

Light gains and physiological capacity of understorey woody plants during phenological avoidance of canopy shade

C. K. AUGSPURGER,[†] J. M. CHEESEMAN and C. F. SALK

Department of Plant Biology, University of Illinois, Urbana, IL 61801, USA

Summary

1. Carbon gain during phenological avoidance of canopy shade by an understorey plant depends on the extent of avoidance, the leaf stage during avoidance, and whether young and old leaves can exploit greater light availability in spring and autumn.

2. For *Asimina triloba* (L.) Dunal., *Aesculus glabra* Willd., *Acer saccharum* Marsh., *Lindera benzoin* (L.) Blume and *Carpinus caroliniana* Walt. in a deciduous forest in Illinois, USA, spring avoidance with leaves at full size ranged from 0 days for *Asimina* to 24 days for *Aesculus*, and brought 36–98% of estimated total annual irradiance. Autumn avoidance was non-existent to minimal in all species.

3. Total chlorophyll reached maximum concentrations at the middle of leaf life span, and declined well before senescence. Leaf nitrogen concentrations and net photosynthetic capacity both peaked in youngest leaves during spring avoidance, and were low in old leaves during autumn avoidance.

4. *Aesculus* had especially high photosynthetic capacity during precanopy closure, while *Asimina* had relatively low capacity in its later developing leaves.

5. Young leaves of species with phenological avoidance can enhance C gain, while old leaves in autumn do not. Thus phenological avoidance in spring may enhance the persistence of understorey woody individuals of some species.

Key-words: canopy closure, chlorophyll, leaf phenology, nitrogen, photosynthetic capacity

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Introduction

Understorey woody plants in deciduous forests receive very little light when leaf cover by canopy trees is complete. Their morphological and physiological adaptations to shade have been well documented. Phenological shade avoidance, where understorey plants have earlier leaf expansion in spring and/or later leaf senescence in autumn than canopy trees, is another means of potentially enhancing carbon gain (Uemura 1994). These periods of high light exposure may result in a significant amount of C gain and enable understorey woody plants to persist in otherwise shaded understoreys (dePamphilis & Neufeld 1989; Gill, Amthor & Bormann 1998; Seiwa 1999a). Quantifying enhanced light gain and physiological capacity during avoidance by understorey woody plants will contribute to understanding C gain and, consequently, growth and survival, as well as species coexistence and the seasonality and magnitude of ecosystem C flux.

Mathematical models predict that alteration of phenology to match light availability brings maximum C gain (Iwasa & Cohen 1989; Kikuzawa 1991). Empirical studies indicate that phenological avoidance in temperate deciduous forests occurs in both seedlings (Seiwa 1999b) and saplings (Gill *et al.* 1998; Seiwa 1999a) of canopy tree species, understorey trees (dePamphilis & Neufeld 1989) and shrubs (Gill *et al.* 1998). In a study in a temperate deciduous forest, spring phenological avoidance by subcanopy individuals occurred in 11 of 13 canopy tree species (Augsburger & Bartlett 2003). In contrast, autumn avoidance was found in only two of 13 species.

The potential for phenological avoidance to enhance C gain depends first on the amount of irradiance received during that period. Irradiance in the understorey is determined by daily changes in solar elevation (affecting attenuation pathways through vegetation) and day-length, as well as cloud cover and canopy cover. Light transmission in spring and autumn is not equivalent (Hutchison & Matt 1977; Gill *et al.* 1998). Light transmitted to shrubs of an oak–hickory forest was 60% of above-canopy incident

radiation prior to bud break of canopy trees, but only 35% after leaf drop of canopy trees (Baldocchi *et al.* 1984). This seasonal asymmetry in light transmission arises because the period of closed canopy begins shortly before the summer solstice, but ends long after it. Therefore, based on the light environment alone, phenological avoidance in spring would be expected to bring more C gain than avoidance in autumn.

Second, phenological avoidance increases C gain only if leaves can utilize the extra light in spring and autumn. Spring physiological capacity requires building the photosynthetic apparatus, including chlorophyll and nitrogen-based enzymes, and the time at which young leaves are fully functionally developed varies among species (Reich, Walters & Ellsworth 1991; Koniger, Harris & Kibler 2000; Morecroft, Stokes & Morison 2003). Autumn physiological capacity depends on the timing of degradation of chlorophyll, N and the photosynthetic apparatus. Generally, leaves of woody species exhibit a loss of photosynthetic capacity as they age (Koniger *et al.* 2000; Wilson *et al.* 2001; Morecroft *et al.* 2003; but see Nelson & Isebrands 1983).

Finally, any potential for C gain via phenological avoidance may be limited by freezing temperatures (Cannell & Smith 1986; Hänninen 1991) that cause leaf damage and xylem cavitation (Wang, Ives & Lechowicz 1992). Young, expanding leaves are especially vulnerable to damage (C.A., personal observation). Therefore the extent of spring avoidance may be constrained.

In this study we estimated the progressions of canopy closure in spring and canopy opening in autumn, based on leaf phenology of 13 canopy tree species in a temperate deciduous forest. Concurrently, understorey individuals of five woody species were examined to document: (1) the extent of phenological avoidance of canopy shade in spring and autumn; (2) the amount and percentage of total annual irradiance received during spring and autumn avoidance *vs* summer shade; and (3) patterns of leaf chlorophyll, N, specific leaf area (SLA) and photosynthetic capacity during avoidance *vs* no avoidance of shade. We also obtained long-term dates of the frost-free period to evaluate whether phenological avoidance is constrained by freezing temperatures. Finally, for two species, similarly sized individuals growing at the forest edge were included. The comparison between edge *vs* understorey individuals allowed a determination of whether the temporal patterns reflected ontogeny, or were specific responses by understorey individuals to the environment created by the phenology of canopy trees.

In spring, we predicted that species with greater phenological avoidance would expand leaves and develop physiological capacity sooner and more rapidly than species with less avoidance. In autumn, we expected senescence would be delayed and physiological capacity retained or enhanced only if phenological avoidance occurs. Finally, we predicted that phenological

avoidance and associated adjustments in physiological capacity would be more evident in spring than in autumn.

