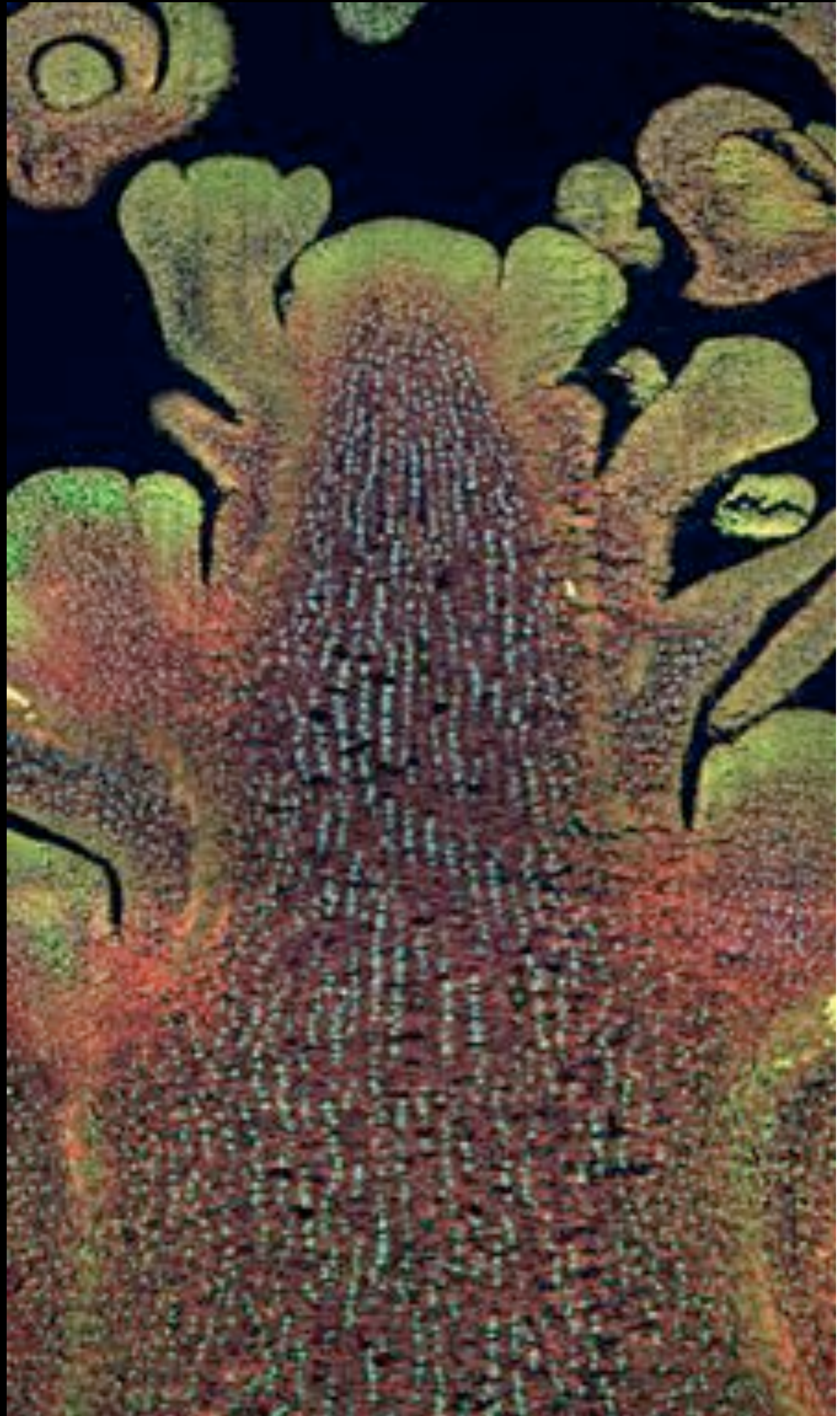
2018 CDB Part IB
Plant Development

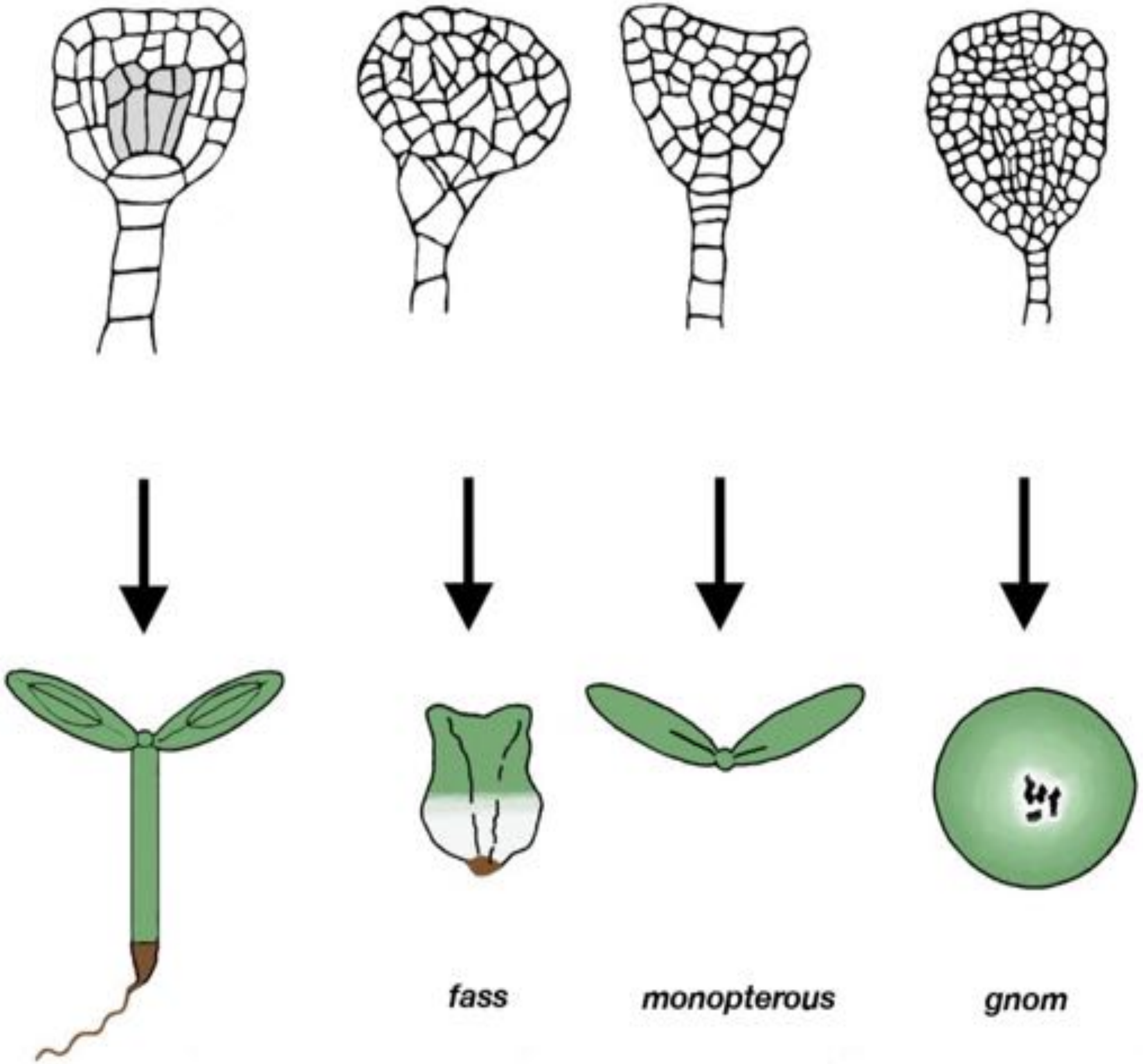
Lecture 2.

Polarity and auxin flow

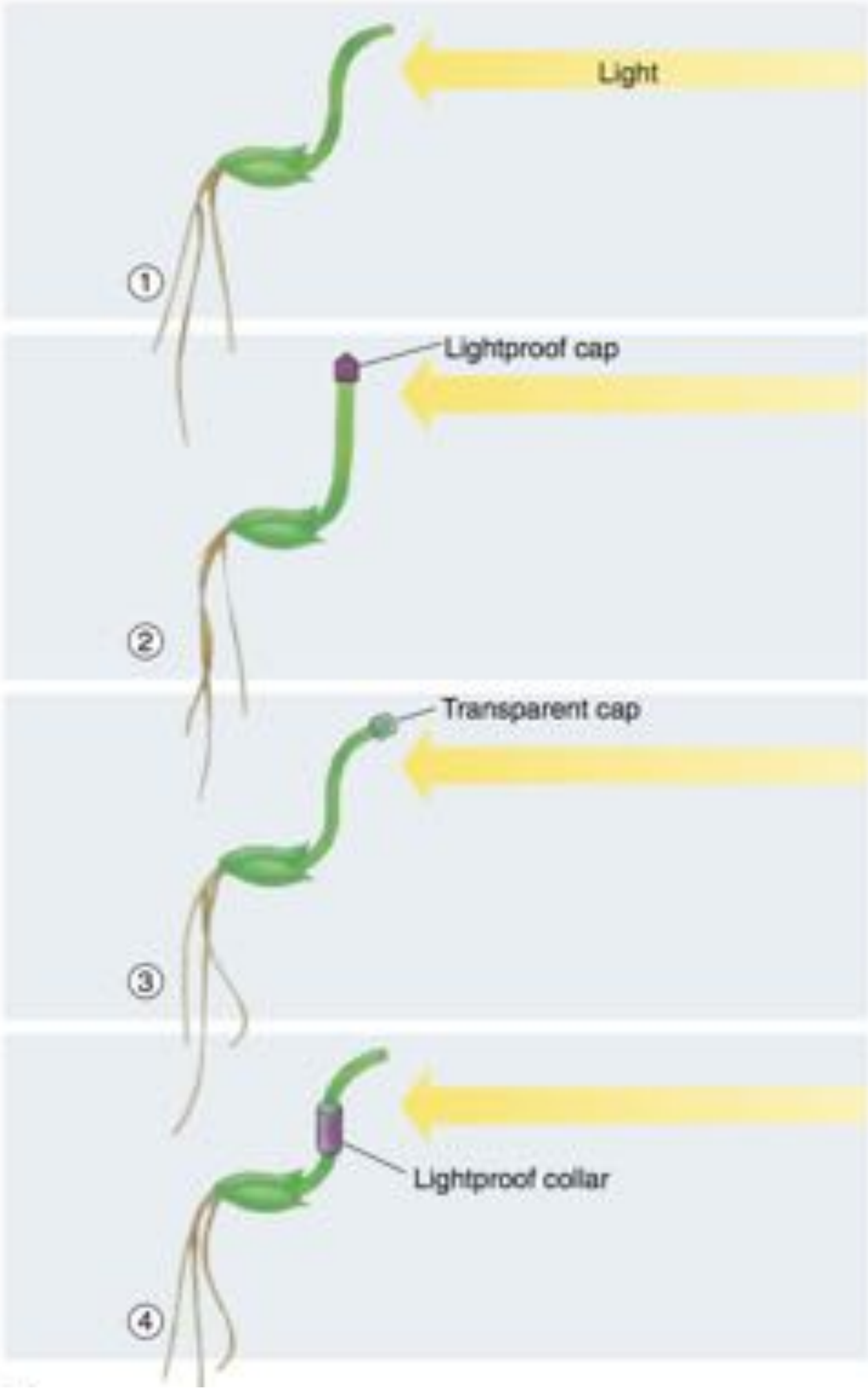


Jim Haseloff
Department of Plant Sciences

Mutations that affect auxin traffic or perception give rise to plants with altered body plans



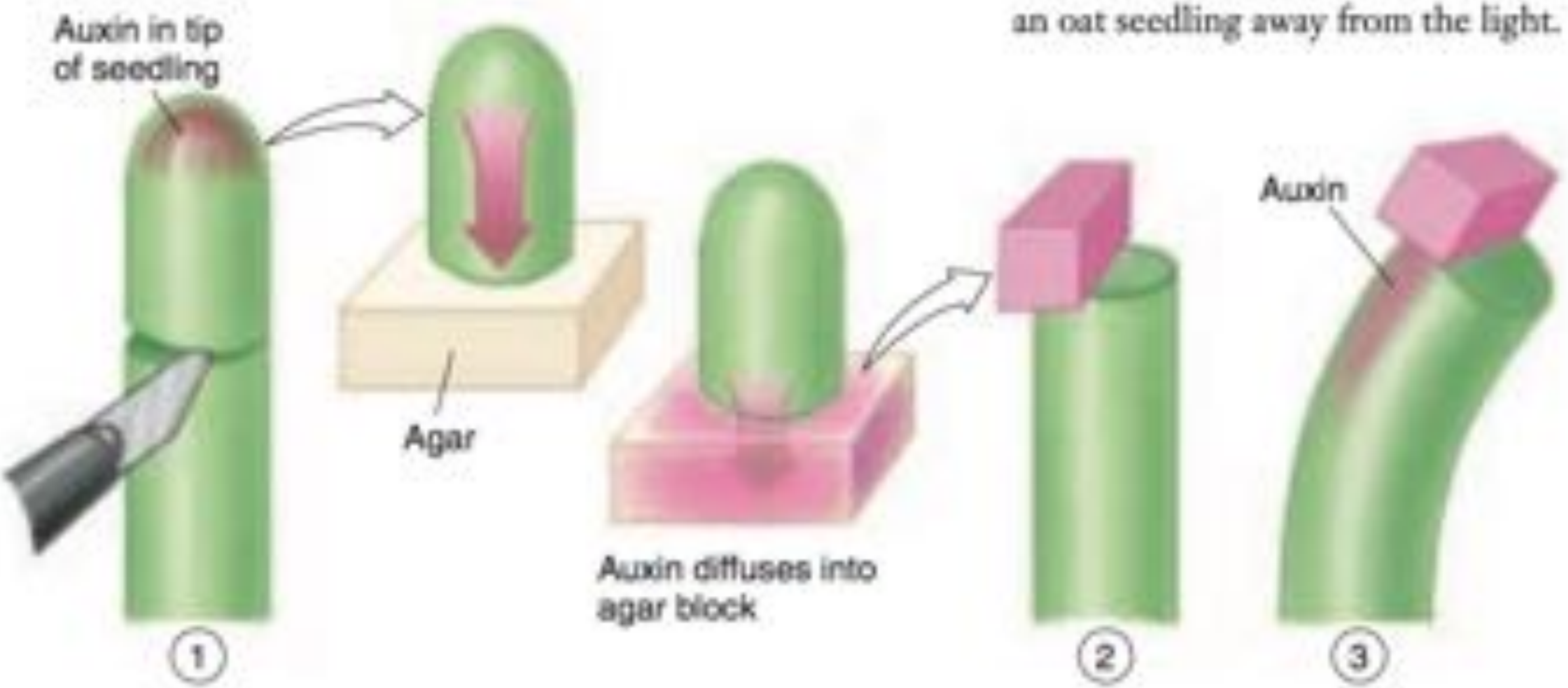
Charles and Francis Darwin's experiments on signalling during plant phototropism

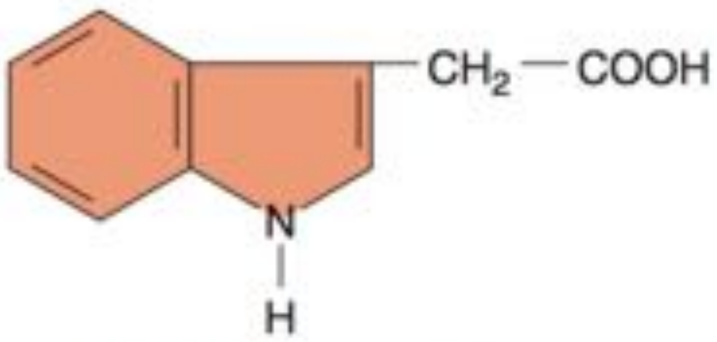


The Darwins' experiment. (*a*) Young grass seedlings normally bend toward the light. (*b*) The bending (*1*) did not occur when the tip of a seedling was covered with a lightproof cap (*2*), but did occur when it was covered with a transparent one (*3*). When a collar was placed below the tip (*4*), the characteristic light response took place. From these experiments, the Darwins concluded that, in response to light, an "influence" that caused bending was transmitted from the tip of the seedling to the area below, where bending normally occurs.

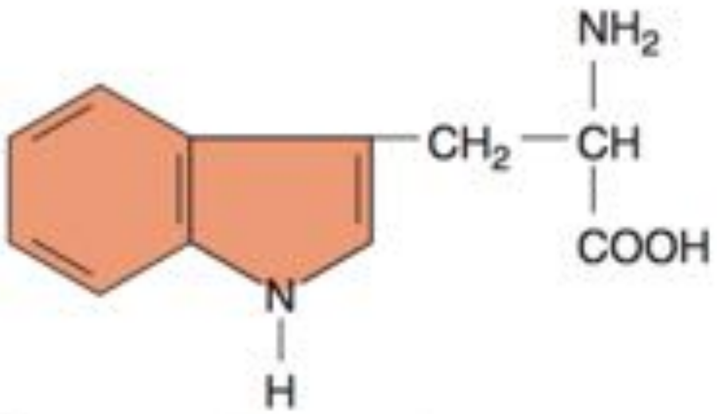
Demonstration of chemical signaling

Frits Went's experiment. (*1*) Went removed the tips of oat seedlings and put them in agar, an inert, gelatinous substance. (*2*) Blocks of agar were then placed off-center on the ends of other oat seedlings from which the tips had been removed. (*3*) The seedlings bent away from the side on which the agar block was placed. Went concluded that the substance that he named *auxin* promoted the elongation of the cells and that it accumulated on the side of an oat seedling away from the light.

