# CDB Part IB
# Plant Development

## Lecture 4

## Patterning of shoot growth

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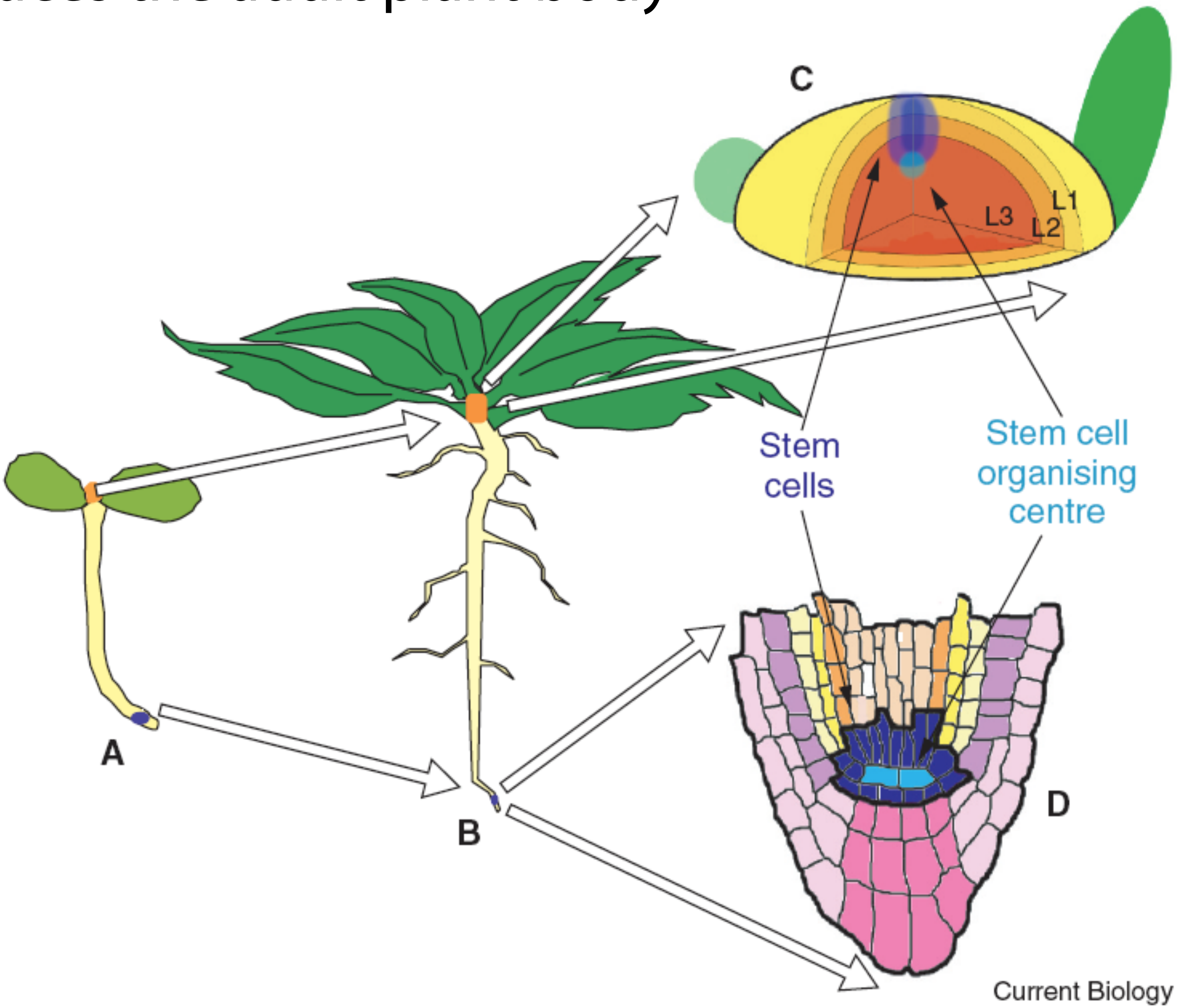
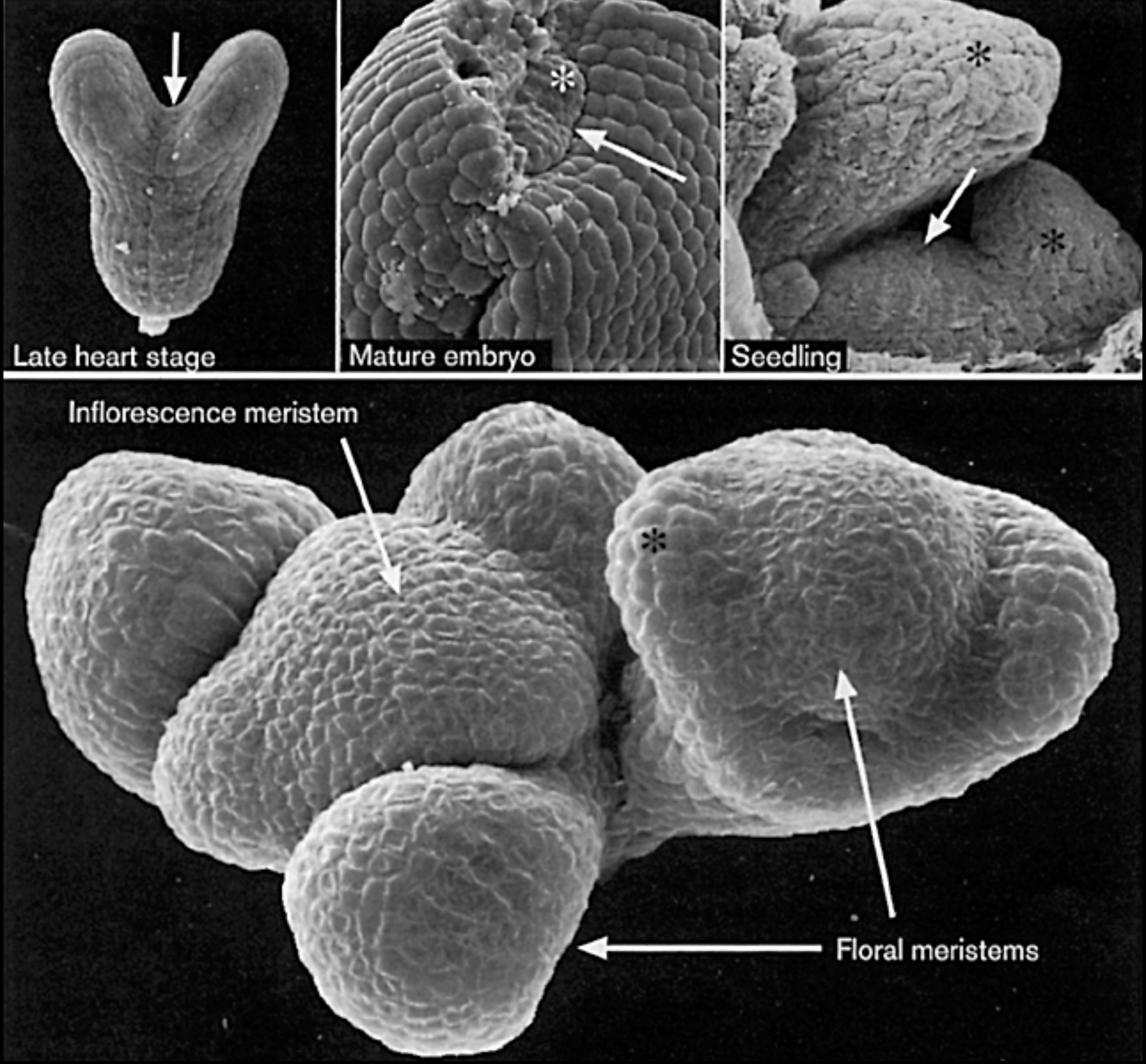
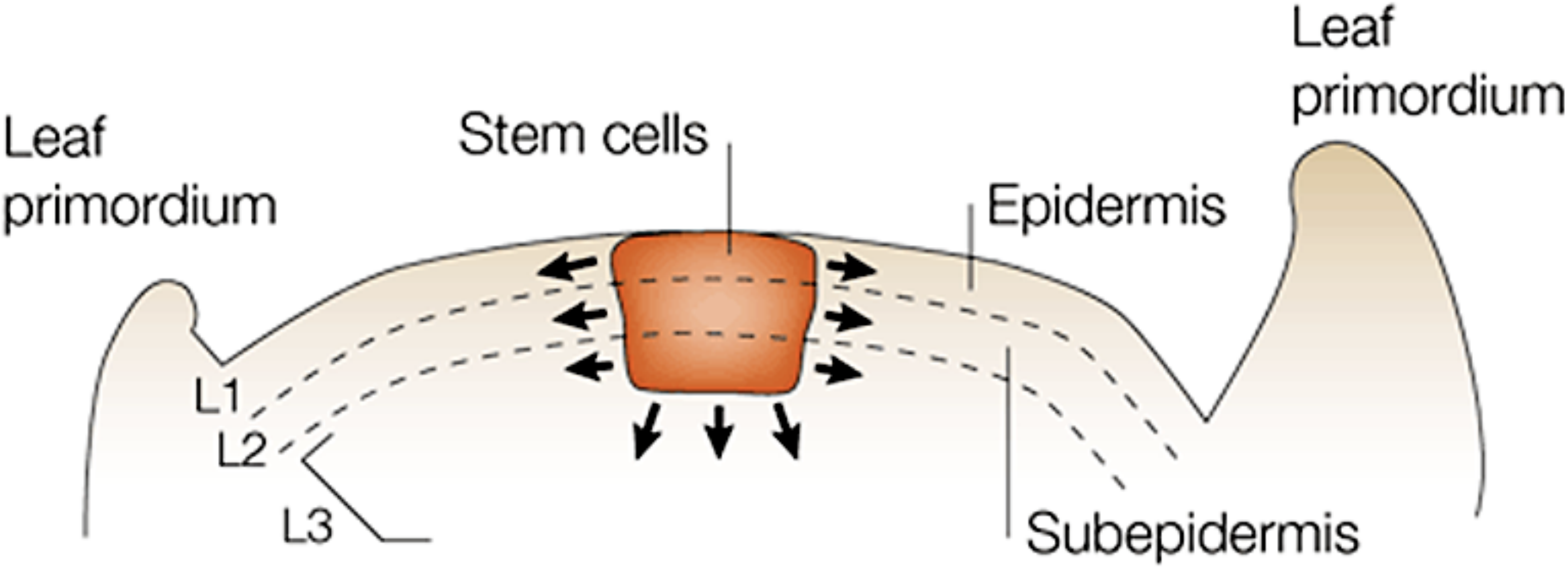
# Continued growth of shoot and root meristems produces the adult plant body



*Arabidopsis apical meristems*

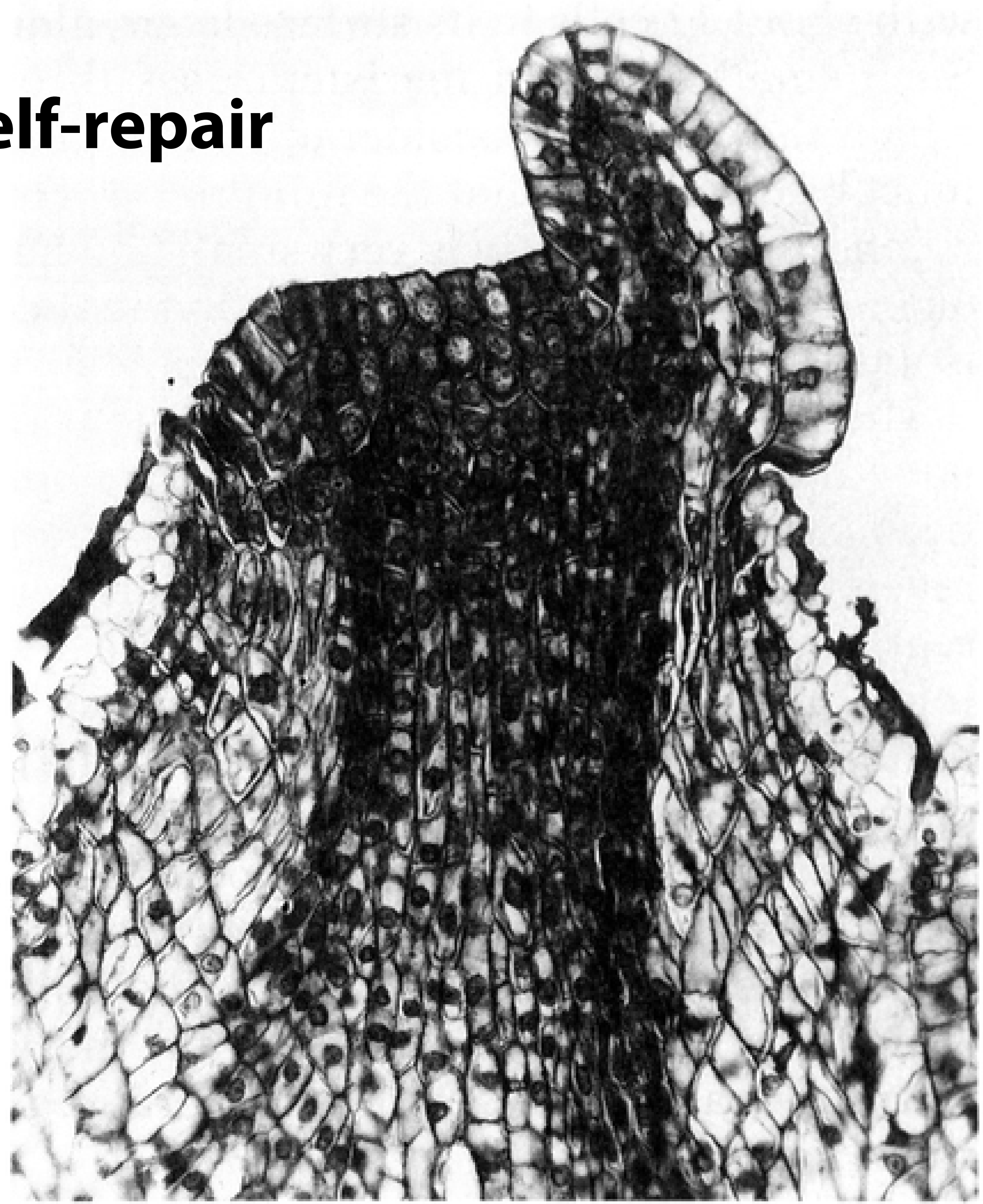



**The shoot meristem is branched and indeterminate, capable of producing lateral primordia at the flanks of the meristem.**



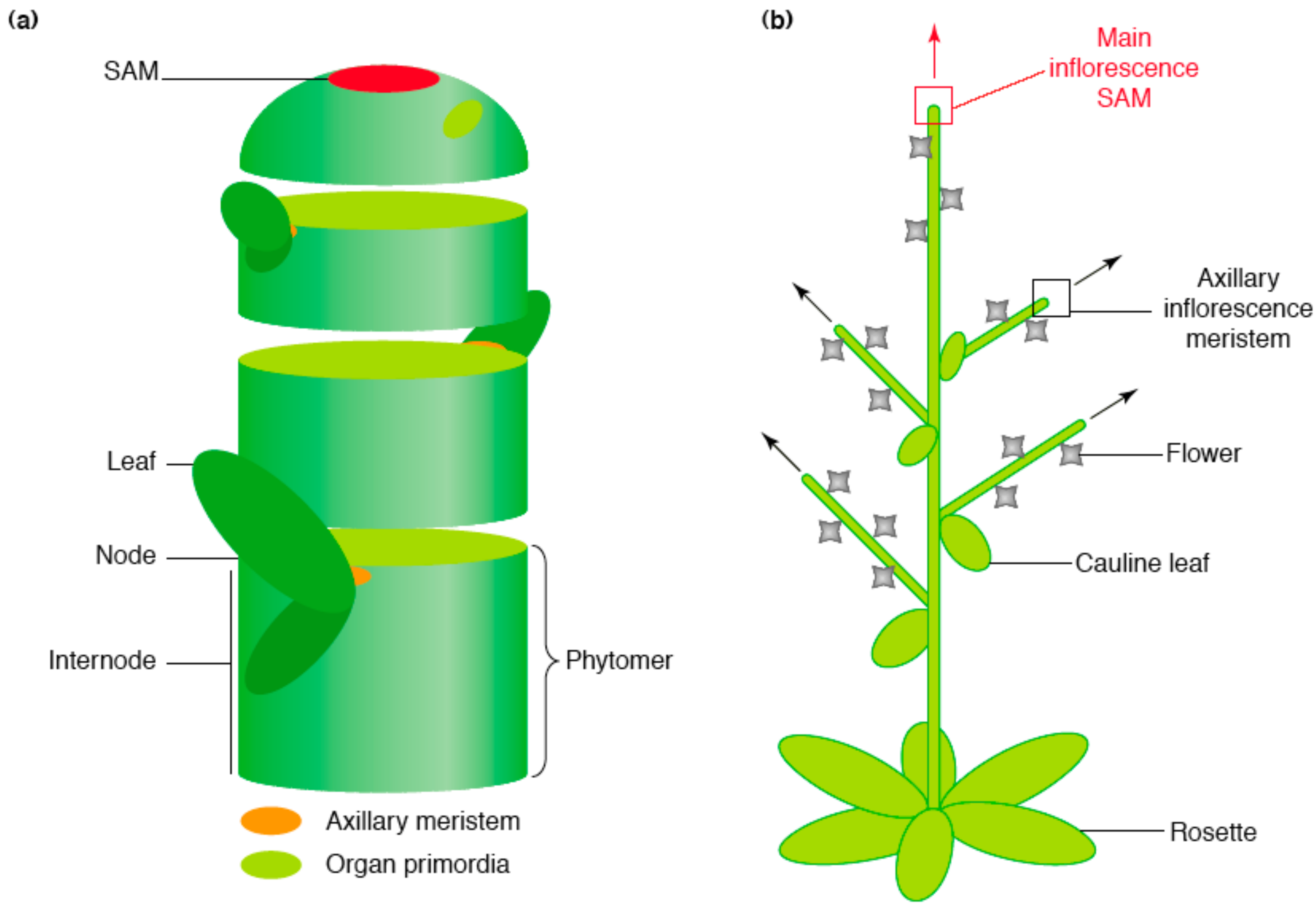

**A meristem is self-organising and renews itself, maintaining a balance between cell proliferation and differentiation**

