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Report

# SPATULA Links Daytime Temperature and Plant Growth Rate

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## Summary

**Plants exhibit a wide variety of growth rates that are known to be determined by genetic and environmental factors, and different plants grow optimally at different temperatures [1, 2], indicating that this is a genetically determined character. Moderate decreases in ambient temperature inhibit vegetative growth, but the mechanism is poorly understood, although a decrease in gibberellin (GA) levels is known to be required [3]. Here we demonstrate that the basic helix-loop-helix transcription factor SPATULA (SPT), previously known to be a regulator of low temperature-responsive germination [4], mediates the repression of growth by cool daytime temperatures but has little or no growth-regulating role under warmer conditions. We show that only daytime temperatures affect vegetative growth and that SPT couples morning temperature to growth rate. In seedlings, warm temperatures inhibit the accumulation of the SPT protein, and SPT autoregulates its own transcript abundance in conjunction with diurnal effects. Genetic data show that repression of growth by SPT is independent of GA signaling and phytochrome B, as previously shown for PIF4 [5]. Our data suggest that SPT integrates time of day and temperature signaling to control vegetative growth rate.**

## Results and Discussion

In order to learn more about growth repression by temperature, we screened known and unknown *Arabidopsis* mutants for the ability to grow fast at moderately low ambient temperatures (here 15°C) while leaving growth rate at higher temperatures (25°C) unaffected, using total rosette leaf area as a measure (this correlates well with fresh weight but can be monitored nondestructively; see Figure S1 available online). We found one such mutant, known as *spatula-2* (*spt-2*), a lesion in a basic helix-loop-helix transcription factor previously characterized for its role in fruit development and temperature-responsive seed germination control [4, 6, 7]. The *spt-2* mutants exhibited near-normal growth at 25°C, but when grown at 15°C, their growth was up to double that of wild-type (Figure 1). We also examined the growth rate of our HA-tagged *SPT* overexpressing lines [4], which complement the germination and growth defects of *spt* mutants. These exhibited wild-type growth rates at constant 25°C but grew as dwarfs when maintained at 15°C (Figures 1A and 1B). The fast growth phenotype of *spt-2* mutants at 15°C was shared with two further *spt* alleles, including *spt-11*, a loss-of-function T-DNA insertion line [8], and *spt-3*, although not by *spt-1*, known to be a weaker allele [6]. This phenotype comprised both increased leaf organ size and leaf initiation rates (Figure S1), coupled with improved fresh weight gain (Figures 1D and 1E). Therefore, we concluded that SPT is a growth repressor with a role in the establishment of the relationship between ambient temperature and growth rate, with increasing SPT levels sensitizing the plant's growth response to decreasing temperature. Analysis of the ability of *spt* mutant plants to withstand subzero temperatures showed that there was no obvious difference from wild-type, suggesting that modification of the SPT-dependent pathway could be used to promote growth in plants grown in field conditions in regions where cool conditions are prevalent (Figure S2).

Because the *spt* growth phenotype is most evident at low ambient temperatures, it seemed likely that either the SPT protein level or activity is increased at these temperatures. Interrogation of published data revealed few changes in SPT expression in response to cold treatments of 4°C except that the highest expression levels occurred later than in the controls [9]. Another study showed a diurnal and circadian regulation of *SPT* transcript abundance, with transcript levels increasing during the light period and decreasing at night [10]. We confirmed this pattern using real-time reverse transcriptase-polymerase chain reaction (RT-PCR) in vegetative tissues under our light/dark conditions (Figure 2A). In addition, we observed a striking interaction between time of day and ambient temperature in the regulation of *SPT* transcript levels. At 25°C, *SPT* levels rose during the day but then declined abruptly at the end of the light period. As the temperature was lowered, the phase and duration of *SPT* expression was altered, so that at 15°C the transcript levels showed an additional and larger peak 4 hr after dusk, only decreasing close to dawn. Interestingly, in the *spt-2* mutant, SPT mRNA shows no temperature-responsive phase shift (Figure S3A). This shows that the activity of the wild-type SPT protein mediates the control of the phase of SPT mRNA oscillations by temperature.

