Modelling Nitrogen Content and Distribution in Cauliflower (*Brassica oleracea* L. *botrytis*)

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A model of nitrogen uptake and distribution is presented which describes these processes in relation to the amount of available soil nitrate and the rate of plant growth. Nitrogen uptake is either sink or source limited. Sink limitation is based on maximum N-concentrations of plant compartments. The N-uptake model is combined with a photosynthesis model based on the productivity-nitrogen relationship at the single-leaf level. The model is parameterized using cauliflower as an example crop. Applied to an independent data set, the combined model was able to predict leaf, stem and inflorescence nitrogen concentrations with correlation coefficients between predicted and simulated values of 0·89, 0·66 and 0·86, respectively. The influence of nitrogen supply and light intensity on leaf nitrate-N could also be predicted with good accuracy ($r^2 = 0.87$). Dry matter production based on the productivity-N relationship and the partitioning into leaf, stem and inflorescence was also reproduced satisfactorily ($r^2 = 0.91$, 0·93 and 0·92, respectively). © 2000 Annals of Botany Company

Key words: *Brassica oleracea* L. *botrytis*, cauliflower, nitrogen, nitrate, nitrogen supply, nitrogen uptake, nitrogen distribution, model.

INTRODUCTION

There are close positive relationships between nitrogen status and plant growth manifested by the close relationship between photosynthesis and nitrogen at the single-leaf level (Evans, 1983) as well as for relative growth rate of the whole-plant (Ågren, 1985). However, positive effects of high N-contents on productivity are accompanied by increased rates of respiration (Penning de Vries *et al.*, 1974; Byrd *et al.*, 1992; Alt *et al.*, 2000) which may partly offset the benefits of high N-contents on net C-accumulation rates. A mechanistic understanding of the interactions between available nitrogen, plant nitrogen content and plant productivity will help to quantify crop N-demand under specific environmental and agronomic conditions. This knowledge will assist the development of fertilizer recommendations and may thereby help to avoid both the environmental risks of excess rates of fertilization and also the effects of N-deficiency in crops.

A decrease in plant nitrogen content per unit dry weight with increasing plant dry weight has been observed for many crops (Greenwood *et al.*, 1990; Lemaire *et al.*, 1992; Vos *et al.*, 1996) and has been termed the 'law of progressive decline' in plant nitrogen concentration during crop growth (LeBot *et al.*, 1998). Greenwood *et al.* (1990) empirically quantified, for many species, a critical nitrogen concentration, n_{crit}, required for maximum growth rate. A comparison of the plant nitrogen concentrations during crop growth with n_{crit} may be used to identify nitrogen deficiency in plants (LeBot *et al.*, 1998). This concept of critical nitrogen concentrations was also used by Greenwood *et al.* (1991) to predict crop growth under optimal and sub-optimal N-supply. However, it is based on plant weight as the only causal factor determining the

change of optimal plant nitrogen concentrations during growth. It does not take into account possible effects of other environmental and agronomic conditions on n_{crit}. Such effects can, for example, be expected from the acclimatization of leaf nitrogen content to a changing radiation environment (Hirose and Werger, 1987). The n_{crit} concept also neglects negative effects of luxury N-consumption on crop productivity, caused by an enhanced protein-turnover and, thereby, increased maintenance respiration.

In a previous paper (Alt *et al.*, 2000), functional relationships between the fundamental physiological processes determining productivity, photosynthesis and respiration, and nitrogen content were established. These relationships were then used in a model-based analysis to quantify effects of given plant N-contents on crop productivity. However, prediction of crop nitrogen demand under certain environmental and agronomic conditions additionally requires prediction of plant nitrogen content and distribution during growth. The objective of the present work is, therefore, to elaborate relationships between nitrogen contents of cauliflower organs and environmental as well as morphological parameters, and to use these relationships in combination with the productivity–nitrogen-relationships presented in Alt *et al.* (2000) to predict nitrogen demand and to estimate crop productivity under different N-supplies.

MODEL

This study assumes that even under optimal N-supply daily nitrogen uptake by the plant is limited, and that this can be characterized by the maximum nitrogen concentrations of plant tissues and organs as they change

with time. This is referred to as the plant's physiological upper limit or as the sink limitation of N-uptake. Source limited N-uptake due to restricted diffusion and mass flow to the roots, even under conditions of optimal water supply, is assumed to be related to the average NO_3-N concentration in the soil exploited by the roots (N_{min}, kg ha^{-1}) (see Appendix for full list of symbols). A detailed mechanistic analysis of soil nitrate availability, taking into account the tempo-spatial variation of root length density (Kage *et al.*, 2000), as well as of soil water and soil nitrate content such as in the approaches presented by De Willigen and Van Nordwijk (1987) and Kage (1997), is beyond the scope of this paper. For the sake of simplicity, and in a first-step analysis, it is postulated that source limitation can be expressed by a factor f_{soil} ranging from one to the asymptotic value of zero as the available amount of nitrate in the soil decreases:

$$f_{soil} = \frac{N_{min}}{K_{Nmin} + N_{min}} \quad (1)$$

where K_{Nmin} (kgN ha^{-1}) is the parameter influencing the curvature of the rectangular hyperbola. At a value of N_{min} equal to K_{Nmin} the value of f_{soil} is 0·5.

Thus, the amount of nitrogen taken up and accumulated in the leaves, N_{leaf}, is assumed to be the maximum N-content corrected for source limitation, i.e.

$$N_{leaf} = n_{max,leaf} W_{leaf} f_{soil}(N_{min}) \quad (2)$$

where $n_{max,leaf}$ denotes the maximal N-concentration of the foliage under non-limiting N-supply and W_{leaf} is leaf mass. Due to senescence and self-shading, nitrogen translocation and remobilization within the cauliflower canopy lead to a decreasing leaf N-concentration with increasing leaf area index (LAI) even under optimal N-supply, assuming continuously increasing leaf area and leaf dry matter until harvest. Thus, $n_{max,leaf}$ may be expressed as a function of LAI:

$$n_{max,leaf} = -m_1 LAI + b_1 \quad (3)$$

where $-m_1$ denotes the decrease in $n_{max,leaf}$ with LAI under non-limiting N-supply, and b_1 is the physiological upper limit of the nitrogen concentration of well fertilized transplants.