Materials and methods

STUDY SITE AND SPECIES

The study site was Trelease Woods, a 24 ha deciduous forest fragment located 5 km north-east of Urbana, IL, USA (40°09' N, 88°10' W). Among 20 canopy species the dominants are *Fraxinus* spp. (Tourn.) L., *Acer saccharum* Marsh., *Celtis occidentalis* L. and *Quercus rubra* L. The study species were juveniles of two canopy tree species, *A. saccharum* (Sugar Maple) and *Aesculus glabra* Willd. (Ohio Buckeye), and adults of three understorey shrubs/treelets, *Lindera benzoin* (L.) Blume (Spicebush), *Asimina triloba* (L.) Dunal. (Pawpaw) and *Carpinus caroliniana* Walt. (Blue Beech, Musclewood). Ten understorey individuals (2–4 m) of each species were chosen haphazardly, avoiding light gaps and edges. In addition 10 individuals of *Acer* and *Aesculus* were selected from the western edge of the fragment.

PHENOLOGY OF UNDERSTOREY AND EDGE PLANTS

Phenological stages were determined weekly and on each physiological sampling day in 1996–98. Three stages were defined: (1) leaf emergence (shoot extending beyond bud scales, leaves visible but folded); (2) leaf expansion (leaf expanding and flattening); and (3) senescence (breakdown of chlorophyll revealing underlying pigments). Within each stage, three substages were recognized: when 1/3, 1/3–2/3, or >2/3 of the stage was completed.

Spring phenological observations were converted to an estimated daily percentage of maximum leaf area, as follows. Leaves were collected at each substage of leaf emergence and leaf expansion. Mean leaf area on each of these six collection dates was quantified using a leaf-area meter (Li-Cor Model 3100, LiCor Inc., Lincoln, NE, USA) on 100 leaves. Leaf area for each substage was expressed as a percentage of maximum leaf area. Interpolation between the measured points was used to estimate daily percentage of maximum leaf area. Leaf phenology in autumn was described by the dates of the three substages of senescence. Interpolation between the measured points was used to estimate daily percentage of total senescence.

CANOPY PHENOLOGY

Concurrently, a phenological census was conducted weekly in 1996–98 on 12–15 canopy individuals of 13 canopy tree species (see Augspurger & Bartlett 2003 for species and methods). Phenological dates relevant

to the determination of canopy closure and opening were: (1) leaf emergence; (2) leaf expansion; and (3) leaf drop. Within each stage three substages were recognized: when 1/3, 1/3–2/3, or >2/3 of the stage was completed.

These observations were used to quantify canopy closure in spring and canopy opening in autumn. First, a springtime canopy closure function was generated for each species. Canopy development was assumed to increase linearly from 0% at the beginning of leaf emergence to 25% half-way between the beginning of leaf emergence and the beginning of leaf expansion, and then to increase linearly to 100% at completion of leaf expansion. Second, the canopy was considered to be 100% closed between the completion of leaf expansion and the beginning of leaf drop. Third, an autumn canopy-opening function was generated for each species. The canopy was assumed to decrease linearly from 100% at the beginning of leaf drop to 0% at the completion of leaf drop.

The contribution of a given species to closure or opening of the entire forest canopy was considered proportional to its relative abundance. Each species' relative abundance was calculated from the basal area of all trees (at 1.4 m) measured during a complete census of the study site in 1996 (J. Edgington, University of Illinois, unpublished data). Following Monk, Child & Nicholson (1970) and Martin *et al.* (1998), it was assumed for all species that the crown area for an individual was proportional to its basal area.

To calculate canopy closure (or opening) for the forest as a whole, percentage canopy closure (opening) for each species was multiplied by the relative abundance of that species. The weighted functions were then summed on each day to create a forest-wide canopy function that summarized the state of the entire forest's canopy from the first date of leaf emergence to the last date of leaf drop. Estimations of canopy closure and canopy opening were made separately for 1996, 1997 and 1998 because measurements of different physiological variables were made in different years.

LIGHT MEASUREMENTS

Total annual irradiance (photosynthetic photon flux density; 400–700 nm) received by plants in full sunlight at this site was quantified. Solar radiation data were collected in the open from 1995 to 2002 by the National Oceanic & Atmospheric Administration's surface radiation programme (www.srrb.noaa.gov/pub/data/surfrad) at Bondville, IL, USA, ≈ 24 km south-west of the study site. One-second samples of irradiance were summed to calculate total daily irradiance for each day of each year. Mean daily irradiance was calculated for the 8 years and a polynomial regression was fitted to estimate mean daily irradiance in the open (Fig. 1a). This was assumed to be the amount of radiation received by

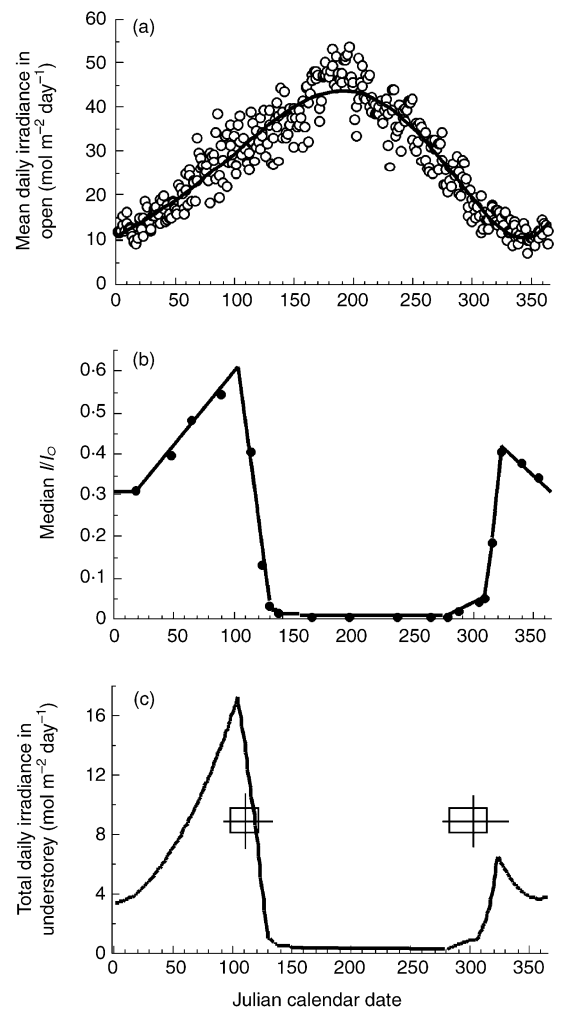


Fig. 1. (a) Total daily irradiance in an open area without canopy cover for each Julian calendar date. Values represent mean of daily values ($\text{mol m}^{-2} \text{ day}^{-1}$) for 1995–2002. Solid line, fitted fifth-degree polynomial regression. (b) Proportion of incident irradiance transmitted to understorey individuals (I/I_0) on 21 sampling dates. Solid line, estimated daily I/I_0 (see Materials and methods). (c) Estimated total daily irradiance received by understorey individuals. Values ($\text{mol m}^{-2} \text{ day}^{-1}$) equal the product of values in (a) and (b). Box-and-whisker plots summarize last spring date of 0 °C and first autumn date of 0 °C from 1971 to 2000. Vertical bar, median date; horizontal line, range; box encompasses dates between which 10 and 90% of first and last dates of 0 °C occurred.

canopy trees and edge individuals of *Acer* and *Aesculus*. No correction was made for the individuals being at the forest's western edge.