To investigate whether temperature affects the SPT protein, we used a previously characterized epitope-tagged line expressed from the 35S promoter [4]. We found that this had markedly increased abundance at 15°C compared to 25°C (Figures 2B and 2C). Measuring at the SPT mRNA levels in these overexpressing plants revealed only a slight increase in transcript levels at 15°C, and only during the dark period (Figure S3B), showing that the control of SPT abundance by temperature is predominantly posttranslational. This substantial temperature regulation of SPT protein levels explains the specificity of the growth phenotype of the overexpressors to low ambient temperatures (Figure 1A). It is possible that temperature controls the abundance of the SPT protein through interaction with a further factor or that temperature regulates an unknown posttranslational modification of SPT. Interestingly, the abundance of SPT at 15°C also shows

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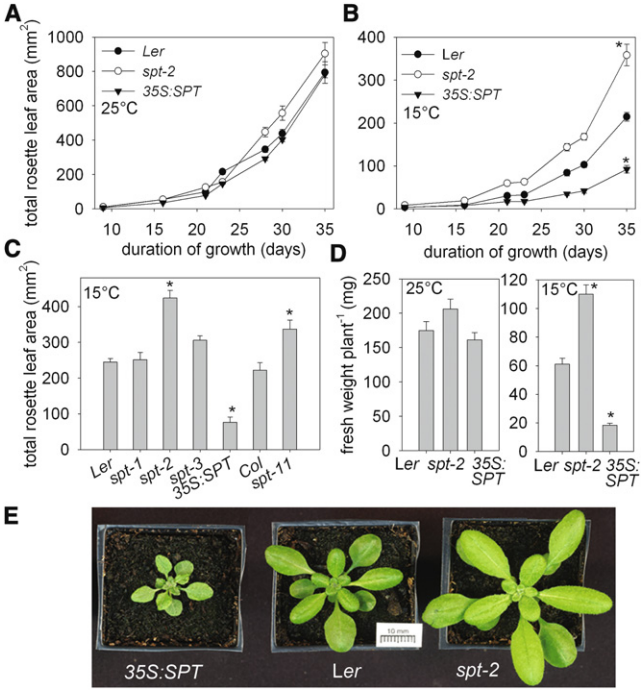


Figure 1. SPT Is a Temperature-Specific Growth Regulator in *Arabidopsis*

(A and B) The growth rates of *SPT* lines at 25°C (A) and 15°C (B), based on total rosette leaf area. The growth rates of the wild-type accession L*er* are also shown in this and subsequent charts.

(C) Rosette leaf area of *spt* mutants and the wild-type accessions L*er* and Col after 36 days of growth.

(D) Biomass (fresh weight) determinations after 36 days of growth.

(E) Overhead pictures of representative 32-day-old plants growing in 12 hr light/dark cycles at 15°C. All data represent the mean and standard error of 10 plants measured per genotype. *$p < 0.05$.

a significant but temporary reduction in the early part of the day. At 25°C, protein levels are much lower at all time points but appear to show a transient increase at dusk. Combining our transcript and protein data, we would expect that at low ambient temperatures SPT would show a predominantly transcriptionally driven increase close to dusk, with protein levels remaining high until just after dawn.

We also analyzed the growth of *Arabidopsis* leaves at 15°C (Figure S3C). We could show that, as has previously been reported for hypocotyls [11, 12], leaves elongated rhythmically with a peak growth rate at dawn in 12 hr light, 12 hr dark cycles. Interestingly, the *spt-2* mutant grew at an increased rate at dawn and during the light period, but the minimum growth rate observed toward the end of the night was similar to that measured in wild-type plants. Therefore, we concluded that *SPT* expression determined the amplitude but not the timing of rhythmic growth. The growth rates of *35S:SPT* plants at 15°C were too low to be measured, consistent with their dwarf phenotype (Figure 1). It is interesting that the time of day at which SPT appears to have the greatest effect on growth rate does not correspond to the peak of *SPT* expression. This suggests that SPT is not directing the rhythmic regulation of growth.

