm mm^{-1})	0.87a	0.72ab	0.54b	0.74ab	1.08a	0.66b	0.97a	0.96a	10.8232**	5.9086**	3.0816*

[a, b, c] $P=0.00011$, 0.05425 and 0.00012, respectively

while LAR and SLA were lowest in HH plants; HL and LH plants showed intermediate values (Table 1).

The thickness of leaf, palisade and spongy parenchyma followed this order: (HH = HL) > (LH = LL), Sicily > Abetone. Significant interactions between treatments and population were found in several cases (Table 1). HH and LL plants did not differ in their leaf anatomy throughout the season.

Discussion

The down-regulation of photosynthesis in plants grown at low irradiance and transferred to high irradiance (as LH plants) can be attributed to the depletion of RuBP by Rubisco, coupled with the reduced energy supply because of slower electron transport (Stitt and Schulze 1994). On the other hand, the increase in leaf conductance shown by LH plants (particularly those from Sicily) may allow high intercellular levels of CO_2, enhancing quantum yield for CO_2 (Pearcy 1987; Tognetti et al. 1997; Johnson et al. 1997), and evaporative leaf cooling (Sims and Pearcy 1991), which all may represent acclimation mechanisms for understorey plants exposed to sunflecks in small canopy gaps.

The ratio between assimilation rate and stomatal conductance suggests a more conservative leaf-level water-use efficiency in HH with respect to LH plants; thus, light-acclimated plants can be considered as more able to survive in a more xeric habitat (such as a gap), resulting from increased evaporative demand and light intensity (Ellsworth and Reich 1992; Johnson et al. 1997). In shaded plants (LL plants) carbon gain is impaired by stomatal closure more than water loss, which is interpretable as acclimation to the light-limited, but not water-limited, understorey environment (Küppers and Schneider 1993; Johnson et al. 1997).

Drastic changes in irradiance may induce photoinhibition in shade-acclimated leaves (Björkman 1981; Ögren and Sjöström 1990; Rosenqvist et al. 1991; Johnson et al. 1997). In agreement with other studies (Wallace and Dunn 1980; Fetcher et al. 1983; Langenheim et al. 1984; Kamaluddin and Grace 1992a, b, 1993; Tognetti et al. 1997; Johnson et al. 1997), when shade-acclimated plants were transferred to high light, the decline in photosynthetic rate was paralleled by a reduction in the photochemical efficiency of PSII, PSII quantum yield and photochemical quenching. According to Havaux et al. (1991) the efficiency of open PSII reaction centres is an approximate measure of their trapping efficiency; under photoinhibitory conditions reduced plastoquinone accumulates, resulting in the inhibition of electron transport (Kyle et al. 1984). The decrease of efficiency of open PSII centres and reduction of photochemical quenching indicates that the fraction of closed PSII centres increased in leaves transferred from low to high light (Chow et al. 1989; Björkman 1987). At a given irradiance, photochemical quenching was different for LH and HH plants; thus, we may argue that the decline in the quantum efficiency of PSII was not only due to the reduction of the efficiency of open PSII centres.

In the case of stepwise exposure to increasing irradiance, the quantum efficiency of PSII electron transport decreased in all treatments. However, under irradiance high enough to saturate photosynthesis, LH plants responded much more than HH plants, and LL plants than HL plants. Thus, it seems that PSII reaction centres of sun and shade beech plants do not have the same intrinsic susceptibilities to photoinhibition. Therefore, our data do not support the hypothesis that photoinhibition of PSII reaction centres is related to long-term down-regulation of photochemistry (Öquist et al. 1992). On the other hand, in all treatments (to a lesser extent in LH seedlings from Abetone compared to those from Sicily), non-photochemical quenching (NPQ), which is related to radiationless photoprotective dissipation processes (Schreiber and Bilger 1987; Osmond et al. 1993), increased with irradiance: at 600 µmol m^{-2} s^{-1} NPQ was between 1 and 1.5 in HH and HL plants, which are values similar to those found for leaves of mature beech trees by Bilger et al. (1995).