We estimated the proportion of irradiance transmitted daily to the understorey (I/I_0 , where I = irradiance reaching understorey individuals and I_0 = irradiance in the open). Instantaneous readings of irradiance were made with a quantum sensor (Li-Cor) on clear sunny days near midday during January–December 2002. The 21 sampling dates were at 3–4-week intervals, but were weekly during rapid canopy closure and opening. Readings were taken in an open field 25 m away from the forest edge at the beginning and end of

each sampling period of ≈ 45 min; the mean of the two measurements was used as I_0 . Irradiance of each understorey plant (I) was measured by holding the sensor horizontally at a fixed point on top of the foliage at ≈ 1.4 m. Readings were taken on leaves not experiencing sunflecks or being self-shaded. The median value of I for all 50 plants was used to calculate I/I_0 for each sample date because the data were non-normal. Different segments of the data in different phenological periods were fitted by regressions to provide a continuous estimate of daily I/I_0 (Fig. 1b). To calculate the estimated daily irradiance received by understorey individuals throughout the year (Fig. 1c), the fitted regression for daily solar radiation (Fig. 1a) was multiplied by the estimated daily I/I_0 (Fig. 1b).

Estimated total annual irradiance reaching an understorey plant was calculated for each study species. From initial leaf emergence until initial leaf senescence, daily irradiance in the understorey in Fig. 1(c) was weighted by the mean daily phenological stage (percentage of maximum leaf area). Daily irradiance after leaf senescence began was not included because photosynthetic capacity was essentially zero then. The same steps were taken for edge individuals, but using total irradiance without canopy cover ($I/I_0 = 1$). Finally, total annual irradiance of each species was subdivided into the percentage prior to canopy closure, during closed canopy, and after the beginning of leaf drop by canopy trees.

FROST-FREE DATES

Long-term dates of the last 0°C temperatures in spring and first 0°C temperatures in autumn were obtained from the Illinois State Water Survey (www.sws.uiuc.edu/atmos/statecli). The data were collected at Urbana, ≈ 5 km south-west of the study site from 1971 to 2000. The temperature was measured at a height of 1.5 m in the open at the edge of an urban heat source.

PHYSIOLOGICAL AND MORPHOLOGICAL MEASUREMENTS

Chlorophyll (1996), N and SLA 1997, and photosynthesis (1998) were measured periodically in each understorey and edge individual. Leaves represented the average phenological stage of the plant on a given sampling date. For all species except *Aesculus* leaf development was not synchronous, thus proximal and distal leaves developed in different light environments. A minimum of four leaves intermediate in location on a branch was used, and different leaves were sampled on each date. Any changes between years in the plant and its environment would have been minimal because individuals were not near gaps with rapidly changing light conditions. Also their growth was very slow, thus keeping them at the same ontogenetic stage between years (C.A., unpublished data).

Chlorophyll *alb* ratio and total chlorophyll ($\mu\text{g cm}^{-2}$) were measured for leaf discs taken weekly from substage 2 of leaf emergence through substage 3 of senescence. Samples were immediately placed in the dark in CaCO_3 -saturated dimethylsulfoxide (DMSO) for chlorophyll extraction. After incubation in a 65°C water bath for 24 h, absorbance was read at 648 and 665 nm in an LKB Ultraspec II spectrophotometer, and chlorophyll content was calculated from the equations in Barnes *et al.* (1992).

Specific leaf area and N were measured on the same leaf samples taken biweekly from substage 2 of leaf emergence through substage 3 of senescence. For SLA ($\text{cm}^2 \text{g}^{-1}$), the area of each fresh leaf was measured with a Li-Cor leaf-area meter, then leaves were dried to constant weight at 65°C . Kjeldahl N was determined on these dried samples. Leaf N content per unit mass (N_{mass} , g g^{-1}) and per unit area (N_{area} , $\mu\text{g cm}^{-2}$) were calculated.

Photosynthetic capacity ($\mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$) was measured biweekly with an infrared gas analyser (Li-Cor 6400) at $2000 \mu\text{mol m}^{-1} \text{CO}_2$ and a photon flux density of $2000 \mu\text{mol m}^{-2} \text{s}^{-1}$ with a red LED light source (Li 6400-02). Measurements were taken between 11:00 and 13:00 on days with a relatively steady temperature, from substage 1 of leaf expansion through substage 3 of senescence. We used $2000 \mu\text{mol m}^{-1} \text{CO}_2$ to overcome stomatal limitations and to come as close to CO_2 saturation as possible. Preliminary studies indicated that raising CO_2 did not result in stomatal closure and that net CO_2 exchange saturated at about $1000 \mu\text{mol m}^{-2} \text{s}^{-1}$, and was not reduced at higher irradiances.

Our objective was not to use gas exchange to estimate total C gain throughout the season, but to compare photosynthetic potential with other, more integrative phenological observations. This protocol reflects a number of compromises, for example the need for a single measurement that could be made on 70 individuals on a biweekly basis, and the lack of control of leaf temperature. It does avoid complexities that would impair data interpretation, for example comparisons based only on net CO_2 exchange under ambient conditions would have been strongly influenced by irradiance and conductance in ways that could not be separated without in-depth, biweekly light- and/or CO_2 -response analyses. Measuring earlier in the morning would have meant comparisons of data collected through the most rapid period of changing air temperature; measuring later in the day would have led to complications from high temperatures and midday stomatal closure. Therefore we decided to make all measurements at midday, at light and CO_2 saturation, using this as a measure of potential photosynthesis, rather than using less interpretable measurements of actual photosynthesis. Given the sensitivity of instantaneous net photosynthesis to light, temperature, stomatal conditions, time of day and season, we did not attempt to model net C gain from the photosynthesis measurements.

Results

PHENOLOGY

Canopy closing in spring lasted about 8 weeks, with most rapid change (10–90% closed) lasting only about 2 weeks (mean Julian calendar dates 124–142) (solid lines in Figs 2–4). Canopy opening in autumn required about 11 weeks (solid lines in Figs 2–4), with rapid opening (10–90%) occurring over 4 weeks (mean dates 289–317). The period of 100% canopy closure was not centred on the summer solstice (date 172) (Figs 2–4), but was >5 weeks later (mean date 209). This asymmetry resulted in the potential for much higher irradiance due to phenological avoidance in spring than in autumn.

