

***From Genome to Phenotype:
Modeling the interaction of physical
and chemical signals in plant
meristems***

**Meyerowitz Lab and
many collaborators**



Needs to understand tissues, morphogenesis and development:

Image Analysis

Computational Models of Chemical
Signals

Physical Models

Connection between Physical and
Chemical Models and Substrate

Connection between Chemical and
Physical Models





0 Gy

100 Gy

ATM/ATM



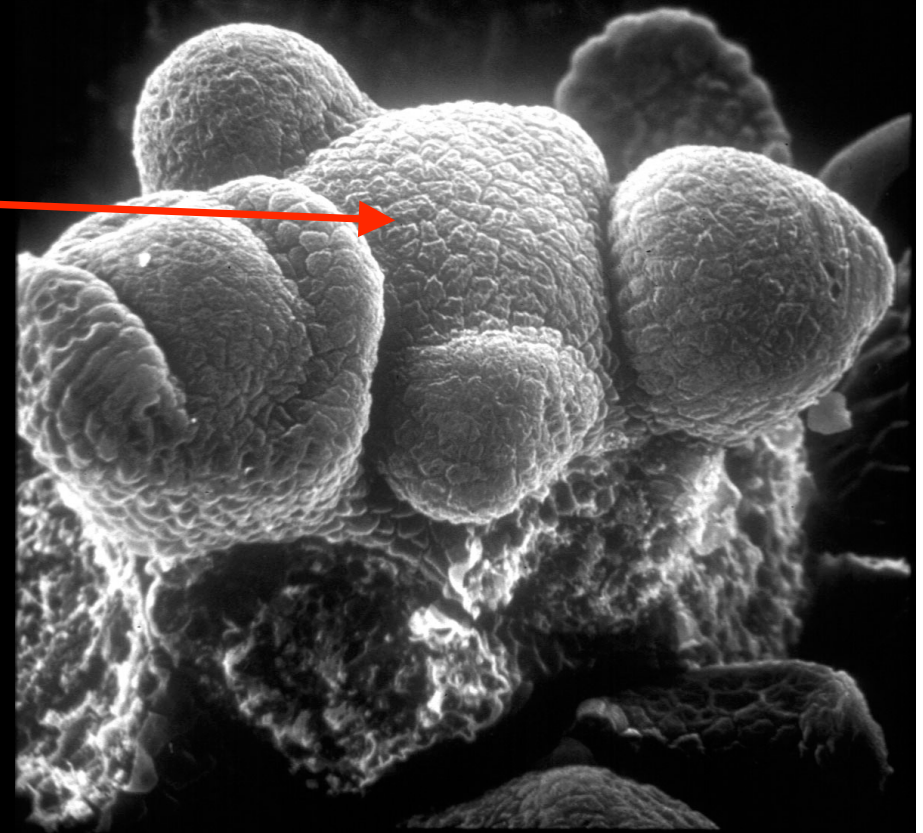
atm-2/atm-2



Where the action is: The *Arabidopsis* shoot apical meristem

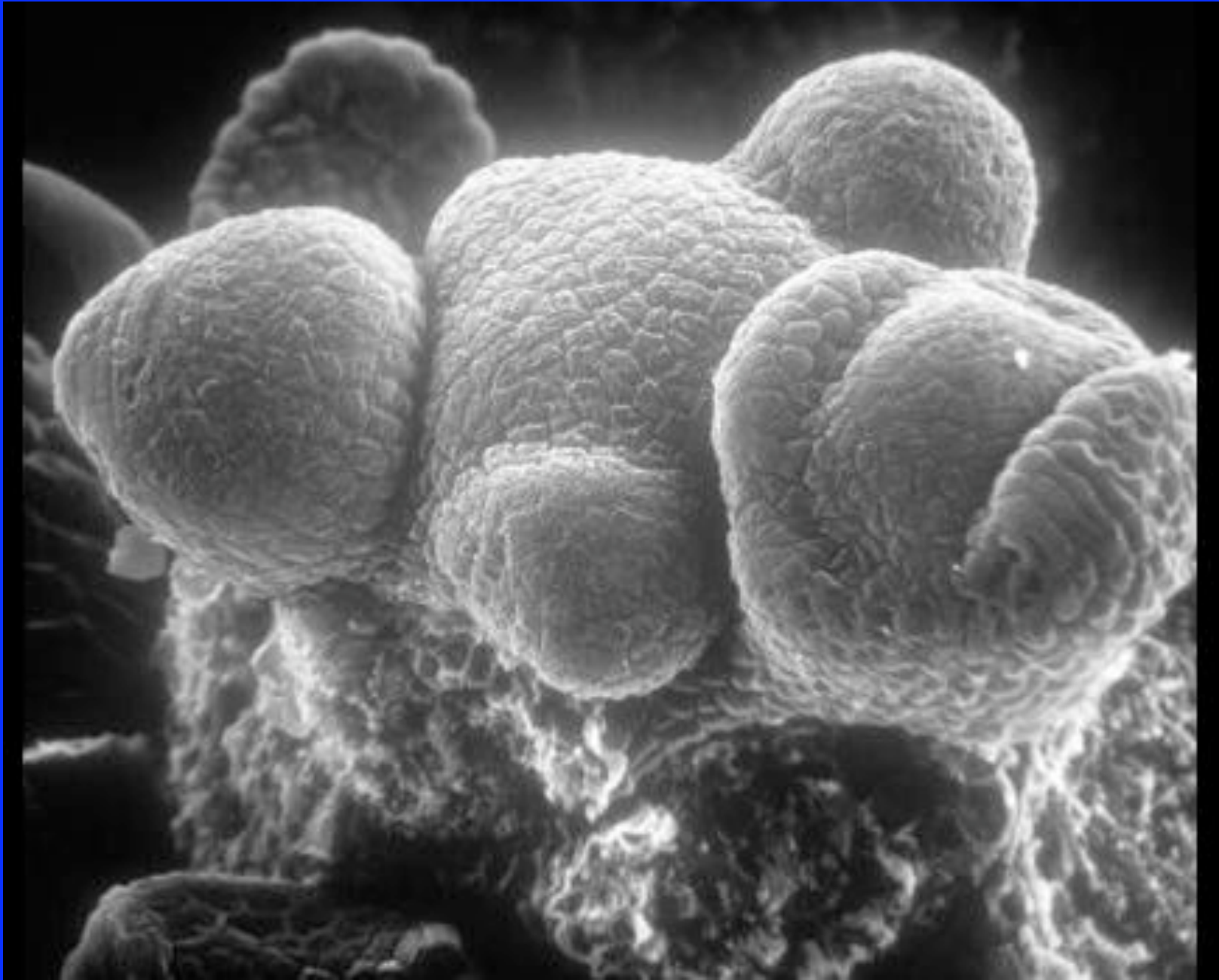


shoot
meristem

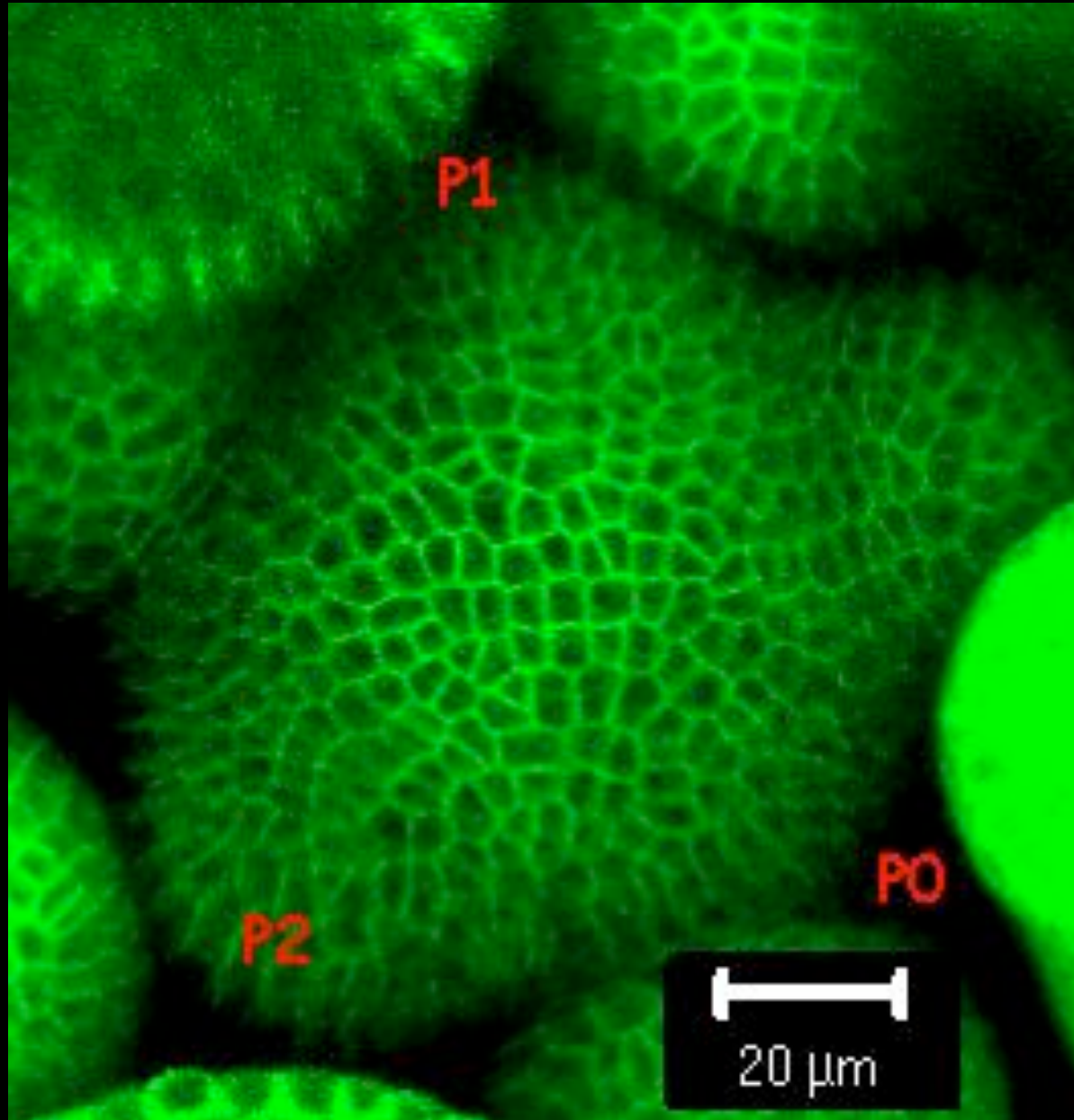


028 000 P1 0.05 5-20

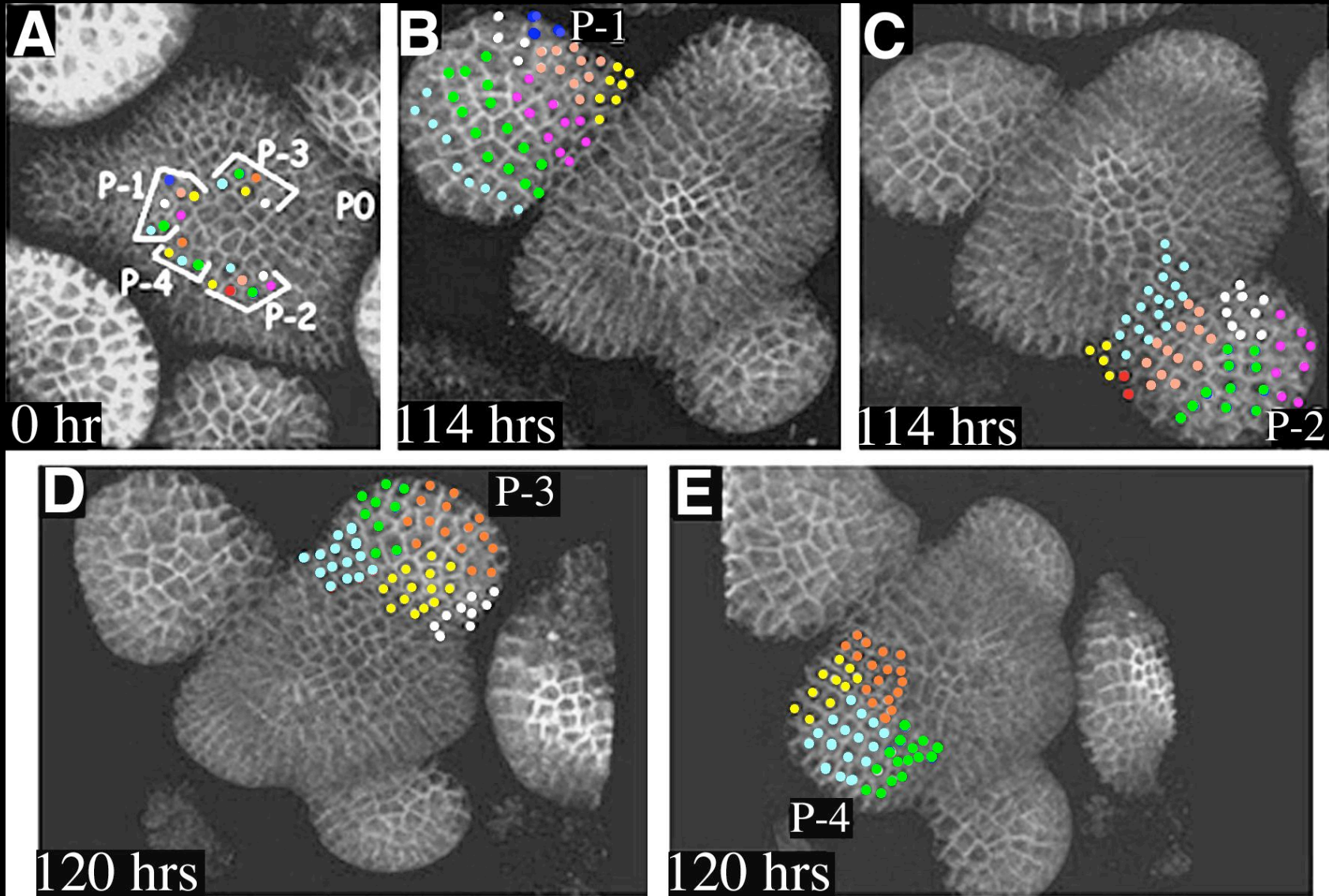
Shoot apical meristem



35S::YFP29-1, every 2 hours and 30minutes 65 hours

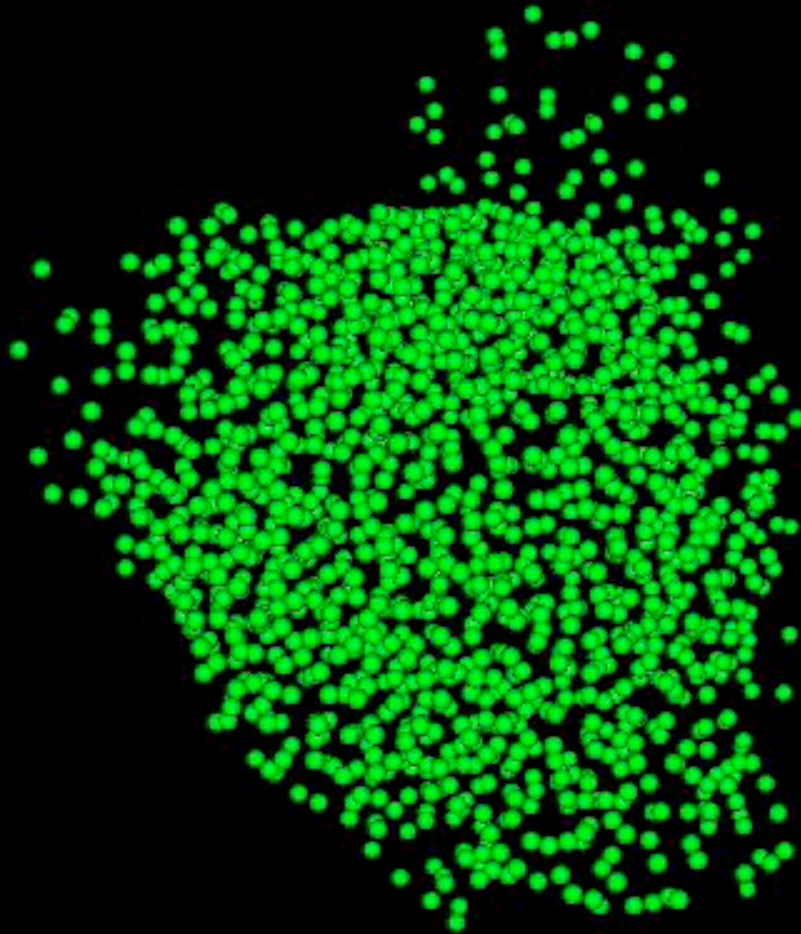


Lineage analysis

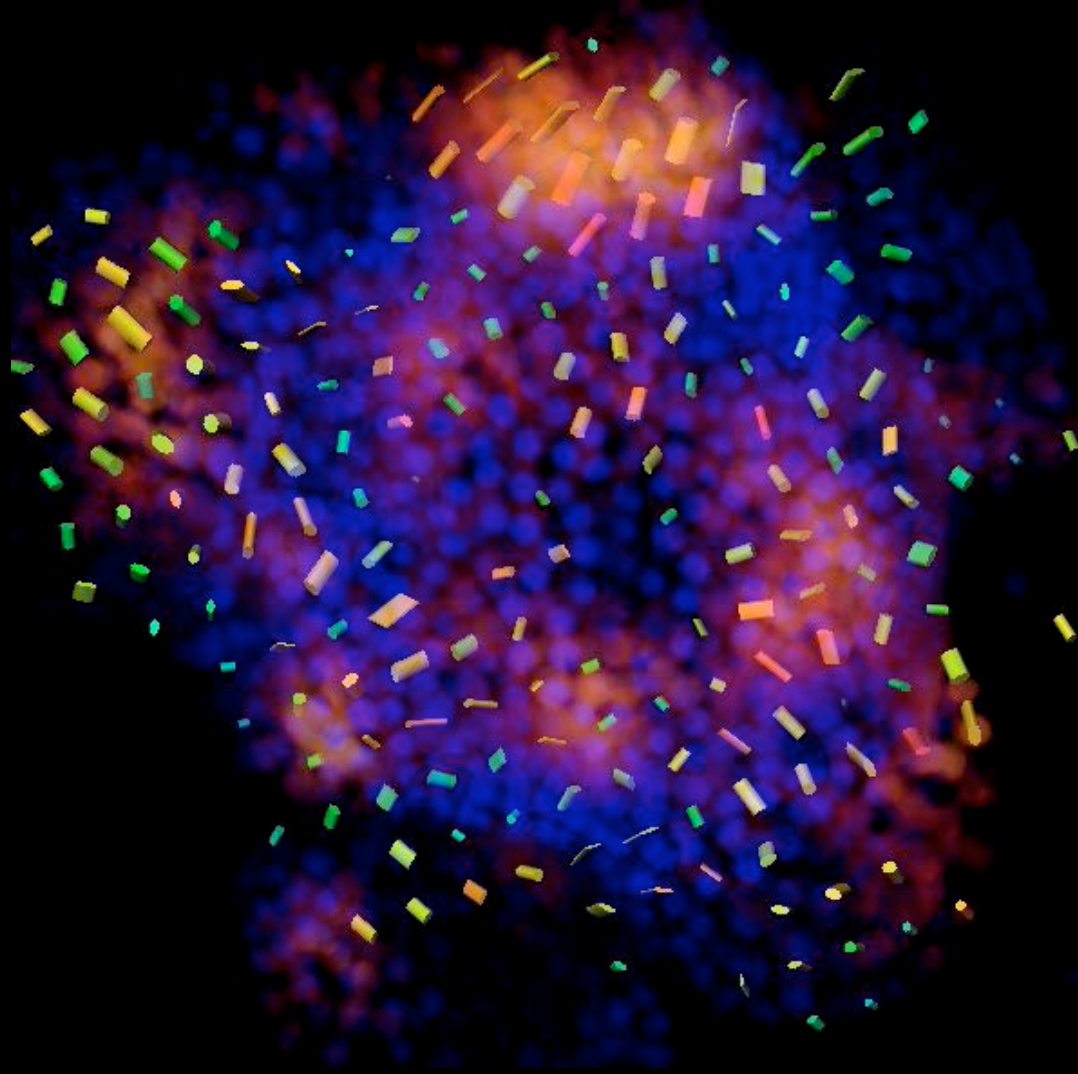


Developing lineages were traced by tracking individual cell divisions over 5 days

