



Bachelor 2: Plant physiology

Lesson 7:

PLANT HORMONE

Instructor: LE Thi Van Anh

le-thi-van.anh@usth.edu.vn

Learning outcome

By the end of this course, students are able to:

- Give definition of “hormone” and the general function of hormone
- List major hormones representing in plant
- Present the biosynthesis of hormone
- Analyze the biological function of auxin, GA and cytokinin
- Describe the process of auxin transport in plant
- Study by them selves the biological of some other hormone:

Concept

Greek *horman* = **to stimulate**

Hormone = Substance or chemical that is transported and causes specific physiological effects

Plant hormones

= phytohormone (phyto = plant): hormone in plant

= plant growth regulators: factors that regulate the growth and development of plant

Features:

- Regulate growth and development
- Mobile throughout plant
- Environment and stress response

Concept: major plant hormones

Auxin – Greek: to grow or increase

Cytokinin – cytokinesis (cell division)

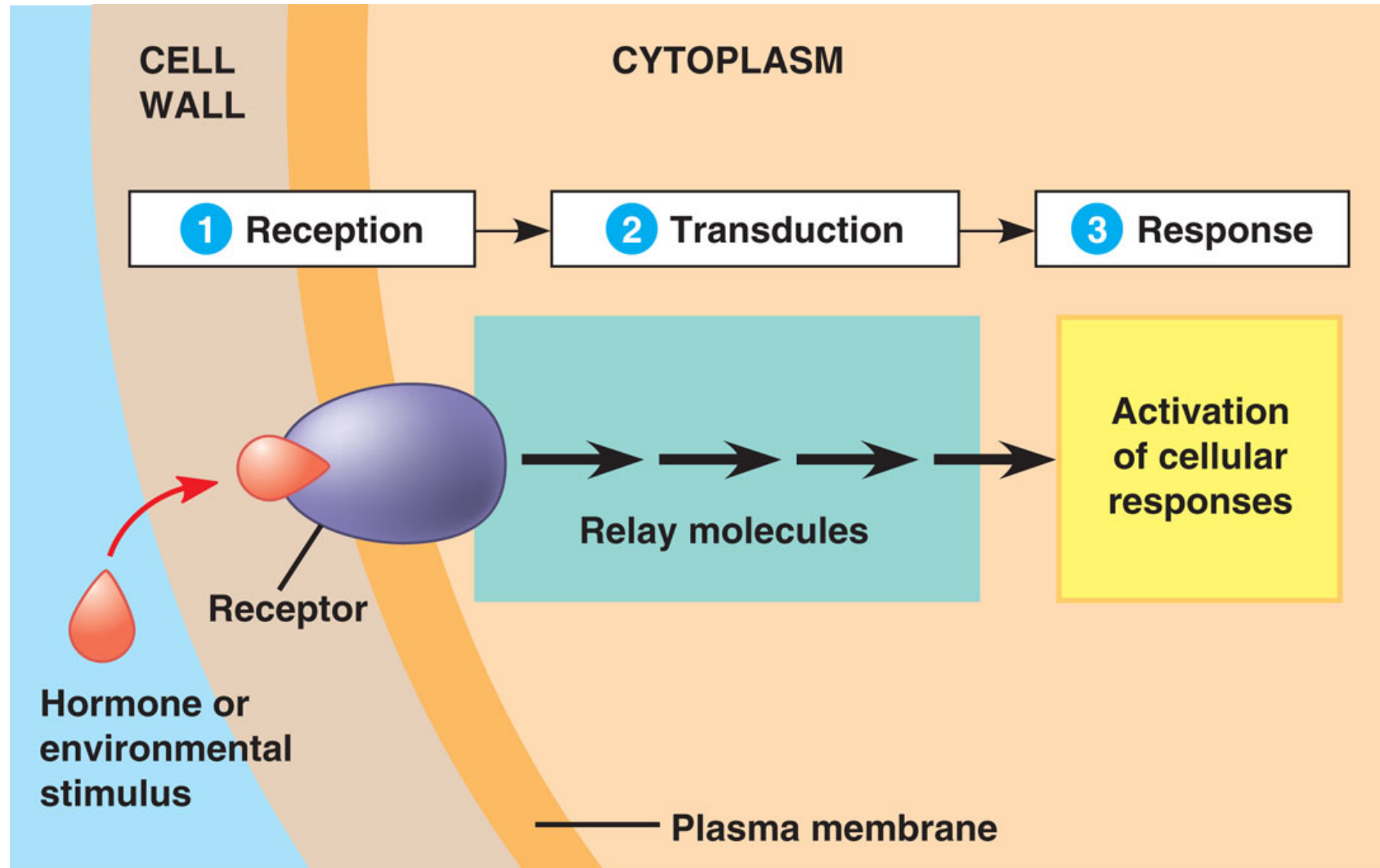
Gibberellic acid – pathogen *Gibberella*

Ethylene – chemical brother to ethanol

Abscisic acid – abscission

Brassinosteroids – derived from *Brassica* spp.

Concept: transduction pathways



Concept: questions to answer...

HOW were they discovered?

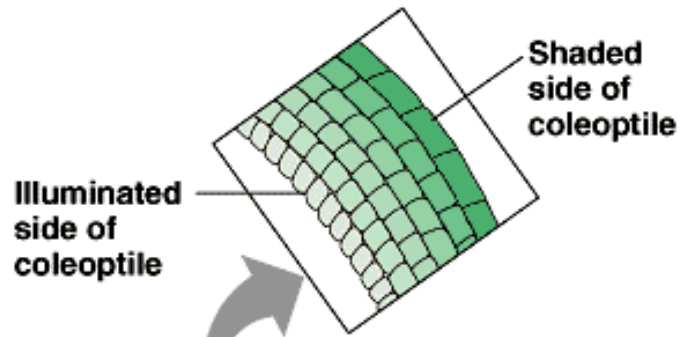
WHERE are they synthesized?

WHAT are their biological functions

AUXIN: discovery

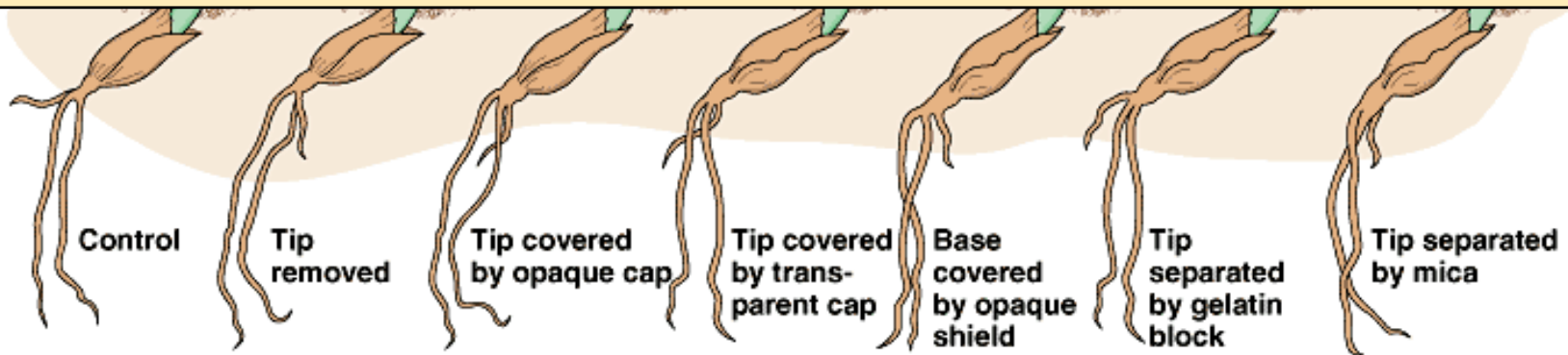
Some sorts of signal:

- **Are produced in the tips**
- **Travel to elongation zone causing curvature**



Light

In 1930s: identification of **Indole-3-acetic acid (IAA)**
as the natural auxin

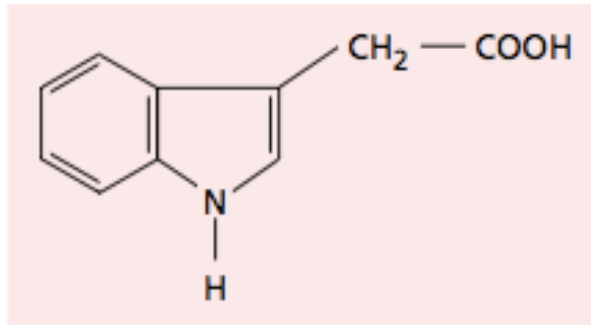


Darwin and Darwin (1880)

