Modelling Ontogenetic Changes of Nitrogen and Water Content in Lettuce

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• Background and Aims It is well established that the nitrogen content of plants, including lettuce, decreases with time. It has also been observed that water content of lettuce increases between planting and harvest. This paper is an attempt at modelling these observations.

• Methods An existing dynamic model (NICOLET), designed to predict growth and nitrate content of glasshouse lettuce, is modified to accommodate the ontogenetic changes of reduced-nitrogen and water contents (on a dry matter basis). The decreasing reduced-N content and the increasing water content are mimicked by dividing the originally uniform plant into ‘metabolically active’ tissue and ‘support’ tissue. The ‘metabolic’ tissue is assumed to contain a higher nitrogen content and a lower water content than the ‘support’ tissue. As the plants grow, the ratio of ‘support’ to ‘metabolic’ tissue increases, resulting in an increased mean water content and a decreased reduced-N content. Simulations with the new model are compared with experimental glasshouse data over four seasons.

• Key Results The empirical linear relationship between water and reduced-N contents, matches, to a good approximation, the corresponding relationship based on the model. The agreement between the two makes it possible to effectively uncouple the estimation of the ‘ontogenetic’ parameters from the estimation of the other parameters. The growth and nitrate simulation results match the data rather well and are hardly affected by the new refinement. The reduced-N and water contents are predicted much better with the new model.

• Conclusion Prediction of nitrogen uptake for the substantial nitrate pool of lettuce depends on the water content. Hence, the modified model may assist in making better fertilization decisions and better estimates of nitrogen leaching.

Key words: Lactuca sativa L, lettuce, ontogenetic changes, nitrogen uptake, chemical composition, nitrogen content, water content, dynamic model, metabolic and support compartments.

INTRODUCTION

When plants are grown to maturity in agricultural stands, an ontogenetic decline of nitrogen content (so called ‘N-dilution’) is generally observed [Greenwood et al. (1990) – several C3 and C4 crops; Justes et al. (1994) – winter wheat; Sheehy et al. (1998) – rice; Colnenne et al. (1998) – winter oilseed rape; Plénet and Lemaire (2000) – maize; Greenwood and Draycott (1989) – various vegetable crops]. This phenomenon is generally modelled by viewing the plants as comprising two types of tissue. One, ‘metabolically active’, responsible for photosynthesis and nutrients uptake, with a high content of nitrogenous compounds; and the other, consisting of mechanical and vascular ‘support’ elements, with a low content of N-compounds (Caloin and Yu, 1984). Hirose and Werger (1987) and Chen et al. (1993) associated the metabolically active tissue with the sunlit leaves, and the support tissue with the shaded leaves and stems. As plants grow, the mass ratio of sunlit leaves to total plant material decreases. Nitrogen-rich compounds, no longer required in the shaded parts, are re-allocated to the younger leaves (Seligman, 1993), resulting in a decline of nitrogen content with depth in the canopy (Charles-Edwards et al., 1987; Grindlay et al. 1995; Alt et al., 2000) and in the canopy as a whole.

This description is inspired by the morphology of ordinary crops, where the young leaves are exposed to solar radiation more than the older leaves. In head-lettuce the young leaves are shaded and yet the same ‘dilution’ curve is evident. One way to treat this apparent inconsistency is to associate, as before, ‘metabolic’ with ‘sunlit’, despite the fact that the sunlit leaves are no longer the young ones. Another option is to assume that the composition of young and old leaves is, in evolutionary terms, a slow-changing trait, compared with leaf morphology (head formation). Finally, it is possible to abandon any attempt at an essential interpretation (for lack of suitable data) and view the model as an instrument, whose task is to produce correct predictions of crop nitrogen content. Our approach here is the last of these, retaining for convenience the description of an ‘ordinary’ crop to develop the lettuce model.

The total nitrogen content of plants is conveniently divided into nitrate (inorganic) and reduced (organic) nitrogen. The ratio nitrate-N:reduced-N has a wide range, from essentially zero (N-stressed plants; Broadley et al., 2003, Linker et al., 2004) to one or more (Table 1). The ‘critical’ nitrogen content, as traditionally plotted in N-dilution curves and believed to be required for maximum growth (Ulrich, 1952), seems to include all the reduced-N and possibly part of the nitrate-N. The extra nitrate, if any, is often considered to result from ‘luxury
Table 1. Summary of lettuce experiments carried out at Beitem during 2000

