Scientia Horticulturae 184 (2015) 106–113

Contents lists available at ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

The phyllochron of *Prunus persica* shoots is relatively constant under controlled growth conditions but seasonally increases in the field in ways unrelated to patterns of temperature or radiation

Anna Davidson*,[1], David Da Silva, Betsy Quintana, Theodore M. DeJong

Department of Plant Sciences, University of California Davis, One Shields Avenue, Davis, CA 95616, USA

ARTICLE INFO

Article history:
Received 23 August 2014
Received in revised form
12 November 2014
Accepted 22 December 2014
Available online 16 January 2015

Keywords:
Plastochron
L-PEACH
Metamer
Leaf appearance rate
Growing degree hour radiation

ABSTRACT

Growth and development of plants are characterized by the addition of repeated units called phytomers that consist of a node, internode, leaf and axillary bud. The time elapsed between the addition of phytomers can be represented by the appearance of leaves at each node, or the leaf appearance rate (LAR). The inverse of LAR is the phyllochron, the chronological time or thermal time elapsed between the appearance of successive leaves along a stem. Plant genotype interactions with environment are responsible for the timing of the phyllochron. Temperature is important for the regulation of many plant processes. To what extent temperature regulates the phyllochron of fruit trees is not clear. Furthermore, the correct thermal time scale to employ for determining the phyllochron has yet to be determined. The objective of this study was to develop a growing degree hour (GDH) thermal time scale model for measuring the phyllochron of *Prunus persica* and to determine if temperature is the primary factor that influences the phyllochron under field conditions. A temperature-controlled growth chamber study was conducted to determine the appropriate temperatures to use in a GDH-based phyllochron model. We developed a model that is linear on both sides of a plateau-shaped optimum. The base temperature was 4 °C, the critical or maximum temperature was 40 °C and the optimal temperature range spanned 18–32 °C. This GDH model was then used to model the phyllochron of proleptic peach shoots of mature grown trees in the field for two years. Even though the GDH model accounted for much of the daily fluctuations in the phyllochron, the phyllochron generally increased as the season progressed in a manner apparently unrelated to seasonal temperature or daily radiation patterns.

Published by Elsevier B.V.

1. Introduction

Development and growth of plants are characterized by the repeated formation, expansion, and subsequent senescence of a basic unit, the phytomer (Gray, 1879). The phytomer is comprised of a node and the tissues and organs derived from it: a leaf, axillary bud(s), and internode. Also known as metamers (White, 1979), phytomers are the building blocks that are used to construct most functional–structural plant models (Allen et al., 2005). The time elapsed between the addition of phytomers can be represented and easily measured by the appearance of leaves at every node (Pagès et al., 1996). The number of leaves emerged per unit of time is termed leaf appearance rate (LAR). The inverse of LAR is termed the phyllochron, the time elapsed between the successive leaves on a stem (Wilhelm and McMaster, 1995) and is most commonly measured in hours, growing degree-days (GDD) or growing degree hours (GDH). A more precise measure is the plastochron, the unit of time corresponding to the interval between two successive leaf primordia on a stem apex (Askenasy, 1880; Erickson and Michelini, 1957; Wilhelm and McMaster, 1995). Because it is impossible to measure the initiation of leaf primordia in the field without imposing damage to the apical meristem, we used the phyllochron concept in this study. Shoot development can be conceived as the addition of successive phytomers, which are added to the shoots during each phyllochron.

The phyllochron is based on growth and development, which are interdependent processes resulting from the interaction of genotype and environment (Callaham, 1962; Lambers et al., 2008). Progress is being made on the genetic factors that control leaf initiation and the plastochron but these topics are still not well understood.

* Corresponding author at: C/O Ted DeJong, Department of Plant Sciences, University of California Davis, Wickson Hall, Mail Stop 1, One Shields Avenue, Davis, CA 95616, USA. Tel.: +1 815 212 4409.
E-mail address: adavidson@ucdavis.edu (A. Davidson).
[1] Permanent address: 615 A St., Davis, CA 95616, USA.

http://dx.doi.org/10.1016/j.scienta.2014.12.033
0304-4238/Published by Elsevier B.V.

Auxin has been shown to induce leaf formation at the shoot apical meristem. Auxin is transported through plant tissues by the polar auxin transport system. A main contributor to the polar auxin transport system is the auxin efflux carrier protein PIN-FORMED1 (PIN1). PIN1 transports auxin through the epidermis to form concentrated points of auxin called auxin maxima on the peripheral zone of the shoot apical meristem to establish the site of leaf initiation. Existing leaf primordia act as sinks, influencing where subsequent leaf initiation sites will occur (Reinhardt et al., 2003). With respect to the plastochron, several genes have been identified: *PLASTOCHRON 1 (PLA1) and PLASTOCHRON 2 (PLA2)* in rice (*Oryza sativa*: Miyoshi et al., 2004; Kawakatsu et al., 2006), *terminal ear1 (te1)* in maize (*Zea mays*; Veit et al., 1998), and ALTERED MERRISTEM PROGRAM1 (AMP1), PHYTOCHROMEB (PHYB), and SERRATE (SE) in *Arabidopsis thailiana* (Helliwell et al., 2001; Prigge and Wagner, 2001). *Pla1*, *te1*, and *amp1* shorten the plastochron. In contrast, *phyB* and *se* show longer plastochrons than the wild type.

In spite of considerable progress in understanding molecular processes that lead to the sequential initiation of new leaves the role of which environmental parameters are most important in the regulation of the phyllochron is also still unclear. Temperature exposure is considered to be the main environmental factor influencing the phyllochron (Dennett et al., 1978; Rawson and Hindmarsh, 1982; Birch et al., 1998) followed by photoperiod (Rawson and Hindmarsh, 1982; Cousens et al., 1992; Rawson, 1993; Slafer et al., 1994b; Kirby, 1995), incident radiation (Slafer, 1995; Bertero, 2001) and water status (Silk, 1980; Mathews et al., 1987). Nitrogen availability (Longnecker and Robson, 1994), salinity (Mass and Grieve, 1990) and atmospheric carbon dioxide (Hofstra and Hesketh, 1975; Rogers et al., 1984) have also been reported to affect the phyllochron, but to a lesser extent.