It has previously been observed that leaf length is slightly decreased with increasing night temperatures [13] and that a high night temperature combined with a cooler day temperature inhibits hypocotyl and pea stem growth, whereas low night temperatures and warmer day temperatures promote growth [3]. However, in certain crop plants, night temperature appears to have little or no effect on growth [14]. Given that *SPT* expression peaks toward the afternoon or dusk, we were interested in the role time of day plays in the temperature regulation of *Arabidopsis* vegetative growth. Hence, we grew *Arabidopsis* plants at 15°C and interrupted the cool temperature with daily timed 8 hr periods of 25°C spanning the middle of the day, the middle of the night, dawn, or dusk (Figure 2D). Strikingly, a 25°C warm pulse was very effective at promoting wild-type growth during the day but was ineffective when applied at night, suggesting that *Arabidopsis* rosette growth is coupled to daytime temperatures. Interestingly, this result is predicted by a recent study modeling *Arabidopsis* growth in real environmental situations [15]. These data also show that *Arabidopsis* has only a weak thermoperiodic growth response [16], because the daytime warm treatment promotes growth only marginally faster than constant warmth alone (compare Figure 1D and Figure 2D). The dusk and dawn warm treatments showed growth intermediate between constant 15°C and the daytime warm treatment; however, *Arabidopsis* growth was significantly more responsive to warmth at dusk than at dawn. We found that the *spt-2* mutant continued to show elevated growth rates compared to wild-type except when warmth was applied during the day, when no difference was observed between wild-type and *spt-2* growth rates. In addition, at dusk, *spt-2* mutant plants are strongly hypersensitive to applied warmth, and *spt-2* mutants grew at the same rate as when warmth was applied in the middle of the day. Therefore, there is a daily cycle in the temperature sensitivity of growth, and SPT has a role in growth repression when daytime temperatures are cool and when warmth is applied only toward dusk. Interestingly, *35S:SPT* plants continued to remain dwarfed at whatever time high temperatures were applied, and the diurnal nature of temperature-responsive growth was reduced. Given that constant 25°C treatment restores *35S:SPT* growth to wild-type levels (Figures 1A and 1D), we conclude that the constitutive presence of high *SPT* abundance can confer a requirement for prolonged warmth for normal growth promotion. Because *spt* mutants remain sensitive to the temperature regulation of growth, particularly at dusk, SPT most likely cooperates with further factors that act to maintain the daily relationship between temperature and growth rate.

To further understand the discrepancy between the effectiveness of dawn and dusk warm treatments, we gave daily shorter 4 hr pulses of 25°C, timed across the day and early night, to plants otherwise maintained at 15°C (Figure 2E). Interestingly, 4 hr warm pulses applied at different times during the light period had identical growth-promoting effects on wild-type plants, in contrast to the effect of the 8 hr pulses (Figure 2D), in which increased sensitivity had been observed at dusk compared to dawn. Furthermore, a 4 hr warm pulse after dusk had no growth-promoting effect, in common with the 8 hr warm pulses applied during the night. Given that the 8 hr treatment that includes 4 hr predusk and 4 hr postdusk warm treatments showed significantly more growth-promoting potential than the 4 hr predusk treatment alone, we concluded that *Arabidopsis* growth is sensitive to warmth during the light period and also in the period immediately after dusk, providing that warmth was first experienced in the period prior to dusk.

We also found that the 4 hr postdawn treatment was equally as effective as the 8 hr treatment applied equally 4 hr before