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>'Flandria'</th>
<th>'Troubadour'</th>
</tr>
</thead>
<tbody>
<tr>
<td>Season</td>
<td>Spring</td>
<td>Summer</td>
</tr>
<tr>
<td>Plant density (plant m⁻²):</td>
<td>13-7</td>
<td>11-1</td>
</tr>
<tr>
<td>Code (as in Figs 4 and 6):</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Mean daily light integral* (mol[PAP] m⁻² d⁻¹)</td>
<td>16-4</td>
<td>24-9</td>
</tr>
<tr>
<td>Mean temperature* (°C)</td>
<td>16-4</td>
<td>19-0</td>
</tr>
<tr>
<td>Mean CO₂ concentration* (mmol[CO₂] m⁻³)</td>
<td>14-7</td>
<td>12-6</td>
</tr>
<tr>
<td>Mean CO₂ concentration* (ppm)</td>
<td>354</td>
<td>285</td>
</tr>
<tr>
<td>Sowing date</td>
<td>16 Mar 00</td>
<td>15 May 00</td>
</tr>
<tr>
<td>Transplanting date</td>
<td>11 Apr 00</td>
<td>31 May 00</td>
</tr>
<tr>
<td>Harvest date</td>
<td>22 May 00</td>
<td>04 Jul 00</td>
</tr>
<tr>
<td>Time in glasshouse (days)</td>
<td>41</td>
<td>34</td>
</tr>
<tr>
<td>Total light integral* (mol[PAP] m⁻²)</td>
<td>673</td>
<td>845</td>
</tr>
<tr>
<td>DM at initial harvest (g[DM] plant⁻¹)</td>
<td>0.219</td>
<td>0.046</td>
</tr>
<tr>
<td>DM at final harvest (g[DM] plant⁻¹)</td>
<td>20.9</td>
<td>20.2</td>
</tr>
<tr>
<td>DMC at final harvest (g[DM] kg⁻¹[FM])</td>
<td>41</td>
<td>46</td>
</tr>
<tr>
<td>DM at final harvest (g[DM] m⁻²)</td>
<td>290</td>
<td>220</td>
</tr>
<tr>
<td>‘Light efficiency’ of growth [g[DM] mol⁻¹(PAP)]</td>
<td>0.43</td>
<td>0.27</td>
</tr>
<tr>
<td>Nitrate at final harvest (mmol[N] kg⁻¹[DM])</td>
<td>1400</td>
<td>1100</td>
</tr>
<tr>
<td>Reduced-N at final harvest (mmol[N] kg⁻¹[DM])</td>
<td>1230</td>
<td>1260</td>
</tr>
</tbody>
</table>

Data are averaged over compartments (two or four), blocks (four) and N treatments (two).

The ‘light efficiency’ is obtained by dividing the total dry mass increase by the total light integral.

Values in bold indicate apparent irregularities.

*Indoor environment.

Fig. 1. Relationship between water and reduced nitrogen, resulting from ontogenetic decrease of N content and increase of water content in soil-grown butterhead lettuce plants. Squares, spring; triangles, summer; diamonds, autumn; circles, winter. Spring and summer data for cultivar ‘Flandria’ and autumn and winter data for ‘Troubadour’. Data provided by P. Bleyaert and M. Breugelmans, Beitem, Belgium. The solid line is the regression through all the points. The dashed lines indicate the range between young (lower right) and mature plants (upper left).
During this early stage of growth the composition of the seedlings is time-invariant and growth is exponential. If the plants are N-stressed, the N:C ratio decreases (Oscarson et al., 1989; Ingestad and Ågren, 1992) as does the water content (Oscarson et al., 1989), resulting in the pattern of Fig. 2.

The data in Figs 1 and 2, for the young, unstressed plants, should coincide. However, in Fig. 1 these are clustered around [2-5, 15], while in Fig. 2 they are clustered around [3, 20]. This discrepancy may be attributed to different lettuce cultivars, different growing conditions (Fig. 1 in soil, Fig. 2 in hydroponics) or, most likely, to different chemical analysis methods.

This paper is an attempt to model quantitatively the data of Fig. 1, by modifying a previously developed dynamic lettuce model, NICOLET (Seginer, 2003a), which is valid for the initial stage of growth, where composition is time-invariant (Fig. 2). The modification extends the validity of the model to a later stage of vegetative growth where ontogenetic composition-changes take place. We assume here, following Caloin and Yu (1984), that viewing the plant as comprising distinct metabolic and support components is essentially correct, and that the trend shown in Fig. 1 is real.

THE BEITEM DATA

During the years 1999 and 2000, experiments were conducted in the Provincial Research Centre at Beitem, Belgium (latitude 50°54'N, longitude 03°07'W, 30 m a.s.l.), in an attempt to quantify the effect of various environmental treatments (supplementing nitrogen, light, CO₂ and heat) on the nitrate content of butterhead lettuce. The crops were soil-grown in glasshouse compartments, following common agricultural practices, except for the environmental treatments. The second-year experiments produced more consistent data than the first year and were, therefore, selected for the present modelling attempt. A detailed description of these experiments is available upon request (Bleyaert and Breugelmans, 2001; unpubl. res.).

Data from experiments where N supply was judged from the results to be ample and for which chemical analyses were available, were used for the current analysis. Since the interest here is not in the effects of the various treatments (which only rarely resulted in significant effects), the data were pooled to produce four distinct data sets, as shown in Table 1. The environmental data were averaged over the two experimental compartments (or four sub-compartments, when appropriate), and the crop data were averaged over the two higher N treatments, over four blocks (replications), as well as over the compartments (climatological treatments). Hence, each point in Fig. 1 and all the averaged fresh mass, dry mass, nitrate content and reduced-N content data (except the initial harvests), are means of at least eight individual determinations. The crops were sampled for analysis five or six times, at approximately uniform intervals of light integral, along the growing period, starting with the day of transplanting and ending at the final harvest.

Table 1 shows that the effects of season and cultivar are partially confounded. Cultivar ‘Flandria’ is associated with spring and summer and cultivar ‘Troubadour’ is associated with autumn and winter (no confounding for seasons within each cultivar). The ‘Flandria’ points in Fig. 1 appear to lie a bit lower than those of ‘Troubadour’. A few apparent irregularities can be found in Table 1 (bold font): (a) the transplants of the spring crop were unusually large; (b) the ‘light efficiency’ of the summer crop was considerably lower than that of the other crops; (c) the reduced-N of the autumn crop is unusually low.