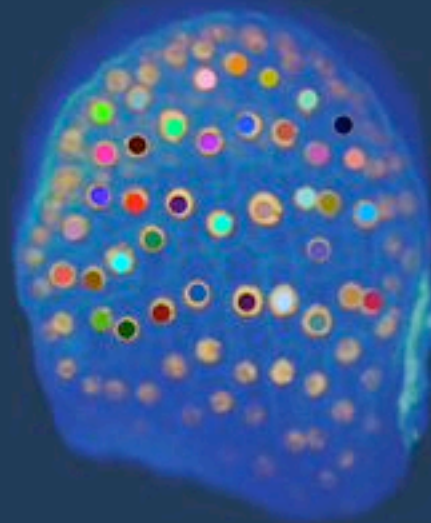
Animation of tracked and smoothed nuclear trajectories

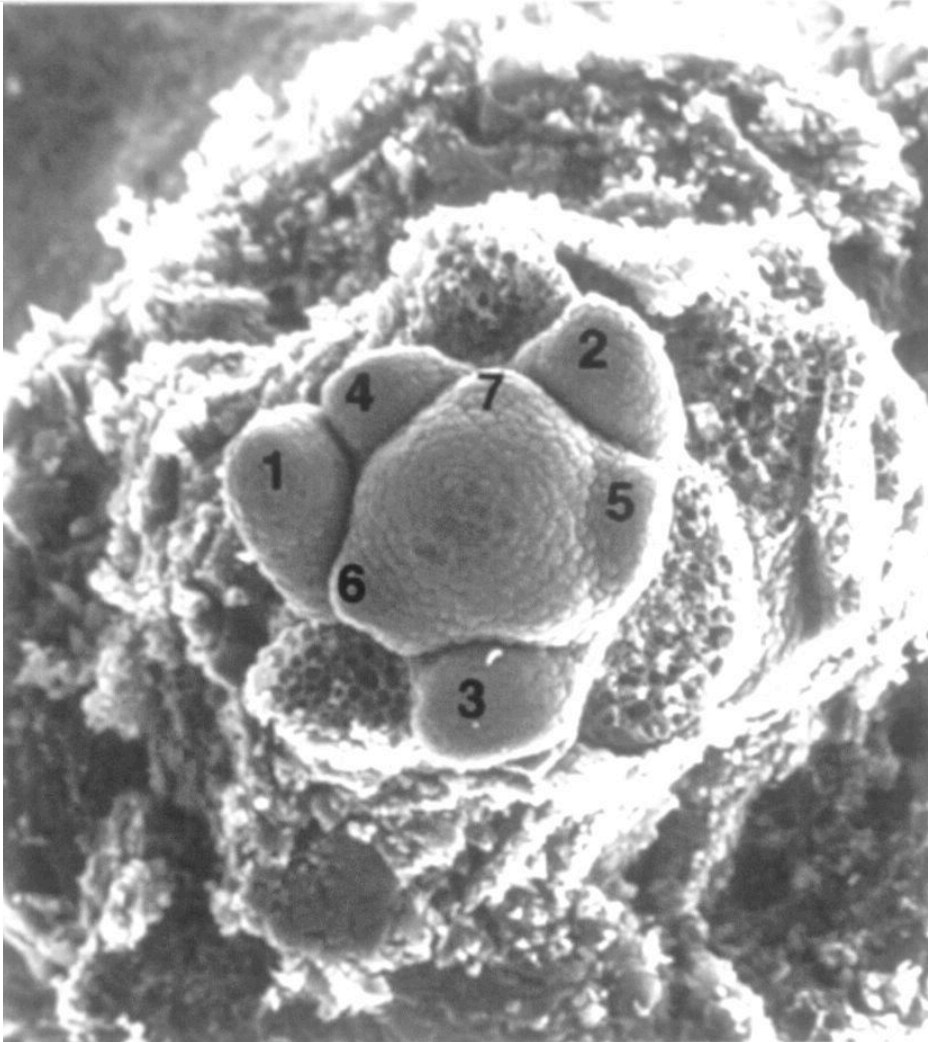


Principal directions of growth

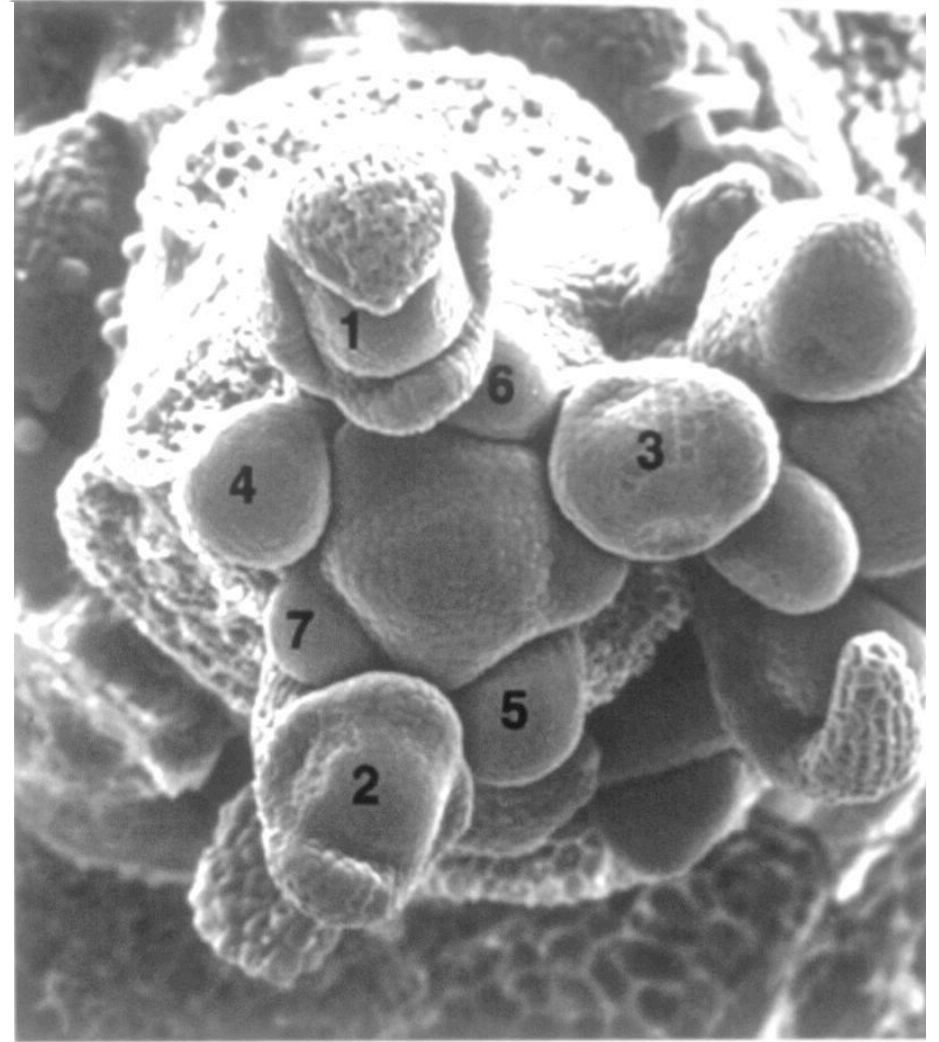


Flower Development, Too

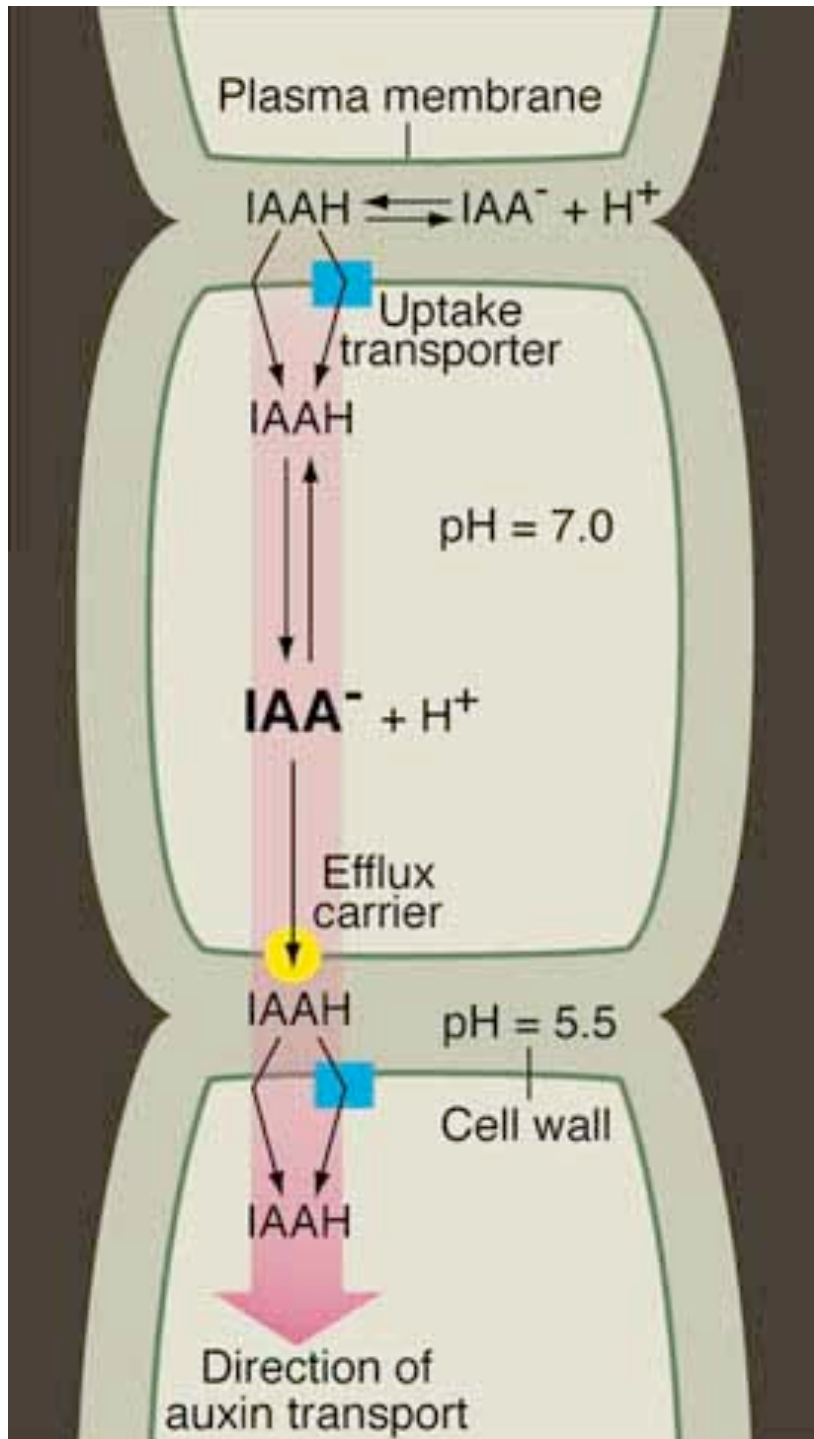




Vegetative SAM



Inflorescence SAM



Jones, A.M. Science (1998) 282, 2201

Previous work has shown:-

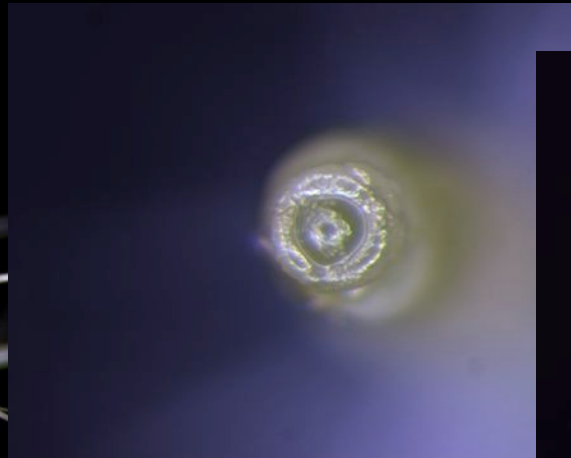
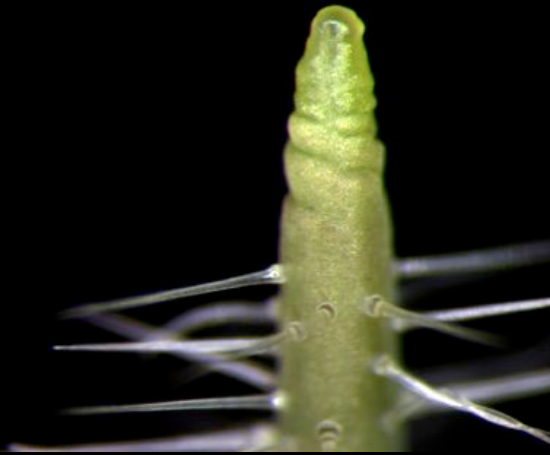
- Application of auxin paste to plant meristems causes lateral organ outgrowth at the site of application (Snow and Snow, 1937).
- When auxin transport is blocked, either in *pin1* mutants or in plants treated with chemical phytotropins lateral organs do not initiate (Okada et al., 1991).
- Auxin application can rescue organ initiation when transport is blocked (Reinhardt et al., 2000).

Taken together, these data suggest that endogenous auxin is required in some way for lateral outgrowth on the meristem flanks.