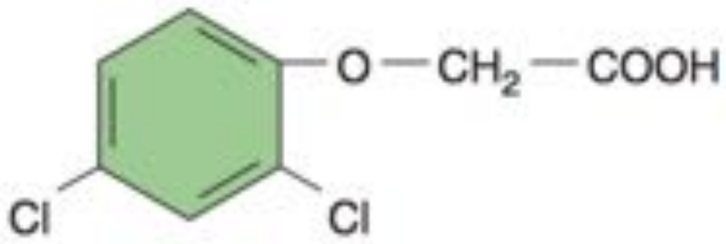




(a) IAA (Indoleacetic acid)



(b) Tryptophan



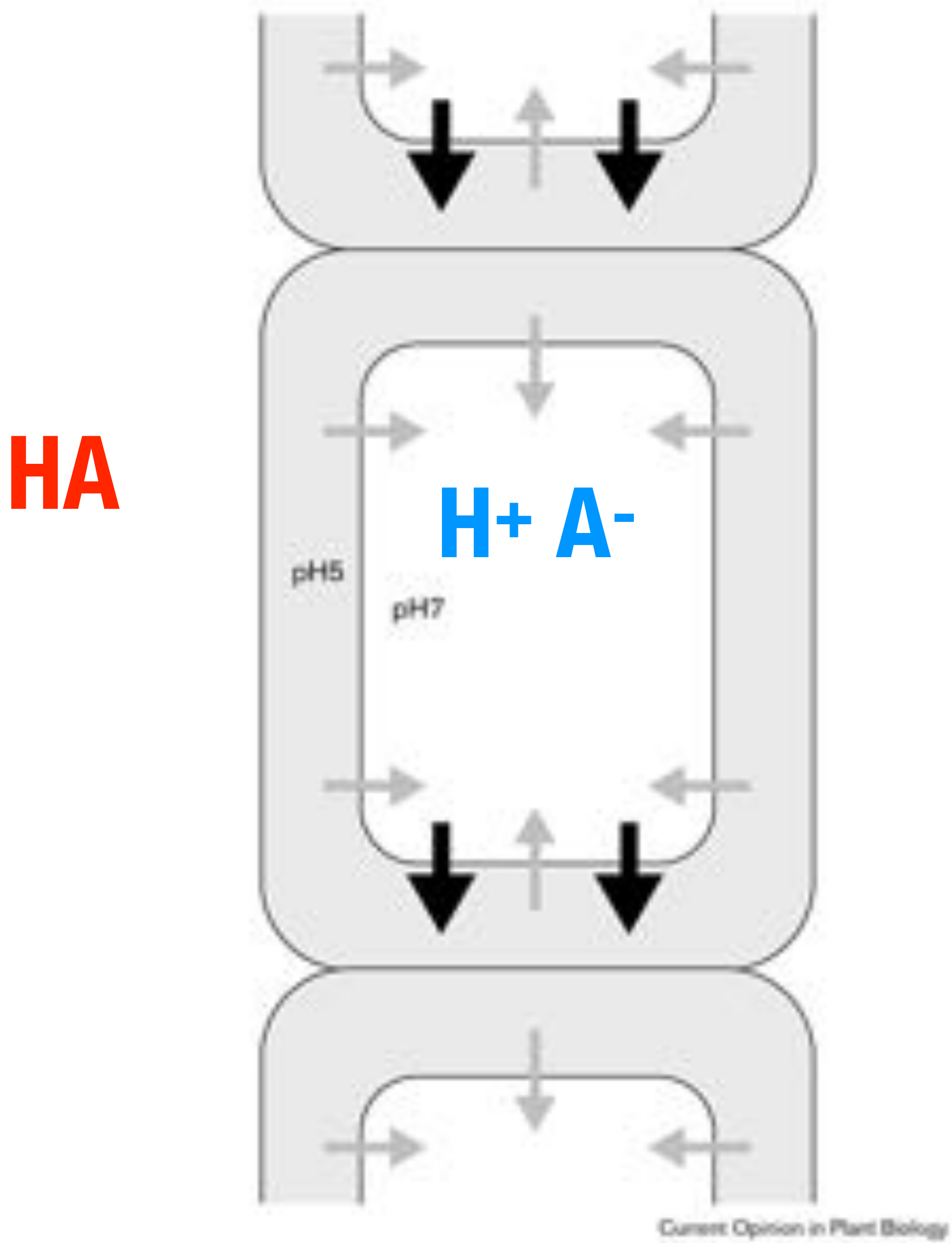
(c) Dichlorophenoxyacetic acid (2,4-D)

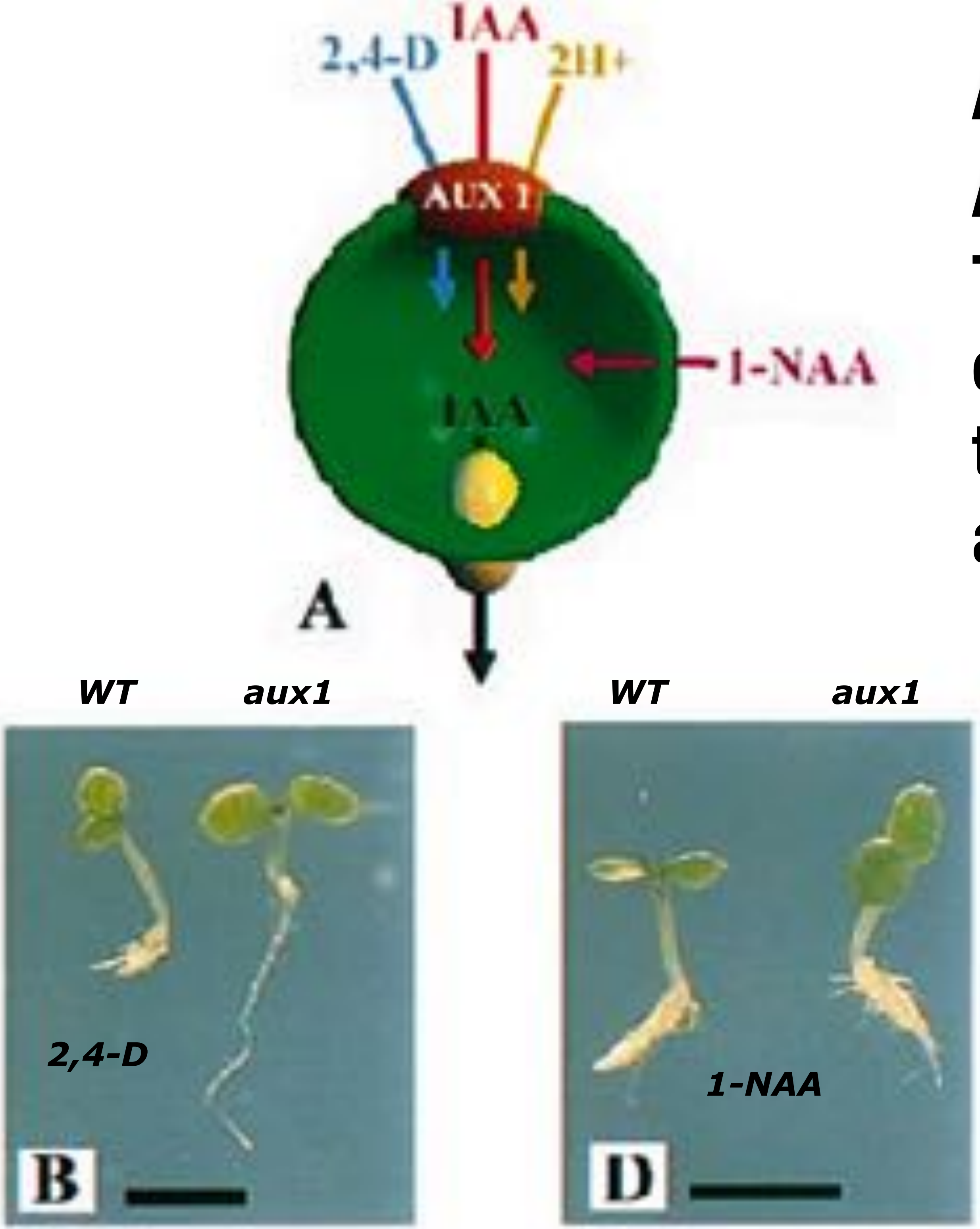
Auxin and apical-basal polarity:

Apical-basal polarity and the coordination of indeterminate growth and branching in plants is maintained by traffic of growth regulators.

These are not passive gradients, but are the product of active cellular transport.

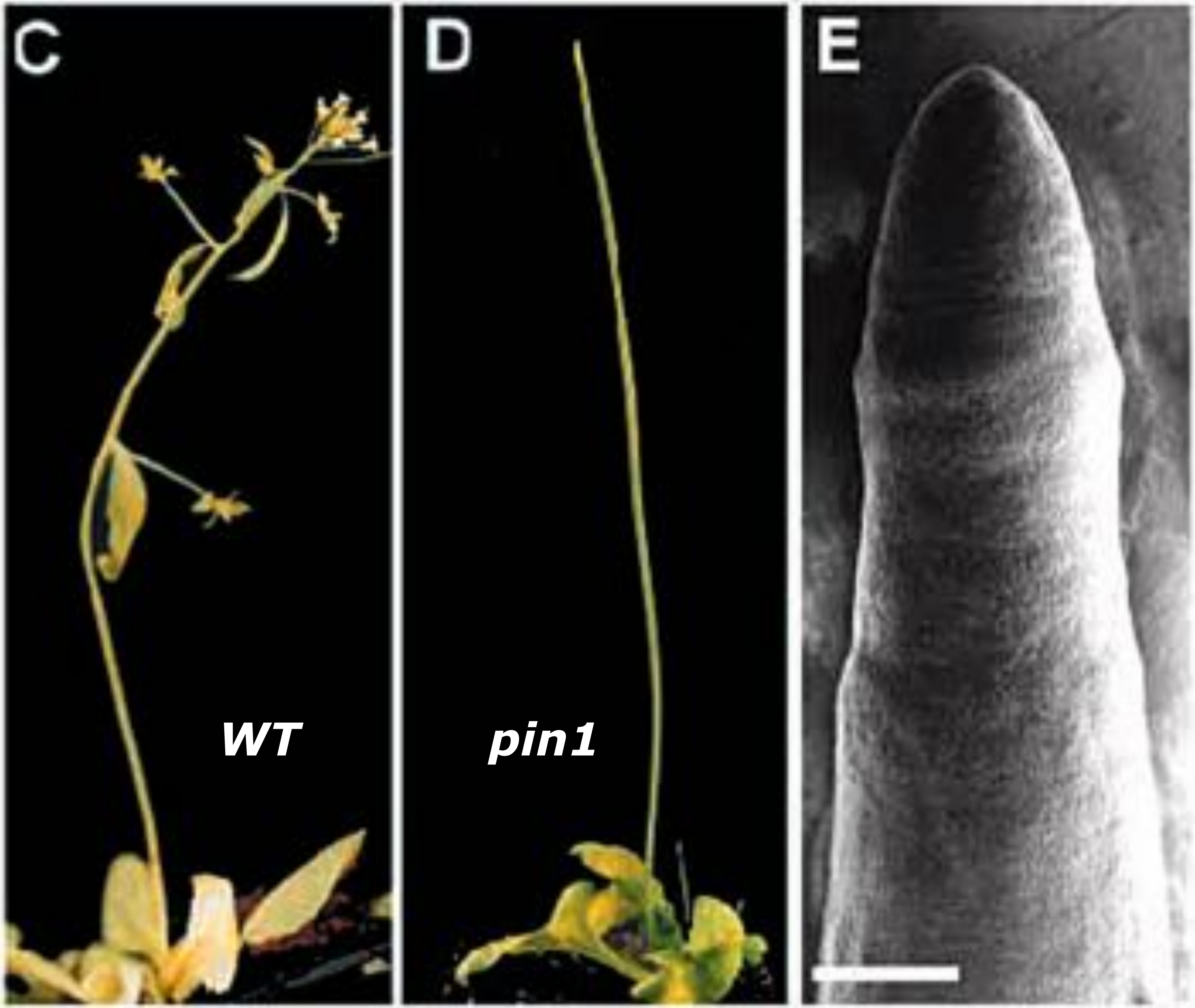
The pathway of auxin traffic through the plant is determined by the activities of influx and efflux carriers.

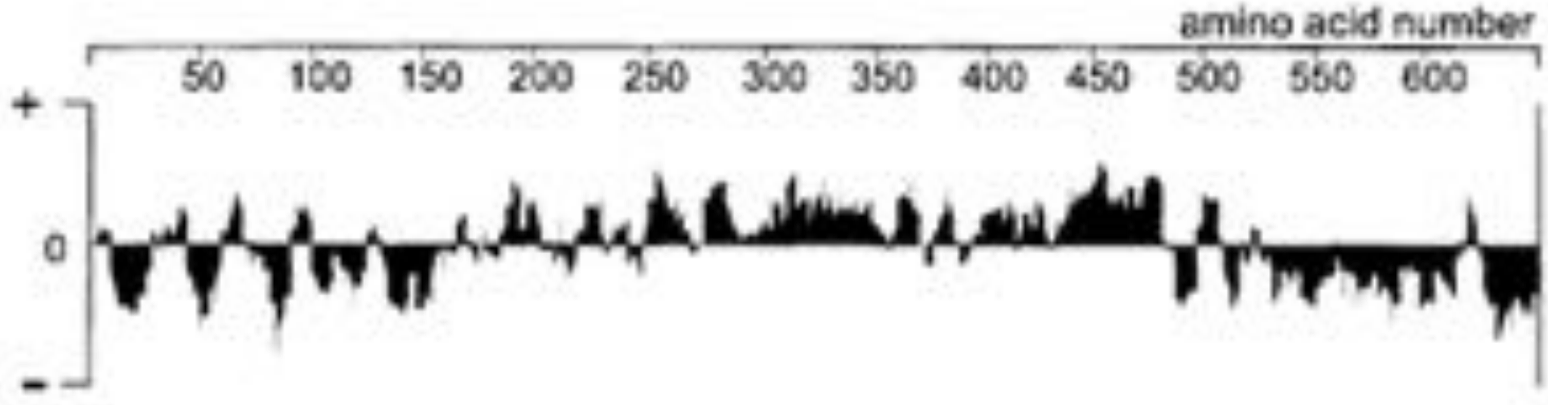




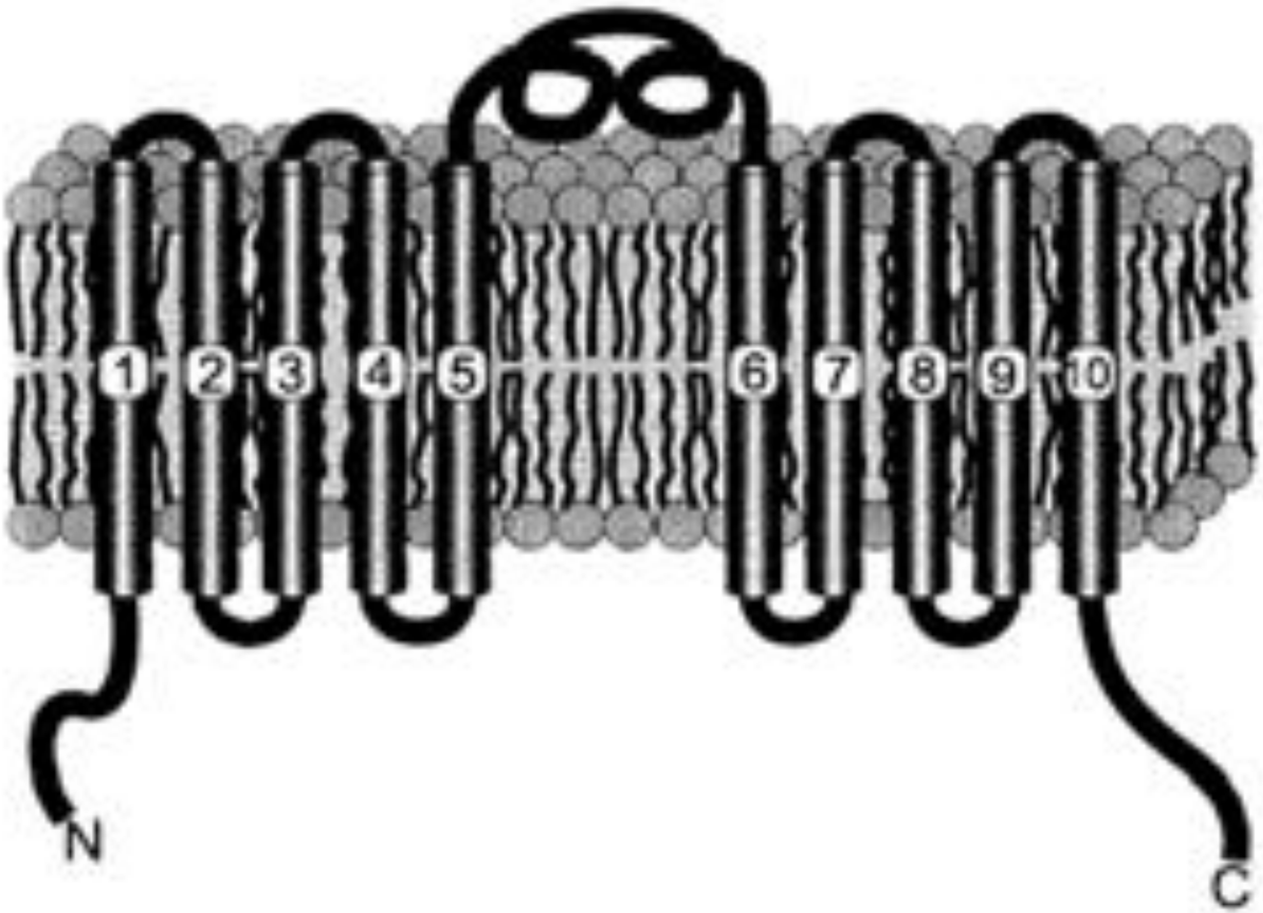
Auxin influx carrier: AUX1

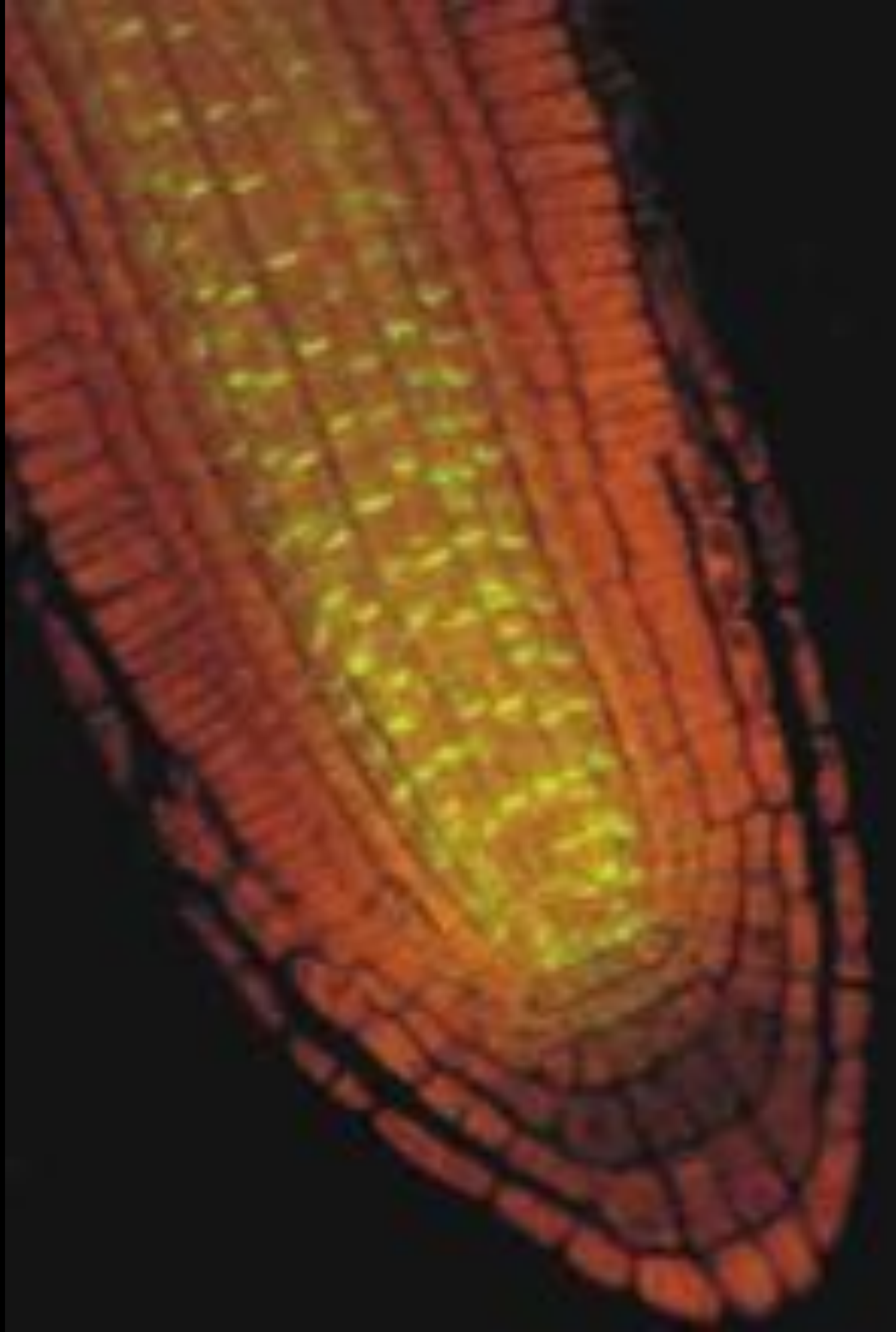
The aux1 mutant confers resistance to the herbicide 2,4-D, an auxin mimic