# …capable of self-repair

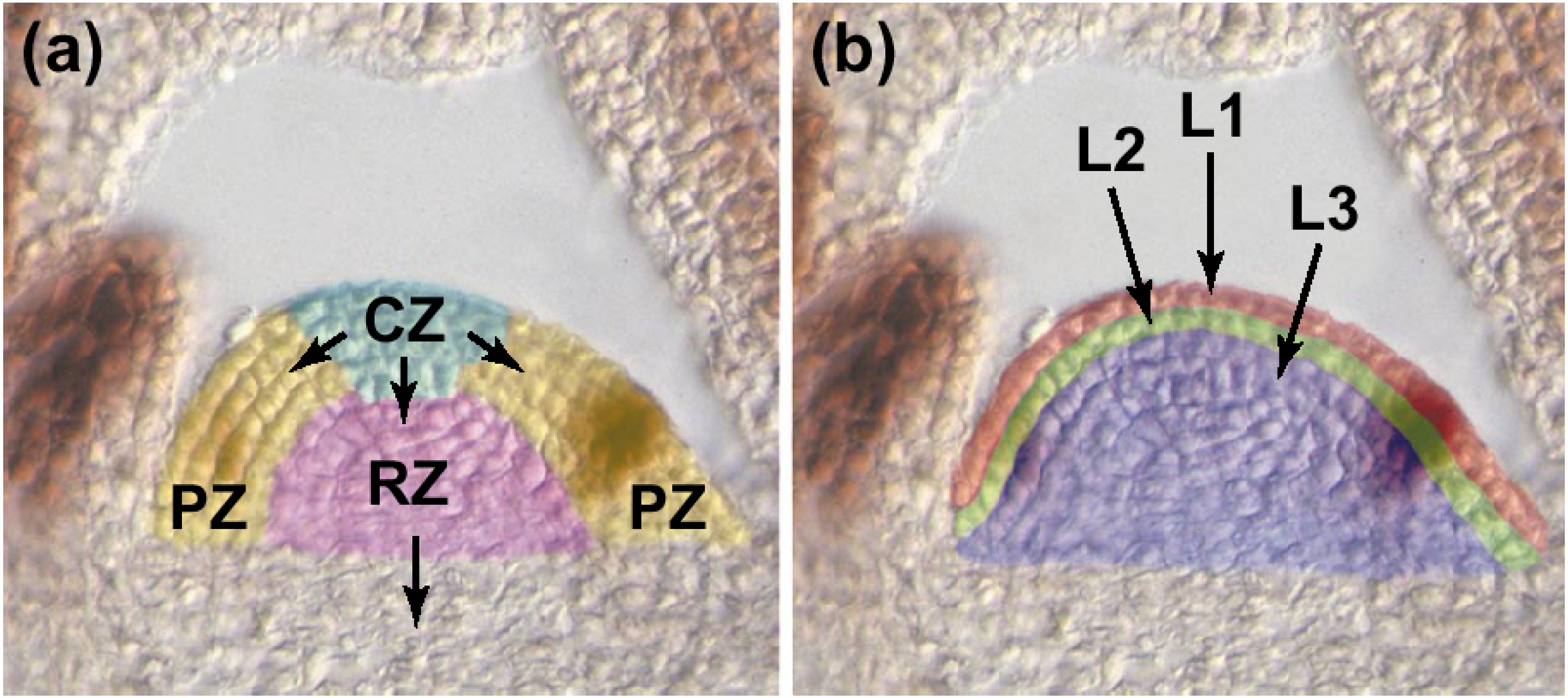


Sussex, Brookhaven Symp. Biol. 16:1–12 (1964)

# Modular growth and production of lateral organs



## The Arabidopsis shoot meristem is divided into functionally distinct zones




**Central zone = undifferentiated cells**
**Peripheral zone = formation of new lateral organs**
**Rib zone = formation of new stem**

**The meristem contains three different layers of cells, L1, L2 & L3. These generally maintain distinct lineages**

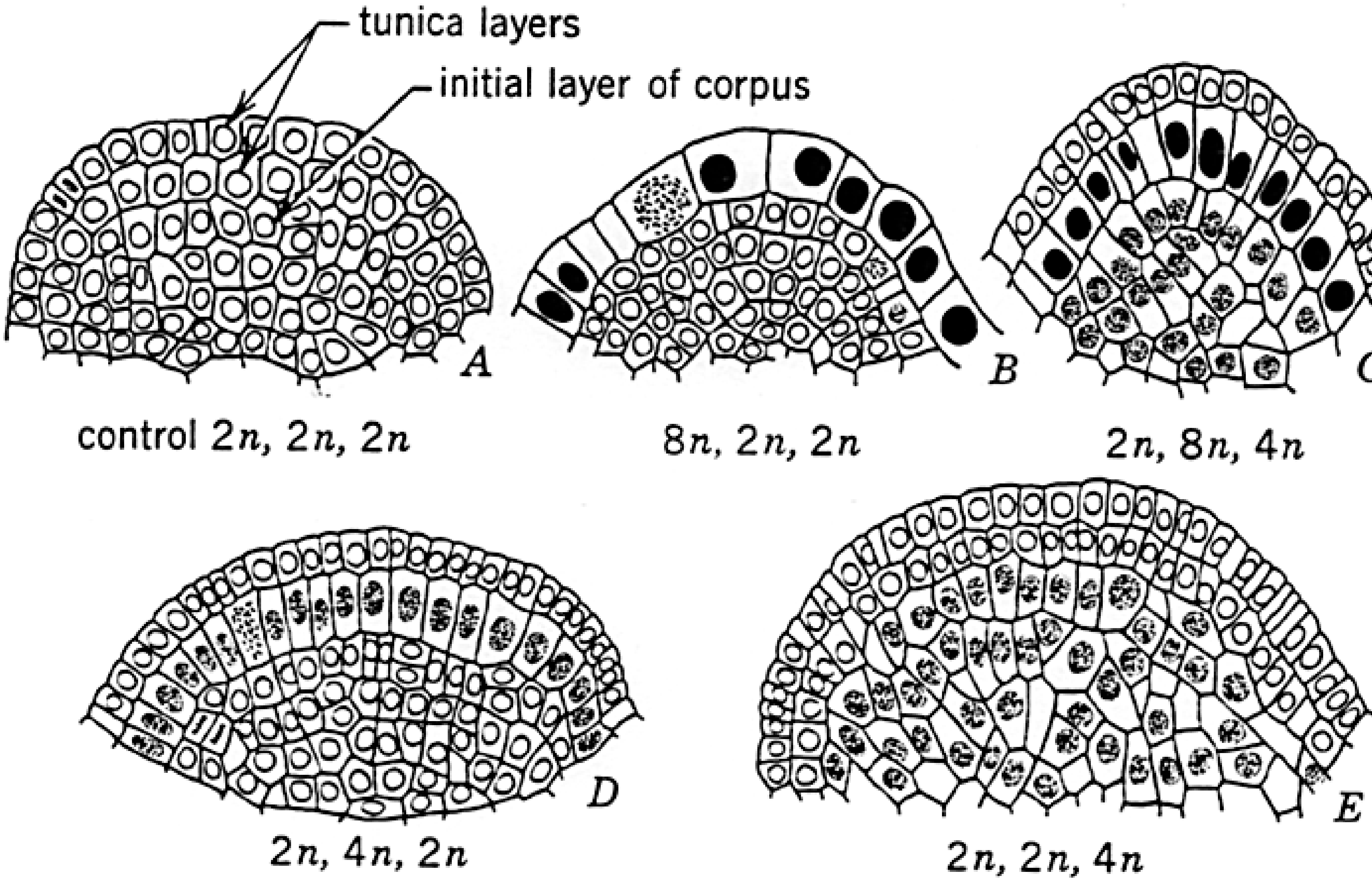
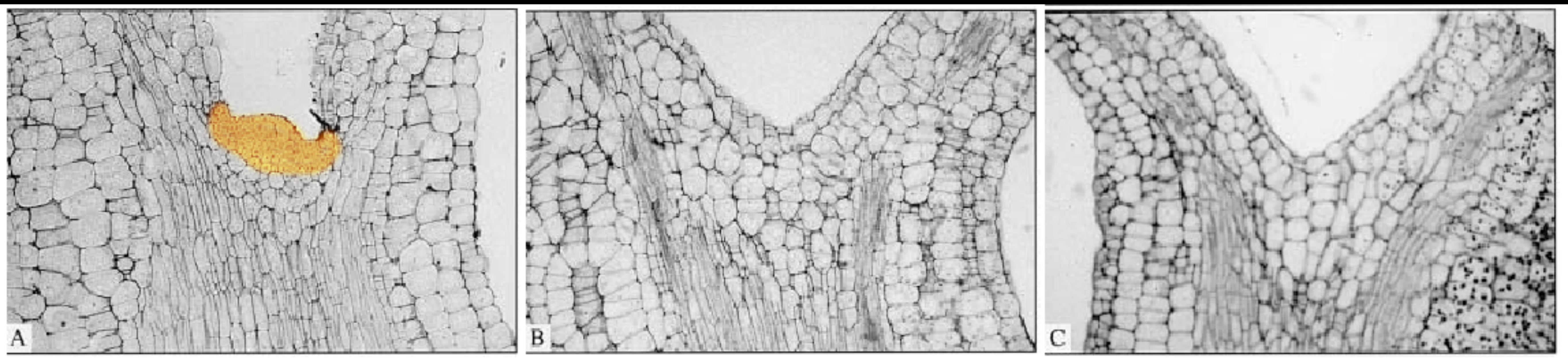



**FIG. 5.2.** Shoot apices of *Datura* from a diploid plant (*A*) and from several periclinal cytochimeras. Chromosomal combinations are indicated by values given below each drawing.

**SHOOT MERISTEMLESS (STM) and WUSCHEL (WUS) are homeodomain genes that are required for formation and maintenance of the shoot apical meristem in Arabidopsis.**

