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Instantaneous photosynthetic responses to temperature of deciduous and evergreen *Nothofagus* species

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Abstract. Deciduous species of *Nothofagus* tend to replace evergreen *Nothofagus* at the highest altitudes. We investigated whether deciduous *Nothofagus* species have higher maximum rates of net photosynthesis (P_{max}) and specific leaf area (SLA) than evergreen species and whether there is an increasing photosynthetic advantage (e.g. higher P_{max}) in deciduous species relative to evergreen species with increasing altitude that might explain their replacement of evergreen species. Net photosynthesis was investigated in (1) five deciduous and five evergreen species of *Nothofagus* grown in a common environment and (2) two co-occurring species, *N. gunnii* (Hook.f.) Oerst. (deciduous) and *N. cunninghamii* (Hook.) Oerst. (evergreen), across a range of altitudes in Tasmania. In the first experiment, the maximum rate of net photosynthesis per leaf mass ($P_{max,\ mass}$) and SLA were higher in deciduous species, whereas the maximum rate of net photosynthesis per leaf area ($P_{max,\ area}$) did not differ between leaf habits. However, in the field-based study, both mass- and area-based P_{max} were higher in *N. gunnii* than *N. cunninghamii* across all sites. The high $P_{max,\ mass}$ of deciduous species may provide a competitive advantage at higher altitudes by maximising carbon gain during the growing season. However, in the study of sympatric populations of *N. gunnii* and *N. cunninghamii* there was no evidence of increasing photosynthetic advantage of the deciduous *N. gunnii* with increasing altitude.

Introduction

There are few cold-deciduous species in the Southern Hemisphere compared with the Northern Hemisphere, although dry-deciduous species are common in tropical areas of the Southern Hemisphere that have a pronounced dry season. The current dominance of temperate Southern Hemisphere vegetation by evergreen species has been attributed to a more temperate, oceanic climate, with adequate precipitation throughout the year and the lack of severe frosts compared with the Northern Hemisphere (Axelrod 1966). Climatic and biogeographic history may also have influenced the current patterns of cold adaptation, particularly in Australia and New Zealand (Read and Francis 1992; Hill and Scriven 1995). Nevertheless, cold-deciduous species in both hemispheres are generally restricted to environments that have a severe winter.

Nothofagus is the predominant tree genus in the Southern Hemisphere containing cold-deciduous species. Of its 35 extant species, there are seven deciduous species, six in South America and one, *Nothofagus gunnii*, in Tasmania, Australia. This genus is useful for studies of the adaptive significance of deciduousness, and leaf lifespan more generally, for several reasons. First, the genus occurs across a wide range of environments (both soil and climate) from 0 to 55°S and from sea level to subalpine environments. Second, it has a long and relatively detailed fossil record that indicates a range of deciduous species occurred in regions where these are now rare. Hence, the disappearance of deciduous species can potentially be linked to changing environments (e.g. Read and Francis 1992; Scriven and Hill 1996). Third, there is considerable variation in leaf lifespan, with a range of *c.* 5 months to 5 years across all species and a range of *c.* 1–5 years just among evergreen species (Table 1). Finally, the co-occurrence of some deciduous and evergreen species is valuable for comparative studies of the adaptive significance of differences in leaf lifespan. For example, in Tasmania, the deciduous *N. gunnii* and the evergreen *N. cunninghamii* often co-occur at intermediate to high altitudes (550–1260 m), but *N. gunnii* can extend up to *c.* 100–250 m above *N. cunninghamii*.

Deciduousness allows the avoidance of costs associated with survival of a stressful season (whether cold or dry) (Chabot and Hicks 1982; Kikuzawa 1991). Both severity and duration of the unfavourable period may influence

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Table 1. Deciduous and evergreen *Nothofagus* species used in this study; their natural distribution, origin and leaf lifespan

Species	Subgenus	Leaf lifespan	Natural distribution	Origin of study plants
Deciduous species				
N. alpina (Poepp. & Endl.) Oerst.	*Lophozonia*	7 months[A]	Chile and Argentina, near sea level–1000 m asl[A]	Chile, no details
N. antarctica (G.Forst.) Oerst.	*Nothofagus*	5–6 months[B]	Chile and Argentina, 0–2000 m asl[A]	Chile, no details
N. gunnii (Hook.F.) Oerst.	*Fuscospora*	5–6 months	Australia (Tasmania) 550–1500 m asl[G]	Mt Field: 42°41′S, 146°38′E, 1020 m Mt Read: 41°51′S, 145°33′E, 760 m
N. obliqua (Mirb.) Oerst.	*Lophozonia*	7 months[A]	Chile and Argentina, near sea level–*c.* 2500 m asl[A,H]	Chile, no details
N. pumilio Reiche	*Nothofagus*	5 months[A]	Chile and Argentina, 0–2000 m asl[A]	Chile, no details
Evergreen species				
N. betuloides (Mirb.) Oerst.	*Nothofagus*	28 months[C]	Chile and Argentina, 0–1200 m asl[A]	Chile, no details
N. cunninghamii (Hook.) Oerst. (Hook.f.) Oerst.	*Lophozonia*	3(–5) years[D]	Australia (Tasmania, Victoria), 0–1450 m asl	Mt Field: as above Mt Read: as above Sumac: 41°9′S, 145°1′E, 180 m
N. fusca (Hook.f.) Oerst.	*Fuscospora*	12 months[E]	New Zealand	New Zealand, no details
N. menziesii (Hook.f.) Oerst.	*Lophozonia*	3(–5) years[E]	New Zealand	New Zealand, no details
N. moorei (F.Muell.) Maiden.	*Lophozonia*	2 years[F]	Australia (NSW and southern Queensland)	New England NP: 30°32′S, 152°24′E, *c.* 1200 m

