A biochemical model of photosynthesis for mango leaves: evidence for the effect of fruit on photosynthetic capacity of nearby leaves

L. URBAN, X. LE ROUX, H. SINOQUET, S. JAFFUEL and M. JANNOYER

Summary  Variations in leaf nitrogen concentration per unit mass ($N_a$) and per unit area ($N_a$), mass-to-area ratio ($M_a$), total nonstructural carbohydrates ($T_a$), and photosynthetic capacity (maximum carboxylation rate, electron transport capacity, rate of phosphate release in triose phosphate utilization and dark respiration rate) were studied within the digitized crowns of two 3-year-old mango trees (Mangifera indica L.) on La Réunion Island. Additional measurements of $N_a$, $M_a$, $T_a$ and photosynthetic capacities were performed on young, fully expanded leaves of 11-year-old mango trees. Leaves of similar gap fractions were taken far from and close to developing fruits. Unlike $N_a$, both $T_a$ and $M_a$ were linearly correlated to gap fraction. Similar relationships were found for all leaves whatever their age and origin, except for $T_a$ for which we found a significant tree effect. Photosynthetic capacity was non-linearly correlated to $N_a$ and a unique relationship was obtained for all types of leaves. Photosynthetic acclimation to light was mainly driven by changes in $M_a$, but allocation of total leaf N between the different photosynthetic functions also played a substantial role in acclimation to the lowest irradiiances. Leaves close to developing fruits exhibited a higher photosynthetic capacity than other leaves, but similar $T_a$. Our data suggest that $T_a$ does not control photosynthetic capacity in mango leaves. We used the data to parameterize a biochemically based model of photosynthesis and an empirical stomatal conductance model, allowing accurate predictions of net photosynthesis of leaves in field-grown mango trees.

Keywords: leaf age, leaf irradiance, leaf nitrogen, Mangifera indica, nonstructural carbohydrates, radiation transfer model.

Introduction  Biochemically based models of leaf photosynthesis are used increasingly to compare photosynthetic performance among plant species (Wullschleger 1993) and to analyze photosynthetic acclimation to high CO$_2$ concentrations (Harley et al. 1992, Peterson et al. 1999) and growth irradiance (Niinemets and Tenhunen 1997, Le Roux et al. 1999a, 2001a). Such models may also be coupled to radiation transfer models to simulate photosynthesis at the canopy level (Harley et al. 1985, Harley and Tenhunen 1991, Harley and Baldocchi 1995, De Pury and Farquhar 1997) or at the individual plant level (Le Roux et al. 2001b, Sinoquet et al. 2001).

Applying biochemically based photosynthesis models at the individual plant level requires representation of the intra-canopy distribution of leaf photosynthetic capacity (i.e., the maximum rate of carboxylation ($V_{\text{max}}$), the light-saturated rate of electron transport ($J_{\text{max}}$), the rate of phosphate release in triose phosphate utilization (TPU) and the mitochondrial respiration rate due to phosphorylative oxidations ($R_{\text{b}}$)). Because proteins of the Calvin cycle and thylakoids represent the majority of leaf nitrogen, photosynthetic capacity is strongly related to leaf nitrogen on an area ($N_a$) or mass ($N_a$) basis (Field and Mooney 1986, Evans 1989a, Kellomäki and Wang 1997, Walscot et al. 1997). Furthermore, for leaves of a given age and for a given nitrogen supply, $N_a$ appears to be strongly related to light exposure (DeJong and Doyle 1985, Le Roux et al. 1999b, 2001a, Rosati et al. 1999, 2000). Therefore, the intra-canopy distribution of leaf photosynthetic capacity has generally been taken into account by documenting (1) the relationships between leaf photosynthetic capacity and $N_a$ and (2) the relationships between $N_a$ and the mean leaf irradiance experienced locally within the canopy. Such an approach is based on the assumption that plants allocate nitrogen resources within the canopy to enhance photosynthetic capacity in locations experiencing high irradiances, thereby maximizing whole-plant carbon gain (Field 1983, Hollinger 1996, Carswell et al. 2000).

Modeling the distribution of photosynthetic carbon gain within individual fruit trees has been suggested as a promising...
tool to predict the intra-crown heterogeneity of fruit growth (Grossman and DeJong 1994, Lescourret et al. 1998, Le Roux et al. 2001b), which is of importance for fruit production quality (Génard et al. 1998). However, several studies have emphasized that leaf characteristics can be influenced by the presence or proximity of developing fruits in tree crowns. Lower leaf nitrogen concentrations have been observed in fruiting peach trees than in non-fruiting peach trees (Taylor and May 1967, Taylor and Van den Ende 1969) and citrus (Lenz 2000). In contrast, leaf N concentration was higher in fruiting apple trees than in non-fruiting apple trees (Thiebus-Kaesberg and Lenz 1994), whereas leaf mass-to-area ratio (M) and chlorophyll concentrations were higher in the leaves of fruit-bearing shoots of olive trees (Proietti 2000). Decreases in leaf N concentration during the fruit growth period could be attributed to N dilution in growing leaves or to remobilization of N from leaves to fruits (Rufat and DeJong 2001). The influence of developing fruits on leaf characteristics could also be explained by differences in carbohydrate accumulation in leaves resulting from local source–sink imbalances, because carbohydrates can exert a feedback inhibition on leaf photosynthesis and leaf photosynthetic capacity (Foyer 1988). Little is known about the effects of local source–sink balance and associated changes in carbon export rate from leaves and leaf total nonstructural carbohydrate (TNC) concentration on photosynthetic capacity within the crowns of field-grown trees. This lack of knowledge may restrict our ability to accurately predict the spatial distribution of local carbon gain and fruit growth within the canopies of fruit trees.

