Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Plant Biology

How boundaries control plant development

Petra Žádníková and Rüdiger Simon

Continuous growth and organ development from the shoot apical meristem (SAM) requires a precise coordination of stem cell proliferation, commitment of stem cell descendants to diverse differentiation pathways and establishment of morphological meristem-to-organ boundaries. These complex biological processes require extensive integration of several components of cell-to-cell signaling and gene regulatory networks whose coordinated actions have an impact on cell division and growth.Here we review the current knowledge of gene networks involved in organogenesis from the SAM in higher plants. We focus on recent advances to show how the interaction between transcriptional regulators, hormonal crosstalk and physical stress regulates the establishment and maintenance of meristem-to-organ boundaries.Continuous growth and organ development from the shoot apical meristem (SAM) requires a precise coordination of stem cell proliferation, commitment of stem cell descendants to diverse differentiation pathways and establishment of morphological meristem-to-organ boundaries. These complex biological processes require extensive integration of several components of cell-to-cell signaling and gene regulatory networks whose coordinated actions have an impact on cell division and growth.Here we review the current knowledge of gene networks involved in organogenesis from the SAM in higher plants. We focus on recent advances to show how the interaction between transcriptional regulators, hormonal crosstalk and physical stress regulates the establishment and maintenance of meristem-to-organ boundaries.

Addresses
Institute of Developmental Genetics, Cluster of Excellence on Plant Sciences (CEPLAS), Heinrich-Heine-University, Universitätsstrasse 1, D-40225 Düsseldorf, Germany

Corresponding author: Simon, Rüdiger (ruediger.simon@hhu.de)

Current Opinion in Plant Biology 2014, **17**:116–125

This review comes from a themed issue on **Growth and development**

Edited by **David R Smyth** and **Jo Ann Banks**

S1369-5266/$ – see front matter, Published by Elsevier Ltd.
http://dx.doi.org/10.1016/j.pbi.2013.11.013

Introduction

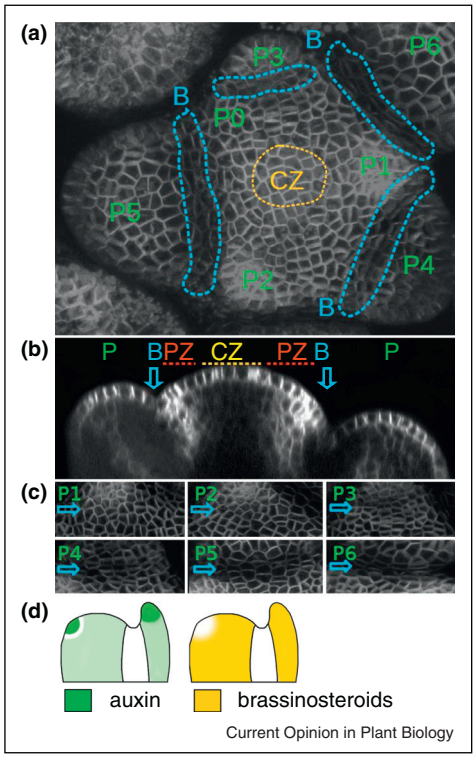
Plant organogenesis is assured by pools of dividing, pluripotent cells that reside in the meristems, the plant's stem cell niches. The shoot apical meristem (SAM), a group of cells at the growing tip of a plant, generates all the aboveground tissues of the plant. The SAM is organized into a central zone, composed of slowly dividing stem cells and a peripheral zone, containing more rapidly dividing cells that can become incorporated into the organ primordia (Figure 1a and b). The initiation of organs from the peripheral zone requires the creation of meristem-to-organ boundaries that separate these two cell groups with very distinct gene expression programs and morphologies. The boundary itself expresses a unique set of transcription factors that play an important role to locally repress cell proliferation, which is a prerequisite for the development of physically separate organs. Loss-of-function mutants often cause organ fusion, but also defects in organ development and altered phyllotactic patterning. This indicates that the meristem-to-organ boundaries also operate as organizing centers that provide information to adjacent cells to control their developmental programs (Figure 1a–c). Recent studies have uncovered how a combination of mechanical forces, transcriptional regulators and phytohormonal inputs interact to establish and maintain functional boundaries at the meristem (Figure 1d).