C
D
E
WT
pin1

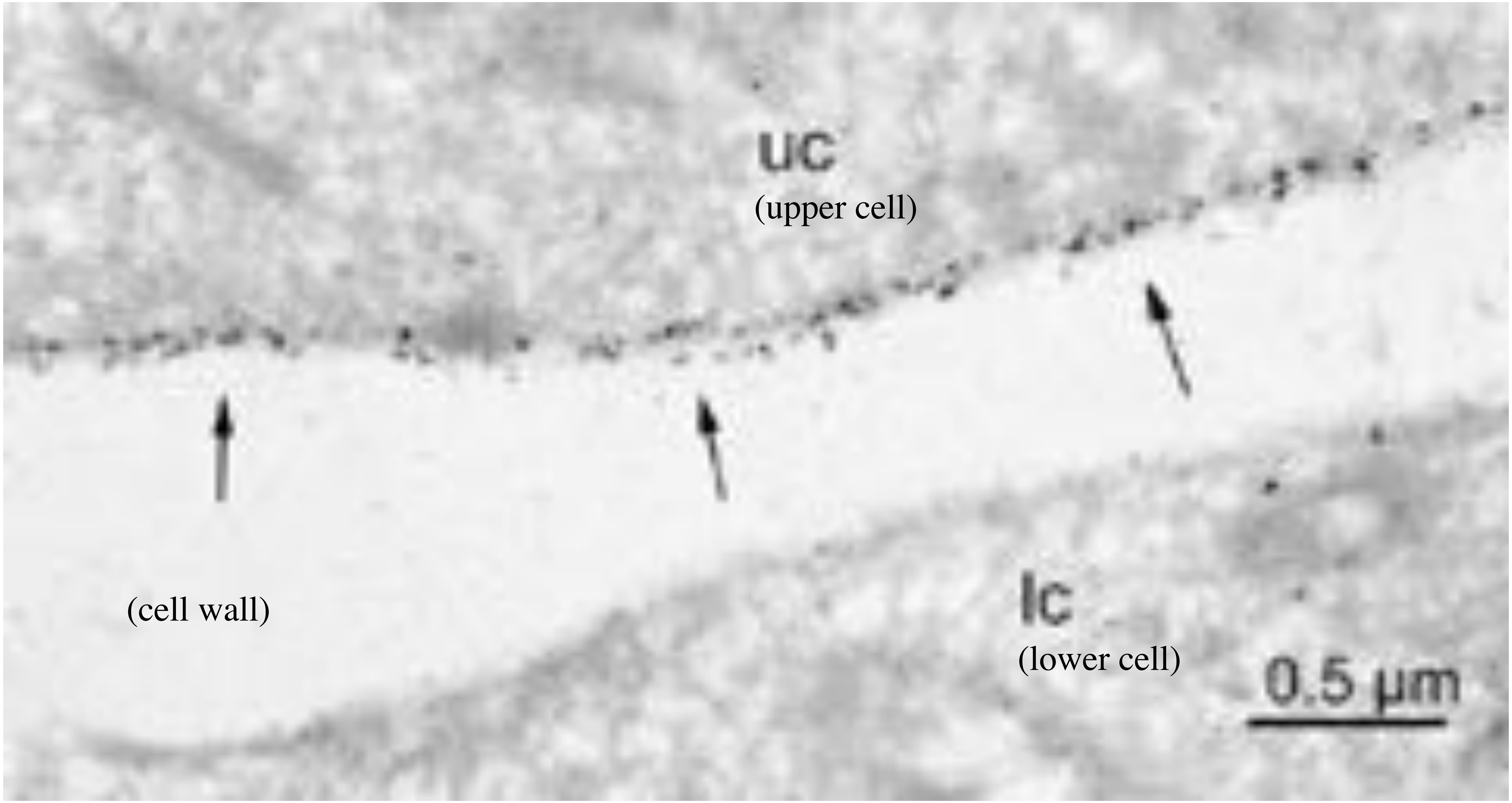


PIN1 auxin efflux carrier

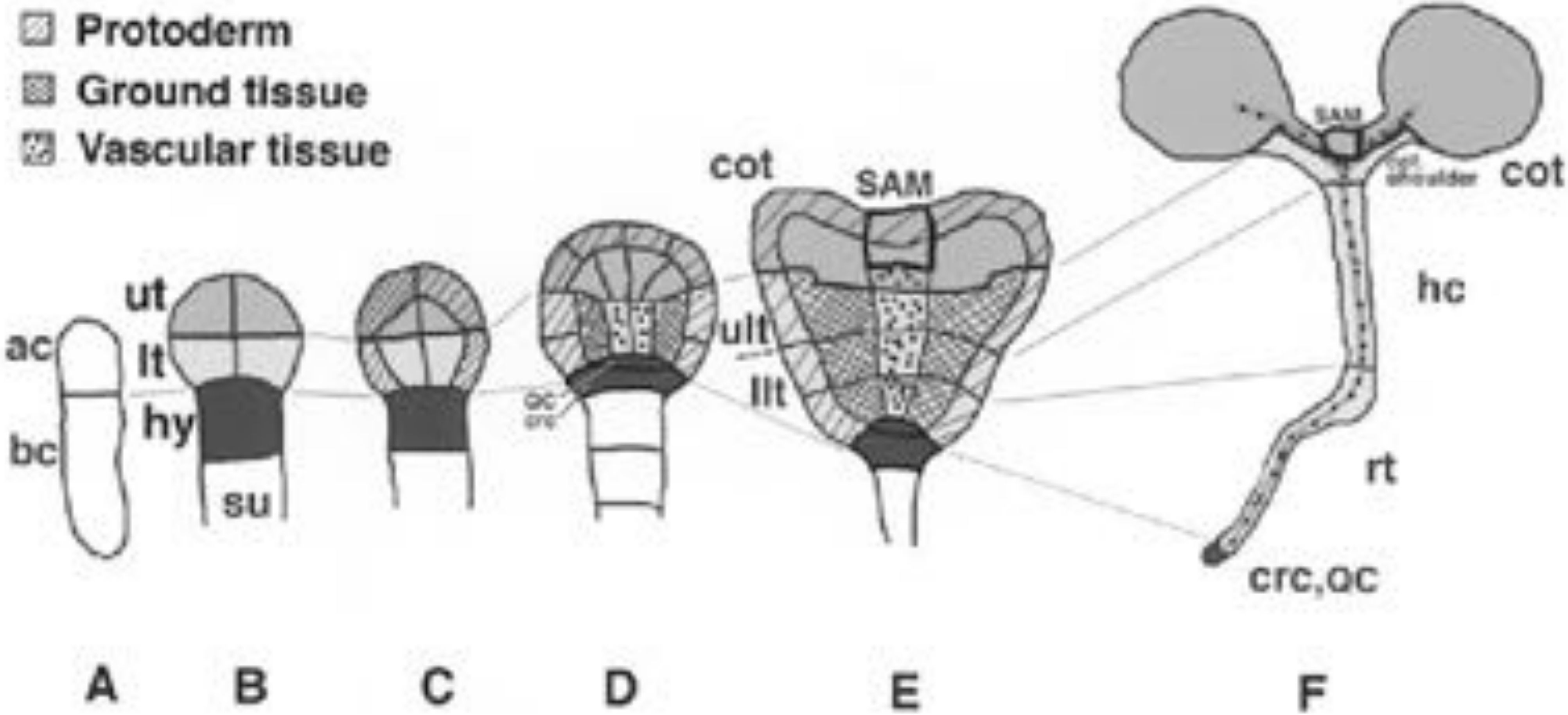




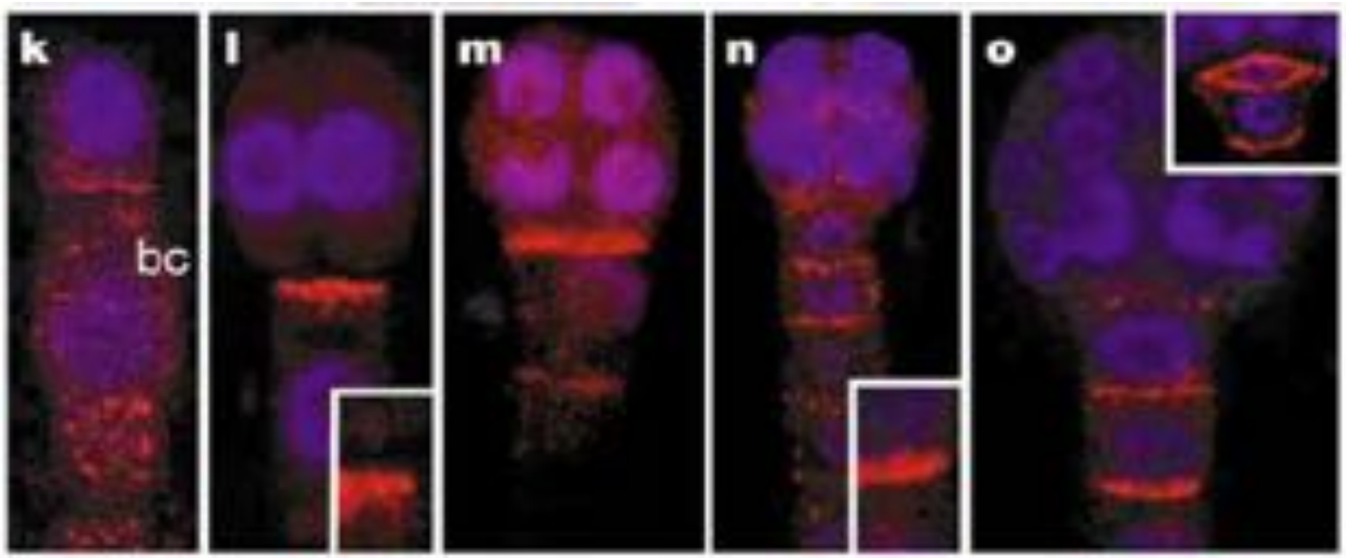
PIN1 is plasma membrane localised with a polar distribution within the cell



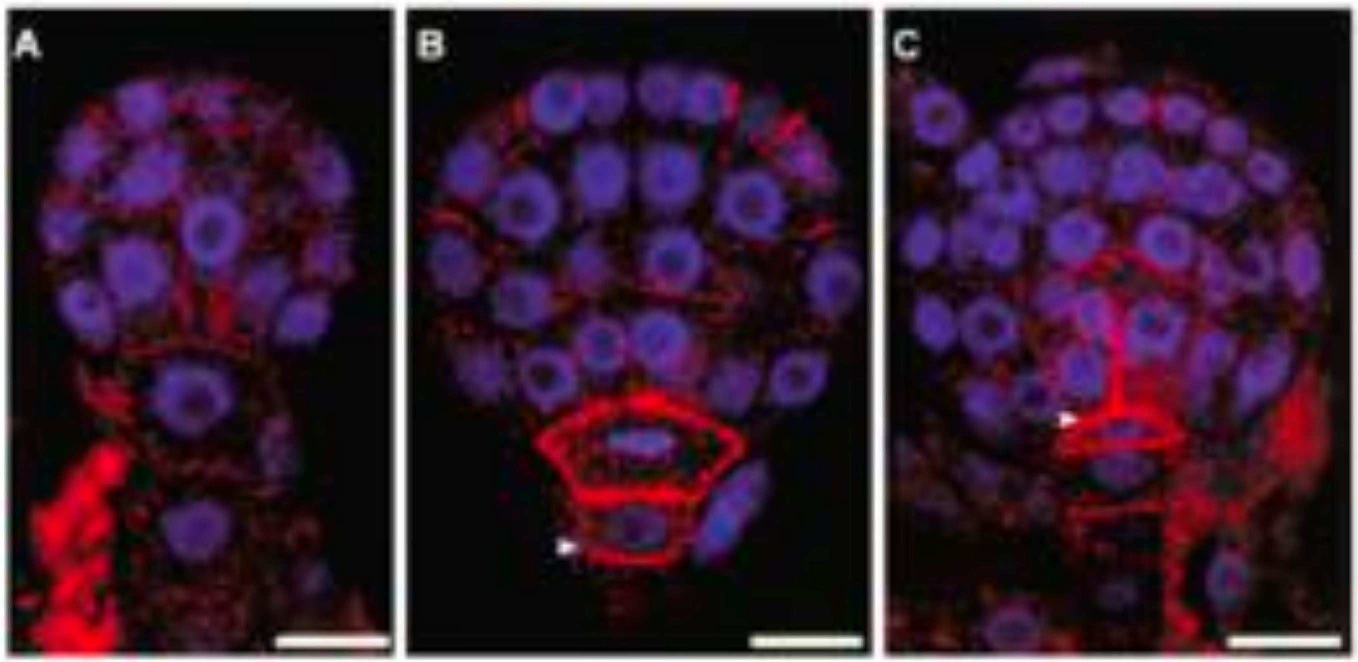
Auxin flow and accumulation regulates patterning in the embryo