The amount of nitrogen taken up per unit of time, dN_{leaf}/dt, is derived from eqn (2):

$$\frac{dN_{leaf}}{dt} = n_{max,leaf} f_{soil}(N_{min}) \frac{dW_{leaf}}{dt}$$
$$+ n_{max,leaf} W_{leaf} \frac{df_{soil}(N_{min})}{dt} \quad (4A)$$

where dW_{leaf}/dt denotes the increase in leaf dry matter. Since f_{soil} is assumed to be a continuous function depending on N_{min} the last term in eqn (4A) is of major importance whenever nitrogen supply in the soil changes very rapidly with time. Since this is not the case in the present study the term will be neglected in what follows. N-accumulation in stem, root and inflorescence has

the same form as the first term in eqn (4A) with N-concentrations in the stem, root and inflorescence, $n_{max,stem}$, $n_{max,root}$ and $n_{max,in}$, respectively:

$$\frac{dN_{stem}}{dt} = n_{max,stem} f_{soil}(N_{min}) \frac{dW_{stem}}{dt} \quad (4B)$$

$$\frac{dN_{root}}{dt} = n_{max,root} f_{soil}(N_{min}) \frac{dW_{root}}{dt} \quad (4C)$$

$$\frac{dN_{in}}{dt} = (n_{max,in} f_{soil}(N_{min}) + n_{red}) \frac{dW_{in}}{dt} \quad (4D)$$

The additional parameter n_{red} in eqn (4D) represents nitrogen redistribution from vegetative into generative organs as observed for many field crops (Booij *et al.*, 1997). All vegetative organs are supposed to contribute to the rate of nitrogen translocation, i.e. $n_{red}(dW_{in}/dt)$ [eqn (4D)], according to their proportion in the total amount of nitrogen stored in leaves, stem and root. The fraction translocated, e.g. from leaves, is given by $n_{red}(dW_{in}/dt)N_{leaf}/(N_{leaf} + N_{stem} + N_{root})$, which must be subtracted from eqn (4A) to give the correct total change in leaf N-content:

$$\frac{dN_{leaf}}{dt} = n_{max,leaf} f_{soil}(N_{min}) \frac{dW_{leaf}}{dt}$$
$$- n_{red} \frac{\frac{dW_{in}}{dt} N_{leaf}}{N_{leaf} + N_{stem} + N_{root}} \quad (5)$$

It is assumed that 1 % of the inflorescence nitrogen concentration results from N-redistribution within the plant, i.e. the value of n_{red} is set to 0·01. Thus, a significant amount (about 1/3) of the total of N in the inflorescence is assumed to result from N-redistribution in the plant.

The profile of protein-nitrogen in the canopy has been examined in detail (Alt *et al.*, 2000), based on the concept of optimal canopy N-distribution with respect to daily growth rate. The protein-N content per unit leaf area was observed to be more evenly distributed within the canopy than predicted by the optimized distribution which follows the life profile in the canopy closely. It is, therefore, assumed here that a linear relationship exists between nitrogen content per unit leaf area, n_a, and cumulative leaf area index, l_c, as determined from the top of the canopy. This relationship was also found in sunflower (Bange *et al.*, 1997). Thus,

$$n_a = -m_2 l_c + n_{a,max} \quad (6)$$

where $n_{a,max}$ is the maximum N-content at the top of the canopy and $-m_2$ denotes the decline with l_c.

Nitrogen contents based on dry weight and leaf area are inter-converted by using the specific leaf area, sla, which depends on the light environment during leaf growth (Björkman, 1981). The sla of growing leaves is assumed to be a function of the daily total of photosynthetic photon flux (PPF) averaged over previous days, I_{ave}:

$$sla = function(I_{ave}) \quad (7)$$

The nitrate-N concentrations in leaves depend on the light environment (Marschner, 1984). Therefore, the maximum nitrate-N concentration of the compartment leaf, $n_{max,NO3-N}$, is assumed to increase with increasing leaf area index due to decreasing average light intensity in the canopy. Thus, under non-limiting N-supply

$$n_{max,NO3-N} = \frac{I_{ref}}{I_{ave}} \text{function(LAI)} \qquad (8A)$$

where the quotient I_{ref}/I_{ave} relates the average light intensity above the canopy to a reference value. This means that the function dependent on LAI in eqn (7A) is parameterized under a certain light condition (I_{ref}) and then adapted to different conditions (I_{ave}) by multiplying by the quotient. To account for a decrease in leaf nitrate concentrations, n_{NO3-N}, due to limiting N-supply (Marschner, 1984; Gardner and Roth, 1990) it is assumed that the change in the nitrate concentration of the plant correlates linearly with N-supply (Gardner and Roth, 1990; Bellaloui and Pilbeam, 1991; Burns, 1994) or, here, f_{soil} as the determinant of source limitation. This assumption predicts that the nitrate concentration of the plant itself, n_{NO3-N}, is proportional to $(f_{soil})^2$ rather than f_{soil}:

$$n_{NO3-N} = n_{max,NO3-N} f_{soil}^2 (N_{min}) \qquad (8B)$$

This expression results in an asymptotic dependency of n_{NO3-N} on N-supply.

Cauliflower development

Leaf development is described in the same way as described by Kage and Stützel (1999); visible leaf number is an expo-linear function of the temperature sum, with base temperature zero, having the two parameters k_1 and k_2. Parameter values were determined from an experiment in 1997, independent of the one described in this study, for the cultivar 'Fremont' to be $k_1 = 0.0032$ $(d°C)^{-1}$ and $k_2 = 0.0350$ $(d°C)^{-1}$ (unpubl. res.). The vernalization process was calculated according to Wiebe (1972) using a daily vernalization rate, dV/dt, as a function of mean daily temperature, T:

$$\frac{dV}{dt} = \begin{cases} 0 & T \leq T1 \\ v_{max}\left(1 - \dfrac{T2 - T}{T2 - T1}\right) & T1 < T \leq T2 \\ v_{max} & T2 < T \leq T3 \\ v_{max}\left(1 - \dfrac{T - T3}{T4 - T3}\right) & T3 < T \leq T4 \\ 0 & T > T4 \end{cases} \qquad (9)$$

where v_{max} is the maximum vernalization rate set to 0.11 d^{-1}. The four temperatures, $T1$ to $T4$, are cultivar dependent and assumed to be 0, 10, 13 and 28°C for 'Fremont' (Wiebe, pers. comm.). The vernalization phase is completed when the sum of daily vernalization rates is one.