THE CURRENT NICOLET MODEL

Description

The NICOLET model is described and justified in detail elsewhere (Seginer, 2003a). It consists of three (virtual) compartments, labelled ‘structure’, ‘vacuole’ and ‘excess-carbon’. The soluble organic and mineral constituents of the vacuole complement each other in contributing to the (constant) osmotic potential, while the carbon and organic-N compounds of the other two compartments are osmotically neutral. The structural N:C ratio and the water:structural-carbon ratio are assumed to be constant. The excess-carbon compartment is assumed to contain only ‘dry’ carbon compounds, and is usually activated only when the crop is N-stressed. Since the available Beitem data are for abundant supply of nitrogen, the excess-carbon compartment and the nutrition uptake formulation are omitted from the summary description provided here, as well as from the computations. A few simplifying notational changes have also been introduced. The resulting two-compartment model is a special case of the (original) three-compartment model. It may be obtained from the latter by setting \( \xi = 0 \) in eqns [27] to [29] of Seginer (2003a). The simplified model is shown in Fig. 3.
The fluxes are controlled by: (a) the environmental conditions – photosynthesis by light and CO₂, and structural growth by temperature; (b) the ground cover of the crop; and (c) attenuation functions, which reflect the inhibition of certain processes. Photosynthesis is modelled as a Michaelis–Menten process, and respiration and growth are formulated as exponential functions of temperature \((Q_{10} = 2)\). The ground cover, to which all fluxes are proportional, approaches one asymptotically, and the attenuation functions control the flows in times of stress.

If the environmental conditions are constant, the current NICOLET model predicts that the composition of the crop equilibrates with these conditions, namely remains constant with time. This will be changed below, to allow for ontogenetic effects.

**System equations**

In the following equations, the symbols denoting masses and fluxes are defined in Fig. 3. The environmental conditions \(I\), \(T\) and \(C_C\) are light flux, ambient temperature and CO₂ concentration in the air. The parentheses \(\{\}\) enclose the arguments of functions, and \(t\) is time. All other symbols are constants, defined in the notation list and explained in Seginer (2003a).

(i) **Carbon balances** (used as state equations)

\[
\frac{dM_{CV}}{dt} = F_{Cp} - F_{CVs} - F_{CG} - F_{Cm} \quad \text{in 'vacuole'} \quad (1)
\]

\[
\frac{dM_{Cs}}{dt} = F_{CVs} \quad \text{in 'structure'} \quad (2)
\]

(ii) **Nitrogen balances**

\[
\frac{dM_{NV}}{dt} = F_{Na} - F_{Nvs} \quad \text{in 'vacuole'} \quad (3)
\]

\[
\frac{dM_{Ns}}{dt} = F_{Nvs} \quad \text{in 'structure'} \quad (4)
\]

(iii) **Compositional relationships**

\[
\beta_{C} M_{CV} + \beta_{N} M_{NV} = \lambda \Pi M_{Cs} \quad \text{'osmotica balance'} \quad (5)
\]

\[
F_{Nvs} = rF_{CVs} \quad \text{in 'structure'} \quad (6)
\]

\[
V = \lambda M_{Cs} \quad (7)
\]

Equation (5) formulates the complementing role of nitrate, \(M_{NV}\), and soluble organic compounds, \(M_{CV}\), in maintaining the osmotic potential; eqn (6) formulates the proportionality between nitrogen, \(F_{Nvs}\), and carbon, \(F_{CVs}\), in the structure; and eqn (7) formulates the proportionality between water, \(V\), and structural carbon, \(M_{Cs}\).

(iv) **Normalized content of soluble carbon-compounds**

\[
\Gamma = \frac{\beta_{C} M_{CV}}{\lambda \Pi M_{Cs}}, \quad 0 \leq \Gamma \leq 1 \quad \text{in 'vacuole'} \quad (8)
\]

(v) **Carbon fluxes**

\[
F_{Cp} = p\{I, C_c\}f\{M_{Cs}\}h_p\{\Gamma\} \quad (9)
\]

\[
F_{Cm} = e\{T\}f\{M_{Cs}\} \quad (10)
\]

\[
F_{CVs} = g\{T\}f\{M_{Cs}\}h_g\{\Gamma\} \quad (11)
\]

\[
F_{CG} = \theta F_{CVs} \quad (12)
\]

Equations (9) to (11) are products of a function of the environment, \(p\), \(e\) or \(g\), a function of the size of the crop, \(f\), and an attenuation function, \(h\), which are given in more detail, as follows:

\[
p\{I, C_c\} = \frac{\epsilon x \sigma C_c}{\epsilon I + \sigma C_c} \quad (13)
\]

\[
e\{T\} = k \exp\{c(T - T^*)\} \quad (14)
\]

\[
g\{T\} = ve\{T\} \quad (15)
\]

\[
\frac{1}{f\{M_{Cs}\}} = 1 - \exp\{-a M_{Cs}\} \quad (16)
\]

where \(f\) is the fraction of light intercepted by the canopy, and all fluxes are proportional to \(f\).
(vi) Attenuation functions

\[ h_p(\Gamma) = \frac{1}{1 + \left(\frac{1 - h_p}{1 - \Gamma}\right)^{s_p}} \]  

\[ h_g(\Gamma) = \frac{1}{1 + \left(\frac{h_g}{\Gamma}\right)^{s_g}} \]  

The attenuation functions protect the vacuole from carbon over-spilling and over-draining.