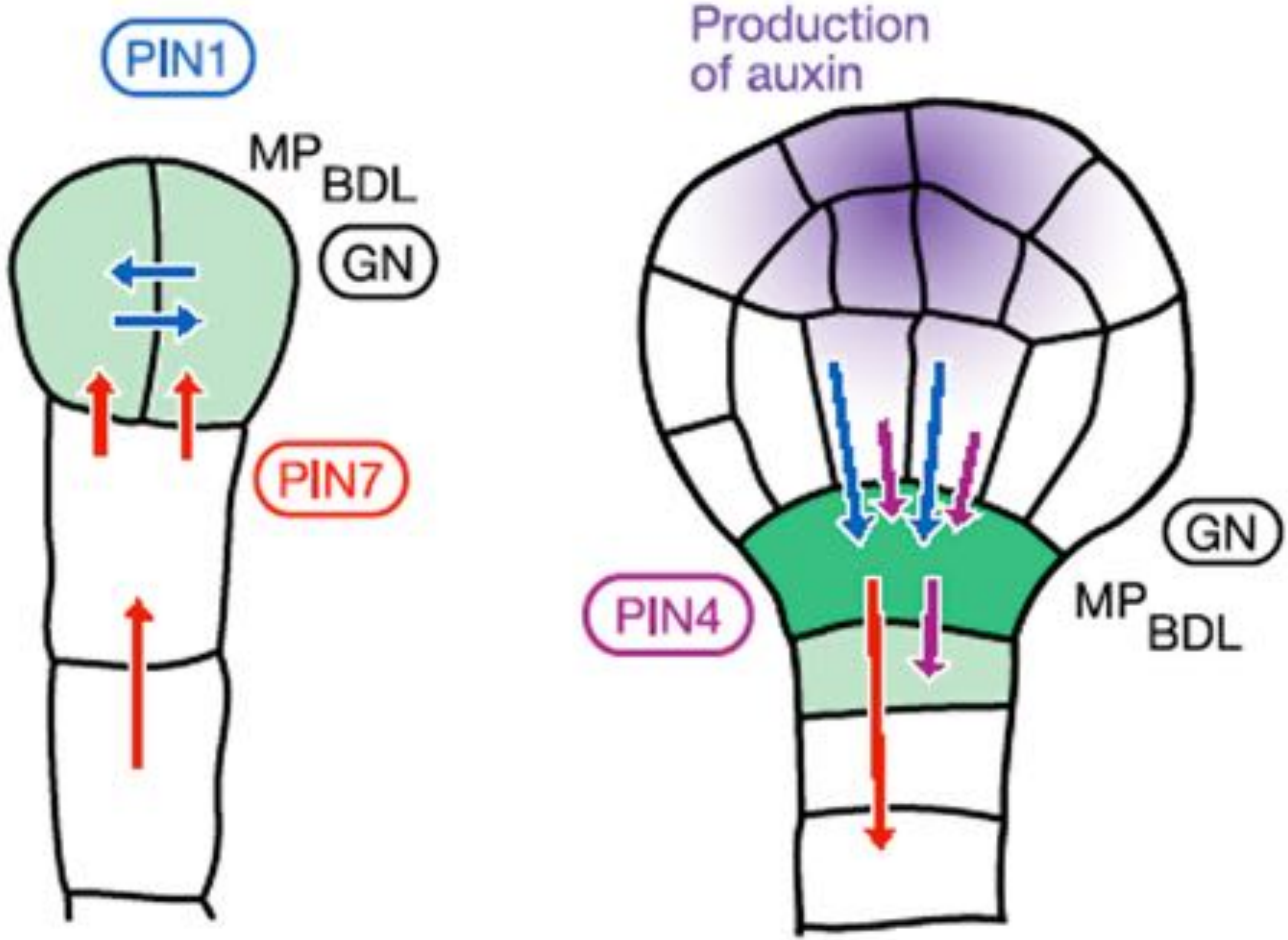


Immunolocalisation of PIN7 in Arabidopsis embryos

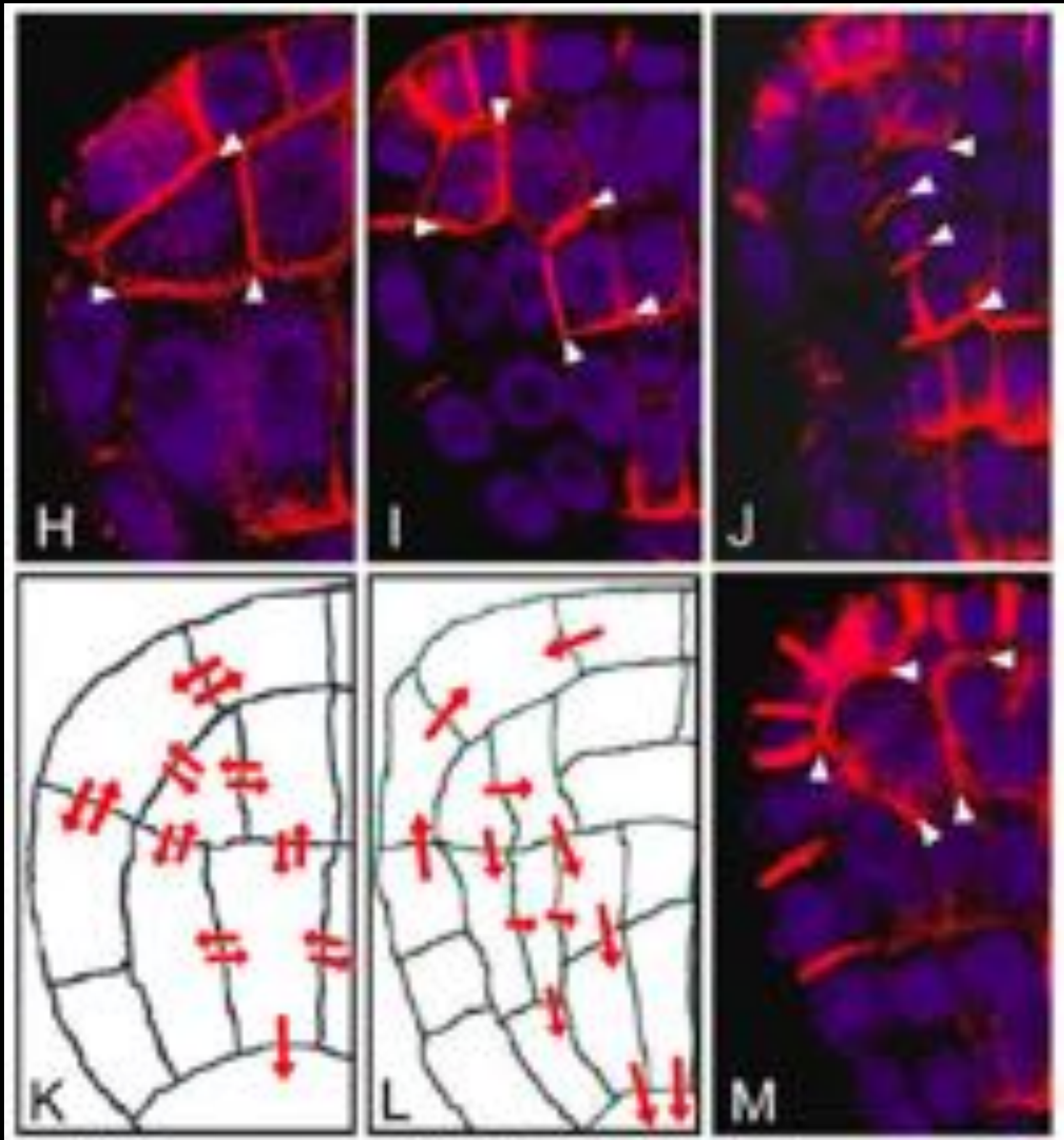


Immunolocalisation of PIN4 in Arabidopsis embryos

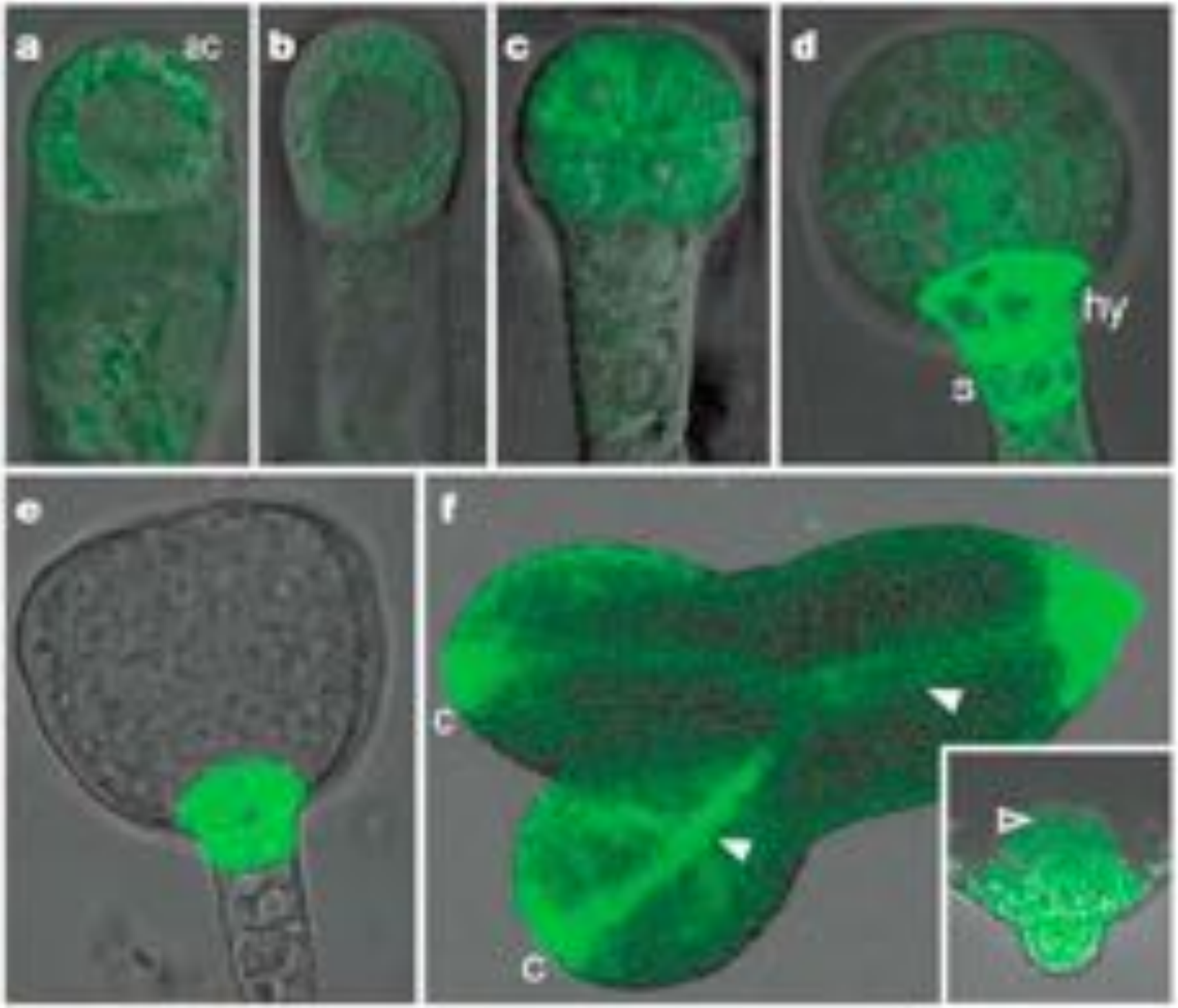



PIN1
MP
BDL
GN
PIN7
Production
of auxin
GN
PIN4
MP
BDL

Changes in PIN1 in distribution during Arabidopsis embryogenesis

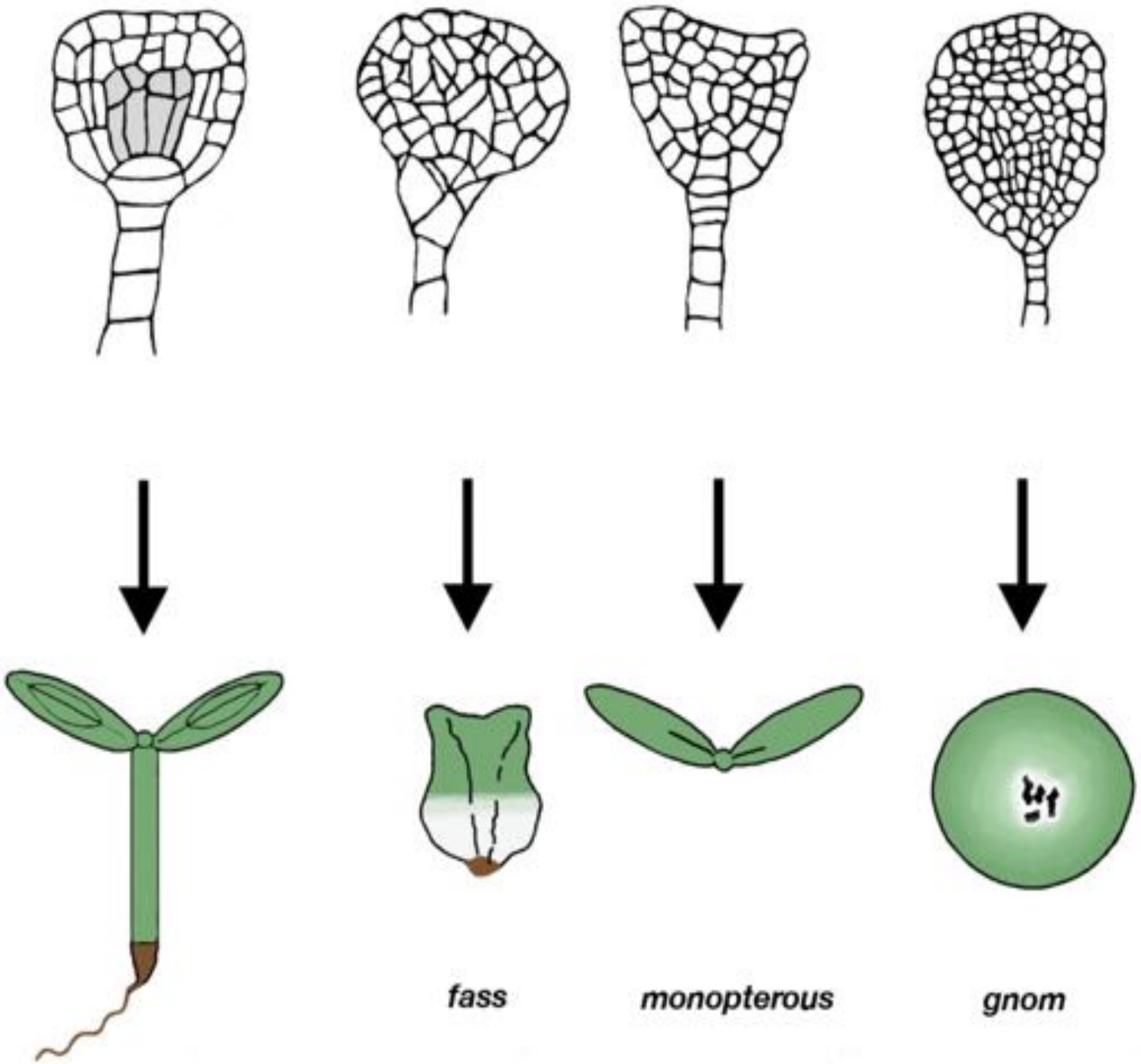


Auxin triggered gene expression during embryogenesis



DR5::GFP

Mutations that affect auxin traffic or perception give rise to plants with altered body plans.



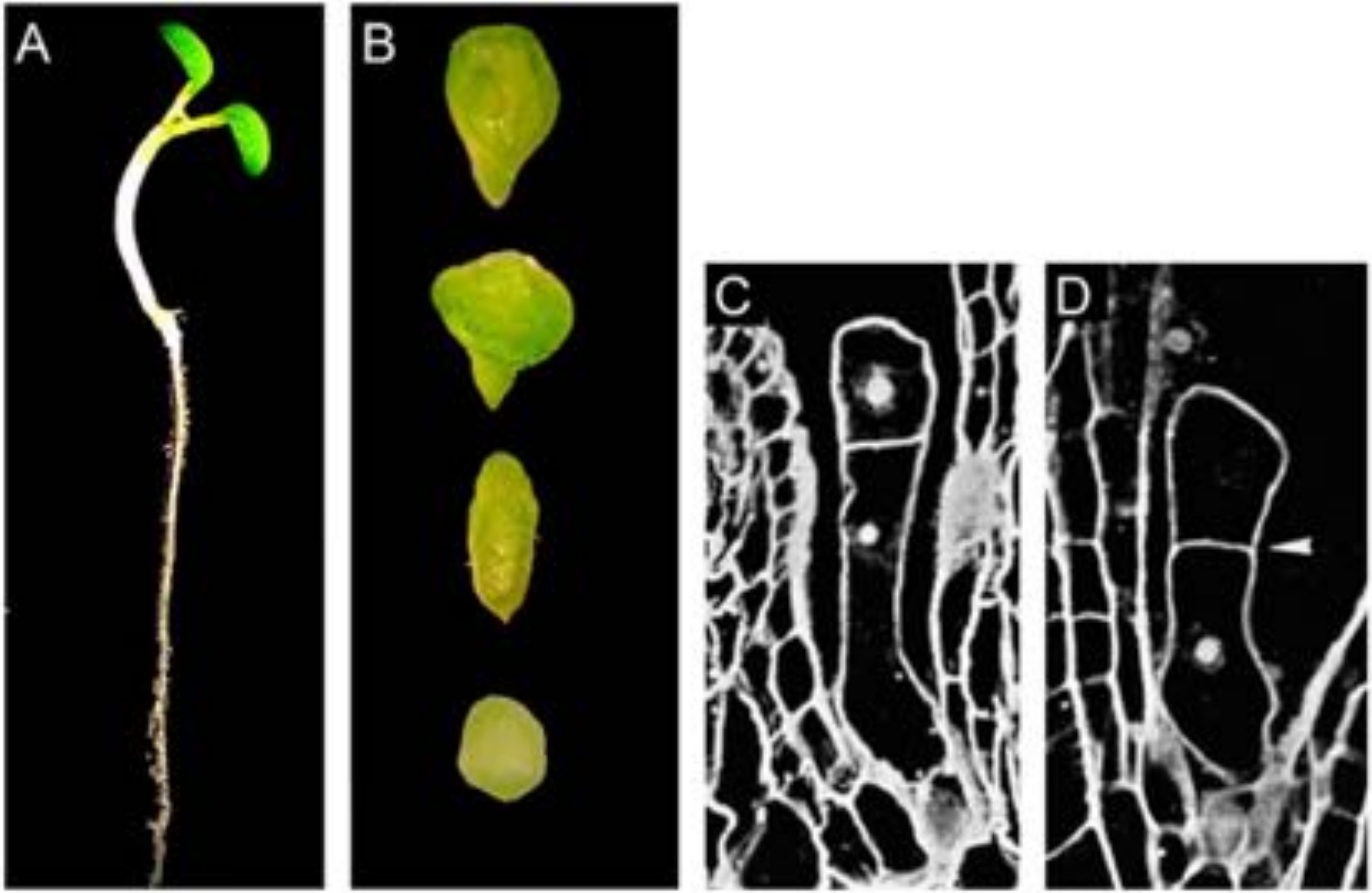
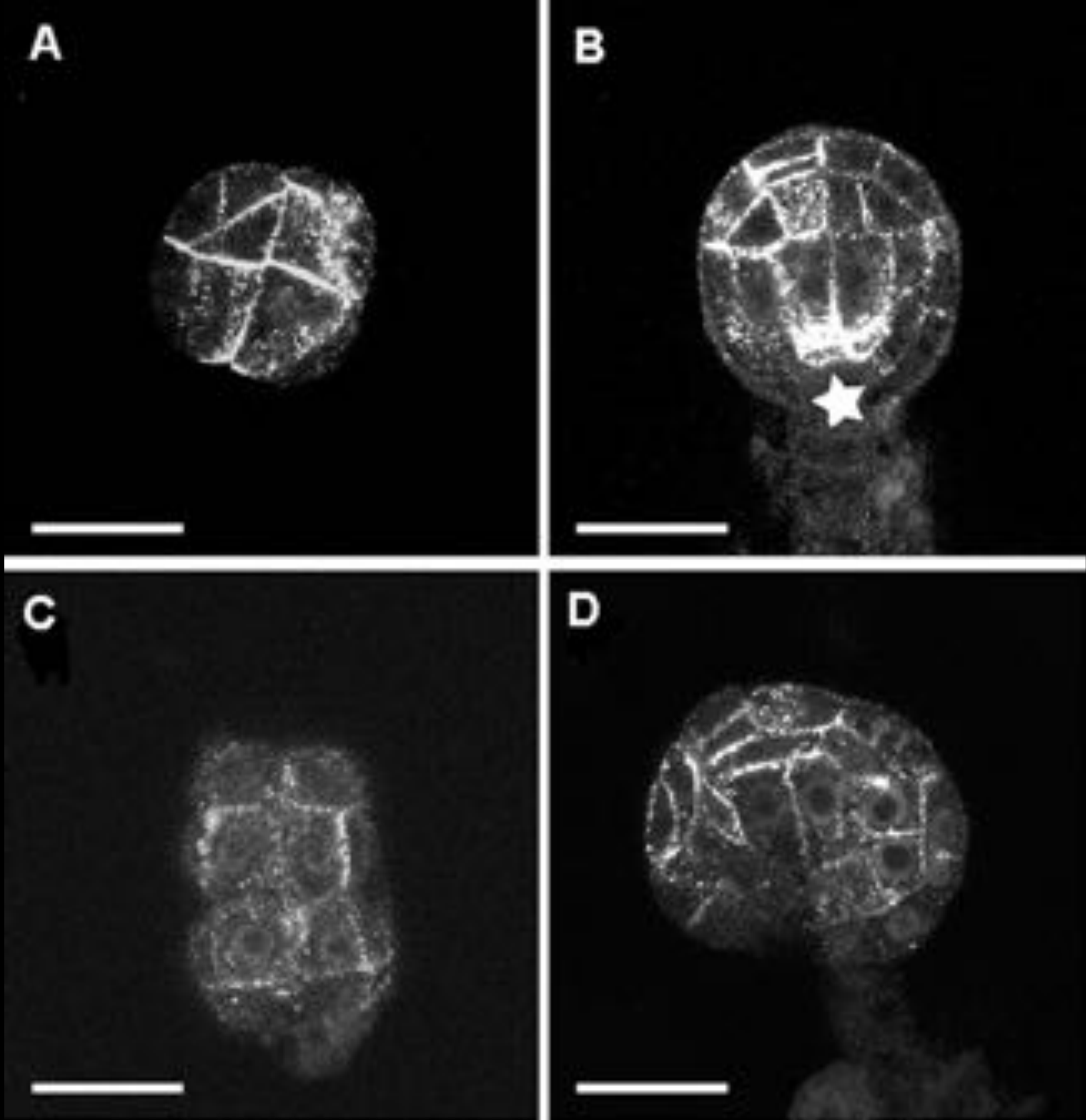


Fig. 1. *gnom* mutant phenotype. (A, C) Wild-type, (B, D) *gnom*. (A, B) Seedling, (C, D) One-cell stage of embryogenesis. Modified after (Mayer et al., 1993).