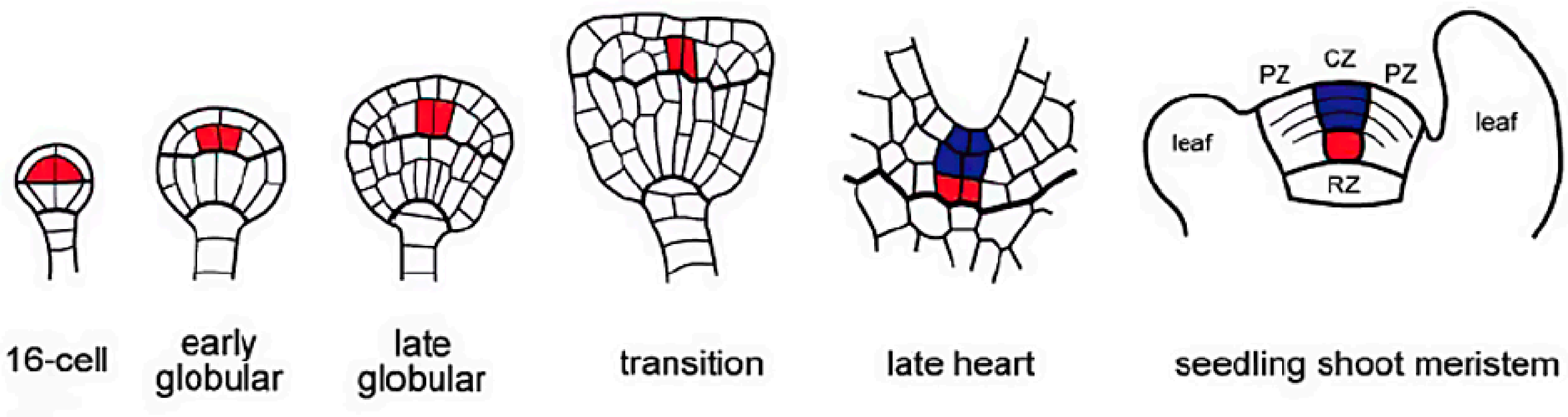


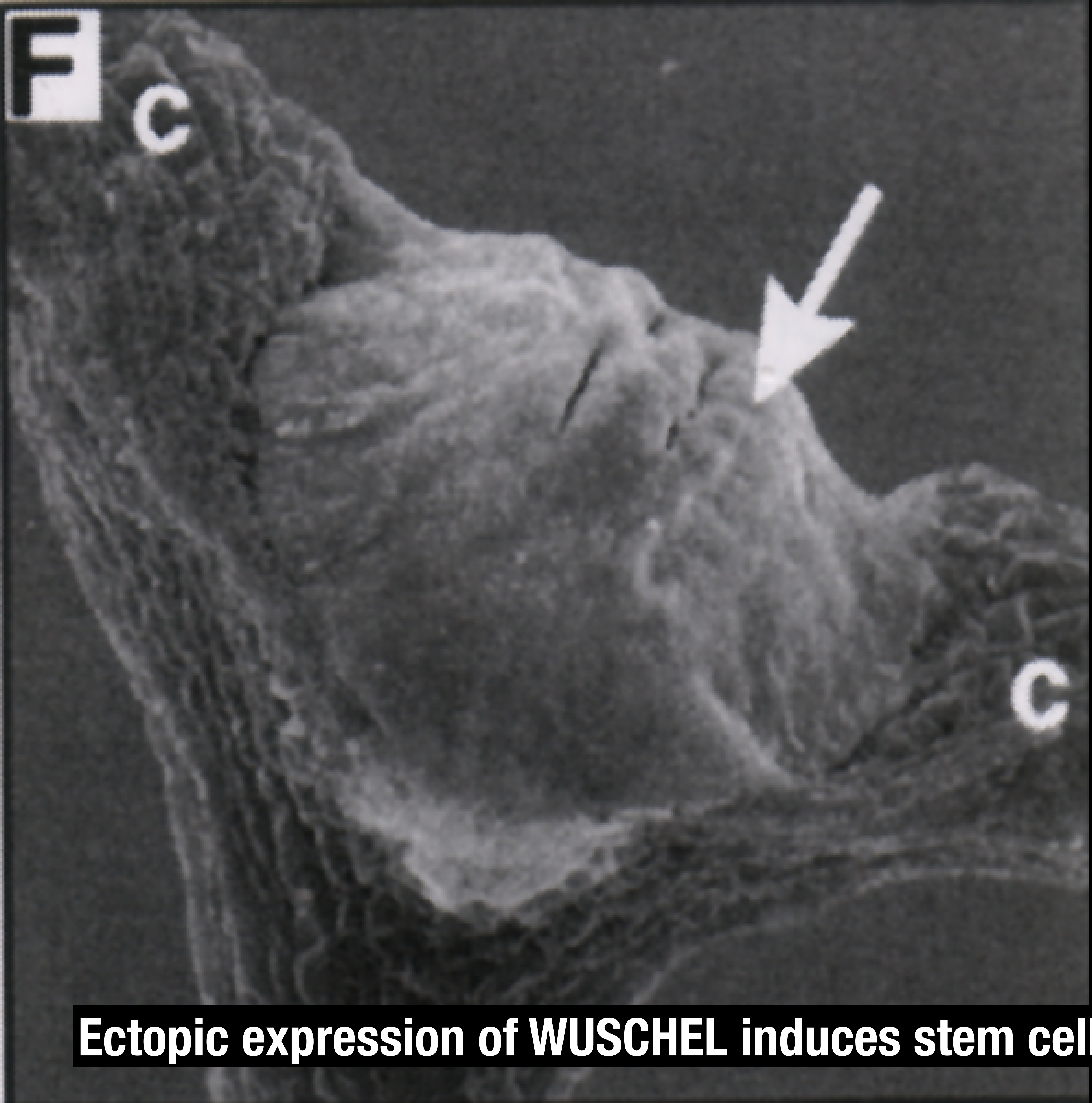

wt

*shootmeristemless (stm)*

*wuschel (wus)*

# WUSCHEL expression






*ANT::WUS*

Ectopic expression of WUSCHEL induces stem cell proliferation

**WUS and STM initiate and maintain meristem growth**

**- but how is the size of the apical meristem constrained?**

**the growth of the shoot meristem is negatively regulated by….**

**the CLAVATA gene pathway**

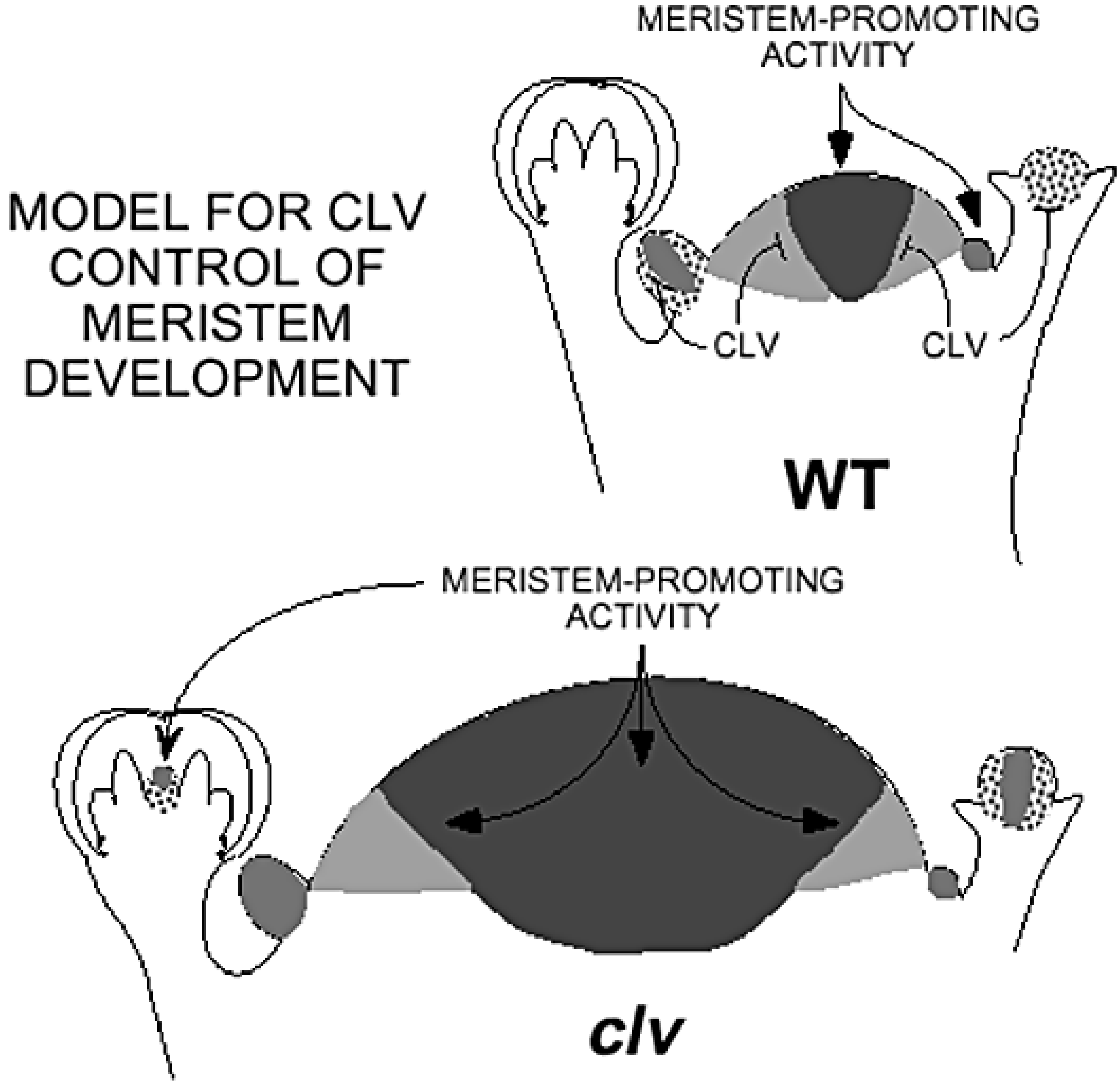
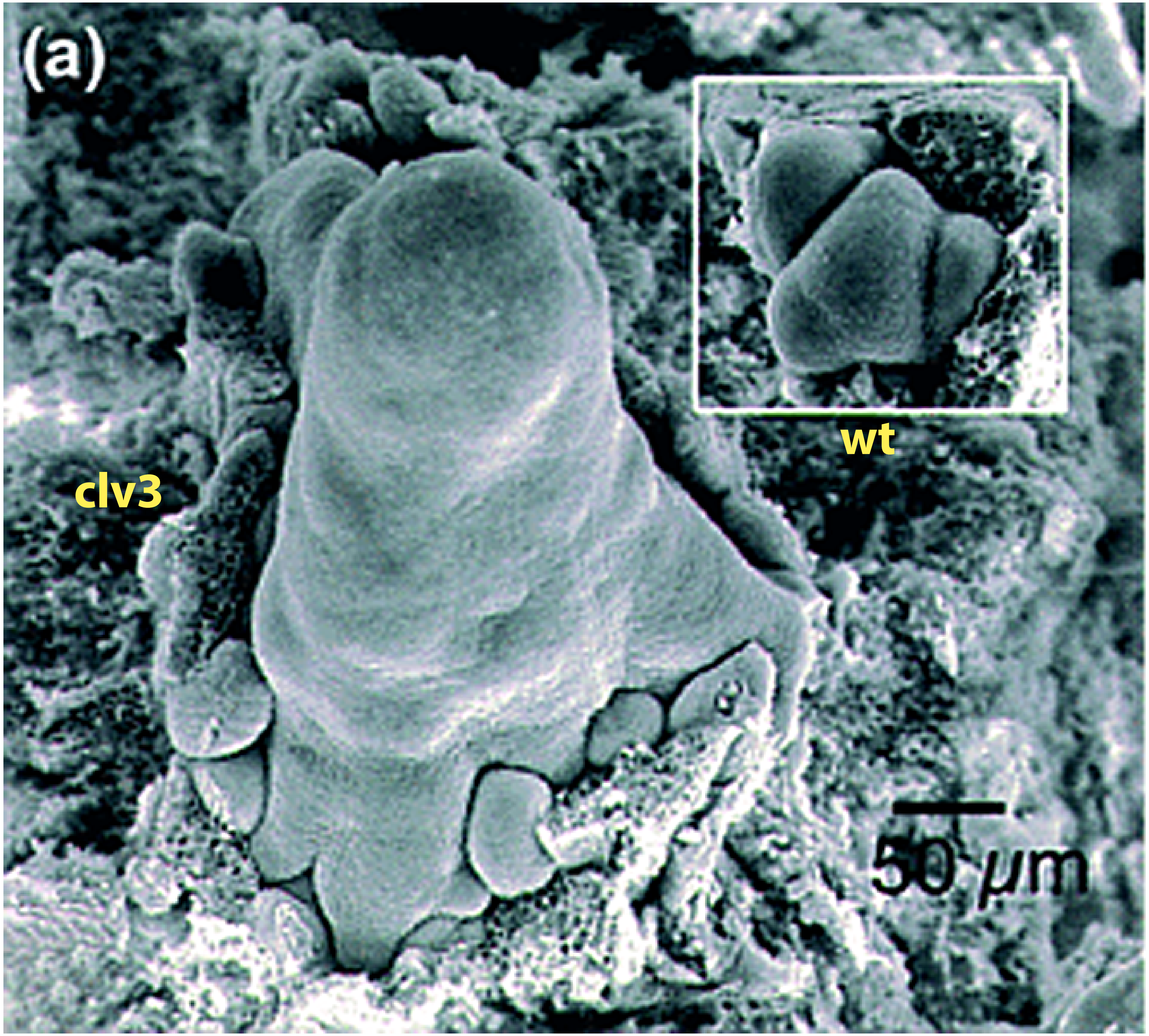


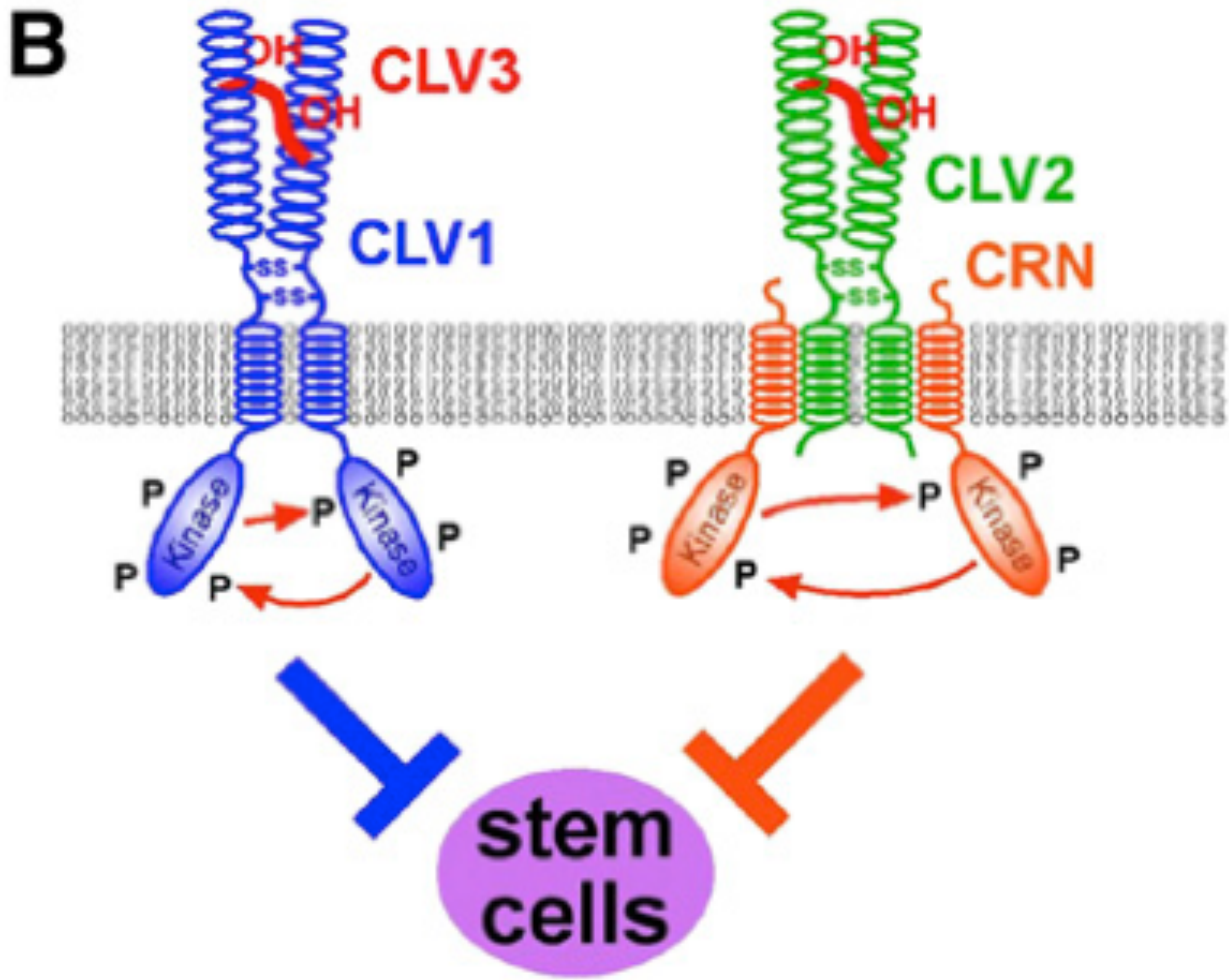

**Fig. 9.** Model for *CLV* action. The model postulates that there is a meristem-promoting activity (MPA) in shoot meristems and young flowers that maintains cells in a proliferative, undifferentiated state.

**Mutations in the genes, *clavata1*, *clavata2*, *coryne* and *clavata3*, produce similar phenotypes - enlarged meristems.**

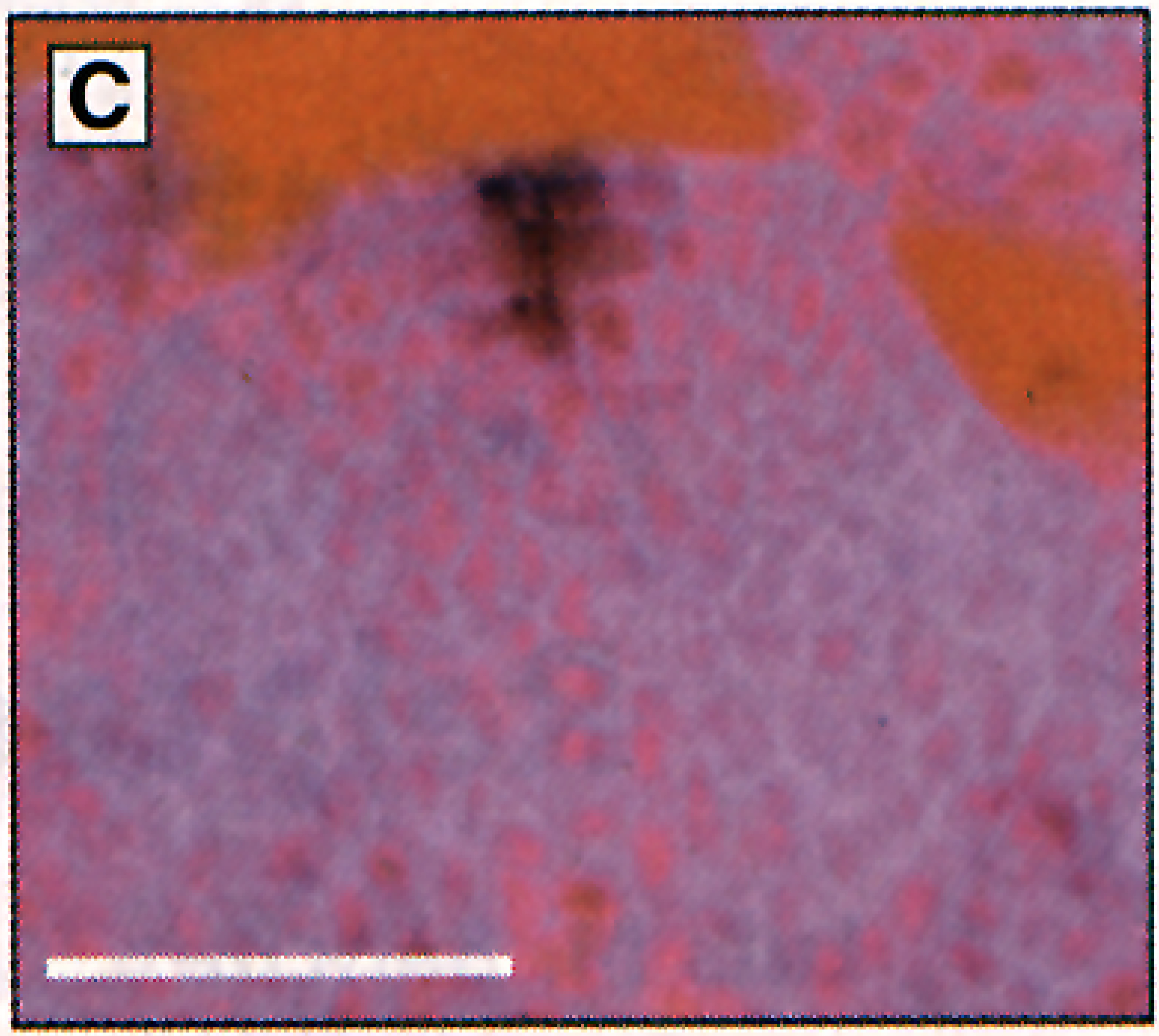



**These genes function in the same regulatory pathway.**

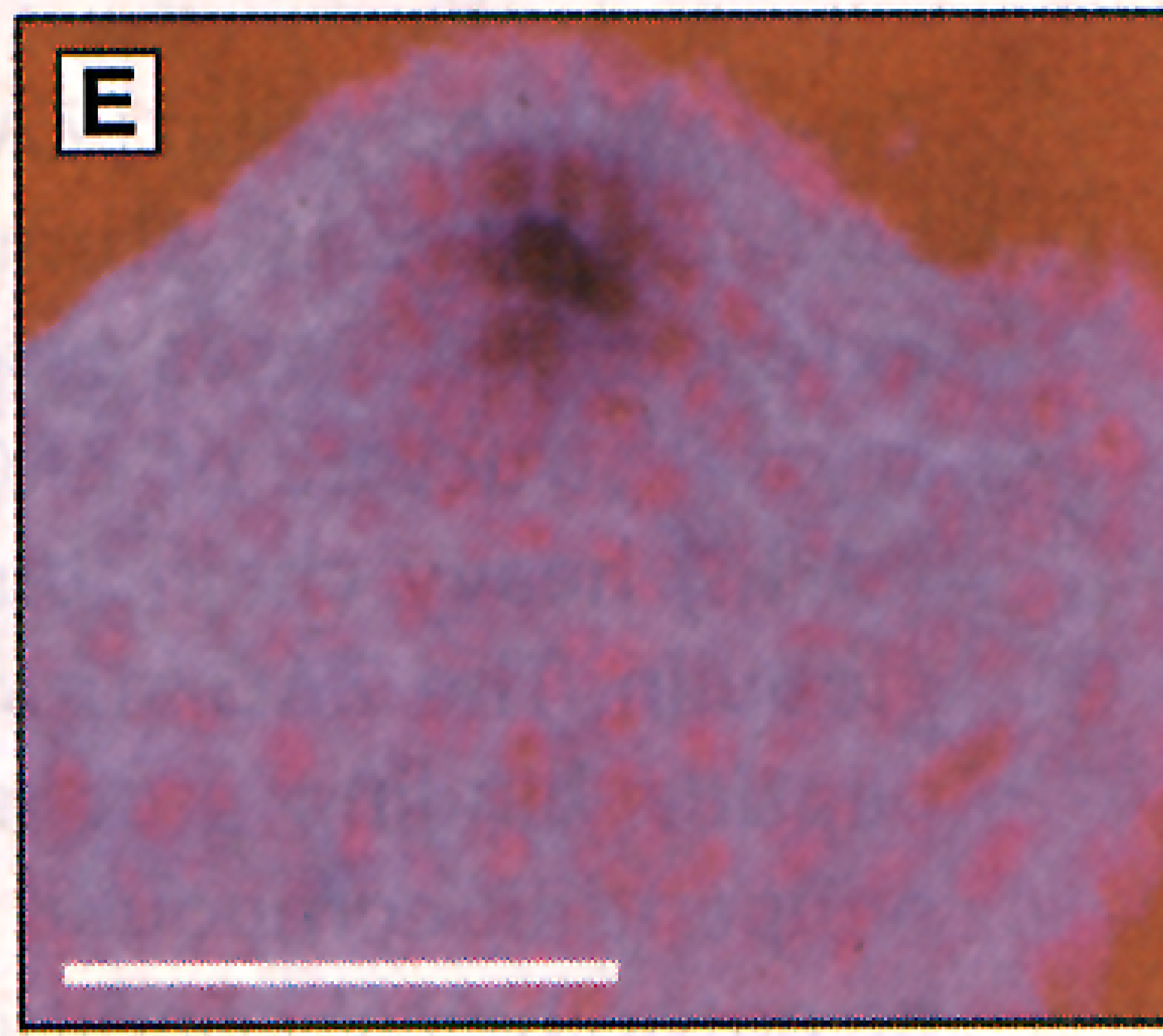
**_Clavata1_, _Clavata2 and Coryne_ are membrane-localised receptor proteins expressed in cells deep in the central zone of the meristem**



