# Simulation of photon transport in a three-dimensional leaf: implications for photosynthesis

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## ABSTRACT

**A model to evaluate photon transport within leaves and the implications for photosynthesis are investigated. A ray tracing model, Raytran, was used to produce absorption profiles within a virtual dorsiventral plant leaf oriented in two positions (horizontal/vertical) and illuminated on one of its two faces (adaxial/abaxial). Together with chlorophyll profiles, these absorption profiles feed a simple photosynthesis model that calculates the gross photosynthetic rate as a function of the incident irradiance. The differences observed between the four conditions are consistent with the literature: horizontal-adaxial leaves, which are commonly found in natural conditions, have the greatest light use efficiency. The absorption profile obtained with horizontal-abaxial leaves lies below this, but above those obtained for vertical leaves. The latter present similar gross photosynthetic rates when irradiated on either the adaxial or abaxial surfaces. Vertical profiles of photosynthetic rates across the leaf confirm that carbon fixation occurs mainly in the palisade parenchyma, that the leaf anatomy is integral to its function and that leaves cannot be considered as a single homogeneous unit. Finally, the relationships between leaf structure, orientation and photosynthesis are discussed.**

*Key-words*: Leaf anatomy; leaf chlorophyll profile; leaf function; leaf light absorption profile; photosynthetically active radiation; ray tracing model.

## INTRODUCTION

It has long been observed that solar radiation regulates the growth and development of plant leaves. The variation in internal leaf structure with environment has fascinated biologists for more than a century (Haberlandt 1884), as has the structure of plant canopies (Schimper 1903; Warming 1909). Natural selection has led to the proliferation of a variety of internal leaf structures that clearly show convergence in ecosystems sharing common climate constraints. Plant leaves are the primary photosynthesizing organs, crucial to biospheric and atmospheric functioning, yet the mechanisms of how leaf structure and orientation interact with the light environment remains incompletely under-

stood. Feedbacks between the light environment during leaf development produce many anatomical and biochemical characteristics that affect subsequent physiological functioning in leaves. One of the most-cited examples is the difference between sun and shade leaves and the concurrent biochemical differences that minimize the potential for photoinhibition in sunlight and maximize light capture in the shade.

On the canopy scale, leaf orientation and stem architecture determine the potential for light interception and penetration of light into the lower canopy. Leaf angles vary from horizontal (planophile) to vertical (erectophile), and many woody species are reported to modulate leaf angle display through plastic environmental responses (King 1997; Smith *et al.* 1997). The significance of leaf orientation for regulating light intensity and physiological responses has also been observed many times (e.g. Ehleringer & Werk 1986; Smith *et al.* 1997; King 1997; Valladares & Pearcy 1998). Pendent leaves reduce the absorption of direct beam midday light intensities and increase absorption of light at low sun angles and the proportion of diffuse light relative to horizontally oriented leaves. Vertical orientation also increases the proportion of light incident on the abaxial side. Correlated with changes in leaf angles are structural alterations that parallel changes observed between sun and shade leaves, for example leaf thickness and size, stomatal density, leaf veination, cell sizes and air spaces and biochemistry [notably Rubisco (ribulose 1·5-bisphosphate carboxylase/oxygenase), chlorophyll *a* and *b*, and accessory pigments; Field *et al.* 1992]. Among other changes, leaf anatomy in vertically oriented sun leaves becomes unifacial or isobilateral, morphology more common in shade leaves, while horizontally oriented sun leaves are strongly bifacial. These and other studies suggest that leaf structure and orientation are integral to the adaptive functioning of leaves in light harvesting and carbon fixation.

Despite the extensive literature relating ecophysiological characteristics to leaf form and function, most studies have not gone beyond qualitatively identifying associated traits to examine the mechanistic bases for the observed patterns between internal leaf structure, leaf orientation and physiological function. This is partly because there have been few studies explicitly relating photon interactions to leaf structure, leaf orientation and photosynthetic potential (although many authors have dealt with various parts of this problem, e.g. Mooney 1972; Mooney *et al.* 1982;

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Parkhurst 1986; Ehleringer & Werk 1986; Myers, Jordan & Vogelmann 1997; Smith, Bell & Shepherd 1998; Sims & Pearcy 1994). In contrast to the extensive literature on plant biochemistry and metabolism, quantitative ultrastructural studies of plant cells or tissues have been largely ignored in recent decades. As a result, the biosynthesis, structure and function of many molecules in the cell are now well known, while the macroscopic description of leaf cells and tissues remains undescribed in any quantitative way. Nonetheless, it is accepted that inside leaves, complex phenomena related to biochemical composition and anatomical features cause light attenuation, particularly in the visible spectrum for photosynthetically active radiation (PAR). In contrast, the epidermis mainly determines the bidirectional reflectance.