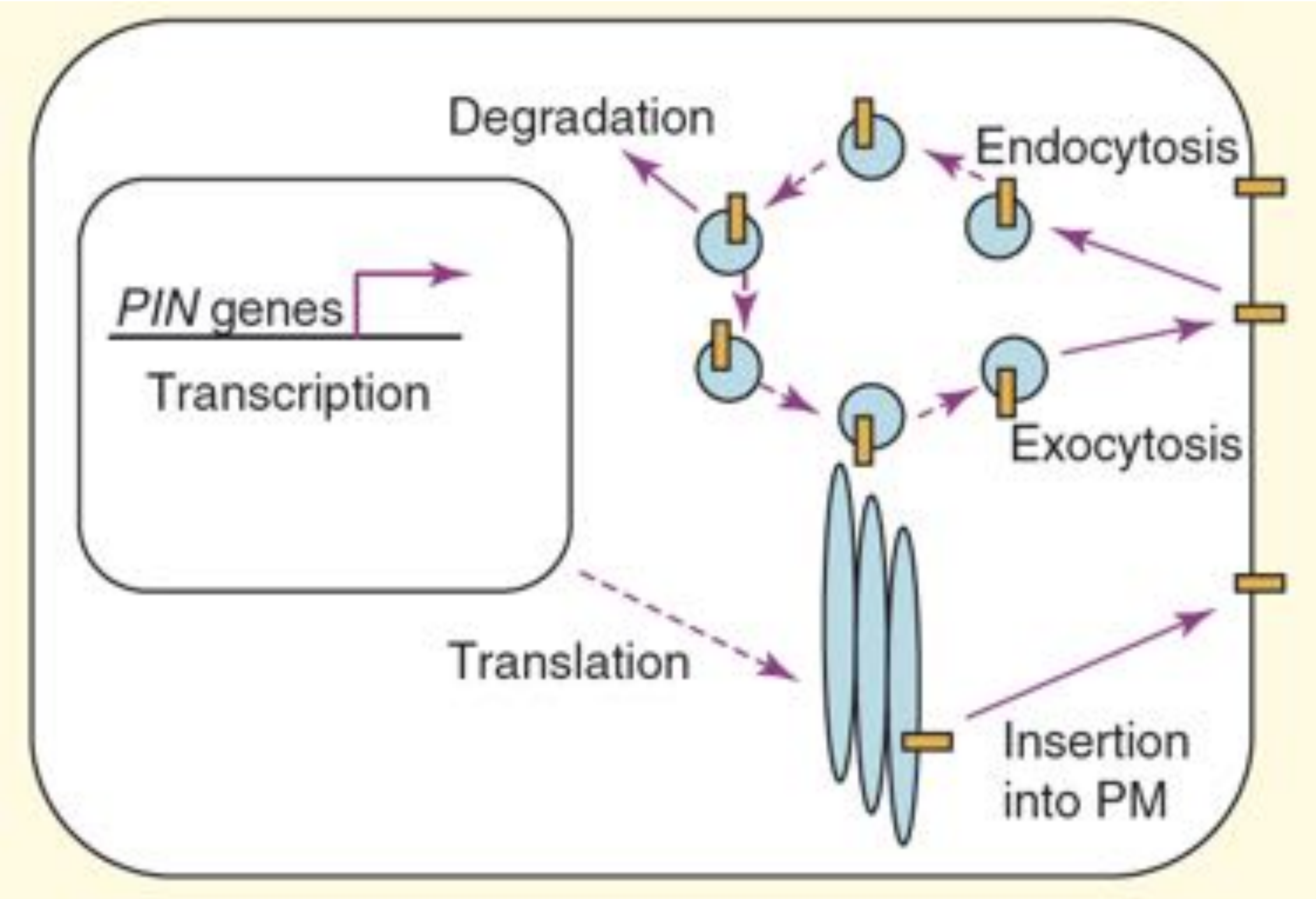


wild type

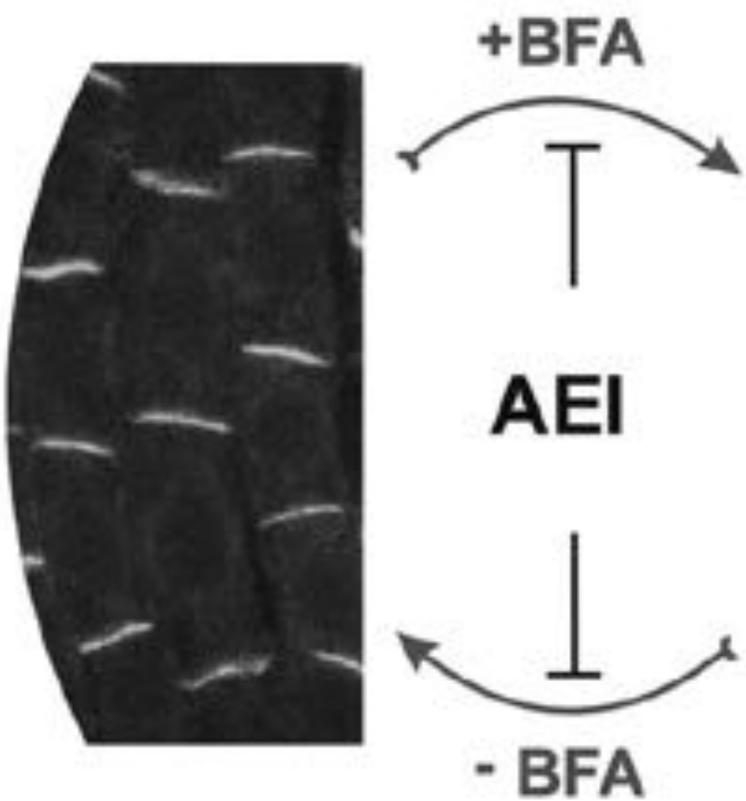
gnom mutant

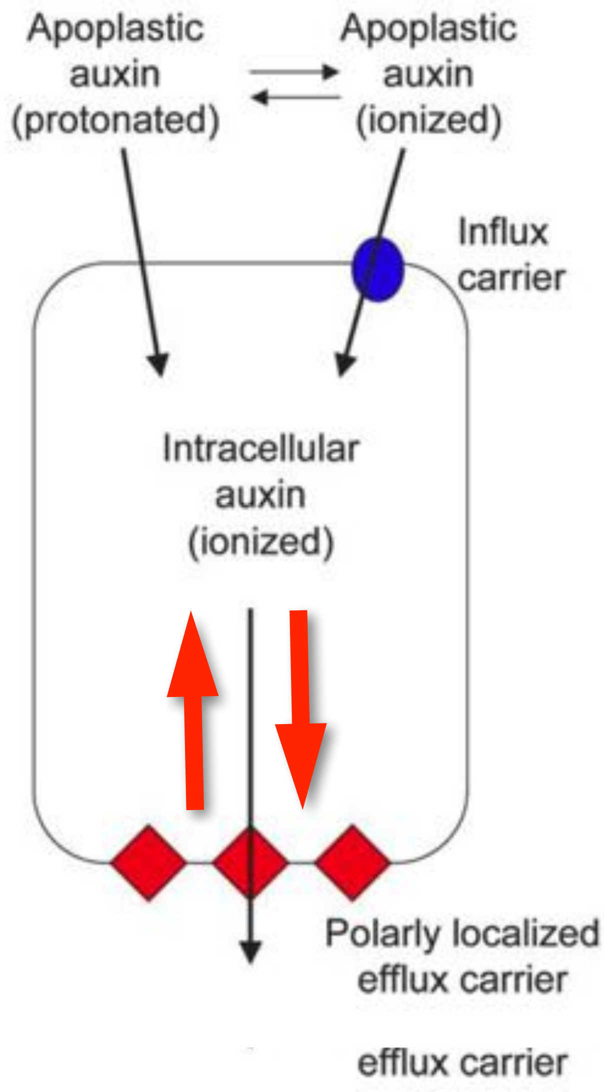
Immunolocalisation of PIN1 in Arabidopsis embryos

Feedback through regulated expression and localisation of PIN genes



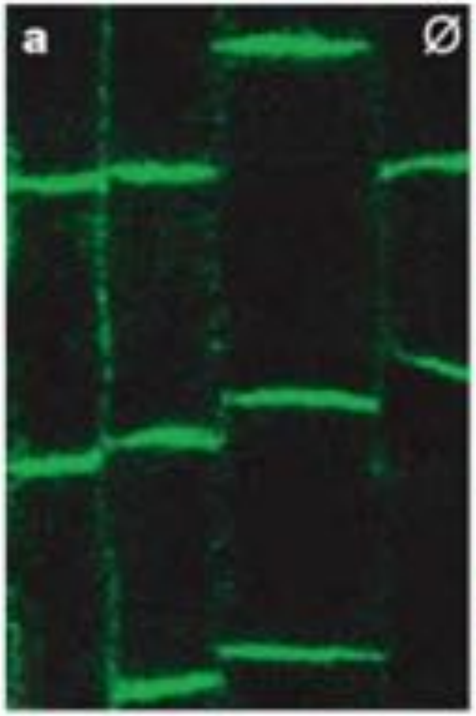
Asymmetric localisation of the PIN1 auxin efflux transporter is a dynamic process and requires the maintenance of polar secretion.

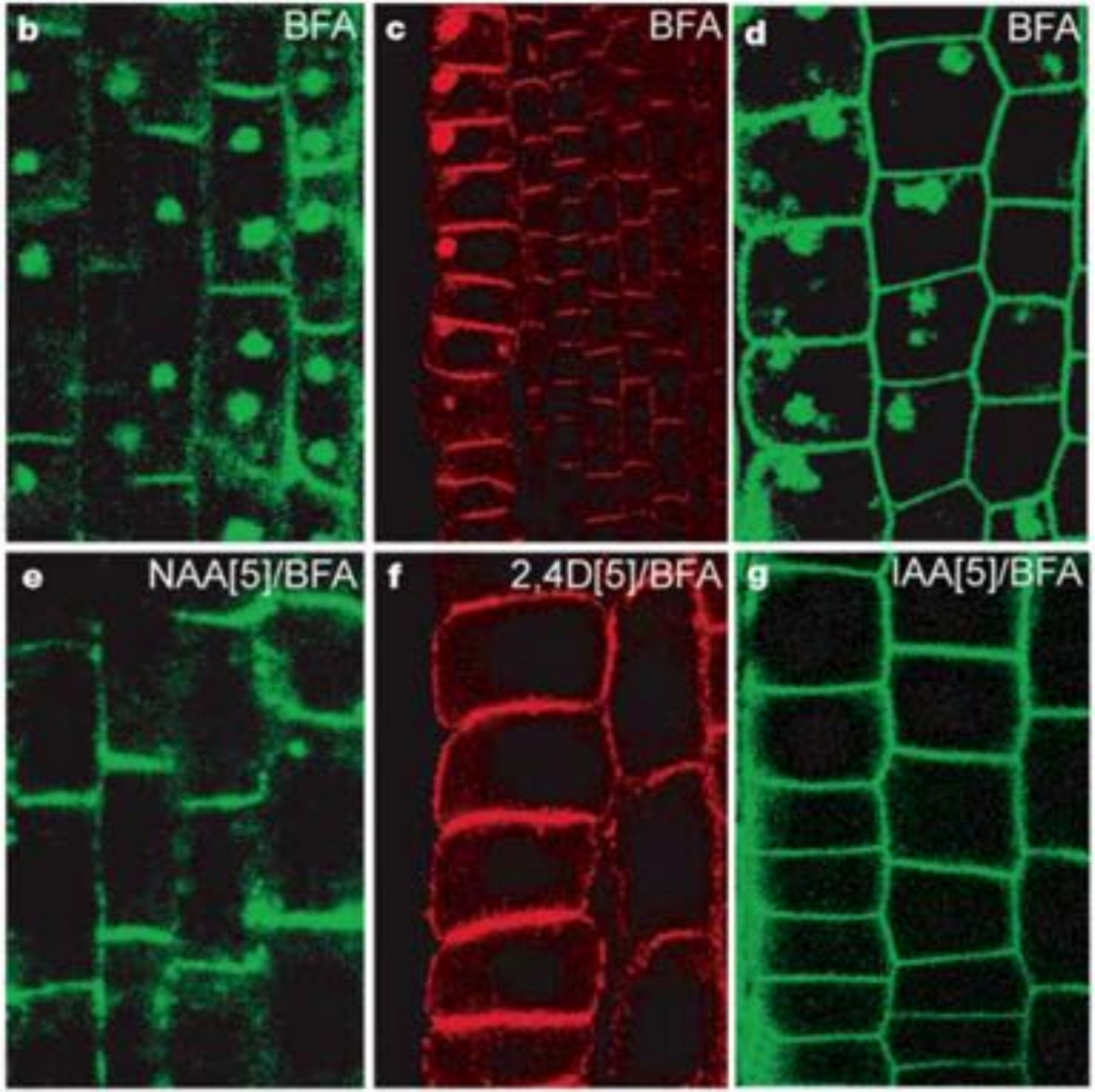




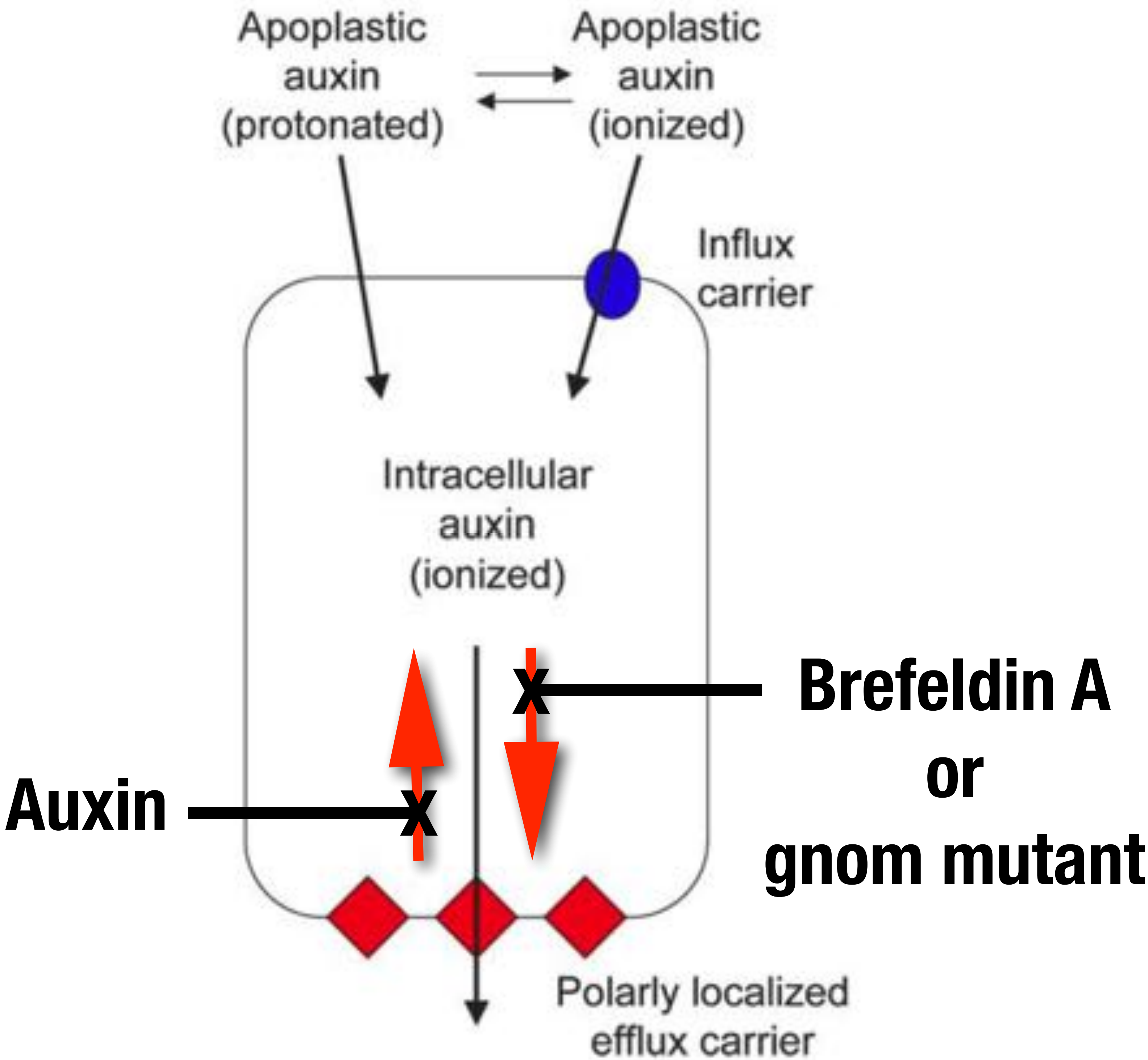
Brefeldin A treatment causes rapid loss of PIN1 localisation.

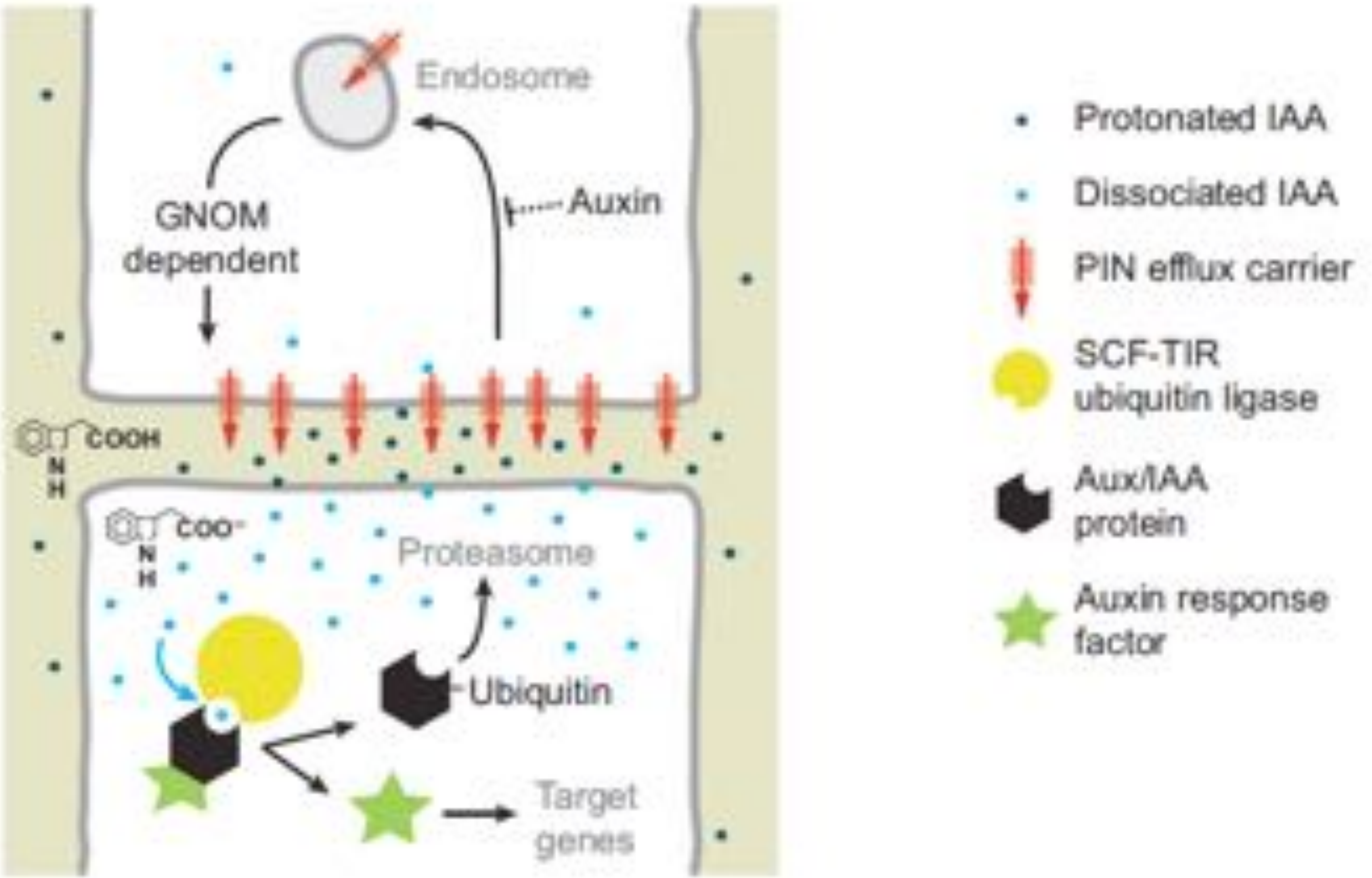
untreated PIN1-GFP | PIN1-GFP | PM-ATPase | PIN2-GFP

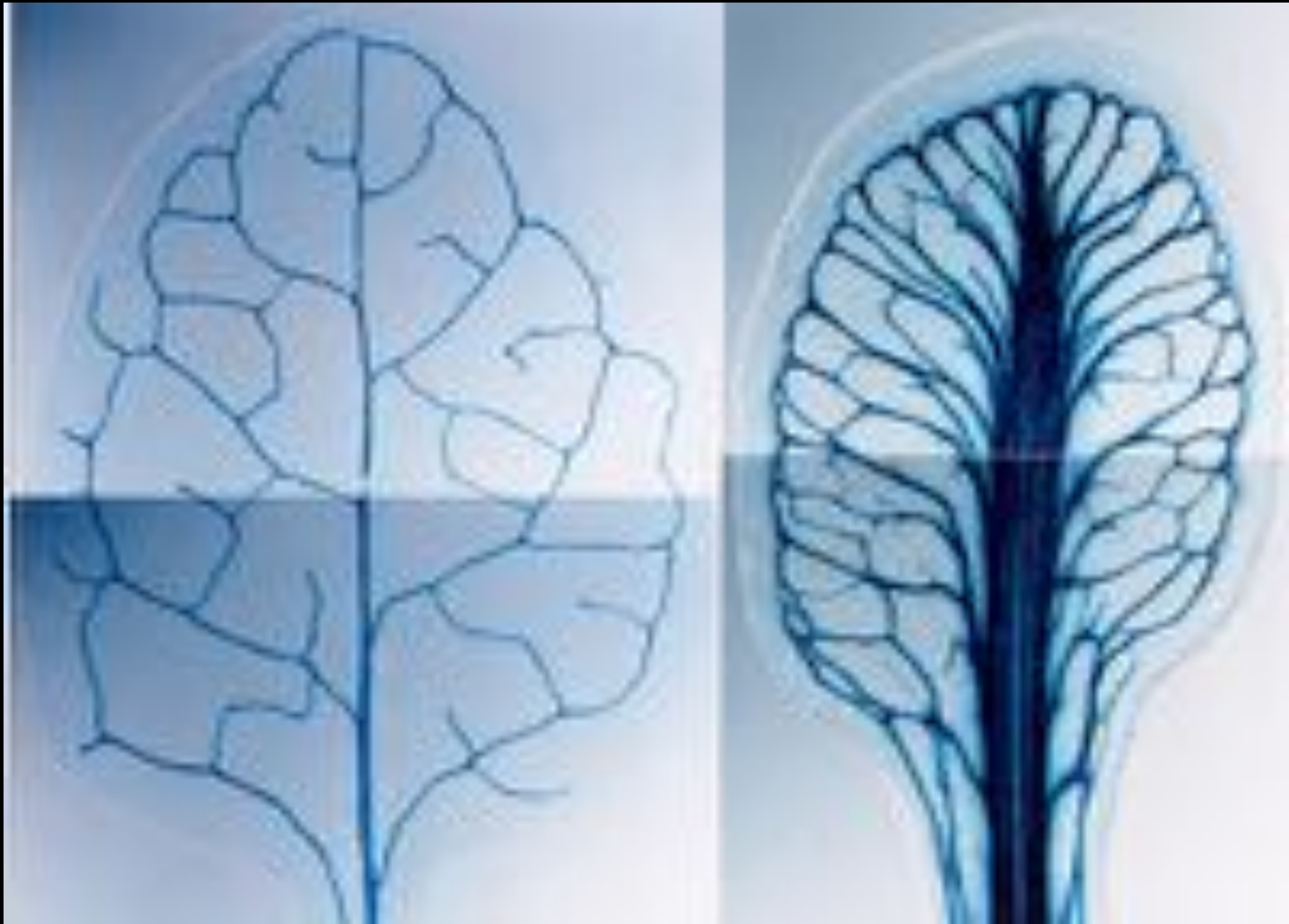




Application of auxin reverses the effect of Brefeldin A (BFA)