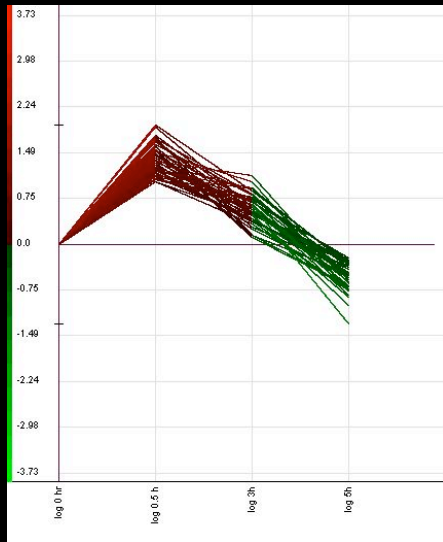
Micro-array analysis of auxin-induced genes in *pin1-1* mutants

Two days after treatment

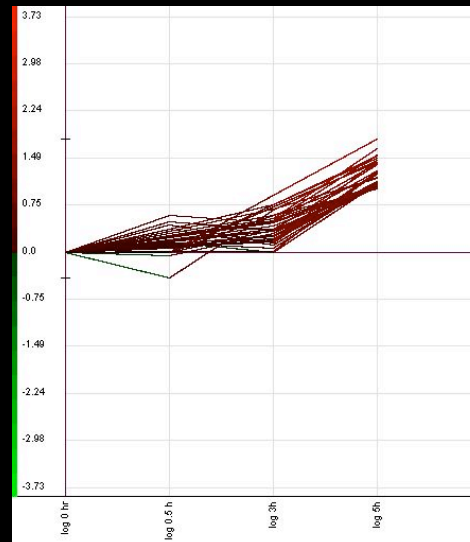
Three days after treatment



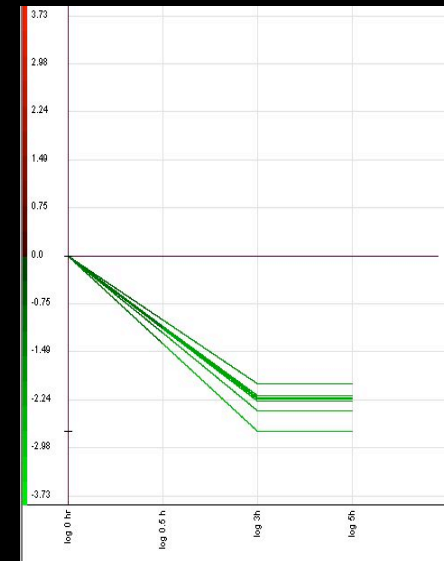
Examples of expression profiles



PIN1
AUX1
IAA4
AP2-like
Zinc finger
SAUR-like



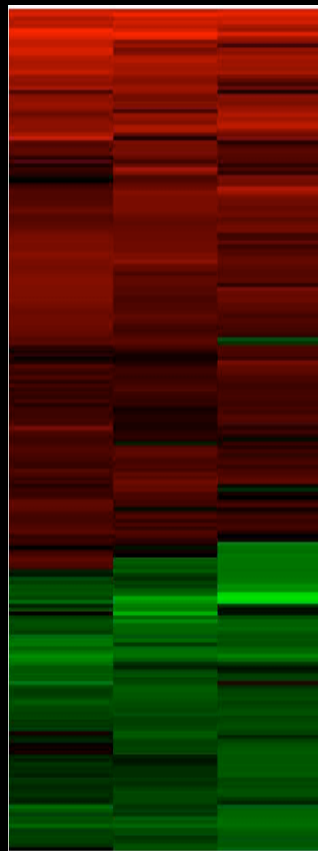
Zinc finger
Giberellin beta-hydroxylase



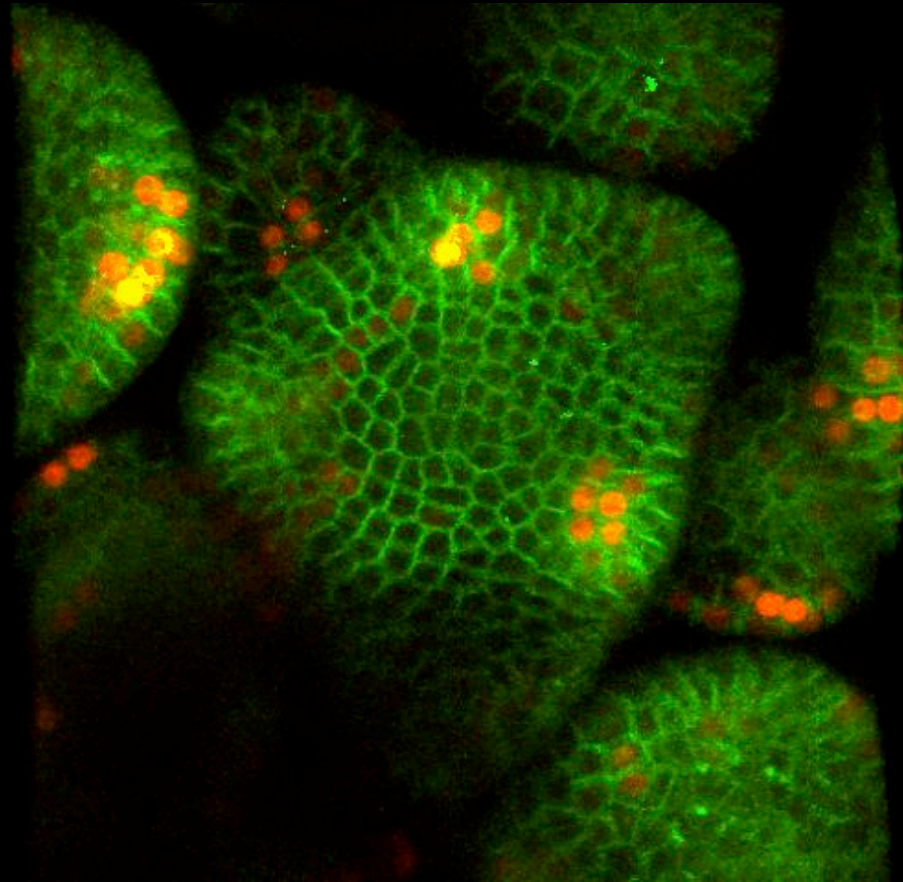
Polyubiquitin

PIN1 expression is auxin induced

Auxin induced genes on
pin1-1 apex



30 mins 5 hrs
 3 hrs

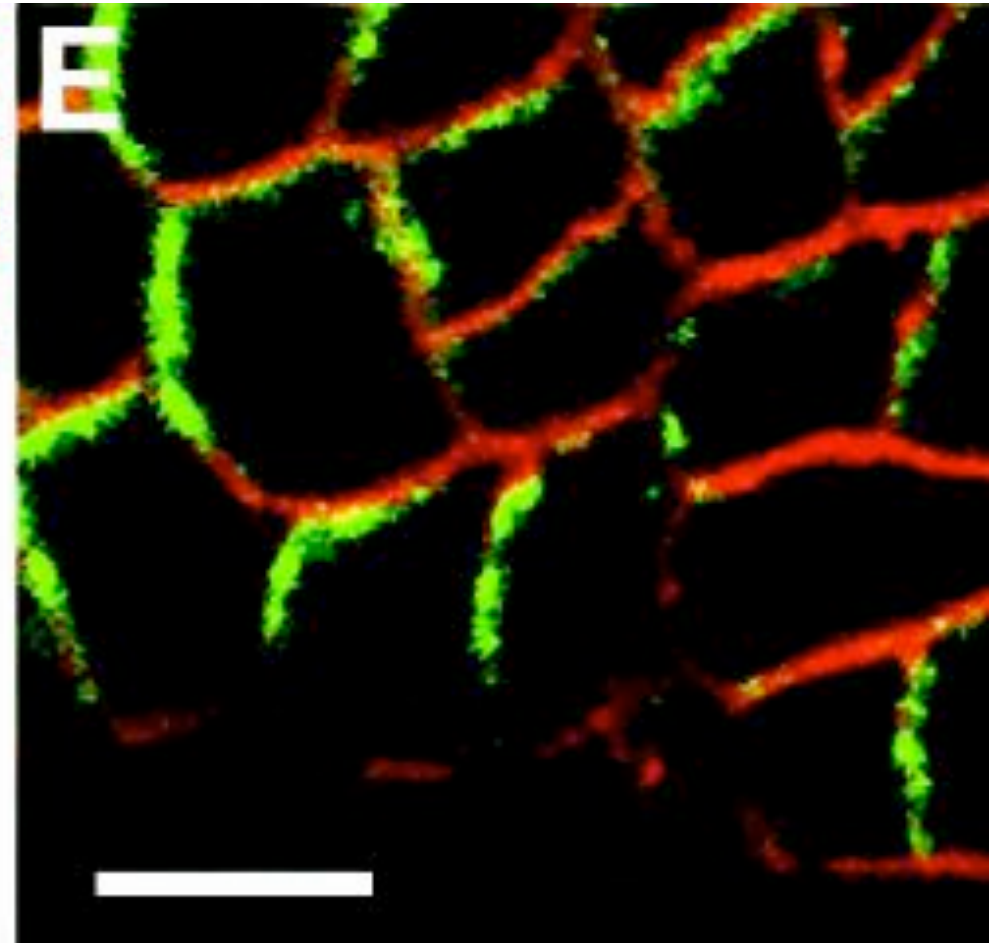
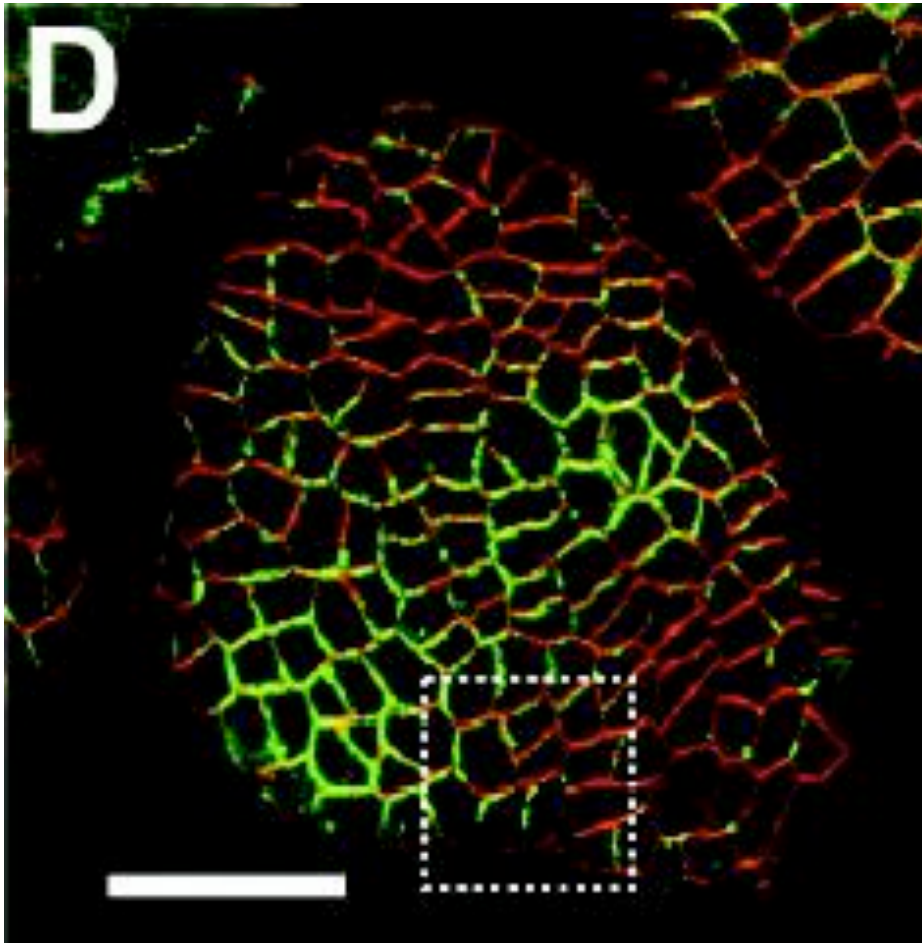


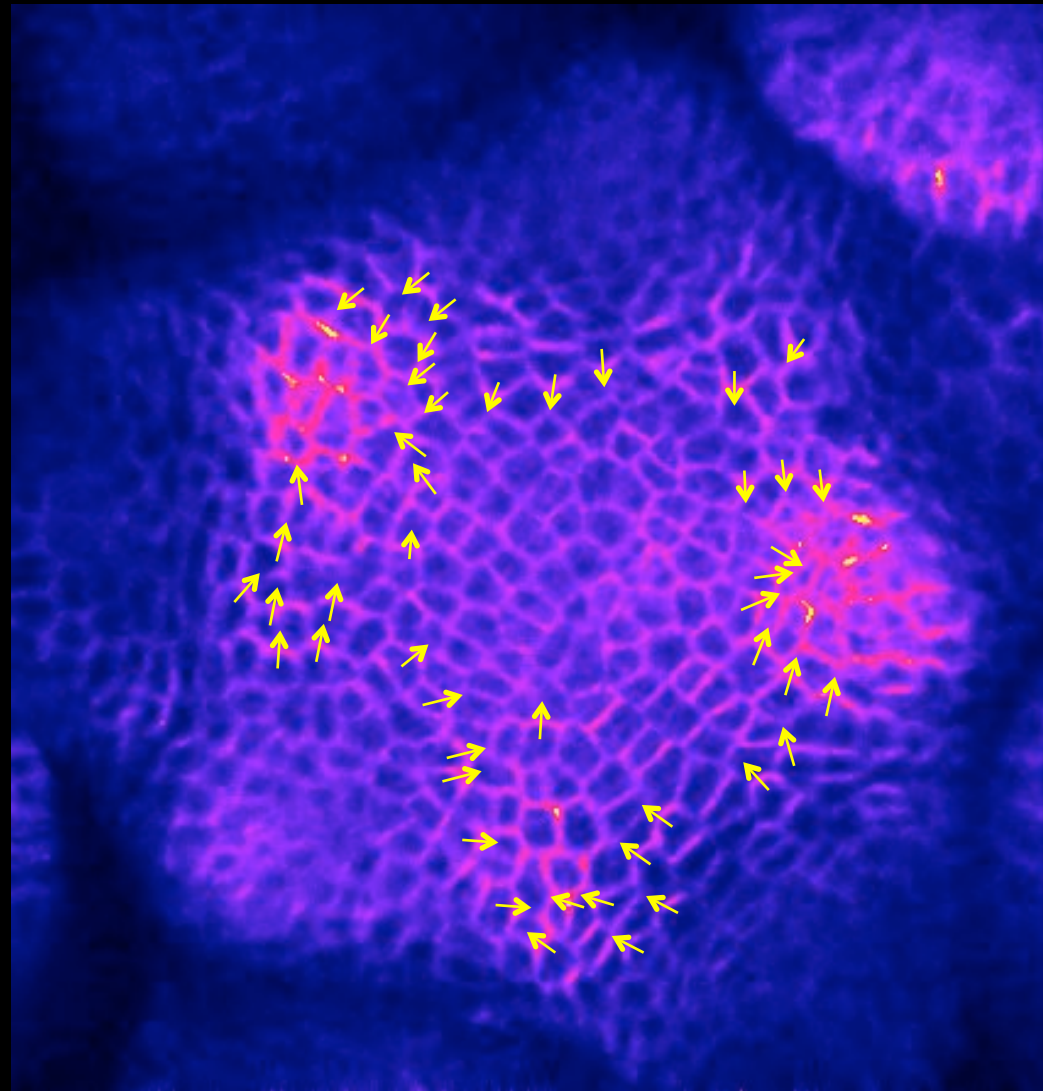
PIN1:GFP and DR5
expression

Time lapse imaging of PIN1GFP in the meristem
confirms expression dynamics



Green: PIN1-GFP
Red: Plasma Membrane Dye





Clues to a model.....

Model

- 1) Auxin efflux carrier moves auxin, and its gene is auxin-induced - so rate of transport from a cell depends on the auxin level in the cell
- 2) Local high auxin concentration causes new primordia, and it gets high locally by transport and diffusion
- 3) Auxin efflux carrier is polarized in cells, and points toward neighboring cells with the highest auxin concentration

Equations

$$\frac{dP_i}{dt} = F_{creation}(a_i) - K_{Pd}P_i$$

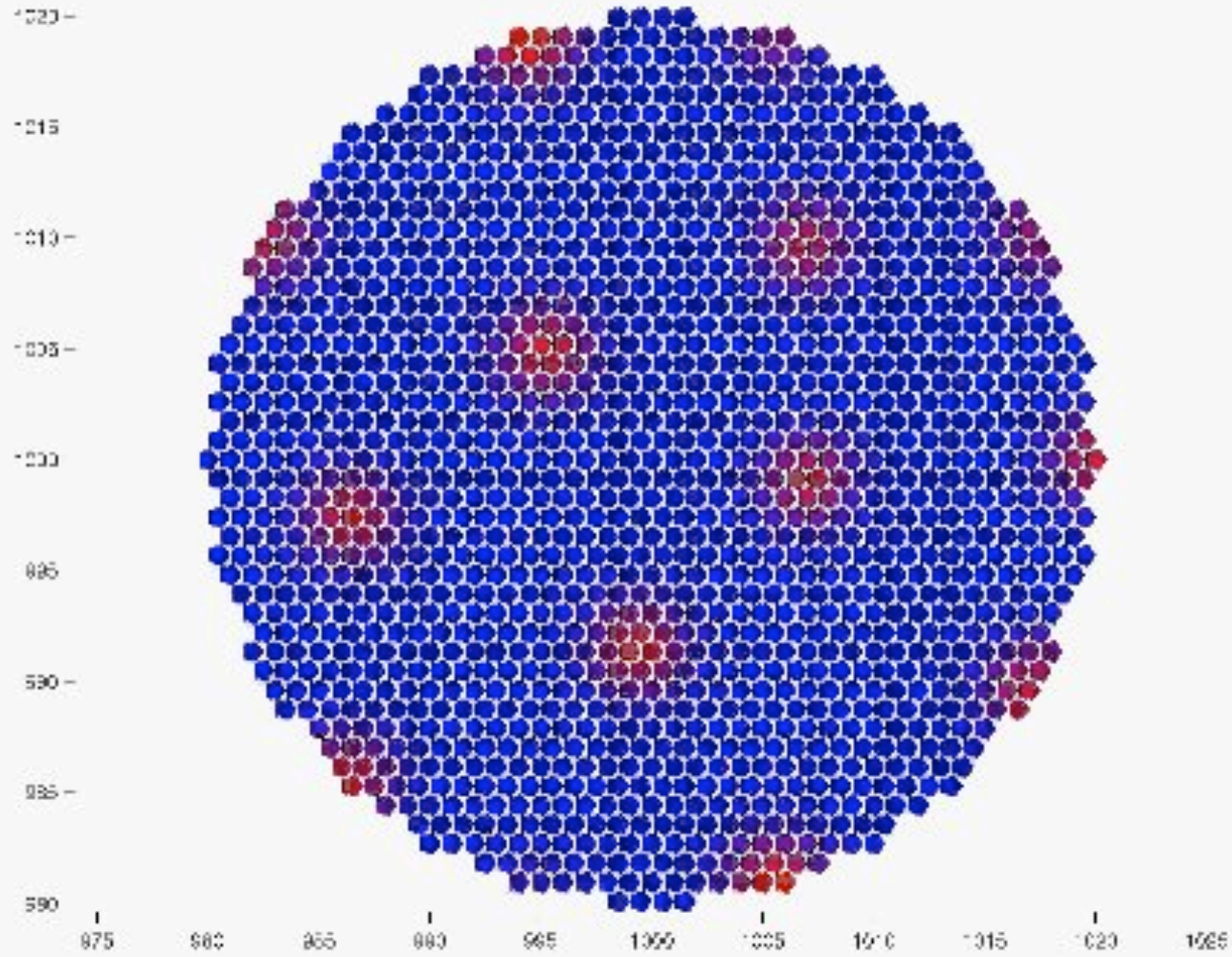
$$\frac{da_i}{dt} = K_p - K_d a_i - T \sum_k^{N_j} (a_i P_{ij} - a_j P_{ji}) + D \left(\sum_j^{N_i} a_j - N_i a_i \right)$$