The objectives of this study were: (1) to quantify spatial variations in leaf photosynthetic capacity (N, Vc max, j max and R) with changes in irradiance in the crowns of mango (Mangifera indica L.) trees growing with non-limiting N supply; (2) to test whether photosynthetic capacities measured at a given irradiance are affected by leaf age and the proximity of developing fruits, and whether leaf TNC accumulation can be used as an indicator of a local source–sink imbalance; and (3) to propose a model of photosynthesis for mango leaves that can be applied to non-fruiting 3-year-old trees and to fruiting 11-year-old trees. The effect of fruit development was studied in 11-year-old trees by comparing leaves with similar light access, but located far from and close to developing fruits.

Materials and methods

Plant material

Measurements were performed on leaves from mango trees, cv. Lirfa, grafted on ‘Maison rouge,’ at two locations on La Réunion Island (20°52′48″ S, 55°31′48″ E). We studied fruiting 11-year-old trees grown in an experimental orchard near Saint-Pierre, and non-fruiting 3-year-old trees grown at a commercial farm near Saint-Paul. The 11-year-old trees were spaced at 5 m (within rows) × 7 m (between rows) and were about 3 m high, with a north-east–south-west row orientation. Access to tree tops was provided by scaffolding. The 3-year-old trees were spaced at 6 m (within rows) × 6 m (between rows) and were about 1.5 m high, with a north-west–south-east row orientation.

Water was supplied every second day on a 100% actual evapotranspiration basis. Variations in stem diameter at Saint-Pierre were monitored with eight linear variable displacement transducers (LVDT; Solartron, Bognor, U.K.). Maximal daytime shrinkage was less than 20 µm for 3- to 4-cm thick stems, indicating that no water stress occurred during our trial (data not shown). The 11-year-old trees received 350 g N tree−1 (urea), 70 g P tree−1 (superphosphate) and 350 g K tree−1 (potassium sulfate) on March 1, 2000 (after harvest), and 700 g N tree−1 (urea), 70 g P tree−1 (superphosphate) and 350 g K tree−1 (potassium sulphate) on October 31, 2000 (at the beginning of the fruit growing phase). Each 3-year-old tree received 210 g urea, 210 g potassium sulfate and 210 g superphosphate in July, 210 g urea and 210 g potassium sulfate in October, and 420 g urea and 420 g potassium sulfate in March. Insects and diseases were controlled according to recommendations commonly accepted for mango orchards.

Photosynthesis model

Net CO2 assimilation rate (A net; µmol CO2 m−2 s−1) in C3 plants is a function of the carboxylation rate (Vc) and the oxygenation rate (Vo) and the rate of CO2 evolution in light that results from processes other than photorespiration (Rd):

\[ A_{\text{net}} = V_{c} - 0.5V_{o} - R_{d} \]

According to a modified version (Harley et al. 1992) of the model proposed by Farquhar et al. (1980), A net can be expressed as:

\[ A_{\text{net}} = (1 - 0.5O/(\tau C_{i}))(w_{1}w_{2}w_{3} - R_{d}) \]

where O represents partial pressure of O2 in the intercellular air spaces (Pa), τ is the specificity factor of ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco), Ci is the partial pressure of CO2 in the intercellular air spaces (Pa), Wj is the carboxylation rate limited by the amount, activation state or kinetic properties of Rubisco (µmol CO2 m−2 s−1), Wc is the carboxylation rate limited by the rate of ribulose bisphosphate regeneration (µmol CO2 m−2 s−1) and Wj is the carboxylation rate limited by triose phosphate utilization in sucrose and starch synthesis (µmol CO2 m−2 s−1). The limitations on Wc can be expressed as:

\[ W_{c} = V_{\text{max}}C_{i}/(C_{i} + K_{i}(1 + O/K_{o})) \]

where Vmax represents maximum rate of carboxylation (µmol CO2 m−2 s−1), and K (Pa CO2) and K (Pa O2) are the Michaelis constants of Rubisco carboxylation and oxygenation, respectively.