Photosynthesis

Photosynthesis was calculated from leaf area and protein-nitrogen content and distribution within the canopy, as well as from the light and temperature environment as described in Alt *et al.* (2000). The instantaneous rate of whole-plant net CO_2-uptake, P_{plant}, for a certain light environment, I, is the integral of single-leaf photosynthesis on a leaf area basis, P_g, over the leaf area, A_1, with whole-plant respiration, R_{plant}, subtracted:

$$P_{plant} = \int_0^{A_1} dl P_g(I, P_m) - R_{plant} \qquad (10)$$

P_g is given by the non-rectangular hyperbola,

$$P_g = \frac{\alpha I + P_m - \sqrt{(\alpha I + P_m)^2 - 4\Theta\alpha I P_m}}{2\Theta} \qquad (11)$$

where α and Θ are two parameters describing the initial slope and curvature of the response curve, respectively. Their values were determined to be $\alpha = 0.056$ µmol CO_2 µmol^{-1} PPF and $\Theta = 0.899$ (Alt, 1999). The single-leaf gross photosynthetic capacity, P_m (µmol CO_2 m^{-2} s^{-1}), varies within the canopy according to the N-content per unit leaf area, n_a (gN m^{-2}) (Alt *et al.*, 2000):

$$P_m = 12.82 \times n_a - 5.07 \qquad (12)$$

R_{plant} (mg CO_2 per plant min^{-1}) has contributions from maintenance respiration, temperature, T (°C), dependent and associated with the amount of protein-N in plant dry matter, $N_{P,plant}$ (g protein-N per plant), and from growth respiration, depending on the increase in dry mass per day, dW_{plant}/dt (g d. wt per plant d^{-1}) (Alt *et al.* (2000):

$$R_{plant} = 1.80 \times N_{P,plant} \times 2^{T-20/10} + 0.40 \times \frac{dW_{plant}}{dt} \qquad (13)$$

Respiration per unit leaf area, $R_{plant,a} = R_{plant}/A_1$, during the night is assumed to decrease linearly with the duration of darkness, t_n(h) (Alt *et al.*, 2000):

$$R_{plant,a}(t_n) = R_{plant,a}(0) \\ - (0.038 \times R_{plant,a}(0) - 0.042) \times t_n \qquad (14)$$

where the rate of decline depends on the initial respiration at the beginning of the night, $R_{plant,a}(0)$, which is determined by eqn (13) divided by A_1.

Light environment

The diurnal course of the photosynthetic photon flux (PPF) above the canopy, I_0, with time, t, is assumed to follow a sine-squared function:

$$I_0(t) = I_{noon} \sin^2\left(\pi \frac{t}{d}\right) \qquad (15)$$

where d denotes the period between sun rise and set, and I_{noon} is the maximum PPF at solar noon calculated from the

daily total PPF, I_{tot}, by $I_{noon} = 2I_{tot}d^{-1}$. I_{tot} is determined from the daily global radiation using a factor of 0·5. The light profile within the canopy is calculated by separating I_0 into diffuse, $I_{0,dif}$, and direct, $I_{0,dr}$, components based on Spitters (1986). The profiles of the diffuse and direct component at a certain depth in the canopy expressed as cumulative leaf area index, l_c, can be calculated as:

$$I_{df}(l_c) = I_{0,df}\,e^{-k_{df} \times l_c} \tag{16A}$$

$$I_{dr}(l_c) = I_{0,dr}e^{-\sqrt{(1-\sigma)} \times k_{bl} \times l_c} \tag{16B}$$

The values of the extinction coefficients for an approximated spherical leaf angle distribution are $k_{df} = 0·8 (1-\sigma)^{0.5}$ and $k_{bl} = 0·5 \sin\beta^{-1}$ (Goudriaan, 1977) where σ is the scattering coefficient, approximately 0·2, and β the solar elevation angle. I_{dr} splits into diffuse, $I_{dr,df}$ and direct, $I_{dr,dr}$, components:

$$I_{dr,df}(l_c) = I_{dr}(l_c) - I_{dr,dr}(l_c) \tag{17}$$

The direct part is given as the profile of light in a non-scattering canopy:

$$I_{dr,dr}(l_c) = I_{0,dr}e^{-k_{bl} \times l_c} \tag{18}$$

The relevant light intensity, I_{sh}, absorbed by the shaded leaf area at canopy height l_c is given by Spitters (1986):

$$I_{sh}(l_c) = k_{df}I_{df}(l_c) + k_{bl}I_{dr,df}(l_c) \tag{19}$$

The sunlit leaf area at canopy height l_c absorbs diffuse and non-scattered direct radiation:

$$I_{sl}(l_c) = I_{sh}(l_c) + (1-\sigma)k_{bl}I_{0,dr} \tag{20}$$

The fractions of sunlit leaf area, f_{sl}, and shaded leaf area, f_{sh}, at canopy height l_c are calculated according to the direct beam profile:

$$f_{sl}(l_c) = e^{-k_{bl} \times l_c} \tag{21A}$$
$$f_{sh}(l_c) = 1 - f_{sl}(l_c) \tag{21B}$$

The photosynthetic rate at canopy height l_c, $P_g(l_c)$ [eqn (11)], is the weighted sum of shaded and sunlit contributions:

$$P_g(l_c) = f_{sl}(l_c)P(I_{sl}(l_c)) + f_{sh}(l_c)P(I_{sh}(l_c)) \tag{22}$$

Dry matter partitioning

The increase in plant dry matter, dW_{plant}/dt, is the net CO_2-uptake P_{plant} [eqn (10)] multiplied by the CO_2–CH_2O-conversion factor of 30/44, and is partitioned between stem, leaf and inflorescence according to Alt (1999). After vernalization has been completed, reproductive growth has priority over vegetative growth in the model in order to satisfy the potential sink capacity of the inflorescence which is the product of its dry matter, W_{in}, and its specific growth rate, rgr_{in} (d^{-1}). The dry matter increase of the inflorescence is limited by its sink capacity at the beginning and, later on, by total available assimilates:

$$\frac{dW_{in}}{dt} = \min\left(\frac{1}{1 + f_{stem}}\frac{dW_{plant}}{dt}, rgr_{in}W_{in}\right) \tag{23}$$

where f_{stem} was experimentally determined to be 0·15 (Alt, 1999). Specific inflorescence growth rate, rgr_{in}, is a function of the nitrogen content per unit leaf area, n_{area}, at the time of inflorescence initiation:

$$rgr_{in} = 0·18 \times n_{area} \tag{24}$$

The growth rate of leaf dry matter is given by:

$$\frac{dW_{leaf}}{dt} = \frac{1}{1 + a \times sla + f_{root}}\left(\frac{dW_{plant}}{dt} - (1 + f_{stem})\frac{dW_{in}}{dt}\right) \tag{25}$$