Most past work on light interactions and anatomy investigated individual cell types, primarily epidermal cells. Haberlandt (1914) and later Gabrys-Mizera (1976) hypothesized that convex epidermal cell surfaces could focus light at the level of chloroplasts in the shaded understorey of tropical forests; this was later demonstrated in a computer simulation by Bone, Lee & Norman (1985). Lee (1986) estimated that light-focusing might produce a 10-fold increase in incident photon flux density on the tropical forest floor. Martin *et al.* (1989) then showed that the epidermal lenses increased light absorption at low sun angles in a high light-adapted cultivar. More recently, Vogelmann and colleagues have examined experimentally light gradients in leaves (Vogelmann *et al.* 1988, 1989, 1991), the relationship to chlorophyll in sun and shade leaves (Cui, Smith & Vogelmann 1991) and shown that columnar palisade parenchyma cells were more efficient than spongy mesophyll cells in channelling collimated light to chloroplasts within the leaf (Vogelmann, Nishio & Smith 1996). The connections between light gradients and photosynthesis have been investigated by Jeje & Zimmermann (1983), Nishio, Sun & Vogelmann (1993) and Sun, Nishio & Vogelmann (1996, 1998), who measured carbon fixation gradients within plant leaves; green radiation, which penetrates deeper in the leaf than blue and red radiation, also moves the location of maximum fixation deeper into the profile. Allen, Gausman & Richardson (1973), building on the work of Allen *et al.* (1969), were the first to model internal leaf structure as intercellular space air and cell walls, characterized by the index of refraction to test the specular and the diffuse reflectance at cell walls within the leaf. Shortly afterwards, Kumar & Silva (1973) showed that actual reflectance and transmittance could be reproduced more accurately by adding representations for cytoplasm and chloroplasts in the model, thereby increasing the internal diffusion and lowering surface reflectance. Yamada & Fujimura (1991) modelled the leaf as a stack of layers, each with specific absorption and scattering coefficients. Parkhurst (1986) demonstrated that the three-dimensional structure of leaves is critical to their gas exchange functions (e.g. for the diffusion of $CO_2$, $H_2O$ and $O_2$) and to light-scattering, based on results from a two-dimensional computer simulation.

More recent advances in solid geometry, radiative transfer modelling and understanding of leaf physiology now make it possible to develop more detailed computer-based simulation models to study the interaction of light within plant leaves. Among possible approaches to modelling leaf function, only ray-tracing models such as Raytran (Govaerts & Verstraete 1998) can account for the complexity of leaf internal structure as it appears in a photomicrograph. While computer-generated synthetic images seem realistic, visualization algorithms are unsuited to extrapolate biophysical and optical properties. Nonetheless, the simulation of photon transport within a leaf with such a technique requires a detailed description of individual cells, their unique arrangement inside leaf tissues, their chemical constituents and their respective physiology. The optical constants of leaf constituents (cell walls, cytoplasm, pigments, air cavities, etc.) must be known for each cell type. Then, using the laws of reflection, refraction and absorption, it is possible to simulate the propagation of individual photons incident on the leaf surface. Once a sufficient number of rays have been simulated, statistical estimates of the radiation transfer within and through a leaf may be produced. Raytran has been applied to simulate the optical properties (spectral and bidirectional reflectance, transmittance and absorption) of mesophytic dicotyledon leaves (Govaerts *et al.* 1996; Jacquemoud *et al.* 1997), which are typical of plants grown in a moderately high-light environment.

In this paper, we demonstrate the application of the Raytran model to examine how variation in leaf angles interacts with leaf anatomy and biochemistry to affect photon gradients and the rate of photosynthesis. The virtual bifacial leaf and the Raytran model are explained in the next section. Four extreme situations have been considered: two leaf-blade orientations (horizontal and vertical) by two leaf surfaces (adaxial and abaxial). They correspond to well-documented experiments in the literature. The second section analyses the model outputs, i.e. the absorption profiles in the leaf that become, in turn, inputs to a photosynthesis model to calculate the response of gross photosynthetic rate to irradiance. Profiles of this rate are presented at the end of the Results section and are compared with carbon fixation gradients measured experimentally.