Parameter Wj is controlled by the rate of electron flow J (µmol electrons m−2 s−1):

\[ W_{j} = JC_{i}/(4(C_{i} + O/\tau)) \]
with

\[ J = \alpha Q/(1 + \alpha^2 Q^2/J_{\text{max}}^{0.5}) \] (5)

where \( Q \) is photosynthetically active photon flux density (\( \mu \)mol m\(^{-2}\) s\(^{-1}\)), \( \alpha \) is apparent efficiency of light energy conversion (mol electrons per mol photons) and \( J_{\text{max}} \) is light-saturated rate of electron transport (\( \mu \)mol m\(^{-2}\) s\(^{-1}\)). The limitations on \( W_p \) can be expressed as:

\[ W_p = 3\text{TPU} + V_p/2 = 3\text{TPU} + V_c 0.5 C_l/(C_l\tau) \] (6)

where TPU is the rate of phosphate release in triose phosphate utilization (starch and sucrose production).

The temperature dependency of \( V_{\text{max}}, J_{\text{max}} \) and TPU is described by:

\[ \text{Parameter}(V_{\text{max}}, J_{\text{max}}, \text{TPU}) = \frac{\exp(-\Delta H_s/(RT_l))}{1 + \exp((\Delta S T_l - \Delta H_s)/RT_l)} \] (7)

where \( c \) is a scaling factor, \( \Delta H_s(J \text{ mol}^{-1}) \) is the activation energy of the given parameter, \( R \) is the gas constant (8.3143 J K\(^{-1} \text{ mol}^{-1}\)), \( T_l \) (K) is leaf temperature, \( \Delta S(J \text{ mol}^{-1}) \) is an entropy term and \( \Delta H_s(J \text{ mol}^{-1}) \) is the deactivation energy of the given parameter. Similarly, the temperature dependency of \( R_\alpha, \tau, K_c \) and \( K_o \) is described by:

\[ \text{Parameter}(R_\alpha, \tau, K_c, K_o) = \exp(-\Delta H_s/(RT_l)) \] (8)

To account for the relationship commonly observed between the parameters defining photosynthetic capacity and \( N_a \) (Field 1983, Leuning et al. 1991, Harley et al. 1992), scaling factors \( c \) may be linearly related to \( \ln(N_a) \):

\[ c = a_N \ln(N_a) + b_N \] (9a)

We also tested the following relationship (see Results):

\[ c = a_N N_a^{-1} + b_N \] (9b)

The model proposed by Niinemets and Tenhunen (1997) was applied to determine the coefficients for leaf nitrogen partitioning into carboxylation, \( P_c \), and bioenergetic pools, \( P_b \). The computation of \( P_c \) (g N in Rubisco per g total leaf N) and \( P_b \) (g N in cytochrome f, ferredoxin, NADP reductase and coupling factors, per g total leaf N) is given by:

\[ P_c = V_{\text{max}} (6.25V_c N_a) \] (10)

\[ P_b = J_{\text{max}} (8.06J_{\text{inc}} N_a) \] (11)

where \( V_c \) is the specific activity of Rubisco (\( \mu \)mol CO\(_2\) g\(^{-1}\) Rubisco) and \( J_{\text{inc}} \) is the potential rate of photosynthetic electron transport per unit of cytochrome f (mol electrons (mol cyt. f\(^{-1}\) s\(^{-1}\)). We used \( V_c = 31.9 \mu \text{mol CO}_2 \text{ g}^{-1} \text{ Rubisco and } J_{\text{inc}} = 180.7 \text{ mol electrons (mol cyt. f)}^{-1}\) at a leaf temperature of 30°C as proposed by Niinemets and Tenhunen (1997). The factor 6.25 (g Rubisco per g N in Rubisco) converts N content to protein content, and the factor 8.06 (mmol cyt. f per g N in bioenergetic pools) assumes a constant 1:1:1.2 molar ratio for cyt. f:ferredoxin NADP reductase:coupling factor.

Parameters \( V_{\text{max}}, J_{\text{max}}, \text{TPU} \) and \( R_\alpha \) were derived from \( A-C_i \) curves taken in January–February 2001 in 11-year-old trees and March–April 2001 in 3-year-old trees (see below). Other parameters were derived from Harley et al. (1992), with the exception of \( \alpha, K_c, \tau, \Delta H_s, \Delta H_o, \Delta A, \alpha, \) and \( \Delta D \), which were preferred to those determined from in vitro leaf gas exchange measurements performed on a Rubisco-antisense line of tobacco (\textit{Nicotiana tabacum} L. cv. W38) by Bernacchi et al. (2001) (Table 1).

