

## Building Virtual Chrysanthemum Based on Sink-Source Relationships: Preliminary Results

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### Abstract

Quality aspects of cut chrysanthemum, such as weight of the plant and number and size of flowers, have been widely studied. However, these are seldom integrated in a single model. A functional-structural model, GreenLab, was used to simulate the geometrical features of chrysanthemum with underlying rules on biomass production and allocation. In this paper, model calibration was conducted based on data from a climate room experiment. Chrysanthemum 'Reagan Improved' was grown at 16°C and 380  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR, with 14 days long day (LD) period (19 h of light), followed by short day (SD) period (11 h of light) until harvest. Detailed measurements included weight and size of leaves and internodes in the main stem, and diameter of flowers, weight of all leaves and that of stems for side shoots. Non-linear least square method was applied to fit the parameters of the GreenLab model, such as the sink strength of the organs. New features introduced into GreenLab are: (1) number of primordia to fill the gap between development and growth; (2) delay function of growth to simulate the top-down flowering sequence; (3) additional sink of growing internodes on main stem to simulate their strong growth after terminal-bud removal; (4) two phases for individual flower growth. Realistic 3D chrysanthemum was simulated, which is a starting point for introducing effects of varying cultivation conditions.

### INTRODUCTION

Empirical models have been developed for chrysanthemum which attempt to describe and predict the development of flowers from flower initiation to anthesis as influenced by daily light integral and temperature (Karlsson et al., 1989; Hidén and Larsen 1994; Pearson et al., 1993). Lee and Heuvelink (2003) present an explanatory photosynthesis-driven model for chrysanthemum, simulating leaf area development based on dry matter partitioning, which was described as a static empirical function. External qualities like flower number and size, plant height and weight, are hardly studied simultaneously (Carvalho et al., 2005) and there is no model linking plant production with 3D-architecture. However, integration of photosynthesis-driven models and architectural models is highly relevant especially for modeling visual quality of ornamental crops like chrysanthemum, where plant architecture is essential (Heuvelink et al., 2004).

GreenLab (Yan et al., 2004) is a functional-structural plant model (FSPM), simulating dynamically the development of plant architecture, biomass production and allocation, the weight and size of individual organs and also the 3D geometrical shape. It has been calibrated for single-stem crops, e.g. maize (Guo et al., 2006). Many papers related to GreenLab have been published in Hu and Jaeger (2003). The aim of the present work is to build a virtual 3D cut chrysanthemum using GreenLab. Some features were introduced to deal with the branching inflorescence structure in chrysanthemum. In this paper, preliminary results of GreenLab calibration at one constant climate condition and 3D-simulation for cut chrysanthemum are presented.

## MATERIAL AND METHODS

### GreenLab Model

The GreenLab model follows the development and growth process of real plants in a discrete way. In each cycle, a new metamer is created as long as organogenesis has not finished. The new and old growing organs (leaves, internodes, flowers) share the assimilates from a common pool. Their competition ability is determined by the relative sink strength of each organ type, and its variation in time, the variation function being described with a Beta law (Yan et al., 2004). Accumulation of obtained biomass results in the weight of an organ. From the weight its size is computed. With the area of each individual leaf known, the biomass production  $Q(i)$  (g) in cycle  $i$  is computed with the following formula (Guo et al., 2006):

$$Q(i) = \frac{E(i)S_p}{r_1 r_2} \left[ 1 - \exp\left(-r_2 \frac{\sum_j s_j}{S_p}\right) \right] \quad (1)$$

$E(i)$  is the potential biomass production (g/g H<sub>2</sub>O) during cycle  $i$ , which can depend on environmental variables,  $S_p$  (cm<sup>2</sup>) is the ground-projected leaf area of the plant,  $s_j$  (cm<sup>2</sup>) is the blade surface of the  $j$ -th living leaf on the plant,  $r_1$  (cm<sup>2</sup>/g H<sub>2</sub>O) and  $r_2$  are empirical parameters,  $r_1$  being the transpiration resistance and  $r_2$  the light extinction coefficient.