[A] Veblen *et al.* (1996).
[B] A. Lara, pers. comm.
[C] R. Godoy, pers. comm.
[D] Usually a little over 3 years (Howard 1973; Read and Brown 1996), but may live up to 5 years (Kohout 2001).
[E] Usually a little over 3 years, but may live up to 5 years (Ogden *et al.* 1996).
[F] Selman and Lowman (1983).
[G] High elevation record from Robertson and Duncan (1991).
[H] Donoso (1996).

leaf performance and longevity. As the duration of the unfavourable period increases, deciduousness becomes more advantageous by avoiding leaf maintenance costs during the unfavourable season when photosynthesis is limited (Kikuzawa 1991; Kikuzawa and Kudo 1995; Kudo *et al.* 2001) and avoiding the costs of building a more robust leaf to survive stress. However, the seasonal absence of leaves can result in loss of assimilation for 2–6 months (Eamus 1999), whereas evergreen species may take advantage of any brief favourable conditions. Duration and quality of the favourable period also affect carbon gain and therefore, the relative advantage of each leaf habit (Chabot and Hicks 1982; Kikuzawa 1989, 1991, 1995; Diemer 1998). For example, a cool or short growing season, or less fertile soils, reduces carbon gain and therefore, potentially limits the capacity of the plant to acquire sufficient resources to replace short-lived leaves (Chabot and Hicks 1982). When it becomes increasingly difficult to pay back leaf construction costs, i.e. when high photosynthetic rates cannot be achieved, long-lived, evergreen leaves are predicted to be more profitable (Schulze *et al.* 1977; Kikuzawa 1991).

Several main predictions can, therefore, be made about trends in net photosynthesis between deciduous and evergreen *Nothofagus* species. First, the shorter lifespan of leaves of deciduous species, together with high annual costs of leaf replacement, predicts that leaves of deciduous species will be less robust, i.e. invest less in leaf structure, with consequently higher mass-based rates of photosynthesis than evergreen species (Chabot and Hicks 1982). Consistent with this prediction, studies have shown that deciduous species generally have higher maximum photosynthetic rates per unit leaf mass than co-occurring evergreen species (Johnson and Tieszen 1976; Medina and Francisco 1994; Chapin and Shaver 1996; Eamus and Prichard 1998), attributed to decreased amounts of non-photosynthetic supportive tissue and higher leaf nitrogen concentrations (Johnson and Tieszen 1976; Chabot and Hicks 1982; Reich *et al.* 1991, 1992; Chapin and Shaver 1996). Second, evergreen leaves may be increasingly robust at higher altitudes due to an increasing demand for leaf protection against both abiotic and biotic damage. If so, then deciduous leaves are predicted to have an increasing advantage in photosynthetic rates per unit leaf mass with altitude, given that robust evergreen leaves may be thicker, with increased resistance to CO_2 diffusion and reduced light penetrating to the lower mesophyll (Field and Mooney 1986; Evans 1989). Third, the presence of leaves throughout the year in evergreen species suggests that they would have a greater capacity for acclimation to temperature and possibly a broader instantaneous temperature-dependence curve. Finally, since the deciduous species are more common in environments with cooler summers, the optimum temperature for photosynthesis is predicted to be lower in deciduous species.

The aim of this study was to test some of the above hypotheses, specifically whether deciduous *Nothofagus* species have (1) higher maximum rates of net photosynthesis (P_{max}) and specific leaf areas (SLA) than evergreen species; (2) an increasing photosynthetic advantage (e.g. higher P_{max}) relative to evergreen species with increasing altitude; (3) a lower optimum temperature for net photosynthesis (T_{opt}) than evergreen species; and (4) a narrower instantaneous temperature-dependence curve (T_{range}) than the evergreen species? Hypotheses 1 and 3 were tested across five deciduous and five evergreen *Nothofagus* species grown in a common environment. Hypotheses 2 and 4 were tested in sympatric populations of *N. cunninghamii* and *N. gunnii* in Tasmania.

Materials and methods

P_{max} and T_{opt} in deciduous and evergreen Nothofagus *species*

Phylogenetic analyses show that leaf habit has evolved several times in *Nothofagus*, with deciduous and evergreen species represented in each of the three temperate subgenera of *Nothofagus* (Hill and Jordan 1993; Martin and Dowd 1993; Manos 1997) (Fig. 1). Five deciduous and five evergreen species were chosen so that both leaf habits of each temperate subgenus were represented (Fig. 1; Table 1). The leaf lifespans used in correlations were the most common lifespan recorded in their natural environment (rather than the maximum values given in Table 1) and were slightly adjusted to take into account that leaves of some species are produced in late spring and drop in late summer–autumn. Estimated leaf lifespans ranged from 5 to 7 months in the deciduous species and from 12 to 39 months in the evergreen species.

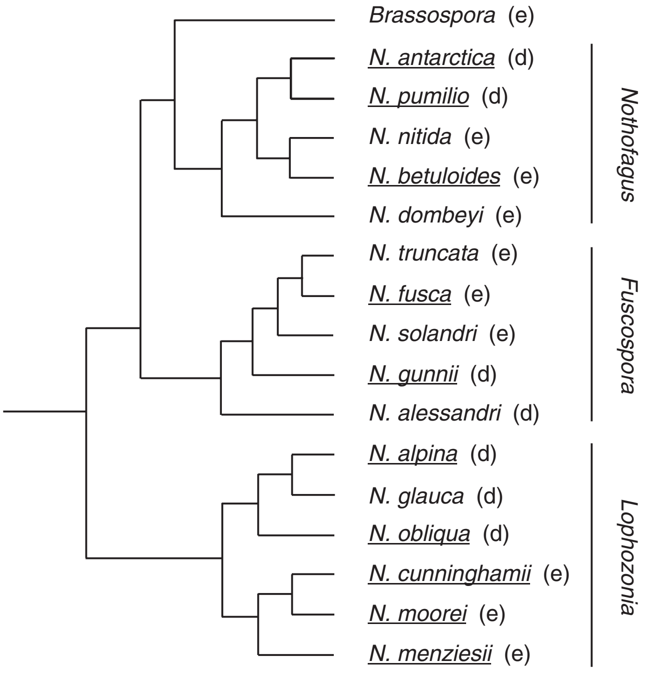

Fig. 1. An hypothesised phylogeny of *Nothofagus* based on combined data from rDNA ITS sequences, *rbc*L sequences and morphological characters (Manos 1997). Only the temperate subgenera are shown in detail. The species underlined are those included in this study. d, deciduous species; e, evergreen species.