\[ \Delta C_i = C_a - A_{\text{net}}/g_b - A_{\text{net}}/g_s \] (13)

where \( C_a \) is the partial pressure of CO\(_2\) (Pa) in ambient air and \( g_b \) is the leaf boundary layer conductance, was used in addition to Equations 2 and 12 to find an analytical solution for the coupled leaf photosynthesis–stomatal conductance model (see Table 1. Values and units of the parameters used in the photosynthesis model for mango (Equations 1 to 9). All parameter values are derived from Harley et al. (1992) and Bernacchi et al. (2001).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Units</th>
<th>Source</th>
</tr>
</thead>
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<tr>
<td>( \alpha )</td>
<td>0.24</td>
<td>mol mol(^{-1})</td>
<td>Harley et al. (1992)</td>
</tr>
<tr>
<td>( c(K_c) )</td>
<td>35.747</td>
<td>dimensionless</td>
<td>Bernacchi et al. (2001)</td>
</tr>
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<td>( c(K_o) )</td>
<td>17.997</td>
<td>dimensionless</td>
<td>Bernacchi et al. (2001)</td>
</tr>
<tr>
<td>( c(\tau) )</td>
<td>-7.458</td>
<td>dimensionless</td>
<td>Bernacchi et al. (2001)</td>
</tr>
<tr>
<td>( \Delta H_s(K_c) )</td>
<td>80470</td>
<td>J mol(^{-1})</td>
<td>Bernacchi et al. (2001)</td>
</tr>
<tr>
<td>( \Delta H_s(K_o) )</td>
<td>14510</td>
<td>J mol(^{-1})</td>
<td>Bernacchi et al. (2001)</td>
</tr>
<tr>
<td>( \Delta H_s(\text{TPU}) )</td>
<td>79500</td>
<td>J mol(^{-1})</td>
<td>Harley et al. (1992)</td>
</tr>
<tr>
<td>( \Delta H_s(T) )</td>
<td>53100</td>
<td>J mol(^{-1})</td>
<td>Harley et al. (1992)</td>
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<tr>
<td>( \Delta H_s(V_{\text{max}}) )</td>
<td>116300</td>
<td>J mol(^{-1})</td>
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<tr>
<td>( \Delta H_s(\text{TPU}) )</td>
<td>-37830</td>
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<td>201000</td>
<td>J mol(^{-1})</td>
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<td>( \Delta H_s(\text{TPU}) )</td>
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<td>J mol(^{-1})</td>
<td>Harley et al. (1992)</td>
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<tr>
<td>( \Delta S(\text{TPU}) )</td>
<td>650</td>
<td>J K(^{-1}) mol(^{-1})</td>
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</tr>
<tr>
<td>( \Delta S(V_{\text{max}}) )</td>
<td>650</td>
<td>J K(^{-1}) mol(^{-1})</td>
<td>Harley et al. (1992)</td>
</tr>
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Baldocchi 1994).

Parameters $g_0$, $a_1$ and $D_0$ were derived from concomitant $A_{\text{act}}$ and $g_s$ measurements performed on leaves from 3- and 11-year-old trees, similar to those used for parameterization of the photosynthesis model.

Gas exchange measurements

Gas exchange was measured with an infrared gas analyzer and leaf chamber system with a red + blue light source (LI-6400, Li-Cor, Lincoln, NE). Calculations were performed according to von Caemmerer and Farquhar (1981).

At Saint-Paul, measurements were made to study the relationship between leaf photosynthetic capacity, total nitrogen per unit leaf area and mean irradiance in two 3-year-old trees. Forty-four leaves encompassing full light and shade conditions were selected within the canopies of the selected trees and tagged. After completion of the tree digitizing procedure (see below), $A$–$C_i$ curves were established for 14 of these leaves from March 19 to April 5, 2001. Leaves were exposed to high irradiance and ambient CO$_2$ concentration for at least 15 min before starting $A$–$C_i$ curves. Thirteen measurements were taken ($C_a = 200, 180, 160, 140, 120, 100, 80, 60, 40, 30, 20, 10$ and $5$ Pa CO$_2$) for each $A$–$C_i$ curve. Conditions in the leaf chamber were controlled ($T = 30 \degree C$, vapor pressure deficit at the leaf surface $= 1.0 \pm 0.2$ kPa). The value of $Q$ was set to maintain the point of transition between the Rubisco-limited and RuBP regeneration-limited segments in the center of the CO$_2$ response curves. We set $Q$ at 2000 mmol m$^{-2}$ s$^{-1}$ for sun leaves and 500 mmol m$^{-2}$ s$^{-1}$ for shade leaves. The best fit $V_{\text{max}}, J_{\text{max}},$ TPU and $R_3$ values were inferred by nonlinear least square regressions (S-Plus 2000, MathSoft International, Bagshot, U.K.). Values of $R_3$ were derived from $A$–$C_i$ curves and found to be in good agreement with values estimated from measurements of CO$_2$ evolution rates after 10 min in the dark.