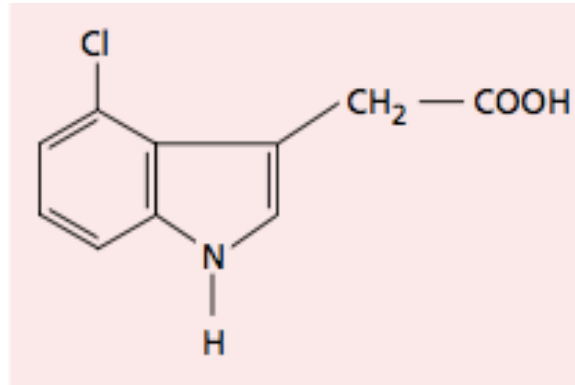
Boysen-Jensen (1913)

7

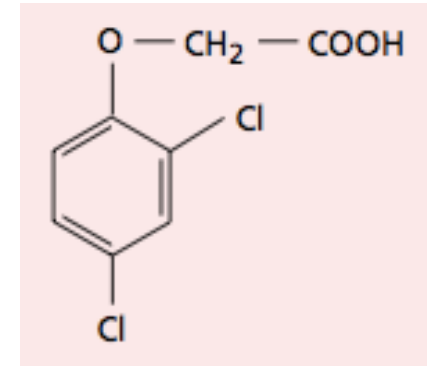
AUXIN: chemical structure



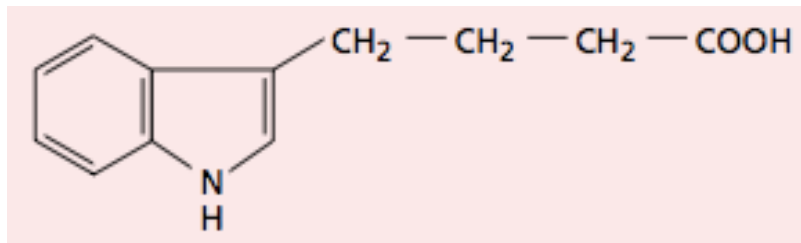
Indole-3-acetic acid
IAA



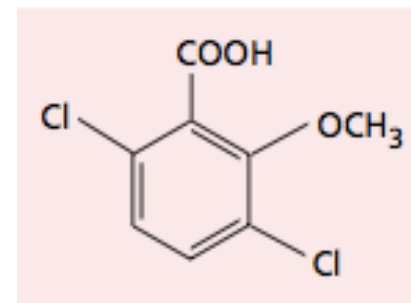
4-chloroindole-3-acetic acid
4-Cl-IAA



2,4-Dichlorophenoxyacetic acid
2,4 D



Indole-3-butyric acid
IBA

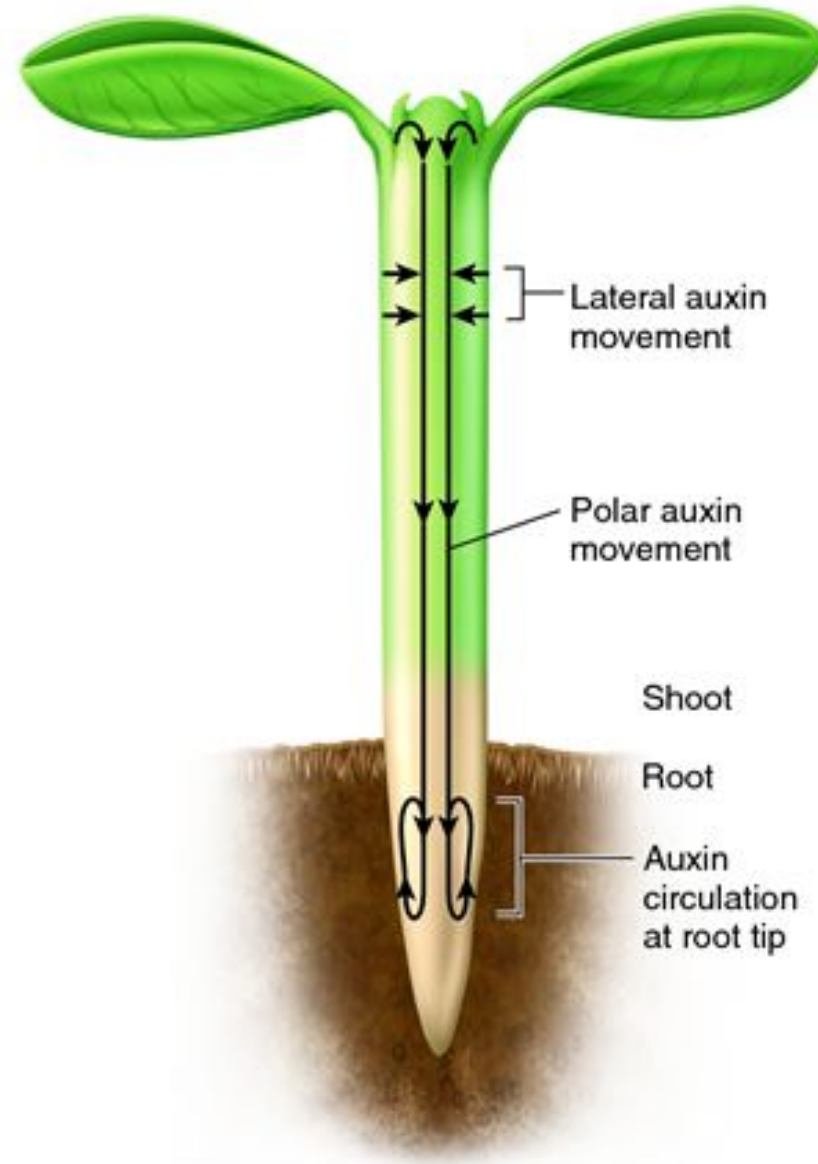


2-Methoxy-
3,6-Dichlorobenzoic acid
dicamba

AUXIN: site of synthesis

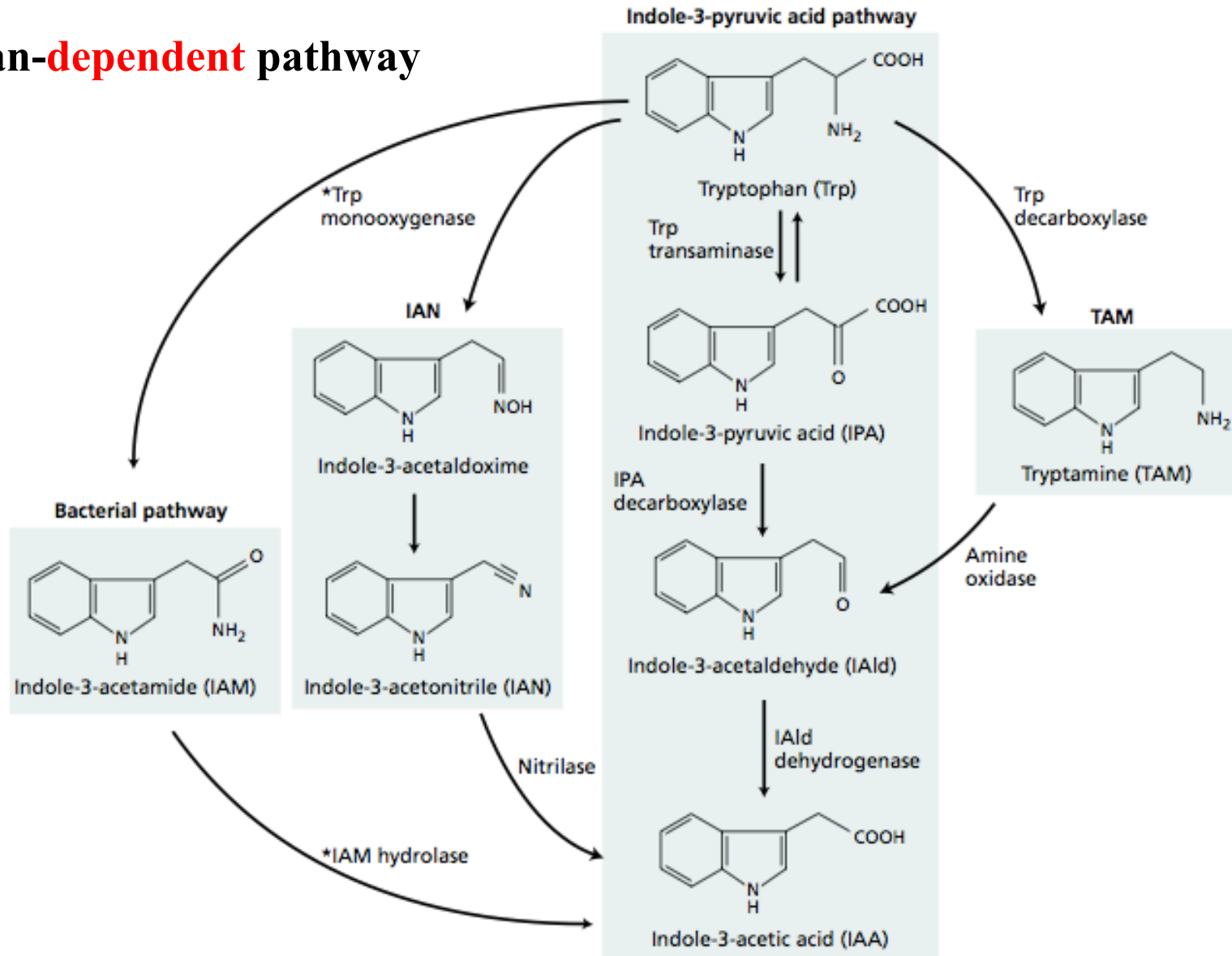
Synthesis in rapidly dividing and growing tissues:

- SAM
- Young leaves
- Developing fruits and seeds



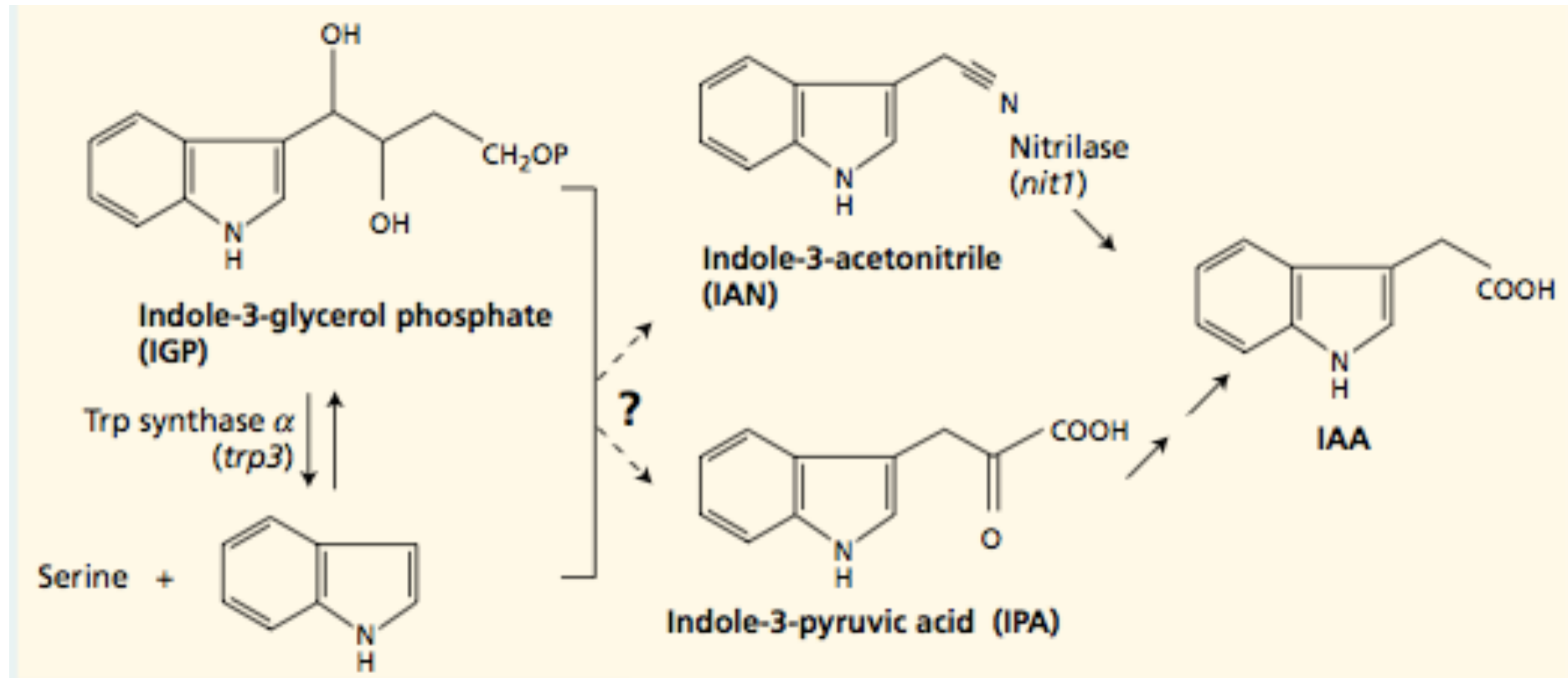
AUXIN: biosynthesis pathway

Tryptophan-dependent pathway



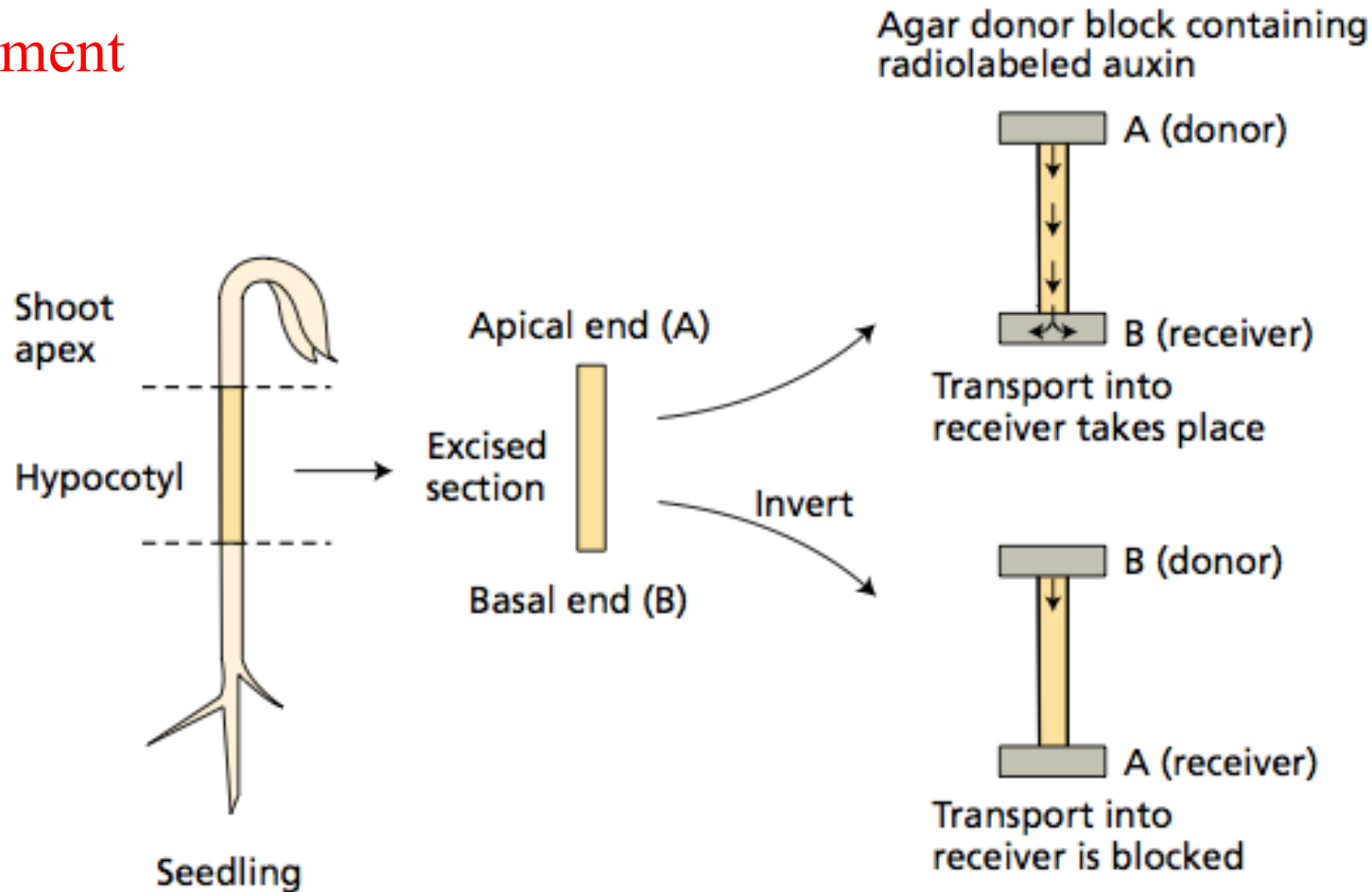
AUXIN: biosynthesis pathway

Triptophan-independent pathway



AUXIN: polar transport

Experiment



Polarity of transport is independent of the orientation of plant tissues:

Transport polarly from “source” to “sink”

AUXIN: polar transport

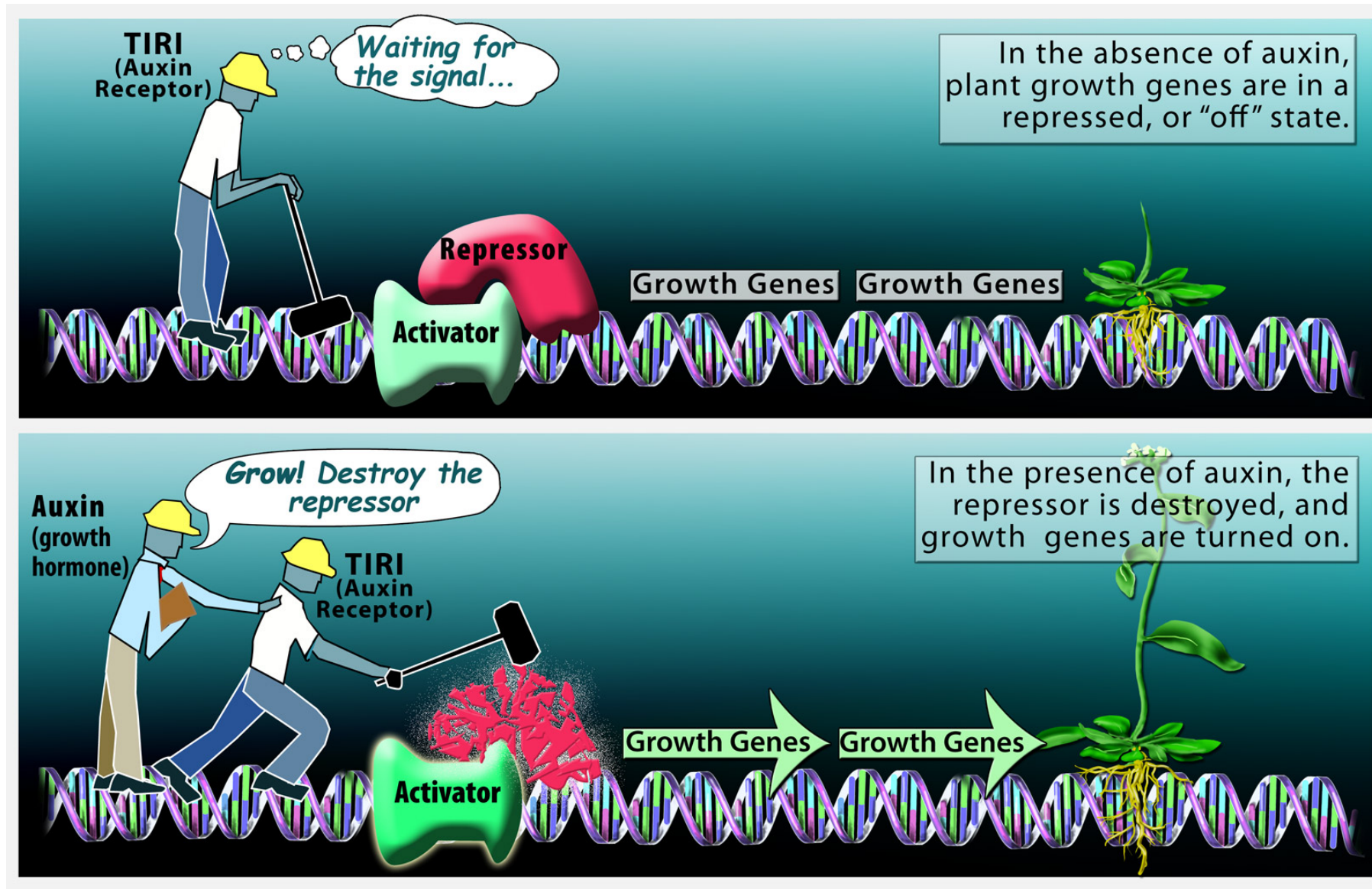


The root always form at the basal end.

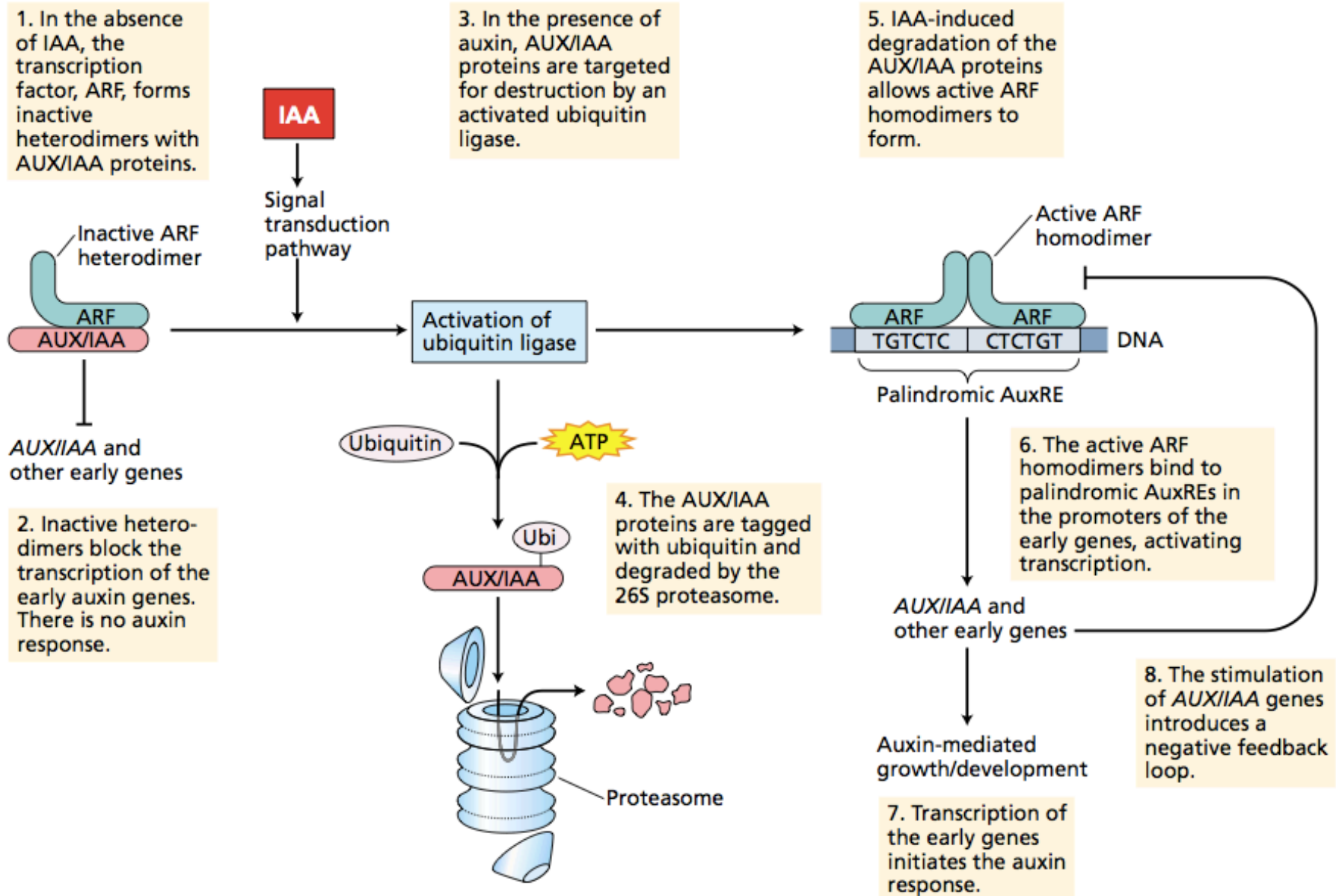


Polar auxin transport is **independent of gravity**

AUXIN: signal transduction pathways



AUXIN: signal transduction pathways



AUXIN: Biological function

- Promote cell elongation
- Promote Shoot growth
- Inhibit root growth (at high concentration)
- Regulate tropism: phototropism, gravitropism
- Regulate apical dominance
- Promote formation of lateral/adventitious root
- Promote fruit development
- Other: delay the onset of leaf abscission; induce vascular differentiation

Cell elongation in response to auxin

Expansins (active at low pH) cleave cellulose microfibrils from polysaccharides. Exposed polysaccharides now accessible to enzymes.