where a was determined from experiments to be 12·93 g stem d.wt m^{-2} leaf area. The term sla denotes the specific leaf area of the newly produced leaf dry matter, and f_{root} is the fraction of root growth rate to leaf growth rate. Thus, stem and root growth rates are given by:

$$\frac{dW_{stem}}{dt} = a \times sla\frac{dW_{leaf}}{dt} + f_{stem}\frac{dW_{in}}{dt} \tag{26}$$

$$\frac{dW_{root}}{dt} = f_{root}\frac{dW_{leaf}}{dt} \tag{27}$$

The partitioning coefficient f_{root} is assumed to vary with nitrogen availability, $f_{root} = 0·15/f_{soil}$. The minimum value of 0·15 for f_{root} was determined from a greenhouse experiment in 1997 (Alt *et al.*, 2000) where cauliflower was cultivated in nutrient solution containing 145 mg N l^{-1}. A similar hyperbolic dependency of f_{root} on N-supply was found for seedlings of different rice cultivars where f_{root} varied from about 0·4 at very low, to about 0·16 at high N-supply (Cook and Evans, 1983).

MATERIALS AND METHODS

Two independent field experiments with cauliflower (*Brassica oleracea* L. convar. *botrytis* var. *botrytis* L. 'Fremont') were conducted on the institute's experimental farm located 15 km south of Hannover, Germany on a typical hapludalf soil derived from loess. The 1996 experiment was used for derivation of model parameters and the 1997 experiment for model evaluation. Seeds were sown in 4 cm^3 peat cubes. When the plants had developed an average of 3·25 and 3·5 visible leaves on June 18 and July 9 in 1996 and 1997, respectively, they were transplanted into the field. The initial dry weight at transplanting was 0·34 g per plant in 1996 and 0·39 g per plant in 1997. Plant spacing was 0·60 m by 0·48 m giving an average density of 3·5 plants m^{-2}. Before planting, chlorfenvinphos (Birlane) and molybdenum sulphate were applied prophylactically. Weeds were controlled by hand. Oxydemeton-methyl (250 g l^{-1} Metasystox) and later parathion (500 g l^{-1} E605 forte) were sprayed for pest control once in both years.

Experimental design

Experiments were laid out as split plots with two different light environments (unshaded, I1 and shaded, I2) as main plots and four different nitrogen-fertilizer levels as sub-plots in four replications. Shaded main plots were covered with a shading net absorbing 40 % of the photosynthetic photon-flux (PPF) placed 1 m above the ground, either immediately after transplanting (1996) or 2 weeks after transplanting (1997). Nitrogen fertilizer was given as ammonium nitrate at the time of transplanting to achieve 0, 150, 300 and 450 kg N ha^{-1}. In both years, soil nitrate content was 10–15 kg ha^{-1} at a depth of 0–60 cm and was allowed for in the applications: N0–N3 denote the 0–450 kg N ha^{-1} fertilizer applications.

Plant growth analysis

At three harvests in 1996, 28, 49 and 69 d after transplanting, and at four harvests in 1997, 26, 47, 68 and 82 d after transplanting, six adjacent plants per plot were collected and separated into stem, leaves (including petioles) and inflorescence. Stems were cut 1 cm below field level and at the onset of inflorescence development. The foliage was further subdivided into groups of five consecutive leaves (1–5, 6–10, etc.). The leaf number corresponded to leaf appearance. Leaf area of every leaf group was measured with a LICOR 3100 leaf area meter (LI-COR Inc., Lincoln, NE, USA). Samples of all plant fractions were oven dried and weighed. Total nitrogen and nitrate nitrogen concentration in dry matter were determined by the micro-Kjeldahl method and a nitrate sensitive electrode, respectively. Plant dimensions (height and width) and curd diameter were measured at all harvests.

Net mineralization

The amount of nitrogen mineralized during the growth period was determined from the N-budget of the N0-treatments as the sum of the measured amount of nitrogen in the above-ground plant parts and N_{min} from 0–120 cm depth at final harvest after subtracting N_{min} at transplanting. The average daily mineralization rate was calculated to be 0·72 kg N ha^{-1} d^{-1} and 0·67 kg N ha^{-1} d^{-1} in 1996 and 1997, respectively. This simplification, which was assumed to be sufficiently accurate under the conditions of the experiments, neglects temperature effects on mineralization during the rest of the season as well as the solute nitrate movement in the soil layers.

Model implementation and statistics

The dynamic model of development, dry matter production and partitioning as well as nitrogen uptake and distribution described above was programmed using the modelling environment ModelMaker (Walker, 1997). The integration was performed by the Euler-algorithm with a time step of 1 h. A non-linear least squares regression analysis is implemented in ModelMaker using a Marquardt optimization algorithm (Marquardt, 1963). Together with

the estimate of the parameter value, the software gives the value of the square root of the diagonal elements of the covariance matrix. Multiplying this value with the square root of the mean square of the residual yields the asymptotic standard error of the coefficient (Gallant, 1987). The non-linear least squares regression analysis implemented in ModelMaker was used to estimate the parameter K_{Nmin} [eqn (1)]. All statistical analyses, if not stated otherwise, were carried out using the procedures NLIN and REG of the SAS software package (SAS Institute, 1988).

RESULTS

Parameterization experiment, 1996

When N-supply was non-limiting, as could be assumed for the N3-treatments at harvests 1 and 2, the leaf nitrogen concentration declined during growth independent of light environment (Table 1). The same treatments caused an increase in nitrate-N concentration with increasing leaf area index during growth. The average daily total PAR of the non-shaded treatment during the entire period of crop growth, $I_{ref} = 7·8$ MJ m^{-2} d^{-1}, was used as the reference value in eqn (8). The nitrate-N contents of the shaded treatment were corrected for light environment by multiplying by a factor of 0·6, characterizing the transmission of the shading net. The linear regression equations were used to parameterize eqns (3) and (8B).