$$P_{ij} = P_i \frac{a_j}{\sum_k^{N_i} a_k} \propto P_i a_j$$

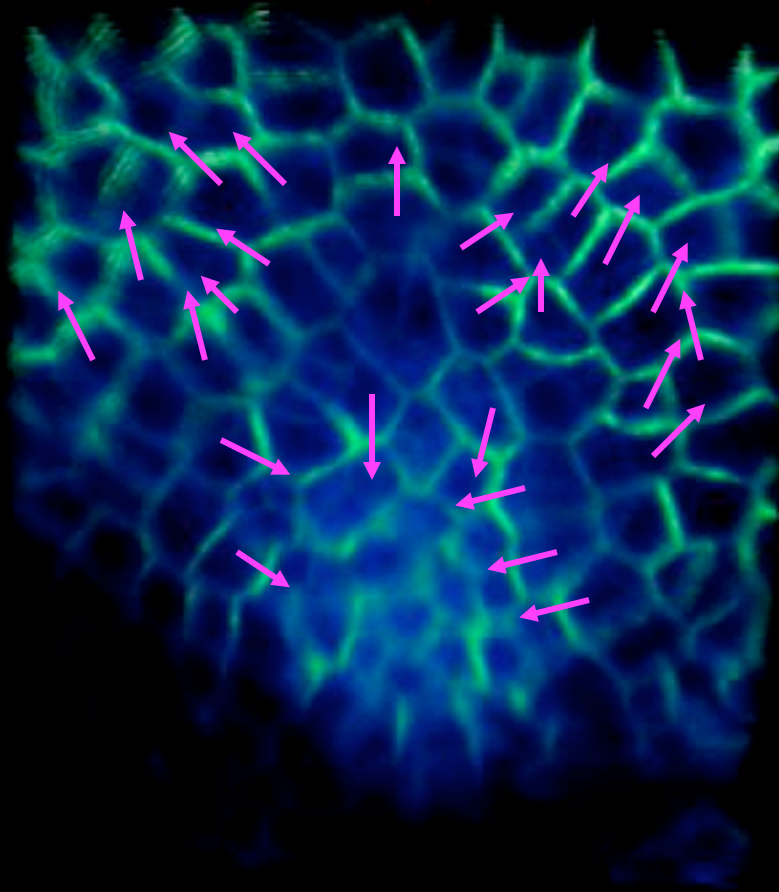
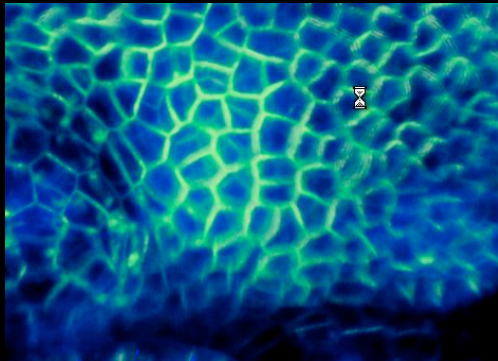
P_i, a_i – PIN1, auxin concentration in cell i

T – transport strength, D – diffusion strength

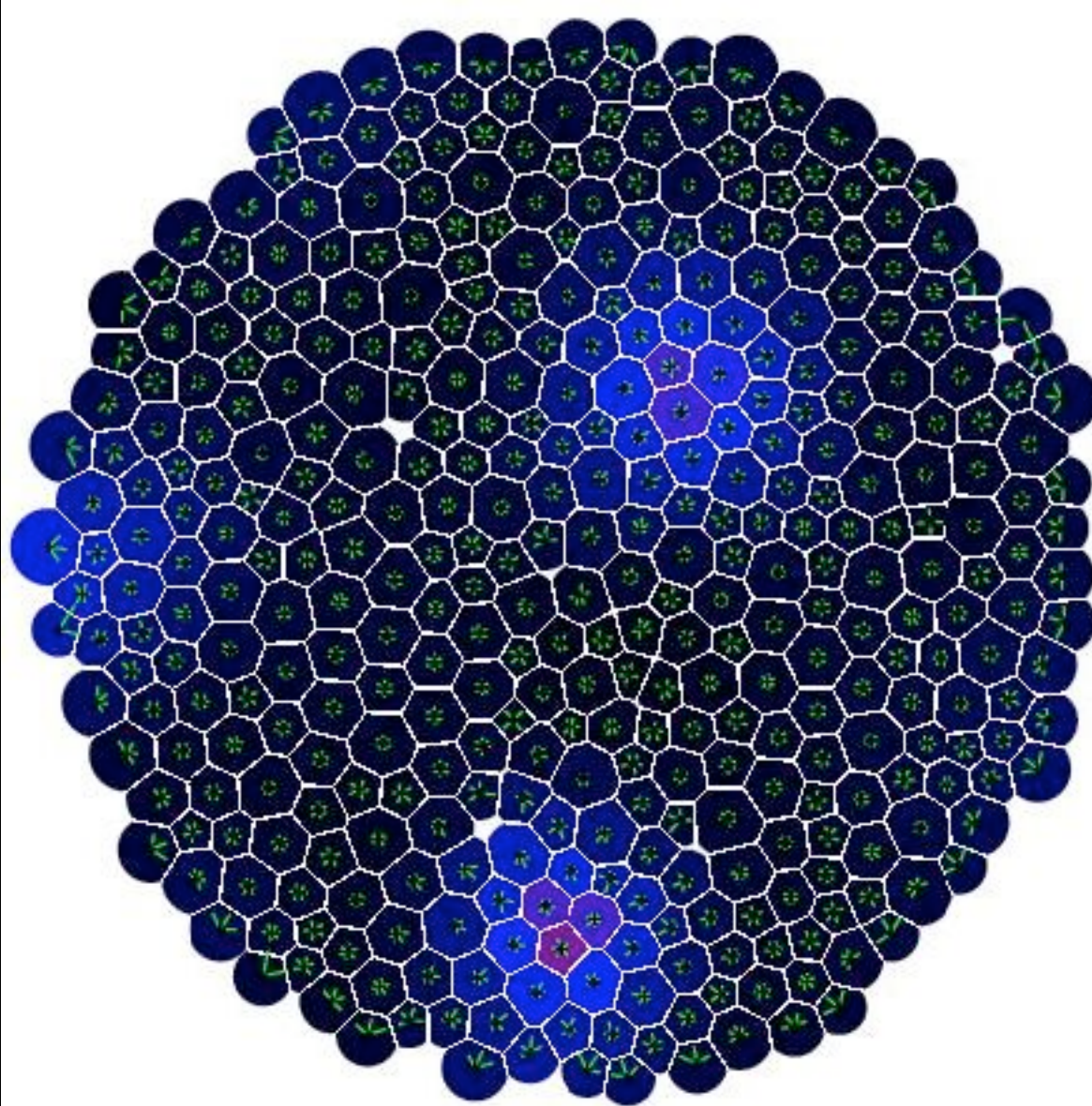
Peak formation including radial growth

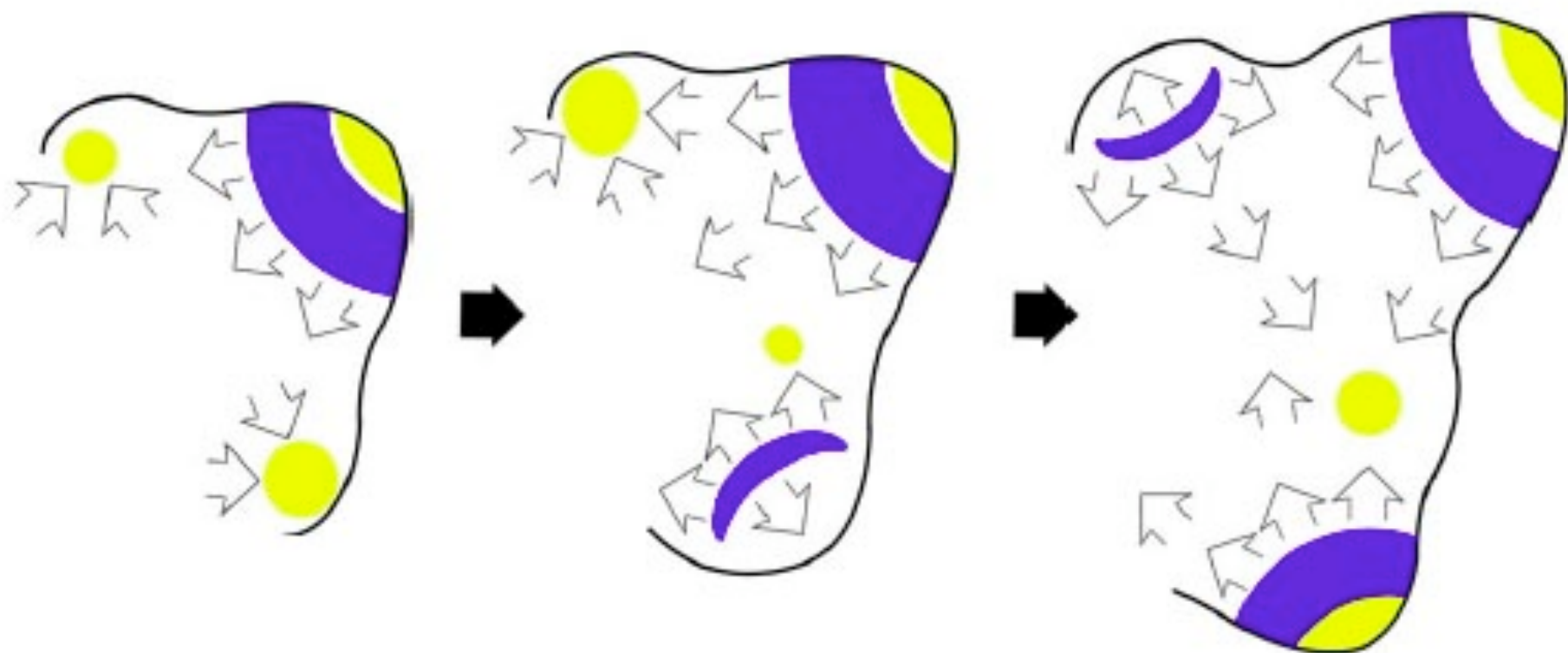


Polarity reversal is abrupt and has a sharp boundary



The auxin concentration model may account for PIN1 reversal



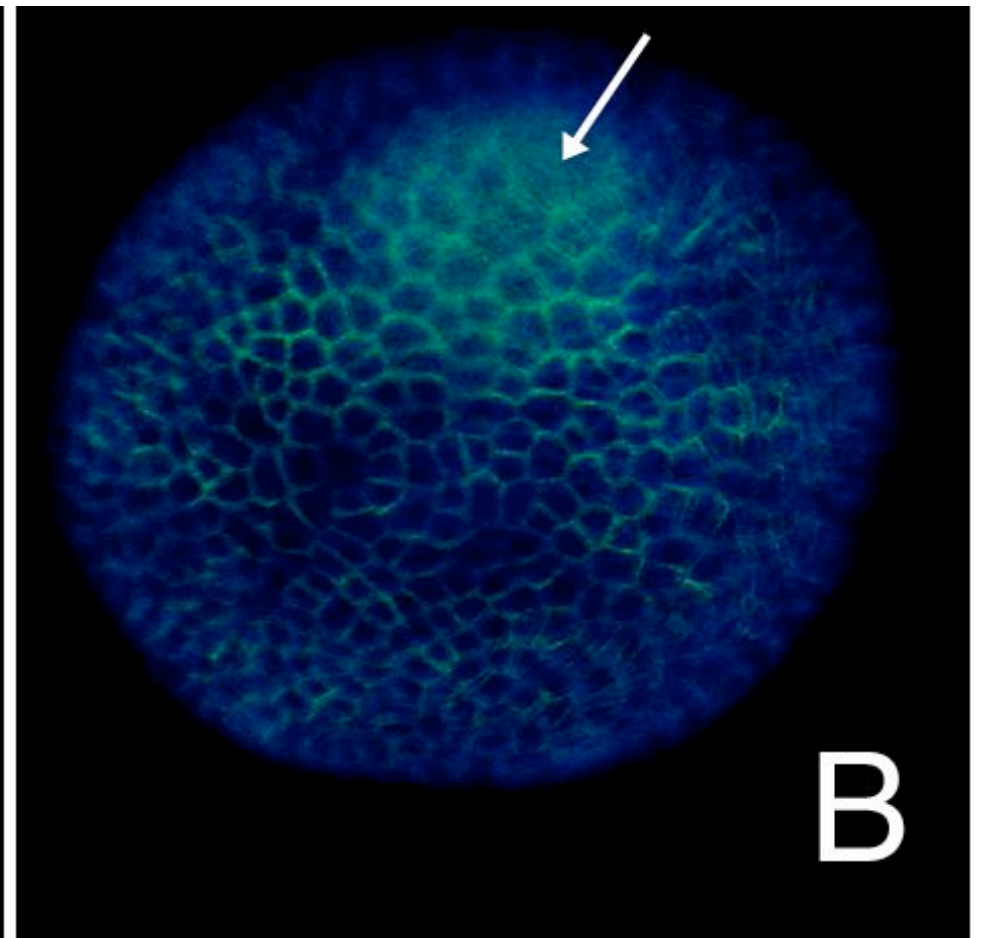
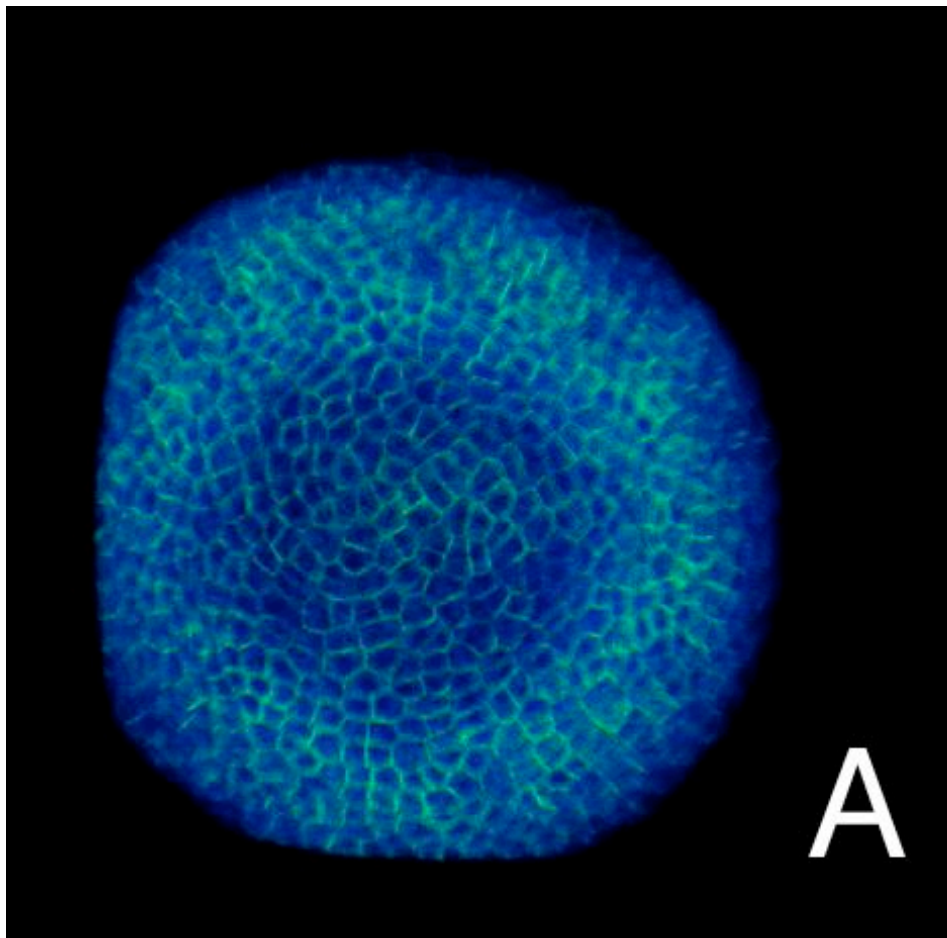