The five species differed sharply in both number of days of phenological avoidance, and phenological stage during avoidance. Three categories were apparent in spring (compare long dashed lines with solid lines in Fig. 2; Table 1). At one extreme was *Aesculus*, with early and rapid leaf expansion. It achieved full leaf expansion prior to 50% canopy closure and had 24 days at full leaf size prior to complete canopy closure. Three species, *Acer*, *Carpinus* and *Lindera*, were intermediate. They had 3–4 weeks of spring avoidance with expanding leaves and 1 week with fully expanded leaves prior to complete canopy closure. At the other extreme was *Asimina*, with late and slow leaf expansion. While leaf expansion began well before canopy

closure, the leaves were small then; full leaf expansion was not reached until after 100% canopy closure.

All species except *Aesculus* had minor phenological avoidance in autumn (compare long dashed line and solid line in Fig. 2; Table 1). They began senescence about 1 week prior to canopy opening and were well advanced in senescence prior to 50% canopy opening.

IRRADIANCE

Incident total daily irradiance was maximal on date 191, 19 days after the summer solstice (date 172) (Fig. 1a). The proportion of incident light reaching understorey individuals (III_o) had two seasonal peaks affected by both the changing angle of the sun and the extent of canopy closure (Fig. 1b). The peak in autumn (0.4) was 67% of the peak in spring (0.6). III_o was 0.006 during closed canopy. Understorey daily irradiance also had seasonal peaks in spring and autumn (Fig. 1c). Peak daily total irradiance in autumn was only 37% of the peak in spring.

IRRADIANCE AS A FUNCTION OF
PHENOLOGY

Total irradiance received by understorey individuals in shade differed greatly among species because of their contrasting phenologies (Table 2). *Asimina*, *Acer*, *Lindera* and *Carpinus* received only 19.7, 30.5, 32.1 and 33.7%, respectively, of the total irradiance received by *Aesculus*. Understorey individuals of *Acer* and *Aesculus* received only 1.2 and 6.9%, respectively, of the total irradiance received by their conspecifics at the edge (Table 2).

Differences in leaf phenology (Figs 2–4) explain species' differences in percentage of total irradiance received in spring, summer and autumn, and their wide differences in total irradiance (Table 2). Only *Aesculus* received irradiance prior to any expansion of canopy leaves (14.5% of total), and received 97.6% of total irradiance prior to 100% canopy closure in spring. At the other extreme, *Asimina* received only 33% of total irradiance in that period and 45.6% of the total during closed canopy in summer. Irradiance received during autumn avoidance ranged from 0 in *Aesculus* to 18.5% for *Asimina*, and all species had senescing leaves when canopy opening reached 50% (Table 1).

FROST-FREE PERIOD AND PHENOLOGY

Over 30 years, the median Julian date of latest spring freezing temperature was 105 (range 83–127) (box-and-whisker plots, Fig. 1c). The median date was very near to when leaves of canopy trees first appeared and irradiance levels in the understorey began to decrease (Julian date 102). Although in the study years only *Aesculus* had leaves prior to the median day of last spring frost, we have seen frost damage to young leaves

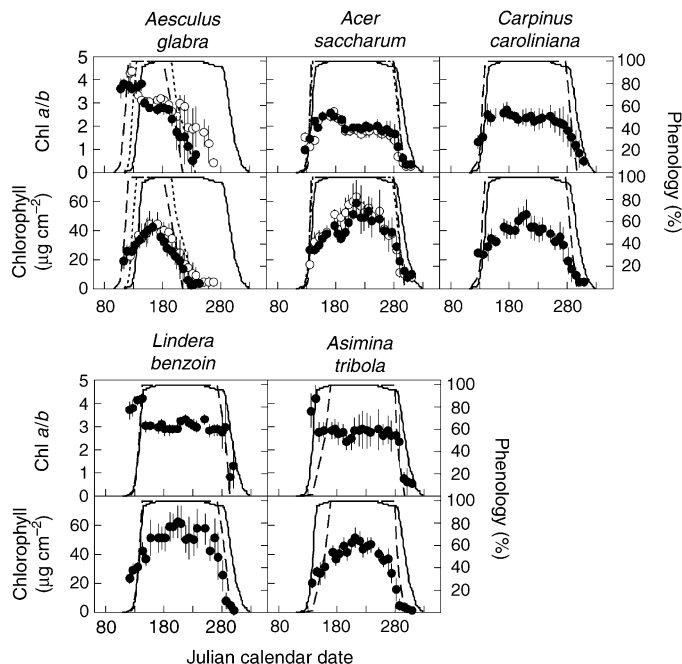


Fig. 2. Seasonal progression of leaf phenology, chlorophyll *a/b* and total chlorophyll ($\mu\text{g cm}^{-2}$) of five woody species. Solid lines, percentage overstorey canopy closure (right vertical axis). Percentage leaf expansion in spring and percentage of maximal green colour remaining in autumn are shown for understorey plants (long dashed lines) and edge plants (short dashed lines for *Aesculus* and *Acer* only) (right vertical axis). Values for chlorophyll, mean ± 1 SD (left vertical axis). For *Aesculus* and *Acer* values are shown for both understorey plants (closed symbols) and edge plants (open symbols).