In this common-garden experiment plants were grown outdoors in pots of sandy loam at Sassafras, Victoria (520 m asl, 37°52′S, 145°22′E) from July 1998 to February 1999, following growth for several years outdoors at Monash University (Melbourne). They ranged in height from 0.5 to 2.0 m and in age from 3 to 5 years old. They experienced full sun for most of the day but light shade in early morning and late afternoon. They were watered using automatic sprinklers daily (twice daily on days exceeding 28°C) and fertilised with a half-strength solution (0.88 g L^{-1}) of Thrive^{TM} fortnightly.

Gas-exchange was measured in February 1999 with an infrared gas analyser (IRGA), possessing an open gas exchange system incorporating a temperature-controlled leaf chamber (Analytical Development Co. LCA4, Hoddesdon, UK). One to several fully expanded sunlit leaves from the current growth season were selected from each of five plants of each species for gas analysis. The attached leaf was arranged in the leaf chamber (surface area of 6.25 cm^2) to allow maximum illumination by the external light source. The light source was a 12V dichroic ‘white light’ lamp with a 20-W globe, providing 800–1000 μmol quanta m^{-2} s^{-1} (above the light saturation point of each species). Air of *c.* 350 μl L^{-1} CO_2 was pumped through the leaf chamber and reference line at 200 μmol s^{-1}. Net CO_2 uptake was measured when stable and the rate of net photosynthesis was recorded at five to six temperatures (with leaf temperature measured by a thermistor), starting at 18°C and increasing by 2°C increments up to 28°C. The maximum rate of net photosynthesis (P_{max}) and the temperature at which it occurred (T_{opt}) were recorded. The vapour pressure deficit was generally maintained at 1.4 kPa for each temperature.

P_{max}, T_{opt} and T_{range} of Nothofagus cunninghamii *and* Nothofagus gunnii *in the field*

Instantaneous temperature-dependence curves of photosynthesis were measured on leaves of mature *Nothofagus gunnii* and *Nothofagus cunninghamii* in the field at five to six sites in February–March 1998. The study sites were within three montane areas in Tasmania, Cradle Mountain (41°38′S, 145°57′E, 920 m asl), Mt Field (42°41′S, 146°38′E, 1020 m asl) and Mt Read (41°51′S, 145°33′E, 550, 760, 1070 and 1120 m asl; Table 1). The species co-occurred at all sites except the highest altitude at Mt Read, where *N. cunninghamii* was absent. The vegetation of the study sites ranged from subalpine heath at high altitudes to closed forest at lower altitudes.

Plants were chosen that had sunlit foliage (in closed vegetation, plants were chosen at edges or in large canopy gaps). Net photosynthesis was measured as described above on attached, fully expanded (current season's growth), sunlit leaves, *c.* 1–1.5 m above ground level. Net CO_2 uptake was measured when stable at 8°C, then at 2°C increments to 30°C. The following equation was used to fit a curve to each set of CO_2 uptake readings:

$$P = (b(t - T_{min})(1 - \exp(c(t - T_{max}))))^2,$$

where P is the net rate of photosynthesis, t is temperature (°C), T_{min} and T_{max} are the temperatures at which net photosynthesis reaches zero and b and c are fitting parameters (Ratkowsky *et al.* 1983). This equation was used to derive (1) maximum rates of net photosynthesis on a mass ($P_{max,\,mass}$) and area ($P_{max,\,area}$) basis, (2) the temperature at which the maximum photosynthetic rate occurred (T_{opt}), (3) the temperature span over which 80% of P_{max} occurred (T_{range}) and (4) the percentage of P_{max}

occurring at 12°C (%P_{max12}) and at 30°C (%P_{max30}). The leaves used were pressed and air-dried in the field, then rehydrated for measurement of leaf area (Bioscan^{TM} image analysis software, Monash University). Trials indicated that rehydration of dried leaves gave 98 ± 0.8% of the area of fresh leaves. The leaves were then oven-dried at 75°C to constant weight and weighed.

Since study sites were located at varying latitudes across Tasmania, the temperature profile rather than altitude was used to investigate trends in photosynthetic variables, with mean annual temperature (MAT) used as the independent variable across a range of analyses. The temperature profile of each site was estimated by the climatic analysis and prediction software ANUCLIM ver. 5.0 (Houlder *et al.* 1999). The relationships between photosynthetic variables and soil nitrogen concentration were also examined. Five samples of surface soil (top 10 cm) were collected at haphazard locations at each site. Samples comprised 4 or 5 subsamples taken equidistantly around the perimeter of a 1-m-diameter circle. Soil nitrogen was measured with a Leco CHN-2000 analyser (LECO Corporation, St Joseph, MI, USA).

Data analysis

The difference in photosynthetic variables between deciduous and evergreen *Nothofagus* was examined using nested ANOVA, with leaf habit as a fixed factor and species as a random factor. Where there was a significant effect of species within leaf habit, one-way ANOVA was used to determine whether the difference was among deciduous or evergreen species. The assumptions of statistical tests were checked before analysis and transformations were used where necessary. Pearson and Spearman correlation was used to test associations among leaf variables.