Protein-nitrogen per unit leaf area, n_a, decreased linearly with increasing depth in the canopy for all treatments as shown for the final harvest in Fig. 1A. Thus, the protein-nitrogen profile can be described by:

$$n_a(l_c) = n_{a,max}\left(1 - m_{na}\frac{l_c}{\text{LAI}}\right) \qquad (28A)$$

where m_{na} describes the slope of the linear decline and l_c is the cumulative leaf area index as determined from the top of the canopy. The maximum value of n_a at the top of the canopy, $n_{a,max}$, is then given by the total amount of leaf protein-N, $N_{P,leaf}$, by integrating eqn (28A) over LAI:

$$n_{a,max} = \frac{N_{P,leaf}}{\text{LAI}\left(1 - \frac{m_{na}}{2}\right)} \qquad (28B)$$

TABLE 1. *Regression equations fitted to the experimental data for maximum leaf N-concentration,* n$_{max,leaf}$, *and nitrate-N concentration,* n$_{max,NO3-N}$, *vs. leaf area index, LAI, and maximum inflorescence N-concentration vs. dry mass,* W$_{in}$, *under non-limiting N-supply in the I2-N3 (450 kg N ha^{-1}) treatment and first two harvests of I1-N3-treatment; experiment 1996*

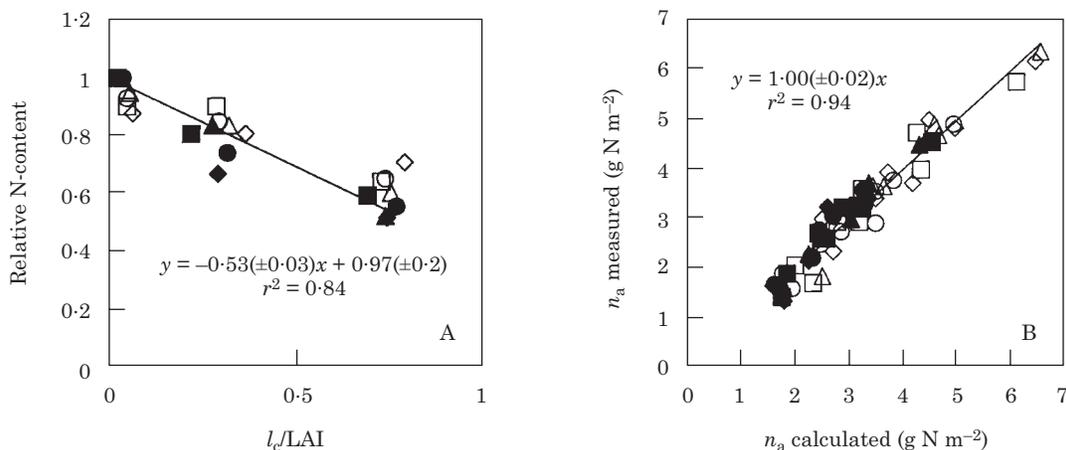| Organ	$n_{max,organ}$	r^2
Leaf-N	$-0·39(\pm 0·05)$LAI $+ 5·80(\pm 0·12)$	0·96
Leaf-NO$_3$-N	$0·08(\pm 0·02)$LAI $+ 0·40(\pm 0·04)$	0·87
Inflorescence	$6·38(\pm 2·29)$W$_{in}$ $- 0·10(\pm 0·02)$	0·96

FIG. 1. A, Measured protein-nitrogen contents per unit leaf area, n_a, relative to the maximum value at the top of the canopy vs. fraction of leaf area above, l_c/LAI, for the third harvest. B, Measured vs. calculated profile [eqn (28)] of leaf protein-nitrogen contents per unit leaf area for all harvests; experiment 1996: (○) I1-N0; (●) I2-N0; (□) I1-N1; (■) I2-N1; (△) I1-N2; (▲) I2-N2; (◇) I1-N3; (◆) I2-N3.
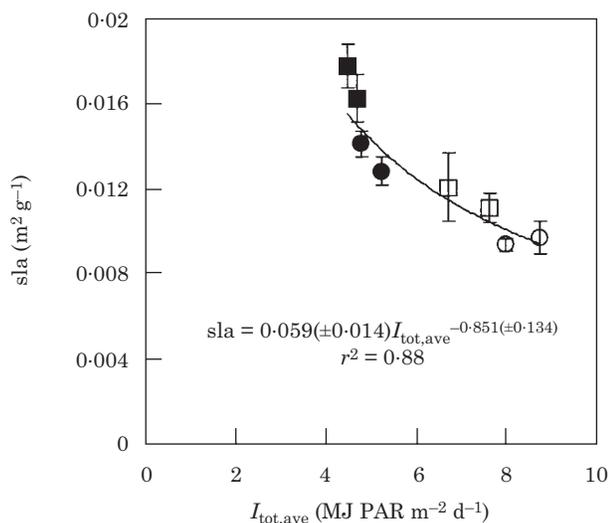

FIG. 2. Experimental relationship between specific leaf area, sla, of the youngest leaf group, and daily total of PPF averaged over 14 d prior to harvests, $I_{\text{tot,ave}}$; data from leaves 1–5 (first harvest) and leaves 11–15 (second harvest) in 1996 (○,●) and 1997 (□,■). (○,□) Non-shaded; (●,■) shaded treatments. Vertical bars denote $\pm$ s.d. of N-treatment means.

The parameter m_{na} was determined by non-linear least-squares regression analysis of measured and calculated protein-nitrogen profiles for all harvests and treatments in 1996. Its value was estimated to be $m_{\text{na}} = 0.53$ (± 0.02). The observed N-profiles for all treatments and harvests were well described by the same parameter value of m_{na} (Fig. 1B), which was used to parameterize eqn (5).

The maximum stem nitrogen-concentration, $n_{\text{max,stem}}$ was determined from the first harvest of the I2-N3-treatment to be 3.4%. The maximum root N-concentration was taken from an independent cauliflower experiment to be $n_{\text{max,root}} = 2.5\%$ (unpubl. res.). The maximum inflorescence N-concentration, $n_{\text{max,in}}$, was found to be a function of the inflorescence dry mass W_{in} (Table 1), parameterizing

eqns (4B)–(4D). Specific leaf area was related to the daily amount of PPF incident on the canopy and averaged over the period between harvests for all treatments (Fig. 2). The regression line was used to parameterize eqn (6). There were no statistically significant differences between the specific leaf areas of the N-treatments for the same light environment at the first two harvests.

The model parameter associated with source limiting processes is f_{soil} [eqns (5) and (8)]. Since the general behaviour of the dependency of f_{soil} on N_{min} is assumed to be smooth, it can be studied by comparing leaf-N contents for the different N-treatments. Maximum nitrogen contents per unit leaf mass as well as per unit leaf area declined with decreasing N-supply for all harvests (Fig. 3). The relationship between f_{soil} and N_{min} for each harvest is described well by a non-rectangular hyperbola [eqn (1)]. The parameter K_{Nmin} was determined by non-linear least-squares regression analysis of measured and simulated leaf, stem and inflorescence N-concentrations of all harvests and treatments in 1996. Its value was estimated to be $K_{\text{Nmin}} = 38.5$ (± 3.0) kg N ha^{-1}, parameterizing eqn (1).