**C*lavata3* encodes a secreted peptide expressed in cells towards the apical surface of the meristem, and is a ligand for the CLAVATA1/ CLAVATA2/CORYNE receptors**




*CLV3 mRNA*




*CLV1 mRNA*

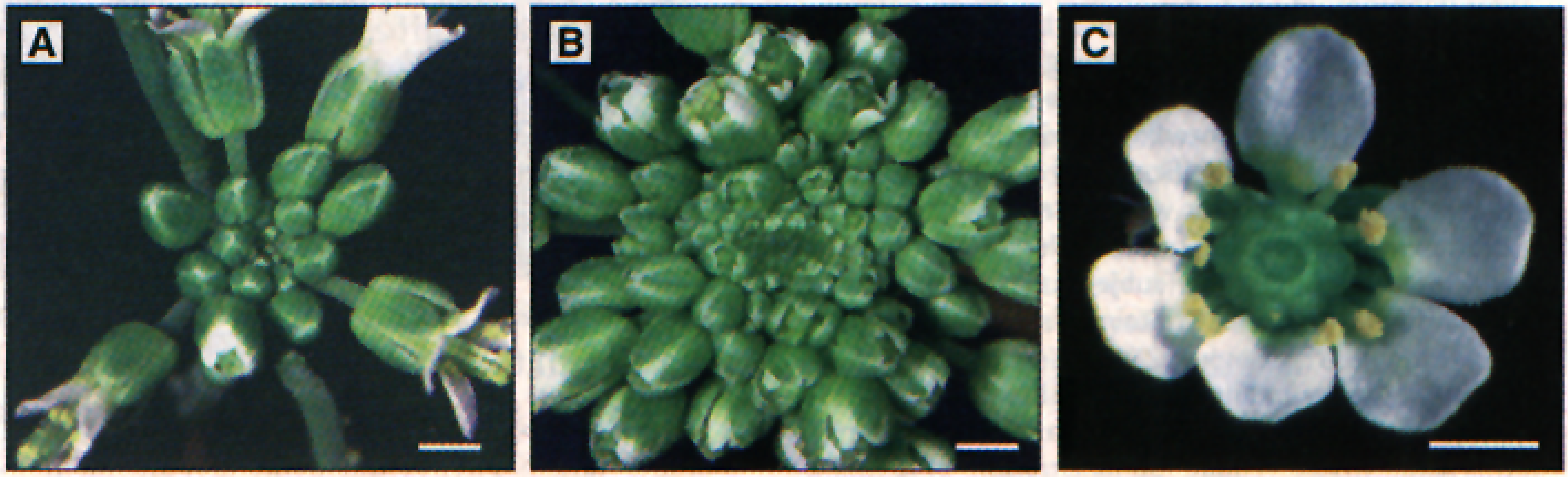



**Fig. 1.** *clv3* shoot and flower phenotypes. (**A**) Wild-type inflorescence meristem. (**B**) *clv3-2* inflorescence meristems undergo fasciation, growing as a ring or line rather than a point. (**C**) *clv3-2* mutant flowers contain extra organs of all types, particularly stamens and carpels. Bars, 1 mm.

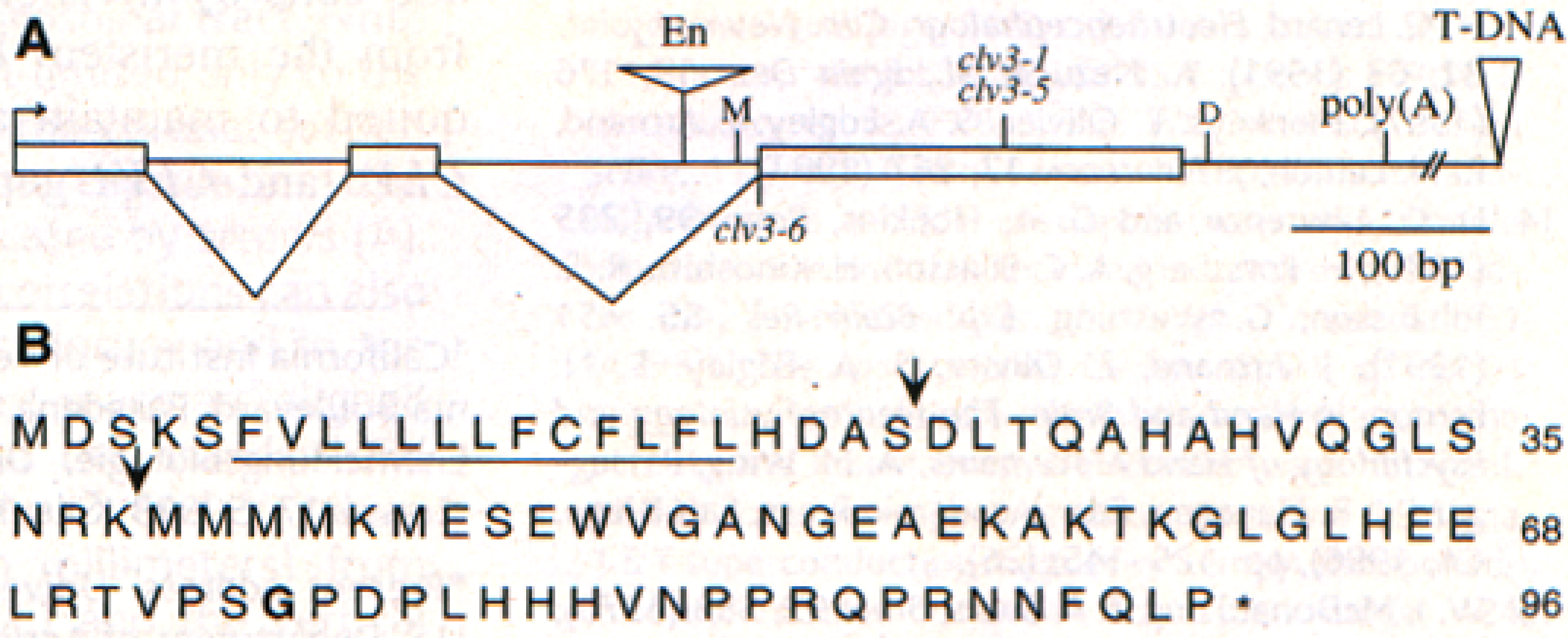
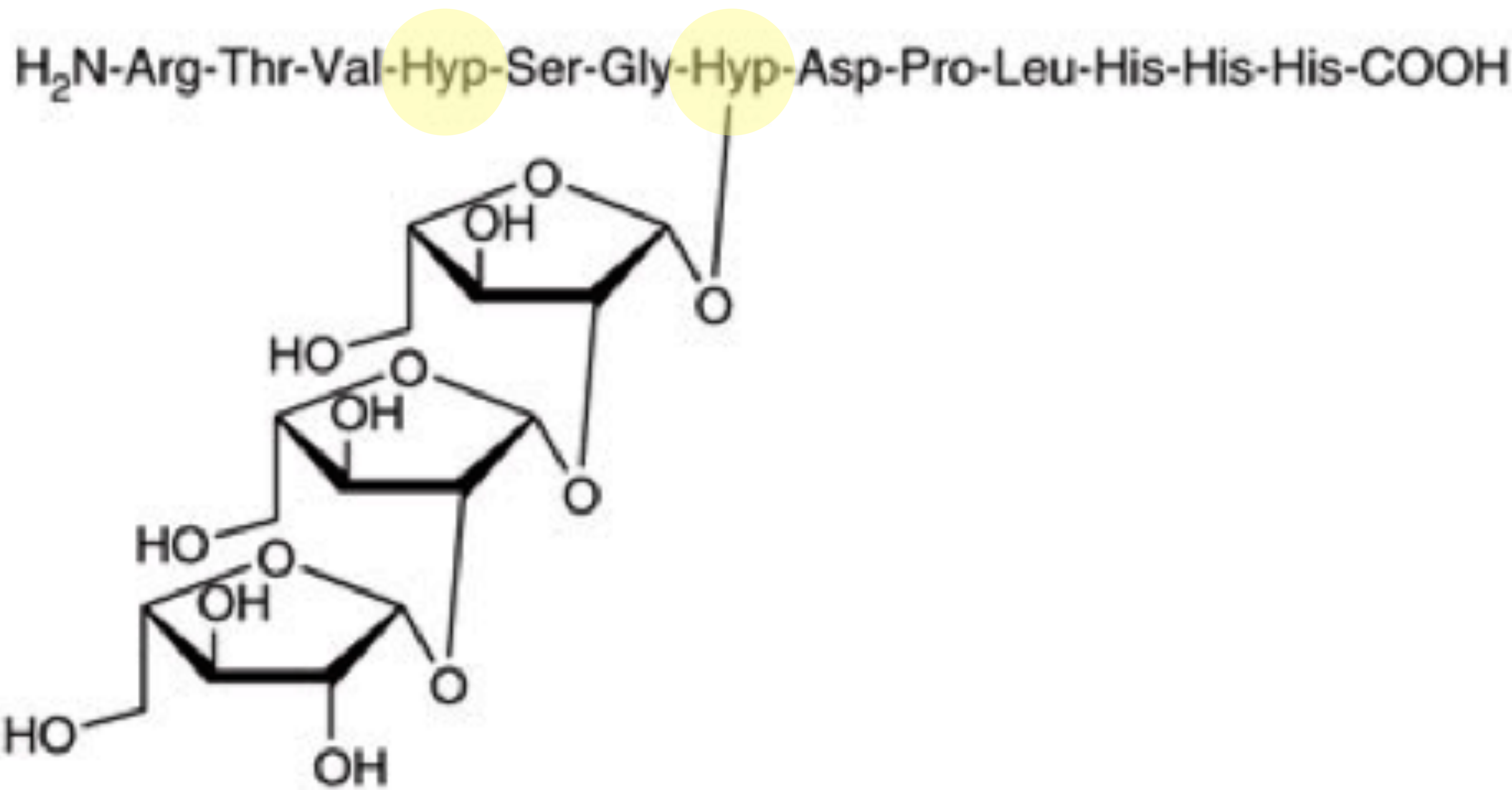
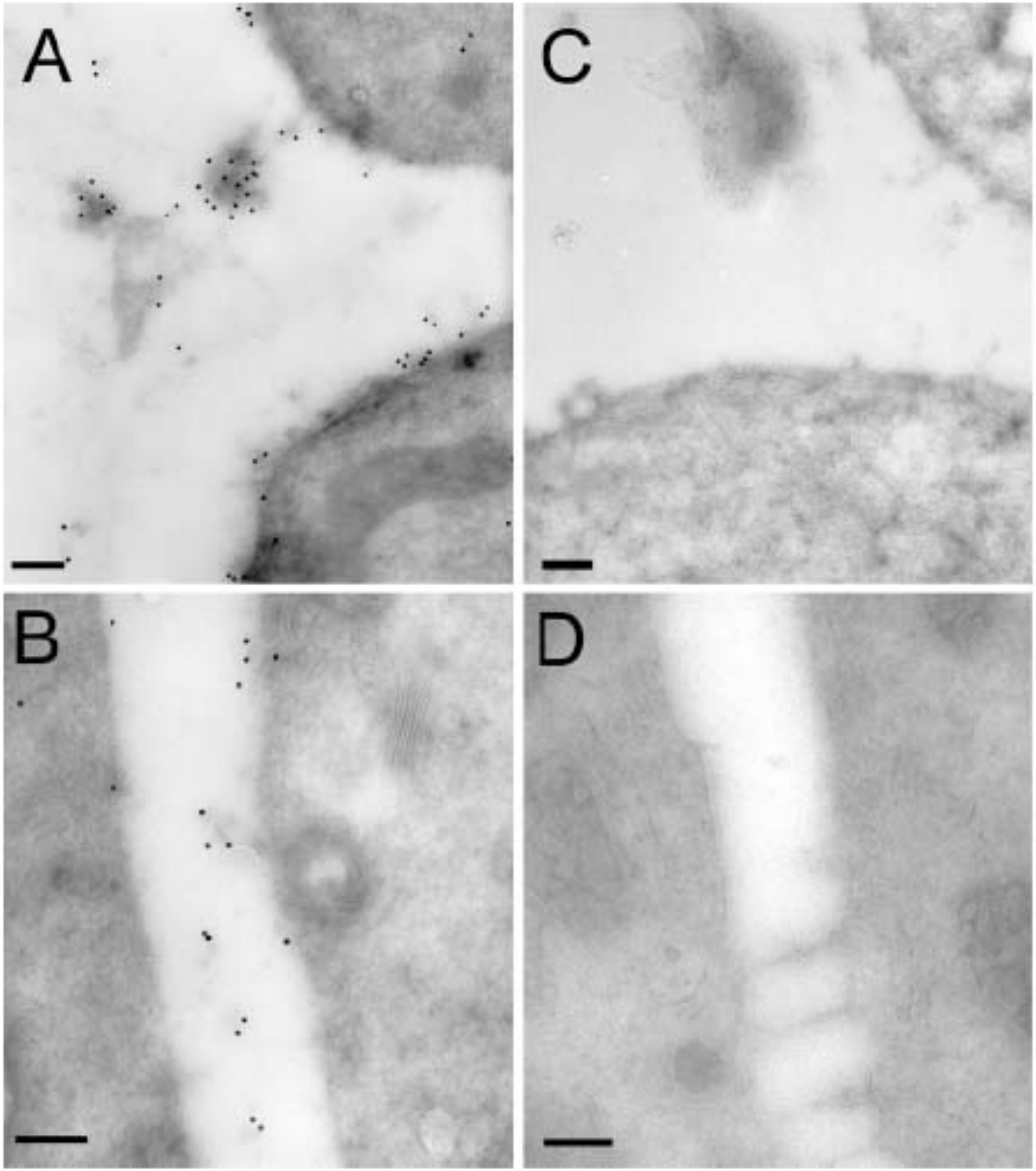



**Fig. 2.** *CLV3* genomic region and peptide sequence. (**A**) The *CLV3* genomic region. The translation start site is denoted by the arrow and the exons by boxes. The relative positions of the *clv3* mutations are shown. Restriction sites: M, Mfe I; D, Dra I. The genom-