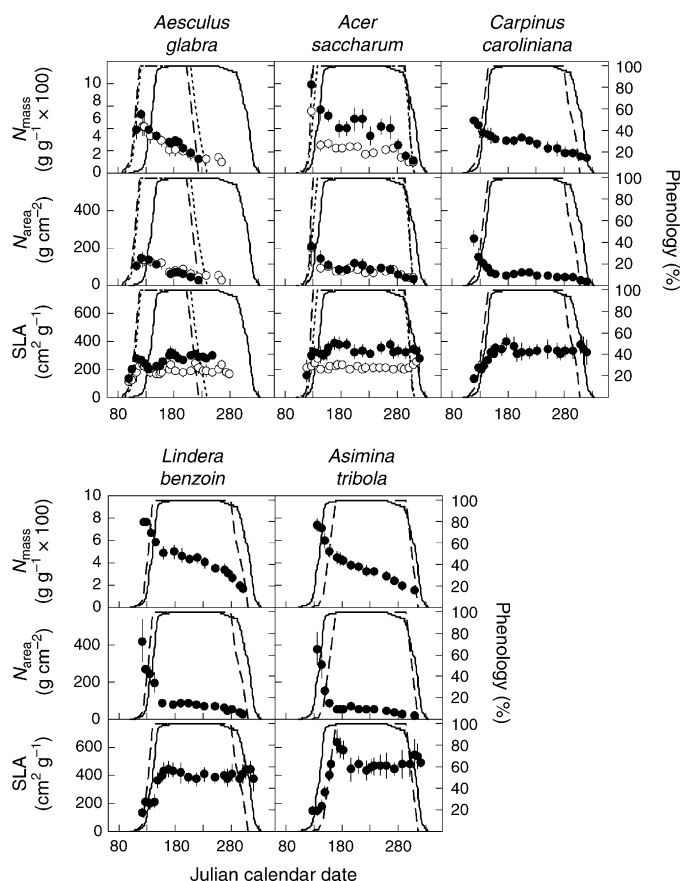


Fig. 3. Seasonal progression of leaf phenology, leaf N content per unit mass [$(N_{\text{mass}}; \text{g N g}^{-1} \text{ DW}) \times 100$]; leaf N content per unit area ($N_{\text{area}}; \mu\text{g cm}^{-2}$); and specific leaf area ($\text{cm}^2 \text{g}^{-1}$) of five woody species. See Fig. 2 for explanation of lines and circles.

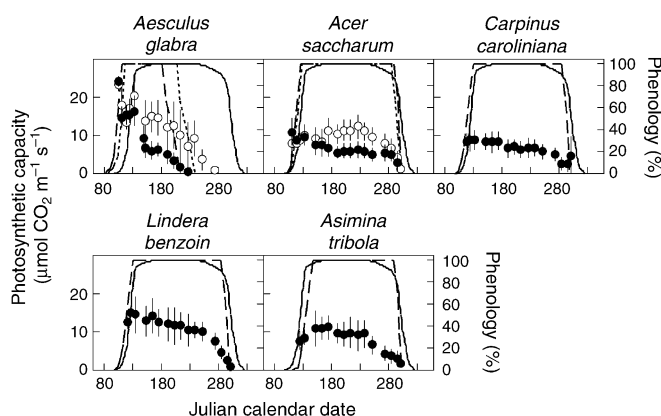


Fig. 4. Seasonal progression of leaf phenology and photosynthetic capacity ($\mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$) at light ($2000 \mu\text{mol m}^{-2} \text{s}^{-1}$) and CO_2 ($2000 \mu\text{mol m}^{-1}$) saturation of five woody species. See Fig. 2 for explanation of lines and circles.

on all species except the late-developing *Asimina* (C.A., personal observation).

The minor amount of phenological avoidance in autumn brings a very minor risk of frost damage prior to senescence. Over 30 years the estimated median date of first autumn freezing temperature was 290 (range 264–316) (Fig. 1c). The canopy was, on average, 87% closed on this date. In the study years all species began

senescence prior to the median date of first autumn frost, and have shown no autumn frost damage except for senescing leaves of *Asimina* (C.A., personal observation).

PHYSIOLOGY AND MORPHOLOGY FOR UNDERSTOREY INDIVIDUALS

Species differed in the seasonal pattern of chlorophyll *alb* ratio, but only during leaf expansion (Fig. 2). The pattern was similar for *Aesculus* and *Asimina*, the species with greatest and least avoidance. The ratio was high initially in *Aesculus*, *Lindera* and *Asimina*, but low in *Acer* and *Carpinus*. All species adjusted quickly to a steady level for the mature phase of leaf life, but ranged from 2.0 in *Acer* to 3.0 in *Lindera*. The ratio declined during senescence in all species.

The seasonal pattern of total chlorophyll was consistent among species and therefore did not reflect their differences in phenological avoidance (Fig. 2). Peak concentration was reached at the middle of leaf life span, and began to decline well before initiation of visible senescence. The maximum level of total chlorophyll ranged from $41.8 \mu\text{g cm}^{-2}$ in *Aesculus* to 63.5 in *Acer*.

The seasonal pattern of leaf N content per unit mass (N_{mass}) was relatively consistent among species (Fig. 3). Regardless of the extent of spring phenological avoidance, it was highest in the youngest leaves and decreased throughout the season. Maximum N content ranged from 0.043 g g^{-1} in *Carpinus* to 0.072 in *Acer*. During the mature leaf stage, values ranged from 0.024 in *Carpinus* to 0.035 in *Acer*, *Asimina* and *Lindera*.

The seasonal pattern of leaf N content per unit area (N_{area}) was also similar among species (Fig. 3). Regardless of the extent of phenological avoidance, N_{area} was highest in the youngest leaf stage, and declined sharply during leaf expansion (except for *Aesculus*) and slowly during mature and senescent leaf stages. The maximum N_{area} ranged from $161 \mu\text{g cm}^{-2}$ in *Aesculus* to 489 in *Lindera*. The values in mature leaves ranged from $55 \mu\text{g cm}^{-2}$ in *Asimina* to 99 in *Aesculus*.

Specific leaf area varied seasonally, but in a similar pattern among species (Fig. 3). It rose rapidly during leaf expansion and remained stable thereafter. Specific leaf area during the mature leaf stage ranged from $286 \text{ cm}^2 \text{g}^{-1}$ in *Aesculus* to 527 in *Asimina*. *Asimina* deviated somewhat from the above pattern, with a decrease following its rapid rise during leaf expansion.

All species except *Asimina* showed a similar seasonal pattern of photosynthetic capacity (light- and CO_2 -saturated CO_2 exchange). It was highest in youngest leaves, and fell shortly after completion of leaf expansion and then continuously throughout the season, markedly so after the beginning of senescence (Fig. 4). This seasonal decline was most pronounced in *Aesculus*. The maximum photosynthetic capacity ranged from $10.4 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ in *Carpinus* and

Table 1. Mean number of days on which individuals of five woody species experience spring shade avoidance, summer non-avoidance and autumn shade avoidance; also shown are median phenological stages of individuals during the three periods

	<i>Aesculus glabra</i>	<i>Acer saccharum</i>	<i>Carpinus caroliniana</i>	<i>Lindera benzoin</i>	<i>Asimina triloba</i>
Mean number of days					
Spring avoidance					
0% canopy closure	23	1	6	9	5
1–50% canopy closure	17	17	17	17	17
51–95% canopy closure	7	7	7	7	7
Summer non-avoidance*	70 (34S)†	138 (3S)	138 (6S)	138 (10S)	138 (25E; 4S)
Autumn avoidance					
5–50% canopy opening	0	15	15	11	13
51–100% canopy opening	0	2	0	0	0
Median percentage of full leaf size					
Spring avoidance					
0% canopy closure	16	1	3	1	<1
1–50% canopy closure	100	27	43	41	1
51–95% canopy closure	100	100	100	100	13
Summer non-avoidance	100	100	100	100	100
Median percentage of complete senescence					
Autumn avoidance					
5–50% canopy opening	–	58	77	67	75
51–100% canopy opening	–	95	–	–	–

*Includes days during 95–100% canopy closure and 0–5% canopy opening.