The above-ground dry matter assimilation of all treatments was well simulated by the model for both years (Fig. 4). Unfortunately, no data on leaf dry weights and leaf area were obtained for the final harvest of the I1-N2- and I1-N3-treatments. Although the fertilized N-treatments at the same light environment showed similar dry matter production, leaf nitrogen contents as well as nitrate-N concentrations differed in early growth (Fig. 5). The leaf nitrate-nitrogen concentrations increased during growth for most of the N-fertilized treatments due to the decreasing average light intensity within the canopy. Limiting N-supply caused NO$_3$-N-concentrations to decline earlier and faster than leaf-N concentrations as found in cabbage (Burns, 1994). Nitrogen concentrations of stem and inflorescence differed similarly between treatments (Table 2) and also declined for all treatments during growth.
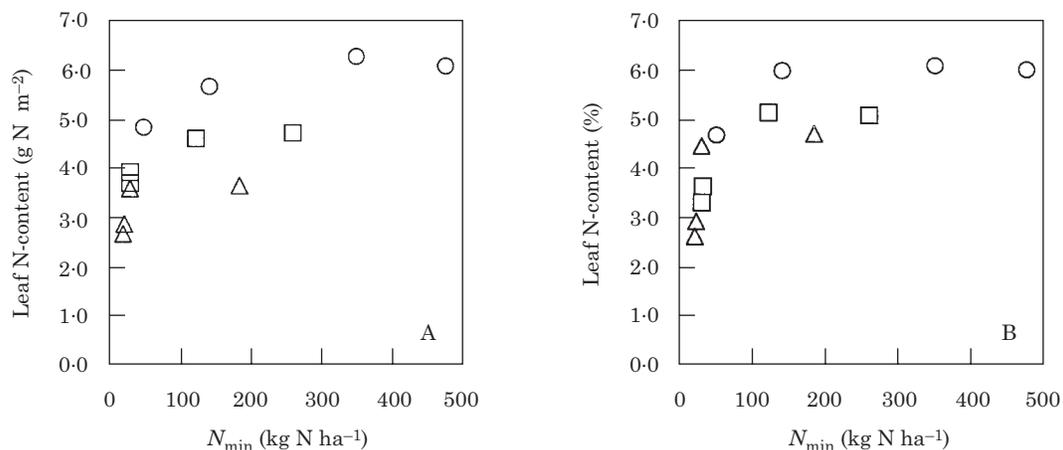
FIG. 3. Leaf area based (A) and mass based leaf nitrogen contents (B) of youngest leaf group *vs.* N_{min} from 0–120 cm for first (○), second (□) and final (△) harvests of non-shaded treatments; experiment 1996.
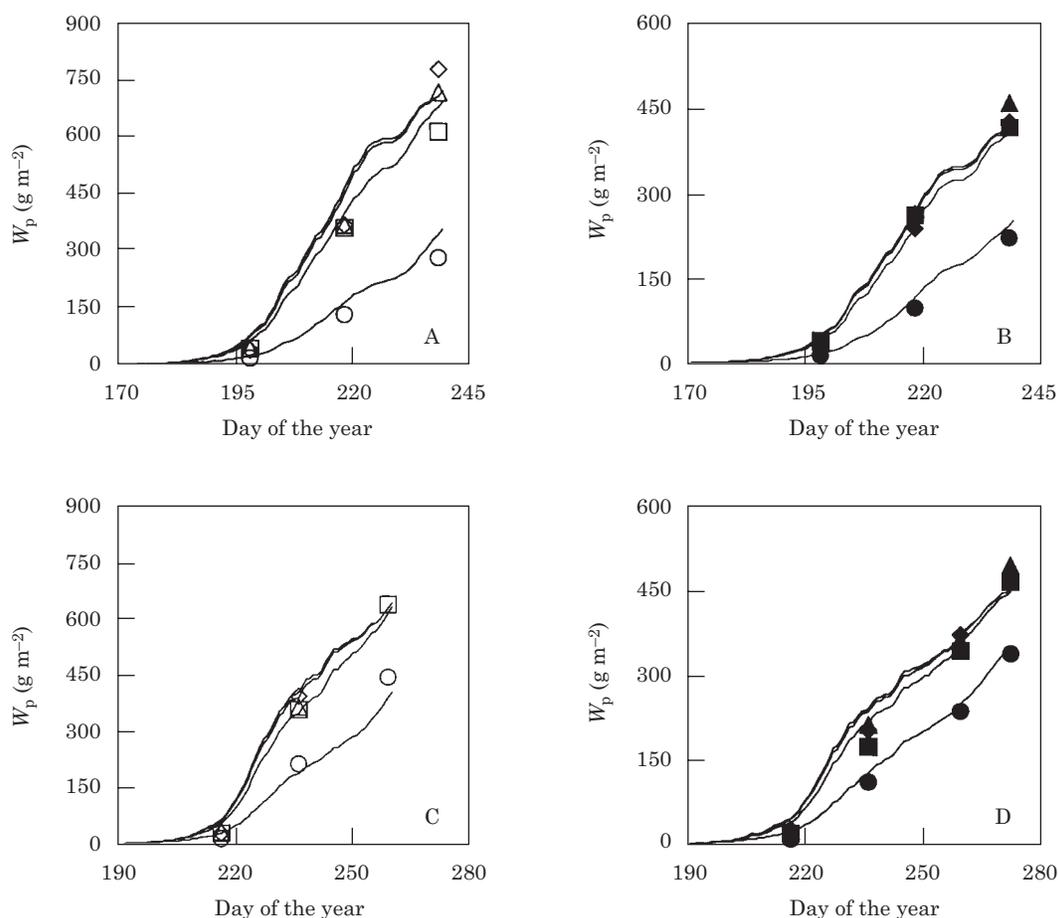

FIG. 4. Measured (symbols) *vs.* simulated (lines) above-ground dry weight, W_p, for all harvests; experiments 1996 (A,B) and 1997 (C,D); symbols as in Fig. 1.