The relationship between photosynthetic capacity and $N_a$, and the effect of developing fruits on the photosynthetic capacity of nearby leaves were measured in January–February 2001 in four 11-year-old trees at Saint-Pierre. We compared $V_{\text{max}}, J_{\text{max}},$ TPU and $R_3$ derived from $A$–$C_i$ curves obtained on seven young mature leaves from the same last flush without fruits nearby (standard leaves), and on eight leaves, also from the last flush, but close to developing fruits (similar to standard leaves in all other aspects). All leaves had the same orientation and were at similar heights (about 1.7 m). Fish-eye pictures were taken to make sure that gap fractions did not differ between treatments (HemiView 3.1 SR1, Delta-T Devices, Cambridge, U.K.). The procedure applied to establish $A$–$C_i$ response curves was the same as that described for the Saint-Paul study.

Measurements for $g_s$ modeling were performed in the irradiance tracking mode to minimize light fluctuations (target value coming from the external quantum sensor, potentially changing every 3 s), on well-exposed young mature leaves similar to those used for the $A$–$C_i$ curves. The partial pressure of CO$_2$ in the air was set at 36 Pa. Leaf gas exchange rates of the same six leaves were monitored (one measurement per leaf during each 90-min period from 0730 to 1800 h). Two sets of data were obtained: one set for standard leaves and leaves close to developing fruits from 11-year-old trees at Saint-Pierre, and the other set for standard leaves from 3-year-old trees at Saint-Paul.

Additional gas exchange measurements were performed to test the model for standard leaves ($N_a = 2.15 + 0.03$ g N m$^{-2}$) and leaves close to developing fruits ($N_a = 2.63 + 0.07$ g N m$^{-2}$) on the 11-year-old trees at Saint-Pierre. Leaves used for model testing were all well-lit, fully developed leaves from the last vegetative flush, experiencing similar gap fractions. During the measurements, $Q$ was in the 70 to 1870 mmol m$^{-2}$ s$^{-1}$ range. Temperature and humidity of the air were not controlled, but the leaf chamber was used in the irradiance tracking mode, and the partial pressure of CO$_2$ in the air was set at 36 Pa to favor rapid stabilization. Measurements were performed on 12 different leaves every 2.5 h from 0730 to 1930 h on one selected clear day.

Leaf nitrogen and nonstructural carbohydrates

Leaves used for $A$–$C_i$ curves and all labeled leaves on the 3-year-old trees were collected and weighed. Leaf areas were measured and the leaves were frozen in liquid nitrogen. Total leaf nitrogen was determined with an elemental analyzer (Carlo Erba Instruments, Milano, Italy). Glucose, fructose and sucrose in the leaves were measured by an enzyme-based analyzer (YSI 2007, Yellow Springs Instruments, Yellow Springs, OH). Starch was determined after enzymatic hydrolysis to glucose (Thievend et al. 1972).

Tree digitizing and leaf irradiance computation

The architecture of the two 3-year-old trees at Saint-Paul was measured with a 3D digitizing technique (Sinoquet and Rivet 1997). Leaf location and orientation on individual shoots were recorded with an electromagnetic 3D digitizer (Fasttrak, Polhemus, Colchester, VT) and the 3A software package (Adam et al. 1999). The spatial coordinates and the orientation of each leaf were recorded at the junction point between petiole and lamina, by setting the digitizer pointer parallel to the leaf lamina (Sinoquet et al. 1998). Concurrently, leaf length was measured and leaf age was recorded.

Leaf samples were used to establish allometric relationships (1) between leaf length ($L; \text{cm}$) and width ($W; \text{cm}$), and (2) between $L$ and leaf area ($A; \text{cm}^2$), calculated from digitized images taken with a standard flat-bed scanner, using Mesurim software (Madre 1998).

$$W = 0.2507L (r^2 = 0.71, n = 34)$$

$$A = 0.7451 L^2 (r^2 = 0.98, n = 108)$$

Directional light interception was computed with VegeSTAR graphics software (Adam et al. 2000). VegeSTAR allows the visualization of 3D digitized plants and computation of light interception from image processing of virtual plant pictures (Sinoquet et al. 1998). The software computes the
projected lit area by counting the colored pixels corresponding to an organ or organ class in the picture. The computation thus discards multiple scattering and treats leaves as black bodies. VegeSTAR input files were obtained from the digitized data (i.e., spatial location and orientation), and leaf width and area were calculated from leaf length by means of the empirically determined allometric relationships given above. Leaf shape was idealized as a polygon obeying allometric relationships (Sinoquet et al. 1998). Each leaf was given a false color in order to identify it on the plant images. Time-averaged irradiance of each leaf was estimated assuming diffuse light conditions. The incident diffuse radiation was approximated by a set of 46 light sources as proposed by Den Dulk (1989). Each light direction was given a fraction of total incident radiation according to the standard overcast sky distribution (SOC, Moon and Spencer 1942). For each light direction, the silhouette-to-total-area ratio (STAR, Stenberg 1996) was calculated as the ratio of projected sunlit leaf area to total leaf area. Mean leaf irradiance was then estimated as the mean of the 46 directional STAR values weighted by the SOC distribution.