## MATERIALS AND METHODS

### Construction of the leaf

The construction of an anatomically realistic leaf was the first challenge to resolve. In two dimensions, the domain of a leaf cross-section has been represented by circular arcs separating media of different refraction indices to simulate its optical properties (Allen *et al.* 1973), or by a polygonal structure with intercellular spaces to simulate gas exchange (Pachepsky & Acock 1996). The change from two to three dimensions required an effort of conceptualization that was missing. The use of the rhombic dodecahedron (10
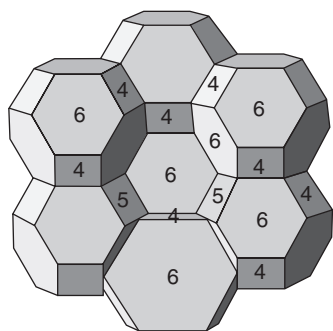
**Figure 1.** A group of orthic tetrakaidecahedra (after Lewis 1923).

parallelogram faces) or the tetrakaidekahedron (8 hexagonal faces and 6 square faces), which conveniently fill space when assembled (Fig. 1), were initially proposed in the first half of the last century (Lewis 1923; Macior & Matzke 1951). Such hexagonal and cubic cells also provide a good representation of the leaf epidermis and can be used to understand and simulate its growth.

However, while some leaf tissues are simple and homogeneous, others are differentiated and cells are not compact and anisotropic. Working on the assumption that polyhedra with many faces can assimilate to sphere-like volumes, Govaerts *et al.* (1996) considered cells to be primitive objects (spheres, ellipsoids, cylinders etc.), which are carved out and assembled using constructive solid geometry techniques to define more complex objects. A typical mesophyll cell is then defined as an object of variable size filled with three concentrically oriented media: cell wall (containing cellulose + hemicellulose + lignin) as the outer layer and cytoplasm with chloroplasts (containing chlorophyll) and tonoplast (containing water) as the inner layer. Since most epidermal cells do not contain chloroplasts, they are defined by only two media. The cells are characterized by size, shape, wall thickness, chloroplast membrane thickness and chemistry. The media composing each cell are homogeneous, i.e. the physical and biochemical properties are uniform and isotropic, but between cells these amounts vary stochastically within the observed ranges reported in the literature. The leaf scattering and absorbing properties are wavelength-dependent and are characterized by the refractive index ($n$) and by the specific absorption coefficient ($k$) of the three main leaf materials (cell wall, chlorophyll and water). Since the optical properties of the other leaf components (such as the endoplasmic reticulum, the nucleus, the mitochondria and plastids, among others) are still largely unknown but transparent in the visible spectrum, they were not considered. Similarly, secondary leaf photosynthetic pigments (carotenoids, xanthophyls, etc.), which have little effect on the optical properties of green leaves, have been ignored.

The geometric construction of the leaf consisted of connecting cells to produce three tissues (epidermis, palisade parenchyma and spongy mesophyll) and assembling tissues to represent the flattened lamina. This representation of the

internal structure was based on common observations and descriptions derived from numerous literature sources (Gibson & Ashby 1988; Martin *et al.* 1989; Poulson & Vogelmann 1990; Niklas 1992; Romberger, Hejnowicz & Hill 1993; Richter & Fukshansky 1994; among others). Figure 2 illustrates the dicotyledon leaf model used with Raytran in this paper: it shows the palisade parenchyma and spongy mesophyll arranged between two layers of epidermal cells. Cells have different sizes and shapes, in contrast to the leaf presented in Govaerts *et al.* (1996). The upper epidermis is 17·5 $\mu$m thick and irregular, while the lower is 25 $\mu$m thick and regular. Other details on the geometric construction of the leaf tissues, their structural and optical properties are found in Govaerts *et al.* (1996).

## The Raytran model

With Raytran, photons are generated from the light source in the forward direction. Ten million rays are generated for each experimental test. After the first interaction with the leaf surface, they are tracked from collision to collision throughout the blade until the ray is either absorbed or escapes from the leaf. The Fresnel formulae are used to determine the direction of propagation when a ray interacts at the interface between media with different refractive indices and Beer's law is followed to determine the probability of whether a ray is absorbed by a medium. The vertical fluxes in the virtual leaf were recorded by 20 horizontal detectors, positioned uniformly along the vertical axis. Each time a ray contacts the upper surface of the sensor, the downward flux counter is incremented; the reverse happens when a ray reaches the lower surface. Rays are divided by the total number of emitted rays to provide relative energy fluxes. For each sensor, the relative net flux is calculated as the difference between the downward and upward fluxes. Finally, to simulate a horizontally infinite leaf, the sampling area is assumed to be surrounded by an equivalent target. When a ray escapes from the upper or lower side of the leaf, it contributes to the estimation of the bidirectional reflectance or transmittance factors, respectively.
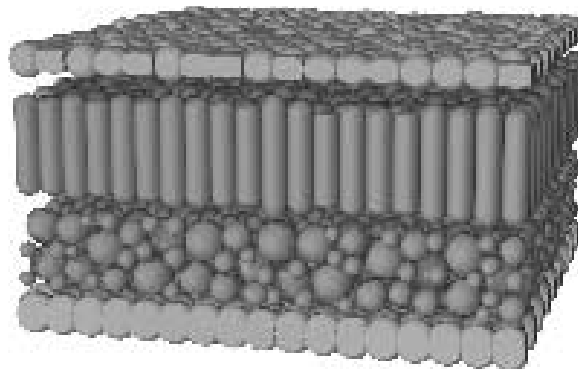