Evaluation experiment, 1997

The model including model parameters derived above from the experiment in 1996 was evaluated with data from an independent field experiment in 1997 (Table 2). The leaf nitrogen-contents again showed differences between treatments soon after transplanting. The leaf nitrate-nitrogen

contents of the evaluation experiment were higher due to a reduction in average daily PPF of 15 % in 1997. The agreement of simulated and measured N-contents is equally high for shaded and non-shaded treatments. The model performance is of similar quality for the evaluation experiment as for the parameterization experiment. Nitrogen
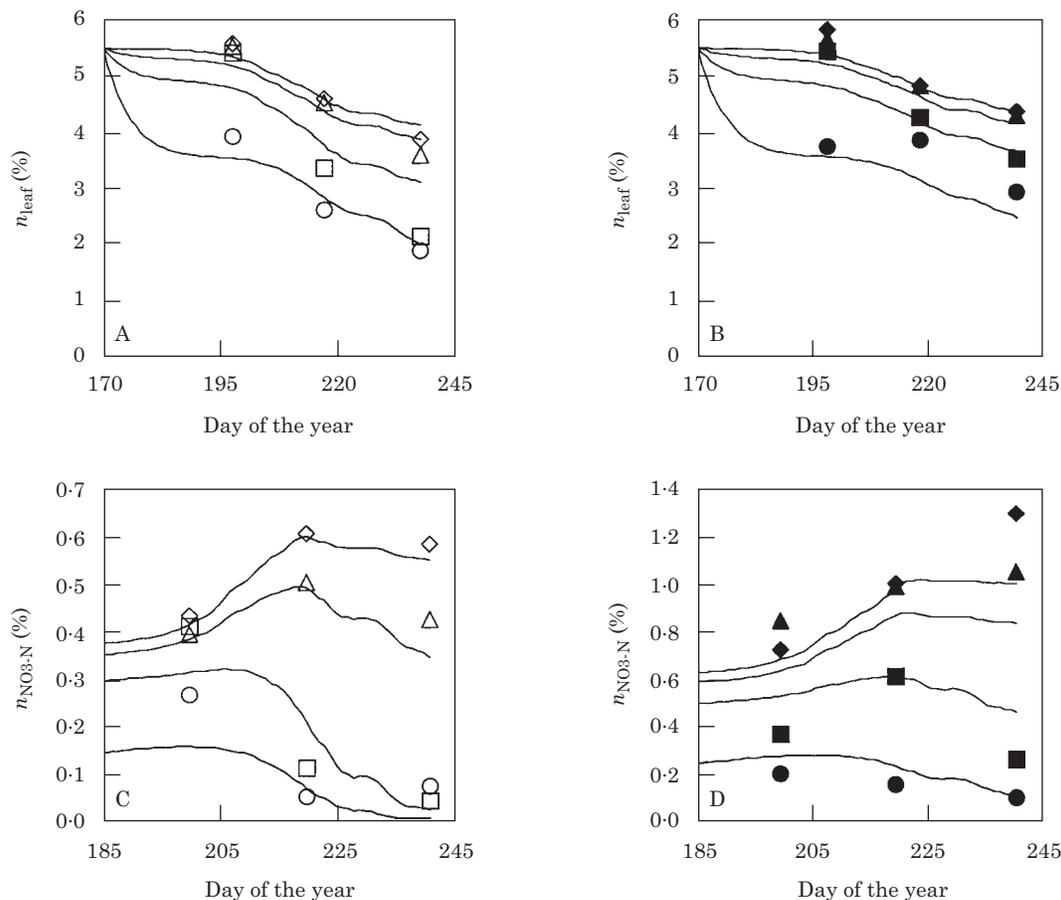
FIG. 5. Time course of measured (symbols) and simulated (lines) leaf-N content, n_{leaf} (A,B), and leaf nitrate-nitrogen content, n_{NO3-N} (C,D), for all harvests of non-shaded and shaded treatments; experiment 1996, symbols as in Fig. 1.

TABLE 2. *Results of the regression analyses* of measured vs. simulated N-concentrations of cauliflower organs in both experiments*

Experiment	Organ	Slope ($\pm$s.e.)	r^2
1996	Leaf-N	1·04 ($\pm$0·02)	0·89
1996	Leaf-NO$_3$-N	1·09 ($\pm$0·04)	0·91
1996	Inflorescence	0·97 ($\pm$0·04)	0·71
1996	Stem	0·95 ($\pm$0·02)	0·77
1997	Leaf-N	0·98 ($\pm$0·01)	0·89
1997	Leaf-NO$_3$-N	1·02 ($\pm$0·02)	0·87
1997	Inflorescence	0·96 ($\pm$0·01)	0·66
1997	Stem	0·99 ($\pm$0·02)	0·86

* In all cases intercepts were not significantly different from zero.

TABLE 3. *Results of the regression analyses* of measured vs. simulated dry matter of cauliflower organs and leaf area in both experiments*

Experiment	Organ	Slope ($\pm$s.e.)	r^2
1996	Above-ground total	1·05 ($\pm$0·03)	0·96
1996	Leaf	0·90 ($\pm$0·02)	0·98
1996	Stem	0·93 ($\pm$0·03)	0·94
1996	Inflorescence	1·11 ($\pm$0·05)	0·95
1996	Leaf area	0·82 ($\pm$0·03)	0·91
1997	Above-ground total	1·03 ($\pm$0·02)	0·96
1997	Leaf	1·05 ($\pm$0·04)	0·91
1997	Stem	1·05 ($\pm$0·03)	0·93
1997	Inflorescence	1·01 ($\pm$0·05)	0·92
1997	Leaf area	1·02 ($\pm$0·03)	0·94

* In all cases intercepts were not significantly different from zero.

concentrations of inflorescence and stem were similar to the data from 1996. A decline in N-concentrations was observed for all treatments and plant compartments during growth. The model predictability of the above-ground dry matter production and partitioning is compared for both experiments in Table 3.

DISCUSSION

The average leaf nitrogen concentration under conditions close to non-limiting N-supply was independent of light environment (Fig. 1) and declined during growth (Fig. 5). This was attributed to a maximum leaf N-concentration as a physiological upper limit that was not maintained during

leaf area growth, possibly due to senescence and self-shading, both increasing with increasing leaf area index. The decline in N-concentration during growth has been found in many species and has been termed the 'law of progressive decline' (LeBot *et al.*, 1998). Such a decline of N concentrations during growth of plants has also been explained by an increasing proportion of structural components with lower N content (Caloin and Yu, 1984). The small effect of shading on relative protein contents (Fig. 1) indicates that this was possibly one of the factors causing the observed decrease of nitrogen contents over time in our experiments.

