A Simple Mass Balance Model for Lettuce – The Water Balance

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Abstract: A simple mass balance model has been developed and tested on lettuce data. A water balance was included in order to predict important fluxes (transpiration, water uptake, etc.) and to consider interactions between the water variables and the metabolism of the plant. A two-stage approach, in which a unique set of yield constants were identified for each stage, was successful in predicting water uptake, carbon dioxide and oxygen concentrations, and final biomass dry weight.

Keywords: Dynamic modelling, parameter identification, biotechnology, identifiability, validation

1. INTRODUCTION

Recent work on the subject of plant growth modelling has largely been focused on making models more closely represent our current knowledge of physiology. This work is very important for furthering our understanding of plant growth and development. However, this progress towards more mechanistic descriptions has resulted in complicated models with large numbers of parameters (Zhu *et al.*, 2007; Laisk *et al.*, 2000; Karlberg *et al.*, 2006). There is also a need for simplified models which focus on model and parameter reliability. This is particularly important when building models for prediction and control, and when dealing with limited datasets, since very often the available data is insufficient to identify a large number of parameters.

One application that requires this type of model is the production of plants in a closed environment system, as is envisioned for regenerative life support systems in space. The MELiSSA (Micro-Ecological Life Support System Alternative) project, developed by the European Space Agency, aims to develop technology for such a system (Godia *et al.*, 2002). The concept is to use microorganisms and plants to regenerate the atmosphere, produce food for the crew, and to contribute to the recycling of some wastes. As part of this work, a higher plant compartment has been designed. Dynamic models of plant growth are required for the monitoring and control of the chamber, and also to facilitate the integration of this compartment into the larger life support system. The control objective will be to ensure a certain desired flow of high quality edible biomass, however other fluxes, including carbon dioxide, oxygen, water and nutrients, should also be predicted.

A simple mass balance model for plant growth has therefore previously been developed and validated on lettuce and beet data from closed environment experiments (Maclean *et al.*, 2011a). The model is fairly successful at predicting carbon dioxide, oxygen and biomass dry weight over the experiments, but does not predict water fluxes, such as transpiration and water uptake, which will be important for

the life support system. The water balance is also highly integrated in the plant's functioning. Therefore, it is expected that the integration of the water balance and consideration of linked environmental factors, such as humidity, will improve the overall model functioning.

Many models that consider the effect of water on plant growth focus on the effect of water limitation (Tardieu *et al.*, 1993; Thornley, 1996) and the influence of humidity on plant metabolism through stomatal conductance (Tardieu *et al.*, 1993; Leuning, 1995; Ball *et al.*, 1987). Stomata are small pores on the surface of leaves through which most of the plant's gas exchange with the atmosphere occurs. Plants need to take up carbon dioxide from the atmosphere while mitigating excessive water loss. This tradeoff is managed by controlling the opening and closing of stomata. Stomatal functioning is influenced by environmental factors such as humidity and carbon dioxide concentration, as well as plant CO₂ requirements (Ball *et al.*, 1987).

In this work, a plant water balance is derived to extend and improve an existing mass balance model of plant growth. The effect of humidity on growth is considered, assuming no water limitation. The approach stresses the importance of maintaining model identifiability, and therefore additions to the model will be evaluated based on the identifiability of parameters and the added predictive value.

2. EXPERIMENTAL DATA

2.1 Experimental Set-Up

Lettuce (*Lactuca sativa* cv. Lively) and red beet (*Beta vulgaris* cv. Detroit Medium Red) experiments performed at the University of Guelph were used for model development. Plants were germinated in a research plant growth room using Rockwool® cubes. The seedlings were moved to a sealed environment plant growth chamber when there was sufficient root exposure to facilitate transplanting to a deep water hydroponic system. The plants remained in the chamber for a growth period of 21 days (lettuce) or 40 days (beets).