Conclusions

- The distribution of auxin is involved in patterning PIN1 expression and can influence PIN1 polarity.
- PIN1 polarity also responds to signals generated by neighboring cells.
- These observations support a proposed model for phyllotaxis based on feedback between PIN1 polarity and auxin levels within neighboring cells.
- THIS IS A NEW CLASS OF DEVELOPMENTAL MODEL - NOT REACTION-DIFFUSION, NOT MUTUAL INHIBITION, BUT REGULATED TRANSPORT OF A MORPHOGEN

pinoid apex, pPIN1::PIN1-GFP

IAA spot added at arrow

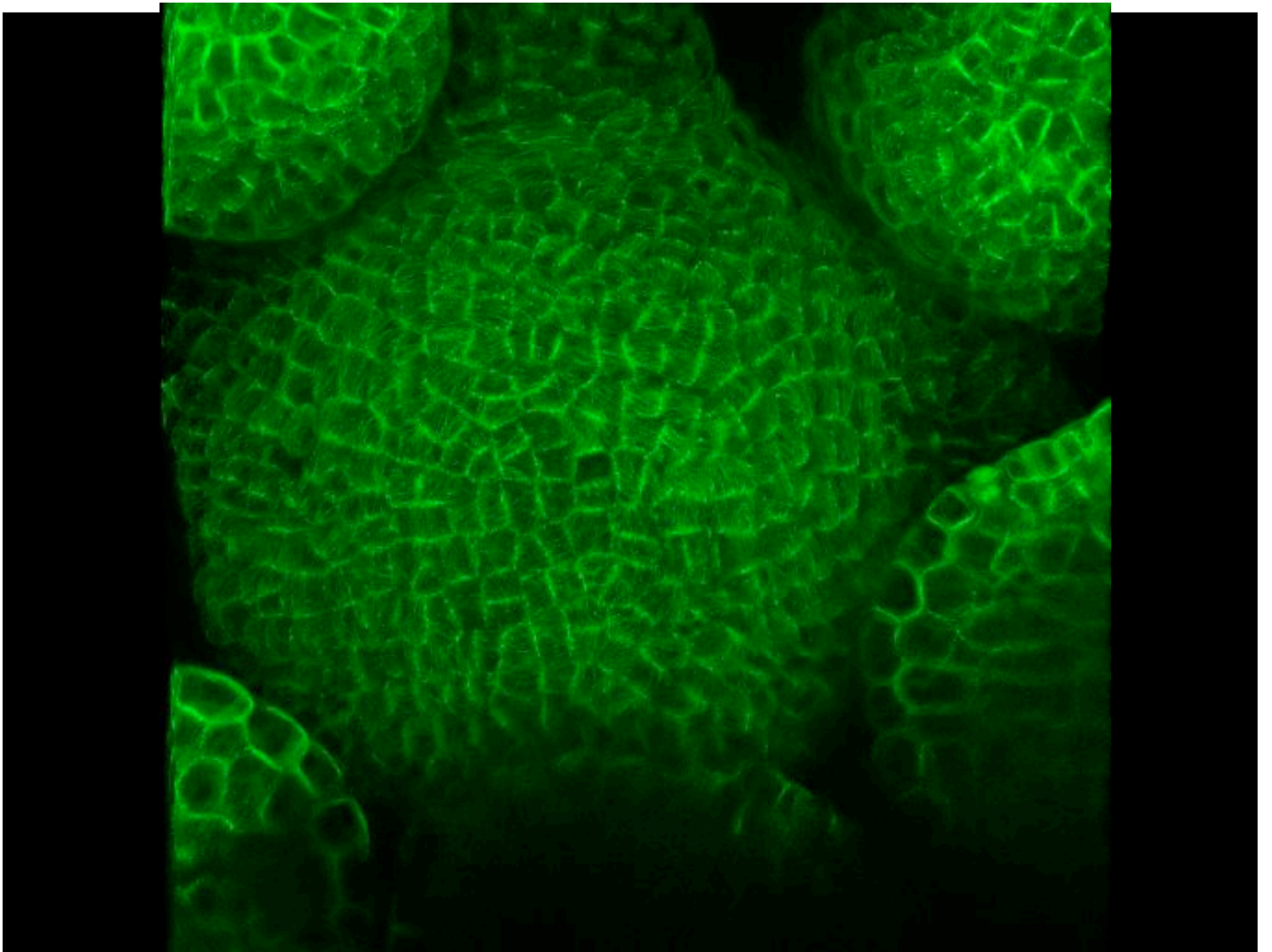


Question

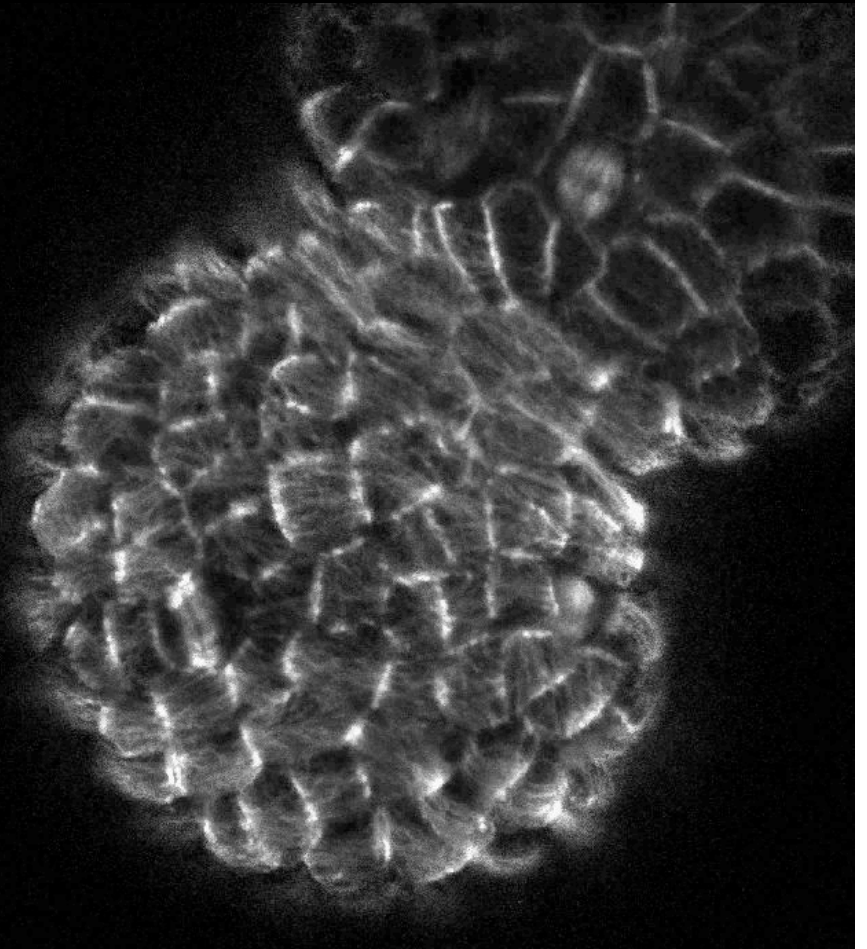
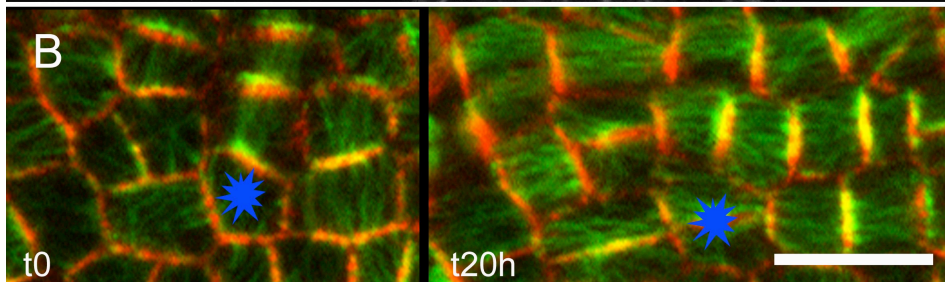
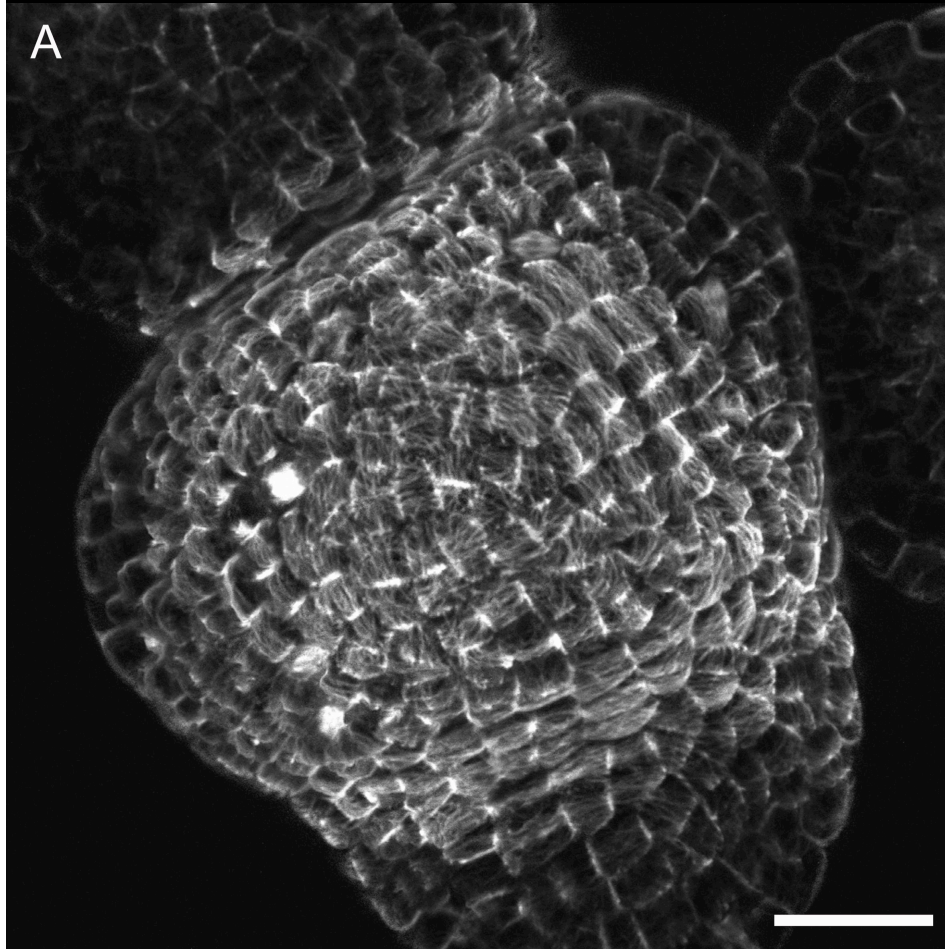
How does a cell *directionally* detect the auxin concentration of its neighbors?

- Auxin itself as a signal?
- Auxin induces a different diffusible substance?
- Auxin induces an ephrin-type cell-to-cell protein?
- Auxin causes cell expansion, physical force affects neighboring cells?: Could this explain expansin and PME experiments?

Can stress really be directionally sensed by meristem cells?



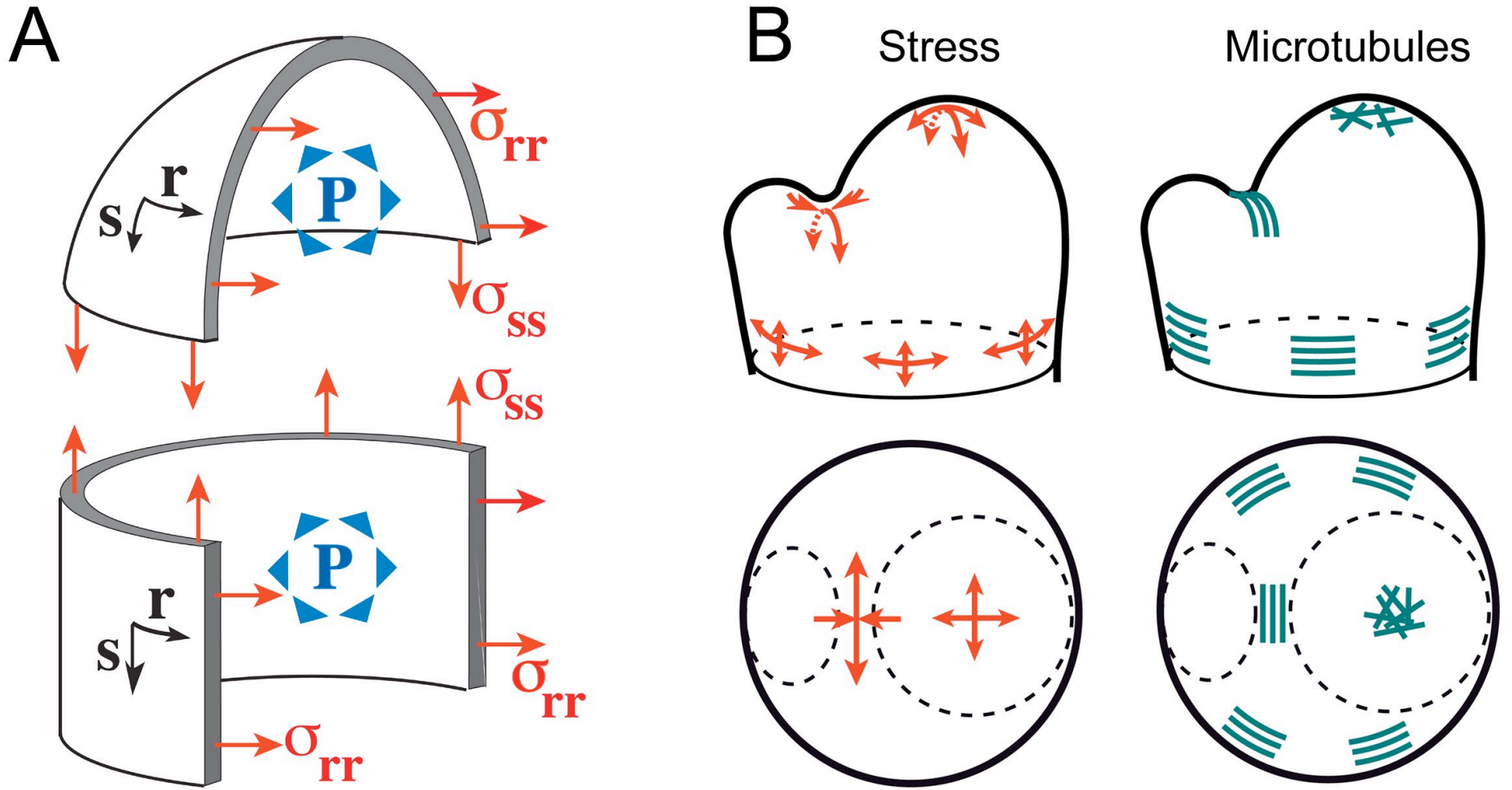
Microtubule patterns in the SAM



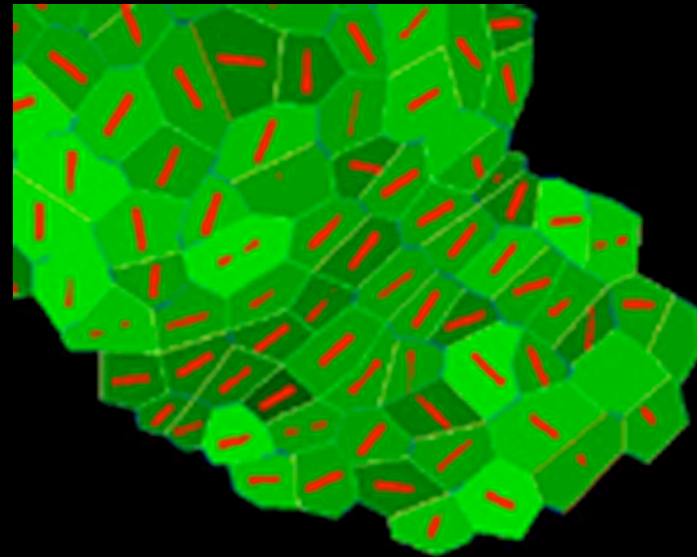
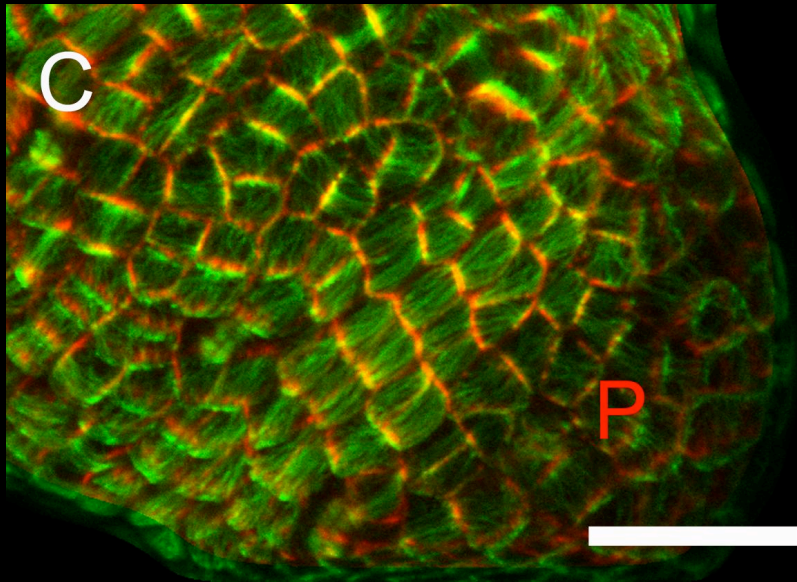
Movie S1