†Number of days during summer non-avoidance in stage of S (senescence) or E (expansion).

Table 2. Estimated total irradiance on edge and understorey individuals of the five species studied and percentage of total irradiance received by understorey individuals during spring shade avoidance, summer non-avoidance and autumn shade avoidance

	<i>Aesculus glabra</i>	<i>Acer saccharum</i>	<i>Carpinus caroliniana</i>	<i>Lindera benzoin</i>	<i>Asimina triloba</i>
Total irradiance at edge (mol m ⁻²)	3569	6211	NA	NA	NA
Total irradiance in understorey (mol m ⁻²)	246	75.0	82.9	78.9	48.6
% Irradiance: spring avoidance					
0% canopy closure	14.5	0	0	0	0
1–50% canopy closure	77.6	42.0	50.3	49.2	21.0
51–95% canopy closure	5.5	17.9	15.9	16.0	14.9
% Irradiance: summer non-avoidance*	2.4	29.5	26.7	28.1	45.6
% Irradiance: autumn avoidance					
5–50% canopy opening	0	10.6	7.1	6.7	18.5
51–100% canopy opening	0	0	0	0	0

*Includes 95–100% canopy closure and 0–5% canopy opening.

Asimina to 28.3 in *Aesculus*. Mature leaves ranged from 7.9 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ in *Carpinus* to 12.6 in *Lindera*. The seasonal pattern of photosynthetic capacity (Fig. 4) was most strongly related to leaf N (Fig. 3) and did not parallel total chlorophyll (Fig. 2).

Aesculus, the species with most spring phenological avoidance and shortest life span, had unusually high photosynthetic capacity during its phenological avoidance (Fig. 4). Its stable period in mature leaves was short as its photosynthetic capacity began to fall rapidly, well in advance of its early senescence. *Asimina*, the species with least spring phenological avoidance, reached its maximal photosynthetic capacity later, at

full leaf expansion, in contrast to the three species with intermediate spring phenological avoidance (Fig. 4). All species, regardless of the extent of autumn avoidance, had quite low photosynthetic capacity during that period.

PHENOLOGY AND PHYSIOLOGY OF EDGE INDIVIDUALS

The phenology of edge individuals of *Aesculus* post-dated that of understorey individuals for both leaf expansion and senescence (compare short- and long-dashed lines in Figs 2–4). The same pattern held for

Acer, although the difference between understorey and edge individuals was much less than for *Aesculus* (Figs 2–4). Augspurger & Bartlett (2003) describe quantitative details of the differences in phenology between understorey and edge individuals of these two species.

Physiological traits of edge individuals showed seasonal patterns similar to understorey individuals, but traits of edge individuals developed later in accordance with their later phenology (Figs 2–4); once leaf development or senescence began, it proceeded at the same rate in both understorey and edge individuals. Overall, relative to understorey individuals, edge individuals of *Aesculus* had a slightly higher chlorophyll *alb* ratio and total chlorophyll (Fig. 2); similar N_{mass} and N_{area} (Fig. 3); lower SLA (Fig. 3); and greater photosynthetic capacity (Fig. 4) throughout their leaf life span. Overall, relative to understorey individuals, edge individuals of *Acer* had a similar chlorophyll *alb* ratio; higher total chlorophyll; much lower N_{mass} ; similar N_{area} ; lower SLA; and greater photosynthetic capacity throughout their leaf life span.

Discussion

The study revealed large differences among species in their extent of spring phenological avoidance, phenological stage during avoidance, and total annual irradiance and its seasonal distribution. Furthermore, as predicted, species that exhibited greater spring avoidance exhibited more rapid expansion of leaves coupled with rapid development of physiological characteristics that would enhance C gain. Autumn avoidance, in contrast, was minimal and would bring little or no C gain. Senescence was well advanced prior to much canopy opening, and leaves did not maintain or enhance their physiological capacity after canopy leaf drop began.

PHENOLOGY

The wide range of phenological avoidance among the study species is representative of juveniles of canopy tree species in the same forest, where spring avoidance by fully expanded leaves ranges from a few days to 3 weeks (Augspurger & Bartlett 2003). Autumn avoidance is uncommon and minimal, as found in this study. Frost damage to young leaves indicates that the species are currently near the limit of spring phenological avoidance in this locality. A similar extent of shade avoidance occurs in understorey woody individuals of other temperate deciduous forests (dePamphilis & Neufeld 1989; Gill *et al.* 1998).

The mechanism underlying phenological avoidance of *Aesculus* is associated with earlier accumulation of thermal degree hours in the understorey than in the canopy (Augspurger 2005). In that study, *Aesculus* seedlings were grown either at ground level under the shelter of the forest canopy, or exposed at near canopy

height. The latter, because of greater radiation cooling at night, accumulated degree hours more slowly and showed delayed leaf development. The mechanism for avoidance in the other species may be similar to *Aesculus*, given that bud break in many temperate woody species has been linked to winter chilling followed by accumulation of thermal degree days (Hunter & Lechowicz 1992; Heide 1993; Chuine & Cour 1999).

Both spring and autumn phenology were initiated earlier in understorey than in edge individuals, especially in *Aesculus*. Temperature accumulation in spring appears to be faster for understorey than for edge individuals. Both juvenile groups, however, predated canopy conspecifics (data not shown). Thus their phenology somewhat reflects ontogeny, but the environment created by canopy trees further modifies it.

The study species differ as to whether they experience phenological avoidance throughout their lifetime. Shrub/treelet species are permanently in the understorey, and presumably display avoidance throughout their lifetime. The phenology of *Acer* and *Aesculus*, in contrast, shifts through time to become more similar to conspecific canopy trees (Augspurger & Bartlett 2003). Their extent of phenological avoidance diminishes as they age.

LIGHT GAINS VIA PHENOLOGICAL AVOIDANCE

A relatively small increase in spring avoidance brings a disproportionate gain in light. Estimated total irradiance of *Aesculus* with the greatest spring avoidance is five times greater than in *Asimina* with the least spring avoidance. Nevertheless, irradiance of understorey individuals of *Aesculus* and *Acer* is still two orders of magnitude lower than edge and canopy individuals.