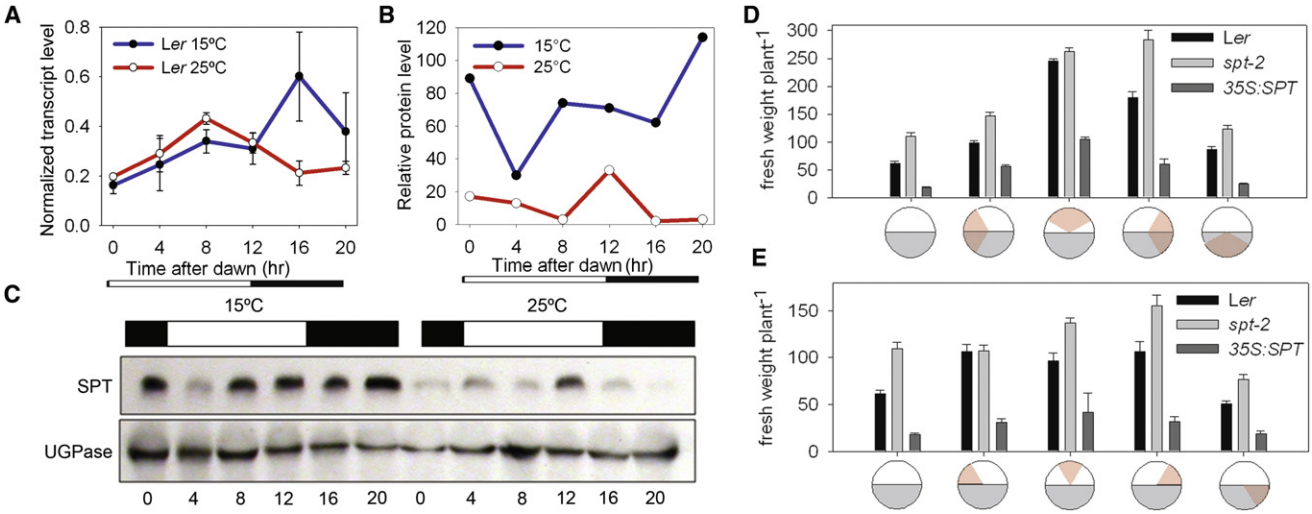


Figure 2. SPT Integrates Time-of-Day and Temperature Information

(A) The transcript levels of *SPT* in 3-week-old wild-type (*Ler*) plants grown at either constant 15°C or 25°C. Data represent the mean of three biological replicates at each time point.

(B and C) Detection of SPT protein from *35S:SPT*-overexpressing plants using the HA tag.

(B) A quantitative description of the data presented in (C).

(C) Top: SPT; bottom: UGPase loading control.

(D) The effect of daily 8 hr long pulses of 25°C on the growth of plants otherwise maintained at 15°C in 12 hr light/dark cycles. The position of the red wedge indicates the time and duration of the 25°C treatment in the daily cycle.

(E) As in (D) above, except that daily 4 hr 25°C pulses were given to plants grown otherwise at 15°C. Fresh weights were determined on day 36 of growth and represent the mean and standard error of 10 plants per genotype.

and 4 hr after dawn (compare Figures 2D and 2E). This suggests that the warmth experienced before dawn in the latter treatment produced no growth response. Therefore, we concluded that the 8 hr warm period during the day was best at promoting growth, not because of special significance of this time of day, but because the warm treatment occurred entirely within the light period. Together, these results show that the daily light period is critical for the generation of a growth-modifying temperature signal in *Arabidopsis*, as has previously been shown in other plants [17].

For the following reasons, we conclude that the action of SPT is to repress growth in response to cool ambient temperatures, particularly when this temperature is experienced by the plant during the earlier part of the light period. First, the 4 hr warm treatment in the morning promotes wild-type growth to that of *spt-2* but does not affect *spt-2* growth (Figure 2E): this shows that warm temperatures and loss of SPT cause an equivalent and epistatic loss of growth repression at this time of day. Second, *spt-2* plants increase growth in response to 4 hr or 8 hr warm periods later in the light period (Figures 2D and 2E). This is likely because in these treatments, without SPT, there is a lack of growth repression in response to cool morning temperatures, combined with growth promotion by warmth later in the day, and the latter is most likely signaled by other factors acting partially redundantly with SPT. It is interesting that in the morning, *SPT* transcript levels start to rise (Figure 2A), and presumably the phase of SPT expression is set.