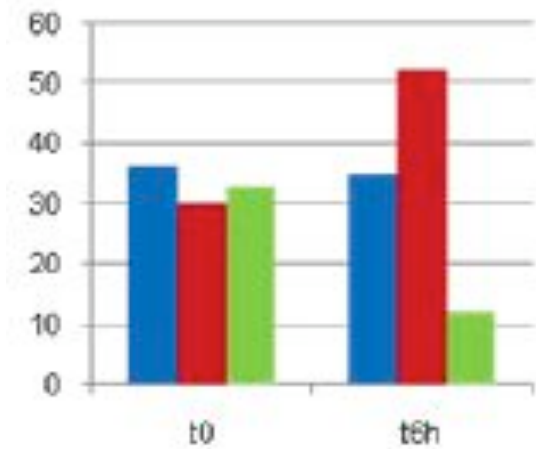
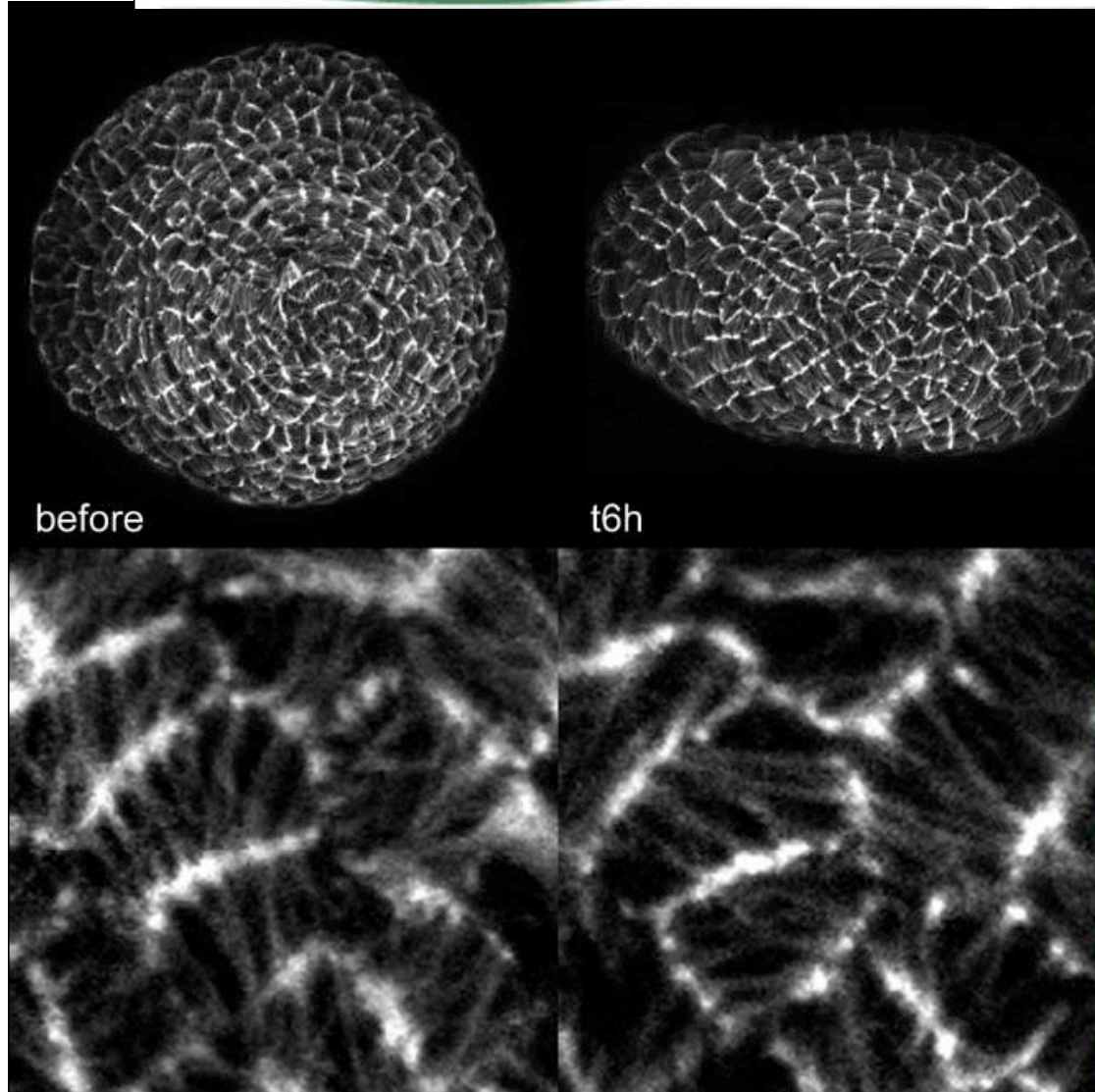
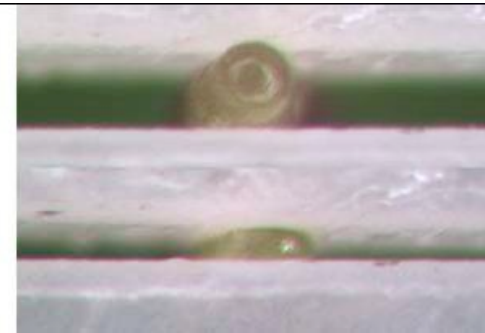
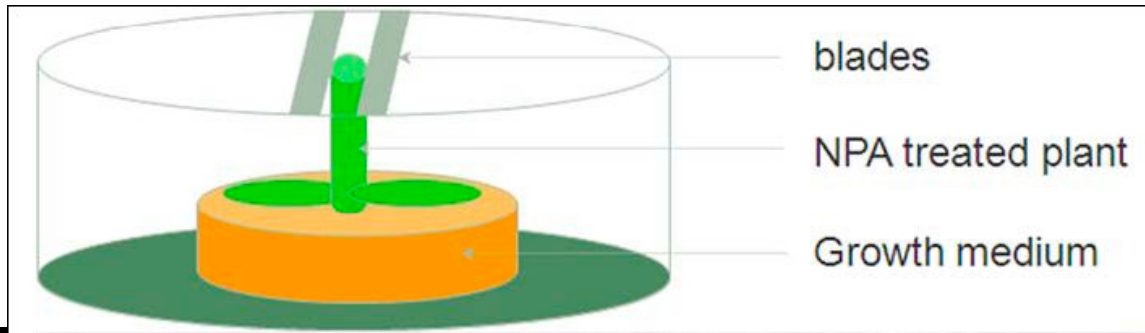
Fig.2

Stress Pattern in SAM



Model surface extracted from real template -
Calculated stress match real microtubules.

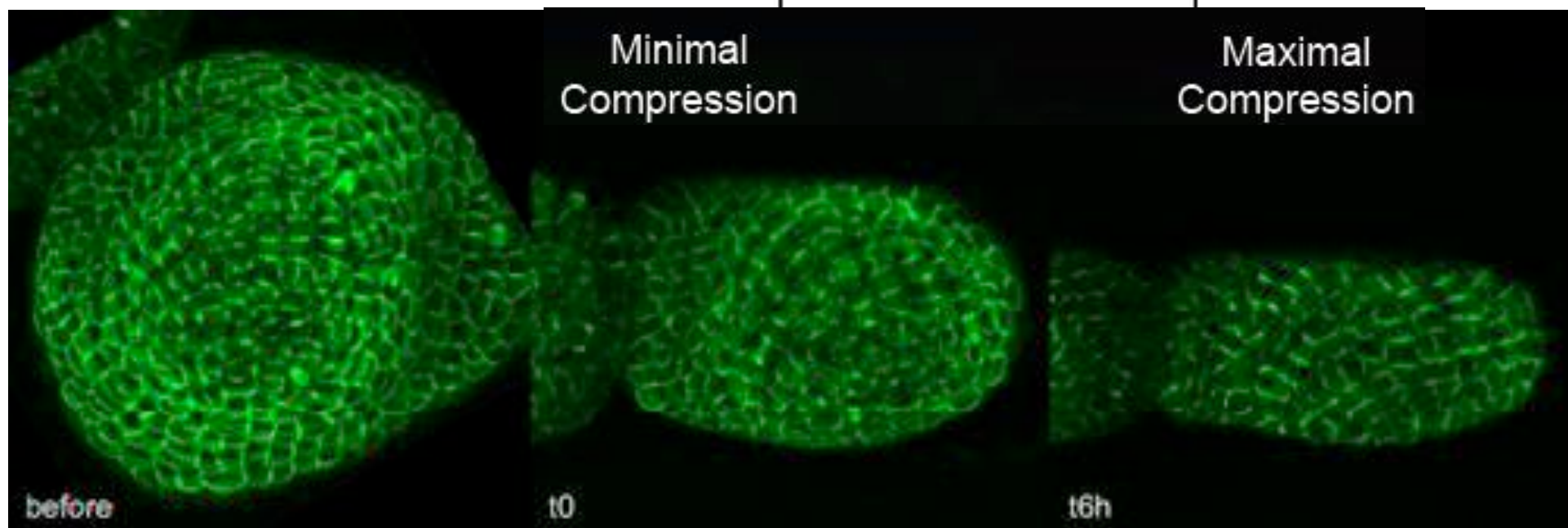
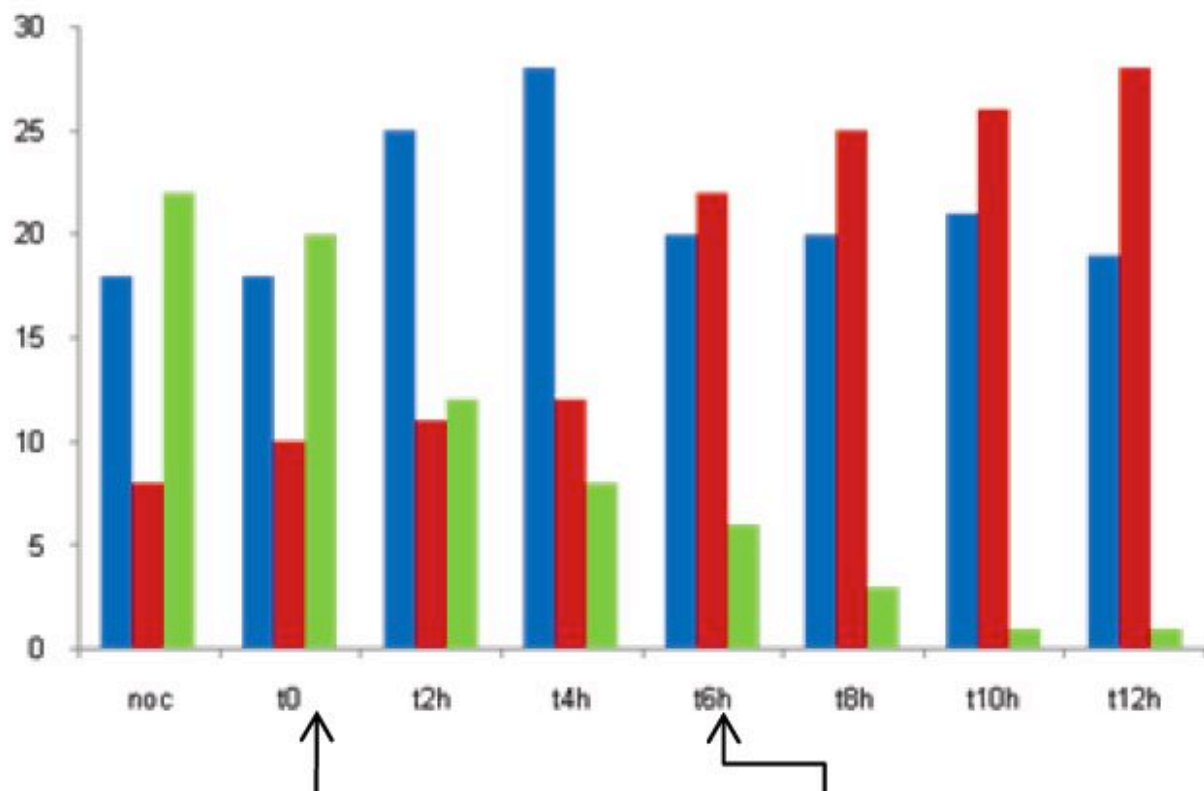
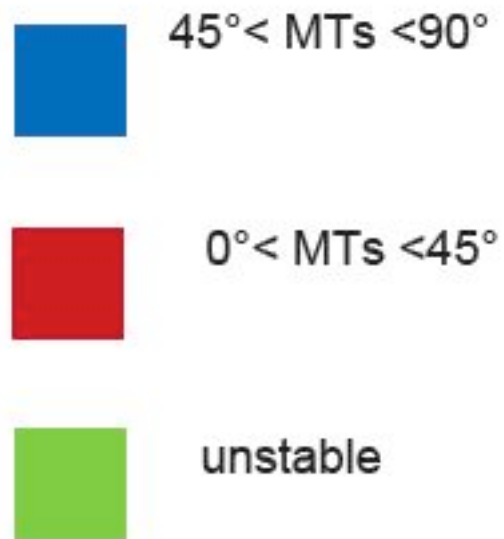




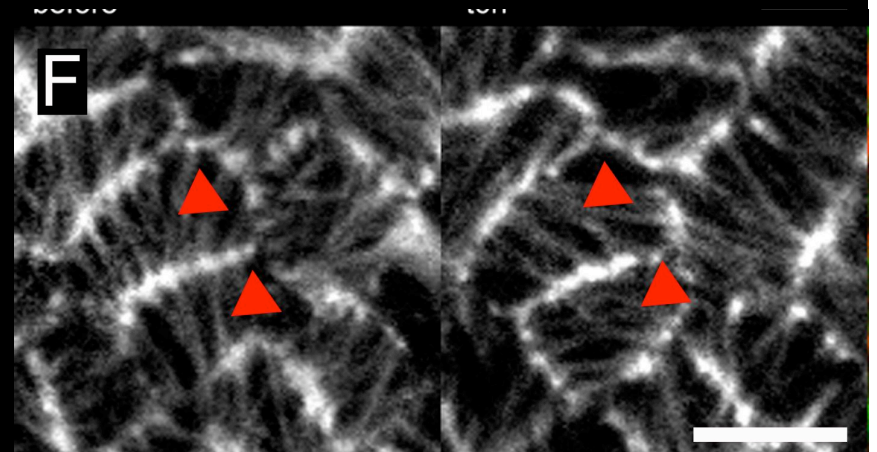
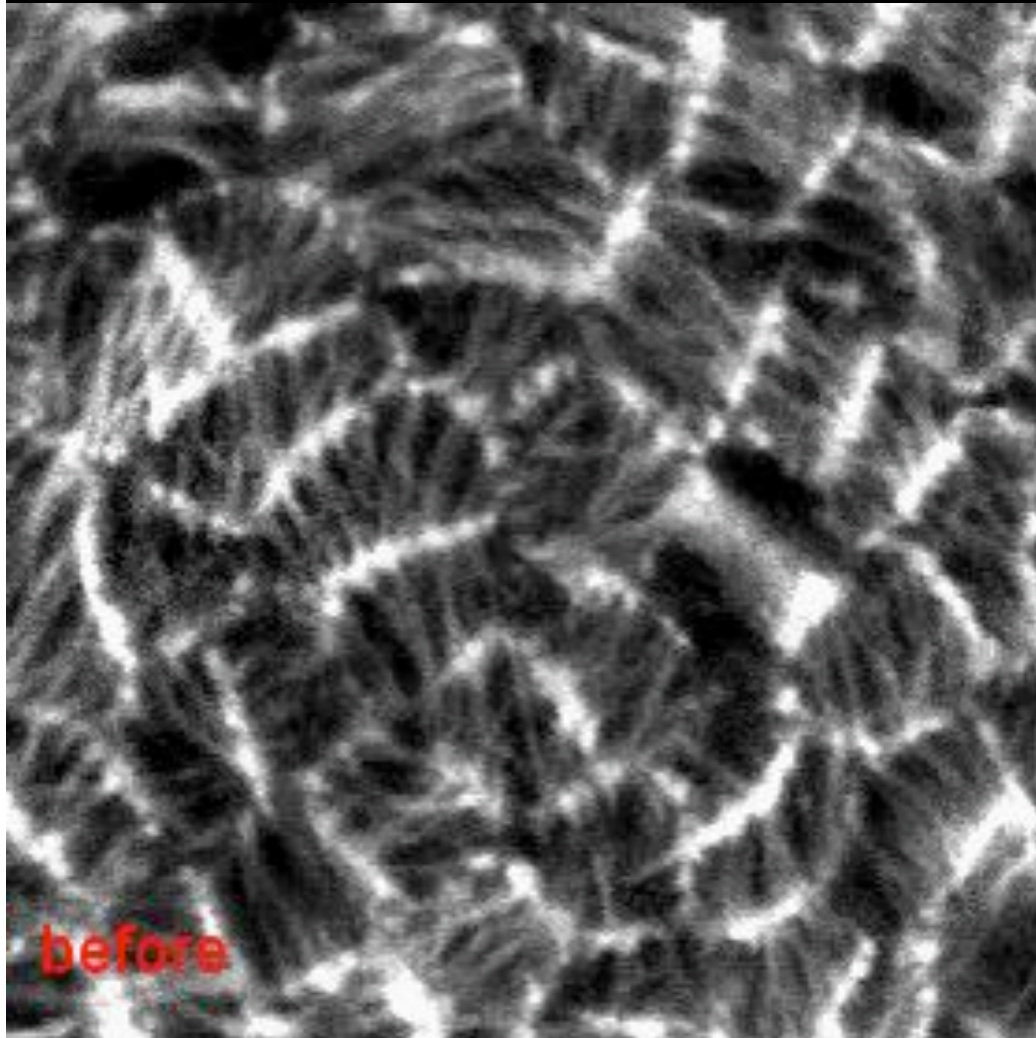
■ $45^\circ < \text{MTs} < 90^\circ$