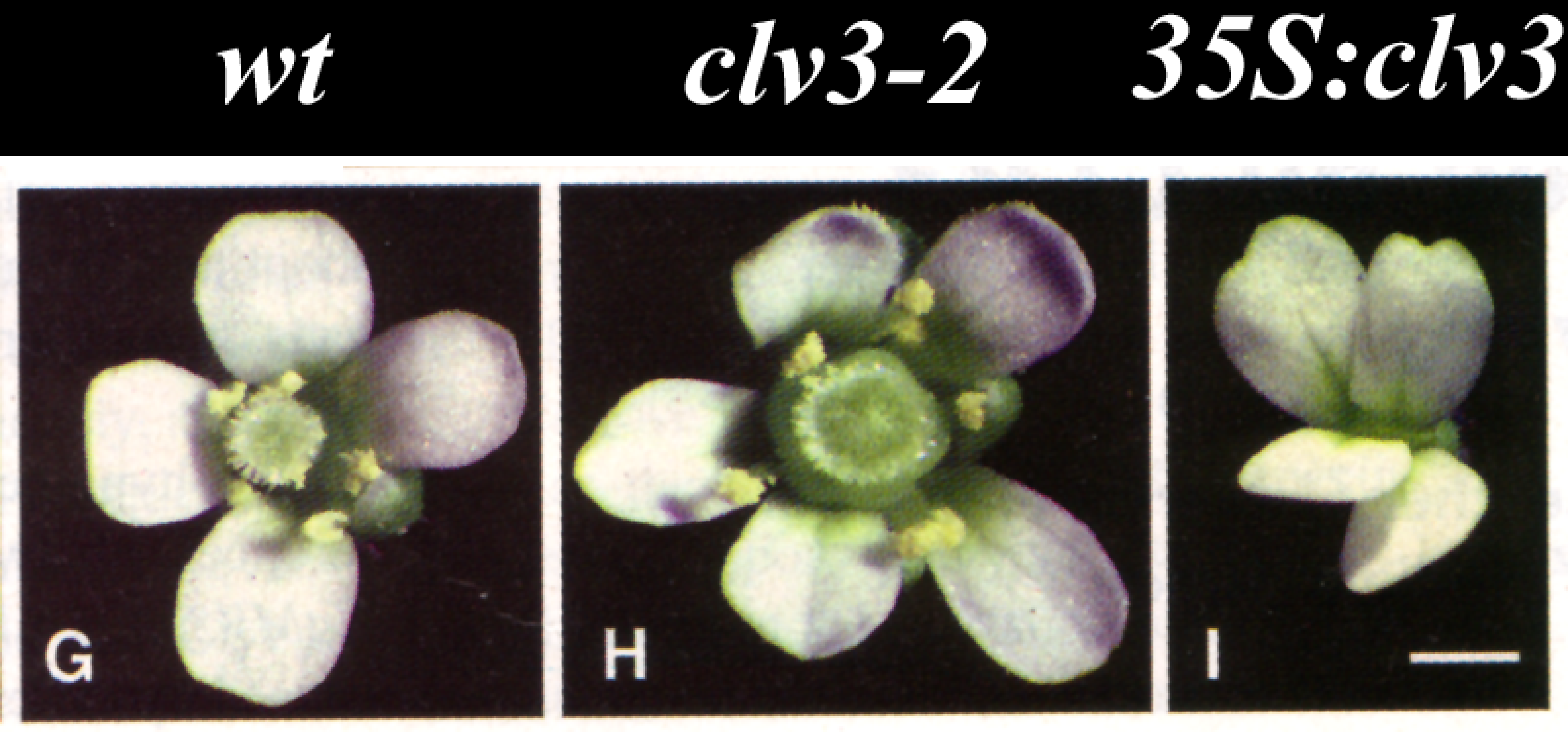
# Clavata3 forms a 13 amino acid arabinosylated glycopeptide




**A glycopeptide regulating stem cell fate in *Arabidopsis thaliana***
Kentaro Ohyama, Hidefumi Shinohara, Mari Ogawa-Ohnishi & Yoshikatsu Matsubayashi
*Nature Chemical Biology* **5**, 578 - 580 (2009)
doi:10.1038/nchembio.182

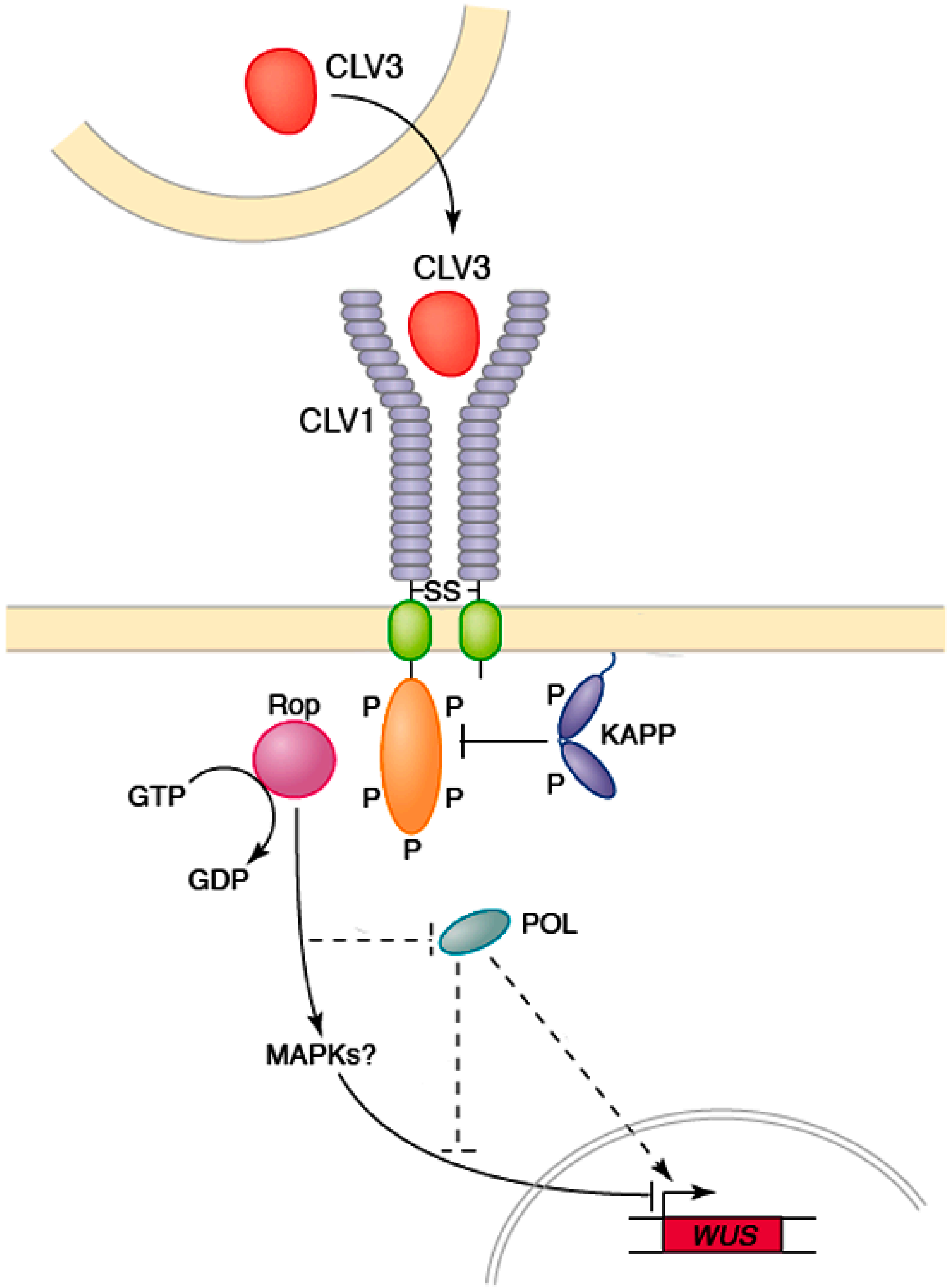


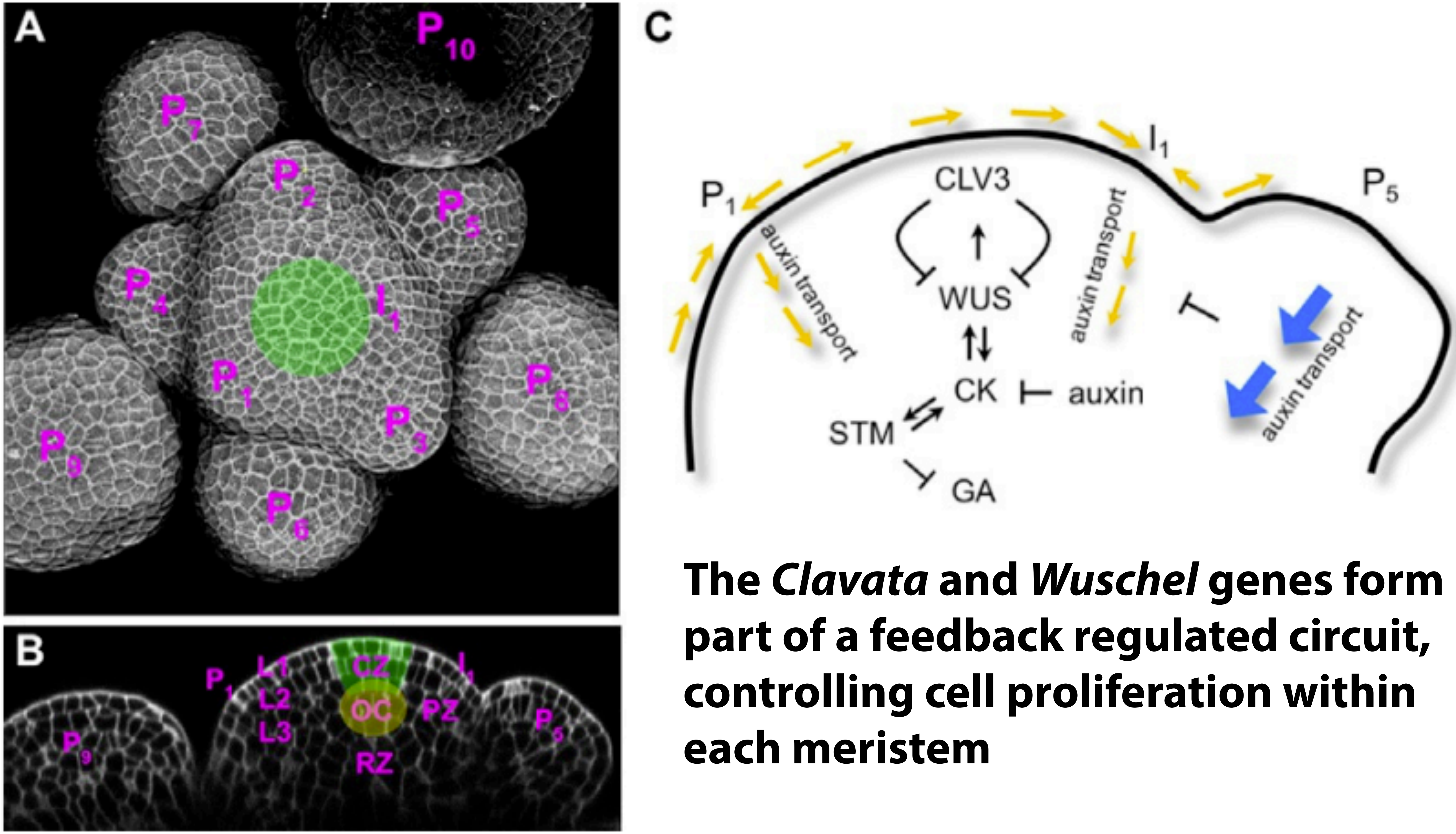

CLV3 Is Localized in the Extracellular Space.




**Clavata 3 gene function is required to maintain meristem size, and sufficient to alter meristem size - Like Wuschel, but in opposition**

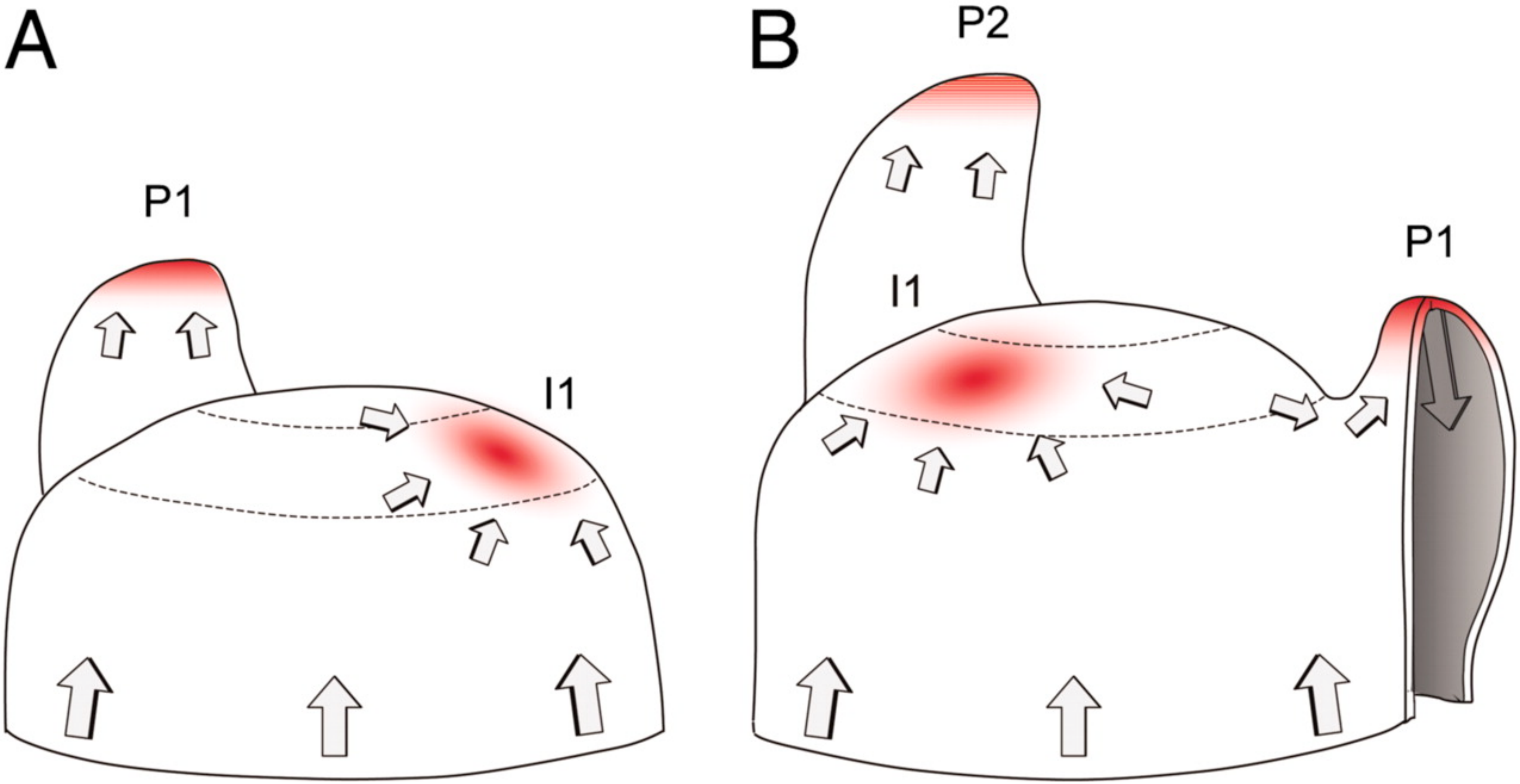
**CLAVATA3 acts as a signal across the meristem, repressing WUSCHEL activity in the central zone of the shoot meristem.**



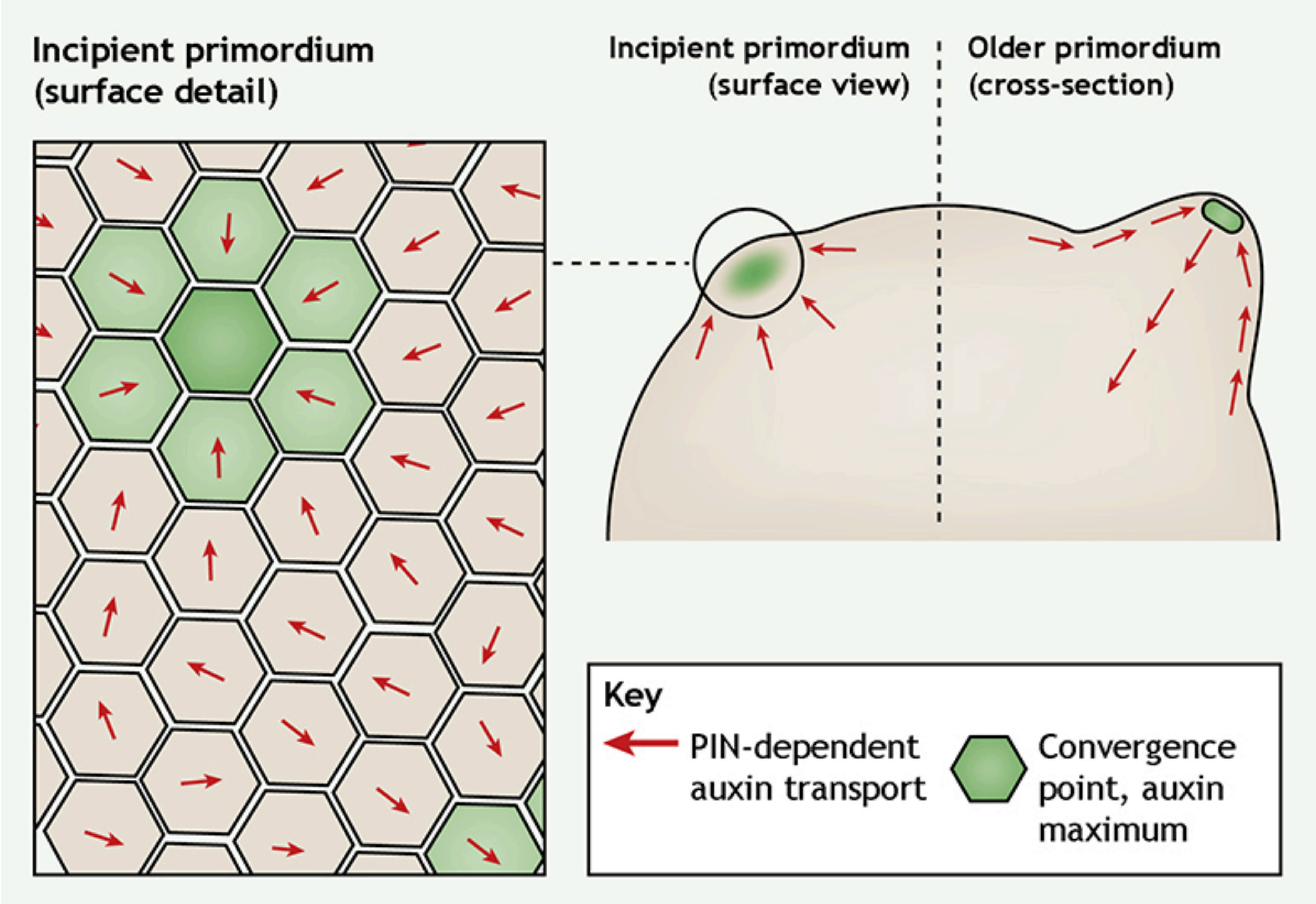



**The *Clavata* and *Wuschel* genes form part of a feedback regulated circuit, controlling cell proliferation within each meristem**