Closely related species may share characteristics because of common ancestry, a feature not recognised by analyses that treat species as independent data points. Data were therefore also analysed with phylogenetically independent contrasts (PICs) computed by CAIC V. 2.6.8 (Purvis and Rambaut 1995) from the *Nothofagus* phylogeny of Manos (1997) that combines data from rDNA ITS sequences, *rbc*L sequences and morphological characters (Fig. 1). Differences between the two leaf habits for each variable were investigated by a one-sample *t*-test on the mean of contrasts calculated in the 'Brunch' option. Under the null hypothesis of no significant difference, the mean value of the contrasts should be zero (Purvis and Rambaut 1995). However, only three contrasts could be computed from the data and these tended to be highly variable (with skewed distributions) because of the intermediate position of *N. fusca* (Hook.f.) Oerst. between deciduous and evergreen species for many of the variables and we did not consider these statistical tests to be reliable. Hence, the only PICs-based analyses we present are regressions of leaf traits with leaf lifespan, using contrasts computed by the 'Crunch' option (nine contrasts). Regressions were only undertaken for the full species dataset; analyses of each leaf habit separately produced too few contrasts.

The field-based study of photosynthesis in *N. gunnii* and *N. cunninghamii* was analysed by two-way ANOVA to determine whether there were differences between species and among sites. This was followed by one-way ANOVA of photosynthetic variables among sites for each species separately, given the additional *N. gunnii* site where *N. cunninghamii* was absent. Pearson correlation, without adjustment for multiple comparisons, was used to examine the associations between P_{max} and each of altitude, ANUCLIM-derived climatic variables and soil nitrogen concentration for each species. A test of homogeneity of slopes was used to test whether the relationship between photosynthetic variables and MAT varied between species, i.e. to test for evidence of increasing advantage in P_{max} in *N. gunnii* on colder sites. A critical value of $\alpha = 0.05$ was used in all hypothesis testing. SYSTAT *v.* 10 was used for all statistical analyses.

Results

P_{max} *and* T_{opt} *in deciduous and evergreen* Nothofagus *species*

There was no significant difference in $P_{max, area}$ between leaves of deciduous and evergreen species, but $P_{max, mass}$ was significantly higher in deciduous species, consistent with their higher SLA (Fig. 2). There was no significant difference in $P_{max, mass}$ among evergreen species, but there was a significant difference among deciduous species (Fig. 2) and a significant difference in $P_{max, area}$ among both deciduous and evergreen species (Fig. 2). $P_{max, area}$ and $P_{max, mass}$ were not significantly correlated with SLA across all species (confirmed by PICs analysis) or among evergreen or deciduous species (Fig. 3). There was little difference in SLA among evergreen species other than *N. fusca*, with its high SLA (153 ± 21 $cm^2 g^{-1}$) associated with a short leaf lifespan (12 months). T_{opt} did not differ between the two leaf habits (Fig. 2), although there were significant differences among both deciduous (highest in *N. alpina*) and evergreen species (highest in *N. moorei*).

The deciduous and evergreen species differed significantly in leaf lifespan ($t = 6.65$, $P = 0.001$, data $\log_e$-transformed). Since there was considerable variation in leaf lifespan among evergreen species (Table 1), SLA and the photosynthetic variables were compared directly to leaf lifespan by using the full dataset and among evergreen species. There was a significant negative correlation of leaf lifespan with $P_{max, mass}$ and SLA across all species (confirmed by using PICs analysis), but not separately among evergreen species (Fig. 4). There were no significant correlations of leaf lifespan with $P_{max, area}$ or T_{opt} either across all species (confirmed by using PICs analysis), or separately among evergreen species (Fig. 4).

Field measurements of N. gunnii *and* N. cunninghamii

$P_{max, mass}$ and $P_{max, area}$ were significantly higher in *N. gunnii* than *N. cunninghamii* (Table 2; Figs 5 and 6). The higher $P_{max, area}$ of *N. gunnii* than *N. cunninghamii* contrasts with the results from the first experiment, where $P_{max, area}$ was 1.5 times higher in *N. cunninghamii* than *N. gunnii*. There was no significant difference in T_{opt}, T_{range} or %P_{max12} between *N. gunnii* and *N. cunninghamii*, but %P_{max30} was significantly higher in *N. gunnii* (Table 2; Fig. 6).

There was no significant difference among sites for any photosynthetic variable in *N. gunnii*, but there was a significant difference in T_{range} among *N. cunninghamii* populations (Fig. 6). The only correlations between photosynthetic variables and any environmental variable or SLA were a positive correlation of $P_{max, mass}$ with SLA in *N. gunnii* and a negative correlation of $P_{max, area}$ with soil N in *N. cunninghamii* (P_{max} correlations presented in Table 3). SLA was not significantly correlated with any environmental variable (Table 3). Since there was no significant variation