EXTENSION TO INCLUDE METABOLIC AND SUPPORT COMPONENTS

Variable \( r \) and \( \lambda \)

The originally uniform plant, with structural N : C ratio \( r \) and water : structural-C ratio \( \lambda \) (eqns 6 and 7), is now divided into ‘metabolic’ and ‘support’ components. The structural N : C ratios of these components are denoted by \( r^{(m)} \) and \( r^{(s)} \), where \( m \) and \( s \) denote ‘metabolic’ and ‘support’, respectively, and \( r^{(m)} \) is larger than \( r^{(s)} \). The water : structural-C ratios are similarly denoted by \( \lambda^{(m)} \) and \( \lambda^{(s)} \), where \( \lambda^{(m)} \) is smaller than \( \lambda^{(s)} \).

By definition

\[ M_{Cs} = M_{Cs}^{(m)} + M_{Cs}^{(s)} \]  

The metabolic component is made proportional to the fraction of intercepted light, given by eqn (16). When the crop is young, namely when \( aM_{Cs} \ll 1 \),

\[ f\{M_{Cs}\} \approx aM_{Cs} \]  

and since at that time essentially all leaves are sunlit, the metabolic component may be defined by

\[ M_{Cs}^{(m)} = \frac{f\{M_{Cs}\}}{a} \]  

With this, the total reduced-nitrogen content becomes

\[ M_{Ns} = M_{Ns}^{(m)} + M_{Ns}^{(s)} = r^{(m)}M_{Cs}^{(m)} + r^{(s)}M_{Cs}^{(s)} \]  

\[ = \left(r^{(m)} - r^{(s)}\right) \frac{f\{M_{Cs}\}}{aM_{Cs}} + r^{(s)}M_{Cs} \]  

(22)

Note that \( M_{Ns}^{(m)} \) is proportional to \( f\{M_{Cs}\} \), which in view of eqns (9) to (12) makes the rate of photosynthesis, and all other metabolic activities, directly proportional to the reduced-nitrogen content of the metabolic compartment. This is in agreement with Agren’s (1985) view of ‘nitrogen productivity’.

Dividing eqn (22) by \( M_{Cs} \), the N : C ratio of the structure as a whole is obtained:

\[ r\{M_{Cs}\} = \frac{M_{Ns}}{M_{Cs}} = \left(r^{(m)} - r^{(s)}\right) \frac{f\{M_{Cs}\}}{aM_{Cs}} + r^{(s)} \]  

(23)

Differentiating eqn (22) with respect to time and replacing state derivatives with the appropriate fluxes, the flux of nitrogen into the structure can be related to the flux of carbon as

\[ F_{Nv} = \left[\left(r^{(m)} - r^{(s)}\right)(1 - f) + r^{(s)}\right] F_{ Cv s } \]  

(24)

which shows that initially the nitrogen flux is a fraction \( r^{(m)} \) of the carbon flux, diminishing towards \( r^{(s)} \) as the canopy closes (as \( f \to 1 \)).

The total water content is, similar to eqn (22)

\[ V = \left(\lambda^{(m)} - \lambda^{(s)}\right) \frac{f\{M_{Cs}\}}{a} + \lambda^{(s)}M_{Cs} \]  

(25)

and the overall \( V : M_{Cs} \) ratio is, similar to eqn (23)

\[ \lambda\{M_{Cs}\} = \frac{V}{M_{Cs}} = \left(\lambda^{(m)} - \lambda^{(s)}\right) \frac{f\{M_{Cs}\}}{aM_{Cs}} + \lambda^{(s)} \]  

(26)

This extension to the original model adds two parameters \( \{r^{(m)}, \lambda^{(m)}\} \) instead of just \( r \) and \( \lambda \), but requires no additional state variables. The original, constant-\( r \) and constant-\( \lambda \) formulation, is a special case of the new model and can be obtained by setting \( r^{(m)} = r \) and \( \lambda^{(m)} = \lambda^{(s)} = \lambda \).

Uptake of nitrogen

Differentiating the osmotica balance, eqn (5), with respect to time, and utilizing eqns (1), (2), (3), (6), (7), (23) and (26), results (Appendix A) in the flux form

\[ G\{f\}F_{ Cv s } - \beta_N F_{ Nu } = \beta_C \left( F_{ Cp } - F_{ Cm } \right) \]  

(27)

where

\[ G\{f\} = \beta_C (1 + \theta) + \beta_N \left( r^{(m)} - r^{(s)}\right)(1 - f) + r^{(s)} \]  

\[ + \Pi \left[ (\lambda^{(m)} - \lambda^{(s)}) \frac{f\{M_{Cs}\}}{aM_{Cs}} + (\lambda^{(m)} - \lambda^{(s)})(1 - f) + \lambda^{(s)} \right] \]  

(28)

Hence, the uptake of nitrogen (when N-supply is abundant) is

\[ F_{Nu} = \frac{G\{f\}}{\beta_N} F_{Cv s } - \frac{\beta_C}{\beta_N} \left( F_{Cp } - F_{Cm } \right) \]  

(29)

CONVERSION BETWEEN MEASUREMENTS AND MODEL STATES

The comparison of model predictions with experimental data requires that the two can be mapped back and forth into each other. The experimental results of Bleyaert and Breugelmans can be expressed in terms of (a) fresh mass per unit ground area, \( W_F \), (b) dry mass per unit ground area, \( W_D \), (c) molar nitrate content on dry-mass basis, \( C_{ nit-N } \), and (d) molar reduced-N content on dry-mass basis, \( C_{ red-N } \).

