Hyperbolic features of the circadian clock oscillations can explain linearity in leaf starch dynamics and adaptation of plants to diverse light and dark cycles

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ABSTRACT

Perfect resource management is the most important factor for plants to guaranty optimal growth and reproduction. To be constantly supplied with sucrose, their most important resource, plants store a part of it as transitory starch during daily photosynthesis, and degrade starch back into sucrose at night, when photosynthesis is not possible. Observations show that starch accumulation and degradation are linear, forming a pyramid shape, and that this shape adapt to various photoperiods to constantly supply the leaf with sucrose. Transitory starch buffer management is performed by a careful control of carbon partitioning, starch degradation and sucrose export key rates, to insure optimal growth whatever the photoperiod. Few theoretical studies have addressed this intriguing starch buffer management. Here we characterize the shape of the three key rates that allow sucrose homeostasis to persist in different photoperiods. We built two models of sucrose and starch dynamics including the three key rates formalized as piecewise continuous functions. First model assumed non-saturating starch degradation (linear model) whereas the second model assumed saturation of starch degradation activity by the substrate (saturating model). In order to obtain sucrose homeostasis we adjusted the shape of the three piecewise continuous rate functions using gradient descent on a sucrose homeostasis payoff function. At convergence of the gradient descent, we always obtained sucrose homeostasis together with the characteristic starch pyramid profile, as observed in empirical data. Surprisingly, in the linear model starch degradation rate at night followed a unique hyperbolic shape whatever the photoperiod, whereas for the saturating model degradation rate was constant at night but globally lower for shorter light periods. As light period shortened starch accumulated faster while degradation speed decreased, consistently with data. For shorter light periods, sucrose homeostasis was achieved at a level lower than the optimal one. We also reproduced an unexpected early dusk experiment in which plants adapted to 12 h light/12 h dark cycle experience earlier dusk at 8 h after dawn. Linear model could reproduce the instantaneous decrease in starch degradation speed during the longer night, as observed in data, whereas saturating model failed. Our results imply that a plant employing a unique hyperbolic starch degradation rate would be able to manage starch buffer in various photoperiod conditions, whereas a saturating starch degradation requires a simple daily constant rate but instantaneous regulation during sudden changes in photoperiod.

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1. Introduction

Although photosynthetic carbon assimilation occurs only in the light, carbon is required at all times for growth and respiration. Transitory starch reserve in leaves enables continuous supply of carbon throughout the day-night cycle (Smith and Stitt, 2007; Yazdanbakhsh et al., 2011). Starch is stored during daily photosynthesis inside chloroplasts and broken down at night to provide sugars to sinks, allowing metabolism and growth regardless of the presence of light (Gibon et al., 2004; Smith and Stitt, 2007; Graf and Smith, 2011). Previous studies have reported that starch is accumulated and is degraded almost linearly, rather than exponentially, drawing a pyramid shape in the day-night cycle (Gibon et al., 2004) (Fig. 1a). More interestingly, the pyramid shape adaptively changes in response to the various lengths of light period (Lu et al., 2005; Smith and Stitt, 2007; Graf et al., 2010; Graf and Smith, 2011). Starch degradation speed immediately decreases while its accumulation speed increases (Fig. 1a) when the light period is shortened from 16 h light/8 h dark to 8 h light/16 h dark (Lu et al., 2005; Graf et al., 2010; Graf and Smith, 2011) and there are no symptoms of carbon starvation throughout the subsequent longer nights (Graf et al., 2010). More surprisingly, when a plant initially placed in a

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a)

b)
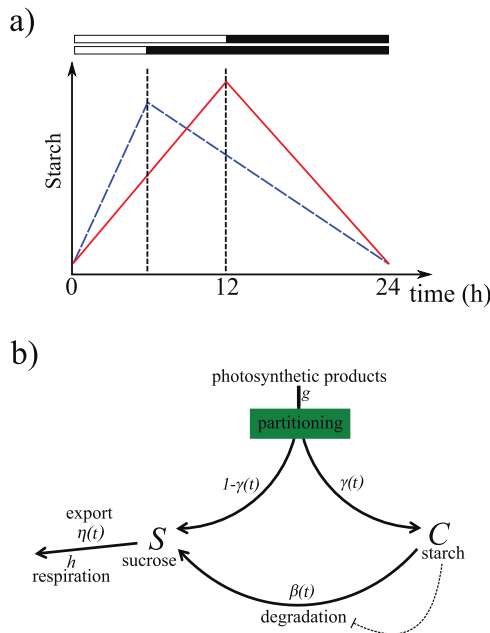
Fig. 1. (a) Typical starch diurnal pattern in long and short day and (b) flow chart of the model. (a) Scheme of observed starch diurnal pattern in long day (12 h/12 LD cycle; red) and short day (6 h/18 h LD cycle; blue) in plant leaves. The rate of starch synthesis is larger in short days while the degradation rate is smaller. This allows leaves to avoid running out of starch, and therefore sucrose, during night. (b) Flow chart of the model. Carbon is captured by photosynthesis with rate g, and a fraction $\gamma(t)$ is partitioned into starch while a fraction $1 - \gamma(t)$ is partitioned into sucrose. Starch is degraded into sucrose with rate $\beta(t)$. Degradation can also be inhibited by too high level of starch (dashed line) creating a saturated degradation. Sucrose is exported and respired with rates $\eta(t)$ and h, respectively. The three rate functions $\gamma(t)$, $\beta(t)$ and $\eta(t)$ can vary during the course of a 24 h cycle, but are assumed to be 24 h-periodic. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of the article.)