Topological parameters mainly include number of buds, leaves, flowers per metamer, and number of metamers in stems, which have clear botanical meaning, and can be observed directly from the plant structure. The number of organs of each type and each age is computed for each cycle. Some parameters, like the functioning duration of leaves, and the expansion duration of organs, are more elaborate to determine. Others can not be estimated directly from the data, like relative sink strength of organs, and the empirical parameters  $r_1$  and  $r_2$ . These values are determined by calibration, fitting the model output with the measured data on the plant architecture.

In fitting with data of chrysanthemum, the following aspects were considered.

**1. The Gap between Development and Growth.** Being a SD (short day) plant, chrysanthemum finishes initiation of new leaves a few days after the start of SD period (Adams et al., 2001). Top meristems are not able to be distinguished or measured until visible bud stage. To simulate the development of plant structure properly, number of primordia in the plant tip,  $n_p$ , is introduced as a new parameter, and organs younger than  $n_p$  are assumed to have neglectable sink strength. The sink strength  $s$  of an organ  $o$  with age  $j$  is thus described as:

$$s = \begin{cases} \varepsilon & j \leq n_p \\ S \varphi(j - n_p) & j > n_p \end{cases} \quad (2)$$

$S$  is the relative sink strength of an organ, being 1 for leaves on the main stem.  $\varphi$  is the Beta law, described by Yan et al. (2004).  $n_p$  is estimated directly from observations, see Results section.  $\varepsilon$  is a neglectable value compared to  $S$ .

**2. The Top-Down Flowering Sequence.** This phenomenon, although visually simulated by passing a signal in the system (Prusinkiewicz et al., 1988), has never been simulated on a functional basis. Side shoots stay dormant with some dwarf leaves until their elongation. In the model, this delay of flowering  $D_p$  depends on their position ( $p$ ), calculated from the oldest node on the main stem upwards:

$$D_p = v_B(M - p) \quad (3)$$

where  $M$  is the maximum number of leaves on the main stem, easily observed at flowering stage;  $v_B$  is a coefficient describing speed of flowering. A side shoot breaks out

when its age is older than the delay. For example, suppose  $v_B=1.6$ ,  $M=32$ , at plant age  $N=41$ . For a branch at  $p=25$ , its age is 16, while  $D_{25}=11$  according to Eqn. (3), thus this branch started growing 5 cycles ago. Normally  $v_B$  is between 1 and 2. Some branches will breakout at the same cycle, sharing the same assimilates, leading to the same weight of organs. Such a cluster of flowers of the same size has been observed by Carvalho (2003). If  $v_B=1$ , all branches breakout at the same cycle. With observed number of elongated branches,  $n_B$ , at a given plant age  $N$ ,  $v_B$  is computed as:

$$v_B = (N - M) / n_B + 1 \quad (4)$$

Eqn. (3) corresponds to the linear relationship between number of laterals at a certain stage and those at final harvest as observed by Schoellhorn et al. (1996).

**3. Strong Internode Growth after Bud Removal.** It was observed that after removal of the terminal bud, when the side branches began to elongate, the corresponding internodes in the main stem grew stronger than the leaves. It is supposed that there is additional sink on the growing internodes at bud break of the corresponding branch. Thus for an internode at position  $p$ , its sink strength is:

$$s_{I,p} = S_I [\varphi(j - n_p) + \alpha\varphi(j - D_p)] \quad (5)$$

where  $\alpha$  is a coefficient to be determined by calibration.