**Figure 2.** Perspective view of a virtual bifacial dicotyledon leaf. The cross-section of the target leaf is $300 \times 300 \times 170$ $\mu$m.
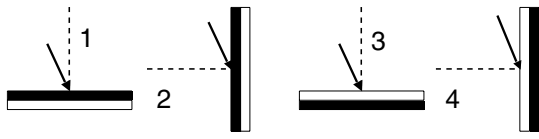
**Figure 3.** Four orientations selected to simulate the absorption profiles of a dorsiventral leaf. The black (filled) box represents the adaxial palisade parenchyma, and the white (open) box the abaxial spongy mesophyll.

## RESULTS

### Effect of leaf orientation on photon flux

To investigate how bifacial leaf anatomy and biochemistry affect light propagation in different orientations, we used the mesophytic dicotyledonous leaf model with Raytran. Simulations were based on differences in leaf angle orientation for vertically and horizontally oriented leaves at 90° and 0°, respectively. Four absorption profiles (corresponding to four orientations) were simulated with a collimated illumination zenith angle of 25° (Fig. 3):

1 horizontal leaf with light incident on palisade parenchyma (adaxial surface);
2 vertical leaf with light incident on palisade parenchyma;
3 horizontal leaf with light incident on spongy mesophyll (abaxial surface), and
4 vertical leaf with light incident on spongy mesophyll.

The illumination configuration for a vertically oriented dorsiventral leaf (as illustrated in profiles 2 and 4) is common, while the leaf orientation illustrated in case 3 only occurs on windy days or in early morning and late afternoon, when the sun is at low angles. Light gradients simulated at 675 nm perpendicular to the leaf surface are shown in Figs 4 and 5. The predicted distributions of light are in close agreement with fibre-optic probe experimental results (Vogelmann *et al.* 1989), including small variations in net flux and

upwelling fluxes at the transition between palisade and spongy mesophyll in the profiles. Collimated incident light becomes completely diffuse before exiting the palisade parenchyma. The relative downward flux through the epidermis (when the palisade parenchyma or the spongy mesophyll are oriented toward the surface for incident photons) can exceed 100% of the incident flux at the epidermis because the same ray may be scattered several times inside an epidermal cell and be counted more than once by a detector. Our results are consistent with microprobe measurements reported by Vogelmann & Björn (1984) and Vogelmann *et al.* (1989, 1991) – that light fluxes within leaves exceeded by three to four times the incident light in the near infrared spectral region – and demonstrate how cell distribution (palisade parenchyma versus spongy mesophyll) affects the distribution of upwelling and downwelling photons in photosynthetically active radiation wavelengths.

Since reflectance + transmittance + absorptance = 1, and since there is almost no transmittance at 675 nm, the total absorptance is the fraction of light not reflected by the leaf, i.e. 0·862, 0·618, 0·900 and 0·706 for profiles 1, 2, 3 and 4, respectively. Net flux profiles vary strongly with leaf orientation and incident light zenith angle. Comparison of transmission through horizontal (Figs 4a & 5a) and vertical (Figs 4b & 5b) leaves shows that the light is more efficiently absorbed in the former case than in the latter. Rotating the leaf from horizontal to vertical amounts to increasing the illumination zenith angle and then the fraction of specular reflectance, as shown experimentally (Brakke 1994) or with simulations (Govaerts *et al.* 1996). In consequence, the effectiveness of light absorption is lower for vertically oriented leaves. With few exceptions, vertical leaves are actually symmetrical, with palisade cells beneath the adaxial and abaxial epidermis (Poulson & DeLucia 1993).

