Light- and seasonal-induced plasticity in leaf morphology, N partitioning and photosynthetic capacity of two temperate deciduous species

Sylvain Delagrange [a,b,*]

[a] University of Quebec in Montreal (UQAM), 141 Av. President Kennedy, P.O. Box 8888, Centre-Ville Station, Montreal, QC, Canada H3P 3P8
[b] INRA Centre of Nancy, UMR1137 INRA/UHP, Forest Ecology and Ecophysiology, 54280 Champenoux, France

ARTICLE INFO

ABSTRACT

Processes involved in leaf photosynthetic acclimation to light and throughout the growing season were investigated in two hardwood species (*Acer saccharum* and *Betula alleghaniensis*), which differed in their level of shade-tolerance. For both species, variation in traits related to (i) leaf morphology (*LMA*, leaf mass:area ratio), (ii) leaf N content (N_A, leaf nitrogen content on an area basis and N_M, N concentration in leaf dry mass), (iii) leaf N partitioning among photosynthetic functions (P_r, N allocated to Rubisco, and P_b, N allocated to bioenergetics), and (iv) leaf photosynthetic capacity (V_{cmax}, maximal carboxylation rates, and J_{max}, maximal light-driven electron flow) were assessed at three different times during the growing season (early, mid- and late summer) and under four contrasting light regimes (40, 17, 6 and 2% of full sunlight). For both species, light-driven variation in most traits was greater than their seasonally driven variation. Furthermore, results showed for both species the pre-eminence of *LMA* changes in the light-driven acclimation of N_A. Importance of N_M to variation in N_A was restricted to seasonal acclimation, especially for the less shade-tolerant species, *B. alleghaniensis*. Similarly, for both species, light-driven acclimation of leaf photosynthetic capacities was tightly related to variation in N_A, which was related to *LMA* changes. However, variation in P_r and P_b better explained seasonally driven variation in V_{cmax} and J_{max}, specifically under lower light levels, where N_A was low. Thus, the great variability observed for leaf activity in response to contrasting light environments was related to efficient morphological adjustments, regardless of species level of shade-tolerance. Finally, physiological adjustments were mainly involved in fine-scale changes observed during seasonally driven acclimation of leaves, when *LMA* was constrained to a slight range of variation.

© 2010 Elsevier B.V. All rights reserved.

1. Introduction

Performance and coexistence of tree species is based on their abilities to acclimate and respond to various environments and to environmental variation. The leaf is one of the most studied among tree organs, in terms of acclimation, due to the critical role of photosynthetic tissues in the carbon balance of the individual. Leaf acclimation to environmental changes occurs mainly through modifications to (i) leaf morphology (i.e., leaf form and display), (ii) leaf anatomy (i.e., leaf density and number and thickness of cell layers), and (iii) leaf physiology (i.e., protein synthesis and activities associated with light capture, electron transport, and carbon fixation). Changes to one, or a combination of these traits, have already been associated with photosynthetic acclimation and patterns of carbon gain maximisation in response to environmental changes, such as variation in light, temperature, and salinity, among others (Larcher, 1995; Dreyer et al., 2001; Poorter et al., 2009). In temperate deciduous species, leaf acclimation to variation in the light environment has been extensively studied (e.g., Walters and Reich, 1999; Niinemets et al., 1998; Frak et al., 2001; Terashima et al., 2006; Valladares and Niinemets, 2008), since this resource is the factor that most limits the regeneration of temperate deciduous seedlings (Coomes and Grubb, 2000; Ricard et al., 2003).

Light-driven changes in leaf morphology and anatomy typically result in a modification of the leaf surface that develops per unit of leaf dry mass invested. For instance, under low light levels, the leaf mass-to-area ratio (*LMA*, g m^{-2}) decreases as a result of decreasing leaf thickness (Terashima et al., 2006). This maximizes the leaf surface exposed to the sun, while reducing overall leaf respiration (Reich et al., 1998; Niinemets, 2006). In contrast, higher *LMA* under high light levels is attained with a greater number or a greater size of palisade cells (Terashima et al., 2006). With such an increase in cell thickness, sun leaves can sustain higher rates of photosynthesis per unit area. A second level of leaf acclimation occurs through the

relationship between leaf N content versus leaf photosynthesis and respiration (e.g., Givnish, 1988; Reich et al., 1998; Niinemets and Tenhunen, 1997; Niinemets, 2006). On a mass basis, high N content (N_M, g N g^{-1} leaf dry mass) increases carbon assimilation through an increase in the pool of enzymes and other proteins involved in light interception (e.g., Chlorophyll a and b), light conversion (e.g., cytochrome f, plastoquinone, plastocyanin, and ferredoxin) and carbon fixation (e.g., Rubisco and other enzymes of the Calvin cycle). However, a larger N_M is also directly associated with greater rates of growth and maintenance respiration (Thornley and Cannell, 2000), suggesting further ecological limitation in the variation of these traits (Valladares et al., 2007). Finally, another level of leaf acclimation is expected through modifications in N allocation to various leaf functions. For instance, under higher light regimes, greater N allocation to carbon fixation and electron transport is usually expected, since light capture is not limiting (Niinemets et al., 1998). In contrast, under low light regimes, N allocation to light capture (e.g., chlorophylls and other pigments) is expected to increase, to the detriment of N allocation to Rubisco or electron transporters (Frak et al., 2001; Evans and Poorter, 2001).