**4. Two Phases of Flower Growth.** Nothnagl et al. (2004) reported that the flower diameter increase is different during bud phase compared to flower opening phase, and the flower stays as a bud for most of the time since it was initiated. Thus the sink variation pattern of a flower is set to be:

$$s_F = \begin{cases} S_F \tau_F & j \leq j_F \\ S_F \varphi(j - j_F) & j > j_F \end{cases} \quad (6)$$

where  $S_F$  and  $\tau_F$  are determined by calibration.  $j_F$  is the number of cycles during which a flower remains a bud, computed according to the critical index as defined in Nothnagl et al. (2004) and the total expansion duration of a flower.

### Model Calibration

While some parameters of the GreenLab model (number of leaves on branches, leaf thickness, duration of expansion) can be measured directly, there are other parameters (e.g. relative sink strength of organs, light extinction coefficient) that need to be estimated by non-linear least square method. Here the Levenberg-Marquardt algorithm was adopted by using the MINPACK library developed by Argonne National Laboratory.

### Experiment

**1. Experiment Setup.** The experiment was conducted in a growth chamber with artificial light ( $380 \mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; Photosynthetically Active Radiation) and controlled temperatures ( $16^\circ\text{C}$ ). Block-rooted stem cuttings of chrysanthemum 'Reagan Improved' were planted in 12 cm pots, filled with commercial potting soil and placed on trolleys at the density of 69 plants/ $\text{m}^2$ .  $\text{CO}_2$  concentration was maintained at ambient level with regular air flow. Relative humidity was maintained at 70%. Plants were grown under LD (long day) condition for 14 days, followed by SD period till commercial harvest. During the LD and SD period assimilation light was provided for 8 h from 6 am to 2 pm using 400W Son-T-Agro and HPI-T plus (1:1 ratio). In addition, during LD period incandescent lamps were lit for 11 h, from 2 pm to 1 am and during SD period these lamps were lit for 3 h, from 2 pm to 5 pm. Fertilization and other cultural practices were done according to common practice throughout the growing period.

**2. Measurements.** Schematic presentation of measurement dates is shown in Fig. 1. After H2, the apical flower bud was removed. Time of harvesting (H4) was decided when 3-4 top flowers were fully opened in the inflorescence. Measurements were done on two plants, detailed destructively measured data mainly include: fresh weight and size of leaves and internodes in the main stem (except that of internodes at planting); fresh weight of all branch leaves and all branch stems; diameter of flower (or flower buds).

## RESULTS

### Observations

**1. Number of Cycles.** The number of leaves increased linearly from H0 to H3 (Fig. 2), giving 2.36 days per cycle. Although 22 leaves were counted at H2, in fact 32 leaves were present on the main stem, as terminal bud was already visible, thus the number of cycles at H2 is 32. Number of primordia in the plant tip ( $n_p$ ) thus equals 10. Number of cycles at the other dates was computed according to the computed number of days per cycle.

The expansion duration of leaves and internodes is about 440°C days (unpublished data of S.M.P. Carvalho). At 16°C, this means the number of cycles for expansion of leaves and internodes is about 12. For the first 8 metamers the expansion duration decreases from bottom, since their expansion was assumed to be stopped at planting. The life duration of leaves was set to a large value (90 cycles), as the leaves remained green during the whole experimental period.

**2. Link between Organ Size and Weight.** These relationships were used in the model to convert from organ weight to organ size.

*1. Leaf.* The specific leaf fresh weight of individual leaves on the main stem was about 0.343 mg/mm<sup>2</sup>, and this was slightly lower for leaves on branches (0.294 mg/mm<sup>2</sup>).

*2. Internode.* An allometric relationship between internode length  $l$  and section area  $s$  existed ( $l = 0.87s^{0.84}$ ). This relationship was determined for young internodes (H1-H3) near the top to exclude the disturbing influence of secondary growth in older internodes.

*3. Flower.* Only diameters of flowers were measured in this experiment, and this is not enough to calibrate GreenLab. However, in a following greenhouse experiment on the same cultivar (not shown), both diameter and fresh weight were measured. Their relationship can be divided into a linear part, for flower buds, and a non-linear one, for opening flowers (Fig. 3). This observation is in line with Nothnagl et al. (2004).