The attenuation of light occurs rapidly when the palisade parenchyma is illuminated. Nearly 100% of the rays are absorbed in this tissue, i.e. within the initial 90 $\mu$m of the adaxial leaf surface. These results are consistent with obser-
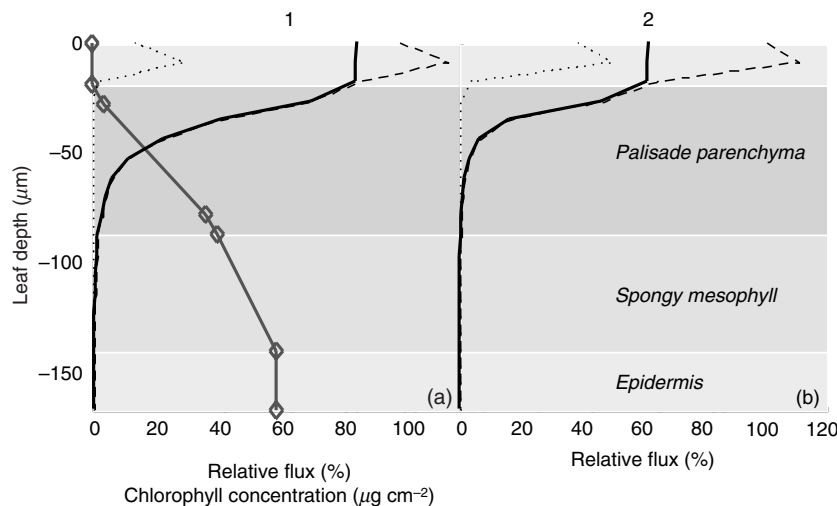


**Figure 4.** Normal distribution of light at 675 nm inside a vertical bifacial dicotyledon leaf for illumination zenith angles of (a) 25° (case 1) and (b) 65° (case 2) with light incident on the palisade mesophyll (net flux, solid line; downward flux, dashed line; upward flux, dotted line). —◇— indicates the cumulative chlorophyll concentration, expressed in $\mu$g cm$^{-2}$.
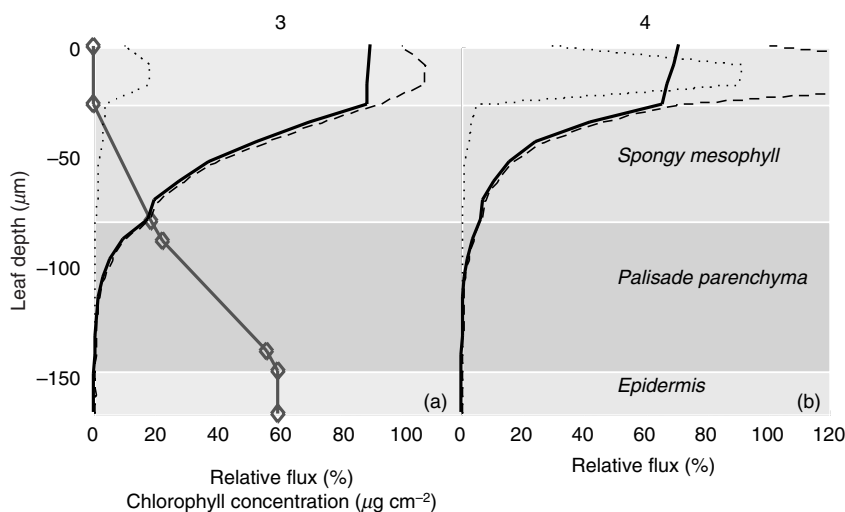
**Figure 5.** Normal distribution of light at 675 nm inside a vertical bifacial dicotyledon leaf for sun zenith angles of (a) 25° (case 3; $\theta = 155°$) and (b) 65° (case 4; $\theta = 115°$) with light incident on the spongy mesophyll (net flux, solid line; downward flux, dashed line; upward flux, dotted line). —◇— indicates the cumulative chlorophyll concentration, expressed in $\mu g\ cm^{-2}$.

vations that palisade cells aid in redistributing collimated light within the leaf for maximum efficiency of photosynthesis. Because the 45% air space in spongy mesophyll greatly increases the photon pathlength, this tissue doesn't attenuate light as fast as the palisade parenchyma when illuminated, but the total absorptance is higher for profile 3 than for 1, and is higher for 4 than for 2. This confirms some experimental observations by DeLucia *et al.* (1991). Furthermore, the absorption profiles of Figs 4 and 5 have been used as inputs of a simple photosynthesis model.

## Effect of leaf orientation on photosynthesis

The photosynthetic response to light absorption of dorsiventral leaves put in contrasting positions has been mea-