Cross-linking cell wall polysaccharides

Cell wall enzymes

Expansin

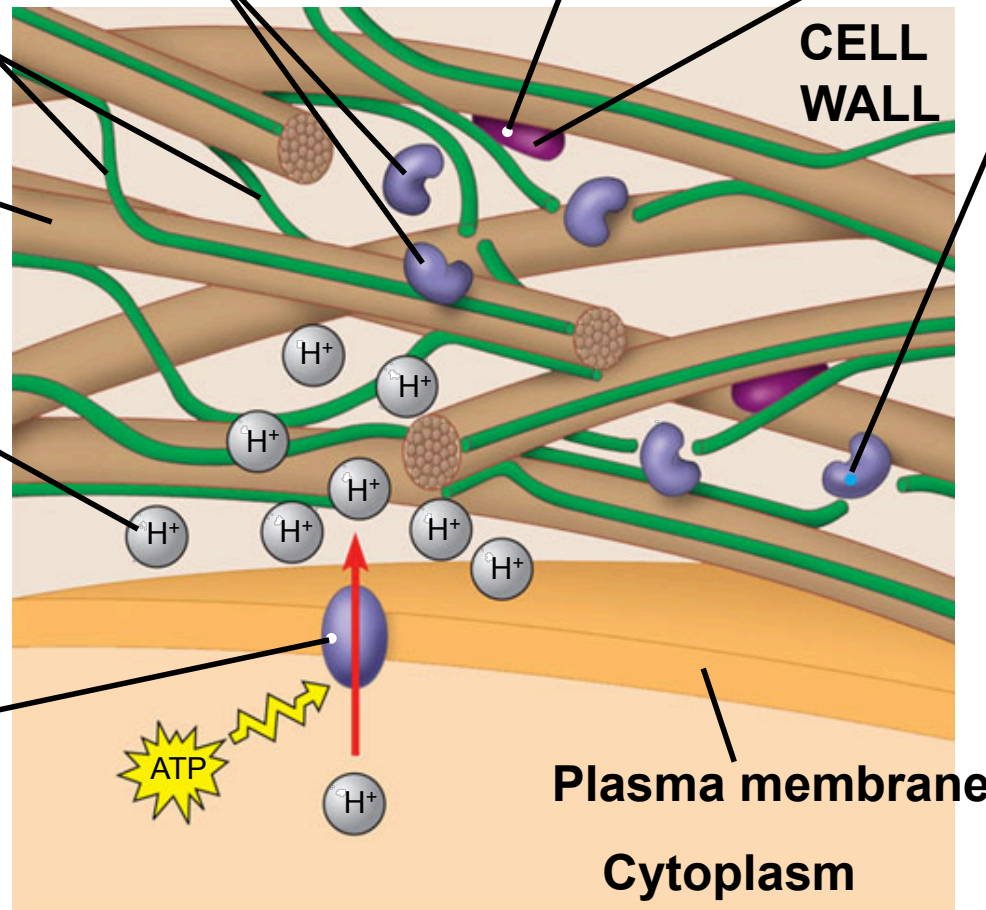
CELL WALL

Microfibril

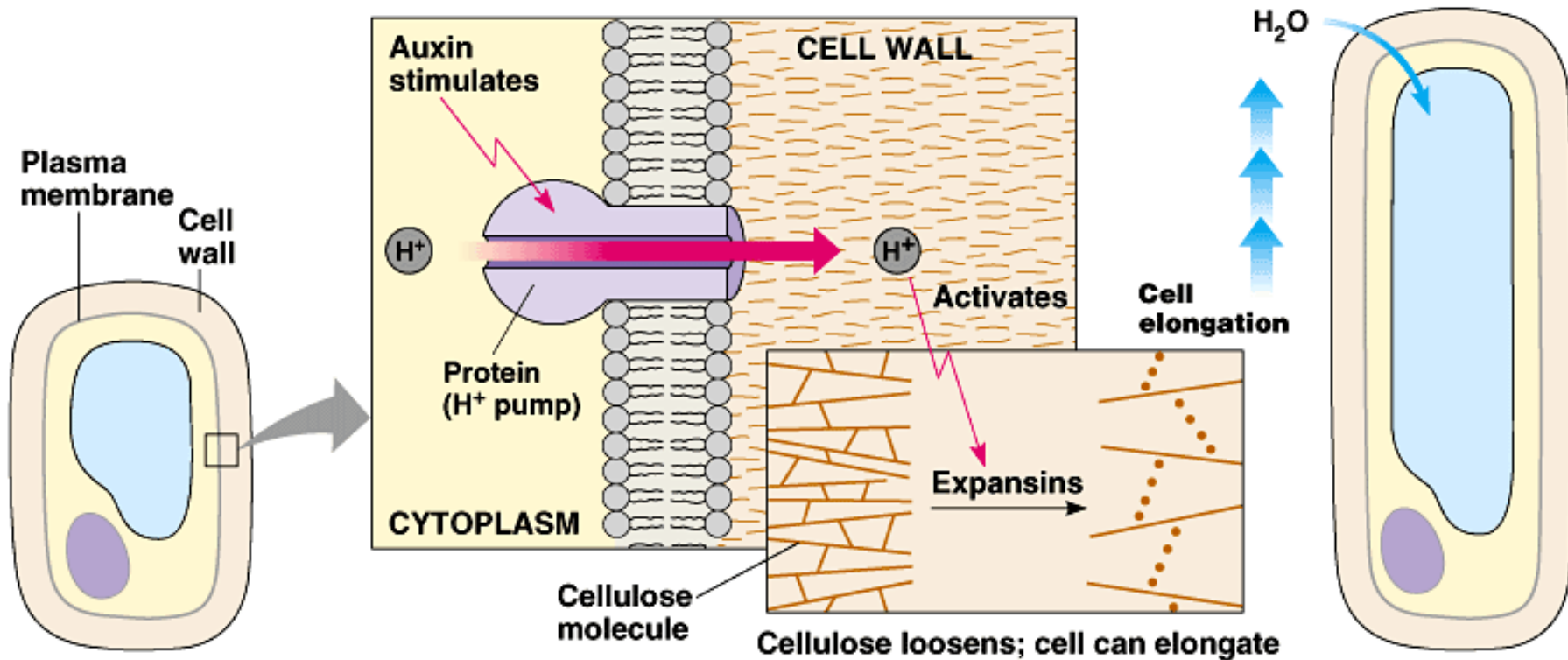
Enzymatic cleavage of polysaccharides allows microfibrils to slide. Cell wall can extend. Turgor causes the cell to expand.

Cell wall becomes acidic.

Auxin increases activity of proton pumps.