The phyllochron has been shown to be constant under constant temperatures (when grown in controlled environments) and fluctuates with changing temperatures in tobacco, barley, wheat, sunflower and potato (Raper and Thomas, 1975; Cao and Moss, 1989; Villalobos and Ritchie, 1992; Cao and Tibbitts, 1995; Fleisher et al., 2006). Rawson and Hindmarsh (1982) studied the effects of temperature on LAR and leaf expansion on five cultivars of sunflower grown in chambers at three temperature regimes and found that leaves appeared more quickly with increasing temperatures. Villalobos and Ritchie (1992), also studying sunflower, measured rates of leaf appearance under nine temperature regimes that ranged from 34 °C to 12/5 °C and found that leaf appearance increased with temperatures reaching a maximum of up to 27 °C, and subsequently declined with increasing temperatures to point when no leaves appeared at 40 °C. When they assessed base temperatures of 5.4, 3.6, 2.3, and 4 °C they found that the most appropriate base temperature was 4 °C. They also reported that the effects of temperature are sensitive to leaf position. Lower leaf positions had higher (longer) phyllochrons (35–43 °Cd) than leaves at higher positions (18–29 °Cd). They found no effect of temperature for leaves 1–6 however, the phyllochron for leaves 7–20 decreased with increasing temperatures. A similar result was observed in both barley and wheat (Cao and Moss, 1989). Cao and Tibbits (1995) indicated that LAR for potato increased slightly with temperatures from 17 to 22 °C. Fleisher et al. (2006) later confirmed that potato LAR increased with temperature until an optimum value of 27.2 °C was reached and then subsequently declined.

Most of the studies of the effects of temperature on the phyllochron have been conducted on annual crops most likely due to their short growing cycle, small size and their ease of cultivation in greenhouses and growth chambers where temperature can be controlled (Dambreville et al., 2013). Very few phyllochron studies have been conducted in woody perennials and even fewer in deciduous fruit trees (Kervella et al., 1995; Pagès et al., 1996). Temperature effects on growth parameters of adult trees in general, have rarely been studied (Dambreville et al., 2013). Modeling the phyllochron of trees as opposed to an herbaceous annual or monocot poses unique challenges because trees are inherently more complex. A tree is a collection of hundreds of shoots, each with apical meristems. A tree also initiates vegetative buds in the late summer followed by a period of winter dormancy. Gordon et al. (2006) found that in peach approximately the first ten leaf primordia are preformed between leaf drop in the fall and bud break in the spring. Subsequent nodes are neoformed after bud break, meaning their abundance is dictated by current season's conditions.

In young potted peach trees grown outdoors, a nonlinear gradual decrease in leaf (Kervella et al., 1995) and metamer emergence rates (Pagès et al., 1996) were reported (plotted in growing degree days), as the number of leaves on shoots increased over the course of the season. Significant differences between main and first order shoots of young potted peach trees were also reported. The same was seen for leaf emergence in grapevine (Schultz, 1992). Cieslak et al. (2011) reported that the kiwifruit vine phyllochron (plotted against average daily temperature) also responded non-linearly to temperature. Their studies showed that the rate of kiwifruit leaf appearance increased with average daily temperature up to an optimum and then subsequently declined.

Understanding the effects of temperature on the phyllochron of peach is an interesting developmental biology question that is still not well understood. Additionally, understanding how temperature affects the timing of the phyllochron is important when building functional structural plant models (DeJong et al., 2011). The L-PEACH model is a 3D functional–structural interactive plant model (Allen et al., 2005; Da Silva et al., 2011; Lopez et al., 2008) that simulates 3D peach tree architecture and solves electrical circuit analogs for carbohydrate flow and allocation based on hourly solar radiation and air temperature. It is comprised of four submodels: movement of carbon, organ functionality, tree architecture and commercial practices.

Currently the L-PEACH model has shoot growth models that need to be improved to incorporate more accurate timing of phytomer appearance. However, quantification of the effects of factors influencing the phyllochron has been insufficient to provide an accurate sub-model for phytomer appearance, which provides the basis for this research.

When modeling the phyllochron in thermal time, an important consideration is the type of thermal time model used. Growing degree-days (GDD) are used for monitoring the effect of temperature on biological processes (McMaster and Wilhelm, 1997). Generally, it is calculated by subtracting a base temperature from the daily average temperature. The base temperature is the temperature below which plant development slows or stops. GDD's are added together resulting in a GDD accumulation, which can be used to assess growth and development stages. Growing degree hours are calculated using similar principles with hourly temperatures. Growing degree hour models for phyllochron studies need to be adjusted for specific species for different objectives. Kervella et al. (1995) and Génard et al. (1994) are the only studies to our knowledge that report GDH minimum and optimum values for *Prunus persica* specifically to study the phyllochron. In their study, they computed a range of temperatures from −2 °C and 4 °C to calculate the base temperature, and a range of temperatures between 20 °C and 30 °C to compute the optimum temperature. They report that the coefficient of correlation between leaf number and thermal unit accumulation at successive observation dates varied minutely over those ranges. However 26 °C corresponded to the highest value of the coefficient of correlation.

To confirm that there was little difference in phyllochron values at 20 °C and 30 °C, we conducted a growth chamber experiment

to observe the phyllochron of one-year-old peach trees grown in two different average daily temperature regimes of 20 °C and 30 °C. Additionally, we were interested in observing how additional growth parameters (i.e. biomass, leaf area, leaf length, shoot length, number of syleptic shoots) responded to the two temperatures. The results of this model were compared to the previous model of Kervella et al. (1995) and hourly phyllochron data. Subsequently we used this growing degree hour model for studying the phyllochron of proleptic shoots on mature bearing peach trees grown in the field in Winters, California during the 2010 and 2011 seasons.

2. Materials and methods

2.1. Temperature growth chamber experiment

To assess temperature effects on the phyllochron a growth chamber experiment was conducted from March 5 to May 5, 2012, at the UC Davis Environmental Horticulture Growth Chamber Facility. One-year-old peach trees, 'Elberta' scions on 'Nemaguard' rootstock, were used. Upon arrival from a commercial nursery, the tree canopies were initially comprised of a trunk and only 3–4 previous year shoots with a few flowers (which were removed) and no emerged vegetative shoot buds. Twenty trees were potted in sixteen-liter buckets in fritted clay. On March 5, ten trees each were put in walk-in growth chamber set at an average temperature of 30 °C, and the other ten trees were put in a growth chamber set at 20 °C. Chambers were set to 12-h days with light flux densities of 500 μmol m^2 s^{-1}. Temperature and humidity were recorded every 30 min with HOBO Data loggers (Onset Computer Corporation, Bourne, MA). The pots were watered to field capacity every other day for the first two weeks then daily starting March 23. One liter of 75% strength Hoagland's solution was added to pots once per week to ensure adequate nutrient availability.