From these, the water volume per unit ground area is determined via

\[ V = \frac{W_F - W_D}{\rho} \]  

(30)
where \( \rho \) is the density of water. Combining this with eqn (26), an implicit expression for one of the state variables, \( M_{C_v} \) (eqn 2), is obtained

\[
M_{C_v} = \frac{1}{\rho \lambda \{M_{C_s}\}} (W_F - W_D) \tag{31}
\]

From the definition of \( C_{\text{nit-N}} \),

\[
M_{N_v} = W_D C_{\text{nit-N}} \tag{32}
\]

and substituting eqns (31) and (32) into eqn (5), an expression for the second state variable, \( M_{C_v} \) (eqn (1)), is obtained

\[
M_{C_v} = \frac{\Pi}{\rho \beta_C} (W_F - W_D) - \frac{\beta_N}{\beta_C} (W_D C_{\text{nit-N}}) \tag{33}
\]

Equations (31) and (33) express the state variables in terms of the measured quantities \( W_F, W_D \) and \( C_{\text{nit-N}} \) as well as some of the model parameters.

The inverse conversion requires, in addition, an assumption regarding the conversion of dry matter, such as

\[
W_D = \eta_C (M_{C_s} + M_{C_v}) + \eta_N M_{N_v} \tag{34}
\]

where the conversion factors, \( \eta_C \) and \( \eta_N \), are determined from the molecular masses of typical compounds.

**ESTIMATING THE PARAMETERS FOR THE ‘METABOLIC’ AND ‘SUPPORT’ COMPONENTS**

**Analysis**

Figure 1 shows a negatively sloping linear relationship between water content and reduced-N content. The empirically fitted line may be expressed as

\[
\frac{\rho V}{W_D} = A \frac{M_{C_s}}{W_D} + B \tag{35}
\]

where \( A \) (negative) and \( B \) are the slope and intercept.

The ontogenetic model is in general agreement with this observation, as can be shown by eliminating \( f(M_{C_s})/\rho M_{C_s} \) between eqns (23) and (26), and multiplying throughout by \( \rho M_{C_s}/W_D \) to obtain

\[
\frac{\rho V}{W_D} = \rho \left( \lambda^{(m)} - \lambda^{(s)} \right) \frac{M_{N_v}}{W_D} \]

\[
+ \rho \left( \lambda^{(s)} - \frac{\lambda^{(m)}}{r^{(m)} - r^{(s)}} \right) \left( \frac{M_{C_s}}{W_D} \right) \tag{36}
\]

The slope is *strictly* constant and negative and the intercept is positive and *nearly* constant, depending on the constancy of \( M_{C_s}/W_D \) over time.

Equating eqns (35) and (36), results in

\[
A = \rho \left( \frac{\lambda^{(m)}}{r^{(m)} - r^{(s)}} \right) \tag{37}
\]

and

\[
B \left( \frac{W_D}{M_{C_s}} \right) = \rho \left( \lambda^{(s)} - \frac{\lambda^{(m)}}{r^{(m)} - r^{(s)}} \right) = \rho \lambda^{(s)} - A r^{(s)} \tag{38}
\]

which places two constraints on the four parameters \( r^{(m)}, r^{(s)}, \lambda^{(m)} \) and \( \lambda^{(s)} \). Hence, given just the line of Fig. 1, two of the four parameters remain free for fitting. Further information can be extracted from the two ends of the data cluster of Fig. 1 if it is assumed that the lower right data points represent young plants with effectively no support tissue and the upper left data points represent mature plants consisting *mainly* of support tissue. For these data, utilizing eqns (23) and (26) and assuming the validity of eqn (20) for the young plants (subscript \( y \)) and \( M_{C_s} \to \infty \) for the mature plants (subscript \( m \)), the result is

\[
\frac{M_{N_v}}{V} = \frac{r^{(m)}}{\lambda^{(m)}} \tag{39}
\]

\[
\frac{M_{N_v}}{V} = \frac{r^{(s)}}{\lambda^{(s)}} \tag{40}
\]

Since \( \rho V/M_{N_v} \) is the slope of lines radiating from the origin in Fig. 1, denoting this slope for the young plants by \( y \) and that for the mature plants by \( m \), eqns (37) to (40), can be used to evaluate the four ‘ontogenetic’ parameters as follows:

\[
\lambda^{(m)} = \frac{B}{\rho (1-A/y)} \left( \frac{W_D}{M_{C_s}} \right) \tag{41}
\]

\[
\lambda^{(s)} = \frac{B}{\rho (1-A/m)} \left( \frac{W_D}{M_{C_s}} \right) \tag{42}
\]

\[
r^{(m)} = \frac{B}{1-A/y} \left( \frac{W_D}{M_{C_s}} \right) \tag{43}
\]

\[
r^{(s)} = \frac{B/m}{1-A/m} \left( \frac{W_D}{M_{C_s}} \right) \tag{44}
\]