**3. Coefficient of Delay of Expansion  $v_B$ .** At H4, the number of elongated branches ( $n_B$ ) is 15, the age of plant (N) is 41, and M=32. Therefore  $v_B=1.6$ , based on Eqn. 4.

### Model Calibration and Simulation

For model calibration fresh weight of leaves, internodes in the main stem and flowers of 10 plants (2 for each harvest date) were used simultaneously. Fitted model parameters are shown in Table 1 and predicted total organ fresh weights are shown in Fig. 4. With these parameters, the dynamic pattern of total weight of the plant and its compartments, and the leaf area index, were accurately described (Fig. 5). The output represents the average plant under the given climatic condition.

Moreover, the shape of plant (Fig. 6) can be simulated not only for the measuring dates, but also for any other stage. The flower buds have appeared at H3, and they grow top-down. The shape of each type of organ is defined manually and normalized with a software developed by Dr. Zhan (LIAMA), before it is scaled according to the computed size. Internodes are linked to form stems, and leaves are attached to the corresponding internodes according to the insertion angle, which is estimated from observation on real plants, and the phyllotactic angle, which was sampled from a negative binomial distribution with parameters  $N=55$  and  $p=0.3$ , according to the observations.

## DISCUSSION

In this study, the GreenLab model is extended with some new features and calibrated for chrysanthemum 'Reagan Improved', whose flowering sequence represents

many similar plants. The model simulated not only the first flowering, but also the development of sequent ones. Nearly all of the external qualities, like number, and size of flowers, plant height and fresh weight, are output of this calibrated function-structural chrysanthemum model. Together with De Visser et al. (2006) this is the first report on such a model for chrysanthemum.

Experience from this study tells that it is very important to know the development and underlying physiological story of real plants to 'teach' the virtual ones how to do. As the model calibration is not simply curve fitting, it would not be possible to get the results in Fig. 4 and 5 if the model didn't follow well the sink-source competition process. For example, it was observed that after pinching the top flower bud, the growing internodes in the main stem become more heavy, reflecting a different sink pattern with that of corresponding leaves (Fig. 4). The substantial increase in weight could not be simulated without introducing a function like in Eqn. (5). Fitting is difficult to reach without sufficient knowledge; in this experiment, for example, as there was no data for the individual leaf and internode weights in branches, their sink strength was difficult to estimate.

A plant is a complex system that changes dynamically, and its key behavior in terms of sink-source relationships should be caught to simulate a virtual one. Although the virtual plant has a dynamic growth pattern, for now it is still rigid in reacting to the environment. For example, instead of using a fixed  $v_B$  in Eqn. (3), it would be more flexible to make this parameter a function of the dynamic biomass supply, such that under better conditions more flowers will be produced (Carvalho, 2003) with smaller  $v_B$ . The current study, although still preliminary, opens possibilities of virtual experiments, such as pruning of plant parts which modify the sink-source relationship, and studying effects of light and plant density on the growth and external quality.

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## Tables

Table 1. GreenLab model parameters calibrated on the measured data from 10 plants at 5 stages (2 plants for each stage).

Para	Description	Estimation
$S_L^I$	Leaf sink strength, for main stem	1
$B_L$	Blade sink variation (parameter for the beta law of leaf)	0.41
$S_I^I$	Internode sink strength (for pith of main stem)	0.66
$B_I$	Internode sink variation (parameter for the beta law of internode)	0.67
$S_c$	Sink strength for secondary growth	0.005
$S_F$	Flower sink strength	0.79
$\tau_F$	Coefficient of bud sink strength, see Eqn. (6)	0.07
$B_F$	Flower sink variation (parameter for the beta law of flower)	0.67
$r_1$	Coefficient for leaf size effect on leaf resistance	0.02
$r_2$	Light extinction coefficient	0.6
$Q_0$	Initial biomass supply at planting	3.8
$\alpha$	Coefficient for sink addition of internode, see Eqn. (5)	2.4