Cultivation conditions were designed to be the same in both experiments. In each experiment 120 plants were grown inside the chambers. A 14/10 h light/dark photoperiod, coupled to a 25/20 °C day/night temperature, was used. Light was provided by 9 high pressure sodium and 6 metal halide lamps, which provided approximately $600 \mu\text{mol m}^{-2} \text{s}^{-1}$ photosynthetically active radiation (PAR) at stand height. Atmospheric CO_2 concentration was controlled at a minimum value of 1000 ppm (it was allowed to increase at night). Relative humidity was intended to be controlled at 70%, however in one lettuce experiment a problem with the control led to lower values. Oxygen concentration in the chamber was not controlled. Therefore, initial O_2 concentration was approximately 21 % (atmospheric) and increased throughout the experiments. The nutrient solution for the hydroponic system was replaced every 5 days, and had the following composition: 1.5 mM PO_4^{3-} , 3.62 mM Ca^{2+} , 4 mM $\text{NH}_4^+\text{-N}$, 11.75 mM $\text{NO}_3\text{-N}$, 5 mM K^+ , 2 mM SO_4^{2-} , 1 mM Mg^{2+} , 0.005 mM Mn^{2+} , 0.025 mM Fe^{3+} , 0.0035 mM Zn^{2+} , 0.02 mM B^{3+} , 0.008 mM Na^+ , 0.0008 mM Cu^{2+} , 0.0005 mM Mo^{6+} .

2.2 Data Availability / Measurements

The following measurements were recorded automatically every 6 minutes in the chamber: CO_2 concentration, the amount of CO_2 added to the chamber to maintain the control set point, light measurements taken by PAR sensors at canopy height, relative humidity, evapotranspiration (condensate collected for control of relative humidity), and temperature. O_2 concentration was measured periodically (every 6 minutes with measurements alternating ON for 6 hours, OFF for 6 hours due to chamber set-up). Average concentrations were calculated for periods over which the data recording was off.

Water and nutrient uptake by the plants was estimated approximately every 5 days (upon changing the nutrient solution), by measuring the loss in volume of the solution provided and the change in concentration of nutrients. Leaf area and total biomass fresh and dry weights were measured on 15 seedlings which were grown together with seedlings transferred to the chamber. At the end of the study all plant material was harvested. Measurements of leaf area, total fresh and dry weights, as well as fresh and dry weights by organ (leaves and roots) were taken for each plant.

3. THE SIMPLE PHOTOSYNTHESIS MODEL

The model is intended to be used to control the production of biomass in a closed chamber environment and should therefore be as simple and reliable as possible while still capturing the main features of plant growth. With this objective in mind, photosynthesis (1), photorespiration (2) and mitochondrial respiration (3) were selected as the most important reactions to consider for their influence on biomass production.

From this reaction scheme, a dynamic mass balance model was written ((4)-(8)) with mass balance equations for biomass dry weight, carbon dioxide and oxygen (Maclean *et al.*, 2011a).

$$\frac{dM_d}{dt} = Y_1 r \quad (4)$$

$$\frac{dC_a}{dt} = \frac{-r}{V_{\text{chamber}}} + \frac{u_1}{V_{\text{chamber}}} \quad (5)$$

$$\frac{dO_a}{dt} = \frac{Y_2 r}{V_{\text{chamber}}} \quad (6)$$

$$r = v_1 C_a I_{\text{intercepted}} - v_2 O_a I_{\text{intercepted}} - v_3 r_{\text{avg, ps-pr}} \quad (7)$$

$$I_{\text{intercepted}} = I_0 (1 - \exp(-k A_{\text{leaf}} / A_{\text{ground}})) \quad (8)$$

In the above equations, M_d is biomass dry mass (g), C_a and O_a are CO_2 and O_2 concentrations in the atmosphere of the chamber (g m^{-3}), r is the reaction rate equation (defined in (7)) with the three terms representing the rates of photosynthesis, photorespiration, and mitochondrial respiration respectively), V_{chamber} is the volume of the plant growth chamber (29 m^3), u_1 is the rate of CO_2 addition to the chamber for control (g s^{-1}), $r_{\text{avg, ps-pr}}$ is the average rate of photosynthesis less photorespiration over the previous 1 day period (g s^{-1}), $I_{\text{intercepted}}$ and I_0 are the intercepted and incident (at canopy height) photon fluxes ($\mu\text{mol PAR m}^{-2} \text{s}^{-1}$), k is the extinction coefficient (0.66, as found in Tei *et al.* (1996)), A_{leaf} is the leaf area (m^2 , estimated as proportional to M_d in the model), A_{ground} is the planting area (5 m^2), v_i are the kinetic rate constants and Y_i are the yields (g g^{-1}). The yields, which relate the rate terms for each of the state variables, should be constants over the full experiments. However, it was found in previous work (Maclean *et al.*, 2011b) that Y_2 decreases with time, and therefore the yields could not be considered constant.