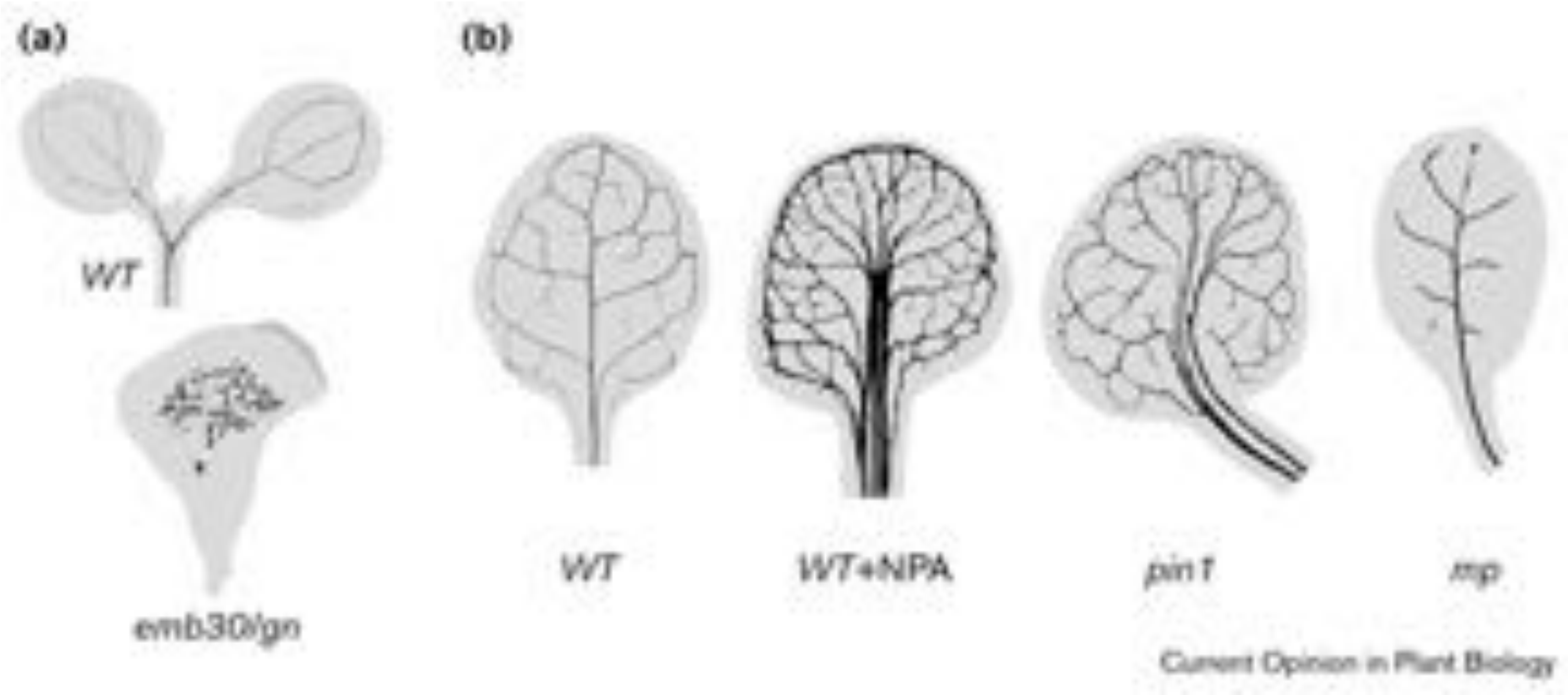

Apoplastic auxin (protonated)
Apoplastic auxin (ionized)
Influx carrier
Intracellular auxin (ionized)
Brefeldin A or gnom mutant
Auxin
Polarly localized efflux carrier

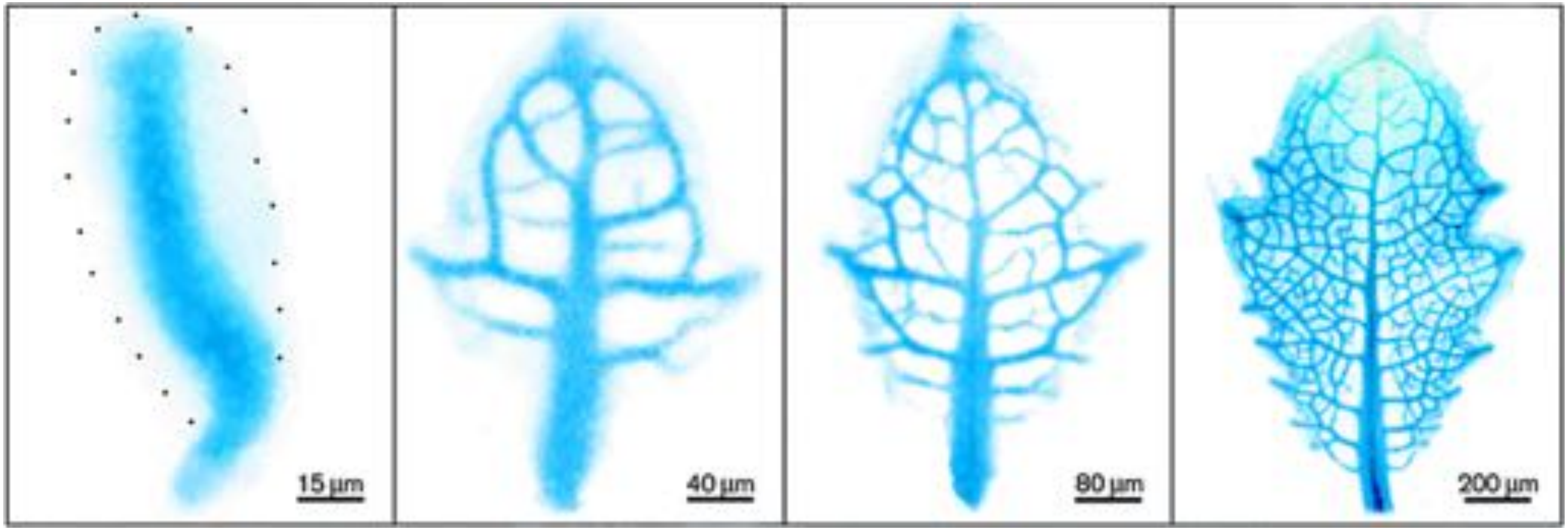

Endosome
GNOM dependent
Auxin
COOH
N H
COO-
Proteasome
Ubiquitin
Target genes
Protonated IAA
Dissociated IAA
PIN efflux carrier
SCF-TIR ubiquitin ligase
Aux/IAA protein
Auxin response factor



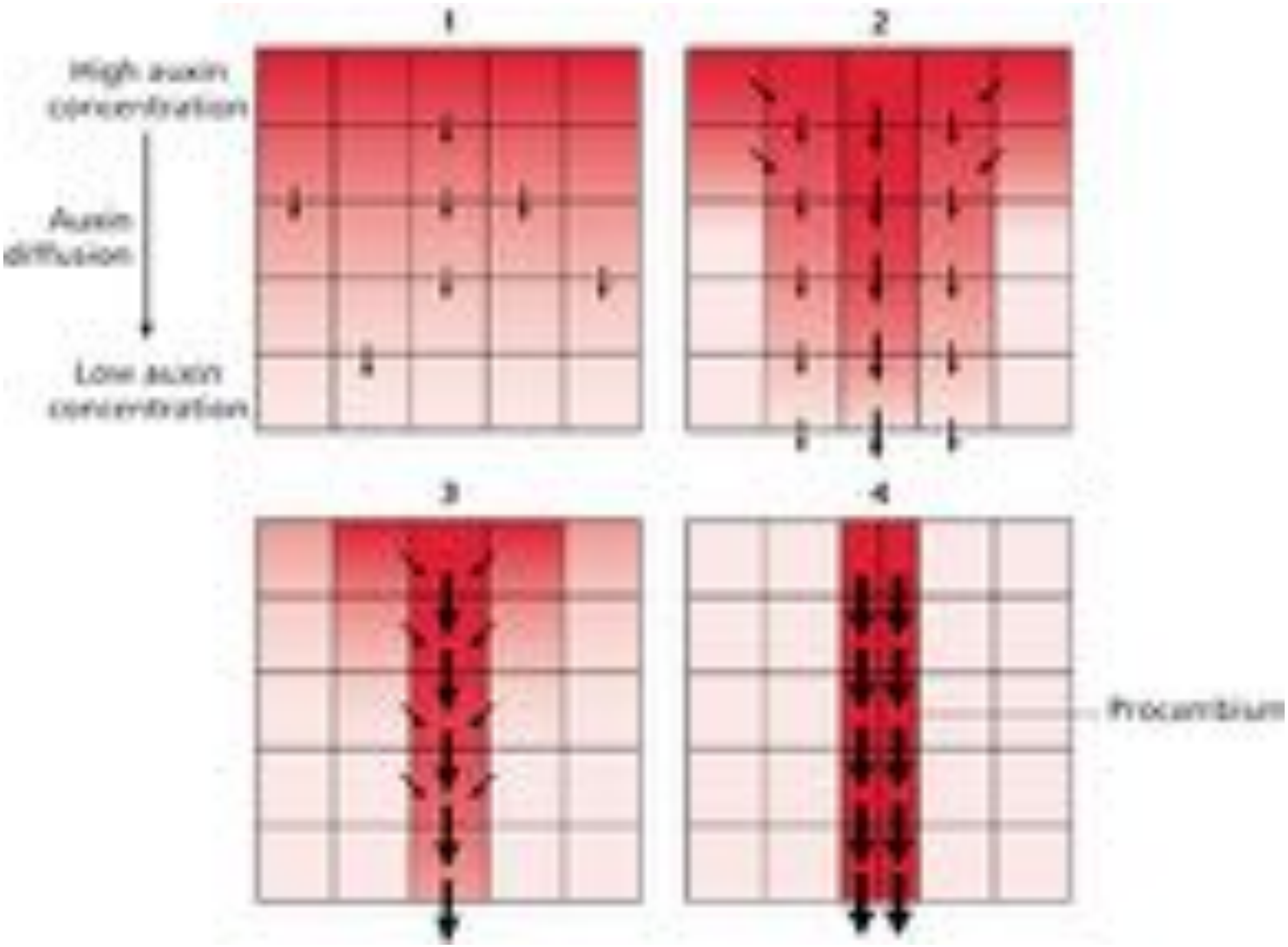
Inhibition of auxin transport by application of NPA

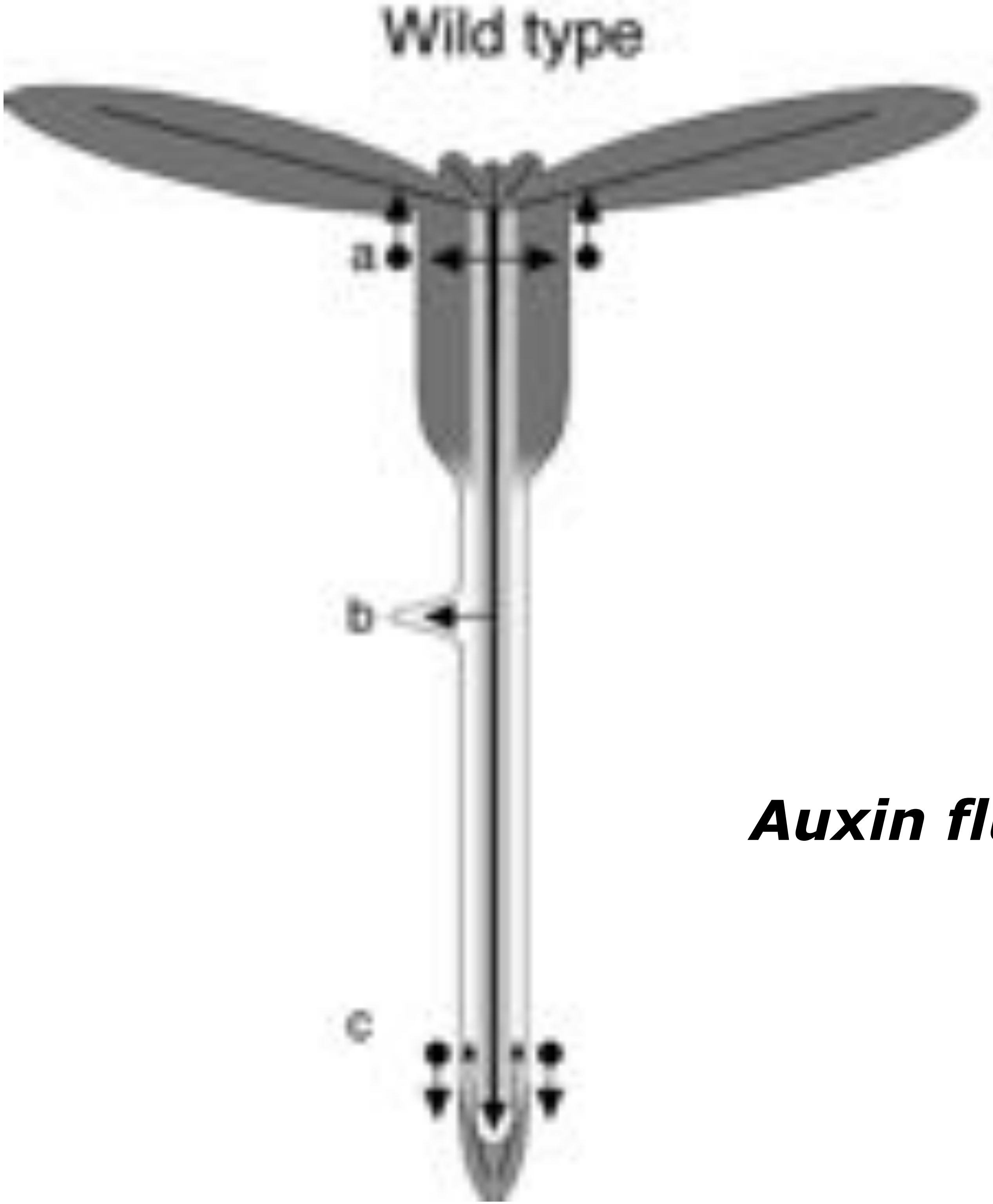
Defects in auxin transport or response affect patterning of the plant vascular system