sured experimentally to study the effect of irradiating adaxial versus abaxial surfaces (Syvertsen & Cunningham 1979; Terashima 1986) and horizontal versus vertical surfaces (Yates 1981; DeLucia *et al.* 1991). Among the equations proposed in the literature, the non-quadratic hyperbola used by Prioul & Chartier (1977) has been chosen to simulate this response. This equation is generally used at the scale of an entire leaf. Terashima & Saeki (1985) applied it to each section of a *Camellia* leaf segmented into 10 vertical layers; the whole leaf photosynthesis (P) was expressed as the sum of the gross photosynthetic rate of each layer (*p*). A similar approach has been used by Fukshanski, Martinez & Remisowsky (1992) on *Catalpa* and *Magnolia* leaves with another photosynthesis model. For layer *j*, the form of the non-quadratic hyperbola model is:
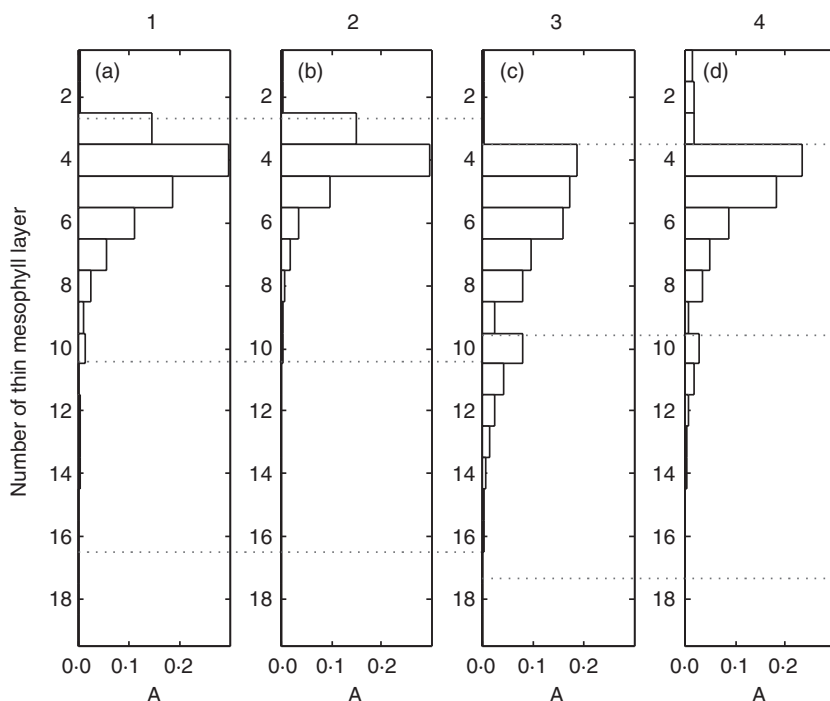


**Figure 6.** Gradients in light absorption ($A_j$) within the leaf when illuminated at 675 nm (a,b) on the adaxial surface (1 and 2) and (c,d) on the abaxial surface (3 and 4). The dashed lines represent the boundaries between the leaf tissues.

$$\theta_j^2 p_j^2 - (\phi_j A_j + p_{\max,j})p_j + \phi_j A_j p_{\max,j} = 0, \qquad (1)$$

where $p_j$ is the gross photosynthetic rate ($\mu$mol $CO_2$ m$^{-2}$ s$^{-1}$), $\theta_j$ is the convexity index ($0 \leq \theta_j \leq 1$), $\phi_j$ is the quantum yield for $CO_2$ fixation ($\mu$mol $CO_2$ $\mu$mol$^{-1}$ photons), $A_j$ is the amount of absorbed light ($\mu$mol photons m$^{-2}$ s$^{-1}$) and $p_{\max,j}$ is the gross photosynthetic rate at light saturation ($\mu$mol $CO_2$ m$^{-2}$ s$^{-1}$). When expressed in $\mu$mol $CO_2$ mg$^{-1}$ chlorophyll s$^{-1}$, the latter must be multiplied by the chlorophyll content $[Chl]_j$ (mg chlorophyll m$^{-2}$). $p_j$ is then the smallest of the two solutions of Eqn 1. To run the photosynthesis model, the gradients within the leaf of the terms described above are required. The leaf is segmented in 19 layers, each having the same thickness: two layers for the upper epidermis, eight for the palisade mesophyll, six layers for the spongy mesophyll and three for the lower epidermis. Figure 6 shows the gradient in absorption ($A_j$) and Fig. 7 the gradient in chlorophyll ($Chl_j$) deduced from Figs 4 and 5.