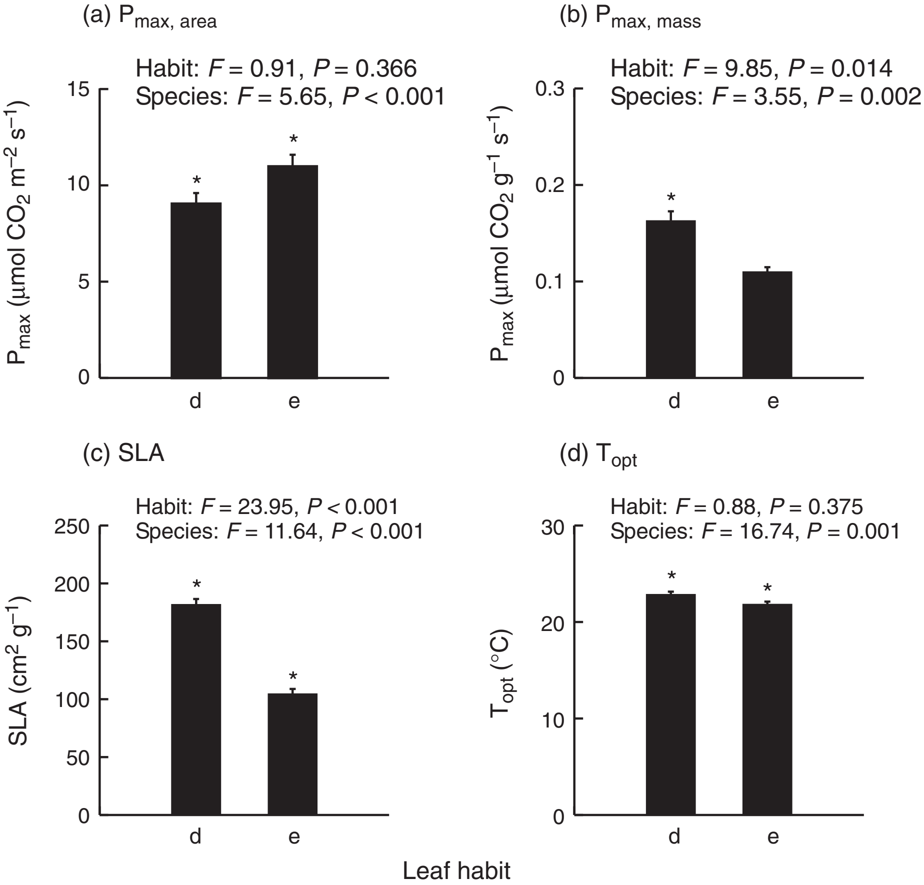

Fig. 2. Comparisons of photosynthetic variables and SLA between deciduous and evergreen species. The data presented are means of each leaf habit (with species as replicates) with standard errors. The results of nested ANOVA are given. Asterisks indicate significant differences among species with that leaf habit. The multiple populations of *Nothofagus cunninghamii* and *N. gunnii* are combined for this analysis.

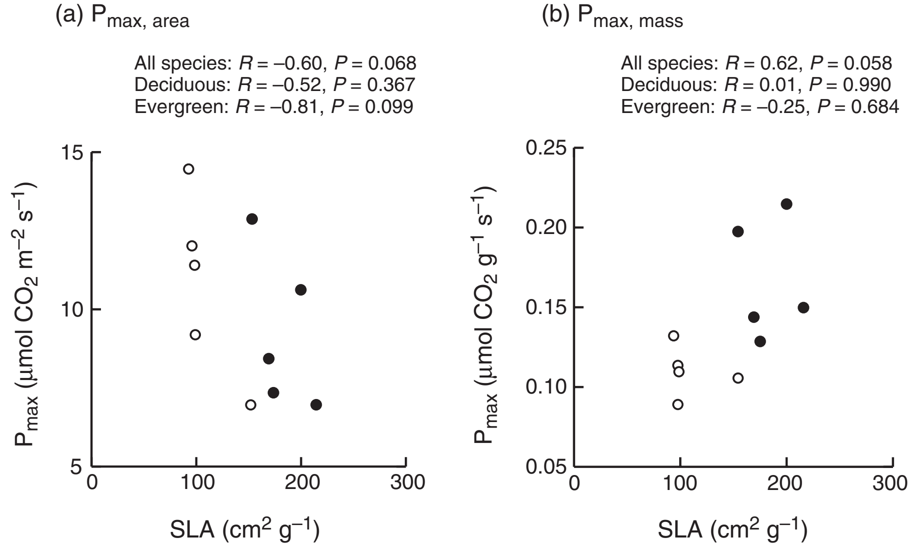

Fig. 3. The relationship of $P_{max, area}$ and $P_{max, mass}$ with SLA among species. The values given are means of at least five replicate plants. The results of Pearson correlation are given for untransformed data. Solid circles, deciduous species; open circles, evergreen species.