Light gains via avoidance are much greater in spring than in autumn. As in other temperate deciduous forests (Hutchison & Matt 1977; Gill *et al.* 1998), the period of closed canopy fell mostly after the summer solstice and peak radiation. The greater light received by understorey individuals in spring than in autumn strongly reflects this asymmetry. It was surprising that peak irradiance in the open was 19 days after the summer solstice. This delay may be due to greater cloud cover in spring.

Light gains in spring are not uniform across the forest due to differential timing of leaf development among species of canopy trees (Shogo & Akira 2002). This study summarized light and canopy closure at the forest level, but an individual understorey plant will experience higher or lower irradiance during spring depending on the specific species of canopy tree above it. The distribution of understorey plants with respect to species of canopy trees is unknown.

Our estimates of total light interception did not take into account transient sunflecks that increase light availability and, potentially, C gain under the closed canopy in summer (Naumburg & Ellsworth 2002).

Canham *et al.* (1994) report that they contribute <10% of the summer radiation beneath late successional species, such as those that predominate at our study site. The estimates of sunflecks were based on hemispheric photographs of the closed canopy, and thus their radiation values did not incorporate light gain during spring or autumn avoidance.

PHYSIOLOGICAL CAPACITY

All species except *Asimina*, the species with minimal avoidance, showed a physiological capacity to capitalize on the high light during spring, but not autumn, avoidance. Photosynthetic capacity, N_{mass} and N_{area} were highest in young, expanding leaves but low in old leaves. Actual photosynthesis, as well as potential, would be higher in spring because soil moisture is more favourable, temperature stress and photorespiration are less, and stomatal conductance is higher (data not shown). In general, older leaves have lower photosynthetic capacity than young leaves (Gill *et al.* 1998; Bond 2000), including in *A. saccharum* (Jurik 1986).

Some forest herbs that spend part of their growing season in canopy shade show photosynthetic acclimation (Rothstein & Zak 2001). The study's woody species, however, did not show evidence of acclimation. *Acer saccharum* has a limited ability to acclimate to increased irradiance (Ellsworth & Reich 1992).

In contrast to the other traits, total chlorophyll did not peak until the middle of leaf life span in all species, as has been shown in shade leaves of *A. saccharum* (Koniger *et al.* 2000). We do not know the reason for the slow accumulation of chlorophyll. The peak occurred at different dates for different species, indicating that it is not related to an increase in leaf area index. The leaf area index peaks at the same time as full expansion of canopy leaves (C.A., unpublished data), while chlorophyll accumulation continues well past that time.

The seasonal pattern of SLA can be used to infer what contributed to the seasonal decline in leaf N. The rapid decline in young leaves was due, at least in part, to a dilution effect from leaf expansion. The continuing, but slow, decline in N after SLA stabilized indicates that N was being translocated from the leaf or leached throughout the mature phase of the leaf, well before the period of leaf senescence. The N did not supply a new cohort of leaves, as all species produced only one leaf cohort. At leaf drop, N content was <1.5% in all species.

The rate of physiological development was similar in understorey and edge individuals of *Acer* and *Aesculus*, with the understorey individuals simply initiating development earlier. Once begun, understorey individuals did not compress leaf development in spring or delay it in autumn, relative to edge individuals. As with phenology, the rate of physiological development appears to be under ontogenetic control, while its initiation is somewhat modified by the environment.

The study predicts that spring phenological avoidance coupled with high physiological capacity will increase C gain and growth. Carbon gain by all the forest's understorey individuals via phenological avoidance should be incorporated into any model considering overall and seasonal C gain of a forest ecosystem (Wilson, Baldocchi & Hanson 2001). Indeed, deciduous juveniles in deciduous forests gain a substantial amount of annual C (Gill *et al.* 1998) and growth (Jones & Sharitz 1989; Seiwa 1998) in periods without canopy closure, but more in spring than in autumn (Harrington, Brown & Reich 1989). In contrast, evergreen understorey plants in deciduous forests can only adjust their physiology to changing light availability. For example, *Juniperus virginiana* has its highest photosynthetic rate in spring, but a high autumnal rate as well (Lassoie *et al.* 1983).

The study has implications for categorizing a species' shade tolerance. *Aesculus glabra* has been classified as shade-tolerant (Wenger 1984). Our study demonstrated, however, that it receives 98% of its light prior to 100% canopy closure; it has young, full-sized leaves with high photosynthetic capacity then, and thus must rely heavily on spring avoidance for C gain during its juvenile stage. Direct evidence for its shade-intolerance has been demonstrated by artificially shading understorey individuals during their normal period of spring phenological avoidance. In that case, we found accelerated leaf senescence, shortened leaf life span, lower growth and higher mortality after only 1 year of treatment (C.A., unpublished data).

The five coexisting species differed greatly in estimated total irradiance, maximal photosynthetic capacity and, presumably, overall C gain. Other things being equal, these differences may result in variation in competitive ability and would be reflected in their relative growth and survival, thus affecting their relative abundance. Indeed, *Aesculus* may assume its prominence in the understorey of this forest because of its great phenological avoidance. *Aesculus* should be less dependent on gaps for recruitment and growth. This temporal partitioning of light may be one mechanism promoting coexistence of these species.

While it is clear that phenological avoidance does not result in total annual light interception or C gain even remotely equivalent to that received by unshaded individuals, its importance for understorey individuals is equally clear. Enhanced C gain via phenological avoidance appears to be the norm for many herbs, saplings of canopy trees, and understorey treelets and shrubs. Phenological avoidance of the low light environment of the closed canopy is thus a common mechanism to enhance C gain of understorey plants in temperate deciduous forests.