Gibberellin (GA) has been shown to be important in thermoperiodic stem elongation responses [3, 18]. The main effects of GA are mediated through the controlled degradation of growth-repressing DELLA proteins [19]. SPT is a member of the PHYTOCHROME INTERACTING FACTOR (PIF) subfamily of bHLH transcription factors, and these have been shown to influence plant development by association with the phytochrome- and DELLA-dependent pathways [20–22], although SPT does not bind phytochrome B [23]. Furthermore, phytochrome, as well as DELLA, is implicated in *Arabidopsis* growth control [24–26], and both have been implicated in the control of temperature-dependent processes [27, 28]. To establish whether these proteins played a role in SPT-mediated growth inhibition, we crossed *spt-2* and *35S:SPT* to the *gai* gain-of-function mutant [29] and *spt-2* to *phyB-1* (Figures 3A and 3B). Analysis of these mutant combinations revealed that SPT and DELLA action was additive in all cases, with *35S:SPT gai* plants smaller than either *gai* or *35:SPT* alone and *spt-2 gai* plants larger than *gai* alone, although still markedly slower growing than *spt-2*. Further supporting this hypothesis, applied exogenous GA has a similar growth-promoting effect on both wild-type and *35S:SPT* plants (Figure 3D). The *spt-2 phyB-1* plants showed characteristics of both *spt-2* mutants and *phyB-1* mutants, exhibiting the large *spt* leaves, but with elongated petioles from *phyB-1*. So, again we found that the two mutants showed an additive affect on growth. We therefore concluded that SPT represses growth through a DELLA- and phyB-independent pathway. A similar conclusion was reached for the function of PIF4 [5], which, like SPT, shows a temperature-responsive growth phenotype, although this phenotype occurs at high rather than low temperatures with PIF4 [5, 30]. It is likely that different members of this transcription factor family act at different temperatures to modulate growth.

We have shown that *Arabidopsis* rosette growth is responsive to warmth during the day, but not during the night. Our data are consistent with a model in which SPT is a growth repressor whose levels are determined primarily by temperature in a process that acts at the protein level. Endogenous *SPT* transcript levels also show regulation by time of day and temperature, with the activity of the SPT protein required for

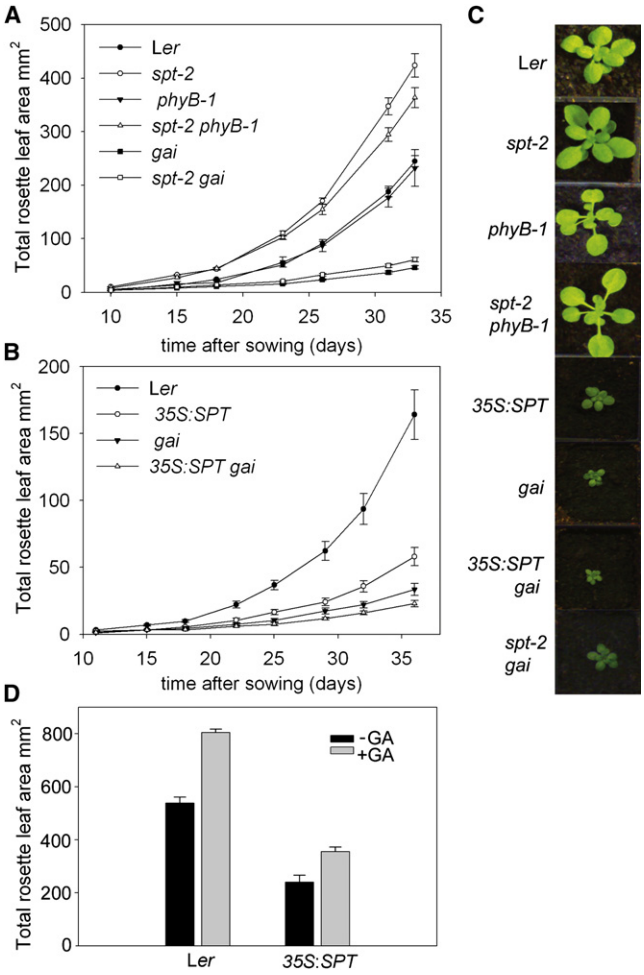


Figure 3. SPT Functions Additively to phyB and DELLAs in Growth Repression

(A and B) Growth curves showing the mean rosette leaf area and standard error for each genotype grown at 15°C. These data represent the mean and standard error of 10 plants measured per genotype.

(C) Representative 27-day-old plants growing at 15°C for each genotype.