The ratio of protein to nitrogen per unit leaf area declined linearly with increasing depth in the canopy, as also found for sunflower (Bange *et al.*, 1997). Theoretical considerations predict a N-distribution close to the light profile (Charles-Edwards *et al.*, 1987). This discrepancy, also found for soybean (Sinclair and Shiraiwa, 1993) and lucerne (Evans, 1993), may be partly due to additional energy consumption required for N-remobilization, which offset possible benefits of N-translocation (Field, 1983). However, the observed decline of N content within the canopy may again be the consequence of increasing proportions of structural leaf components of older leaves situated deeper in the canopy.

The specific leaf area of newly produced tissue was related to the average light intensity above the canopy (Fig. 2), which is closely related to the PPF incident on growing leaves at the top of the canopy. Although the changed wind conditions may have contributed to the observed differences in sla between shaded and non-shaded light environments (Retuerto and Woodward, 1992), they have not been taken into account. Their influence may be regarded to be of minor importance because the dependence of sla on PPF successfully described leaf area development for all treatments.

The reduction in N-uptake was well described by the factor f_{soil}, which is hyperbolically dependent on the amount of soil nitrate. The factor f_{soil} associated with N-uptake by the total root system can be compared to single-root influx rates which are widely accepted to be described by Michaelis–Menten kinetics (Peuke and Kaiser, 1996). The value of K_{Nmin} found in this study exceeds the values of the Michaelis–Menten constants obtained from nutrient solution experiments by one or two orders of magnitude (Peuke and Kaiser, 1996). This suggests that K_{Nmin} depicts mainly effects of a limited nitrate transport from the bulk soil to the root surface which was found for faba bean (Kage, 1997).

The nitrate-nitrogen contents in leaves were related to light environment and nitrogen supply. They were affected first by limiting N-supply and declined earlier than leaf protein-N contents. Although dry matter production was similar for the N-fertilized treatments in the same light environment in both years (Fig. 4), nitrogen-concentrations differed substantially soon after transplanting (1996 data: Fig. 5A and B). Both of these experimental findings, also found for many field crops (Booji *et al.*, 1996; Van den Boogaard and Thorup-Kristensen, 1997), were simulated from the model. This was achieved by increased photosynthetic capacity of the plant due to increased leaf protein-nitrogen content, which was offset by increased maintenance respiration due to enhanced protein-turnover under the given light environments.

In conclusion, the model presented was able to predict nitrogen uptake and distribution in cauliflower from plant growth rate, available soil nitrate-nitrogen content and radiation environment. The observed nitrogen concentrations declined in all plant organs during growth. The maximum leaf nitrogen content under non-limiting N-supply was independent of light environment. Although decreased leaf nitrogen concentrations due to limiting N-supply were observed soon after transplanting, dry matter production was affected later and the effect of N was smaller than on N-concentration. The increased photosynthetic capacity of plants due to increased leaf protein-nitrogen contents was mainly offset by increased maintenance respiration due to enhanced protein-turnover.

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APPENDIX

Symbols, definitions and units

Symbol	Definition	Unit
a	Increase in stem dry matter per increase in leaf area	g m^{-2}
A_1	Leaf area	m^2 per plant
d	Period between sunrise and sunset	h
dV/dt	Daily rate of vernalization	d^{-1}
f_{root}	Root partitioning coefficient	–
f_{sl}, f_{sh}	Fraction of sunlit and of shaded leaf area	–
f_{soil}	Factor denoting source N-limitation	–
f_{stem}	Stem partitioning coefficient	–
k_{bl}, k_{df}	Light extinction coefficient of direct, diffuse radiation	m^2 m^{-2}
K_{Nmin}	Curvature factor of the N_{min}-response curve	kg N ha^{-1}
LAI	Leaf area index	m^2 m^{-2}
l_c	Cumulative leaf area index	m^2 m^{-2}
m_{na}	Coefficient of protein-N profile	–
n_a	Protein-N content per unit leaf area	g N m^{-2}
$n_{a,max}$	Maximum protein-N content per unit leaf area	g N m^{-2}
$n_{max,NO3-N}$	Maximum leaf NO_3-N-content	g NO_3-N g^{-1}
$n_{max,x}$*	Maximum N-content	g N g^{-1}
N_{min}	Available soil nitrate-N	kg N ha^{-1}
n_{NO3-N}	Leaf NO_3-N-content	g NO_3-N g^{-1}
$N_{P,plant}$	Total of plant protein-N	g protein-N per plant
n_{red}	Coefficient of N-redistribution	g N g^{-1}
n_s	Structural leaf N-content per unit leaf area	g N m^{-2}
N_x*	Total amount of nitrogen in the plant	g N per plant
n_x*	N-amount in dry matter	g N g^{-1}
P_g	Rate of gross photosynthesis	μmol CO_2 m^{-2} s^{-1}
P_m	Rate of gross photosynthetic capacity	μmol CO_2 m^{-2} s^{-1}
P_{plant}	Rate of whole-plant net CO_2-uptake	μmol CO_2 per plant s^{-1}
rgr_{in}	Relative growth rate of inflorescence	d^{-1}
R_{plant}	Rate of whole-plant respiration	μmol CO per plant s^{-1}
$R_{plant,a}$	Rate of whole-plant respiration per unit leaf area	μmol CO_2 per plant m^{-2} s^{-1}
sla	Specific leaf area	m^2 g^{-1}
T	Temperature	°C
V	Vernalization	d^{-1}
v_{max}	Maximum daily rate of vernalization	d^{-1}
W_x*	Dry weight	g per plant
I	Photosynthetic photon flux (PPF)	μmol m^{-2} s^{-1}
I_0	PPF above the canopy	μmol m^{-2} s^{-1}
I_{ave}	Daily total of PPF averaged over 14 d	MJ m^{-2} d^{-1}
I_{noon}	Maximum PPF at solar noon	μmol m^{-2} s^{-1}
I_{ref}	Reference value of daily total of PPF	MJ m^{-2} d^{-1}
$I_{0,dr}, I_{0,df}$	Direct, diffuse PPF above canopy	μmol m^{-2} s^{-1}
I_{dr}, I_{df}	Direct, diffuse component of PPF	μmol m^{-2} s^{-1}
I_{sl}, I_{sh}	PPF of sunlit, shaded leaves	μmol m^{-2} s^{-1}
σ	Scattering coefficient	–
θ	Curvature factor of the light-response curve	–
α	Initial slope of the light-response curve	μmol CO_2 μmol^{-1} PPF

* index x: in, inflorescence; leaf, leaf; plant, whole-plant; root, root; stem, stem.