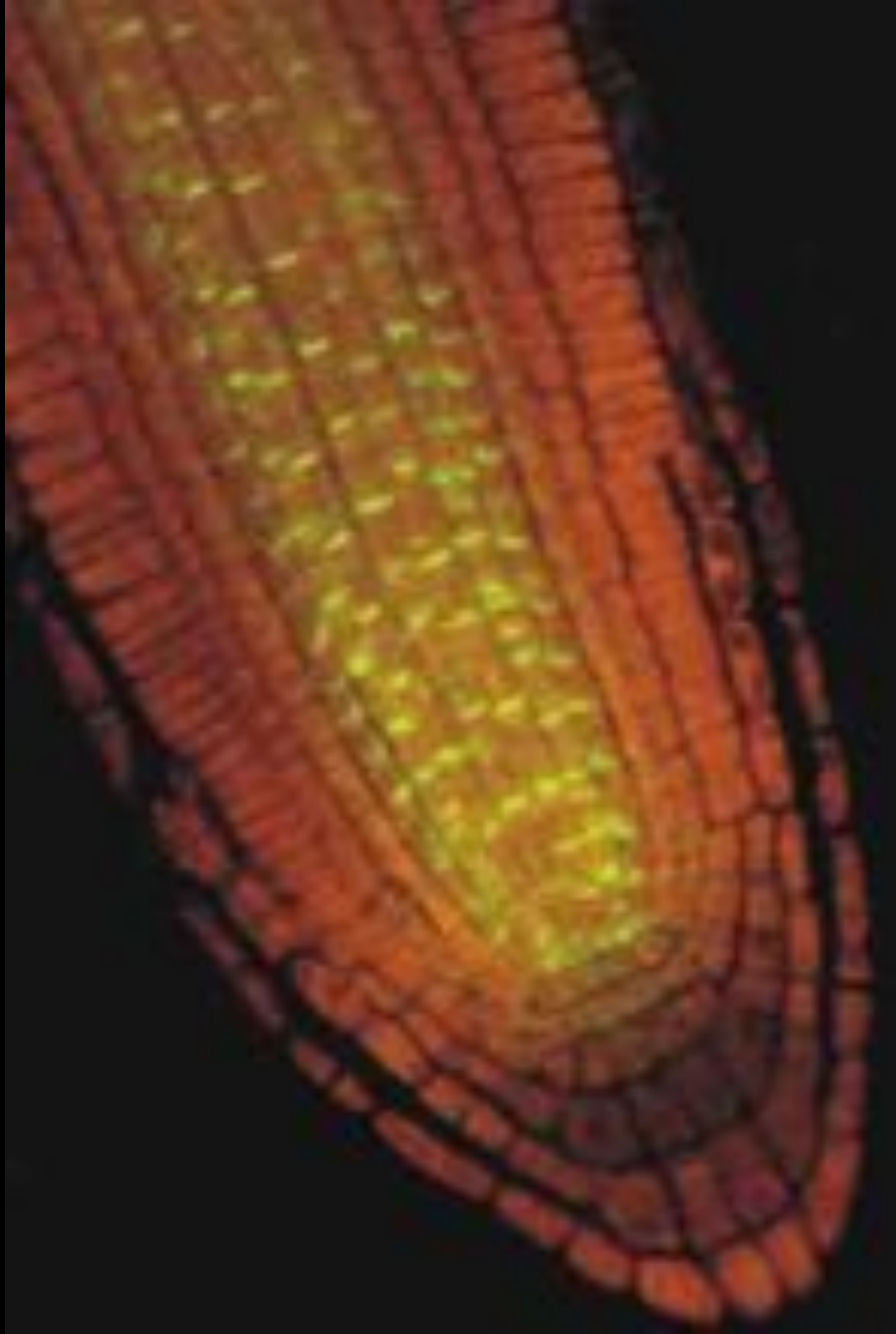

15 µm
40 µm
80 µm
200 µm

"Canalisation" of auxin flow





Auxin flux



PIN1 localisation

Family of PIN genes in the Arabidopsis root

pin2 mutant seedlings show loss of gravitropism in the root

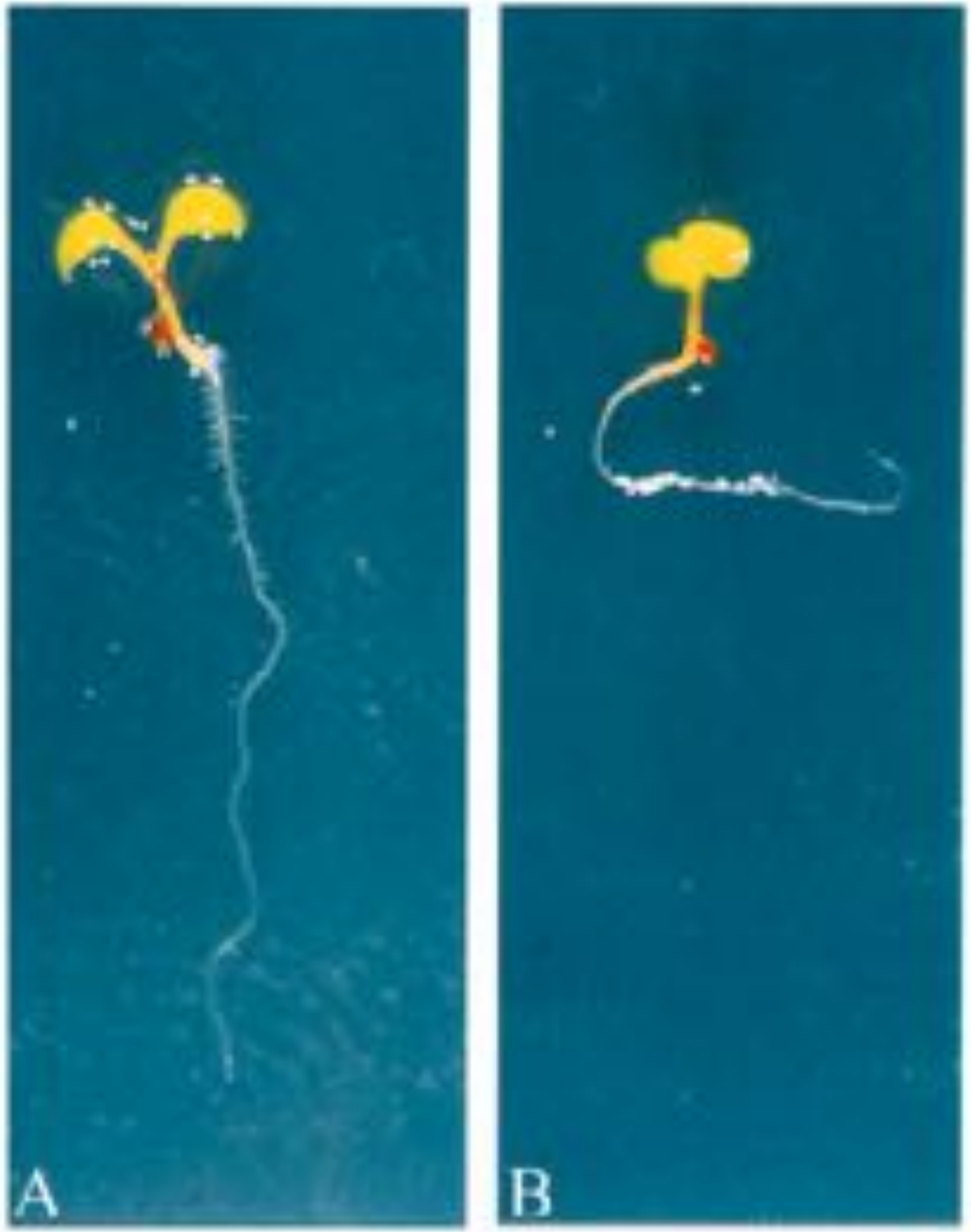
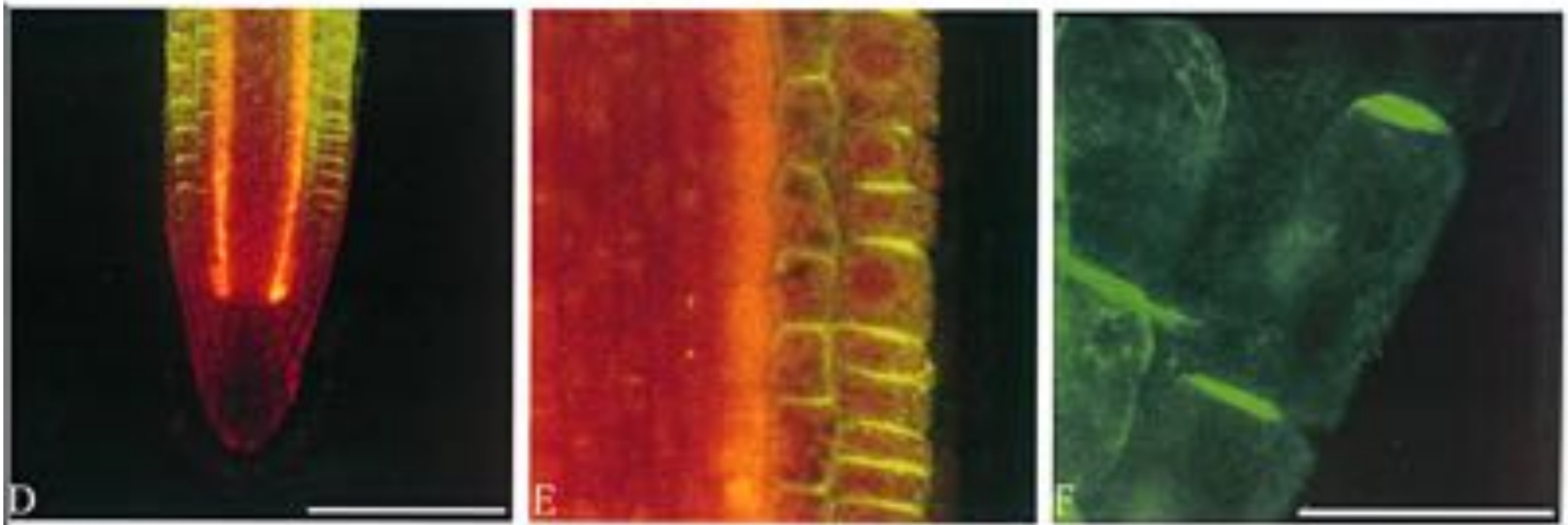
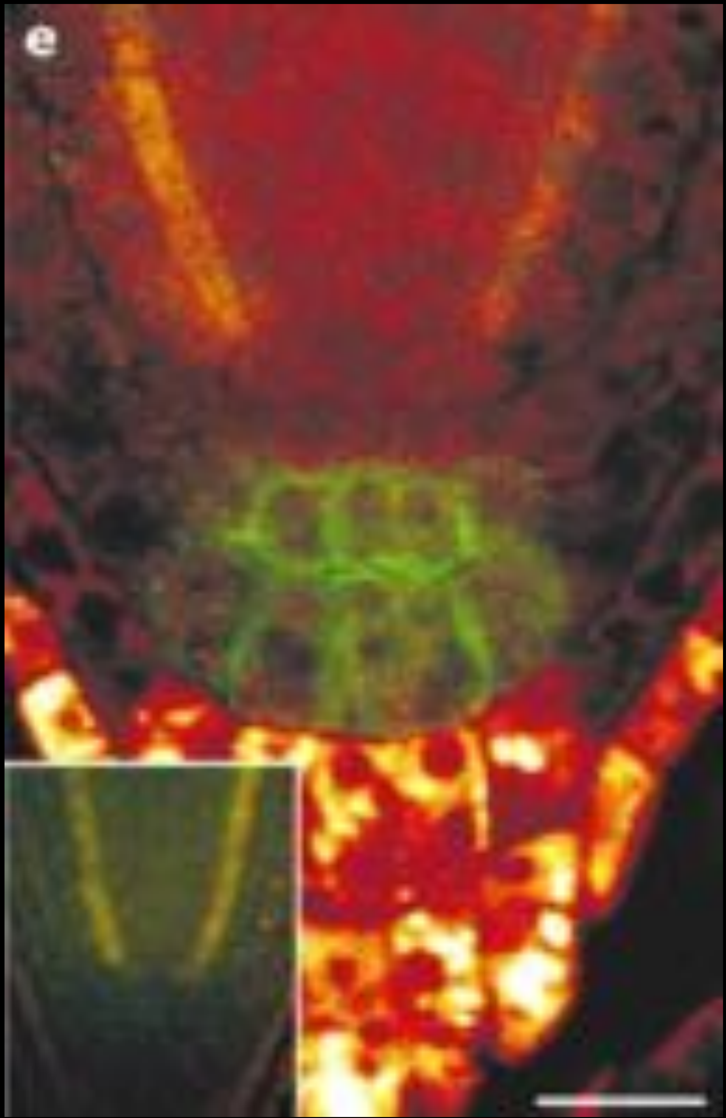


Fig. 3. Mutations in the *AtPIN2* gene alter root growth and gravitropism. Homozygous 5-day-old Columbia-0 wild-type seedlings (**A**) and *Atpin2::En701* mutant seedlings (**B**) were grown vertically on agar plates.

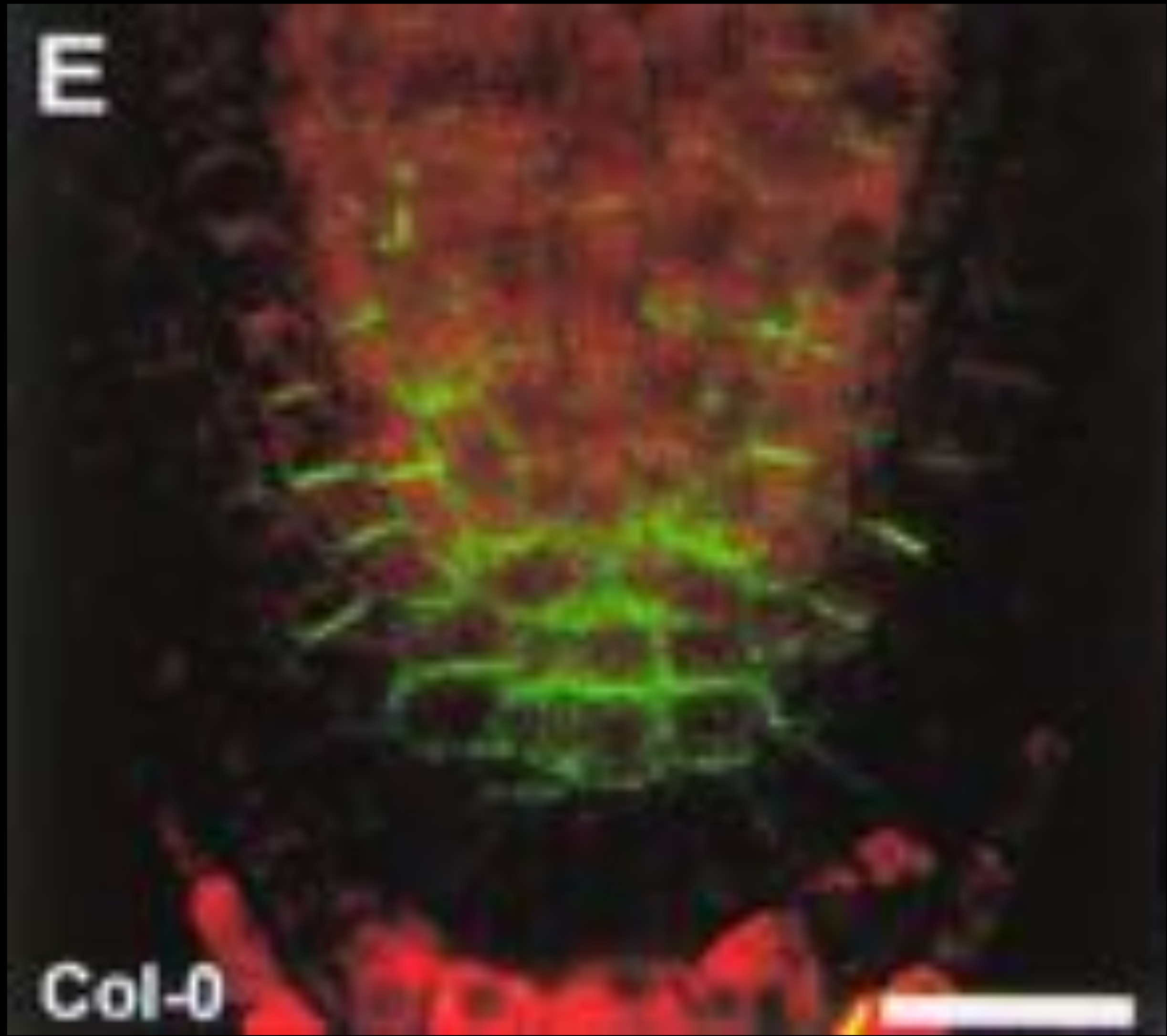


Localization of AtPIN2p in 4-day-old *Arabidopsis* seedling root tips.

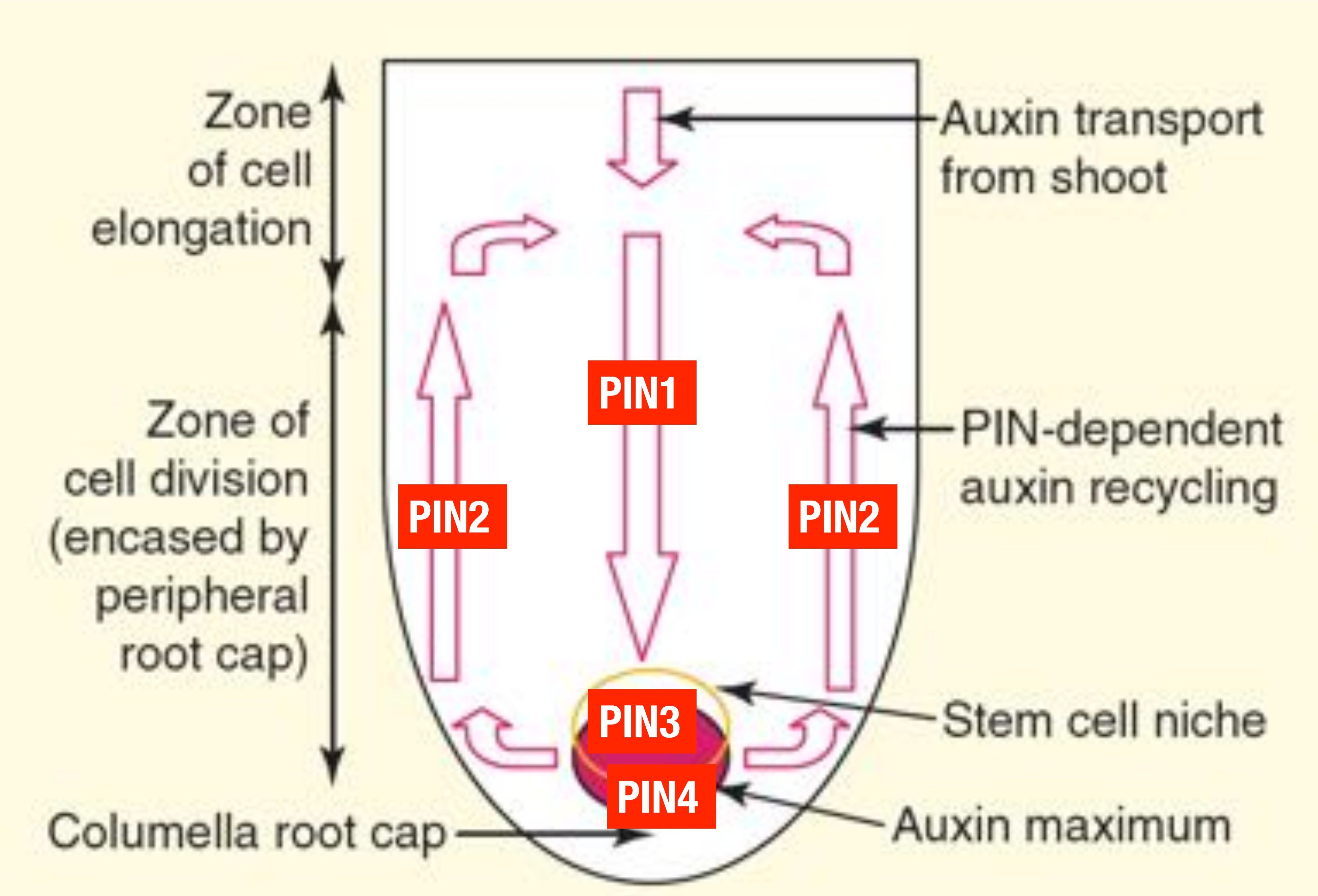
PIN2 localisation



PIN3 localisation



PIN4 localisation


Zone of cell elongation
Zone of cell division (encased by peripheral root cap)
Columella root cap
Auxin transport from shoot
PIN1
PIN-dependent auxin recycling
PIN2
PIN2
Stem cell niche
PIN3
PIN4
Auxin maximum

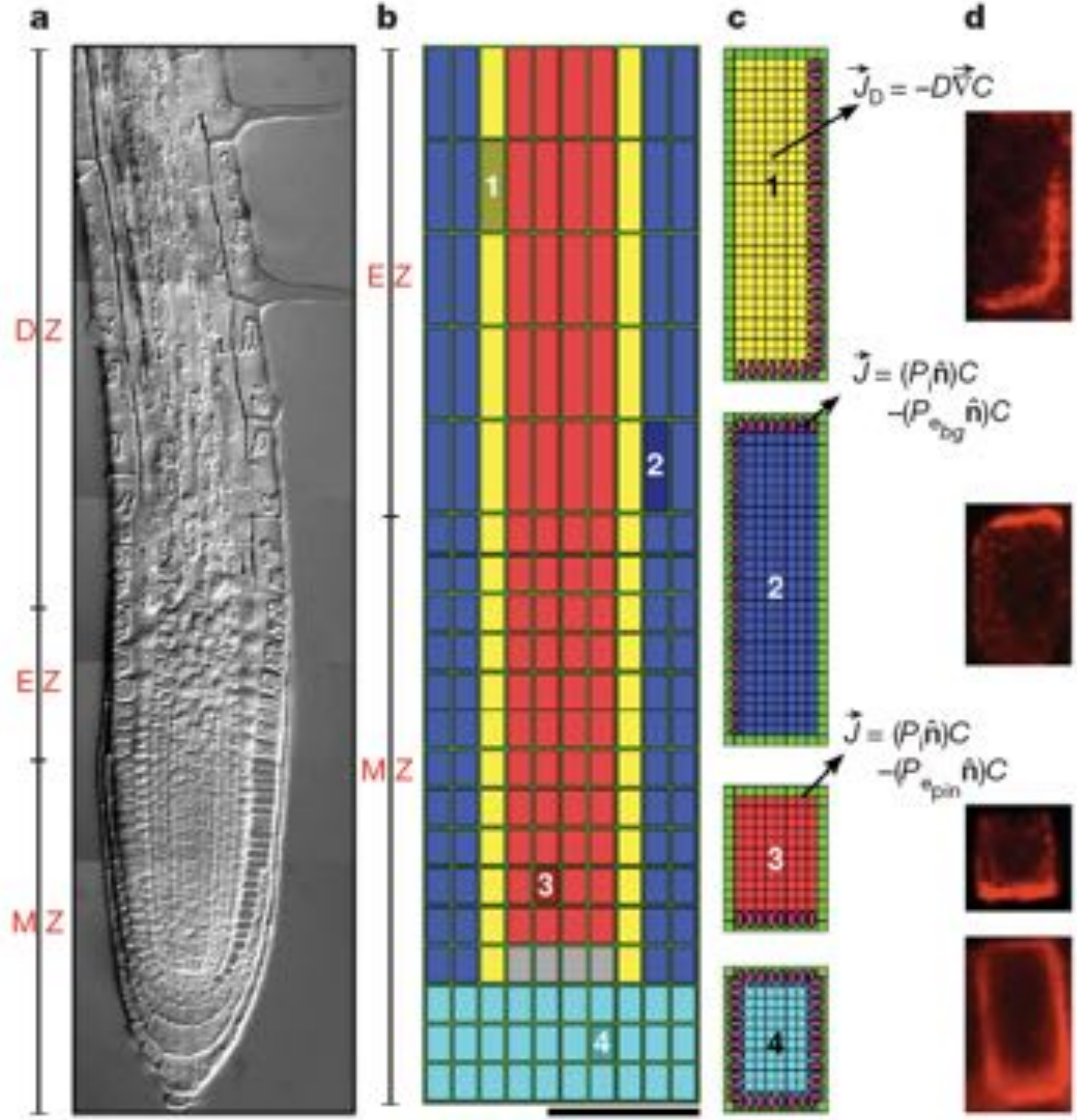


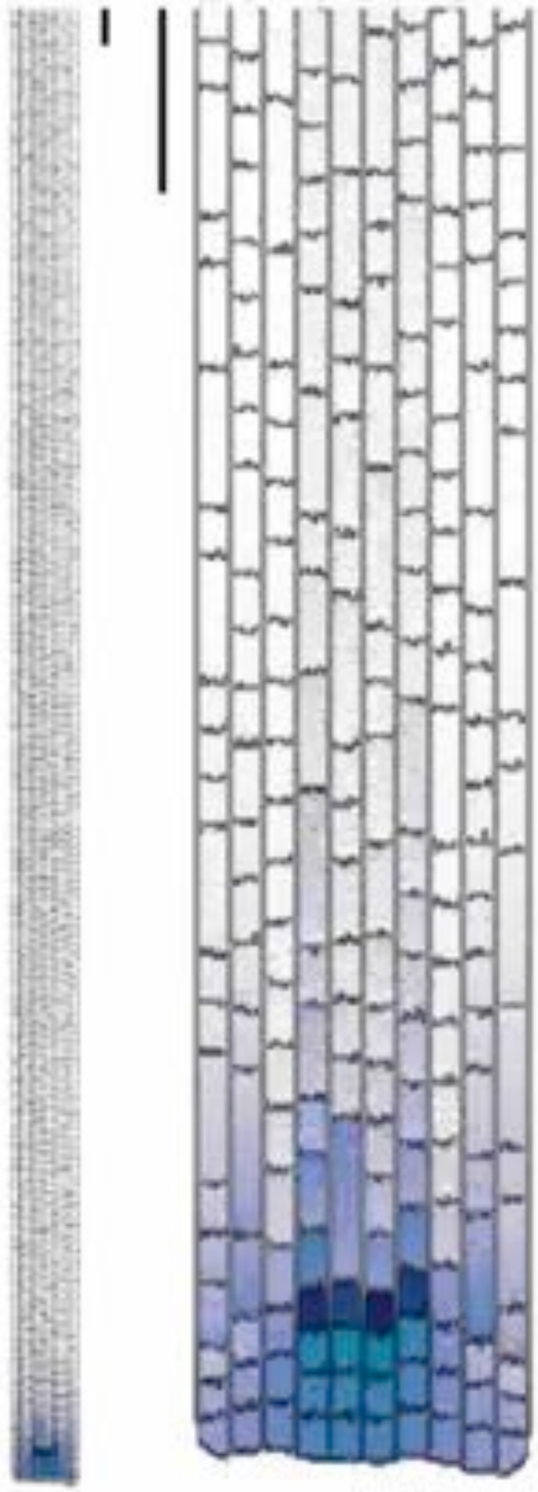
Figure 1 | Mesoscopic model for polar auxin transport. a, The *Arabidopsis* root. DZ, differentiation zone; EZ, elongation zone; MZ, meristematic zone.