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References

- Augspurger, C.K. (2004) Developmental vs. environmental control of early leaf phenology in juvenile Ohio buckeye (*Aesculus glabra*). *Canadian Journal of Botany* **82**, 31–35.
- Augspurger, C.K. & Bartlett, E.A. (2003) Differences in leaf phenology between juvenile and adult trees in a temperate deciduous forest. *Tree Physiology* **23**, 517–525.
- Baldocchi, D., Hutchison, B., Matt, D. & McMillen, R. (1984) Seasonal variations in the radiation regime within an oak–hickory forest. *Agricultural and Forest Meteorology* **33**, 177–191.
- Barnes, J.D., Balaguer, L., Manrique, E., Elvira, S. & Davison, A.W. (1992) A reappraisal of the use of DMSO for the extraction and determination of chlorophylls *a* and *b* in lichens and higher plants. *Environmental and Experimental Botany* **32**, 85–100.
- Bond, B.J. (2000) Age-related changes in photosynthesis of woody plants. *Trends in Plant Science* **5**, 403–409.
- Canham, C.K., Finzi, A.C., Pacala, S.W. & Burbank, D.H. (1994) Causes and consequences of resource heterogeneity in forests: interspecific variation in light transmission by canopy trees. *Canadian Journal of Forest Research* **24**, 337–349.
- Cannell, M.G.R. & Smith, R.I. (1986) Climatic warming, spring budburst, and frost damage on trees. *Journal of Applied Ecology* **23**, 177–191.
- Chaine, I. & Cour, P. (1999) Climatic determinants of budburst seasonality in four temperate-zone tree species. *New Phytologist* **143**, 339–349.
- Ellsworth, D.S. & Reich, P.B. (1992) Leaf mass per area, nitrogen content and photosynthetic carbon gain in *Acer saccharum* seedlings in contrasting forest light environments. *Functional Ecology* **6**, 423–435.
- Gill, D.S., Amthor, J.S. & Bormann, F.H. (1998) Leaf phenology, photosynthesis, and the persistence of saplings and shrubs in a mature northern hardwood forest. *Tree Physiology* **18**, 281–289.
- Hänninen, H. (1991) Does climatic warming increase the risk of frost damage in northern trees? *Plant, Cell & Environment* **14**, 449–454.
- Harrington, R.A., Brown, B.J. & Reich, P.B. (1989) Ecophysiology of exotic and native shrubs in southern Wisconsin (USA). I. Relationship of leaf characteristics, resource availability, and phenology to seasonal patterns of carbon gain. *Oecologia* **80**, 356–367.
- Heide, O.M. (1993) Daylength and thermal time responses of budburst during dormancy release in some northern deciduous trees. *Physiologia Plantarum* **88**, 531–540.
- Hunter, A.F. & Lechowicz, M.J. (1992) Predicting the timing of bud-burst in temperate trees. *Journal of Applied Ecology* **29**, 597–604.
- Hutchison, B.A. & Matt, D.R. (1977) The distribution of solar radiation within a deciduous forest. *Ecological Monographs* **47**, 185–207.
- Iwasa, U. & Cohen, D. (1989) Optimal growth schedule of a perennial plant. *American Naturalist* **133**, 480–505.
- Jones, R.H. & Sharitz, R.R. (1989) Potential advantages and disadvantages of germinating early for trees in floodplain forests. *Oecologia* **81**, 443–449.
- Jurik, T.W. (1986) Seasonal patterns of leaf photosynthetic capacity in successional northern hardwood tree species. *American Journal of Botany* **73**, 1331–1338.
- Kikuzawa, K. (1991) A cost–benefit analysis of leaf habit and leaf longevity of trees and their geographical pattern. *American Naturalist* **138**, 1250–1263.
- Koniger, M., Harris, G.C. & Kibler, E. (2000) Seasonal changes in the physiology of shade leaves of *Acer saccharum*. *Journal of Plant Physiology* **157**, 627–636.
- Lassoie, J.P., Dougherty, P.M., Reich, P.B., Hinckley, T.M., Metcalf, C.M. & Dina, S.J. (1983) Ecophysiological investigations of understory eastern red cedar in central Missouri. *Ecology* **64**, 1355–1366.
- Martin, J.G., Kloeppel, B.D., Schaefer, T.L., Kimbler, D.L. & McNulty, S.G. (1998) Aboveground biomass and nitrogen allocation of ten deciduous southern Appalachian tree species. *Canadian Journal of Forest Research* **28**, 1648–1659.
- Monk, C.D., Child, G.I. & Nicholson, S.A. (1970) Biomass, litter, and leaf surface area estimates of an oak–hickory forest. *Oikos* **21**, 138–141.
- Morecroft, M.D., Stokes, V.J. & Morison, J.I.L. (2003) Seasonal changes in the photosynthetic capacity of canopy oak (*Quercus robur*) leaves: the impact of slow development on annual carbon uptake. *International Journal of Biometeorology* **47**, 221–226.
- Naumburg, E. & Ellsworth, D.S. (2002) Short-term light and leaf photosynthetic dynamics affect estimates of daily understory photosynthesis in four tree species. *Tree Physiology* **22**, 393–401.
- Nelson, N.D. & Isebrands, J.G. (1983) Late-season photosynthesis and photosynthate distribution in an intensively-cultured *Populus nigra* × *Populus laurifolia* clone. *Photosynthetica* **17**, 537–549.
- dePamphilis, C.W. & Neufeld, H.S. (1989) Phenology and ecophysiology of *Aesculus sylvatica*, a vernal understory tree. *Canadian Journal of Botany* **67**, 2161–2167.
- Reich, P.G., Walters, M.B. & Ellsworth, D.S. (1991) Leaf age and season influence the relationships between leaf nitrogen, leaf mass per area and photosynthesis in maple and oak trees. *Plant, Cell & Environment* **14**, 251–259.
- Rothstein, D.E. & Zak, D.R. (2001) Photosynthetic adaptation and acclimation to exploit seasonal periods of direct irradiance in three temperate, deciduous-forest herbs. *Functional Ecology* **15**, 722–731.
- Seiwa, K. (1998) Advantages of early germination for growth and survival of seedlings of *Acer mono* under different overstorey phenologies in deciduous broad-leaved forests. *Journal of Ecology* **86**, 219–228.
- Seiwa, K. (1999a) Changes in leaf phenology are dependent on tree height in *Acer mono*, a deciduous broad-leaved tree. *Annals of Botany* **83**, 355–361.
- Seiwa, K. (1999b) Ontogenetic changes in leaf phenology of *Ulmus davidiana* var. *japonica*, a deciduous broad-leaved tree. *Tree Physiology* **19**, 793–797.
- Shogo, K. & Akira, K. (2002) Spatial and seasonal heterogeneity in understory light conditions caused by differential leaf flushing of deciduous overstorey trees. *Ecological Research* **17**, 687–693.
- Uemura, S. (1994) Patterns of leaf phenology in forest understory. *Canadian Journal of Botany* **72**, 409–414.
- Wang, J., Ives, N.E. & Lechowicz, M.J. (1992) The relation of foliar phenology to xylem embolism in trees. *Functional Ecology* **6**, 69–75.
- Wenger, K.F. (1984) *Forestry Handbook*. John Wiley, New York, NY, USA.
- Wilson, K.B., Baldocchi, D.D. & Hanson, P.J. (2001) Leaf age affects the seasonal pattern of photosynthetic capacity and net ecosystem exchange of carbon in a deciduous forest. *Plant, Cell & Environment* **24**, 571–583.

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