**Wus activates a local cytokinin response in the meristem**

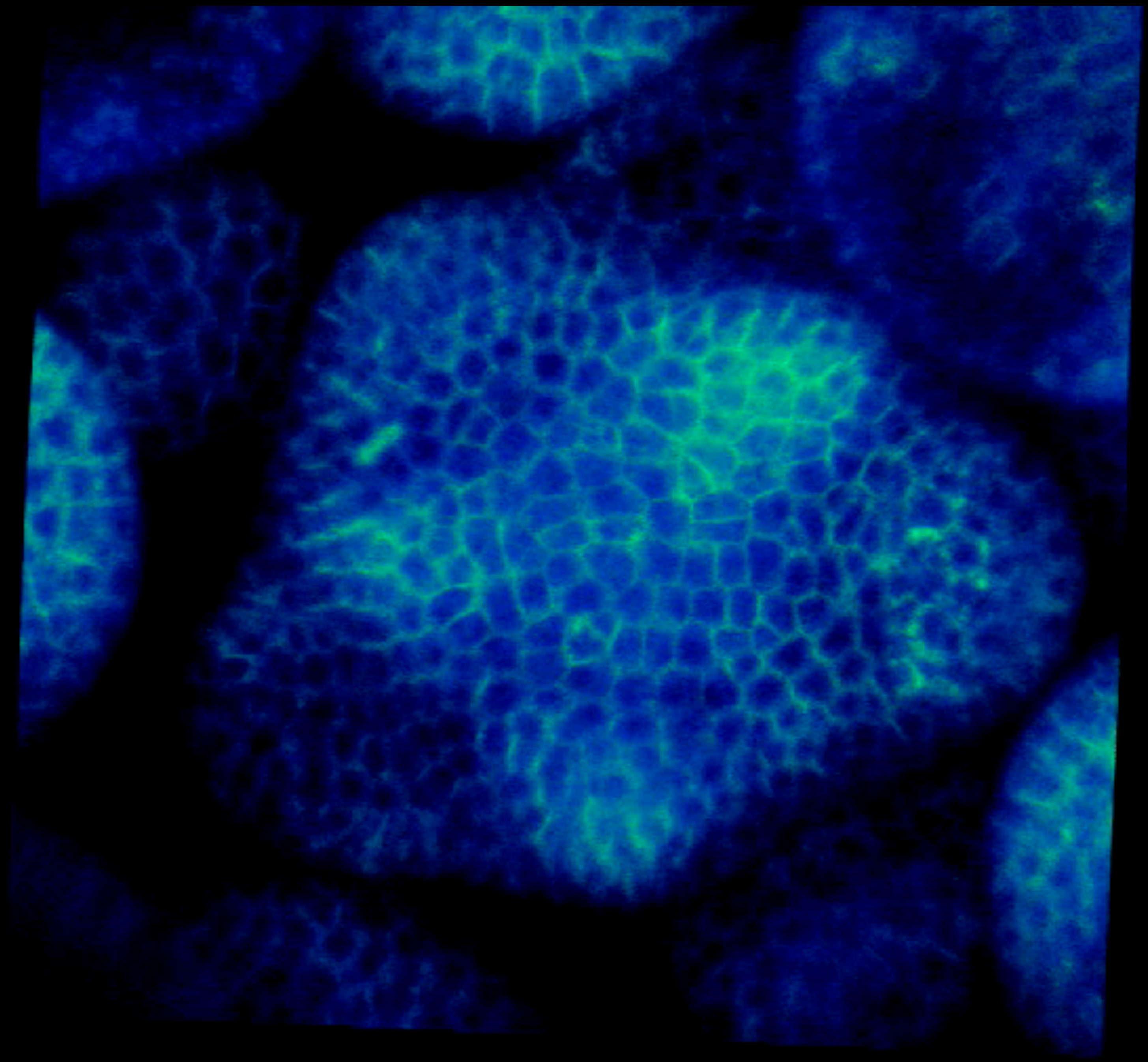


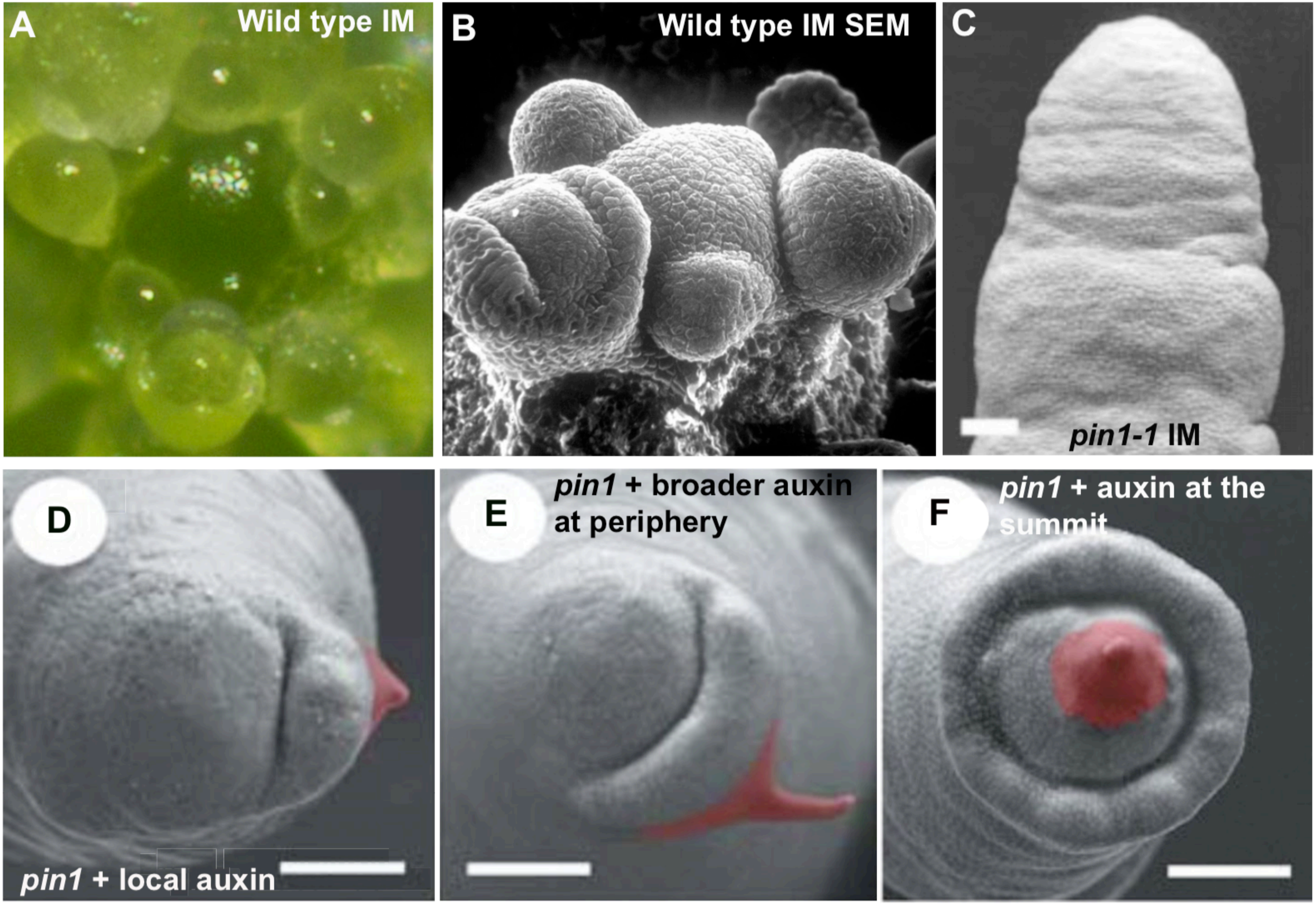

**Auxin regulated feedback initiates meristem outgrowth**




**Competitive interactions between self-reinforcing flows of auxin are responsible for initiation and spacing of shoot meristem primordia**

# PIN1:GFP distribution in the Arabidopsis shoot apical meristem






**Local application of auxin induces outgrowth of primordia in *pin1* mutants**

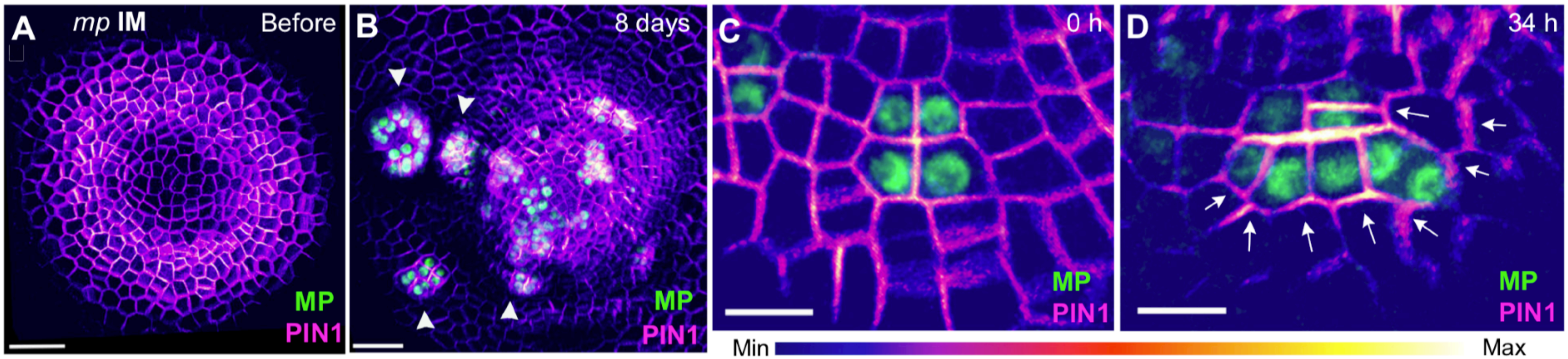



**Fig. 6. MONOPTEROS (MP) orients PIN1 polarity non-cell-autonomously.** (A,B) Confocal projection of *mpB4149* mutant IM before (A) and 8 days after (B) the induction of MP-YPet clones (green). PIN1 expression is in magenta. Arrowheads indicate organs initiating from cells expressing MP clones. (C) Magnified view of an *mpB4149* mutant apex containing a 4-cell MP-YPet clone (green), showing PIN1-GFP polarity and expression (magenta) 2 days after induction (i.e. when the clone was first visible). (D) Magnified view of the MP-YPet clone shown in C 36 h later, showing an increase in PIN1 expression within the clone and polarization of PIN1-GFP (magenta) in neighboring cells directed towards the clone (arrows). Scale bars: 30 μm A,B; 10 μm in C,D. Adapted from Bhatia et al. (2016).