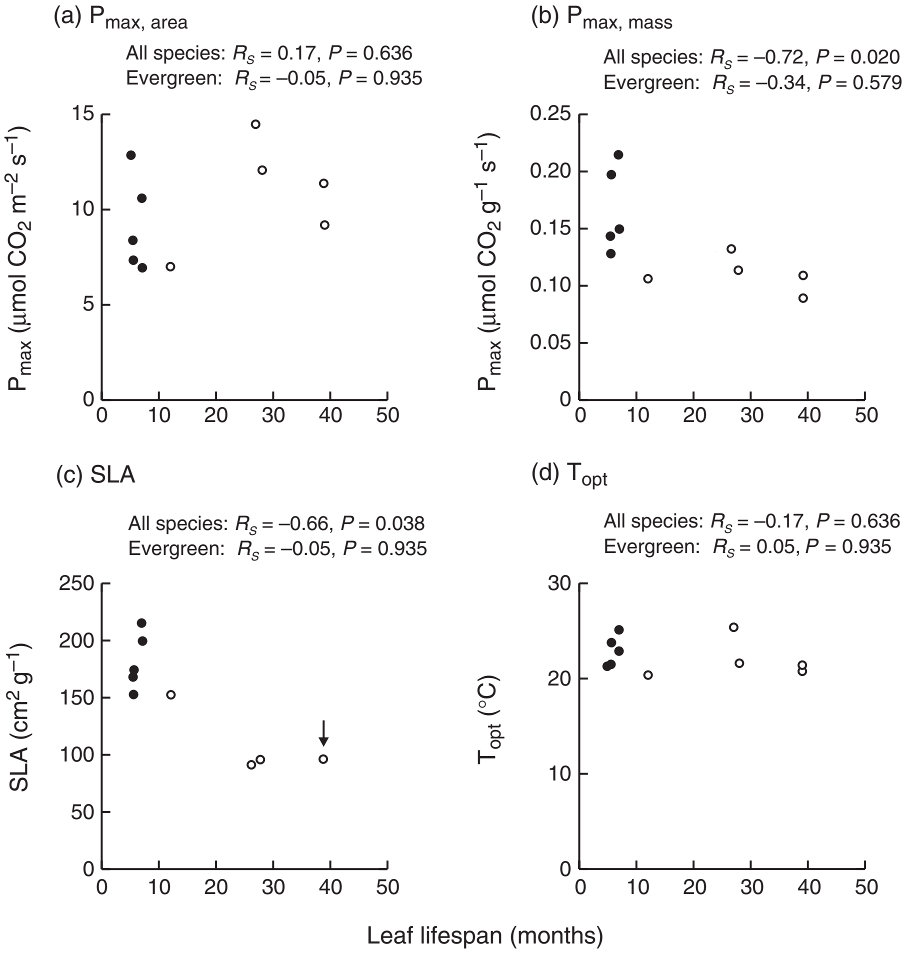

Fig. 4. The relationship between leaf lifespan and each of (*a*) $P_{max, area}$, (*b*) $P_{max, mass}$, (*c*) SLA and (*d*) T_{opt} among species. The values given are means of at least five replicate plants. Spearman rank correlations (R_S) are given for the relationships across all species and among evergreen species. They are not given for deciduous species because of insufficient variation in lifespan. The arrow in (*c*) indicates two coinciding datapoints. Solid circles, deciduous species; open circles, evergreen species.

in P_{max} among sites in *N. gunnii* and no correlation between P_{max} and MAT in either species, we do not report the tests of homogeneity of slopes.

Discussion

Deciduous *Nothofagus* species showed significantly higher rates of $P_{max, mass}$ than did evergreen species, consistent with previous comparisons of species with contrasting leaf habits in regions experiencing severe cold or dry seasons (Johnson and Tieszen 1976; DeLucia and Schlesinger 1995; Eamus and Prichard 1998). The same trend in $P_{max, mass}$ with respect to leaf lifespan was recorded by PICs analysis, indicating correlated evolution among these variables rather than just shared characteristics through shared ancestry. Field-based measurements also demonstrated significantly higher $P_{max, mass}$ in *N. gunnii*, but also a higher $P_{max, area}$ than *N. cunninghamii* over a range of altitudes. The high photosynthetic rate per unit leaf mass allows carbon assimilation to be maximised during the growing season to compensate for any carbon costs of being leafless for a season and the cost of annual replacement of the canopy (Orians and Solbrig 1977; Kikuzawa 1991). Since the extension of leaf lifespan beyond the summer growth period is limited in deciduous species, a positive carbon balance may be primarily achieved by high photosynthetic rates (Kudo *et al.* 2001) per unit mass. In comparison, evergreen species are potentially able to assimilate carbon throughout the year, depending on climatic conditions, and across multiple years (Schulze *et al.* 1977; Chabot and Hicks 1982).

The contribution of the high SLA to the high $P_{max, mass}$ in deciduous species is uncertain. The deciduous species may have higher mesophyll-level photosynthetic rates than evergreen species, independent of tissue arrangements. However, $P_{max, mass}$ is also generally negatively correlated with leaf longevity (Reich *et al.* 1991, 1992). Longer-lived evergreen leaves generally have lower SLA, with

Table 2. Results of two-way ANOVA of photosynthetic variables in *Nothofagus gunnii* and *N. cunninghamii* at sites where both species occur

L, data $\log_e$-transformed

Variable	Factor	*F*-ratio	*P*
$P_{\text{max, mass}}$ (L)	Species	170.69	<0.001
	Site	1.70	0.183
	Species × site	2.11	0.110
$P_{\text{max, area}}$	Species	49.57	<0.001
	Site	2.02	0.123
	Species × site	1.65	0.195
T_{opt}	Species	0.39	0.537
	Site	1.04	0.408
	Species × site	0.41	0.803
T_{range}	Species	0.94	0.342
	Site	0.98	0.436
	Species × site	1.07	0.395
$\%P_{\text{max12}}$	Species	0.01	0.907
	Site	0.58	0.682
	Species × site	0.81	0.529
$\%P_{\text{max30}}$	Species	10.46	0.004
	Site	1.45	0.248
	Species × site	1.13	0.364

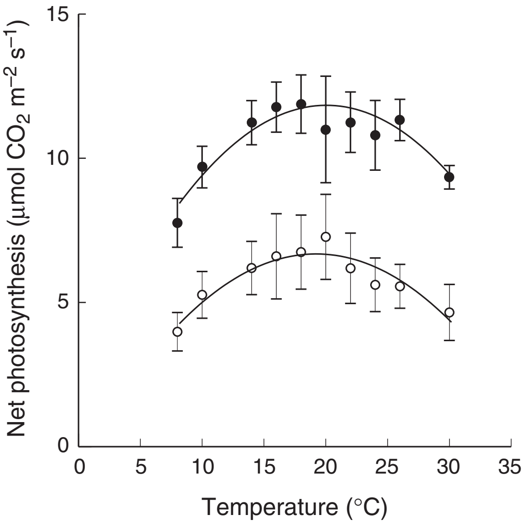

Fig. 5. Instantaneous temperature-dependence curves of net photosynthetic rate for *Nothofagus gunnii* and *N. cunninghamii*. The points are the means with standard errors of all sampled populations from sites where the species co-occur. Solid circles, *N. gunnii*; open circles, *N. cunninghamii*.

higher investment in non-photosynthetic tissue for structural and chemical defence, which must reduce mass-based photosynthetic rates (Field and Mooney 1983; Reich *et al.* 1991; Gower *et al.* 1993; Reich 1993; Aerts 1995). In addition to dilution by non-photosynthetic tissue, low-SLA leaves are often thicker, with potentially increased resistance to CO_2 diffusion and reduced light penetration to the lower mesophyll (Field and Mooney 1986; Evans 1989). However, although a significant difference in $P_{\text{max, mass}}$ was recorded between deciduous and evergreen species in this study, no trend occurred among evergreen species with respect to leaf lifespan, despite *c.* 3-fold variation in the latter. Nor was there any correlation of P_{max} (either mass-based or area-based) with SLA. The absence of a simple relationship between $P_{\text{max, mass}}$ and leaf lifespan or SLA among these species may be due to there being various ways (anatomical and morphological) that plants can maximise carbon gain. Confounding factors, such as shade tolerance, may also affect the relationship of P_{max} with SLA and leaf lifespan across species. For example, in our study, *N. alpina* (Poepp. & Endl) Oerst. showed a low P_{max} (mass-based and area-based) compared with other deciduous species, probably related to its higher shade tolerance (Read and Hill 1985).