(D) *SPT*-overexpressing plants have a wild-type growth response to gibberellin application. Plants grown at 15°C were sprayed weekly with 100 μM GA, starting in week 2 after transfer to soil, and growth was monitored as total rosette leaf area. These data represent the mean leaf area of 20 plants per treatment after 47 days. Error bars indicate the standard error.

the regulation of the phase of *SPT* transcription by temperature. The daily increase of *SPT* expression coincides with the time at which SPT is most important for growth control by temperature. Meanwhile, constitutive expression of SPT confers a requirement for constant warmth for growth promotion by high temperatures. Therefore, the increased growth phenotype of *spt* mutants at 15°C can be interpreted as exhibiting a constitutive response to morning warmth in the absence of the signal, which leads to faster but still rhythmic growth. One possibility is that temperature during the morning is important for the autoregulation of the phase of *SPT* expression, because this is the time of day that SPT transcription starts to rise. In this scenario, low morning temperatures would increase *SPT* transcription at the later time of day when the SPT protein is apparently most stable (Figures 2B and 2C). Our work shows that, alongside the transcriptional induction of PIF4 [5], the regulation of SPT protein levels at the translational or posttranslational level coordinates the plant's growth response to temperature. However, because *spt* mutants show an attenuated but not absent diurnal pattern of growth responses to temperature and because *spt* mutants retain a growth response to warmth, SPT must cooperate with further factors in the maintenance of the daily temperature-response rhythm. This work gives important insights into a new mechanism through which plants control temperature signaling, regulating key developmental programs.

## Experimental Procedures

The *spt-1*, *spt-2*, *spt-3* [6], *spt-11* [8], and *SPT* overexpressing plants in a wild-type background and including the HA epitope tag [4] have been described previously. The wild-type accessions used were Columbia (Col) and Landsberg erecta (L*er*), as stated. Plants were sown first on Murashige and Skoog (MS) agar plates and transferred to 40-well trays of John Innes No. 2 compost after the first 7–10 days of growth. Growth conditions were 12 hr white light (75–100 $\mu molm^{-2}s^{-1}$), 12 hr dark at 70%–80% relative humidity. Recording of growth by leaf area began on day 10, 11, or 12 using a CreativeLive webcam. At the growth stages investigated here, there is very little overlap between leaves, meaning that measurement of leaf area in this way can be used to obtain an accurate measure of total leaf area. Total rosette area was calculated for a total of 10 plants per genotype using ImageJ. Experiments were terminated at the first sign of flowering, because this marks the cessation of exponential growth [2].

RNA was isolated from 12-day-old seedlings grown at either 15°C or 25°C (identical conditions to the growth experiments) using a QIAGEN RNeasy kit, and *SPT* transcripts were measured by real-time RT-PCR as described previously [4] and were normalized to mean expression of both *UBIQUITIN 10* and *TUBULIN 2* control genes. These have been used as the internal control for RT-PCR in several studies, including several looking into effects of temperature and circadian clock [31–34]. Protein was isolated from seedlings grown for 7 days at 22°C and then switched to either 15°C or 25°C for 5 days, using the protocol of Duek et al. [35]. The HA tag was detected by probing the membrane with a rat anti-HA antibody (3F10, Roche) at a dilution of 1:1000 followed by a HRP-conjugated sheep anti-rat (Abcam) at a dilution of 1:5000. Loading was checked by directly reprobing membranes using a goat anti-UGPase antibody (Agrisera). Protein levels were quantified using the histogram function of Adobe Photoshop.

## Supplemental Information

Supplemental Information includes three figures and can be found with this article online at doi:10.1016/j.cub.2010.07.028.

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## References

1. Briggs, G., Kidd, F., and West, C. (1920). A quantitative analysis of plant growth. Ann. Appl. Biol. *7*, 202–223.
2. Grime, J., and Hunt, R. (1975). Relative growth rate: Its range and adaptive significance in a local flora. J. Ecol. *69*, 393–422.
3. Stavang, J.A., Lindgård, B., Erntsen, A., Lid, S.E., Moe, R., and Olsen, J.E. (2005). Thermoperiodic stem elongation involves transcriptional regulation of gibberellin deactivation in pea. Plant Physiol. *138*, 2344–2353.
4. Penfield, S., Josse, E.-M., Kannangara, R., Gilday, A.D., Halliday, K.J., and Graham, I.A. (2005). Cold and light control seed germination

through the bHLH transcription factor SPATULA. Curr. Biol. *15*, 1998–2006.