As shown earlier, the total absorptance equals 0·862, 0·618, 0·900 and 0·706 for profiles 1, 2, 3 and 4, respectively. A decrease in the incidence angle from 65° to 25° decreases absorptance by 24·4% when the adaxial surface is illuminated and by 19·4% when the abaxial surface is illuminated. In the latter case, the pattern of light absorption is slightly inverted at the transition from spongy mesophyll to palisade parenchyma, probably because of the higher absorption coefficient in this tissue.
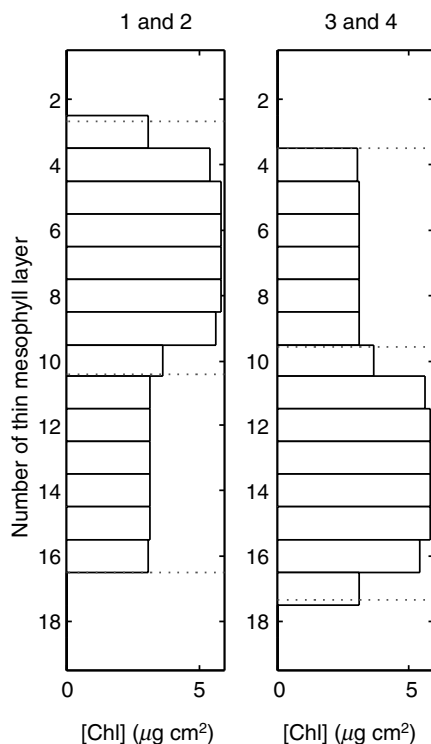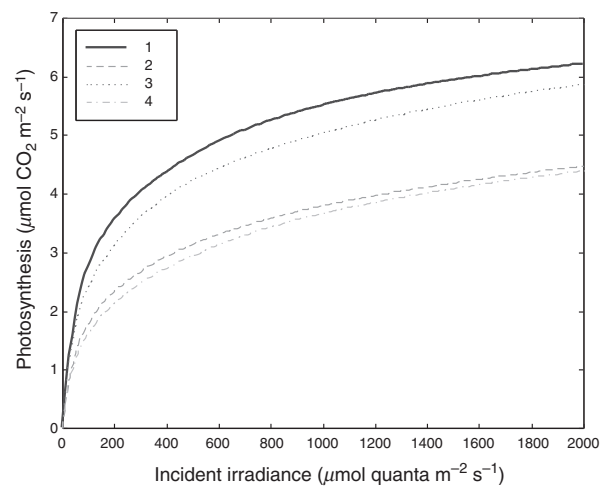


**Figure 8.** The response of gross photosynthetic rate to irradiance incident on the adaxial (1 and 2) and abaxial (3 and 4) leaf surfaces, oriented horizontally (1 and 3) and vertically (2 and 4).



**Figure 7.** Gradients in chlorophyll concentration $[Chl]_j$ within the leaf. The dashed lines represent the boundaries between the leaf tissues.

Some assumptions are necessary to calculate the gross photosynthetic rates of the layers. For instance, although $\phi_j$ and $p_{\max,j}$ depend on many physiological parameters, such as the internal temperature and the $CO_2$ and $O_2$ concentrations at the intercellular spaces, we model them as constants according to Terashima & Saeki (1985), where $\phi_j = \phi = 0\cdot05$ $\mu$mol $CO_2$ $\mu$mol$^{-1}$ photons for all the layers, $p_{\max,j} = p_{\max 1} = 0\cdot0125$ $\mu$mol $CO_2$ mg$^{-1}$ chlorophyll s$^{-1}$ for the palisade parenchyma and $p_{\max,j} = p_{\text{ma}}^{\text{x2}} = 0\cdot01$ $\mu$mol $CO_2$ mg$^{-1}$ chlorophyll s$^{-1}$ for the spongy mesophyll. This difference accounts for the higher electron transport activity in the first tissue (Terashima & Inoue 1984). The convexity index $\theta_j$ was set to $\theta = \theta_j = 0\cdot95$. Note that the precision of these values is not very important for this comparative study, since the virtual leaf in Fig. 2 is not a real case but a statistical representation of a typical dorsiventral leaf.

As expected, the horizontal adaxial leaf (1) in Fig. 8 shows the highest gross photosynthetic rate. This result is consistent with experimental measures performed on dorsiventral leaves (Terashima 1986; DeLucia *et al.* 1991). Considering that vertical leaves receive 47% less total incident irradiance than horizontal leaves for purely geometrical reasons (Yates 1981), and that their orientation increases their total reflectance (decreases absorptance) by 20–25% as seen earlier, their gross photosynthesis should be reduced: this is confirmed by the results shown in Fig. 9. Once again, incident light on the adaxial surface seems to be advantageous, although the difference is small in this case.