Photosynthetic performance became more equivalent between leaf habits when expressed on an area basis. Although the thicker evergreen leaves may increase resistance to CO_2 diffusion and reduce light penetrating to the lower mesophyll (Field and Mooney 1986; Evans 1989), there may be more assimilatory tissue per unit leaf area (Garnier *et al.* 1999). The advantage of the robust leaves to the evergreen species may lie in leaf protection from abiotic and biotic factors (Chabot and Hicks 1982). However, the higher rates of net photosynthesis per unit leaf area that may result from having denser or thicker leaves also potentially reduces support (branching) costs (King 1999), since less leaves are needed to produce the same carbon gain.

Nothofagus gunnii showed a significantly higher $P_{\text{max, area}}$ than *N. cunninghamii* in field-grown plants, in contrast to the trend in the common-garden experiment. The reversal of the trend was due to a greater reduction in SLA in *N. gunnii* than *N. cunninghamii* in field-grown plants and an increase in $P_{\text{max, mass}}$ in contrast to the decrease shown by *N. cunninghamii*. The cause of these opposing trends is unclear, but could relate to differences in climate, plant nutrition, plant genotype and plant age between the two experiments.

There was no significant difference in T_{opt} or T_{range} between *N. gunnii* and *N. cunninghamii*, but there was a significantly higher percentage of P_{max} reached at 30°C ($\%P_{\text{max30}}$) in *N. gunnii* than *N. cunninghamii* in the field-based study. Read and Busby (1990) recorded a similar or lower T_{opt} (from instantaneous temperature-dependence curves) in *N. gunnii* than in *N. cunninghamii*, in seedlings acclimated to various temperatures in growth cabinets and a lower T_{range}. The low T_{range} in that study may be related to the constant temperatures under which seedlings were grown, in contrast to those experienced in the field environment in this study. However, in the same study, a higher optimum acclimation temperature was recorded in *N. gunnii* (23°C) than *N. cunninghamii* (17°C) seedlings from the same collection site (Mt Field, 980 m asl) and the optimal (80% of P_{max}) acclimatory temperature range

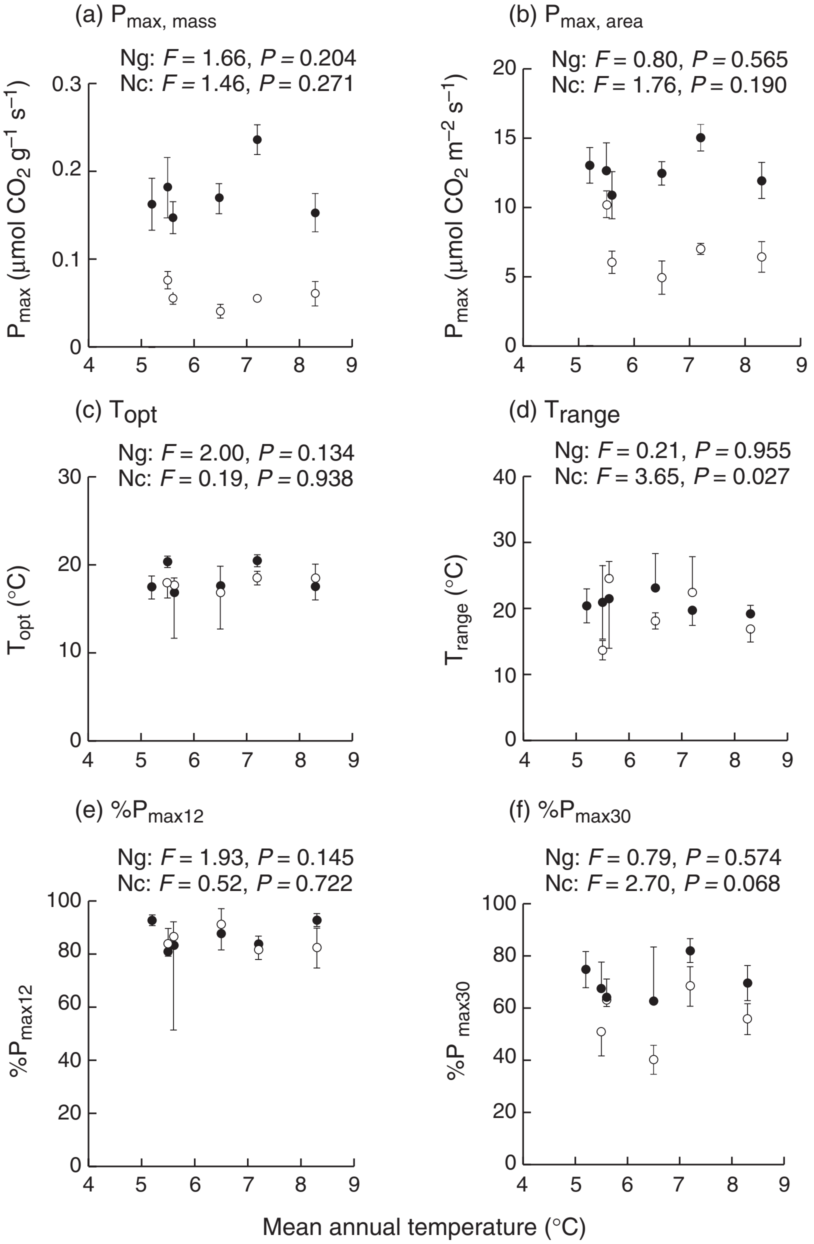

Fig. 6. Comparison of photosynthetic variables in *Nothofagus gunnii* (Ng) and *N. cunninghamii* (Nc). Data presented are means with standard errors of each sampled population, derived from the instantaneous temperature-dependence curves measured at each site and plotted against mean annual temperature of the study site. The results of one-way ANOVA are given. Solid circles, *N. gunnii*; open circles, *N. cunninghamii*.