**Evaluation for the Beitem data**

The values of \( A, B, y \) and \( m \) can be read directly from Fig. 1, resulting approximately in

\[
A = -7 \text{ kg[water] mol}^{-1}[N]
\]

\[
B = 30 \text{ kg[water] kg}^{-1}[\text{DM}]
\]

\[
y = 5 \text{ kg[water] mol}^{-1}[N]
\]

\[
m = 38 \text{ kg[water] mol}^{-1}[N]
\]
The value of \( m \) is an underestimate, since even at harvest the metabolic component is not negligible. Estimating that its appropriate value is about \( m = 70 \text{[kg][water]} \text{[mol]}^{-1} \text{[N]} \), using \( \rho = 1000 \text{[kg][water]} \text{[m]}^{-3} \), and substituting into eqns (41) to (44), the mean results over the two cultivars and over all seasons become

\[
\lambda \text{[m]} = 0.013(W_D/M_{C_S}) \text{[m]}^3 \text{[water]} \text{[mol]}^{-1} \text{[C]}
\]

\[
\lambda \text{[s]} = 0.027(W_D/M_{C_S}) \text{[m]}^3 \text{[water]} \text{[mol]}^{-1} \text{[C]}
\]

\[
r \text{[m]} = 2.50(W_D/M_{C_S}) \text{[mol]} \text{[N]} \text{[mol]}^{-1} \text{[C]}
\]

\[
r \text{[s]} = 0.39(W_D/M_{C_S}) \text{[mol]} \text{[N]} \text{[mol]}^{-1} \text{[C]}
\]

The common factor, \( W_D/M_{C_S} \), which depends to some extent on model parameters and environmental conditions, increases, as the simulated crop grows, from about 0.04 to about 0.05 \( \text{[kg][DM]} \text{[mol]}^{-1} \text{[C]} \), since the increasing water content implies more soluble compounds per unit of structural carbon. Multiplying the metabolic parameters (superscript \( m \)) by 0.04 and the support parameters (s) by 0.05 \( \text{[kg][DM]} \text{[mol]}^{-1} \text{[C]} \), yields the values of the four parameters as shown in Table 2, columns 1 and 2.

**SIMULATIONS**

*Variable compositional ratios*

The model was fitted to the available data, by changing some of the original NICOLET parameters, selected according to previous experience with the model (Broadley et al., 2003; Linker et al., 2004). It turns out that the two cultivars require somewhat different values for two of the parameters, \( \varepsilon \) and \( \nu \), as shown in columns 1 and 2 of Table 2. Apparently, 'Troubadour' has a somewhat higher photosynthetic efficiency, \( \varepsilon \), and a somewhat lower partitioning to growth, \( \nu \), than 'Flandria'; however, the differences are rather small and may result from the partial confounding between cultivar and season. The values of the compositional ratios \( r \) and \( \lambda \) are the same for the two cultivars, as derived in the previous section from the common regression line of Fig. 1. All other parameters were the same for all simulations.

The results of the simulations are presented in Fig. 4, showing fair to good agreements between the measured and simulated values. The first two variables, dry and fresh mass, are cumulative and are predicted rather well. The last three variables are ratios, with noisier signals and looser fits. The logarithmic scale of the fresh mass and the linear scale of the dry mass together show how the initially exponential growth changes gradually into linear. The model was able to mimic the rather different shapes of the growth curves, resulting from different weather, cultivar and spacing. The prediction of nitrate content, the main focus of the original NICOLET model, is rather sensitive to short (daily) and medium term (weekly) environmental changes. In view of this, the fit to the various trends, including the nearly constant level of the winter experiment, should be considered as rather good.

The focus of this study is on the changes with time of the water and reduced-N content. The model predicts an increase with time of the former (decrease of DMC) and decrease of the latter. This is generally the trend of both data and simulation, but the details of the fit are less clear, partly because several data points are missing and partly because the scatter around the line in Fig. 1 is rather large. Figure 5 summarizes the simulations of water and reduced-N contents. Although the individual trajectories, starting at the lower right and ending at the upper left, do not closely agree with the data, the general behaviour in Fig. 5 is as expected.

*Comparison with fixed compositional ratios*

The question now is whether the new model produces better predictions than the original NICOLET model, where the water and reduced-N ratios were fixed (single \( r \) and \( \lambda \); original model). Selecting mean values for these ratios and adjusting slightly \( \varepsilon \) and \( \nu \) (Table 2), the results of Fig. 6 were obtained. The figure shows that the trajectories of dry mass, fresh mass and nitrate content, are generally similar to those
of Fig. 4. The dry matter and reduced-N contents, however, are rather different. The difference between the models is particularly striking when viewed in terms of the co-ordinates of Figs 1 and 5, as shown in Fig. 7.

Comparison of Figs 5 and 7 shows clearly that the model with fixed compositional ratios cannot capture the changes occurring in water and reduced-N contents as the crop grows.

**DISCUSSION**

**Reduced-N and water contents**

The new model, with ontogenetically decreasing reduced-N content and increasing water content, is a refinement of a previous model, NICOLET, which had been designed to predict growth and nitrate content of lettuce. The division of the ‘ontogenetic’ plant model, into ‘metabolic’ and ‘support’ tissue with high and low nitrogen contents, respectively, corresponds well with the morphology and nitrogen...
distribution of ‘ordinary’ crops. However, the extension of the concept to head lettuce and to water content can at the moment, while detailed data (of individual organs over the growing season) are not available, only be supported by the overall trends, such as shown in Fig. 1.

Figures 1 and 2 show the wide range of possible combinations of water and reduced-N content in lettuce. The negative correlation of Fig. 1 is due to ontogeny, while the positive correlation of Fig. 2 is due to N-stress. It may be of theoretical interest to explore what might be possible and impossible trajectories in the water versus reduced-N plane. The data of Fig. 1 are likely to mark the upper bound of the feasible region, while the data of Fig. 2 are likely to mark the lower bound.

