

Blue light regulated shade avoidance

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Most plants grow in dense vegetation with the risk of being out-competed by neighboring plants. These neighbors can be detected not only through the depletion in light quantity that they cause, but also through the change in light quality, which plants perceive using specific photoreceptors. Both the reduction of the red:far-red ratio and the depletion of blue light are signals that induce a set of phenotypic traits, such as shoot elongation and leaf hyponasty, which increase the likelihood of light capture in dense plant stands. This set of phenotypic responses are part of the so called shade avoidance syndrome (SAS). This addendum discusses recent findings on the regulation of the SAS of *Arabidopsis thaliana* upon blue light depletion. Keller et al. and Keuskamp et al. show that the low blue light attenuation induced shade avoidance response of seedling and rosette-stage *A. thaliana* plants differ in their hormonal regulation. These studies also show there is a regulatory overlap with the R:FR-regulated SAS.

Plants perceive the threat of competing neighbors through various signals, such as specific changes in the light quality. Plants carry sophisticated photoreceptor systems to perceive these signals and subsequently activate a complex network of various hormones and transcriptional regulators.^{1–4} Perception of competition signals results in increased growth of the hypocotyl, stem and petioles, and an increased leaf angle (hyponasty), which are all part of the so-called shade avoidance syndrome (SAS).⁵ Plants use specific changes in the light quality as signals of potential or

actual competition for light. A reduction in blue light fluence rate is an indicator of actual shading, whereas the reduction in the red (R) to far-red (FR) ratio (R:FR) of canopy light can be used by plants as an early warning signal of future competition.⁶ Leaves absorb blue and R light for photosynthesis, whereas FR radiation is not absorbed and it is either transmitted or reflected. A significant body of work has concentrated on the mechanisms and ecological implications of the R:FR-regulated SAS responses.⁵ In contrast, the role of blue light signals in the control of plant developmental plasticity has received only limited attention. This addendum discusses the hormonal regulation of the SAS responses of *Arabidopsis thaliana* plants to blue light depletion based on two recent papers.^{7,8}

Photoreceptors Mediate the Shade Avoidance Response

Plants carries specific photoreceptors to detect the blue, R and FR radiation. The photoreceptor families of cryptochromes and phototropins, are sensitive to changes in blue light, whereas phytochromes are sensitive to R and FR radiation.^{9–12} Until now most research was focused on the R:FR and thus phytochrome regulated SAS. Recent studies showed that cryptochromes are not only involved in blue light de-etiolation and phototropism of seedlings,^{13,14} but are also the major mediators in blue light regulated SAS.^{7,15}

In the study of Keller et al.⁷ petiole elongation and hyponasty of rosette-stage *A. thaliana* plants were investigated, whereas in Keuskamp et al.⁸ hypocotyl elongation of *A. thaliana* seedlings was used as a readout of SAS. Interestingly,

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both studies found that blue light attenuation induced robust SAS responses, which in many regards resembled the phenotypes of seedlings and plants grown under low R:FR ratio.^{7,8} In these studies light treatments were used that had reduced blue light fluence rates (low blue) through wavelength-specific filters and these were compared against control light conditions. Keller et al.⁷ showed that *cry1* is required for the SAS response to low blue for rosette plants, whereas in seedlings, elongation responses to blue light depletion are controlled by both *cry1* and *cry2*.¹⁵ The mechanisms that mediate the SAS responses induced by *cry* inactivation have been much less intensively studied than those involved in the regulation of phyB responses, and the papers of Keuskamp et al.⁸ and Keller et al.⁷ have addressed the involvement of several hormones known to participate in the control of elongation.

The Hormonal Regulation of the Low-Blue-Induced SAS

Auxin is a well-studied plant hormone that has been associated with various elongation responses, such as low R:FR-induced SAS¹⁶⁻¹⁸ and is known to be regulated by cryptochrome as well.¹⁹ However, its role in plant responses to blue light attenuation has not been fully investigated. Auxin biosynthesis under the control of *TRYPTOPHAN AMINOTRANSFERASE OF ARABIDOPSIS1 (TAA1)* is upregulated in response to low R:FR.¹⁷ Mutations in the *TAA1* gene, such as in the *sav3-2* or *wei8-1*, prevent SAS induction by low R:FR.^{17,18,20} In Keuskamp et al.⁸ it was shown that *wei8-1* had a reduced (but still significant) elongation response to blue light attenuation. Likewise, the *sav3-2* mutant at the rosette stage displayed a nearly normal petiole elongation response to blue light attenuation, and retained a full leaf hyponastic response.⁷

In a previous study, Keuskamp et al.¹⁸ demonstrated that Polar Auxin Transport (PAT) is required for the elongation response to low R:FR of petioles and hypocotyls. They showed that low R:FR treatments produced a change in the sub-cellular localization of PIN-FORMED3 (PIN3), which is a facilitator of auxin transport.²¹ This relocation would lead to

a change in PAT, leading to auxin accumulation throughout the hypocotyl and eventually an increased elongation rate. Seedlings of the *pin3-3* mutant failed to respond to a low R:FR treatment with auxin accumulation in the hypocotyl and hypocotyl elongation.¹⁸ These data show the significance of auxin transport in SAS response elicited by low R:FR, as proposed by Morelli and Ruberti.¹⁶ Interestingly, PAT can be regulated by cryptochrome.¹⁹

Inhibition of auxin transport (or blocking the auxin perception) in seedlings, did not fully inhibit the elongation response induced by blue light attenuation⁸ and, in the rosette phase, *pin3-3* plants showed completely normal SAS responses to reduced blue light levels.⁷ The observed inhibition of the petiole elongation response upon blue light attenuation by 1-N-Naphthylphthalamic acid (NPA), an inhibitor of PAT, could imply involvement of other PINs,⁷ as was shown for seedlings.⁸ Interestingly, inhibiting PAT did not affect low blue-induced hyponasty, indicating that these two responses are regulated through partly separate mechanisms.