was 14–29°C in *N. gunnii* compared with 9–26°C in *N. cunninghamii* (Read 1990; Read and Busby 1990). The greater difference between these species in acclimatory than instantaneous responses is consistent, with leaves of co-occurring *N. cunninghamii* and *N. gunnii* experiencing similar diurnal ranges of temperature, but a very different seasonal range of temperatures, given that *N. gunnii* is leafless in winter. The adaptation of photosynthetic acclimation in *N. gunnii* to warmer temperatures is consistent with the deciduous habit, i.e. with carbon gain and growth optimised towards temperatures experienced during the warmer summer months. However, *N. gunnii* appears to be sensitive to high temperatures (Read and Busby 1990), which may limit its distribution at lower altitudes. The slightly higher acclimatory capacity recorded in *N. cunninghamii* (80% of maximum across a 17°C span of acclimation temperatures) than *N. gunnii* (15°C) (Read and Busby 1990) is likely to be important in maximising carbon gain, given the greater seasonal variability in temperature experienced by leaves of evergreen species (Berry and

Table 3. The relationship of $P_{max,\ mass}$, $P_{max,\ area}$ and SLA of *Nothofagus gunnii* and *N. cunninghamii* with altitude, ANUCLIM-derived temperature variables and soil N concentration of the study sites and SLA
The data given are the Pearson correlation coefficient, R_P, with asterisks indicating significance ($P < 0.05$).
n, number of sites; L, data $\log_e$-transformed

Environmental variable	*N. gunnii* ($n = 6$)			*N. cunninghamii* ($n = 5$)		
	$P_{max,\ mass}$ (L)	$P_{max,\ area}$	SLA	$P_{max,\ mass}$	$P_{max,\ area}$ (L)	SLA
Altitude	−0.38	0.49	−0.17	−0.16	0.12	−0.41
Mean annual temperature	0.41	0.52	0.19	0.14	−0.11	0.34
Max. temp. warmest month	0.35	0.43	0.19	<0.01	−0.26	0.36
Min. temp. coolest month	0.57	0.76	0.16	0.33	0.20	0.08
Soil N (L)	−0.43	−0.61	0.01	−0.82	−0.90*	−0.02
SLA	0.82*	0.47		0.21	−0.27	

Björkman 1980). A broad instantaneous photosynthetic temperature optimum (T_{range}) might also increase carbon assimilation (Battaglia *et al.* 1996), but no difference was recorded between the species.

The lack of clear relationships between the field photosynthetic responses to temperature and site climate may be due to the complexity of photosynthetic processes. Many factors, both intracellular and extracellular, influence photosynthetic responses and their influence can be difficult to separate (Pearcy *et al.* 1987; Ferrar *et al.* 1989). In addition, while temperature varied with altitude, other environmental factors that influence photosynthesis, such as soil and atmospheric moisture, light and soil nutrients, may be varying in a complex manner, thereby obscuring influences of temperature. For example, soil nitrogen concentration was 0.14–0.18% at the Mt Read sites, but 0.48–0.69% at the other sites. This effect is evident in the negative correlation of $P_{max,\ area}$ with soil nitrogen concentration in *N. cunninghamii*. In addition, these factors may also influence SLA, which did not decline in *N. cunninghamii* with increasing altitude (decreasing MAT) as predicted, but was consistently lower at the less fertile Mt Read sites.

This study shows that $P_{max,\ mass}$ is higher (1) for deciduous *Nothofagus* species than for evergreen species and (2) for *N. gunnii* than for *N. cunninghamii* where they co-occur across a range of altitudes. The higher mass-based photosynthetic rates of *N. gunnii* (and the other deciduous *Nothofagus* species) may substantially increase their competitiveness in these cold climates, although these gains will be partly offset by the costs of annual leaf replacement. Comparisons of *N. gunnii* and *N. cunninghamii* across a range of altitudes showed no evidence of increasing assimilatory superiority with decreasing MAT in *N. gunnii* to explain the absence of the evergreen *N. cunninghamii* at the highest altitudes, or vice versa for the absence of *N. gunnii* at lower altitudes. Instead, the differential distribution of deciduous *v.* evergreen *Nothofagus* species may be influenced by factors related to rates of carbon gain at the level of whole plant, rather than at the level of leaf tissue (Poorter and Remkes 1990; Körner 1991; Givnish 2002). For example, *N. cunninghamii* may have a greater decline in leaf area ratio with increasing altitude relative to *N. gunnii*. In addition, the evergreen habit of *N. cunninghamii* renders it prone to leaf damage by frost on the coldest sites, thereby incurring large replacement costs. We have seen substantial leaf loss, probably due to winter frost damage, in *N. cunninghamii* at its upper altitudinal limit on Mt Read. Co-occurring *N. gunnii* was not visibly affected, presumably because the frost event(s) occurred while *N. gunnii* was leafless. Hence, the advantages of deciduousness in *Nothofagus* in environments that experience an unfavourable season are likely to be more complex than simply rates of leaf-based carbon gain. Some of these factors will be explored in later papers that compare allocation patterns and nutrient-use efficiency between deciduous and evergreen *Nothofagus* species.

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