## Figures

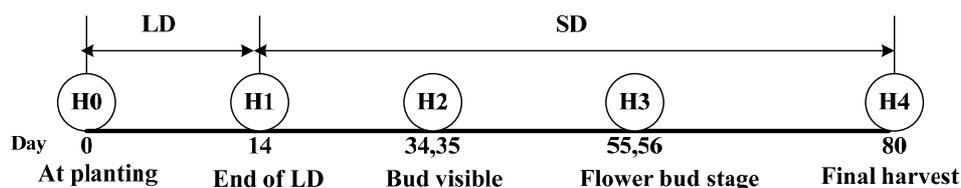


Fig. 1. Schematic representation of destructive measurement dates.

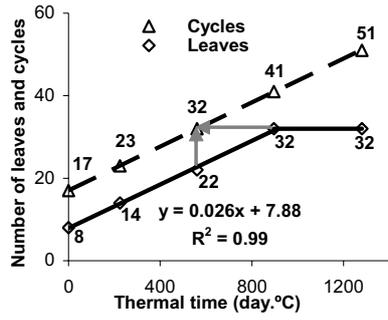


Fig. 2. Number of cycles, related to the number of leaves at each harvest.

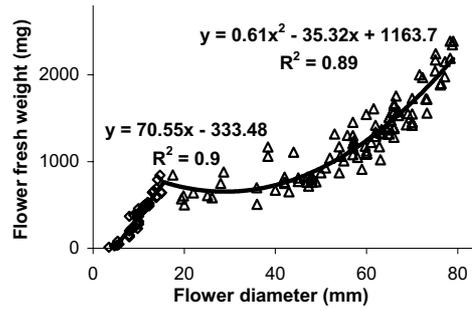


Fig. 3. Relationship between size and weight of chrysanthemum flowers.

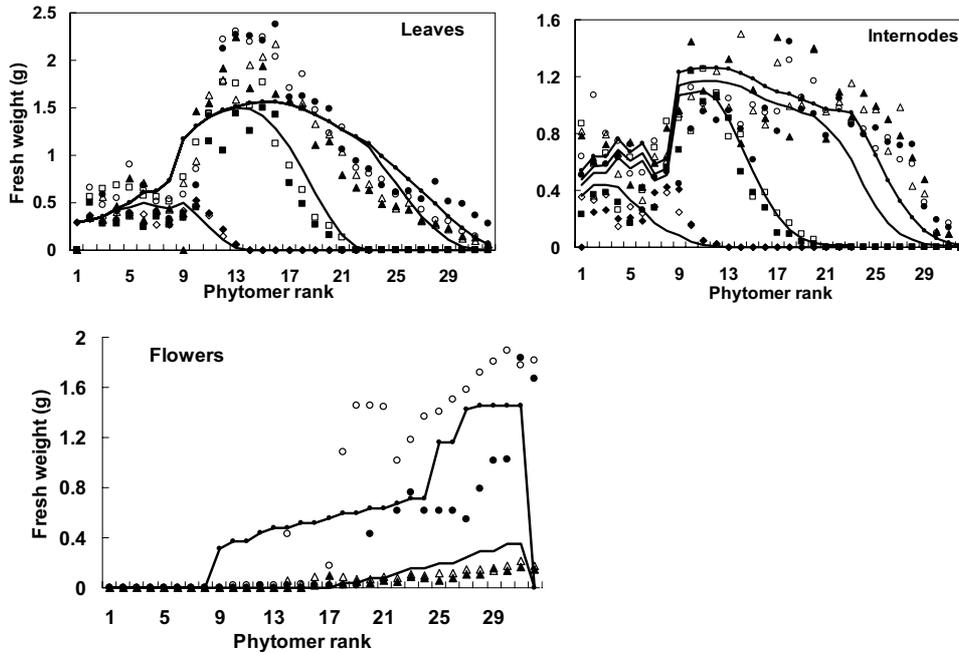


Fig. 4. Fresh weight of organs at each measurement date, for model output (solid lines) and measured data (symbols:  $\diamond$ —H1,  $\square$ —H2,  $\Delta$ —H3,  $\circ$ —H4, empty and solid for two plants).