No carotenoid (total concentration) biosynthesis, interpretable as a photoprotective strategy (Demmig-Adams and Adams 1992), can be suggested for LH seedlings. LH Abetone plants were found to be more susceptible to photoinhibitory damage, as they showed limited capacity of thermal dissipation at the PSII level and retained a more reduced plastochinone pool at high light. Following Bilger et al. (1995), apparent electron transport rate (ETR) was estimated to be in the range 200–300 µmol m^{-2} s^{-1} for HH seedlings and of about 100 µmol m^{-2} s^{-1} for LL seedlings; low ETR values were more lasting in the Abetone plants. Studying the same populations, Tognetti et al. (1997) showed a tendency for prolonged photoinhibition of PSII following changes in irradiance, suggesting no readily reversible damages to PSII centres; similar results are reported by Layne and Flore in *Prunus cerasus* (1993).

The highest chlorophyll concentration was found in shaded plants, in accordance with previous results on beech (Johnson et al. 1997; Minotta and Pinzauti 1996) and on sugar maple (Ellsworth and Reich 1992). Substantial nitrogen cost is linked to high chlorophyll contents in shaded plants (Evans 1989), and adjustment to low light regimes can be interpreted as a switch in the allocation of nitrogen resources from Rubisco and electron transport proteins towards associated chlorophyll proteins (Field 1983), thus improving light interception.

An only slight decline of chlorophyll concentrations was found in LH seedlings, thus providing a chance for long-term partial recovery of shade-acclimated leaves. A more rapid and substantial decline in chlorophyll concentrations was observed, after transfer to high irradiance, in shade-acclimated leaves originated from sun-leaf primordia (Tognetti et al. 1994). The increase of chlorophyll concentration observed in HL seedlings from Sicily may allow them to couple with changing light conditions (for instance sunflecks in canopy gaps) better than seedlings from Abetone, which did not show adjustments of chlorophyll content with decreasing irradiances.

Differences between treatments in the chlorophyll *a*/*b* ratio suggest that leaves were structurally different; the lower chlorophyll *a*/*b* ratio in HL and LL plants may be related to increased thylakoid membrane stacking (shade-type chloroplasts) containing predominantly ligth-harvesting units (Virzo de Santo et al. 1984). It is worth noting that chlorophyll *a*/*b* ratio in HL plants exhibited a more pronounced decrease in Abetone with respect to the Sicily population.

Seedlings grew bigger under high light conditions as a result of enhanced net assimilation rate, despite a lower leaf area ratio. Significant differences in carbon allocation have been observed between light treatments. In LL, HL and LH seedlings, in which photosynthesis was a limiting factor, carbon was preferentially allocated to shoot growth to improve light interception (Cannell 1985; Johnson et al. 1997; Minotta and Pinzauti 1996). The higher proportion of carbon allocated to roots in Sicily plants, with respect to Abetone plants, could represent an adaptation to drought conditions at their geographic origin (Borghetti et al. 1993).

Even fully differentiated and mature leaves can acclimate to new light environments through re-organisation of photosynthetic machinery and leaf anatomy (Bauer and Thöni 1988; Kamaluddin and Grace 1992a, b, 1993). However, significant ‘carry over’ effects of ‘previous’ light environment on leaf morhology and plant allometry have often been reported (Oberbauer and Strain 1985; Kamaluddin and Grace 1993; Bongers et al. 1988; Fetcher et al. 1983), and may explain why in the present experiment leaves differentiated and produced before the transfer to a new light environment maintained their anatomy and physiology to a large extent. ‘Carry over’ effects can be related also to the environment under which leaf primordia have developed; since the induction of sun and shade leaves in beech occurs during the previous year (Eschrich et al. 1989), HL and LH seedlings had leaves originating from sun-leaf and shade-leaf primordia, respectively. Photoinhibition damage might have also prevented leaves of LH seedlings from anatomical rearrangements and new tissue formation (Bauer and Thöni 1988; Kamaluddin and Grace 1992a, b, 1993). The higher photosynthetic capacity observed in HH and Sicily plants can be related with their thicker palisade tissue and denser mesophyll, implying a higher number of chloroplasts and a greater photosynthetically active mass per unit leaf area (Björkman 1981). It is worth noting that LH plants from Sicily had a higher palisade/spongy ratio than LH plants from Abetone.