Regarding the rate of ‘maturation’, in the sense of rate of N ‘dilution’ (moving from right to left in Fig. 1), this rate presumably depends on the rate of growth (faster growth leading to faster ‘dilution’). To some extent, however, it may also depend on plant spacing, isolated plants having
higher reduced-N content than dense canopies of similar age (Seginer, 2003b).

Perhaps the most noteworthy point regarding the ontogenetic model is the similarity between eqns (35) and (36). The first is an empirical linear relationship between water and reduced-N contents, and the second is the model-formulated relationship, which is approximately linear. The correspondence between the two expressions makes it possible to effectively uncouple the estimation of the ‘ontogenetic’ parameters $r^{(m)}$, $r^{(s)}$, $\lambda^{(m)}$ and $\lambda^{(s)}$ (utilizing Fig. 1), from the estimation of the other parameters.

Uptake of nitrogen

The new model has the potential advantage of estimating nitrogen uptake more accurately than before. Partitioning the total uptake into structural reduced-nitrogen and vacuolar nitrate, Nicolet treats the former strictly as a function of the size of the plant (as measured by $M_C$) and the latter as a function of both size and environment. Regarding reduced-N, the new version predicts uptake rates for mature plants which are lower than those predicted by the original model, since in the new model the mature plants recycle much of their structural nitrogen ($r^{(s)} < r^{(o)}$). On the other hand, the increasing water content ($\lambda^{(o)} > \lambda^{(s)}$), results in an increased demand for nitrate to maintain the required osmotic potential. Judging by the potentially large nitrate content of lettuce, changes in the environment may have a significant effect on the uptake of nitrogen.

The accuracy of the uptake calculations depends on the accuracy of the predicted size and nitrogen content of the crop. At the moment, as shown by Fig. 8 (which may be regarded as a cumulative version of Fig. 1), there is some disagreement between model and data with respect to reduced-N accumulation in the crop. According to the model, all the points of Fig. 8 should lie on a single curve, obtained by plotting eqn (25) (times $\rho$) against eqn (22). The data, however, show that two of the autumn points are considerably off the trend of the other three seasons (evident also in the reduced-N frames of Fig. 4). This may be blamed on the model (insufficient detail) and/or on the data (insufficient accuracy). An optimistic view would be to emphasize the good agreement of the data of three of the seasons, but definitely more data are required to establish a reliable set of the relevant parameters [$r^{(m)}$, $r^{(s)}$, $\lambda^{(m)}$, $\lambda^{(s)}$ and $a$].

Good estimation of the nitrate content is even more difficult, since it strongly depends on the environmental conditions. Considering the potentially high nitrate content of lettuce, the accuracy of its estimation should also be improved before the model can be used reliably for N-uptake calculations. For that, a good estimate of the ontogenetic change in water content is required.

Further points

The model was able to fit data of different seasons with the same set of (cultivar specific) parameters, even when the differences were as significant as between autumn and winter for the same cultivar (Troubadour; Fig. 4). The fit (Fig. 4) was generally good, despite significant differences in initial size of transplants (Flandria), in light efficiency (Flandria) and in final reduced-N content (Troubadour), as pointed out in conjunction with Table 1.

Comparison of Figs 4 and 6 shows that the new model does not improve significantly the prediction of growth and nitrate content of lettuce. It does, however, improve the prediction of water and reduced-N contents (Figs 5 and 7). Whether this improvement is useful to horticultural practice depends on the effect of these two attributes on lettuce quality and on the ability to make better nitrogen uptake predictions.

Most of the other tests of the Nicolet model have been against data from hydroponics (Broadley et al., 2003; Linker et al., 2004). The present data are of soil-grown lettuce which, however, was assumed to have a plentiful supply of nitrogen. The predictability for the soil-grown lettuce seems as good as for the hydroponics lettuce.

In conclusion, the new model is an improvement, as a comparison of Figs 5 and 7 demonstrates. It has the potential for improving the estimates of nitrogen uptake, thus leading to more accurate calculation of fertilizer needs and leaching potentials.

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LITERATURE CITED


Hansen H. 1976. The content of nitrate and protein in lettuce (Lactuca sativa var. capitata (butterhead lettuce)) grown under different conditions. Tidsskrift Planteavl 80: 370–380 [In Danish with summary, figures and tables in English].


APPENDIX A: DERIVATION OF EQN (27)

The time derivative of eqn (5) is

$$\frac{d\beta_c}{dt} = \frac{dM_{CV}}{dt} + \beta_N \frac{dM_N}{dt}$$

From (26) and (16), respectively

$$\frac{d\lambda}{df} = \left(\frac{m}{a_{MC}} - \frac{l}{a_{MC}}\right)$$

$$\frac{dM_{CV}}{dt} = a \exp\left(-aM_{CV}\right)$$

Using eqns (16), (26), (A2) and (A3) to substitute into eqn (A1), yields

$$\frac{dM_{CV}}{dt} \beta_c = \frac{dM_N}{dt} \beta_N$$

$$= \Pi \left(\left[\frac{m}{a_{MC}} - \frac{l}{a_{MC}}\right] \left\{M_{CV}\right\} \frac{dM_{CV}}{dt}\right)$$