The trees were given one week to acclimate to growth chamber conditions and to initiate shoot growth before the start of data collection. On March 12 two shoots per tree (a total of 40 shoots) were selected and followed for the remainder of the experiment. All the leaves on each monitored shoot were counted and measured every other day.

At the end of the experiment the trees were harvested by separating the incremental canopy growth from the main scaffolds that existed previous to the start of the experiment. The tagged shoots followed for phyllochron measurements were separated from the rest of the canopy. The lengths of these shoots and their secondary lateral neoformed syleptic shoots were measured and counted. The stem biomass was separated from the leaf biomass for all shoots of the canopy for each tree. All canopy material was dried at 50 °C for one week and weighed.

Leaves representing six size classes were randomly selected from non-tagged shoots (120 leaves per treatment) were measured for leaf length and area. A polynomial equation, $LA = 0.1889L^2 + 0.2735L$ was fit to estimate leaf area (LA, cm^2) non-destructively from measurements of leaf lamina length (L, mm). Leaves were scanned using a portable scanner (CanoScan LIDE110) and an image analysis program (ImageJ, U. S. National Institutes of Health, Bethesda, MD, USA) that calculated leaf area. Treatment difference in harvest measurements was evaluated by analysis of variance using JMP version 10 (SAS Institute Inc., Cary, NC, 1989–2010).

2.2. Field experiment

This research was conducted during the 2010 and 2011 growing seasons at the UC Davis Wolfskill Experimental Orchards in Winters, CA. Four-year and subsequently five-year-old fully mature peach trees of the Lorri May cultivar (unreleased) grafted on Controller 9 rootstock growing in a Yolo sandy clay loam were used in this study. Trees were spaced 1.83 m apart in the row, with 5.18 m between rows and trained to the Kearney Agricultural Center perpendicular-V system (KACV) (DeJong et al., 1994). Nitrogen was applied twice per year, 112 kg/ha in February and 56 kg/ha in September. Following the cessation of winter rains, trees were irrigated an average of 5.84 cm approximately every two weeks using microsprinklers. To ensure that the trees were not water limited, plant water status was regularly monitored by taking mid-day stem water potential (Ψ_{ST}) readings from each studied tree using a pressure chamber as described by McCutchan and Shackel (1992).

2.3. Experimental design

Two north-south oriented rows of trees (in the middle of a total of eleven rows) were organized into a randomized complete block design with three blocks, six replications (trees) per block. Two proleptic shoots located at breast height were randomly selected from both the west and the east-facing sides of the tree (a total of 36 shoots per season). If the shoots became damaged or ended growth uncharacteristically early, they were replaced by nearby proleptic shoots.

2.4. Leaf appearance rate measurements

In order to assess leaf growth rates and the phyllochron, incremental measurements of every leaf grown on selected shoots were made using a metric ruler. These repeated measurements were collected three times per week from April 5 to September 29, 2010, and from April 13 to August 17 in 2011. Hourly temperatures were recorded by two HOBO data loggers (Onset Computer Corporation, Bourne, MA) located in the orchard and confirmed by the local California Irrigation Management Information System (CIMIS) weather station located on site.

2.5. Data analysis

Leaf lengths recorded in the field were analyzed using Python 2.7 (http://www.python.org/) and matplotlib library (http://matplotlib.org/). When taking field measurements of very small leaves it was impossible to capture the exact initiation point of the leaf without imposing damage. Additionally, the appearance of leaves sometimes occurred between days of data collection. Therefore, leaf appearance was normalized to the time when a new leaf was 2.0 cm long. This was achieved by first plotting the incremental measurements of each leaf to create a leaf growth rate.

The normalized leaf appearance time was estimated by fitting the leaf length growth data to a classical growth curve using the Gompertz model $y(t) = a.e^{b.e^{ct}}$ where a is defined as the upper asymptote, b is related to the speed of growth, c the lower asymptote, and t the time in hours (Gompertz, 1825). We chose 2.0 cm as the leaf initiation point to avoid the initial curve of the Gompertz model (Fig. 1).

2.6. Development of a growing degree hour model

With the normalized leaf appearance times we estimated the time interval between two successive leaves in chronological time (hours) and in thermal time (GDH). We based our GDH model on previous GDH models developed by Richardson et al. (1975) and Kervella et al. (1995). We modified the optimal temperature based on the results of the growth chamber experiment (see Section 3). We developed a GDH model as linear on both sides of a plateau-shaped optimum. The base temperature (T_B) was 4 °C, the critical or maximum temperature (T_C) was 40 °C.

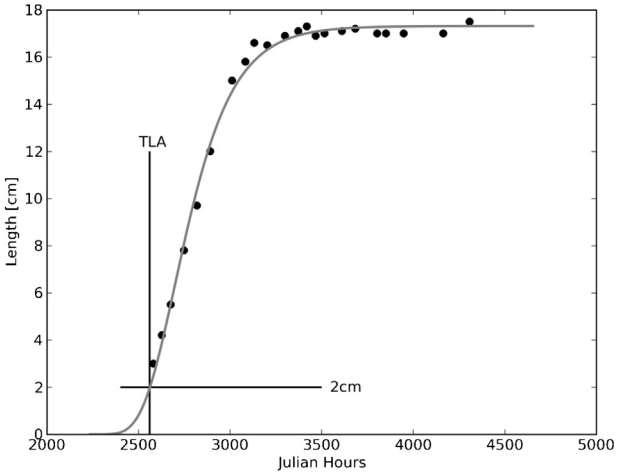

Fig. 1. An example of a Gompertz growth curve fitted to the original leaf growth rate data to estimate the normalized time of leaf appearance, defined as 2.0 cm. TLA = time of leaf appearance.

Because temperatures in the growth chamber reached as low as 18 and as high as 32 °C we used a span of 18–32 °C (T_{O1} and T_{O2}). Therefore, when the current temperature (T_H) was below T_B or above T_C nothing was added to the GDH accumulation. When $T_B < T_H < T_{O1}$, $T_H - T_B$ was added to GDH accumulation. When $T_{O2} < T_H < T_C$, $(T_C - T_H)^*(T_{O1} - T_B)/(T_C - T_{O2})$ was added to the GDH accumulation. When $T_{O1} \leq T_H \leq T_{O2}$, $T_{O1} - T_B$ was added to GDH. Fig. 2 shows the value the GDH value associated with an hour at a given temperature.