In conclusion, small forest gaps may represent a favorable environment for photosynthesis and growth of beech seedlings, given their capability to cope with the moderate levels of light stress in small canopy openings, where sunflecks play a major role (Johnson et al. 1997). Overall, it is reasonable to assume that survival and growth of beech seedlings following gap formation may depend more on competition from successional species with high photosynthetic rates than on acclimation of the photosynthetic apparatus of pre-existing shade acclimated seedlings. Seedlings from the Abetone population were confirmed to be more susceptible to changing light conditions than those from Sicily (Tognetti et al. 1997). In Sicily beech generally grows in a more open habitat and may have developed physiological and morphological mechanisms to utilise more effectively a high light environment.

Acknowledgements Research supported by MURST 40% (Effetto sugli ecosistemi forestali dei cambiamenti climatici e del territorio) and EU-LTEEF project (PL931511) ‘Long-term effects of CO_2 increase and climate change on the European forests’. We thank G. Scarascia-Mugnozza (University of Tuscia, Viterbo, Italy) for supplying the PAM 2000 modulated fluorimeter. We are grateful to J.D. Johnson (Intensive Forestry Program, Washingotn State University, Puyallup, USA) for useful comments on the manuscript.

References

ReferencesAnderson JM, Osmond CB (1987) Shade-sun responses: compromises between acclimation and photoinhibition. In: Kyle DJ, Osmond CB, Arntzen CJ (eds) Photoinhibition. Elsevier, Amsterdam, pp 1–38

Bauer H, Thöni W (1988) Photosynthetic light acclimation in fully developed leaves of the juvenile and adult phases of *Hedera helix*. Physiol Plant 73: 31–37

Bilger W, Björkman O (1990) Role of xanthophyll cycle in photoprotection elucidated by measurements of light-induced absorbance changes, fluorescence and photosynthesis in leaves of *Hedera canariensis*. Photosynth Res 25: 173–186

Bilger W, Schreiber U, Bock M, (1995) Determination of the quantum efficiency of photosystem II and non-photochemical quenching of chlorophyll fluorescence in the field. Oecologia 102: 425–432

Björkman O (1981) Responses to different quantum flux densities. In: Lange OL, Nobel PS, Osmond CB, Ziegler H (eds) Encyclopedia of plant physiology, NS, vol 12A. Springer, Berlin Heidelberg New York, pp 57–107

Björkman O (1987) High-irradiance stress in higher plants and interaction with other stress factors. In: Biggens J (ed) Progress in photosynthesis research, vol IV. Martinus Nijhoff, Dordrecht, pp 11–18

Boardman NK (1977) Comparative photosynthesis of sun and shade plants. Annu Rev Plant Physiol 28: 355–377

Bongers F, Popma J, Iriarte-Vivar S (1988) Response of *Cordia megalantha* seedlings to gap environments in tropical rain forest. Funct Ecol 2: 379–390

Borghetti M, Giannini R (1982) Indagini preliminari sulla variazione di alcuni caratteri in piantine di faggio di provenienza diversa. Ann Acc It Sci For XXXI: 119–134

Borghetti M, Leonardi S, Raschi A, Snyderman D, Tognetti R (1993) Ecotypic variation of xylem embolism, phenological traits, growth parameters and allozyme characteristics in *Fagus sylvatica*. Funct Ecol 7: 713–720

Bussotti F, Bottacci A, Bartolesi A, Grossoni P, Tani C (1995) Morpho-anatomical alterations in leaves collected from beech trees (*Fagus sylvatica* L.) in conditions of natural water stress. Env Exp Bot 35: 201–213

Butler WL (1978) Energy distribution in the photosynthetical apparatus of photosynthesis. Annu Rev Plant Physiol 29: 345–378

Cannell MGR (1985) Dry-matter partitioning in tree crops. In: Cannell MGR, Jackson JE (eds) Attributes of trees as crop plants. Institute of Terrestrial Ecology, Huntingdon, pp 160–193

Chazdon RL (1988) Sunflecks and their importance to forest understorey plants. Adv Ecol Res 18: 1–63