Substituting from eqns (1), (2), (3) and (12) into eqn (A4)

$$\beta_c \left(F_{CP} - F_{CM}\right) - \beta_c (1 + \theta) F_{CVS} + \beta_N F_{Nav}$$

$$= \frac{\left(r_m - r_s\right)}{(1 - f) + \left(r_s\right)} F_{CVS}$$

$$= \Pi \left[\left(\frac{m}{a_{MC}} - \frac{l}{a_{MC}}\right) \left\{M_{CV}\right\} \frac{dM_{CV}}{dt}\right]$$

$$= \frac{\beta_c (1 + \theta) + \beta_N \left(r_m - r_s\right)(1 - f) + \left(r_s\right)}{(1 - f) + \left(r_s\right)}$$

$$= F_{CVS} - \beta_N F_{Nav}$$

or

$$\frac{\left(r_m - r_s\right)}{(1 - f) + \left(r_s\right)} F_{CVS} - \beta_N F_{Nav}$$

$$= \beta_c \left(F_{CP} - F_{CM}\right)$$

where

$$G \left(F_{CVS}\right) = \frac{\beta_c (1 + \theta) + \beta_N \left(r_m - r_s\right)(1 - f) + \left(r_s\right)}{(1 - f) + \left(r_s\right)}$$

(A7)

(A8)
## APPENDIX B: NOTATION

### MAIN SYMBOLS

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>SI units</th>
</tr>
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<tbody>
<tr>
<td>$A$</td>
<td>slope of data line in Fig. 1</td>
<td>kg[water] mol$^{-1}$[N]</td>
</tr>
<tr>
<td>$a$</td>
<td>light extinction coefficient</td>
<td>m$^2$[ground] mol$^{-1}$[C]</td>
</tr>
<tr>
<td>$B$</td>
<td>intercept of data line in Fig. 1</td>
<td>kg[water] kg$^{-1}$[DM]</td>
</tr>
<tr>
<td>$b_k$</td>
<td>attenuation border of process $k$</td>
<td>–</td>
</tr>
<tr>
<td>$C_C$</td>
<td>CO$_2$ concentration in the air</td>
<td>mol[C] m$^{-3}$[air]</td>
</tr>
<tr>
<td>$c$</td>
<td>exponent in respiration equation</td>
<td>K$^{-1}$</td>
</tr>
<tr>
<td>$e$</td>
<td>maintenance respiration rate of a closed-canopy crop</td>
<td>mol[C] m$^{-2}$[ground] s$^{-1}$</td>
</tr>
<tr>
<td>$F_{Ck}$</td>
<td>carbon flux due to process $k$</td>
<td>mol[C] m$^{-2}$[ground] s$^{-1}$</td>
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<tr>
<td>$F_{p}$</td>
<td>flux of constituent $J$ from compartment $i$ to compartment $j$</td>
<td>mol[J] m$^{-2}$[ground] s$^{-1}$</td>
</tr>
<tr>
<td>$F_{Na}$</td>
<td>uptake rate of nitrogen</td>
<td>mol[N] m$^{-2}$[ground] s$^{-1}$</td>
</tr>
<tr>
<td>$f$</td>
<td>fraction of light intercepted by canopy</td>
<td>–</td>
</tr>
<tr>
<td>$G$</td>
<td>a collection of parameters</td>
<td>m$^3$[ground] Pa mol$^{-1}$[C]</td>
</tr>
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<td>$g$</td>
<td>uninhibited growth of closed canopy</td>
<td>mol[C] m$^{-2}$[ground] s$^{-1}$</td>
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<tr>
<td>$b_k$</td>
<td>attenuation function of process $k$ when no attenuation</td>
<td>–</td>
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<tr>
<td>$I$</td>
<td>light flux</td>
<td>mol[PAP] m$^{-2}$[ground] s$^{-1}$</td>
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<td>maintenance respiration rate at $T = T^*$</td>
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<td>$m$</td>
<td>slope of mature-plants line in Fig. 1</td>
<td>kg[water] mol$^{-1}$[N]</td>
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<td>$M_{ij}$</td>
<td>molar mass of constituent $J$ in compartment $j$</td>
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<td>$T^*$</td>
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<tr>
<td>$t$</td>
<td>time</td>
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<tr>
<td>$V$</td>
<td>volume of water</td>
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<td>$W_D$</td>
<td>mass of dry matter of crop</td>
<td>kg[DM] m$^{-2}$[ground]</td>
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<td>mass of fresh matter of crop</td>
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<td>$\beta_J$</td>
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<td>m$^3$[water] Pa mol$^{-1}$[J]</td>
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<td>$\Gamma$</td>
<td>normalized carbon concentration in vacuole</td>
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<td>$\varepsilon$</td>
<td>photosynthetic efficiency</td>
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<td>$\eta_D$</td>
<td>dry-mass equivalent of model components</td>
<td>kg[DM] mol$^{-1}$[J]</td>
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<td>growth respiration as fraction of growth</td>
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<td>$\lambda$</td>
<td>water associated with unit of structure</td>
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<td>$\nu$</td>
<td>ratio $g:e$</td>
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<tr>
<td>$\Pi$</td>
<td>osmotic potential in vacuole</td>
<td>Pa</td>
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<tr>
<td>$\sigma$</td>
<td>leaf conductance to CO$_2$</td>
<td>m/s</td>
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<tr>
<td>$\rho$</td>
<td>density of water</td>
<td>kg[water] m$^{-3}$[water]</td>
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### SUBSCRIPTS

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<td>u</td>
<td></td>
<td>uptake</td>
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### SUPERSCRIBTS

| (m) | metabolically active component of plant |
| (s) | support component of plant |

### Acronyms

- **DAP** days after (trans)planting
- **DM** dry matter
- **DMC** dry matter content
- **FM** fresh matter
- **PAP** photosynthetically active photons

### Notes

- indicates dimensionless quantities
- used exclusively to contain the arguments of functions.