The mean phyllochron values were calculated for each shoot and then averaged again to obtain the mean phyllochron of all shoots in a given ten-day time interval. Ten-day time intervals were repeated throughout the season. We divided the season into ten-day time intervals to obtain a running average that represented the periods during which at least 3–4 leaves were produced. Phyllochrons were calculated in both chronological time and thermal time scales to determine if temperature had a significant impact on the phyllochron and if so, to negate the effects of temperature. Analysis of variance with one standard error from the mean was calculated using JMP version 10 (SAS Institute Inc., Cary, NC, 1989–2010).

For comparison, we plotted our data using the GDH model developed by Kervella et al. (1996) and Génard et al. (1994), which consisted of a minimum of 0 °C and an optimum of 26 °C. We also

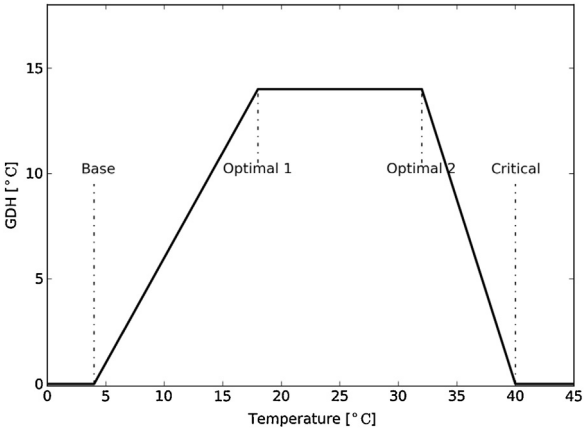

Fig. 2. Growing degree hour model based on Richardson (1975), where the base temperature (T_B) is 4 °C, optimal temperatures range from 18 °C (T_{O1}) to 32 °C (T_{O2}) and the critical temperature (T_C) is 40 °C.

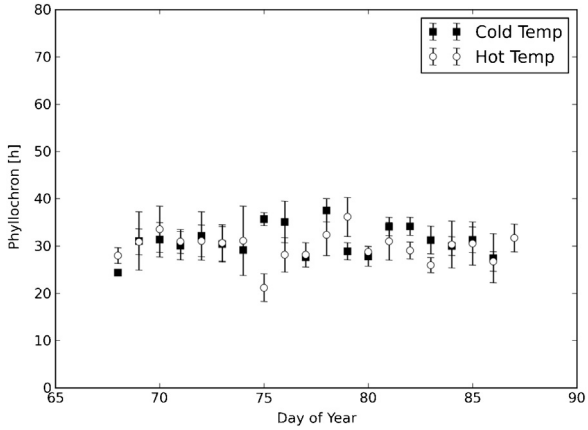

Fig. 3. The mean phyllochron of shoots on peach trees grown in cold and warm temperature growth chamber treatments plotted daily in hours. Error bars represent the standard error from the mean.

plotted our field data against daily radiation to investigate its effect on the phyllochron over the season.

3. Results

3.1. Temperature growth chamber experiment

In both temperature treatments, which maintained a constant average temperature, the phyllochron remained relatively constant (Fig. 3). There were no overall significant differences in phyllochron values between temperature treatments when plotted in hours (Fig. 3). The mean phyllochron value was 31.4 ± 1.64 h for the 30 °C temperature treatment and 32.1 ± 1.72 (Table 1) hours for the 20 °C temperature treatment.

We then applied the GDH model developed by Kervella et al. (1995) to our hourly growth chamber data which consisted of a minimum of 0 °C and an optimum of 26 °C. The fit to this model can be observed in Fig. 4.

The temperature treatment effects on the phyllochron were then plotted using a GDH scale with the cardinal temperatures: T_B = 4 °C, T_O = 18–32 °C and T_C = 40 °C (Fig. 5). With these parameters the phyllochron displayed the same pattern as when plotted in hours. When plotted on our GDH scale (Fig. 5), mean phyllochrons were 414.1 ± 21.71 and 447.23 ± 22.80 GDH for warm and cool treatments, respectively (Table 1). Because our GDH scale better fit

Table 1
Summary of final growth data for growth chamber experiment.

Growth chamber temperature experiment:	30 °C	20 °C
Phyllochron (GDH)	414.1 ± 21.71a	447.2 ± 22.80a
Phyllochron (h)	31.6 ± 1.64a	32.1 ± 1.72a
Leaf length (cm)	11.3 ± 0.30a	12.8 ± 0.29b
Leaf area (cm^2)	28.2 ± 1.53a	36.3 ± 1.53b
Leaf dry weight (g) (total canopy)	15.2 ± 1.36a	27.9 ± 1.36b
Stem dry weight (g) (total canopy)	3.3 ± 0.44a	6.3 ± 0.44b
Tagged shoot length (cm)	20.8 ± 1.54a	24.3 ± 1.54a
# syleptics on tagged shoots	3.2 ± 0.92a	1.8 ± 0.92a
Length of syleptics on tagged shoots (cm)	12.4 ± 1.49a	9.5 ± 1.11a
# syleptics per tree	8.1 ± 1.65a	5.3 ± 1.65a
# additional first order shoots	7.2 ± 1.50a	10.1 ± 1.50a
# syleptics on additional first order shoots	2.6 ± 0.64a	1.7 ± 0.64a
Length of additional first order shoots (cm)	20.8 ± 1.86a	24.4 ± 1.76a
Length of syleptics on additional first order shoots (cm)	20.5 ± 2.18a	9.7 ± 3.04b

All values are means ± standard error. Means followed by letters that are the same are not significantly different at according to LS means Student's *t*-test ($P < .005$).