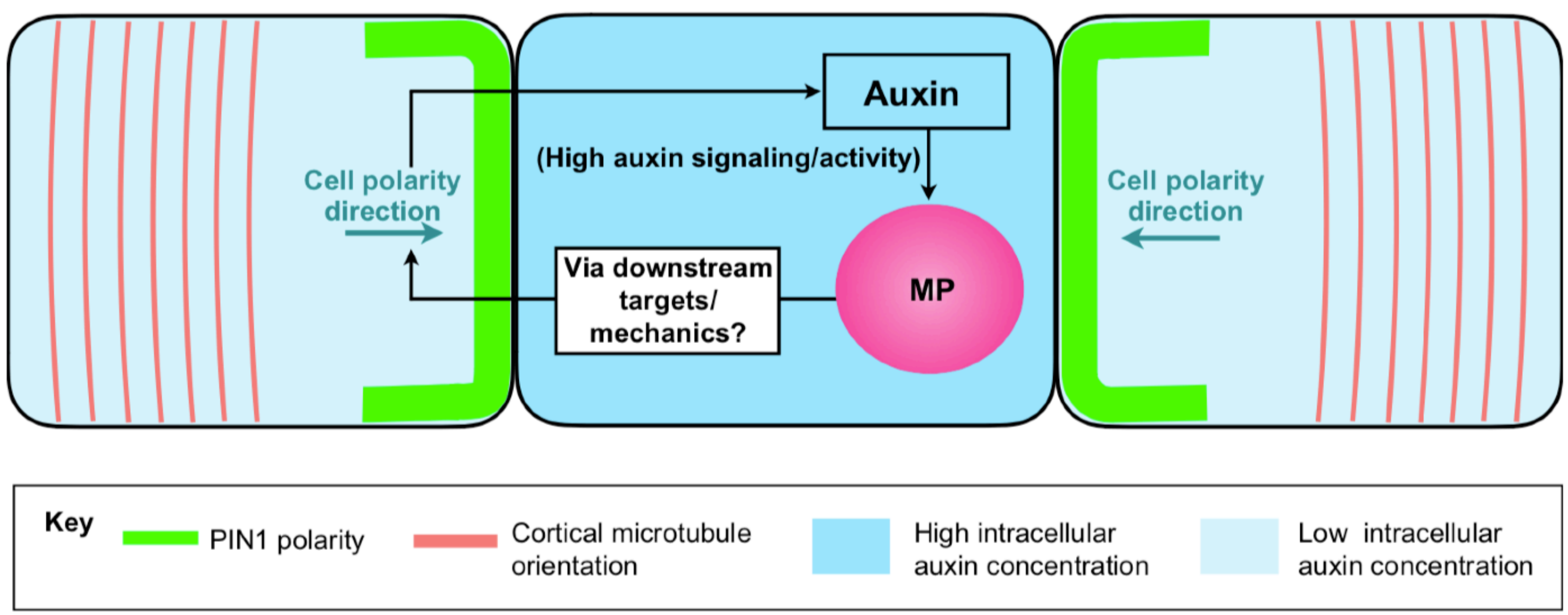
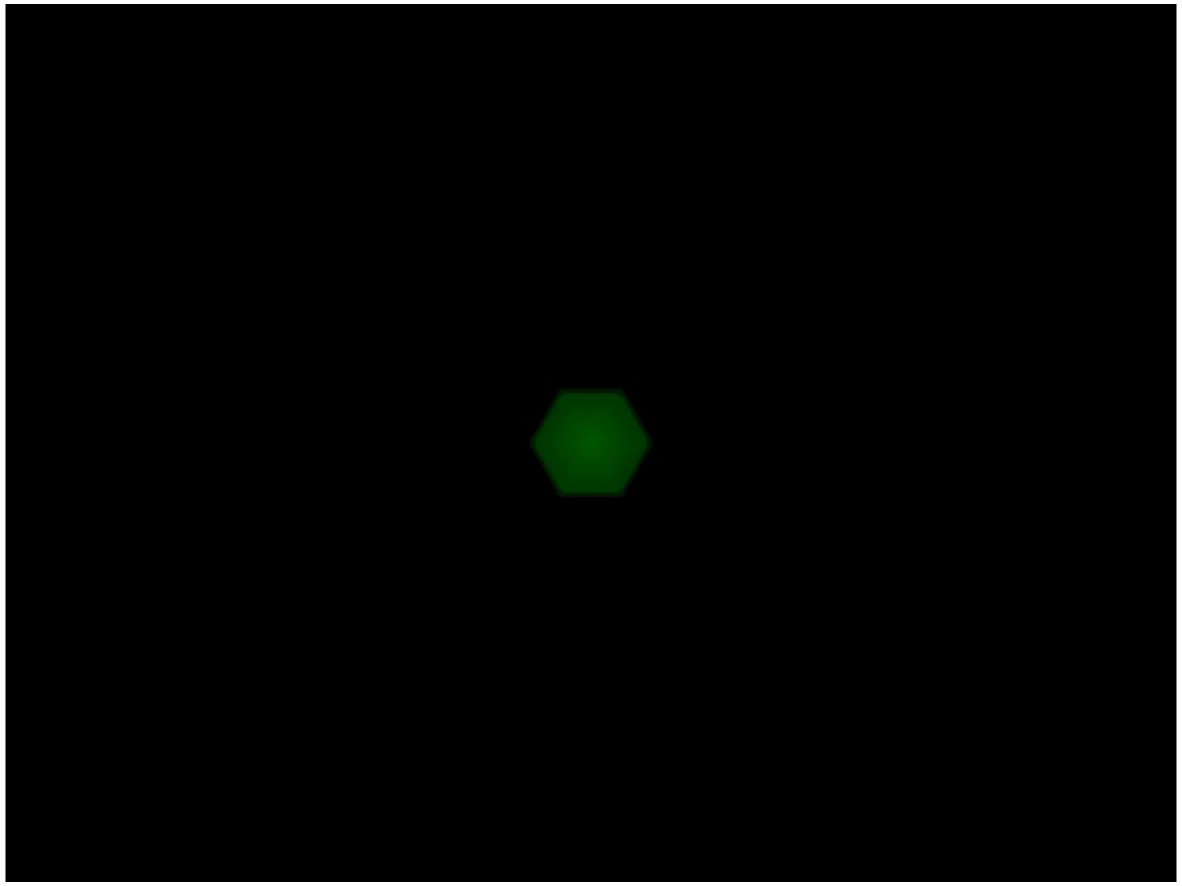
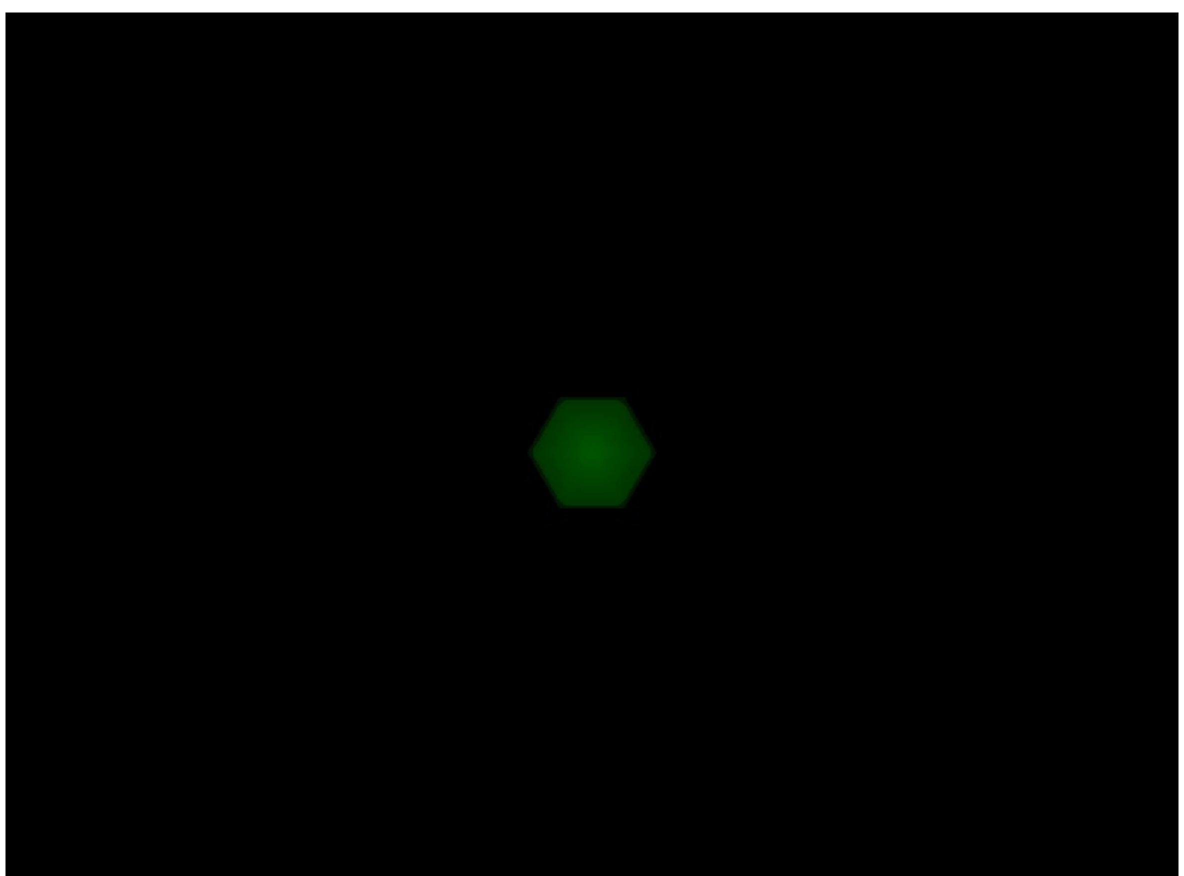
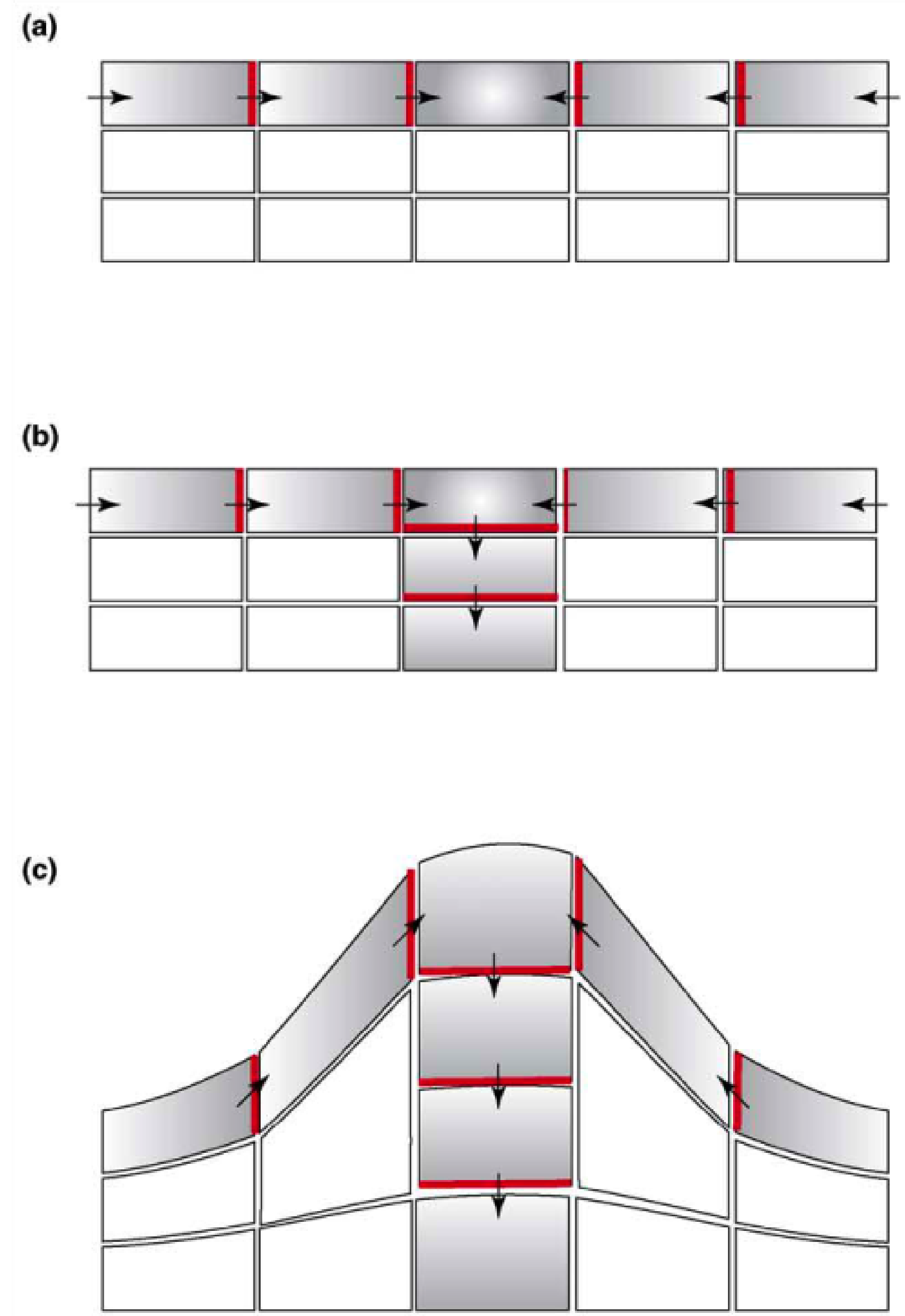



**Fig. 8. A feedback loop at the cell-cell communication level generates cell polarity patterns at the tissue level.** A positive-feedback loop between auxin abundance and its transport orients PIN1 polarity and microtubule orientations in the neighboring cells non-cell-autonomously, towards the cell with high auxin. This loop acts via a localized auxin transcriptional response mediated by MP activity. This substantiates the proposed up-the-gradient model in organizing complex cell polarity patterns underlying visible phyllotactic patterns. How MP acts to mediate such a response is yet to be discovered; it might act through its downstream targets or via altering cellular mechanics but these hypotheses need to be tested *in vivo*. See Bhatia et al. (2016).

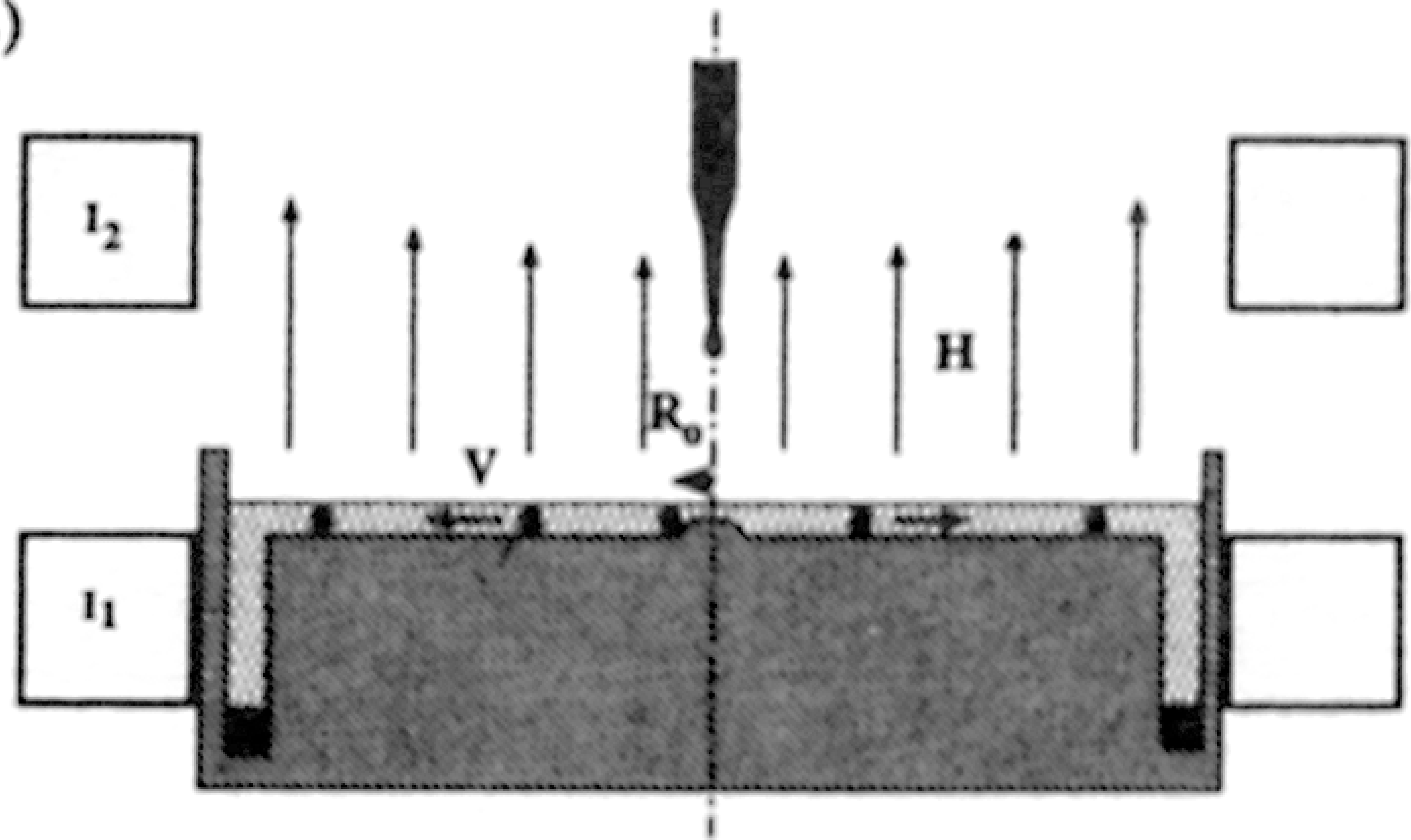
# Primordial outgrowths in the meristem are triggered by local influxes of auxin

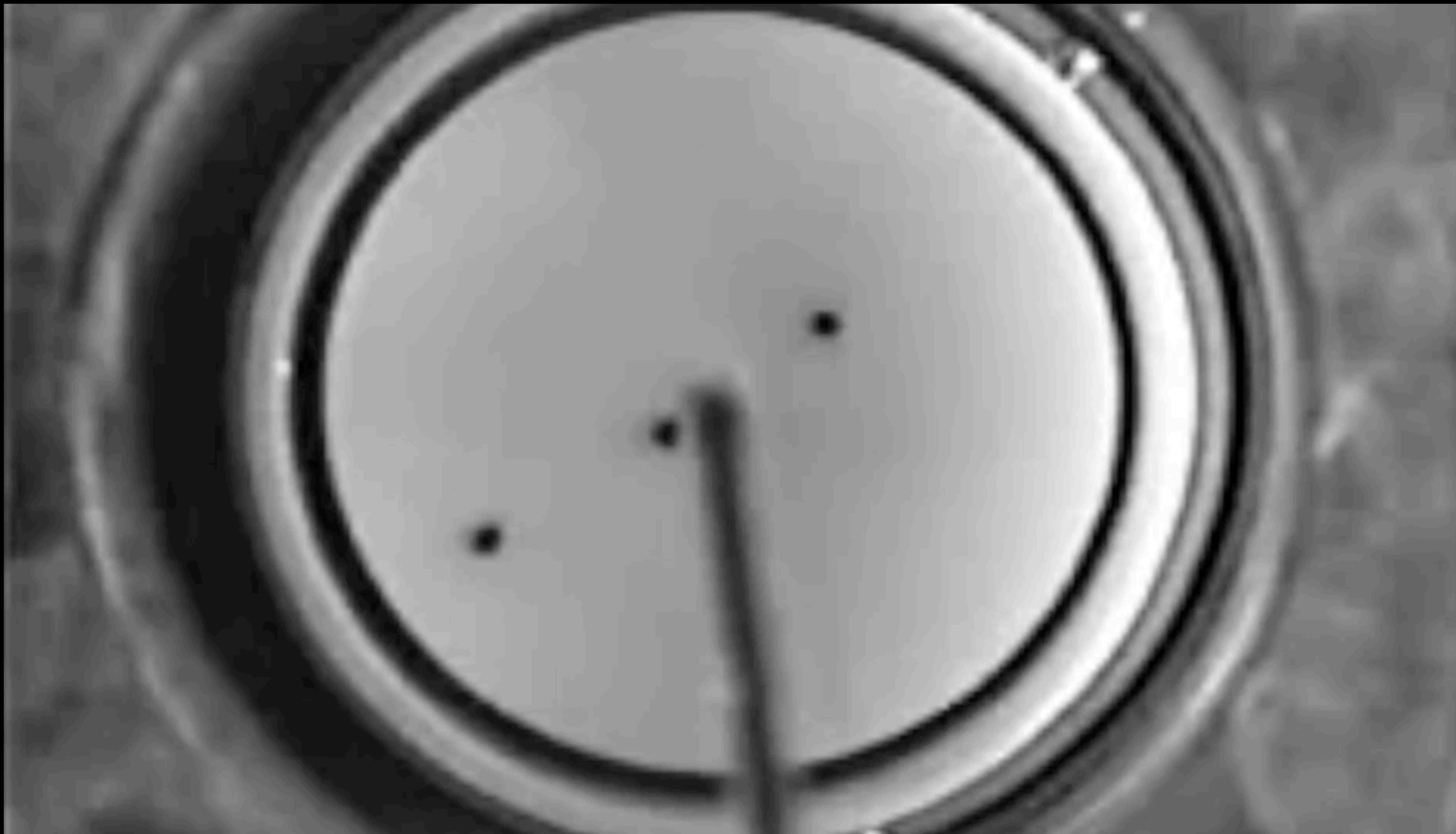


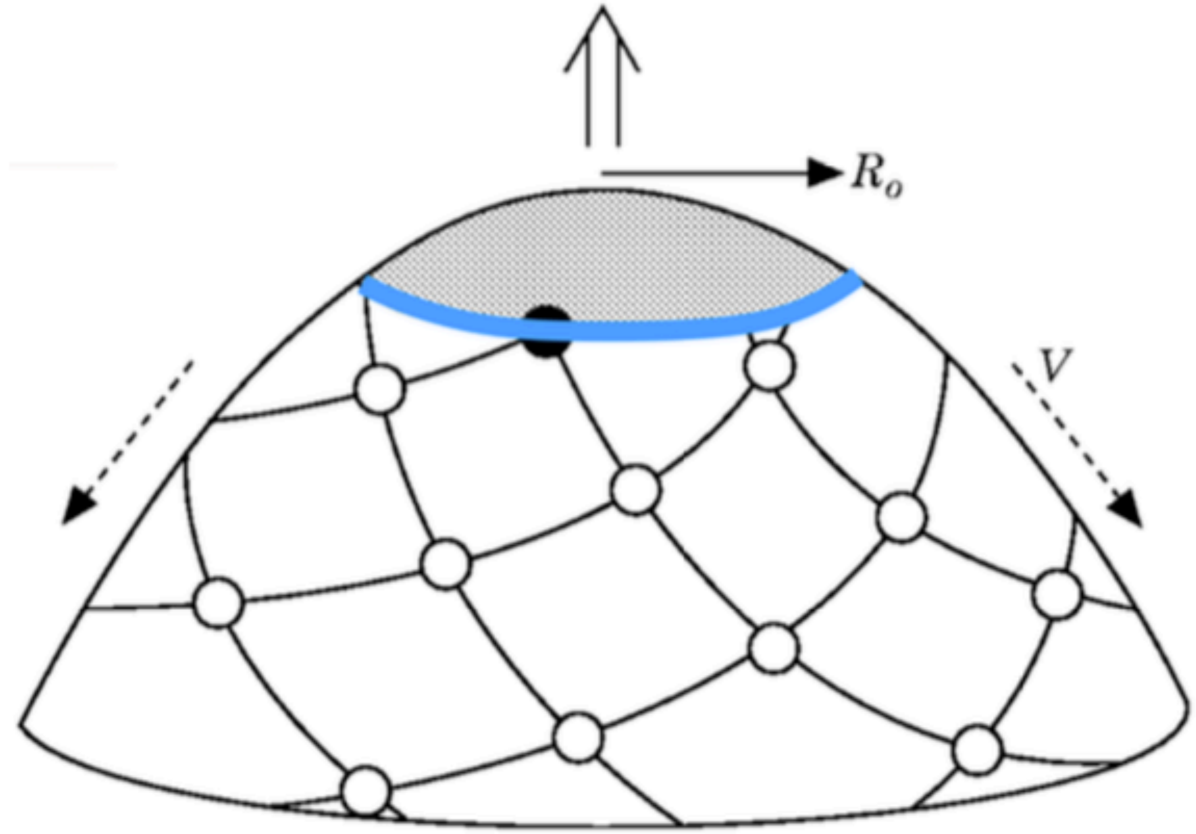




(b)