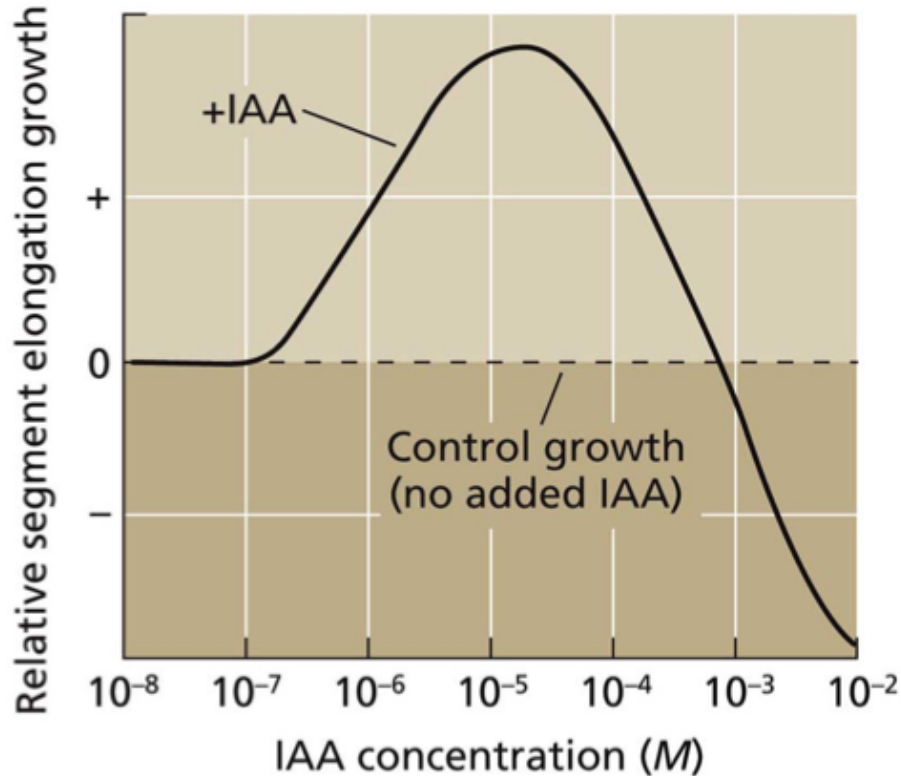
Cell elongation in response to auxin



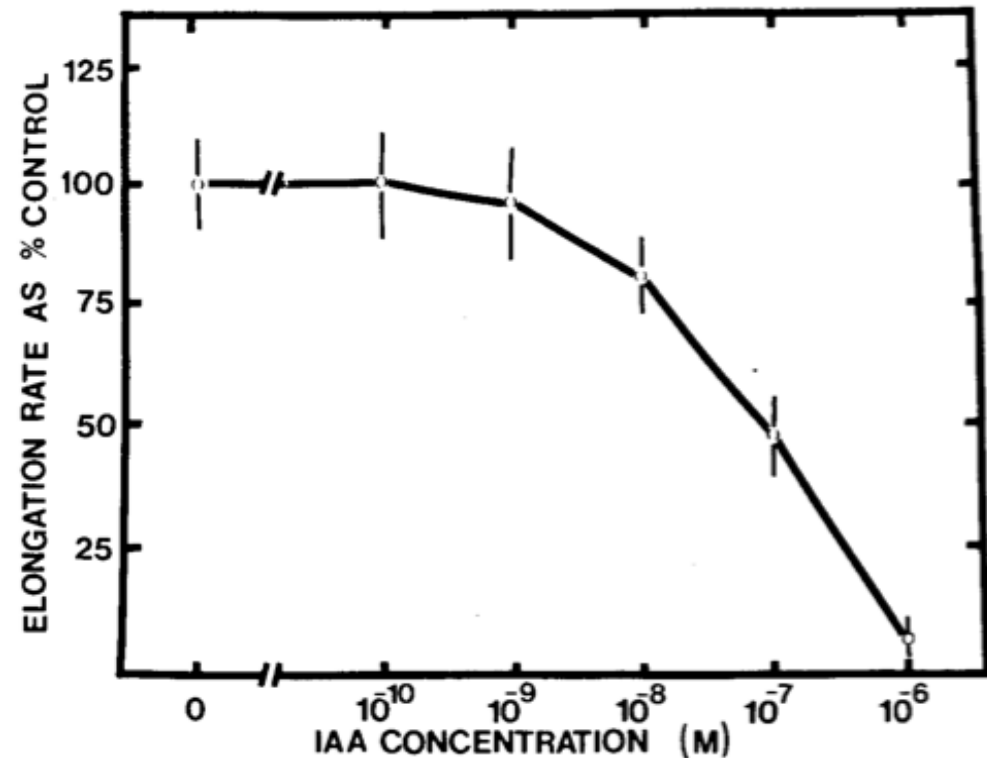
Auxin cause the cellulose loosened, thus the cell can elongate