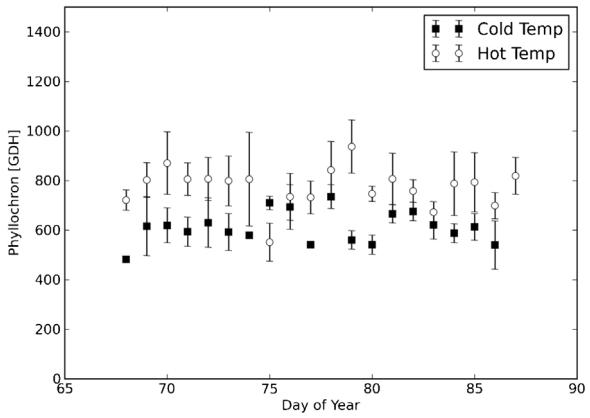

Fig. 4. The mean phyllochron of shoots on peach trees grown in cold and warm temperature growth chamber treatments plotted daily in GDH using the "Kervella Model" with a minimum of 0 °C and an optimum of 26 °C. Error bars represent the standard error of the mean.

the growth chamber data in hours compared to the model developed by Kervella et al. (1995), we decided to employ this model to our field data.

While there were no significant differences in the phyllochron, the total incremental canopy biomass (leaf dry weight and mean stem dry weight) was greater for trees in the 20 °C treatment. This was likely related to both the significantly greater leaf length and leaf area in the 20 °C treatment (Table 1). Final mean length of the tagged shoots was also greater in the cool treatment, although not significantly. The number of first order shoots that grew in addition to the tagged shoots was greater in the 20 °C treatment with a mean of 10 compared to only 7 additional shoots in the warm treatment. The average length of these shoots was significantly higher in the 30 °C treatment. Syleptic shoots were also counted, measured and harvested. Both the mean number of syleptics per tree and mean length of sylpetics were higher in the 30 °C treatment (Table 1).

3.2. Field experiments

Although the proleptic shoots initiated their growth around the same time in the spring they grew for a longer length of time in 2011 than in 2010. Field temperatures early in the season were somewhat cooler in the spring of 2010 than 2011.

In spite of the fact that the phyllochron of field grown shoots was calculated using a thermal time model (Fig. 6) there was a clear

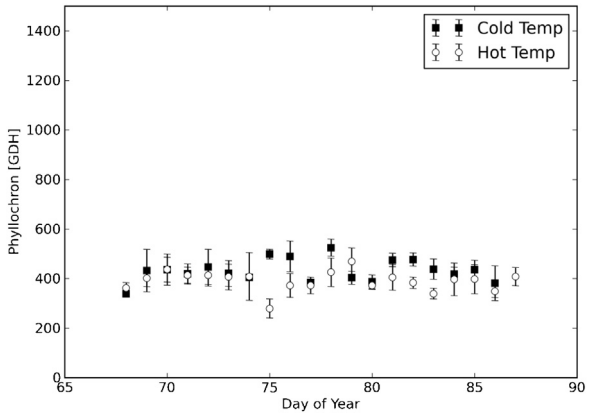

Fig. 5. The mean phyllochron of shoots on peach trees grown in cold and warm temperature growth chamber treatments plotted daily in GDH using the temperatures T_B = 4 °C, T_O = 18–32 °C and T_C = 40 °C. Error bars represent the standard error of the mean.

Table 2
Phyllochron values for proleptic shoots grown in the field during the 2010 and 2011 field season expressed in hours and GDH.

Phyllochron values	Year 2010	Year 2011
Mean phyllochron (H)	85.3 ± 3.34	88.5 ± 2.68
Mean phyllochron (GDH)	857.6 ± 32.91	988.0 ± 30.18

All values are means ± standard error.

trend toward increasing phyllochrons over the growing season in spite of the fact that temperatures also increased over that period. This pattern of increasing phyllochron with increasing temperature was apparent regardless of whether phyllochrons were calculated using chronological time or thermal time (using our GDH scale or that of Kervella et al. (1995)). However the patterns were most consistent using our modified GDH model.

Annual phyllochron grand means in both chronological time and thermal time were similar for 2010 and 2011 (Table 2). The slightly higher mean values in 2011 are likely a function of the fact that measurements proceeded later in 2011 than 2010 and the phyllochron tended to be longer as the season progressed.

Patterns of daily radiation and day length in the field steadily increased until about day 180, after which they began to steadily decline indicating a normal seasonal trend. However, the phyllochron continued to increase throughout the season despite the change in patterns in daily radiation (Fig. 7).

4. Discussion

The 10 °C difference in average daily temperature in the growth chamber treatments did not affect the phyllochron when calculated using chronological time (Fig. 3). This corresponds with the report of Kervella et al. (1995) who also reported a minimal effect of temperatures between 20 °C and 30 °C on leaf emergence rate for peach but constructed a GDH model with an optimal temperature of 26 °C because that corresponded to the highest correlation coefficient for their data. The Kervella model did not fit our growth chamber hourly data as we expected, therefore we constructed our own GDH model with a maximum temperature with the goal of achieving a better fit. The growth chamber settings for the temperature experiments were set at 20 and 30 °C, but when the measured temperatures inside of the growth chambers were analyzed we determined that the mean temperatures in the chamber were close to 18 and 32 °C so we used these temperatures for establishing the optimal temperature range of our GDH model. This model fit our growth chamber data very well (Figs. 3 and 4). Therefore we also used it to calculate the phyllochron for the field experiments. Temperatures in the field in California do not normally go above 40 °C or below 4 °C, which were designated as the T_c and T_b values respectively.

If temperature were the only factor affecting the phyllochron in the field, as in the growth chamber, then when the phyllochron is calculated using a thermal time model (accounting for temperature effects) then one might expect the phyllochron to be steady over the course of the entire season, creating a flat line across time. However, there was a general upward trend throughout the season indicating that temperature was not the only factor increasing the phyllochron (Fig. 6). This seasonal trend toward longer phyllochrons was also apparent when the phyllochron was calculated using chronological time or the Kervella et al. (1996) thermal time model (data not shown).