Chow WS, Osmond CB, Huang LK (1989) Photosystem function and herbicide binding sites during photoinhibition of spinach chloroplasts in-vivo and in-vitro. Photosynth Res 21:17–26

Demmig-Adams B, Adams WW III (1992) Photoprotection and others responses of plants to high light stress. Annu Rev Plant Physiol Plant Mol Biol 43: 599–626

Ellsworth DS, Reich PB (1992) Leaf mass per area, nitrogen content and photosynthetic carbon gain in *Acer saccharum* seedlings in contrasting forest light environments. Funct Ecol 6: 423–435

Eschrich W, Buchardt R, Essiamah S (1989) The induction of sun and shade leaves of the European beech (*Fagus sylvatica* L.): anatomical studies. Trees 3: 1–10

Evans JR (1989) Partitioning of nitrogen between and within leaves grown under different irradiances. Aust J Plant Physiol 16: 533–548

Fetcher N, Strain BR, Oberbauer SF (1983) Effects of light regime on the growth, leaf morphology and water relations of seedlings of two species of tropical trees. Oecologia 58: 314–319

Field C (1983) Allocating leaf nitrogen for the maximisation of carbon gains: leaf age as a control on the allocation program. Oecologia 56: 341–347

Genty B, Briantais JM, Baker NR (1989) The relationship between the quantum yield of phptosynthetic electron transport and quenching of chlorophyll fluorescence. Biochim Biophys Acta 990: 87–92

Havaux M, Strasser RJ, Greppin H (1991) A theoretical and experimental analysis of the qP and qN coefficients of chlorophyll fluorescence quenching and their relation to photochemical and nonphotochemical events. Photosynth Res 27: 41–55

Johnson JD, Tognetti R, Michelozzi M, Pinzauti S, Minotta G, Borghetti M (1997) Ecophysiological responses of *Fagus sylvatica* L. seedlings to changing light conditions. II. The interaction of light environment and soil fertility on seedling physiology. Physiol Plant 101: 124–134

Kamaluddin M, Grace J (1992a) Photoinhibition of photosynthesis and light acclimation in seedlings of *Bishofia javanica*, a tropical forest tree from Asia. Ann Bot 69: 47–52

Kamaluddin M, Grace J (1992b) Acclimation in seedlings of a tropical trees, *Bishofia javanica*, following a stepwise reduction in light. Ann Bot 69: 557–562

Kamaluddin M, Grace J (1993) Growth and photosynthesis of tropical forest tree seedlings (*Bishofia javanica* Blume) as influenced by change in light availability. Tree Physiol 13: 189–201

Kooten O van, Snel JFH (1990) The use of chlorophyll fluorescence nomenclature in plant stress physiology. Photosynth Res 25: 147–150

Küppers M (1994) Canopy gaps: competitive light interception and economic space filling - a matter of whole-plant allocation. In: Caldwell MM, Pearcy RW (eds) Exploitation of environmental heterogeneity by plants. Academic Press, San Diego, pp 111–144

Küppers M, Schneider H (1993) Leaf gas exchamnge of beech (*Fagus sylvatica* L.) seedlings in lightflecks: effects of fleck lenght and leaf temperature in leaves grown in deep and partial shade. Trees 7: 160–168

Kyle DJ, Ohad I, Arntzen CJ (1984) Membrane protein damage and repair: selective loss of a quinone-protein function in chloroplast membranes. Proc Natl Acad Sci USA 81: 4070–4074

Langenheim JH, Osmond CB, Brooks A, Ferrar PJ (1984) Photosynthetic responses to light in seedlings of selected Amazonian and Australian rainforest tree species. Oecologia 63: 215–224

Layne DR, Flore JA (1993) Physiological responses of *Prunus cerasus* to whole-plant source manipulation. Leaf gas exchange, chlorophyll fluorescence, water relations and carbohydrate concentrations. Physiol Plant 88: 44–51

Leonardi S, Menozzi P (1995) Genetic variability of *Fagus sylvatica* L. in Italy: the role of postglacial recolonization. Heredity 75: 35–44