5. Koini, M.A., Alvey, L., Allen, T., Tilley, C.A., Harberd, N.P., Whitelam, G.C., and Franklin, K.A. (2009). High temperature-mediated adaptations in plant architecture require the bHLH transcription factor PIF4. Curr. Biol. *19*, 408–413.
6. Alvarez, J., and Smyth, D.R. (1999). CRABS CLAW and SPATULA, two Arabidopsis genes that control carpel development in parallel with AGAMOUS. Development *126*, 2377–2386.
7. Heisler, M.G.B., Atkinson, A., Bylstra, Y.H., Walsh, R., and Smyth, D.R. (2001). SPATULA, a gene that controls development of carpel margin tissues in Arabidopsis, encodes a bHLH protein. Development *128*, 1089–1098.
8. Ichihashi, Y., Horiguchi, G., Gleissberg, S., and Tsukaya, H. (2010). The bHLH transcription factor SPATULA controls final leaf size in Arabidopsis thaliana. Plant Cell Physiol. *51*, 252–261.
9. Kilian, J., Whitehead, D., Horak, J., Wanke, D., Weinl, S., Batistic, O., D'Angelo, C., Bornberg-Bauer, E., Kudla, J., and Harter, K. (2007). The AtGenExpress global stress expression data set: Protocols, evaluation and model data analysis of UV-B light, drought and cold stress responses. Plant J. *50*, 347–363.
10. Michael, T.P., Mockler, T.C., Breton, G., McEntee, C., Byer, A., Trout, J.D., Hazen, S.P., Shen, R., Priest, H.D., Sullivan, C.M., et al. (2008). Network discovery pipeline elucidates conserved time-of-day-specific cis-regulatory modules. PLoS Genet. *4*, e14.
11. Dowson-Day, M.J., and Millar, A.J. (1999). Circadian dysfunction causes aberrant hypocotyl elongation patterns in Arabidopsis. Plant J. *17*, 63–71.
12. Nozue, K., Covington, M.F., Duek, P.D., Lorrain, S., Fankhauser, C., Harmer, S.L., and Maloof, J.N. (2007). Rhythmic growth explained by coincidence between internal and external cues. Nature *448*, 358–361.
13. Thingnaes, E., Torre, S., Ernstsen, A., and Moe, R. (2003). Day and night temperature responses in Arabidopsis: Effects on gibberellin and auxin content, cell size, morphology and flowering time. Ann. Bot. (Lond.) *92*, 601–612.
14. Frantz, J.M., Cometti, N.N., and Bugbee, B. (2004). Night temperature has a minimal effect on respiration and growth in rapidly growing plants. Ann. Bot. (Lond.) *94*, 155–166.
15. Wilczek, A.M., Roe, J.L., Knapp, M.C., Cooper, M.D., Lopez-Gallego, C., Martin, L.J., Muir, C.D., Sim, S., Walker, A., Anderson, J., et al. (2009). Effects of genetic perturbation on seasonal life history plasticity. Science *323*, 930–934.
16. Went, F. (1944). Plant growth under controlled conditions. II. Thermoperiodicity in growth and fruiting of the tomato. Am. J. Bot. *31*, 135–150.
17. Erwin, J.E., and Heins, R.D. (1995). Thermomorphogenic responses in stem and leaf development. HortScience *30*, 940–949.
18. Grindal, G., Ernstsen, A., Reid, J.B., Junttila, O., Lindgård, B., and Moe, R. (1998). Endogenous gibberellin A1 levels control thermoperiodic stem elongation in Pisum sativum. Physiol. Plant. *102*, 523–531.
19. Jiang, C., and Fu, X. (2007). GA action: Turning on de-DELLA repressing signaling. Curr. Opin. Plant Biol. *10*, 461–465.
20. Ni, M., Tepperman, J.M., and Quail, P.H. (1998). PIF3, a phytochrome-interacting factor necessary for normal photoinduced signal transduction, is a novel basic helix-loop-helix protein. Cell *95*, 657–667.
21. de Lucas, M., Davière, J.M., Rodríguez-Falcón, M., Pontin, M., Iglesias-Pedraz, J.M., Lorrain, S., Fankhauser, C., Blázquez, M.A., Titarenko, E., and Prat, S. (2008). A molecular framework for light and gibberellin control of cell elongation. Nature *451*, 480–484.