Two other potentially important factors reported to effect the phyllochron; radiation (Bertero, 2001) and photoperiod (Rawson and Hindmarsh, 1982; Rawson, 1993), also appear to be unrelated to the phyllochron in field-grown peach trees because the usual expected effect of the increasing radiation and photoperiod would

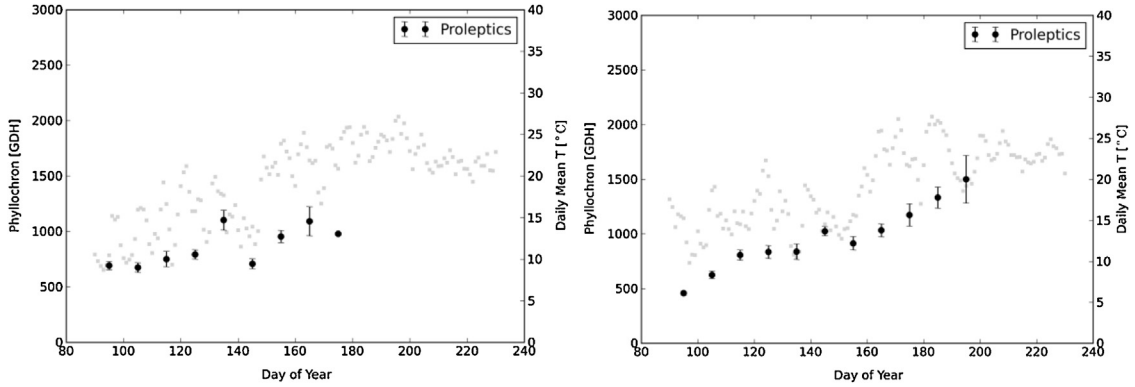

Fig. 6. The mean phyllochron (in GDH) of proleptic peach shoots grown in the field during the 2010 (left) season and 2011 season (right) plotted for ten-day intervals. Error bars represent the standard errors from the mean. Mean daily temperature is plotted as gray dots in the background.

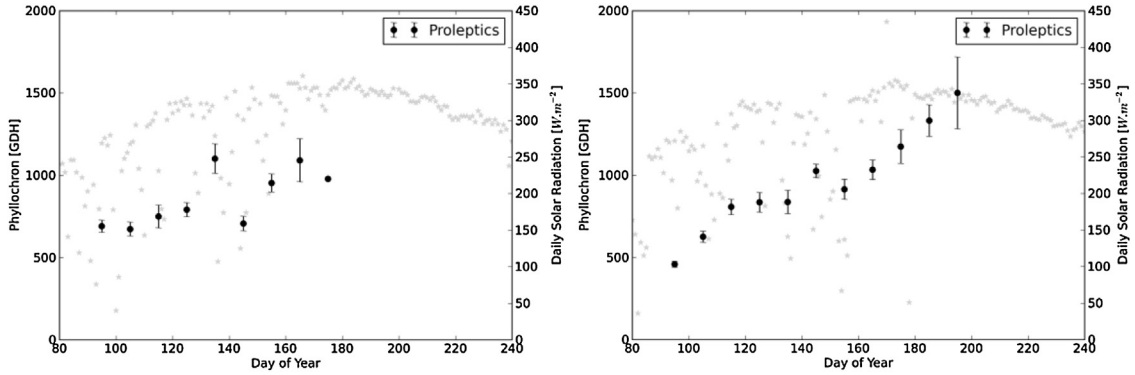

Fig. 7. The mean phyllochron of proleptic peach shoots grown in the field during the 2010 (left) and 2011 (right) season plotted for ten-day intervals in GDH. Daily radiation is represented by gray stars. Error bars represent the standard errors from the mean.

be a shortening of the phyllochron and the opposite was the case in both the 2010 and 2011 experiments prior to midseason. Observations in wheat, barley and *Avena fatua* (Volk and Bugbee, 1991; Cousens et al., 1992; Cao and Moss, 1994; Kirby, 1995; Mosaad et al., 1995; Slafer and Rawson, 1997) indicated that longer photoperiods shortened the phyllochron. However, longer photoperiods did lengthen the phyllochron in quinoa (Bertero et al., 2000). After midseason (day 180) in 2011 the peach shoot phyllochron continued to lengthen even though days became shorter and daily radiation decreased. Thus, the general evidence indicates that the peach shoots were relatively insensitive to total daily radiation of photoperiod.

Therefore, we hypothesize that the slowing of the phyllochron in field-grown peach shoots was due to an endogenous rank effect; meaning that the speed of the phyllochron may be a reflection of the number of leaves previously emerged. Thus the phyllochron is shorter at lower insertions (Villalobos and Ritchie, 1992), and as more phytomers are added over the season, the phyllochron lengthens (Kervella et al., 1995; Pagès et al., 1996). Kervella et al. (1995) studied genotypic differences in leaf emergence (phyllochron) and Pagès et al. (1996) the metamer emergence rate, of four peach and five nectarine cultivars that were chip budded on one-year-old peach rootstock. Leaf emergence was followed twice per week for the duration of the season on main and first order shoots. The phyllochron and metamer emergence decreased steadily from the start of the season until the end. This trend was consistent, though there were differences among cultivars. In most cases the phyllochron lengthened more slowly for trees with long initial phyllochrons. Phyllochrons were always longer for first-order shoots than for main shoots, suggesting shoot order and shoot type are important considerations when modeling the phyllochron. Nonlinear relationships between leaf emergence/phyllochron and thermal time have also been reported for many other crops, including sorghum (Clerget et al., 2008) and kiwi (Morgan et al., 1985; Seleznyova and Halligan, 2006).

A rank effect was not observed in the growth chamber experiments. The phyllochron was nearly constant over the course of these short-term experiments (Figs. 3 and 4) in contrast to the field studies. As previously mentioned, there are numerous growth chamber studies that report a constant phyllochron when grown in constant temperatures, but fluctuate with changing temperatures as in tobacco, barley, wheat, sunflower and potato (Raper and Thomas, 1975; Cao and Moss, 1989; Villalobos and Ritchie, 1992; Cao and Tibbitts, 1995; Fleisher et al., 2006) but, as stated above, changing temperatures and radiation were not correlated with changes in the phyllochron in the field. Thus there must be something about the overall field environment or plant material that induced a rank effect in the field but not the growth chamber. It may be that the different ages of the plant material or severities of pruning prior to the onset of the measurements in the growth chamber and field experiments may be responsible for these differences in phyllochron patterns. More research is needed to clearly explain these differences in plant behavior (Fig. 7).

In addition to the phyllochron being unaffected between growth chamber temperature treatments, final shoot length also did not differ significantly. The number of secondary syleptic shoots that grew simultaneously with the tagged shoots was greater in warmer temperatures, but this difference was not statistically significant (Table 1). The total number of first order shoots, average leaf length and leaf area were greater in the 20 °C treatment. These differences reflect the significantly greater total canopy leaf and stem dry weight in the 20 °C treatment (Table 1). These total dry matter

growth characteristics are consistent with the fact that peach leaves are reported to be relatively insensitive to temperatures between 20 °C and 30 °C (DeJong and Moing, 2008) while total plant respiration rates likely doubled over a similar increase in temperature (Grossman and DeJong, 1994). Thus the temperature difference between 20 °C and 30 °C did have an effect on the overall growth of the tree, while the phyllochron remained insensitive to these temperature differences.