Auxin promote shoot and inhibit root growth

IAA induction on shoot growth



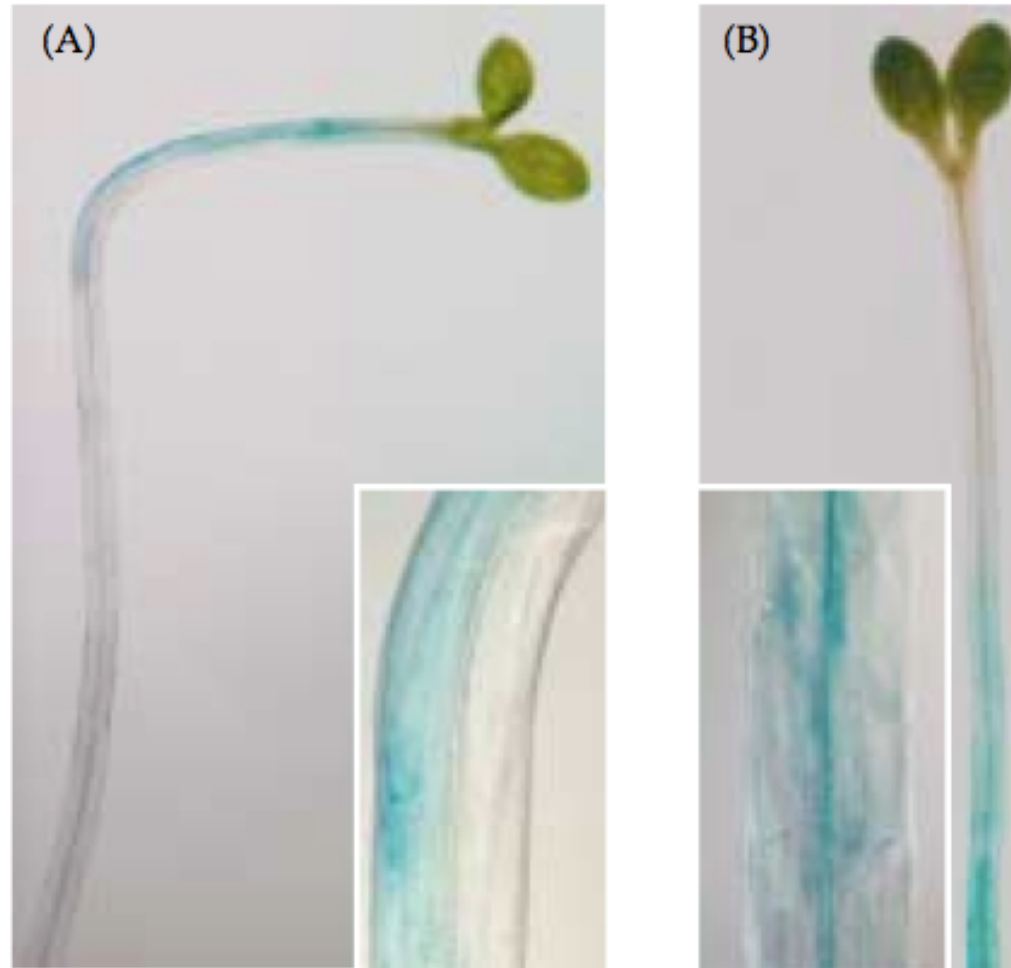
IAA induction on root growth



AUXIN: induce plant phototropism

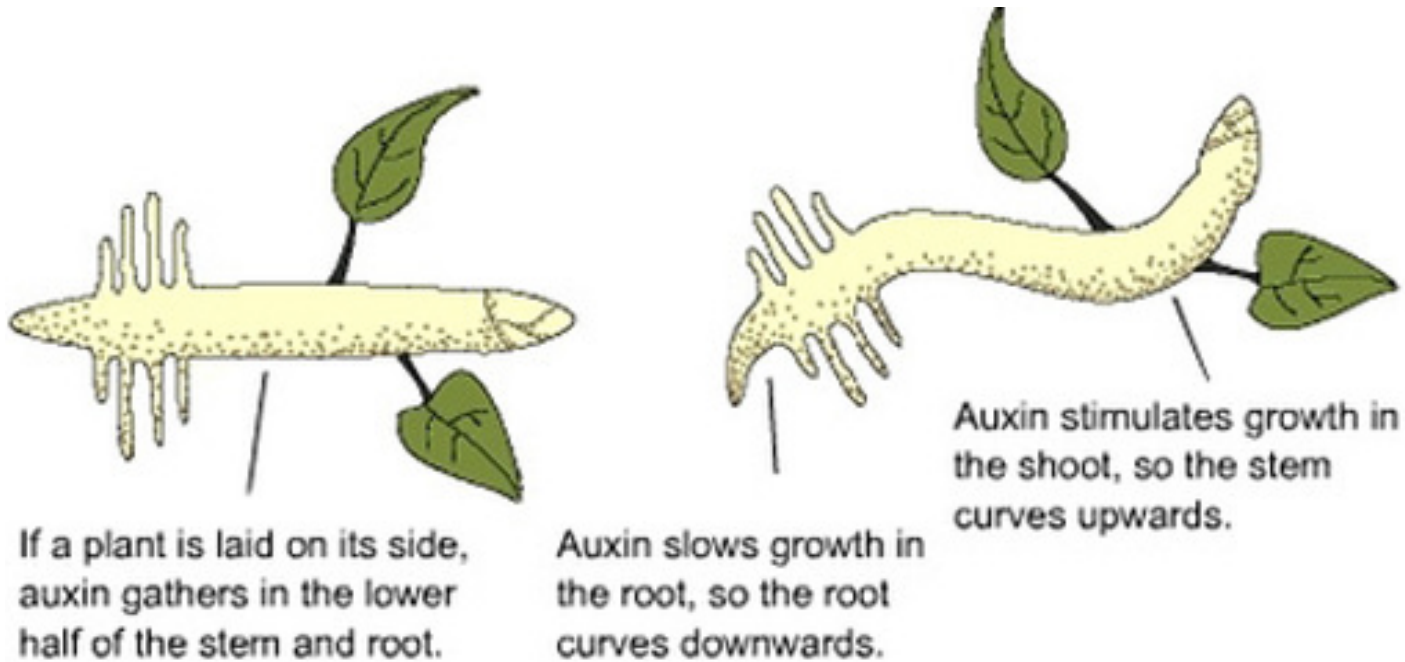
Transformed plant
with DR5::GUS

A: no NPA treatment
B: with NPA treatment



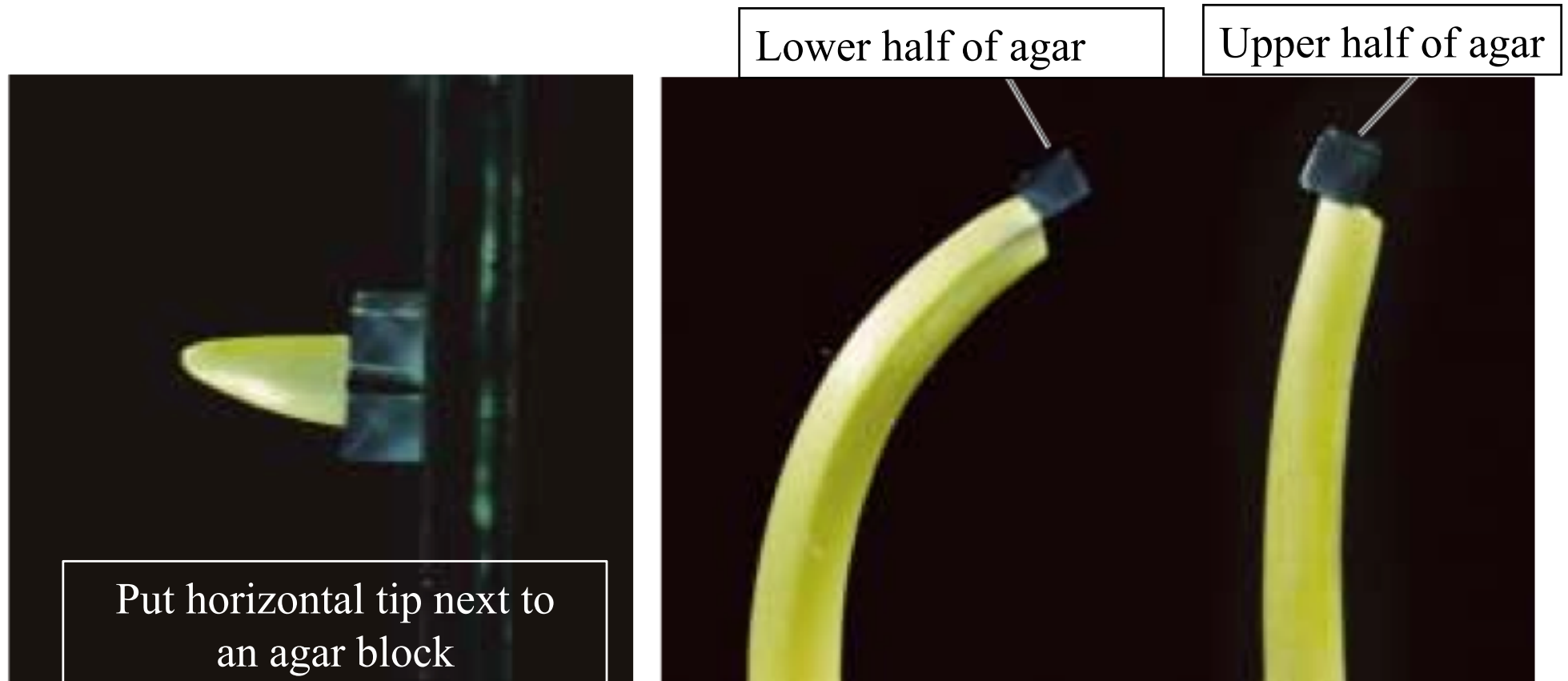
There is lateral redistribution of auxin during phototropism

AUXIN: induce gravitropism



Root has positive gravitropism
Shoot has negative gravitropism

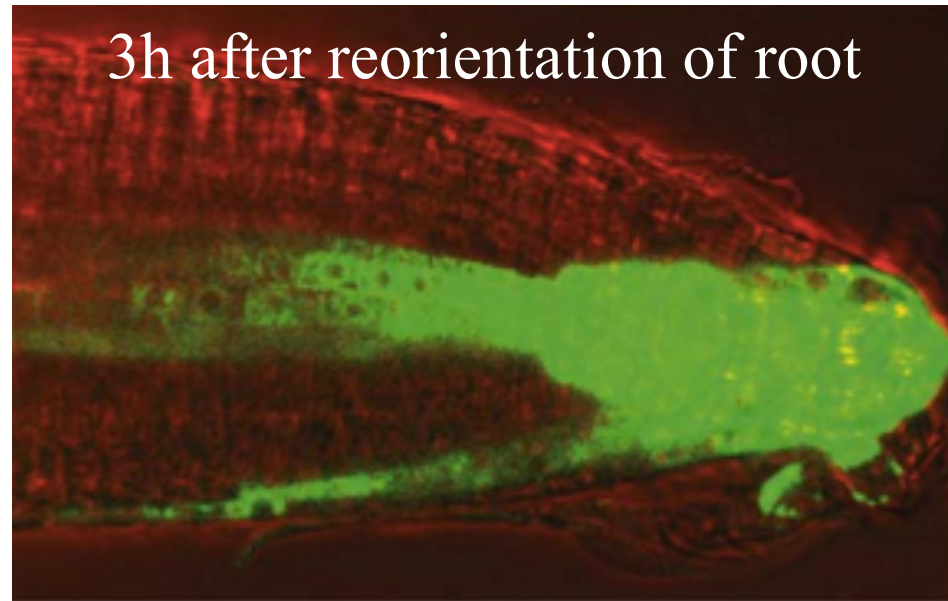
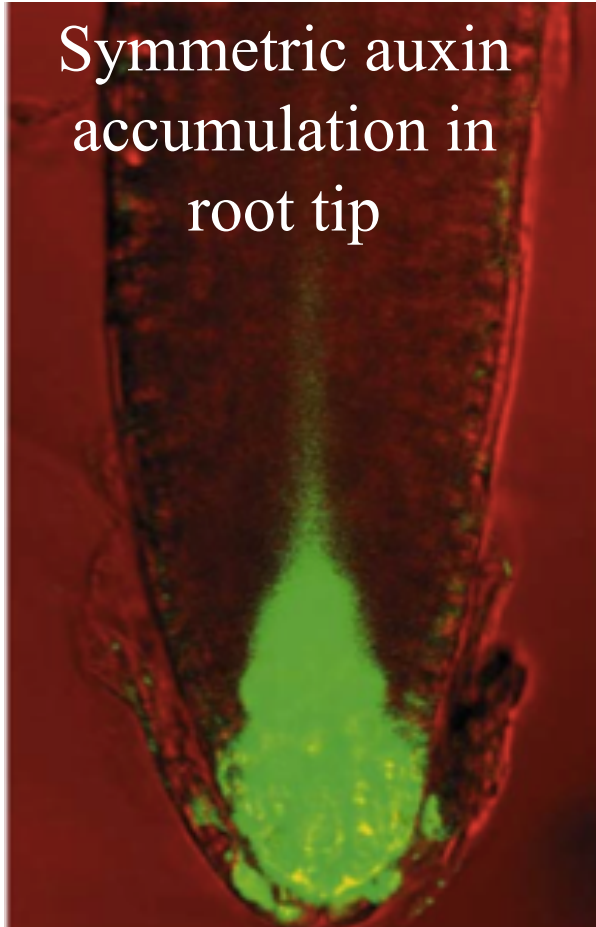
AUXIN: induce shoot negative gravitropism