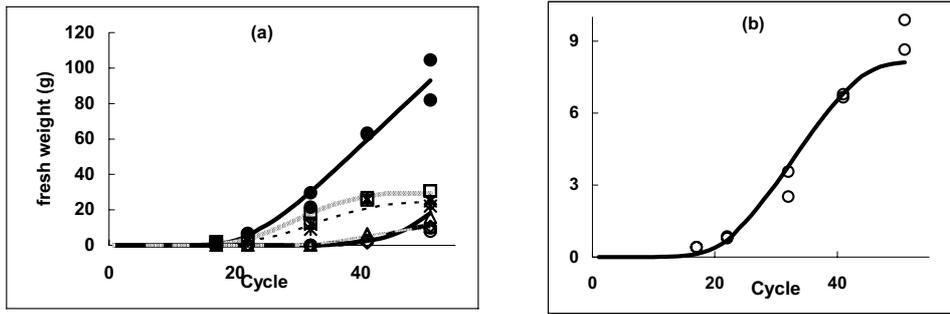


Fig. 5. (a) Weight of the plant and its allocation during growth, for model output (lines) and measured data (symbols), ●—Plant, □—Leaf MS, \*—Internode MS, ◇—Flower, Δ—Leaf B, ○—Internode B. MS=main stem, B=branch; (b) Leaf area index.

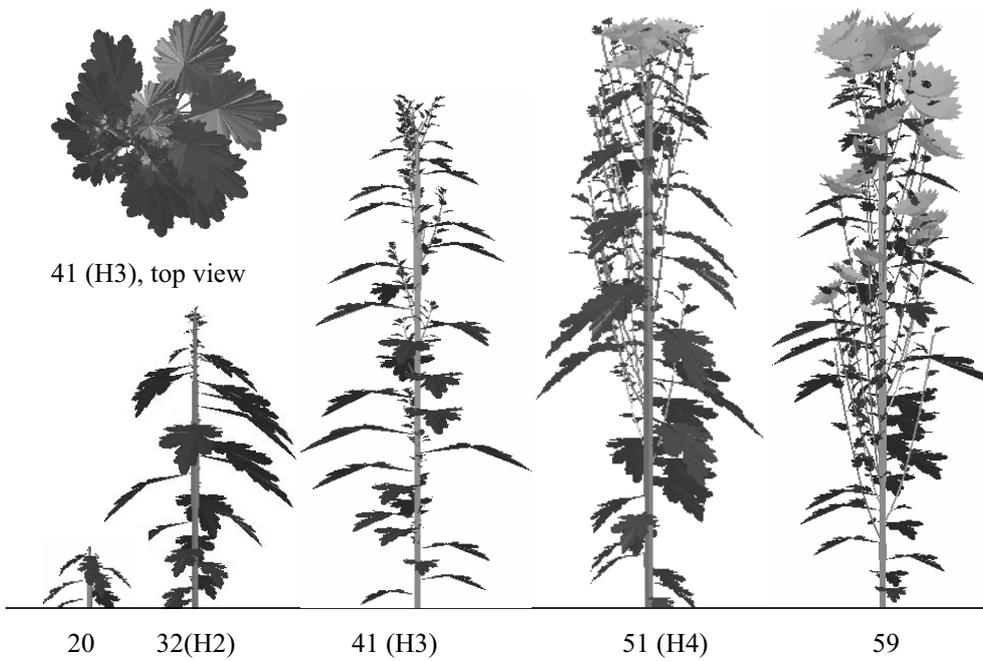


Fig. 6. Simulated 3D structure of chrysanthemum at measurement and other stages (cycles).