Few studies have discussed the relative contribution of variation in LMA, N_M, and N partitioning in overall leaf response (Niinemets et al., 1998; Reich et al., 1998; Le Roux et al., 2001; Frak et al., 2001). Changes in leaf N content are generally reported on an area basis (N_A, g m^{-2}), which is the product of LMA and N_M. However, the increasing use of this integrated foliar trait to describe variation in leaf N (especially for deciduous species) generally has provided no clear understanding of the patterns of leaf N_A variation that arise in response to the environment. Moreover, variation in N_M in response to light acclimation, both within- and between-species, has not yet been clearly elucidated. Most deciduous species have displayed very stable N_M in response to light (e.g., Casella and Ceulemans, 2002; Niinemets et al., 1998; Evans and Poorter, 2001). These observations in deciduous species have generally led to the conclusion that LMA takes a preeminent role in controlling light-induced variation in N_A (Niinemets et al., 1998; Le Roux et al., 2001; Frak et al., 2001). However, several species such as *Populus nigra*, *P. deltoides*, *Corylus avellana*, *Betula pendula*, and *Eucalyptus grandis* (Casella and Ceulemans, 2002) tend to contradict this theory by showing light-driven changes in N_M. Finally, little is known about the importance of possible adjustments in N partitioning among leaf functions as a response to light acclimation (Frak et al., 2001). Several studies have already reported that this partitioning of N to different leaf functions (e.g., CO$_2$ fixation, electron transport, cell structure) may be critical to leaf carbon gain abilities, since they are competing for the same available N (Evans and Poorter, 2001; Onoda et al., 2004; Feng et al., 2009; Hikosaka and Shigeno, 2009).

This study also aims to assess seasonally driven acclimation of leaves grown under contrasting light regimes. During the growing season, large morphological adjustments are not expected after the leaf expansion phase (Ellsworth and Reich, 1992; Poorter et al., 2009). However, several studies have shown strong seasonal variation in leaf photosynthetic capacities and CO$_2$ assimilation, which were associated with physiological modifications (Kull et al., 1998; Frak et al., 2001; Muller et al., 2005; Yasumura et al., 2006; Katahata et al., 2007), but no clear patterns have yet been reported for temperate deciduous species. Furthermore, Naidu and DeLucia (1997) already showed little to no seasonal adjustment under low light for more shade-tolerant species such as *Acer saccharum*, while they reported adjustments for the more shade-intolerant *Quercus robur*. Seasonal acclimation to light transfer was assessed in *Juglans regia* (Frak et al., 2001) and was associated with morphological and physiological adjustments, depending if the light availability was increased or decreased, respectively.

Thus, to help determine the importance of morphological and physiological adjustments, which co-occur, in leaf acclimation to light and seasonally driven changes, key traits involved in leaf performance were investigated, and more specifically, the origin of their variation. This was examined in two hardwood species that differ in their functional ecology, but co-occur in the same temperate forest stands of eastern North America: sugar maple (*A. saccharum* Marsh.), which is a shade-tolerant species; and yellow birch (*Betula alleghaniensis* Britt.), which is an intermediate shade-tolerant species. Based on literature reports, more shade-intolerant species are expected to rapidly adjust their physiology to efficiently use variation in irradiance and enhance leaf carbon gain, while shade-tolerant species tend to adjust their leaf morphology with little modification to their physiology (Bazzaz and Carlson, 1982; Valladares et al., 2002).

2. Materials and methods

2.1. Plant material and growing conditions

Seeds of the two species were provided by the Ministry of Natural Resources and Wildlife of Quebec (MRNFQ) from its Duchesnay forest station (46°55′N, 71°40′W), which is located in the west-central region of the province. During early spring (2001), the seeds were germinated in a glasshouse, which was located in Champenoux (48°44′N, 6°14′E), near Nancy (France), using a cold room pre-treatment for *A. saccharum* and a liquid fertiliser addition for *B. alleghaniensis*. One hundred seedlings of each species were planted into ten-litre pots filled with a peat-sand mixture (1:3, v/v). All pots were placed in a nursery under nets providing four contrasting light regimes: 40%, 17%, 6%, and 2% of full sun. Shading was provided by neutral aluminised nets, which decreased light quantity without modifying its spectral composition (Delagrange et al., 2006). Seedlings were fertilised twice a year (beginning of May and beginning of August) with 4 g L^{-1} soil Nutricote 100 (13–13–13 N–P–K with micro-elements; Plant Products Co. Ltd., Brampton, ON) and watered to field capacity twice a day using drip irrigation.

2.2. Leaf sampling

Leaf sampling occurred under the four light regimes during the second year of growth (2002), and was repeated over three time periods of the growing season. These periods corresponded to "early summer" (5–20 June), "mid-summer" (15–30 July), and "late summer" (1–15 September). Under each light regime and for each period of the season, six healthy individuals were randomly chosen from among available individuals for both species. It should be noted that, when an individual was selected for measurements, it was automatically removed from the available pool of individuals for the subsequent samplings to avoid any effect of repeated leaf sampling. Since *A. saccharum* experienced significant mortality in 2001 under the lowest light regime (2% of full sunlight) due to excessive moisture, no individuals were sampled in early summer 2002 under this light treatment. For the same reasons, subsequent sampling of *A. saccharum* under the 2% LR consisted of selecting four individuals (instead of 6). Consequently, a total of 62 and 72 fully developed leaves of *A. saccharum* and *B. alleghaniensis*, respectively, were used for measurements of leaf traits.

2.3. Leaf gas exchange and photosynthetic capacity

Selected leaves were used to generate A/C_i curves with an infrared gas analyser system (Model 6400, LI-COR, Lincoln, NE, USA). Conditions in the chamber were 25 °C for leaf temperature, between 60% and 75% RH, and saturating irradiance (between 1000 and 1700 μmol m^{-2} s^{-1} PAR for the lowest and highest light regimes, respectively). Generation of the A/C_i curves began with a