Therefore, a two-stage approach was developed to represent the changing metabolism of the plant without adding unnecessary complexity. Two distinct growth stages, each stage having a unique set of constant yields, were detected using the following condition for the transition between stages:

$$\text{Transition if} \quad \frac{d}{dt} (\Delta_{\text{flux}}) < \alpha \text{ for } \beta \text{ hours and } |\Delta_{\text{flux}}| > \gamma \quad (9)$$

Where Δ_{flux} (mol) is the difference between the moles of oxygen produced and carbon dioxide consumed, and α , β and γ are constant parameters such that $\alpha = 6.603 \times 10^{-6} \text{ mol s}^{-1}$, $\beta = 0.646 \text{ days}$, $\gamma = 3.937 \text{ mol}$ (Maclean *et al.*, 2011b).

4. THE WATER BALANCE

A water balance should be included in the model to predict important fluxes for the life support system and to improve model predictions. Therefore, a mass balance on water (10) was derived and added to the original model ((4)-(8)).

$$\frac{dW_p}{dt} = -Y_3 r - E + U \quad (10)$$

In the equation above, W_p is the water stored in the plants (g), E is the rate of transpiration (g s⁻¹, to be defined), U is the water uptake rate (g s⁻¹, to be defined), Y_3 is a new yield parameter, and r is again the reaction rate term (7).

Several terms in the water balance require further definition. The transpiration rate (E) can be considered a standard transfer term (11) and is driven by a vapour density gradient ($\rho_{vi} - \rho_{va}$ (g m⁻³) where ρ_{vi} is the vapour density inside the leaf and ρ_{va} is the density in the atmosphere of the chamber).

$$E = g_w A_{leaf} (\rho_{vi} - \rho_{va}) \quad (11)$$

The constant g_w is a conductance parameter which could depend on environmental factors affecting the boundary layer and also on stomatal conductance.

Water uptake is driven by the difference in the water potential (ψ – potential energy of water per unit volume relative to pure water at reference conditions) between the roots or hydroponic solution and the leaves (Thornley *et al.*, 2000). Water potential is difficult to estimate accurately, and would require measurements that are not available for this dataset. Therefore, the water balance was rearranged, so that uptake would be predicted by solving the equation (12).

$$U = \frac{dW_p}{dt} + Y_3 r + g_w A_{leaf} (\rho_{vi} - \rho_{va}) \quad (12)$$

With this modification, the water storage in plants (W_p) must be modelled. The amount of water that a plant stores but does not metabolise depends mainly on the water and nutrient availability. However other factors, such as the carbon assimilation rate and related environmental conditions, can also have an effect (Seginer, 2003). Given that the experiments were performed under non-limiting nutrient and water conditions, it was assumed that the water in the plant could be correlated to biomass dry weight by a constant ω . Based on these assumptions and considerations, the water balance was rewritten in its final form as (13).

$$U = (\omega Y_1 + Y_3) r + g_w A_{leaf} (\rho_{vi} - \rho_{va}) \quad (13)$$

5. MODEL IDENTIFIABILITY & YIELD IDENTIFICATION

Now that the model equations have been fully defined, the next step is to identify model parameters, but first we must test whether the model is structurally identifiable. Model identifiability is an important consideration in building a reliable model, and an often overlooked issue in plant modelling. Testing for structural identifiability asks whether, based on the structure of the model, all parameters can be given unique values (Dochain, 2008).

The model elaborated in (4) – (8) and (13) has 3 yield parameters (Y_i), 3 kinetic parameters (v_i), an extinction coefficient (k), and two new parameters from the water balance (ω and g_w). Three of the four states can be assumed to be known (C_a , O_a and U), since these variables are measured with time. In this case, an analysis of the terms reveals that ω and Y_3 cannot be identified uniquely, since

they do not occur independently in the model. Y_1 is also unidentifiable since biomass dry weight measurements are only available at the beginning and end of the experiment (and are therefore not available with time). This shows that even for a simple model, with relatively few parameters and many measured variables, the identifiability of the model should not be taken for granted.