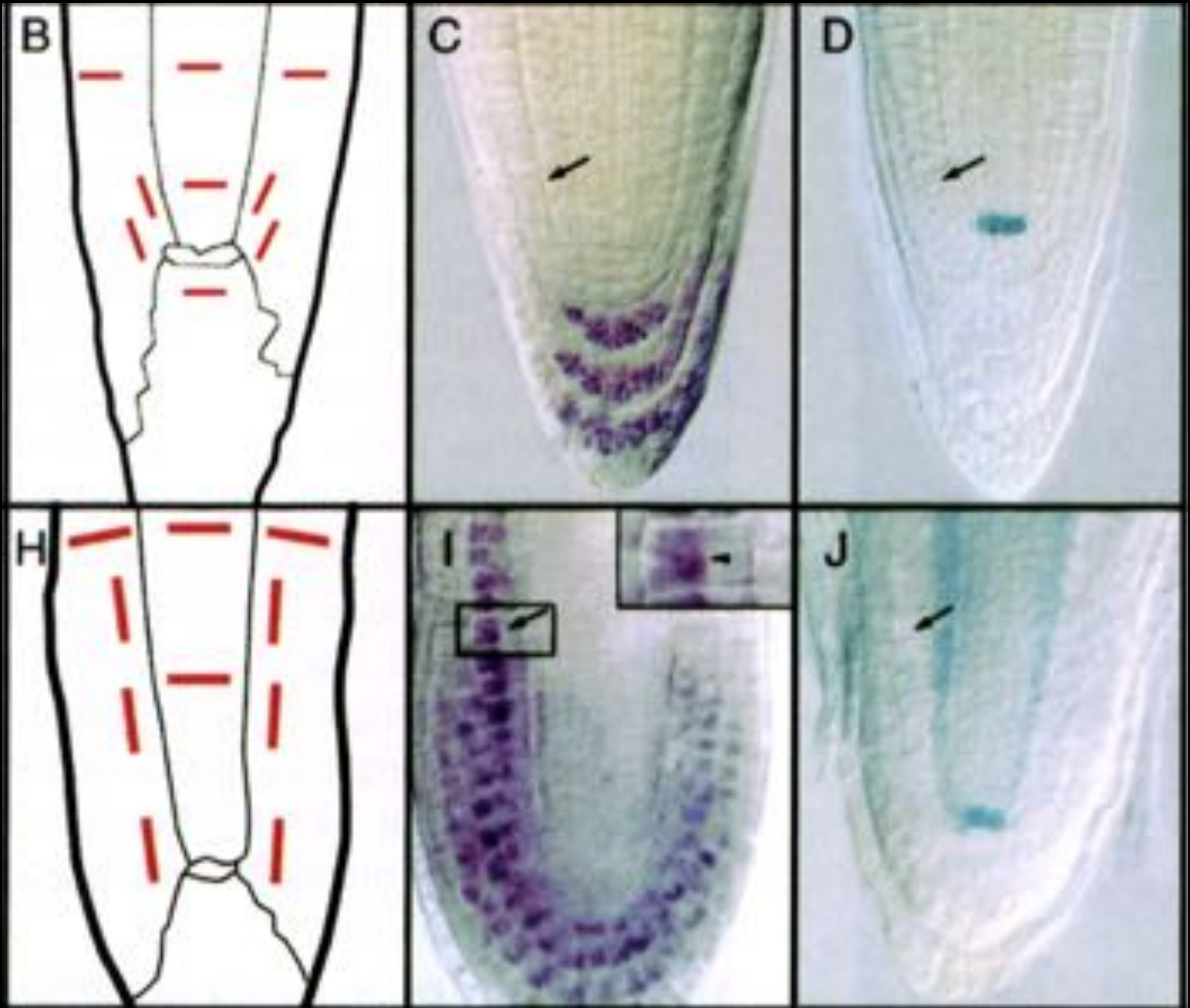
Grieneisen *et al.*

Supplementary Movie 1

Establishment of the auxin maximum in a root receiving shoot-derived auxin influx (simulation of Fig. 2b). Relative auxin concentrations according to the colour bar of Fig. 2d. Scale bar 100 μm.

-00:00

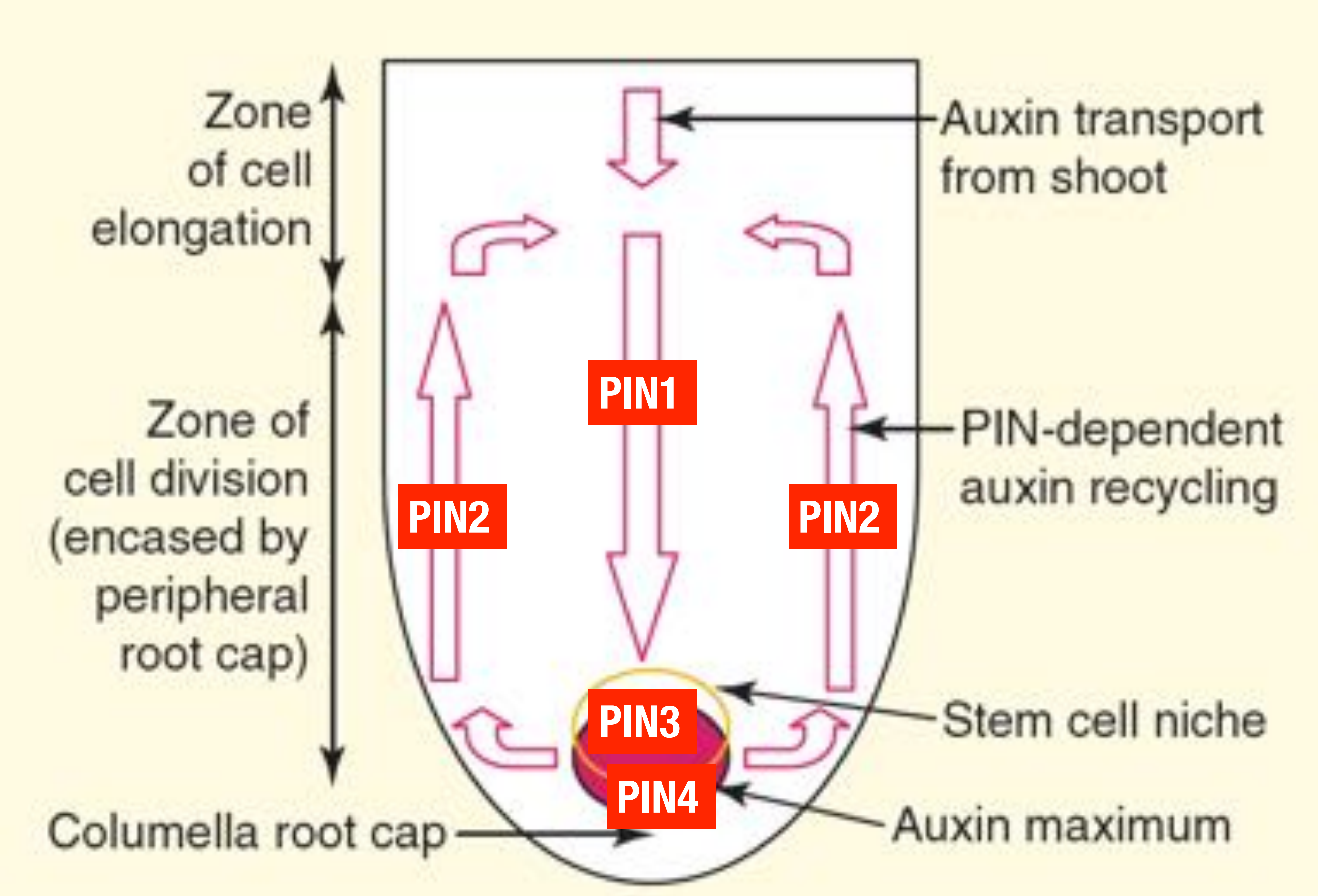




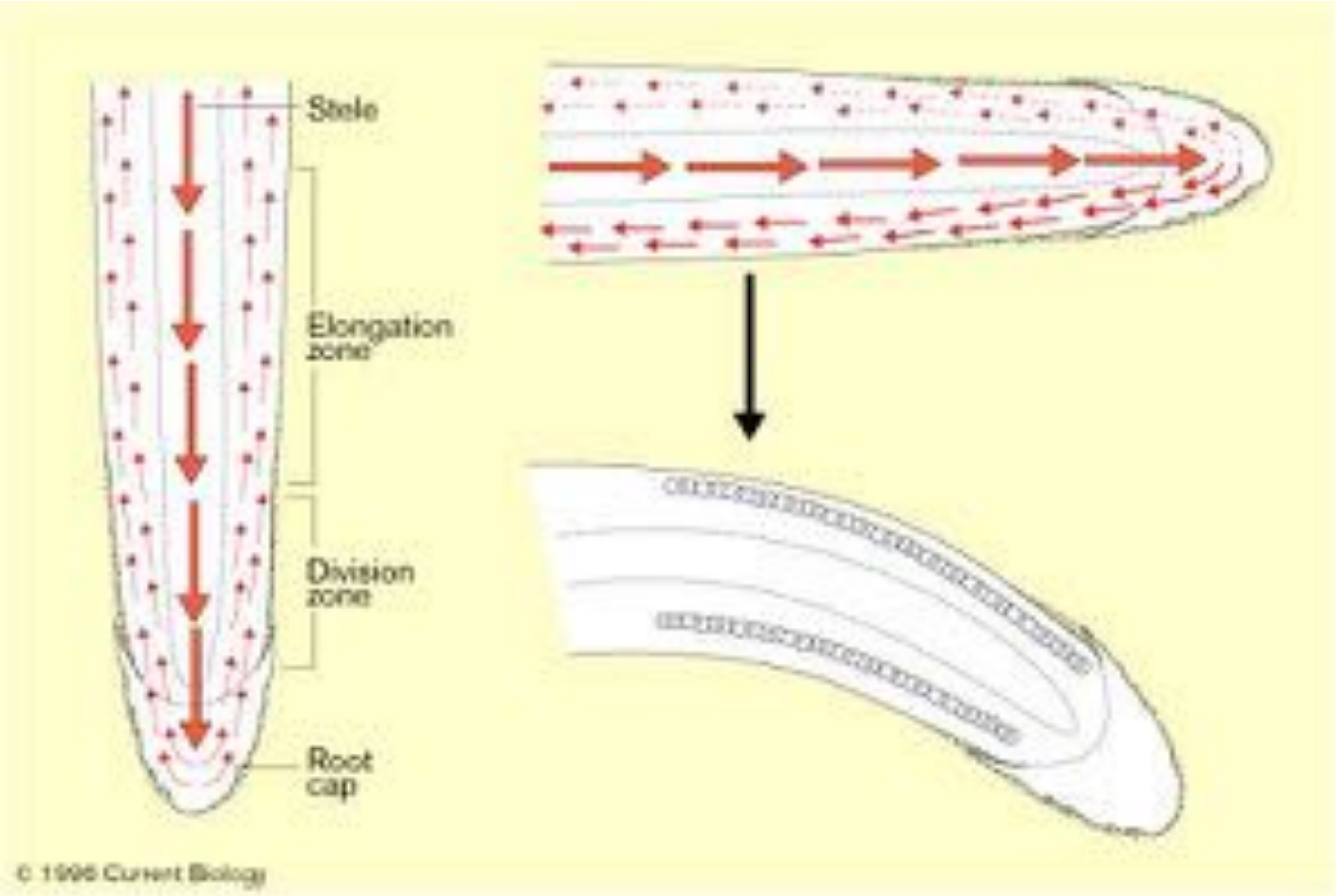
control

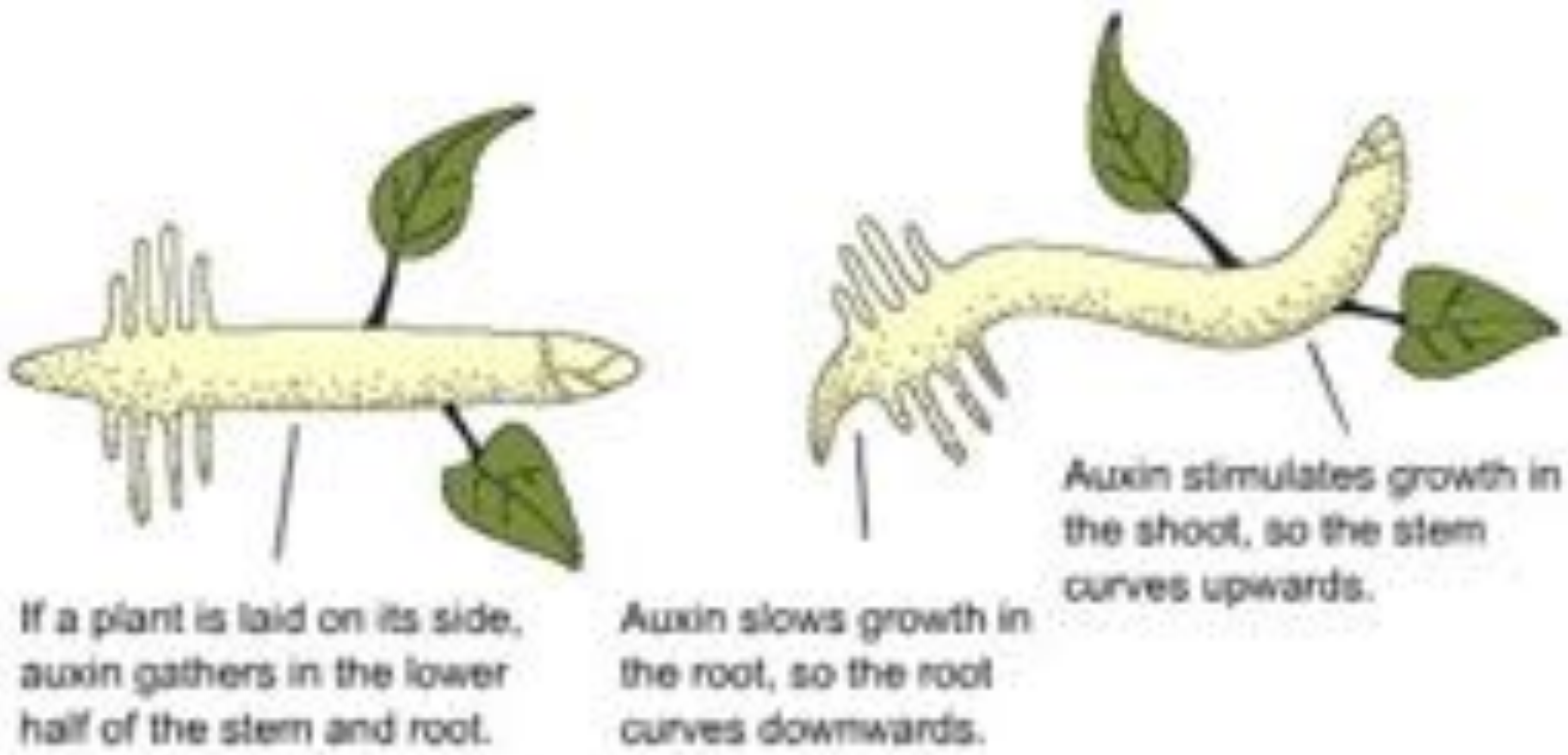
+ NPA
auxin flux
inhibitor

Polar flux of auxin controls cell fate and organisation in the Arabidopsis root


Zone
of cell
elongation
Zone of
cell division
(encased by
peripheral
root cap)
Columella root cap
Auxin transport
from shoot
PIN1
PIN2
PIN2
PIN-dependent
auxin recycling
PIN3
Stem cell niche
PIN4
Auxin maximum

Gravity and PIN3 mediated redirection of auxin flow at the root tip regulates the direction of root growth

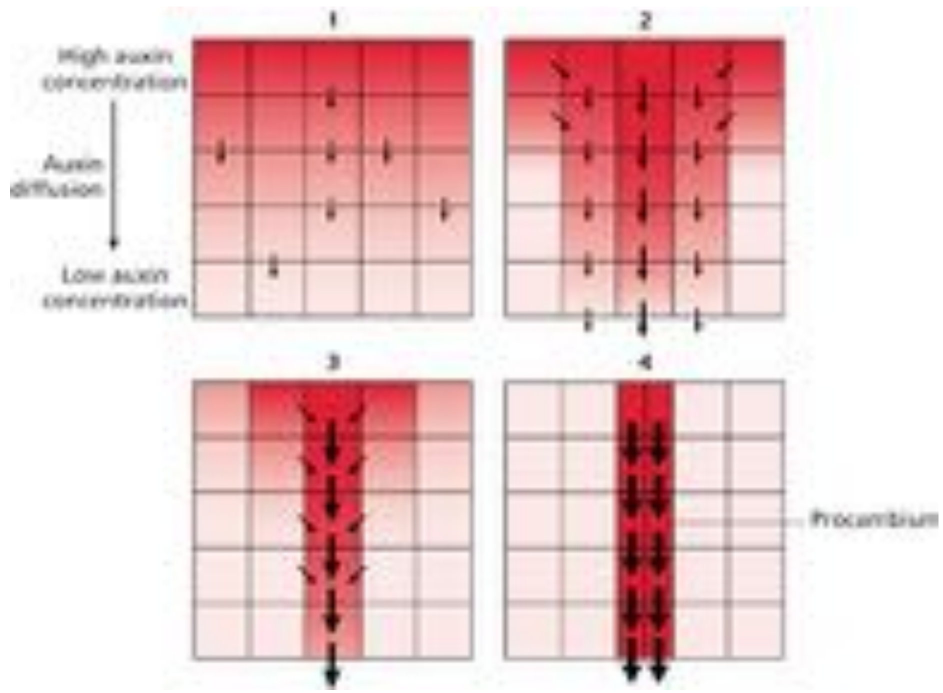



If a plant is laid on its side, auxin gathers in the lower half of the stem and root.
Auxin slows growth in the root, so the root curves downwards.
Auxin stimulates growth in the shoot, so the stem curves upwards.

Feedback-regulated traffic of auxin coordinates polar growth of plant cells

It provides both long-range coordination of plant architecture, and a short-range mechanism for controlling individual cell fates.

- Embryo polarity and outgrowth
- Root and shoot meristem outgrowth
- Vascular development



"Canalisation" of auxin flow

Traffic of auxin regulates many aspects of plant growth and development