The profiles of gross photosynthetic rate (shown in Fig. 9), related to carbon fixation within leaves, do not follow within-leaf light gradients closely, as pointed out by Jeje & Zimmermann (1983), Nishio *et al.* (1993) and Sun *et al.* (1996, 1998). For adaxial illumination (profiles 1 and 2), one can see a maximum in the middle of the palisade parenchyma that is in agreement with experiments. Since no
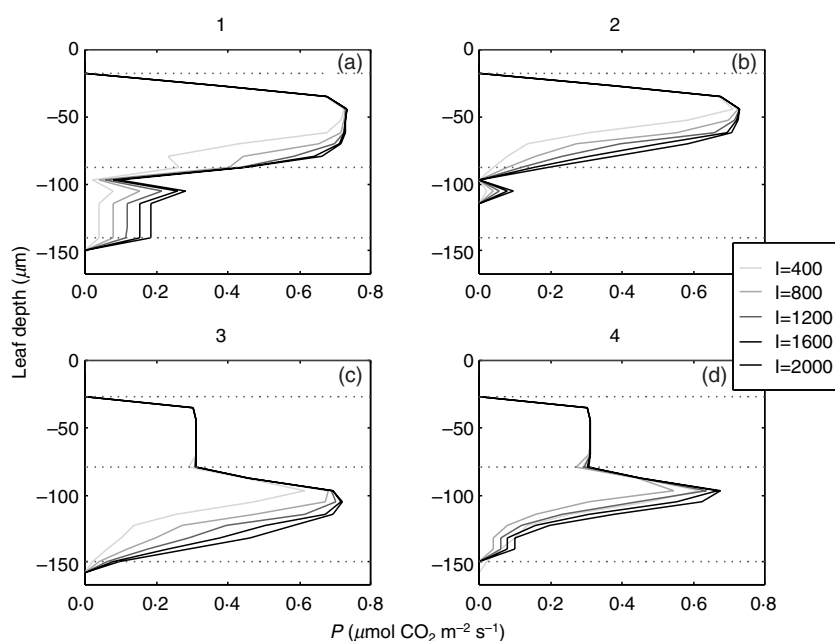
**Figure 9.** Profiles of gross photosynthetic rate within the leaf for different incident irradiation intensities (a,b) on the adaxial surface (1 and 2) and (c,d) on the abaxial surface (3 and 4). The dashed lines represent the boundaries between the leaf tissues.

hypotheses have been made on the Rubisco distribution, our simulations show that the latter is not the only factor controlling carbon fixation. Moreover, an increase in light irradiance from 400 to 2000 $\mu$mol photons m$^{-2}$ s$^{-1}$ leads to a saturation in the first half of this tissue, while abaxial illumination induces a saturation in the entire spongy mesophyll. This means that orientations 1 and 2 are better adapted to increasing light intensities than 3 and 4. Figure 9a also shows that $CO_2$ fixation is very low in the spongy mesophyll (almost nil for vertical leaves; Fig. 9b) when the adaxial surface is illuminated. However, the palisade parenchyma is still important for light absorption when the abaxial surface is illuminated. This pattern is consistent with the results of physiological models (Parkhurst 1986; Pachepsky & Acock 1998); these authors found significantly higher photosynthetic rates in simulated leaves containing only spongy mesophyll cells than in leaves containing only palisade parenchyma.

## DISCUSSION

Understanding the radiative environment at the leaf level is of great importance to the scaling of observable electromagnetic features if we want to scale ecophysiological processes to the plant canopy or larger. Leaves are complex structures that regulate the uptake of light at multiple levels, from the biophysical construction to the orientation of leaves. Significant advances in modelling the radiative transfer in virtual leaves allows us to test hypotheses and quantify how cellular structure, orientation and chemistry control physiological processes and how these are altered by leaf orientation. The intrinsic optical properties of chlorophyll and pigment–protein complexes are still poorly known and our assumption of a homogeneous distribution

inside the pigment membrane is clearly inappropriate for producing more accurate simulations of photon flux and physiological processes in leaves. Chloroplast function depends on the orientation and number of grana stacks, and varies with the distribution and composition of pigment complexes in the light-harvesting antenna. Such information on ultrastructure of leaves is presently unavailable for the construction of more accurate leaf representations or for comparison against real leaves.

This paper is a first attempt to simulate the photosynthetic response of a virtual leaf entirely described as a three-dimensional object. In spite of many approximations in the construction of the leaf and in the carbon fixation properties, the absorption profiles simulated by Raytran and the ensuing patterns of photosynthetic activity are remarkably consistent with data in the literature. These models open new opportunities to test the relationships between leaf structure and function and should provide a powerful tool for studying the effect of leaf anatomy and leaf orientation on leaf and plant photosynthesis. Note that simple models, based on the Kubelka–Munk theory for instance, have a lower level of complexity and therefore flexibility, limiting their application to real leaves. The most recent experimental results showing the influence of these two variables upon leaf functioning is confirmed in these simulations. Models have the advantage of testing and validating various hypotheses at a low cost and within a limited time: for example, testing the effect of adding another layer of palisade cells, increasing the chlorophyll concentration or increasing cell wall thickness on carbon fixation. However, improved realism in the results requires additional work, because detailed anatomical and biochemical data to drive the model are largely unavailable. In particular, the description of the leaf internal structure must be improved. and clearly adding factors to the model related to $CO_2$ dif-

fusion, concentration of Rubisco (Sun, Nishio & Vogelmann 1996, 1998) and other biochemical resistances in the cytoplasm and chloroplast will refine these simulations.

## ACKNOWLEDGMENTS

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