We can improve the identifiability of the model by taking into account the unique model structure, which allows us to identify the yields separately from the other parameters. This approach, demonstrated by Chen (1996) uses a state transformation (Bastin *et al.*, 1990) to transform the model into one which does not depend on the reaction kinetics. The resulting equations are shown below:

$$\frac{dM_d}{dt} = Y_1 \left(u_1 - V_{chamber} \frac{dC_a}{dt} \right) \quad (14)$$

$$\frac{dO_a}{dt} = Y_2 \left(\frac{u_1}{V_{chamber}} - \frac{dC_a}{dt} \right) \quad (15)$$

$$\frac{dW_p}{dt} = Y_3 \left(V_{chamber} \frac{dC_a}{dt} - u_1 \right) + U - E \quad (16)$$

For each of these equations all of the data required to identify the yields is available, however the frequency of measurement varies. For example, the measurements required to solve for Y_2 (15) were taken every 6 minutes, while M_d and W_p were only measured at the start of the experiment and at harvest and therefore Y_1 and Y_3 can only be identified based on two datapoints per experiment (where one is essentially a zero point). However, the data was sufficient to perform a least squared identification to identify the yields. It should be noted that in the two-stage approach, a unique value of Y_2 is identified for each stage by performing an identification which solves for values of the parameters associated with the transition condition (9) and Y_2 together (described in detail in Maclean *et al.*, 2011b). Y_1 and Y_3 could not be identified by the same approach since limited data was available on biomass dry weight and water storage in the plant. Therefore, Y_1 was estimated assuming a constant yield of biomass on oxygen over the full experiment, while Y_3 was estimated assuming a constant yield of water on CO₂ (assumptions were made based on available data).

This approach greatly simplifies the structural identifiability analysis, as these constants can now be considered known. After this simplification, it is clear that all the parameters should be structurally identifiable based on the data available.

Another important consideration is practical identifiability, which considers whether the quality of the data is adequate to identify unique values of the parameters (Dochain, 2008). It has been shown (work not included) that even with this limited number of parameters, several are highly correlated. Therefore, to improve the confidence intervals, some additional knowledge on the values of the parameters was taken into account. Tei *et al* (1996) performed experiments on lettuce, red beet and onion and calculated the extinction coefficients directly from measured data. They found an extinction coefficient of 0.66 ± 0.22 for lettuce, which agrees well with ranges reported in the literature (Thornley *et al.*,

2000). Therefore, the extinction coefficient will be treated as a known constant in the model. Similarly, we can calculate an approximate ω directly from harvest measurements taken in the experiments. Therefore, ω was taken to be a known constant of $11.74 \text{ gW}_p \text{ gM}_d^{-1}$.

6. EFFECT OF RELATIVE HUMIDITY ON PLANT GROWTH

Water variables also have a direct impact on growth. The current model only considers the effect of light, CO_2 and O_2 concentrations on the metabolism of the plant (as shown in (7)). The water status of the plant and the relative humidity should also have an important effect. However, the inclusion of the water status was considered unnecessary for modelling the available data at this time because water was provided continually through a hydroponic system and is therefore assumed to be non-limiting for growth.

The main environmental difference between the two datasets was in the relative humidity (Figure 1). The effect of relative humidity on transpiration has already been taken into account in the water balance. However, relative humidity also impacts the growth of the plant through its effect on stomatal conductance. The opening and closing of stomata (pores through which gas exchange occurs) is the plant's main method of managing the trade-off between their CO_2 requirements and avoiding excess water loss and can therefore have a large impact on the concentrations of carbon dioxide and oxygen available at the sites of photosynthesis and respiration reactions.

It was hypothesized that by accounting for this seemingly important effect on plant growth, the model could be improved. Several methods were tested. Theoretically, separate mass balance equations could be written for the carbon dioxide and oxygen concentrations inside the leaves (in addition to the balances on CO_2 and O_2 in the atmosphere, which are currently included). However, no data was available to validate internal gas concentrations, and therefore a simpler approach was taken. As a first test, it was assumed that the most important effect of humidity was its influence on CO_2 availability (effect on O_2 availability was also considered, but results are not included here). Therefore, the rate equation was amended as shown in (17).

$$r = v_1 C_a f_c I_{\text{intercepted}} - v_2 O_a I_{\text{intercepted}} - v_3 r_{\text{avg}, ps-pr} \quad (17)$$

In the above equation, f_c is a factor which acts on C_a to give an approximation of the internal CO_2 concentration (C_i). Several different equations for f_c were tested (see Table 1). The first two equations were derived by making some assumption about stomatal conductance. In the first case it was assumed that stomatal conductance for CO_2 transport could be related to the conductance for water transport (g_w) by a constant p_1 , and therefore the conductance could be approximated from the transpiration equation (Lambers *et al.*, 2008). In the second case a Ball-Berry-type model for stomatal conductance was assumed (Ball *et al.*, 1987). The remaining two cases are simply empirical curves which were derived based on the known boundary conditions ($C_i = C_a$

Fig. 1. Relative humidity for two lettuce experiments.