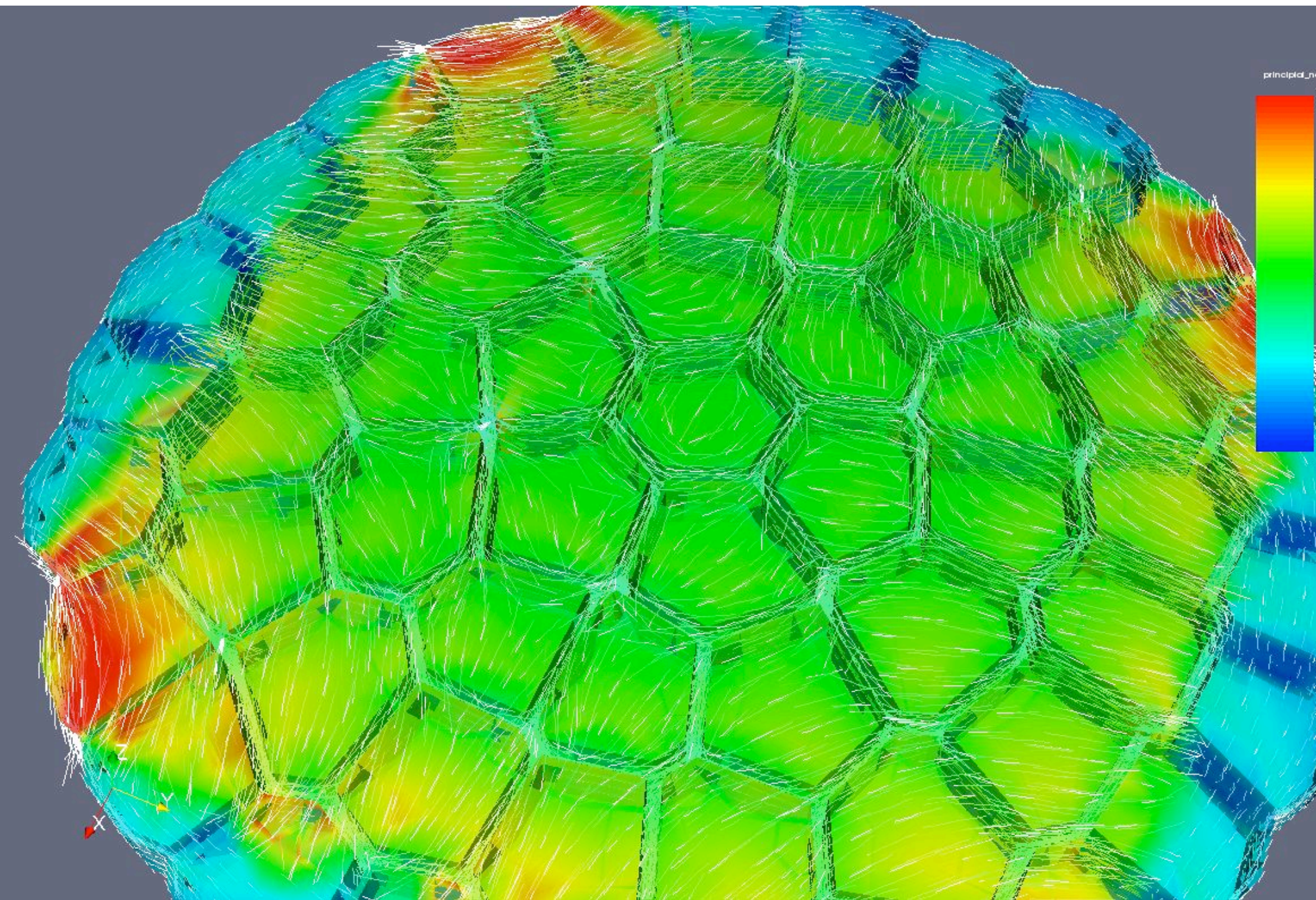
■ $0^\circ < \text{MTs} < 45^\circ$

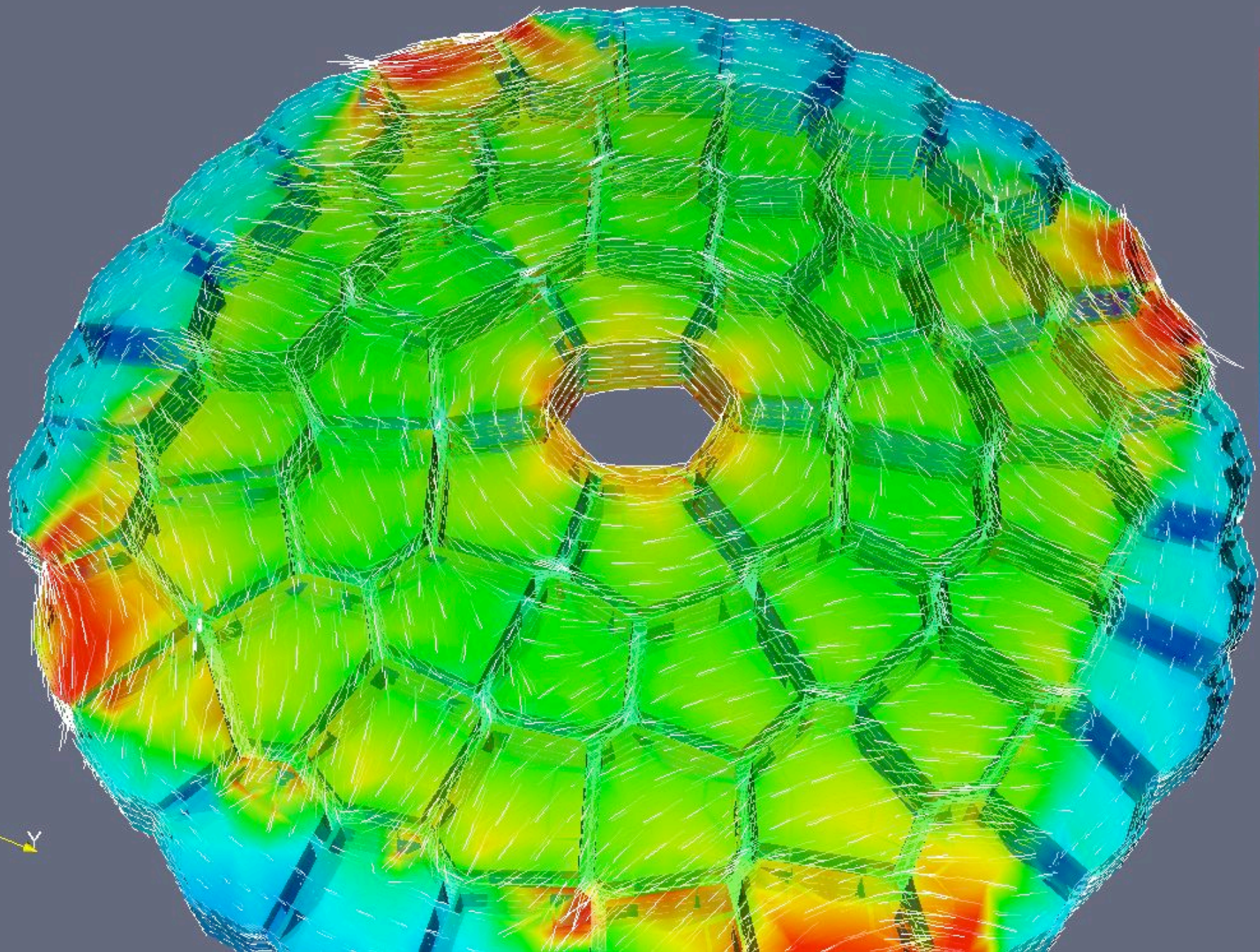
■ unstable

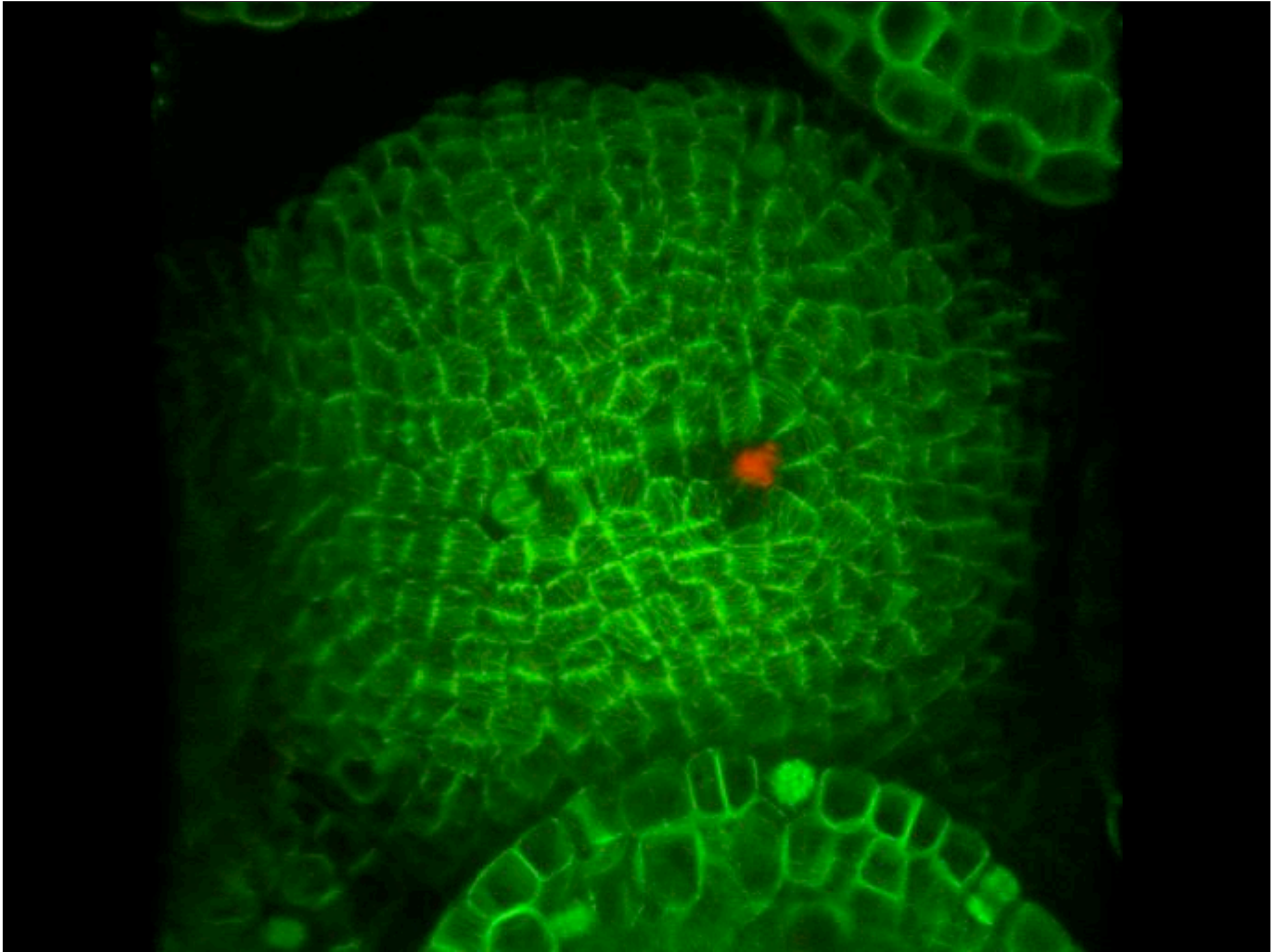


MT orientations 6 hrs following compression

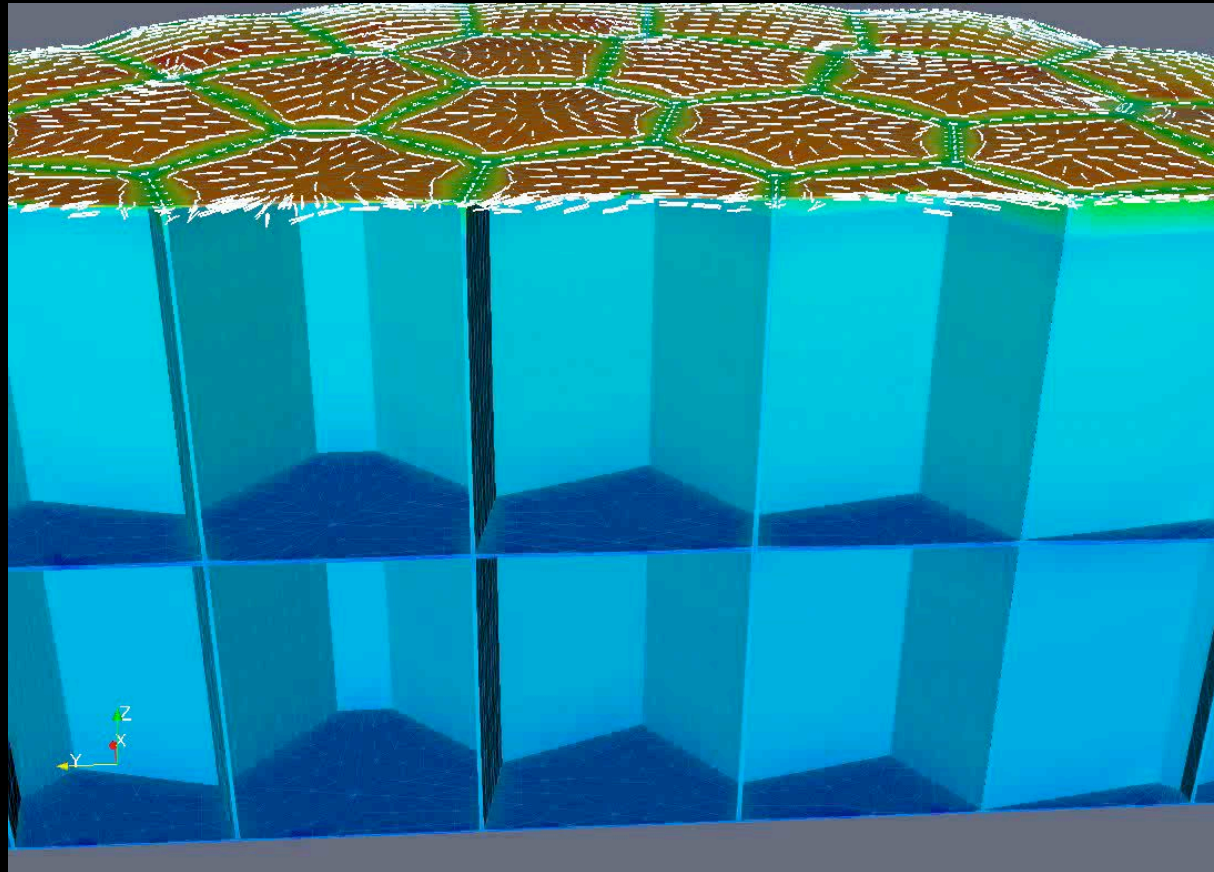
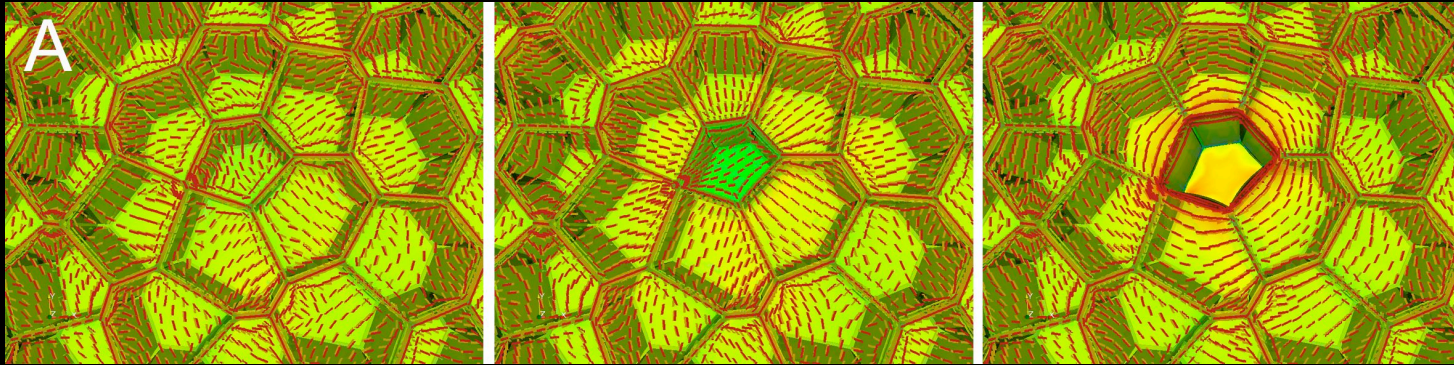




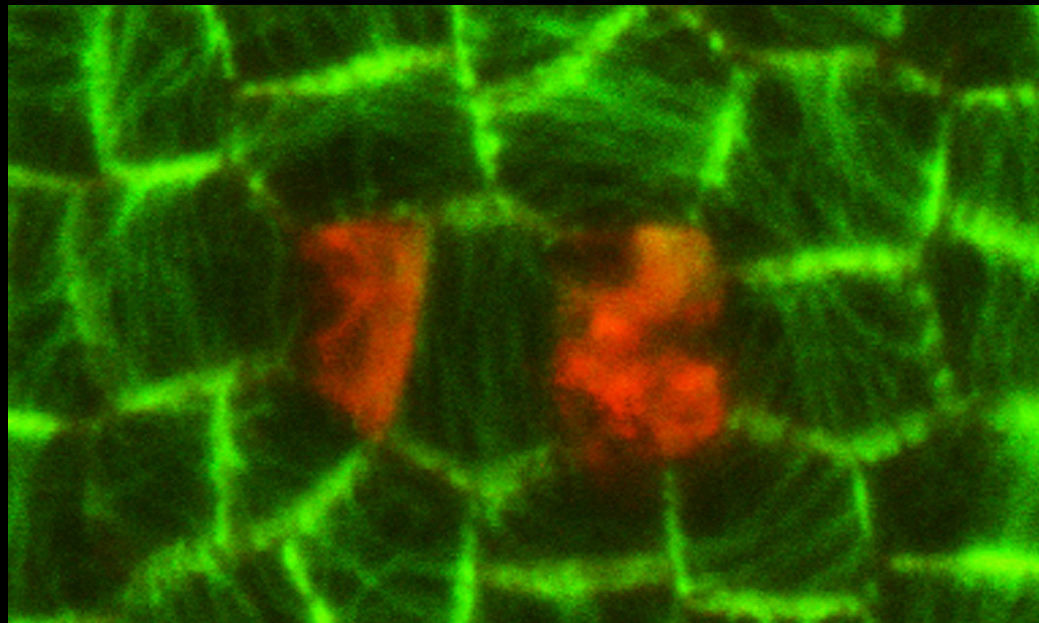
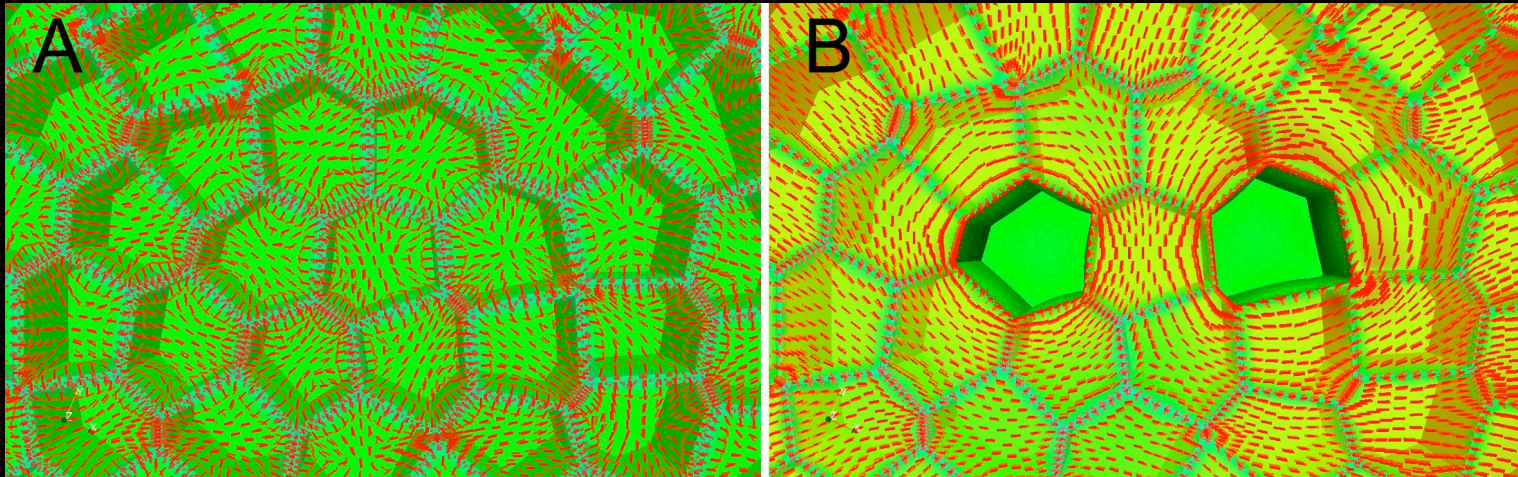