22. Feng, S., Martinez, C., Gusmaroli, G., Wang, Y., Zhou, J., Wang, F., Chen, L., Yu, L., Iglesias-Pedraz, J.M., Kircher, S., et al. (2008). Coordinated regulation of Arabidopsis thaliana development by light and gibberellins. Nature *451*, 475–479.
23. Khanna, R., Huq, E., Kikis, E.A., Al-Sady, B., Lanzatella, C., and Quail, P.H. (2004). A novel molecular recognition motif necessary for targeting photoactivated phytochrome signaling to specific basic helix-loop-helix transcription factors. Plant Cell *16*, 3033–3044.
24. Reed, J.W., Nagpal, P., Poole, D.S., Furuya, M., and Chory, J. (1993). Mutations in the gene for the red/far-red light receptor phytochrome B alter cell elongation and physiological responses throughout Arabidopsis development. Plant Cell *5*, 147–157.
25. Peng, J., Carol, P., Richards, D.E., King, K.E., Cowling, R.J., Murphy, G.P., and Harberd, N.P. (1997). The Arabidopsis GAI gene defines a signaling pathway that negatively regulates gibberellin responses. Genes Dev. *11*, 3194–3205.
26. Achard, P., Cheng, H., De Grauwe, L., Decat, J., Schoutteten, H., Moritz, T., Van Der Straeten, D., Peng, J., and Harberd, N.P. (2006). Integration of plant responses to environmentally activated phytohormonal signals. Science *311*, 91–94.
27. Halliday, K.J., Salter, M.G., Thingnaes, E., and Whitelam, G.C. (2003). Phytochrome control of flowering is temperature sensitive and correlates with expression of the floral integrator FT. Plant J. *33*, 875–885.
28. Achard, P., Gong, F., Cheminant, S., Alioua, M., Hedden, P., and Genschik, P. (2008). The cold-inducible CBF1 factor-dependent signaling pathway modulates the accumulation of the growth-repressing DELLA proteins via its effect on gibberellin metabolism. Plant Cell *20*, 2117–2129.
29. Koornneef, M., Elgersma, A., Hanhart, C.J., van Loenen-Martinet, E.P., van Rign, L., and Zeevaart, J.A.D. (1985). A gibberellin insensitive mutant of *Arabidopsis thaliana*. Physiol. Plant. *65*, 33–39.
30. Stavang, J.A., Gallego-Bartolomé, J., Gómez, M.D., Yoshida, S., Asami, T., Olsen, J.E., García-Martínez, J.L., Alabadí, D., and Blázquez, M.A. (2009). Hormonal regulation of temperature-induced growth in Arabidopsis. Plant J. *60*, 589–601.
31. Gould, P.D., Locke, J.C.W., Larue, C., Southern, M.M., Davis, S.J., Hanano, S., Moyle, R., Milich, R., Putterill, J., Millar, A.J., and Hall, A. (2006). The molecular basis of temperature compensation in the *Arabidopsis* circadian clock. Plant Cell *18*, 1177–1187.
32. Wang, Z.-Y., and Tobin, E.M. (1998). Constitutive expression of the CIRCADIAN CLOCK ASSOCIATED 1 (CCA1) gene disrupts circadian rhythms and suppresses its own expression. Cell *93*, 1207–1217.
33. Blázquez, M.A., Ahn, J.H., and Weigel, D. (2003). A thermosensory pathway controlling flowering time in *Arabidopsis thaliana*. Nat. Genet. *33*, 168–171.
34. Koo, S.C., Bracko, O., Park, M.S., Schwab, R., Chun, H.J., Park, K.M., Seo, J.S., Grbic, V., Balasubramanian, S., Schmid, M., et al. (2010). Control of lateral organ development and flowering time by the Arabidopsis thaliana MADS-box Gene AGAMOUS-LIKE6. Plant J. *62*, 807–816.
35. Duek, P.D., Elmer, M.V., van Oosten, V.R., and Fankhauser, C. (2004). The degradation of HFR1, a putative bHLH class transcription factor involved in light signaling, is regulated by phosphorylation and requires COP1. Curr. Biol. *14*, 2296–2301.