Table 1: Definitions of f_c for several cases.

Case	f_c	Description
0	1	Original model - no effect of humidity on r .
1	$1 - A\Delta\rho_{va} / (p_1 EC_a)$	Assumes $g_c = p_1 g_w$
2	$1 - A / (C_a A_{leaf} g_c)$ where $g_c = p_1 + \frac{p_2 A h_a}{C_a}$	Ball-Berry-type model for stomatal conductance (Ball <i>et al.</i> , 1987)
3	$1 - \Delta\rho_{va} / (\Delta\rho_{va} + p_1)$	Empirical
4	$(1 + p_1) / (1 + \lambda e^{p_2 \Delta\rho_{va}})$	Empirical (logistic)

A is the net CO_2 assimilation rate (g s^{-1}), h_a is the relative humidity in the atmosphere of the plant chamber (%), and p_1 and p_2 are new constant parameters to be identified. All other variables are as defined previously.

when $\Delta\rho_{va} \rightarrow 0$ and $C_i \rightarrow 0$ when $\Delta\rho_{va} \rightarrow \infty$). Only the two-stage approach was tested with the modified rate equation (17) since this amendment will have no impact on the yields. The new parameters introduced will not affect the structural identifiability of the model, but may affect practical identifiability, as discussed below.

7. PARAMETER IDENTIFICATION AND MODEL VALIDATION

Parameters (v_1 , v_2 , v_3 , g_w , and parameters associated with f_c where applicable) were identified by minimizing the weighted sum of squared errors (SSE) between measurements and model predictions on C_a , O_a and U , where $\hat{\theta}$ are the parameter estimates, N_i represents the number of measurements of each variable (required since uptake is measured less frequently than CO_2 and O_2), and μ_i are the mean values of the measured states:

$$\begin{aligned} SSE = & \frac{1}{N_c} \sum_i \frac{1}{\mu_c^2} (C_{a,i} - C_{a,i}(\hat{\theta}))^2 \\ & + \frac{1}{N_o} \sum_j \frac{1}{\mu_o^2} (O_{a,j} - O_{a,j}(\hat{\theta}))^2 \\ & + \frac{1}{N_u} \sum_k \frac{1}{\mu_u^2} (U_k - U_k(\hat{\theta}))^2 \end{aligned} \quad (18)$$

For each case, the parameters were identified on one dataset, and then validated on another. This procedure was then repeated, switching the identification and validation datasets, for further validation of the model. Note that vapour density

inside the leaves (ρ_a) is estimated as the saturated vapour density at atmospheric temperature. The results from the analysis (prediction errors are shown in Table 2) do not show a clear overall improvement for any of the four cases in comparison to the original model. The addition of new parameter(s) to the model also decreases practical identifiability (increased confidence intervals and/or decreased sensitivity). Overall, it was concluded that accounting for the effect of humidity on the metabolism of the plant made no significant improvement to the two-stage model, and thus the factor f_C was excluded.

Table 2: Weighted sum of squared errors from parameter identification (SSE_i) and validation (SSE_v) for several definitions of f_C (see Table 1). Trial 2 shows results when identification and validation datasets in were exchanged.

	Trial 1		Trial 2	
	SSE_i	SSE_v	SSE_i	SSE_v
Case 0 (original)	7.71E-03	1.01E-02	3.33E-02	3.62E-02
Case 1	7.71E-03	9.49E-03	3.33E-02	5.35E-02
Case 2	5.55E-03	5.06E-02	1.02E-02	3.02E-02
Case 3	6.83E-03	1.01E-02	4.34E-02	3.53E-02
Case 4	6.78E-03	1.01E-02	4.16E-02	3.61E-02

There are several potential explanations for these results. Firstly, it is possible that the data available was not sufficient to identify an effect due to relative humidity. It is possible that with additional data an effect of relative humidity on the rate equation would become important and practically identifiable.