Role of mechanical stress in meristem patterning and organ initiation

Cells at different positions within the dome-shaped apical meristem experience different growth and expansion rates and therefore are subjected to mechanical stress. In response to this local mechanical stresses, they modify their growth rate and anisotropy accordingly. Growth rate and anisotropy depend on the cell wall properties, since cell growth is driven by internal turgor pressure, which is limited by the stiffness of the wall. The rigid cellulose microfibrils provide the necessary reinforcements that counteract cellular turgor. However, not all cells in the meristem are subjected to the same mechanical stress, indicated by the observation that the outer cell wall is approximately five-times stiffer at the tip of the meristem than on its flanks [1•]. Anisotropic growth is mediated by the parallel alignment of cellulose microfibrils in plant cell walls, which is controlled by cortical microtubules. These drive the local insertion and processing trajectory of the cellulose synthase complex (CESA) at the plasma membrane [2,3]. An intimate connection between microtubules and the cellulose synthase complex is essential for organ phyllotaxis: absence of the linker protein CESA INTERACTIVE PROTEIN 1 (CSI1) that connects CESA with cortical microtubules [4,5] cause twisting of the rapidly elongating part of the stem after primordia have arisen, and subsequently affecting phyllotaxy [6•]. This revealed that the overall mechanical properties and local resistance to stress within meristem can affect organ positioning [1•,6•].

Local growth heterogeneity during organ initiation requires a differential response of neighboring cells. In

Figure 1



The inflorescence meristem (IM) of *Arabidopsis thaliana*. The shoot apical meristem (SAM) is responsible for the production of rosette leaves and, after transition to the inflorescence meristem (IM), for the production of the stem, cauline leaves, lateral meristems and flowers of the inflorescence, which arise in the axils of cryptic bracts. **(a)–(c)** Reconstructed views of the SAM expressing membrane-localized *PIN1::PIN1-GFP* in the epidermis (a, top view; b, optical section; c, detailed view of the top, focused on different stages of meristem-to-organ boundary formation). The localization of *PIN1::PIN1-GFP* indicates auxin transport towards young flower bud primordia (named P1-P6 from the youngest to the oldest organ primordium). The functional zones are represented and highlighted in different colors. At the meristem summit the central zone (CZ, yellow) comprises the stem cells, primordia (P, green) are initiated in the peripheral zone (PZ, red). The zone between the peripheral zone and the primordium represents the meristem-to-organ boundary (B, blue). (D) Schematic representation of the likely distribution of auxin and brassinosteroids in the SAM.

the SAM, cortical microtubules continuously reorganize to remain parallel to the direction of maximal stress, and the ability to reorient these cortical microtubules in response to mechanical forces is crucial to allow differential growth. This is achieved through proteins like KATANIN that fragment microtubules. In *katanin* mutants, neighboring cells have the tendency to grow more frequently in the same direction, and the characteristic dome-like shape of the shoot tip inverts [7••], reviewed in [8].

Cells in boundaries exhibit a very low growth rate and high anisotropy, compared to the neighboring meristem and primordium, which correlates with stiff cell walls and a parallel orientation of the microtubules [9•], reviewed in [10]. Microtubules quickly reorient upon wounding of the SAM, which locally alters the mechanical forces. Interestingly, relocalization of the auxin efflux carrier protein PINFORMED1 (PIN1) was shown to parallel microtubule reorientation to the direction of the largest stress thereby affecting auxin distribution in the tissue. However, PIN1 was still stably localized after microtubule depolymerization through oryzalin treatment, indicating that microtubules are not causing the PIN1 localization in a direct manner.

In boundaries, microtubules are oriented along the major axis of cells coinciding with PIN1 localization at anticlinal walls. This orientation of PIN1 will then result in a depletion of auxin from the boundary domain and export towards the organ initial and the remainder of the meristem. Importantly, auxin itself affects the mechanical properties of the cell wall controlling the expression of cell-wall remodeling enzymes such as expansins, and auxin maxima allow primordia to grow out. Thus, a positive feedback loop is created whereby auxin transport is affected by tissue mechanics, which is in turn controlled by auxin through its effect on cell wall characteristics [9•].

Transcriptional regulation of boundary functions

Organ initiation is closely related to auxin accumulation in organ founder cells due to PIN1-mediated directional auxin transport. Subsequently, when primordia start to grow, PIN1 polarity reverses to form a new auxin maximum at a distant position. The shifts in the auxin transport direction are temporally and spatially correlated with the establishment of the boundary between the new primordium and the meristem, as well as with auxin depletion from boundaries [9•].

The complex interplay of regulatory gene networks involved in boundary establishment and maintenance will now be discussed (see Table 1 for a list of genes involved). The role of some of these genes, and the phytohormone auxin, is illustrated in Figure 2.

CUP-SHAPED COTYLEDON (*CUC1,2,3*) genes in *Arabidopsis* and the *NO APICAL MERISTEM* (*MtNAM*) gene in *Medicago truncatula* [11] encode NAC transcription factors that, together with the homeobox gene *SHOOTMERISTEM LESS* (*STM*), regulate the formation of shoot meristems. Later in development, *CUC* genes also control the specification of organ boundaries. Typical for genes regulating boundary functions, *cuc1 cuc2* double mutants display organ fusions and growth arrest [12•], reviewed in [13].

Earlier studies showed that there is interdependence between auxin-dependent organ initiation and *CUC* gene expression. Mutations in *PIN1* produce naked inflorescence stems resulting from the ectopic expression of