Leaf age was derived from direct observations made on the studied trees in March and December 2000, and in March 2001, during the flushing periods.

Results

Effects of leaf irradiance and leaf age on leaf nitrogen and total nonstructural carbohydrates

There was considerable variation in \( N_m \) among leaves (from 1.1 to 1.97%), but \( N_a \) was not correlated with the fraction of intercepted light or with leaf age (Figure 1A). Both \( N_a \) and \( T_a \) were linearly related to the fraction of intercepted light (Figures 1B and 1C). A common relationship was observed between \( N_a \) and \( T_a \) for the two 3-year-old trees studied. In contrast, the relationship between \( T_a \) and irradiance differed significantly between the two 11-year-old trees. At a given irradiance and over the whole range of irradiances, leaves of Tree 1 exhibited higher \( T_a \) than leaves of Tree 2. The relationship between \( N_a \) or \( T_a \) and irradiance was unaffected by leaf age; \( N_a \) was high in well-lit leaves older than 12 months.

Relationships between leaf photosynthetic capacity and leaf nitrogen

Unlike \( N_m \), \( N_a \) was proportional to \( M_a \), and a common relationship was observed for leaves of 3-year-old trees experiencing different local light regimes, and well-lit leaves far from and close to developing fruits of 11-year-old trees (Figures 2A and 2B). The lowest \( N_a \) and \( M_a \) values were observed in shade leaves sampled within the canopies of 3-year-old trees. Although \( T_a \) was proportional to \( M_a \) in leaves of both the 3-year-old trees at Saint-Paul and the 11-year-old trees at Saint-Pierre (Figure 2C), \( T_a \) (slope of the relationship) was significantly higher \((P < 0.01)\) in leaves of Tree 1 at Saint-Paul, as was the starch to sucrose ratio. Sucrose was the major nonstructural carbohydrate in mango leaves, and starch concentrations were less than 10% of sucrose concentrations (Table 2).

Nonlinear fits accounted slightly better for the relationship between the four key parameters defining leaf photosynthetic capacity (i.e., \( V_{\text{cmax}}, J_{\text{max}}, \text{TPU} \) and \( R_d \)) and \( N_a \) than linear fits, with the exception of \( R_d \) (Figure 3). Equation 9b was thus more effective than Equation 9a for describing the relationship between \( V_{\text{cmax}}, J_{\text{max}}, \text{TPU} \) and \( N_a \). At 30 °C, values of \( V_{\text{cmax}}, J_{\text{max}}, \text{TPU} \) and \( R_d \) ranged from 37.6 to 122.4 \( \mu \text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1} \), 62.8 to 205.0 \( \mu \text{mol electrons m}^{-2} \text{ s}^{-1} \), 3.9 to 14 \( \mu \text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1} \) and 1.5 to 3.7 \( \mu \text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1} \), respectively. The lowest values of \( V_{\text{cmax}}, J_{\text{max}}, \text{TPU} \) and \( R_d \) were found in leaves sampled deep in the crown of the 3-year-old trees. The ratios \( J_{\text{max}}/V_{\text{cmax}} \) and \( \text{TPU}/V_{\text{cmax}} \) remained constant (around 1.7 and 0.07, re-
spectively) over a large range of \( N_a \) values (Figure 4). In contrast, \( R_d/V_{\text{cmax}} \) decreased from 0.04 to 0.02 when \( N_a \) increased from 1.5 to 3.4 g m\(^{-2}\).

The leaf N allocation coefficients \( P_c \) and \( P_b \) increased by 50\% (from 0.1 to 0.19 g N in Rubisco per g total leaf N), and by 40\% (from 0.03 g to more than 0.04 g N in cytochrome f, ferredoxin, NADP reductase and coupling factors per g total leaf N), respectively, in leaves from 3-year-old trees when \( N_a \) increased from 1.4 to 1.8 g m\(^{-2}\) (Figure 5). But both coefficients remained relatively constant (around 0.19 g N in Rubisco per g total leaf N, and 0.04 g N in cytochrome f, ferredoxin, NADP reductase, and coupling factors, per g total leaf N, respectively) when \( N_a \) increased from 1.8 to 3.4 g m\(^{-2}\), for all types of leaves.

In 11-year-old trees, several leaf characteristics differed significantly between standard leaves and leaves close to developing fruits (Table 2). For similar gap fractions, leaves close to developing fruits exhibited higher values of \( M_a, N_a, V_{\text{cmax}}, J_{\text{max}}, \) and \( \text{TPU} \), and lower values of \( R_d/V_{\text{cmax}} \) than standard leaves. In contrast, \( N_m \) and \( T_a \) values did not differ between the two leaf types.

### Parameterization of the stomatal conductance model

The linear relationship between \( g_s \) simulated with the Leuning (1995) model and measured \( g_s \) was unaffected by the presence of fruit or tree age (Figure 6). This relationship also held for gas exchange measurements made over a large range of growing conditions, with the notable exception of the pre-flowering and flowering periods (data not shown).