5. Conclusions

The GDH model that we developed for modeling the phyllochron of peach shoots was consistent with the growth chamber based phyllochron data and the phyllochron remained relatively constant over time over the course of the growth chamber experiments. When the thermal time model was used to calculate the phyllochron of shoots on mature field-grown peach trees there was a clear lengthening of the phyllochron over each growing season in two sequential years that appeared to unrelated to seasonal changes in temperature, daily radiation and photoperiod. Thus there appeared to be a rank effect on the phyllochron of shoots on field grown mature peach trees such that as additional leaves were sequentially added to shoots the phyllochron lengthened. A similar rank effect was not seen in the shorter growth chamber experiments. Thus predicting the phyllochron in field grown peach trees is much more complicated than simply using phyllochron models based on data collected on young trees under controlled growth conditions. This makes accurate modeling of shoot growth of peach trees more complex than previously thought.

Acknowledgments

The authors would like to thank Duarte's Nursery for their generous donation of the peach trees used in the growth chamber study. Special thanks to Kevin Coe and Randi Golding for their assistance with data collection and Dr. Neil Willits for his statistical advice.

References

Allen, M.T., Prusinkiewicz, P., DeJong, T.M., 2005. Using L-systems for modeling source–sink interactions, architecture and physiology of growing trees: the L-PEACH model. New Phytol. 166, 869–880.
Askenasy, E., 1880. Uber eine neue methode, um die ver- theilung der wachsthumsintensitat wachsenden theilen zu bestimmen. Verh. Naturh.-Medic. Ver. Heidelb. 2, 70–153.
Bertero, D.H., 2001. Effects of photoperiod, temperature and radiation on the rate of leaf appearance in quinoa (*Chenopodium quinoa Willd.*) under field conditions. Ann. Bot. 87, 495–502.
Bertero, H.D., King, R.W., Hall, A.J., 2000. Modelling photoperiod and temperature responses in quinoa (*Chenopodium quinoa Willd.*). Field Crops Res. 63, 19–34.
Birch, C.J., Vos, J., Kiniry, J., Bpos, H.J., Elings, A., 1998. Phyllochron responds to acclimation to temperature and irradiance in maize. Field Crops Res. 59, 87–104.
Callaham, R.Z., 1962. In: Tree growth., Kozlowski, T.T. (Eds.), Geographic Variability in Growth in Forest Trees. Ronald Press Company, pp. 311–325, 1962.
Cao, W., Moss, D., 1989. Temperature and daylength interaction on phyllochron in wheat and barley. Crop Sci. 29, 1046–1048.
Cao, W., Moss, D.N., 1994. Sensitivity of winter wheat phyllochron to environmental changes. Agron. J. 86, 63–66.
Cao, W., Tibbitts, W., 1995. Leaf emergence on potato stems in relation to thermal time. Crop Sci. 87, 474–477.
Cieslak, M., Seleznyova, A.N., Hanan, J., 2011. A functional–structural kiwifruit vine model integrating architecture, carbon dynamics and effects of the environment. Ann. Bot. 107, 747–764.
Clerget, B., Dingkuhn, M., Gozé, E., Rattunde, H.F.W., Ney, B., 2008. Variability of phyllochron, plastochron and rate of increase in height in photoperiod-sensitive sorghum varieties. Ann. Bot. 101, 579–594.
Cousens, R.D., Johnson, M.P., Weaver, S.E., Martin, T.D., Blair, A.M., 1992. Comparative rates of emergence and leaf appearance in wild oats (*Avena fatua*), winter barley (*Hordeum sativum*) and winter wheat (*Triticum aestivum*). J. Agric. Sci. 118, 149–156.
Da Silva, D., Favreau, R., Auzmendi, I., DeJong, T.M., 2011. Linking water stress effects on carbon partitioning by introducing a xylem circuit into L-PEACH. Ann. Bot. 41, 433–447.
Dambreville, A., Normand, F., Lauri, P.E., 2013. Plant growth co-ordination in natura: a unique temperature controlled law among vegetative and reproductive organs in mango. Funct. Plant Biol. 40, 280–291.
DeJong, T.M., Moing, A., 2008. Carbon assimilation, partitioning and budget modelling. In: Layne, D.R., Bassi, D. (Eds.), The Peach: Botany, Production and Uses. CAB International, Oxfordshire, UK, pp. 224–263.
DeJong, T.M., Day, K.R., Doyle, J.F., Johnson, R.S., 1994. The Kearney Agricultural Center Perpendicular V (KAC-V) orchard system for peaches and nectarines. HortTechnology 4, 362–367.
DeJong, T.M., Da Silva, D., Vos, J., Escobar-Gutierrez, A.J., 2011. Using functional–structural plant models to study, understand and integrate plant development and ecophysiology. Ann. Bot. 108, 987–990.
Dennett, M.D., Auld, B.A., Elston, J., 1978. A description of leaf growth in *Vicia faba* L. Ann. Bot. 47, 350–351.
Erickson, R.O., Michelini, F.J., 1957. The plastochron index. Am. J. Bot. 44, 297–305.
Fleisher, D.H., Shillito, R.M., Timlin, D.J., Kim, S.H., Reddy, V.R., 2006. Approaches to modeling potato leaf appearance rate. Agron. J. 98, 522–528.
Génard, M., Pagès, L., Kervella, J., 1994. Relationship between sylleptic branching and components of parent shoot development in the peach tree. Ann. Bot. 74, 465–470, 1994.
Gompertz, B., 1825. On the nature of the function expressive of the law of human mortality, and on a new method of determining the value of life contigencies. Philos. Trans. Roy. Soc. 115, 513–583.
Gordon, D., Damiano, C., DeJong, T.M., 2006. Preformation in vegetative buds of *Prunus persica*: factors influencing number of leaf primordia in overwintering buds. Tree Physiol. 26, 537–544.
Gray, A., 1879. Structural Botany. Ivsion, Blakeman, Taylor and Company, New York.
Grossman, Y.L., DeJong, T.M., 1994. Carbohydrate requirements for dark respiration by peach vegetative organs. Tree Physiol. 14, 37–48.
Helliwell, C.A., Chin-Atkins, A.N., Wilson, I.W., Chapple, R., Dennis, E.S., Chaudhury, A., 2001. The Arabidopsis *AMP1* gene encodes a putative glutamate carboxypeptidase. Plant Cell 13, 2115–2125.
Hofstra, G., Hesketh, J.D., 1975. The effects of temperature and CO_2 enrichment on photosynthesis in soybean. In: Marcelle, R. (Ed.), Environmental and Biological Control of Photosynthesis. Springer, The Netherlands, pp. 71–80.
Kawakatsu, T., Itoch, J., Miyoshi, K., Kurata, N., Alvarez, N., Veit, B., Nagato, Y., 2006. PLASTOCHRON2 regulates leaf initiation and maturation in rice. Plant Cell 18, 612–625.
Kervella, J., Pagès, L., Génard, M., 1995. Genotypic differences in the leaf emergence rate of young peach trees. J. Am. Soc. Hortic. Sci. 120, 278–282.
Kirby, E.J.M., 1995. Factors affecting rate of leaf emergence in barley and wheat. Crop Sci. 35, 11–19.
Lambers, H., Chapin III, S.F., Pons, T.L., 2008. Plant Physiological Ecology, 2nd ed. Springer, New York, NY, pp. 321.
Longnecker, N., Robson, A., 1994. Leaf emergence of spring wheat receiving varying nitrogen supply at different stages of development. Ann. Bot. 74, 1–7.
Lopez, G., Favreau, R.R., Smith, C., Costes, E., Prusinkiewicz, P., DeJong, T.M., 2008. Integrating simulation of architectural development and source–sink behaviour of peach trees by incorporating Markov chains and physiological organ function submodels into L-PEACH. Funct. Plant Biol. 35, 761–771.
Mass, E.V., Grieve, C.M., 1990. Spike and leaf development in salt stressed wheat. Crop Sci. 30, 1309–1313.
Mathews, M., Anderson, M., Schultz, H., 1987. Phenologic and growth responses to early and late season water deficits in *Cabernet franc*. Vitis 26, 147–160.
McCutchan, H., Shackel, K.A., 1992. Stem-water potential as a sensitive indicator of water stress in prune trees (*Prunus domestica* L cv French). J. Am. Soc. Hortic. Sci. 117, 607–611.
McMaster, G.S., Wilhelm, W.W., 1997. Growing degree days: one equation, two interpretations. Agric. For. Meterol. 87, 291–300.
Miyoshi, K., Ahn, B.O., Kawakatsu, T., Ito, Y., Itoh, J., Nagato, Y., Kurata, N., 2004. PLASTOCHRON1, a timekeeper of leaf initiation in rice, encodes cytochrome P450. Proc. Natl. Acad. Sci. U. S. A. 101, 875–880.
Morgan, D.C., Warrington, I.J., Halligan, E.A., 1985. Effect of temperature and photosynthetic photon fluxdensity on vegetative growth of kiwifruit (*Actinidia chinensis*). N. Z. J. Agric. Res. 28, 109–116.
Mosaad, M.G., Oritz-Ferrara, G., Mahalakshmi, V., Fischer, R.A., 1995. Phyllochron response to vernalization and photoperiod in spring wheat. Crop Sci. 35, 168–171.
Pagès, L., Kervella, J., Genard, M., 1996. Modelling variations of metamer emergence rate in peach trees. Acta Hortic. 416, 29–38.
Prigge, M.J., Wagner, D.R., 2001. The *Arabidopsis* serrate gene encodes a zinc-finger protein required for normal shoot development. Plant Cell 13, 1263–1279.
Raper, C.D., Thomas, J.F., 1975. Temperatures in early post-transplant growth: Influence on leaf and floral initiation in tobacco. Crop Sci. 15, 732–733.
Rawson, H., 1993. Radiation effects on rate of development in wheat grown under different photoperiods and high and low temperatures. Aust. J. Plant Physiol. 20, 719–727.
Rawson, H., Hindmarsh, J., 1982. Effects of temperature on leaf expansion in sunflower. Aust. J. Plant Physiol. 9, 209–219.
Reinhardt, D., Pesce, E.R., Stieger, P., Mandel, T., Baltensperger, K., Bennett, M., Traas, J., Friml, J., Kuhlemeier, C., 2003. Regulation of phyllotaxis by polar auxin transport. Nature 426, 255–260.
Richardson, E.A., Schuyler, D.S., Walker, D.R., 1975. Pheno-climatography of spring peach bud development. HortScience 10, 236–237.
Rogers, H.H., Cure, J.D., Thomas, J.F., Smith, J.M., 1984. Influence of elevated CO_2 on growth of soybean plants. Crop Sci. 24, 361–366.