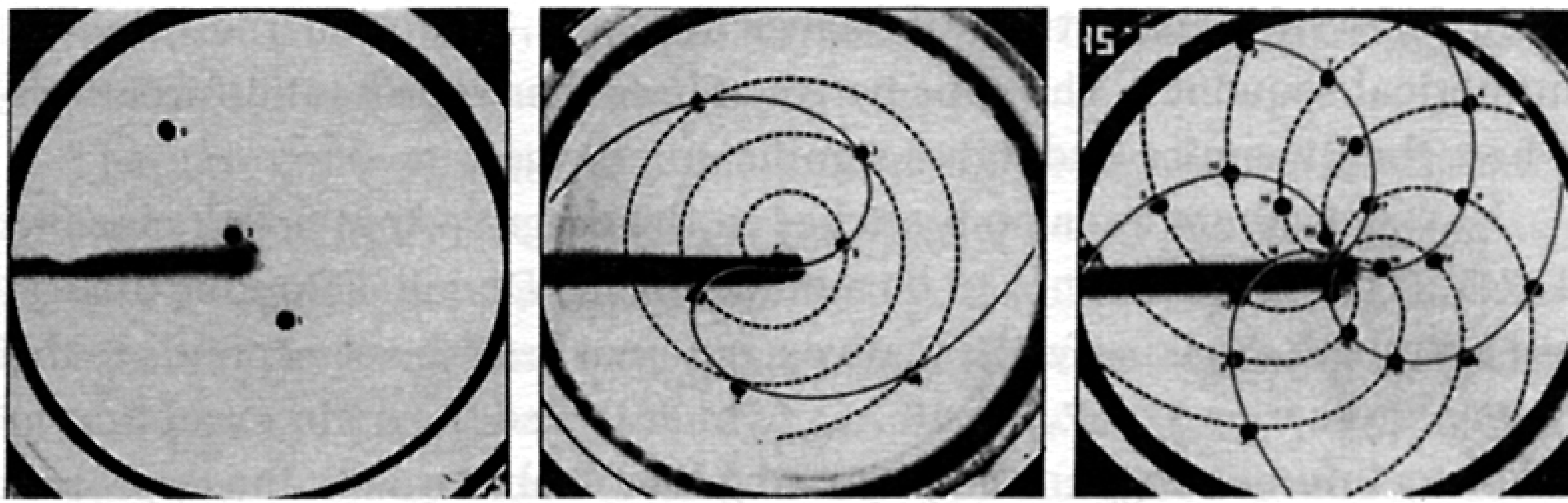
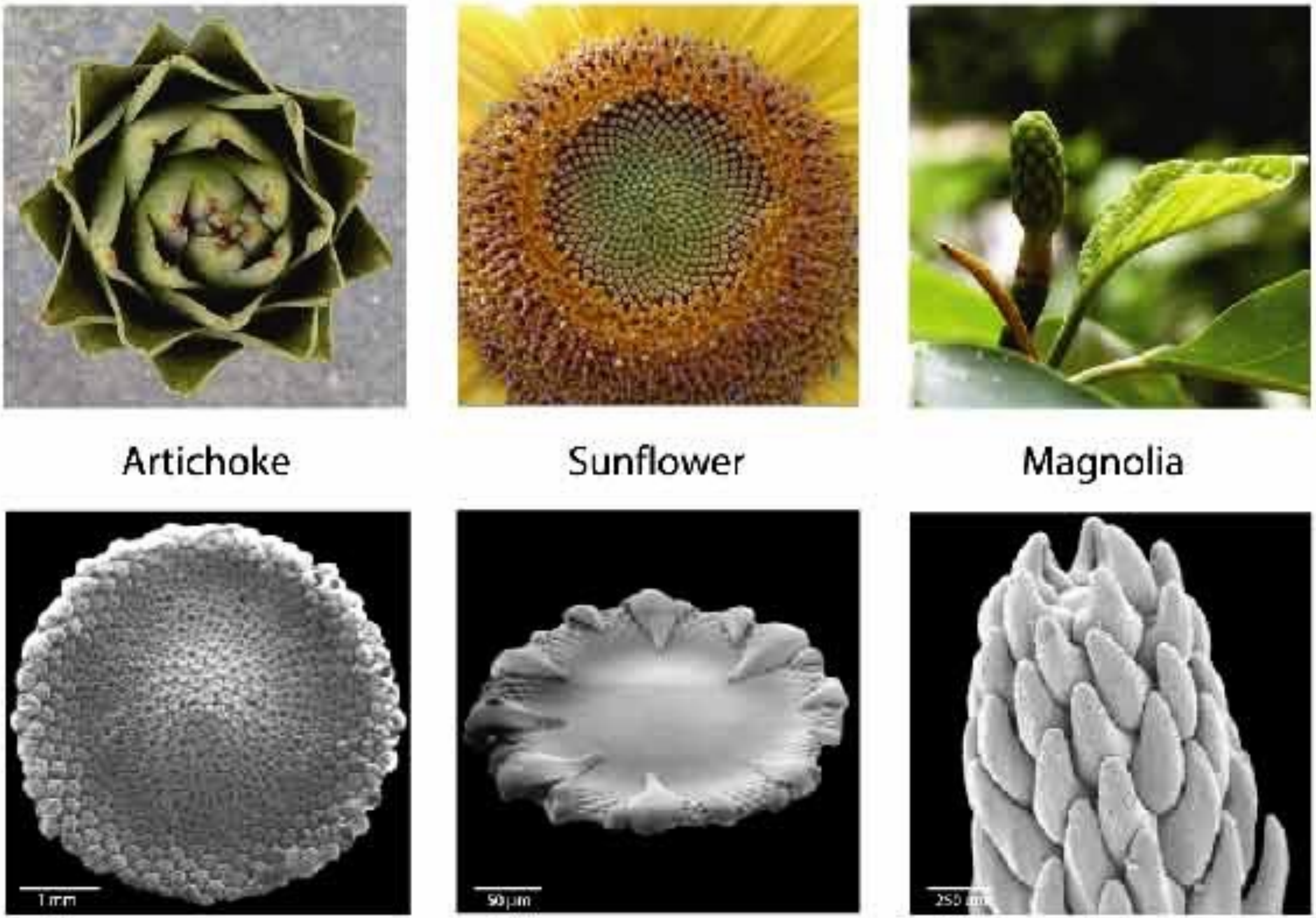



**Figure 50**

Fibonacci spirals observed in an experiment with electrically charged oil drops.

# Emergence of patterns at microscopic scales

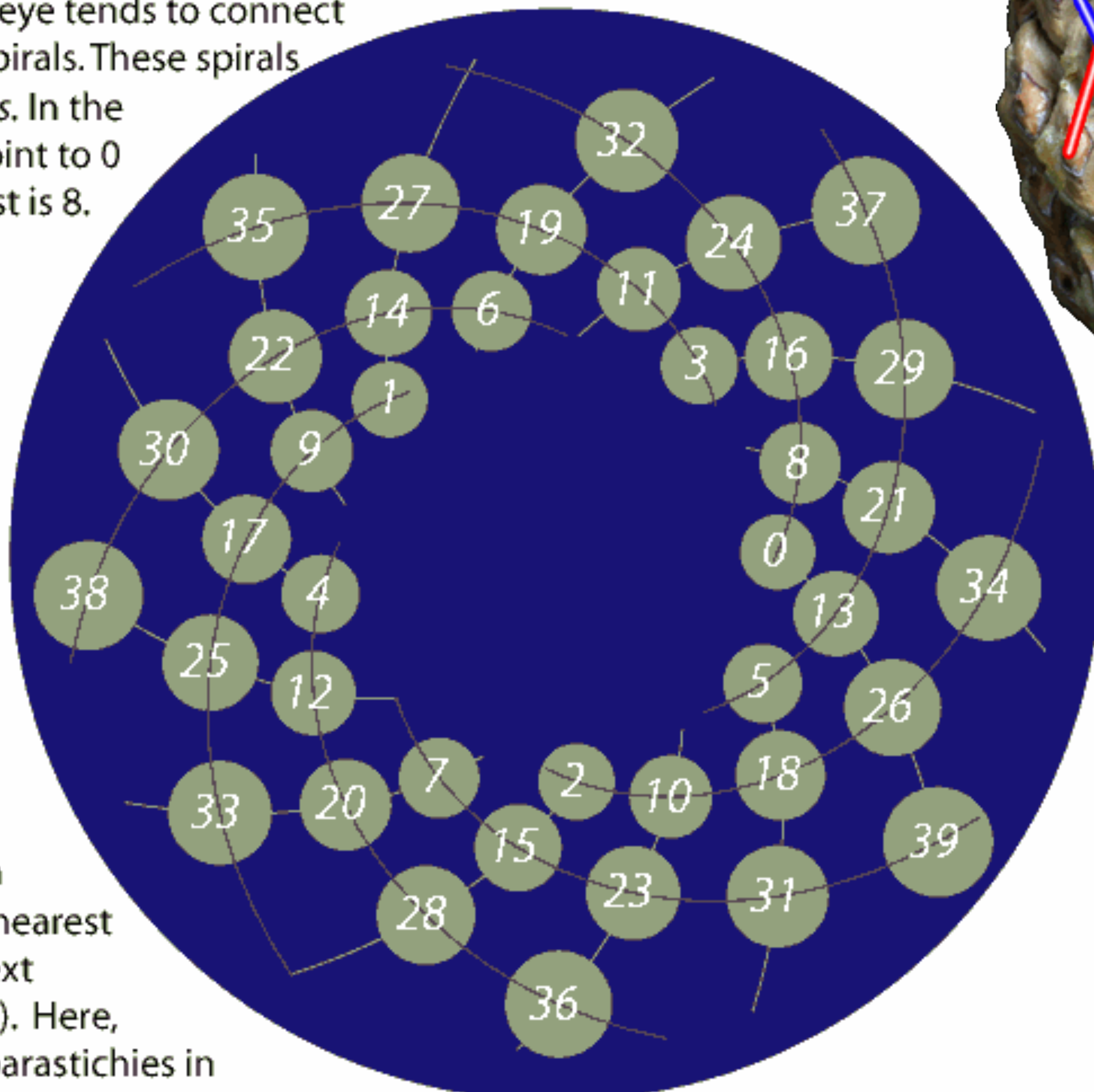


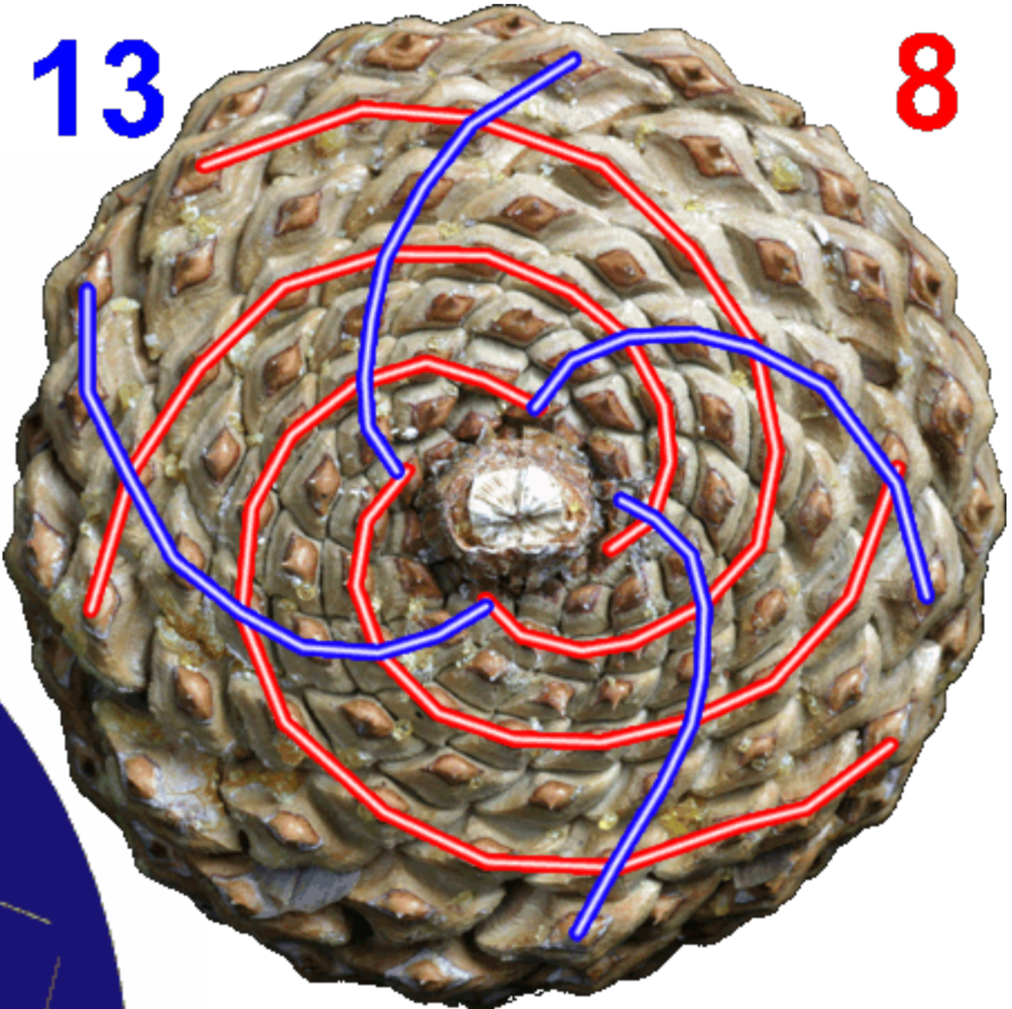
# Visible Spirals: Parastichies

In a spiral lattice, the eye tends to connect nearest points into spirals. These spirals are called *parastichies*. In the figure, the nearest point to 0 is 13; the next nearest is 8.

There are two sets of parastichies winding in different directions. In the figure, 0, 8, 16 … winds in one direction and 0, 13, 26 … in the other.

The number of parastichies in each direction is the difference between a point, e.g., 7, and its nearest neighbor (20) and next nearest neighbor (15). Here, there are 15 - 7 = 8 parastichies in one set and 20 - 7 = 13 in the other.




Spiral lattices are classified according to the number of parastichies in each set. This lattice is (8, 13).

## Plant organs and the Fibonacci series:

**1 petal:** Lily
**2 petals:** Euphorbia
**3 petals:** Iris
**4 petals:** Arabidopsis, Fuchsia (decussate arrangement, not spiral)
**5 petals:** Buttercup, wild rose, Larkspur, columbine (Aquilegia), pinks
**8 petals:** Delphinium
**13 petals:** Ragwort, corn marigold, cineraria, some daisies
**21 petals:** Aster, black-eyed susan, chicory
**34 petals:** Plantain, Pyrethrum
**55, 89 petals:** Michaelmas daisies, the Asteraceae family

(With some variation in numbers due to noise)

# Feedback-regulated auxin traffic and responses play a key role in coordination of whole plant growth

1. Plant body plans are flexible, and are built step-wise through a series of local interactions.
2. Auxin is a mobile informational molecule and its directed traffic plays an important role in establishing key landmarks during cellular development in plants.
3. Auxin triggers specific genetic responses in cells via Aux/IAA and ARF pathways.
4. Genetic responses can trigger coordinated behaviour in adjacent cells, creating feedback systems e.g. PIN1-MP, WUS-CLV3 across meristems
5. Competition for auxin can cause lateral inhibition and result in spatial patterning in responsive tissues, e.g. phyllotactic patterning.
6. PIN-mediated transport of auxin results in coupling of cells, and formation of long-distance interactions that can regulate the balance of growth across tissues or the entire plant.