Simulation of cell ablation



Double cell ablations also match and argue against chemical morphogen model



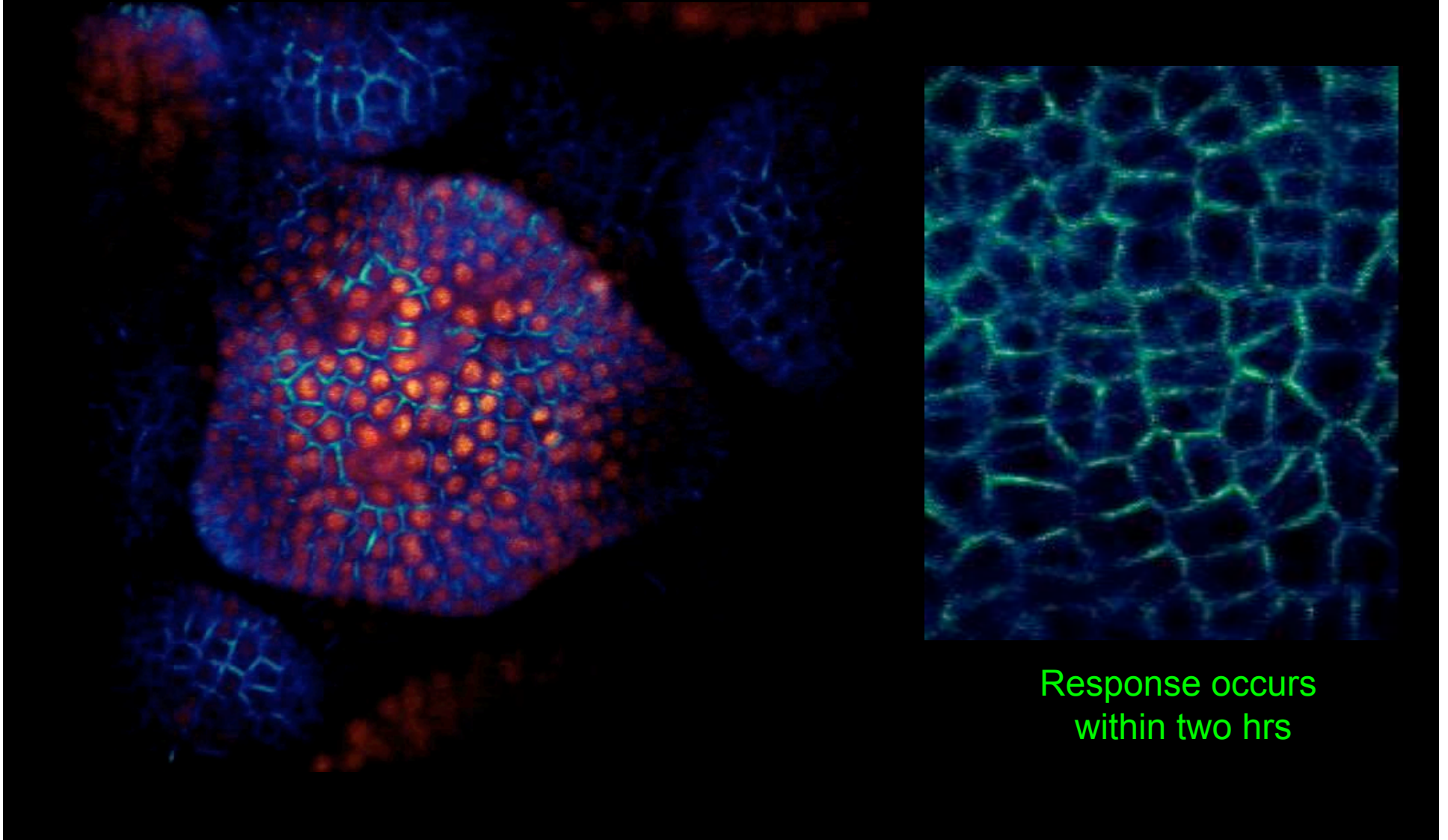
Question

How does a cell *directionally* detect the auxin concentration of its neighbors?

Why do cell wall relaxers have the same effect as auxin in inducing new primordia?

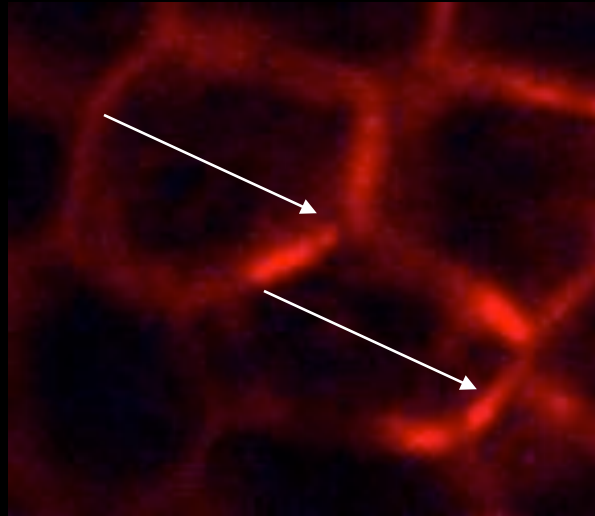
- Auxin causes cell expansion, PIN1 moves to the membrane adjacent to the most stressed wall?

Responses to single cell ablations indicate
local signaling can modulate PIN1 polarity

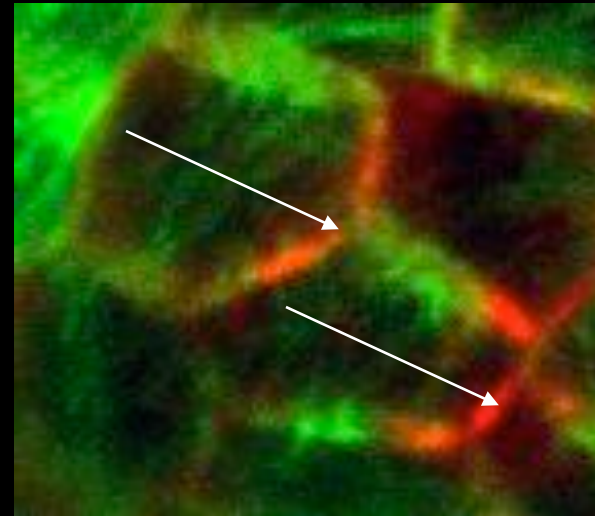


Response occurs
within two hrs

PIN1 and microtubule interphase are generally aligned



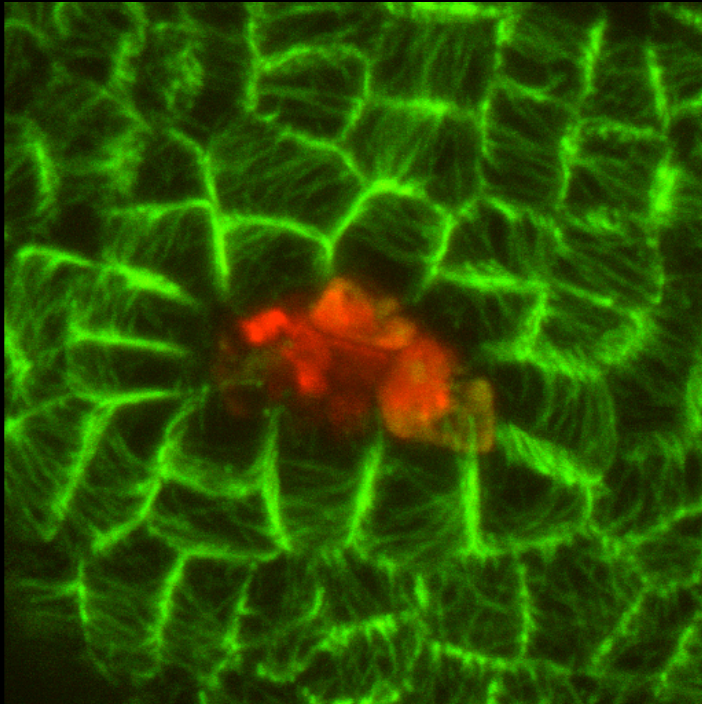
PIN1



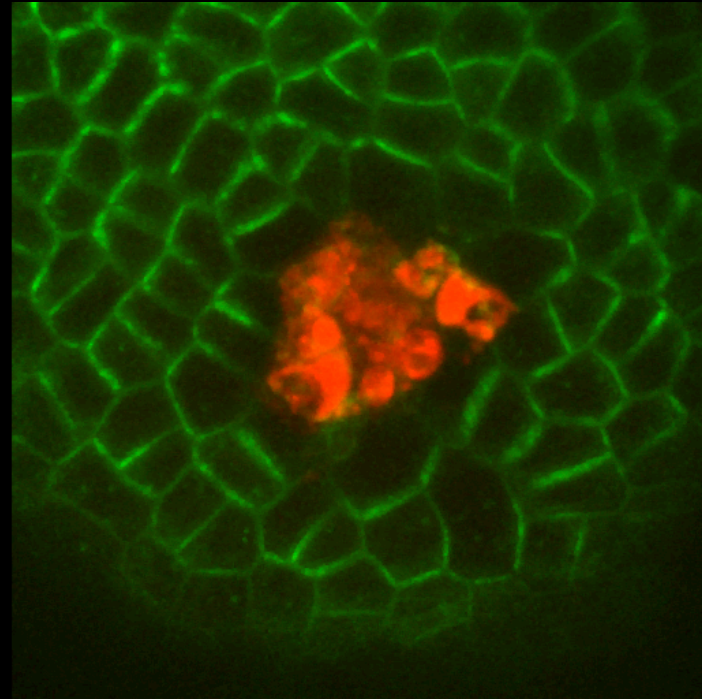
PIN1 MTs

Reorientation (at a distance) is NPA insensitive

MTs

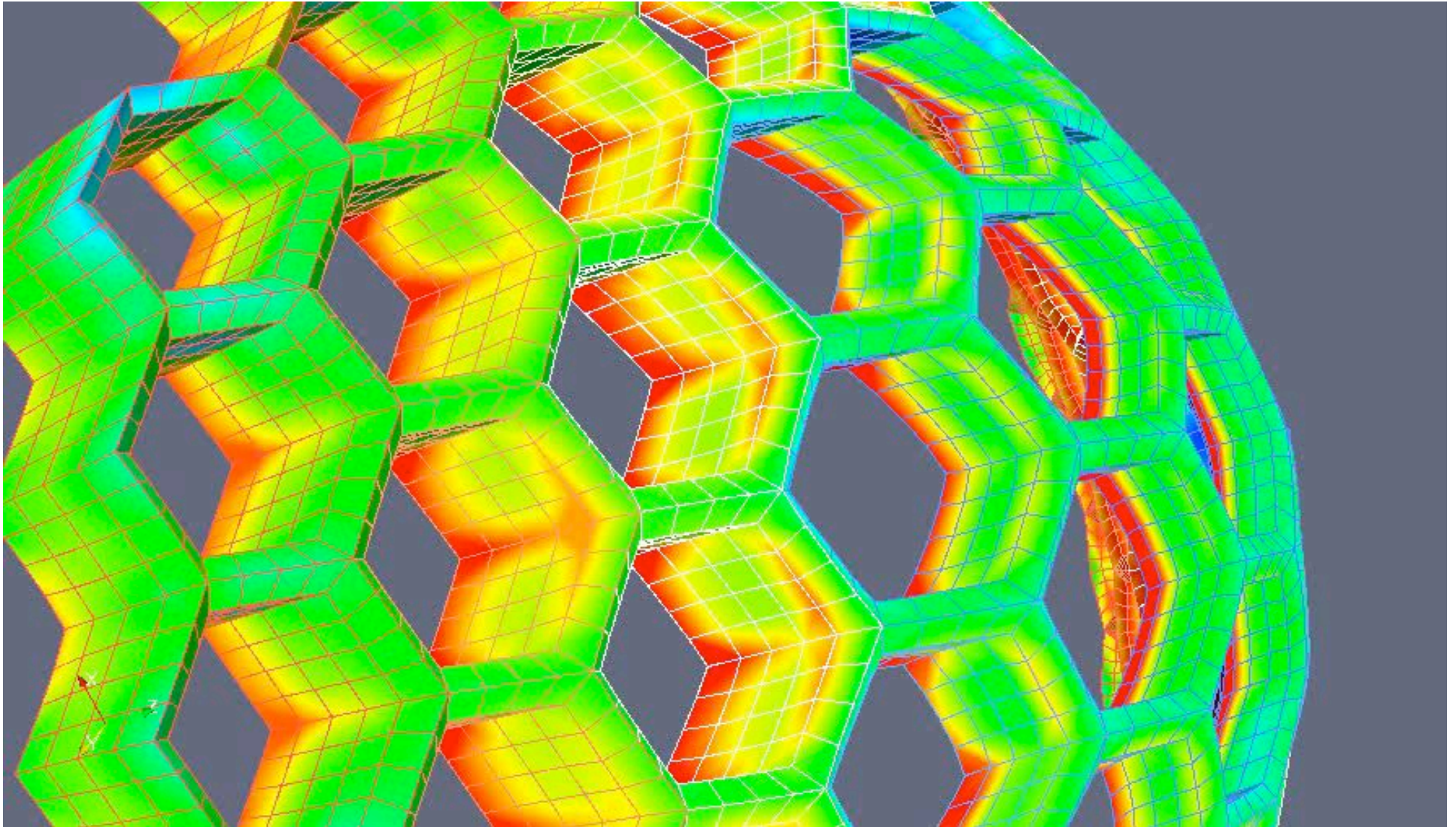


PIN1



Plants treated with 100uM NPA for 24 hrs

Most stressed wall/PIN1?



Conclusion

- Cell polarity in response to ablation is not easily perturbed by inhibition of auxin transport but fits patterns predicted by mechanical models
- Phyllotaxis is likely to involve a mechanical-chemical coupling

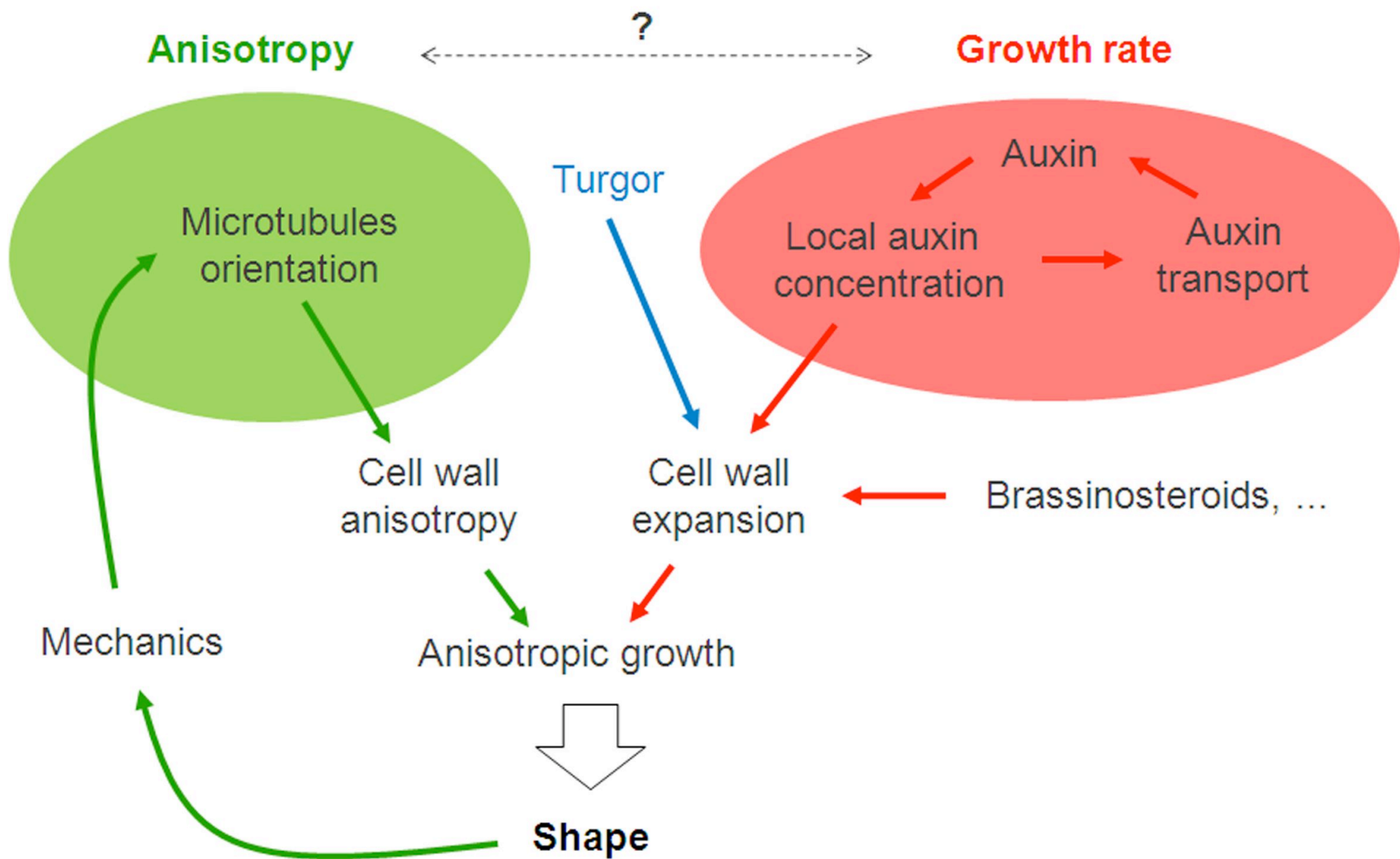


Fig. S1

Needs to understand tissues, morphogenesis and development:

Image Analysis

Computational Models of Chemical
Signals

Physical Models

Connection between Physical and
Chemical Models and Substrate

Connection between Chemical and
Physical Models

<http://www.computableplant.org>



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