Schultz, H.R., 1992. An empirical model for the simulation of leaf appearance and leaf area development of primary shoots of several grapevine (*Vitis vinifera* L.). Sci. Hortic. 52, 179–200.
Seleznyova, A.N., Halligan, E., 2006. Modeling effect of temperature on area expansion at the leaf, the shoot, and the whole plant level. Acta Hortic. 707, 167–174.
Silk, W.K., 1980. Plastochron indices in cantaloupe grown on an irrigation line source. Bot. Gaz. 141, 73–78.
Slafer, G.A., 1995. Wheat development as affected by radiation at two temperatures. J. Agron. Crop Sci. 175, 249–263.
Slafer, G.A., Rawson, H.M., 1997. Phyllochron in wheat as affected by photoperiod under two temperature regimes. Aust. J. Plant Physiol. 24, 151–158.
Slafer, G.A., Halloran, G.H., Connor, D.J., 1994b. Development rate in wheat as affected by duration and rate of change of photoperiod. Ann. Bot. 74, 427–467.
Veit, B., Briggs, S.P., Schmidt, R.J., Yanofsky, M.F., Hake, S., 1998. Regulation of leaf initiation by the terminal ear 1 gene of maize. Nature 393, 166–168.
Villalobos, F.J., Ritchie, J.T., 1992. The effect of temperature on leaf emergence rates of sunflower genotypes. Field Crops Res. 29, 37–46.
Volk, T., Bugbee, B., 1991. Modeling light and temperature effects on leaf emergence in wheat and barley. Crop Sci. 31, 1218–1224.
White, J., 1979. The plant as a metapopulation. Ann. Rev. Ecol. Syst. 10, 109–145.
Wilhelm, W.W., McMaster, G.S., 1995. Importance of the phyllochron in studying the development and growth of grasses. Crop Sci. 35, 1–3.