Spotlight

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# Spatial control of plant steroid signaling

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**Two recent studies disclose the interaction between brassinosteroids (BRs) and cell-identity genes in establishing organ boundaries. BR signaling is downregulated by LATERAL ORGAN BOUNDARIES (*LOB*), whereas active BR signaling prevents LATERAL ORGAN FUSION1 (*LOF1*) and CUP SHAPED COTYLEDON (*CUC*) expression in the growing primordia, pointing to cell-specific hormone signaling as part of the mechanisms controlling fate acquisition.**

Hormones regulate almost every aspect of a plant's life. In particular, plant steroids, the brassinosteroids (BRs), are instrumental for the establishment of morphogenetic programs through their interaction with developmental genetic circuits, with different roles in different tissues. Despite the detailed information gathered so far on the underlying molecular events of the BR signaling pathway, an intriguing question has not yet been answered: what mechanisms restrict hormone function depending on the cell type? Recently, two reports have unveiled how BR signaling is integrated with cell-type specific information to establish organ boundaries in the *Arabidopsis* (*Arabidopsis thaliana*) shoot through a molecular strategy that may operate in various developmental contexts [1,2].

Morphogenesis relies on the concerted action of signals that promote cell division and expansion in certain cells, with signals that prevent growth in neighboring cells. The mechanical tensions created through this interplay also play an important role in shaping an organ [3]. A critical event in this context is the precise establishment of boundaries between different organs. For instance, newly emerging primordia arise at the shoot apex as groups of proliferating cells derived from the meristem, that soon create a bulge separated from the rest of the apex by a boundary of small cells with reduced division rate [4]. Boundary cell identity and maintenance is conferred by transcription factors such as CUC1, CUC2, CUC3, LOF1, and LOB, whose loss of function results in organ fusion [4,5]. The molecular targets of these transcription factors are largely unknown, yet some of them participate in the control of auxin flux across the shoot apex. Auxin maxima determine organ *anlagen* at the apex [6], whereas other phytohormones such as BRs and gibberellins (GAs) are necessary for growth of the new primordia. For instance, BRs promote cell cycle progression and cell elongation that jointly contribute to the control of organ size [7].

To identify new targets of boundary-specific transcription factors, the authors of [1] searched for genes induced shortly after translocation of LOB to the nucleus in plants expressing a LOB:GR fusion protein under the control of a constitutive promoter. Among the ca. 300 genes identified, several were related to hormone metabolism and signaling. One of the heavily induced transcripts, *BAS1*, encodes a cytochrome P450 enzyme involved in BR inactivation, and changes in its expression level had been linked previously to changes in the levels of active BRs in the plant [8]. Interestingly, plants expressing LOB:GR ectopically displayed a general dwarfism that mimicked the phenotype of BR-deficient mutants, and this defect was partially rescued by *bzr1-1D* mutations that enhance BR signaling, but not by exogenous application of BRs. The actual proof that establishment of a correct boundary requires local reduction of BR levels in boundary cells was suppression of the organ fusion defects of *lob* mutants by forced expression of *BAS1* under the control of the *LOB* promoter [1].

These results suggest that cell division in emerging primordia is regulated in part by BRs, whereas boundary-specific genes restrict growth in boundary cells by decreasing BR levels (Figure 1). This idea is elegantly supported by the results on the BR-activated transcription factor BRZ1 showing that BZR1:GFP accumulation is lower in boundary cells than in the neighboring cells of shoot apex and the emerging primordia, and the accumulation of the dominant bzr1-1D in boundary cells causes organ fusion phenotypes [2]. However, an intriguing question arising from this model is how the expression of boundary-specific genes is precisely restricted to boundary cells. A possible answer is provided by the authors of [2], who demonstrated that active BZR1 in emerging primordia binds to promoters of the *CUC* genes and represses their expression, thereby closing a regulatory loop that helps establish a boundary territory of non-dividing cells between new organs and the shoot apex (Figure 1). The involvement of BRs in the establishment of boundaries at the shoot apex very likely also operates in other morphogenetic processes. For instance, a high number of *bzr1* mutant plants also display abnormal anther fusion.

Importantly, the new evidence presented by the two recent studies [1,2] not only unveils new roles of BRs in plant development, but also points to at least two mechanisms for cell-type specific action of BRs. The first mechanism implicates a spatial regulation of hormone signaling based on local accumulation of BRs. Given the important role of BRs in cell division [7], BR depletion from cells that show a different cellular behavior would be a simple way to prevent BR-mediated signaling in these cells. In the case

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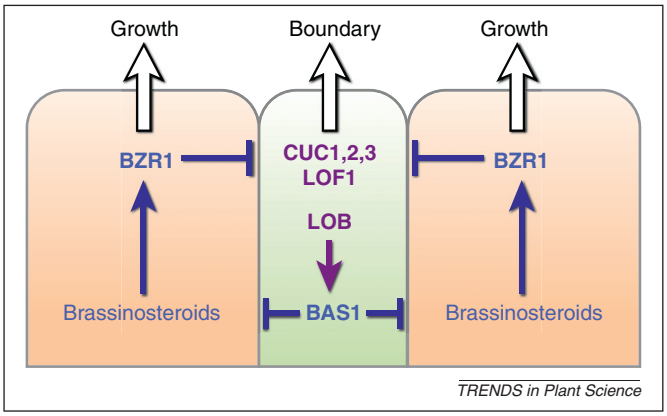


**Figure 1**. Model for spatial regulation of boundary formation by brassinosteroids. In growing cells, active BZR1 targets and represses the transcription of boundary-specific genes *CUC* and *LOF1* that control organ separation. Conversely, the boundary specific transcription factor LOB represses the expression of *BAS1*, which encodes a BR-degrading enzyme, ensuring the reduced BR-signaling levels in these cells necessary to restrain growth in the boundary region. This mutual regulation between BR signaling and boundary genes has two readouts: BR activity becomes strongly dependent of the cell's identity, and BR signaling is also part of the mechanism that establishes cell identity.

reported here, a boundary-specific transcription factor, LOB, directly promotes local induction of a BR inactivating enzyme. Similarly, evidence has shown that KNOX genes repress GA biosynthesis specifically in the shoot apical meristem,whereas allowing higher GA levels in emerging primordia, and precise localization of GAs in the vasculature at the apex is required for normal development in trees.

The second mechanism would require the presence of cell-type specific signaling components. An indication that this mechanism occurs during the establishment of boundaries is that exogenous BR application induces *LOB* expression, but always within the boundary domain, indicating that this effect of BR signaling is restricted by yet unknown cell-type specific factors. Two recent studies have illustrated the existence of this mechanism in stomata development. BRs influence the stomata differentiation program in epidermal cells – and not in any other tissue – because the ubiquitous BR-dependent BIN2 protein kinase phosphorylates YODA and SPEECHLESS, two epidermis-specific elements in the stomata differentiation program [9,10].

Collectively, these studies provide a step forward in the understanding of hormone action with cellular resolution. Furthermore, they open a new path to study the mechanisms through which BR signaling exerts different roles depending on the cell type and/or organ, similar to the better known control of BR-mediated cell expansion and overall plant growth. The combination of *avant-garde* quantitative cellular analyses with molecular genomic tools will be crucial to unravel the molecular mechanisms that regulate plant morphogenesis through the integration, at the cellular level, of hormone signaling with other genetic circuits governing cell identity. The show has just begun.

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