### Testing of the coupled model

The stomatal conductance–photosynthesis model parameterized from measurements made on standard leaves and leaves
close to developing fruits, accurately predicts net photosynthesis of both standard leaves and leaves close to developing fruits, experiencing similar gap fractions (Figure 7).

**Discussion**

**Photoacclimation in mango leaves**

Leaf photosynthetic capacities of the 3-year-old mango trees exhibited strong spatial variations (e.g., from 37.6 to 122.4 µmol CO$_2$ m$^{-2}$ s$^{-1}$ for $V_{cmax}$). Parameters $V_{cmax}$, $J_{max}$ and TPU were strongly related to $N_a$ and nonlinear fits accounted slightly better for the relationships than linear fits. Linear relationships have been reported between photosynthetic capacities and $N_a$ in the chaparral shrub, *Lepechini a calycina* (Benth.) Epl. (Field 1983), *Eucalyptus grandis* W. Hill. (Leuning et al. 1991), cotton (Harley et al. 1992), *Nothofagus fusca* (Hook. F.) Orst. (Hollinger 1996), *Pinus radiata* D. Don (Walcroft et al. 1997), *Pinus pinaster* Ait. (Porté and Loustau 1998), *Juglans regia* L. (Le Roux et al. 1999a) and *Prunus*...
persica (L.) Batsch. (Le Roux et al. 2001a). The nonlinearity of the relationships observed for mango was weak, and should be tested with additional data. The dependency of $R_d$ on $N_a$ was consistent with the relationships generally observed between leaf photosynthetic capacity and $R_d$ reported by Ceulemans and Saugier (1991) and Reich et al. (1998). Our observations thus support the idea that all parameters related to the major biochemical limitations to photosynthesis vary in parallel, because leaf N has a major role in controlling each process involved, and because allocation of N is regulated in such a way that these limitations remain balanced as total N varies (Chen et al. 1993). However, a substantial increase in $R_d/V_{\text{max}}$ with increasing $N_a$ was observed (Figure 4), which contradicts the assumption that leaf $R_d$ is proportional to $V_{\text{max}}$ (Farquhar et al. 1980, Niinemets and Tenhunen 1997). Further data are needed to assess whether the observed increase in $R_d/V_{\text{max}}$ with increasing $N_a$ was associated with measurement errors or indicates actual changes in the balance between $R_d$ and photosynthetic capacity.

Photosynthetic light acclimation of leaves generally results from changes in $N_{\text{in}}, M_s$ and changes in partitioning of total leaf nitrogen among the different pools of the photosynthetic machinery (Evans 1989b). When mean leaf irradiance increased from 0.02 to 0.90 of incident irradiance, $M_s$ increased by about 90\% (from 92 to 173 gDM m$^{-2}$), and $N_{\text{in}}$ increased by 20\% (from 1.39 to 1.67\%), resulting in a more than doubling of $N_a$ (from 1.32 to 2.78 g m$^{-2}$). Concurrently, the relative amount of leaf N allocated to carboxylation ($P_c$) and electron carriers ($P_b$) increased by 50 and 40\%, respectively. However, changes in total leaf N allocation seemed to play a more important role in light acclimation at low $N_a$ (Figure 5), because...
increases in \( N_a \) values above 1.8 g m\(^{-2} \) had no significant effect on photosynthetic light acclimation. In mango, photosynthetic light acclimation thus results mainly from changes in \( M_b \) and, to a lesser extent, from changes in allocation of total leaf N at low irradiance, whereas changes in \( N_{in} \) play only a minor role. Changes in \( M_b \) are also of major importance for photoacclimation in other tree and shrub species (Gulmon and Chu 1981, Niinemets et al. 1998, Le Roux et al. 1999b, Rosati et al. 1999). In contrast, although adjustment of leaf N partitioning among different pools of the photosynthetic machinery is expected from optimization theories (i.e., for optimizing daily carbon gain of individual leaves in different light regimes: Hikosaka and Terashima 1995), such an adjustment is of minor importance for species like Alocasia macrorrhiza (L.) G. Don (Seemann et al. 1987) and Juglans regia (Le Roux et al. 1999a, 1999b), but significant for species like Phaseolus vulgaris L., Cucumis sativus L. (Seemann et al. 1987) and Prunus persica (Le Roux et al. 2001).

Factors affecting the relationship between photosynthetic capacity and light regime

Effect of age Leaf characteristics such as photosynthetic capacity and \( N_a \) generally exhibit strong patterns with leaf age, with maximum values observed when leaves have just completed full expansion (Constable and Rawson 1980, Marshall and Biscoe 1980, Dwyer and Stewart 1986, Field 1987, Wilson et al. 2000, Frak et al. 2001). However, we observed that the relationship between \( N_a \) and irradiance was unaffected by leaf age in mango and that \( N_a \) values remained high in old leaves experiencing high irradiance. This suggests that changes in \( N_a \) were influenced by irradiance rather than age during the first year of growth in mango. Furthermore, the tight correlation observed between photosynthetic capacity and \( N_a \) for leaves of different age classes is consistent with previous studies reporting that changes in both variables are closely correlated during leaf aging (Field and Mooney 1983, Vos and Oyarzun 1987). Such observations suggest that aging represents a period of redistribution of N resources and not uncontrolled deterioration of leaf biochemical characteristics (Field and Mooney 1983).