Recent studies have shown that brassinosteroids (BR) also play a role in the SAS response of *A. thaliana* plants triggered by phyB inactivation.^{8,22} Furthermore, auxin and brassinosteroids (BR) are linked to many of the same growth processes, including cell elongation which is the driving force behind hypocotyl elongation.²³ BRs were found to be required for the elongation response of seedlings to blue light attenuation. When BR biosynthesis or perception was blocked, the elongation response to low blue was reduced. Interestingly, only when both auxin and BR were blocked simultaneously, the response to low blue was fully inhibited.⁸ In petioles, the combined involvement was not studied, but BR appeared to be more important for petiole elongation than for leaf hyponasty.⁷

These data show that the hypocotyl elongation response upon low blue treatment can be fully explained by the combined action of auxin and brassinosteroid,⁸ whereas the leaf morphology responses of rosette-stage plants do not seem to be regulated in the same way.⁷ Interestingly, Both auxin and BR

are important regulators of the petiole elongation and hyponasty responses elicited by low R:FR.^{18,20,22} In short, this means that the hormonal regulation of SAS is not only organ- and/or developmental stage-dependent but also photoreceptor-dependent.

GA is a key regulator of cell elongation, and an important player in SAS responses.^{2,5,15,24} Low R:FR enhances both GA biosynthesis²⁵ and responsiveness,²⁶ representing a direct link between GA and SAS. In addition, GA-related mutants do not only present reduced hypocotyl elongation responses to low R:FR treatments but also to blue light depletion.^{15,24} The signal transduction of GA requires ubiquitination of DELLA proteins,²⁷ and previous studies show that DELLA abundance is affected during the SAS response.^{15,24} Although the stability of DELLAs is reduced in the hypocotyl when seedlings are exposed to low blue light levels,¹⁵ this was not the case for petioles in plants at the rosette stage.⁷ In addition, the DELLA gain-of-function mutant *gai-1*, which has a reduced GA responsiveness due to enhanced DELLA stability, displayed only a slightly reduced petiole elongation response compared with wild-type, and a completely-normal hyponastic response.⁷ These data suggest that DELLA stability is not affected in petioles that are exposed to low blue light levels, which is different from the situation in seedlings or petioles exposed to low R:FR.¹⁵

In the absence of GA, DELLA proteins accumulate to higher levels and interact with PHYTOCHROME INTERACTING FACTORS (PIFs), a family of growth-promoting transcription factors, and prevent these PIFs from regulating gene expression associated with cell elongation.^{28,29} The PIF family of proteins is involved in the signal transduction of phytochromes and can affect the complex network of hormone interactions.²⁹⁻³¹ Interestingly, two PIFs that are required for low R:FR-induced SAS, PIF4 and PIF5, also appeared to be essential for low blue-induced petiole elongation and hyponasty.⁷ Although it remains to be studied whether *pif4* and *pif5* mutants also have impaired low blue-induced hypocotyl elongation, these data imply that PIFs can

be recruited by photoreceptor-signaling pathways other than those controlled through phytochromes.

Conclusions

The studies of Keller et al.⁷ and Keuskamp et al.⁸ show that there are differences in the hormonal regulation of SAS response elicited by low blue light between seedling and rosette-stage *A. thaliana* plants. The hypocotyl elongation response to blue light depletion can be explained by a combined action of auxin and BR, whereas the hormonal regulation of petiole elongation and hyponasty appears to be more complex. The elongation response of the

petiole to blue light attenuation seems to be (partly) regulated by auxin and brassinosteroids, whereas hyponasty could not be explained on the basis of the tested hormonal pathways. Ethylene could be a possibility, as ethylene is a potent inducer of hyponastic growth. However, in *A. thaliana* ethylene does not appear to contribute to hyponastic growth in response to low light intensity.³² Further work is needed to evaluate the interplay between light signals and hyponastic growth in *A. thaliana*.

Keller et al.⁷ identified PIF4 and PIF5 as novel regulators of SAS in response to blue light depletion. Much is known about the interaction between phytochromes

and PIFs,³¹ but it remains to be elucidated how PIFs are recruited to elicit a growth response when cry1 is inactivated by blue light depletion.

This addendum discussed the fact that although the SAS phenotype induced by low R:FR or low blue light is similar in many regards, the hormonal regulation of these responses overlap only partly. More research is needed especially on the blue light regulated SAS. From an ecophysiological point of view, it would be interesting to investigate possible interactions between R:FR and blue light regulated SAS, and how plants respond when the light treatments are combined.

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