Madsen P (1995) Effects of soil water content, fertilization, light, weed competition and seedbed type on natural regeneration of beech (*Fagus sylvatica*). For Ecol Manage 72: 251–264

Minotta G, Pinzauti S (1996) Effects of light and soil fertility on growth, leaf chlorophyll content and nutrient use efficiency of beech (*Fagus sylvatica* L.) seedlings. For Ecol Manage 86: 61–71

Moran R (1982) Formulae for determination of chlorophyllous pigments extracted with *N*,*N*-dimethylformamide. Plant Physiol 69: 1376–1381

Mulkey SS, Pearcy RW (1992) Interactions between acclimation and photoinhibition of photosynthesis of a tropical forest understorey herb, *Alocasia macrorrhiza*, during simulated canopy gap formation. Funct Ecol 6: 719–729

Oberbauer SF, Strain BR (1985) Effects of light regime on the growth and physiology of *Pentaclethra macroloba* (Mimosaceae) in Costa Rica. J Trop Ecol 1: 303–320

Ögren E, Sjöström M (1990) Estimation of the effect of photoinhibition on the carbon gain in leaves of a willow canopy. Planta 181: 560–567

Öquist G, Chow WS, Anderson JM (1992) Photoinhibition of photosynthesis represents a mechanism for long-term regulation of photosystem II. Planta 186: 450–460

Osmond CB, Ramus J, Levavasseur G, Franklin LA, Henley WJ (1993) Fluorescence quenching during photosynthesis and photoinhibition of *Ulva rotundata* Blid. Planta 190: 97–106

Pearcy RW (1987) Photosynthetic gas exchange responses of Australian tropical forest trees in canopy, gap and understory micro-environments. Funct Ecol 1: 169–178

Popma J, Bongers F (1991) Acclimation of seedlings of three Mexican tropical rain forest species to a change in light availability. J Trop Ecol 7: 85–97

Rosenqvist E, Wingsle G, Ögren E (1991) Photoinhibition of photosynthesis in intact willow leaves in response to moderate changes in light and temperature. Physiol Plant 83: 390–396

Schreiber U, Bilger W (1987) Assessment of stress effects on plant leaves by chlorophyll fluorescence measurements. In: Tenhunen JD, Catarino FM, Lange OL, Oechel WC (eds) Plant response to stress - functional analysis in Mediterranean ecosystems, NATO Advanced Science Institute series. Springer, Berlin Heidelberg New York, pp 27–53

Sims DA, Pearcy RW (1991) Photosynthesis and respiration in *Alocasia macrorrhiza*, following transfers to high and low light. Oecologia 86: 447–453

Stickan W, Zhang X (1992) Seasonal changes in CO2 and H2O gas exchange of young European beech (*Fagus sylvatica* L.). Trees 6: 96–102

Stitt M, Schulze E-D (1994) Does Rubisco control the rate of photosynthesis and plant growth? An exercise in molecular ecophysiology. Plant Cell Environ 17: 465–487

Tognetti R, Michelozzi M, Borghetti M (1994) Response to light of shade-grown beech seedlings subjected to different watering regimes. Tree Physiol 14: 751–758

Tognetti R, Johnson JD, Michelozzi M (1995) The response of European beech (*Fagus sylvatica* L.) seedlings from two Italian populations to drought and recovery. Trees 9: 348–354

Tognetti R, Johnson JD, Michelozzi M (1997) Ecophysiological responses of *Fagus sylvatica* L. seedlings to changing light conditions. I. Interaction between photosynthetic acclimation and photoinhibition during simulated canopy gap formation. Physiol Plant 101: 115–123

Turnbull MH (1991) The effects of light quantity and quality during development on the photosynthetic characters of six Australian rain forest tree species. Oecologia 87: 110–117

Virzo De Santo A, Ligrone R, Alfani A, Fioretto A, Russo G (1984) CAM activity and day/night changes in the ultrastructure of stem chlorenchyma *of Cissus quadrangolaris* L. as influenced by drought. Plant Cell Environ 7: 105–112

Wallace LL, Dunn EL (1980) Comparative photosynthesis of three gap phase successional tree species. Oecologia 45: 331–334