Effect of the local source–sink balance We observed that photosynthetic capacity was substantially higher in leaves close to developing fruits than in control leaves experiencing the same light regime. Source–sink imbalances can exert feedback inhibition on leaf photosynthesis through carbohydrate accumulation in leaves (Foyer, 1988). For instance, transient accumulations of carbohydrates in leaves, which have been observed during the diurnal period, may impair the rate of electron transport (Pammenter et al. 1993). Changes in photosynthetic capacity, not just assimilation rates, are more likely to be observed in association with lasting source–sink imbalances. One hypothesis is that high concentrations of carbohydrates repress the expression of genes coding for several photosynthetic enzymes (Krapp and Stitt 1995, Koch 1996, Drake et al. 1997). Alternatively, carbohydrates may interact with hormonal signals to control gene expression (Thomas and Rodriguez 1994). There is also some evidence that photosynthetic capacity is related to leaf carbohydrate status through the effect of the latter on phosphate availability (Riesmeier et al. 1993, Sun et al. 1999). However, we found increased photosynthetic capacity in leaves close to developing fruits compared with control leaves, whereas no significant difference in \( T_s \) was observed, suggesting that \( T_s \) was probably not the signal promoting photosynthetic capacity in leaves close to developing fruits. The common relationship observed between \( N_a \) (i.e., photosynthetic capacity) and irradiance for the two trees studied, and the different relationships observed between \( T_s \) and irradiance for these trees, provide further evidence that \( T_s \) was not a major factor driving photosynthetic acclimation in mango. This conclusion is inconsistent with the assumption of recent models of photosynthetic acclimation (Dewar et al. 1998, Kull and Kruijt 1999).

Differences in photosynthetic capacity between standard leaves and leaves close to developing fruits were mainly attributable to differences in \( M_b \) rather than in \( N_{in} \). A fruit effect on photosynthetic capacity was not expected because there were no differences in mean leaf irradiance and leaf age (the usual driving forces behind photosynthetic capacity) between the two leaf types. We have no explanation for the observed fruit effect.

Potential and limits of the stomatal conductance–photosynthesis model

The existence of an almost unique relationship between photosynthetic capacity and \( N_{in} \), whatever the origin of leaves (leaves selected from 3-year-old trees, standard leaves and leaves close to developing fruits of 11-year-old trees) suggests that this relationship is robust. Similarly, the linear relationship between \( g_s \) and \( A_{net} \) also seems to be robust, at least in the range of leaf temperatures of our gas exchange measurements (from 25 to 35 °C) and in the absence of water stress. Because changes in stomatal conductance may not parallel those of assimilation rate in response to temperature (Bunce 2000), the relationship between \( g_s \) and \( A_{net} \) needs to be reassessed at temperatures lower than 25 °C and higher than 35 °C, to extend the predictive abilities of our model. Although the effects of soil drought on stomatal conductance have been demonstrated in many field and laboratory experiments (Baldocchi 1997), the two most common models of \( g_s \), the Ball-Berry model (Ball et al. 1987) and the Leuning (1995) model, do not specifically incorporate a water stress function. This is a clear shortcoming of our model of leaf photosynthesis because it relies on the original Leuning model of \( g_s \). However, the robustness of the relationships reported here augurs well for the ability of our model to predict photosynthesis of mango leaves from well-irrigated trees of different ages and phenological stages.

Another shortcoming of our model is that most parameters were estimated from measurements made at 30 °C. Previous studies have shown the importance of the temperature response of leaf photosynthetic capacity, and the existence of interspecific differences in responses obtained from different species or under different environmental conditions (Dreyer et al. 2001, Medlyn et al. 2002). The temperature dependency of
the parameters of photosynthetic capacity will thus have to be studied for mango to improve the predictive ability of the model.

Conclusion
Our data suggest that there is a fruit effect on photosynthetic capacity of nearby leaves that does not seem to be driven by $T_e$. Modeling the distribution of photosynthetic carbon gains within fruit trees will have to take such an effect into account in the future. Because the distribution of carbon sources has potentially important consequences on the distribution of fruit quality in the tree, the mango leaf model will also need to be scaled up to the fruiting branch and tree canopy levels, for instance by using the RATP model (Sinoquet et al. 2001). Scaling up the model could help assess effects on carbon acquisition at the fruiting unit level of: (1) plant and orchard geometry, namely the spatial distribution of fruiting units in the canopy; (2) environmental variables; and (3) leaf physiological properties.

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References