12 h light/12 h dark cycle experiences an unexpected early dusk at 8 h after dawn, an instantaneous decrease in starch degradation speed is observed, as if the plant knew that the night would be longer than usual because dusk came earlier. The mechanism behind the pyramid built by plants and adaptation to diverse photoperiods is mysterious and little is known about the significance of this characteristic shape.

Feugier and Satake (2013) previously proposed the hypothesis that the feedback between circadian clock and starch and sucrose metabolisms is the key mechanism to generate linearity in starch diurnal patterns and adaptation to various photoperiods. They developed a model linking the circadian clock to sucrose and starch dynamics. In the model, three key processes, carbon partitioning between sucrose and starch, starch degradation, and sucrose export, were described as cosine functions that approximate circadian oscillations. They demonstrated that cosine generated oscillations adjusted in time to minimize sucrose starvation are sufficient to reproduce almost all the starch profiles observed in experiments, showing that the circadian clock regulation may operate these key rates. However, when the photoperiod was as short as 8 h, the model predicted non-linear decay and depletion of starch before the end of the night, which was not consistent with the observations.

They explained that this discrepancy between the model and data would be caused by the choice of the cosine function to represent circadian oscillations: to create a perfect linear decay of starch at night degradation rate should follow an accelerating increase. Thus the portion of cosine function having this feature would produce a linear decay while others will make the decay diverge from linear.

In the present study we relax the assumption of cosine functions by using piecewise continuous rate functions whose shapes are undetermined. We also introduce a sucrose homeostasis objective function whose minimization will characterize the shape of the three piecewise continuous rate functions. This new framework allows decreasing the number of assumptions, making the model more general, and sucrose homeostasis introduces more objectively the dilemma between starch accumulation to prevent starvation at night, and sucrose export to promote growth. That is to say, the life time dilemma of all organisms: should I spare or should I grow?

Our aim is to characterize the optimal shape of the three time dependent key rates that would reproduce the characteristic pyramid shape of starch profile in various photoperiods without any assumption about starch profile itself.

2. Methods

2.1. Model for sucrose and starch dynamics in leaves

We start from the work from Feugier and Satake (2013). In their model the three key rates of carbon partitioning between sucrose and starch, starch degradation, and sucrose export are formalized by cosine functions summarizing the oscillating regulation from the clock. They estimated the maximum amplitude of the oscillations from available data, whereas the time shift was searched by minimization of sucrose starvation cost function using gradient descent.

Here we use sucrose and starch dynamics from Feugier and Satake (2013) (Fig. 1b), while the three rate functions will be redefined in next section. The gross production rate of sucrose by photosynthesis is the constant speed of carbon capture g weighted by the light availability function $L(t)$ taking value 1 for light and 0 for dark. We consider a constant carbon capture rate, rather than considering naturally varying light intensities, to be consistent with empirical studies. During light period a fraction $\gamma(t)$ of carbon assimilated by photosynthesis is partitioned into starch (C) – which accumulates in the leaf through the day – and a fraction $1 - \gamma(t)$ is partitioned into sucrose (S) – immediately available for growth and respiration. No partitioning occurs at night as there is no photosynthesis ($L(t) = 0$). Starch is degraded into sucrose with rate $\beta(t)$ providing the only source of sucrose to support leaf respiration and growth at night. Sucrose is exported with rate $\eta(t)$ to non-photosynthetic tissues such as roots and growing organs via the phloem. The three time dependent rates $\gamma(t)$, $\beta(t)$ and $\eta(t)$ are aggregate parameters of multiple processes (such as cascades of regulations; transcription and translation of various enzymes involved in metabolism) and are treated here as equivalent to the activity of carbon partitioning, activity of starch degradation, and activity of sucrose export, respectively.

Taken together, temporal dynamics of sucrose and starch concentrations in leaves are represented by the following equations (Fig. 1b):

$$\dot{S} = (1 - \gamma(t))gL(t) + \beta(t)C - (h + \eta(t))S, \qquad (1)$$

$$\dot{C} = \gamma(t)gL(t) - \beta(t)C. \qquad (2)$$

The rate of change in sucrose concentration (S) in (1) is equal to the fraction $1 - \gamma(t)$ of carbon flux that is distributed into sucrose pathway plus the flux of sucrose that comes from starch breakdown (with rate $\beta(t)$), minus the loss due to leaf maintenance and respiration (with constant rate h) and export to non-photosynthetic tissues (with rate $\eta(t)$). Similarly the rate of change in starch concentration (C) in (2) is equal to the complementary fraction $\gamma(t)$ of the carbon flux distributed into starch pathway, minus the loss due