Another possibility is that the two-stage approach itself is capturing some of the effect of changes in environmental parameters. For example, for the two lettuce datasets available, the yield of oxygen on carbon dioxide changed (signalling the transition to the second stage of growth) at quite different times. This change was detected through carbon dioxide and oxygen measurements (9) rather than being predicted. However, it is clear that environmental factors should contribute to metabolic changes. From an examination of the lettuce data available, it was hypothesized that higher relative humidity values might lead to an earlier transition to the second stage of growth (with higher CO_2 consumption relative to O_2 production). To investigate this further, we can compute a developmental variable, h (19), which represents the developmental progress in terms of some environmental variable (x) (Thornley et al., 2000).

$$h = \sum_{i=1}^j [(x_i - x_0)] \quad (19)$$

In this equation x_0 is a base value, below which development is assumed to be zero. Typically, this method is used for calculating a temperature sum ($h = \sum_i (T_i - T_0)$), since temperature is usually the most important factor for development (especially under field conditions). However, in our case temperature is well controlled and follows the same

profile for each experiment, and therefore its inclusion is not necessary. However, using the same approach, a developmental variable based on the vapour density deficit ($\Delta\rho_{va}$) and a ratio from Ball-Berry's (Ball et al., 1987) model for stomatal conductance (Ah_s/C_s) can be derived which may be useful in determining whether humidity or stomatal functioning influences development.

Figure 2 shows the results of this analysis. In this figure Δ_{flux} (the difference between moles of O_2 produced and CO_2 consumed) is plotted against time and the two potential measures of development discussed above. This variable can be used to detect the transition from the first to second stage of growth (9). Therefore, the more similar the curves from different experiments are, the more likely that the developmental variable being tested is a good measure for development.

Fig. 2. Δ_{flux} variable (used to detect transition between stages) plotted against time and two developmental variables ($\Delta\rho_{va}$ sum and g_c sum) for two lettuce datasets (red and blue lines)

When Δ_{flux} is plotted against time (Figure 2), the curves are quite different for the two datasets. Therefore the transition between the two stages does not occur at the same time and so time is not a good developmental variable. This is to be expected since some environmental variables (most notably humidity) are quite different between the two datasets. When humidity is taken into account through the vapour density deficit ($\Delta\rho_{va}$ sum on the x-axis), the curves are more similar, and finally, when the ratio employed in Ball-Berry's model of stomatal conductance is used (g_c sum), the curves match very closely. This suggests that the changes in metabolism are somehow linked to stomatal conductance (possibly through internal CO_2 concentration, which may affect metabolism), however at this stage, this hypothesis is rather speculative and should be further tested with additional data. If this hypothesis holds, it would theoretically be possible to predict the transition to the second stage of growth by monitoring this ratio for stomatal conductance. However, for our application, this prediction is unnecessary as we should have online data for CO_2 and O_2 that we can monitor directly. Therefore, by monitoring these important metabolic outputs, we can inherently account for some changes to metabolism due to environmental variables without explicitly taking these interactions into account. This is an important benefit of the two-stage approach.

Therefore, although relative humidity does seem to effect growth, through its influence on the yields, the two-stage approach inherently captures this effect. Figure 3 shows the validation of the original model with the water balance.

Results from both the single-stage and two-stage approaches are shown for comparison.

Fig. 3. Results from validation on independent measured data (black) using single stage (predictions in blue) and two-stage (predictions in red) model with water balance.

The results demonstrate that the two-stage model accurately predicts carbon dioxide, oxygen, water uptake and final biomass dry weight. The two-stage approach improves the prediction of water uptake compared to the single-stage method, due largely to an improved prediction of leaf area (which is estimated based on biomass dry weight). Overall, the parameter values for the rate constants of the two stage model (v_1 , v_2 , v_3) were essentially unchanged by the addition of the water balance and water cycle data, which suggests that the growth model is quite robust.

8. CONCLUSIONS

The approach to model development proposed here, which focuses on identifiability and validation on independent data, is a useful method for developing simple reliable models and is particularly important when dealing with limited datasets. By using this approach, we have developed a simple mass balance model able to predict biomass dry weight, CO_2 and O_2 concentration, as well as water uptake by the plant accurately for two lettuce experiments. Any additional model complexity would likely lead to identifiability problems unless additional data is used for model development.

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