Auxin is transported to the lower side of a horizontally oriented oat coleoptile tip

AUXIN: induce root positive gravitropism

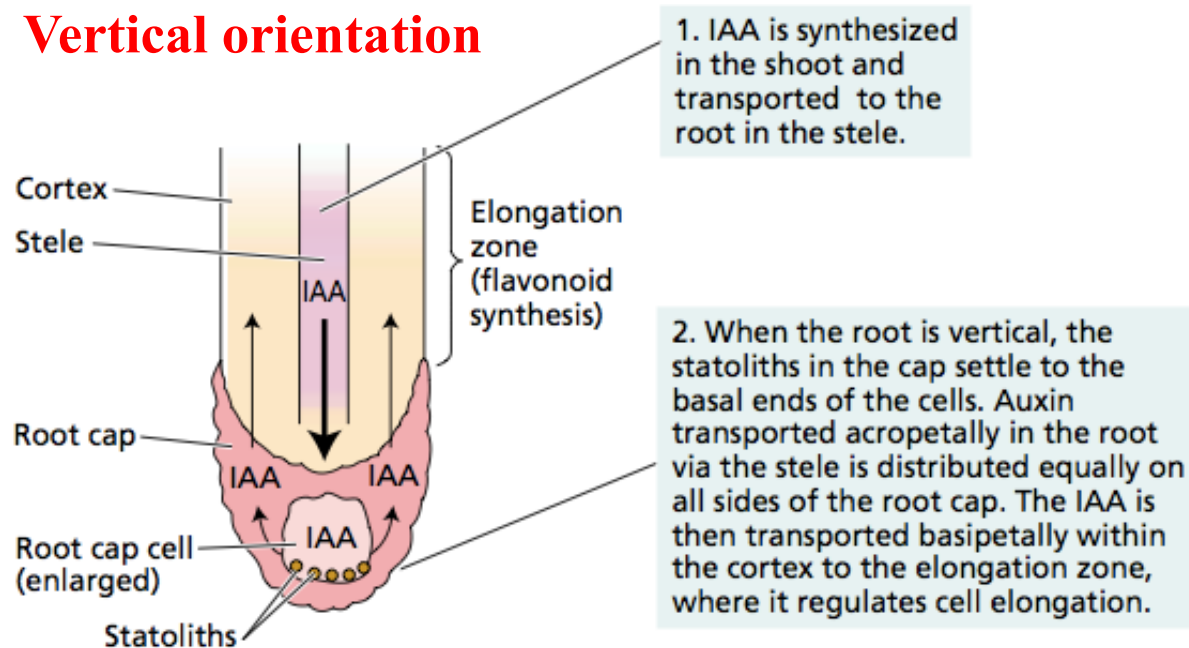
Transgenic plant DR5::GFP



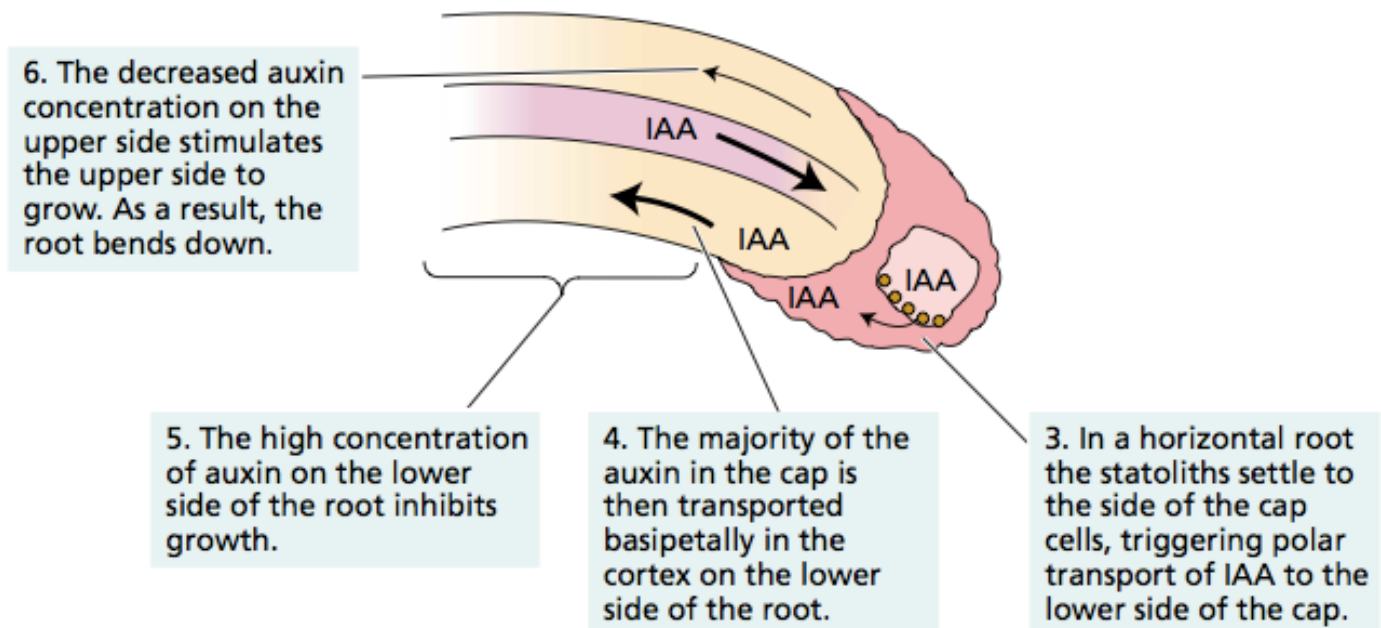
Auxin is redistributed to the lower side after reorientation of root

AUXIN: induce root positive gravitropism

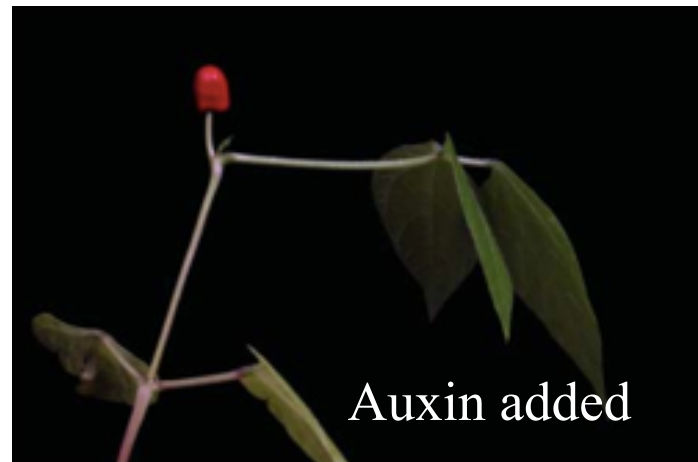
Vertical orientation



horizontal orientation

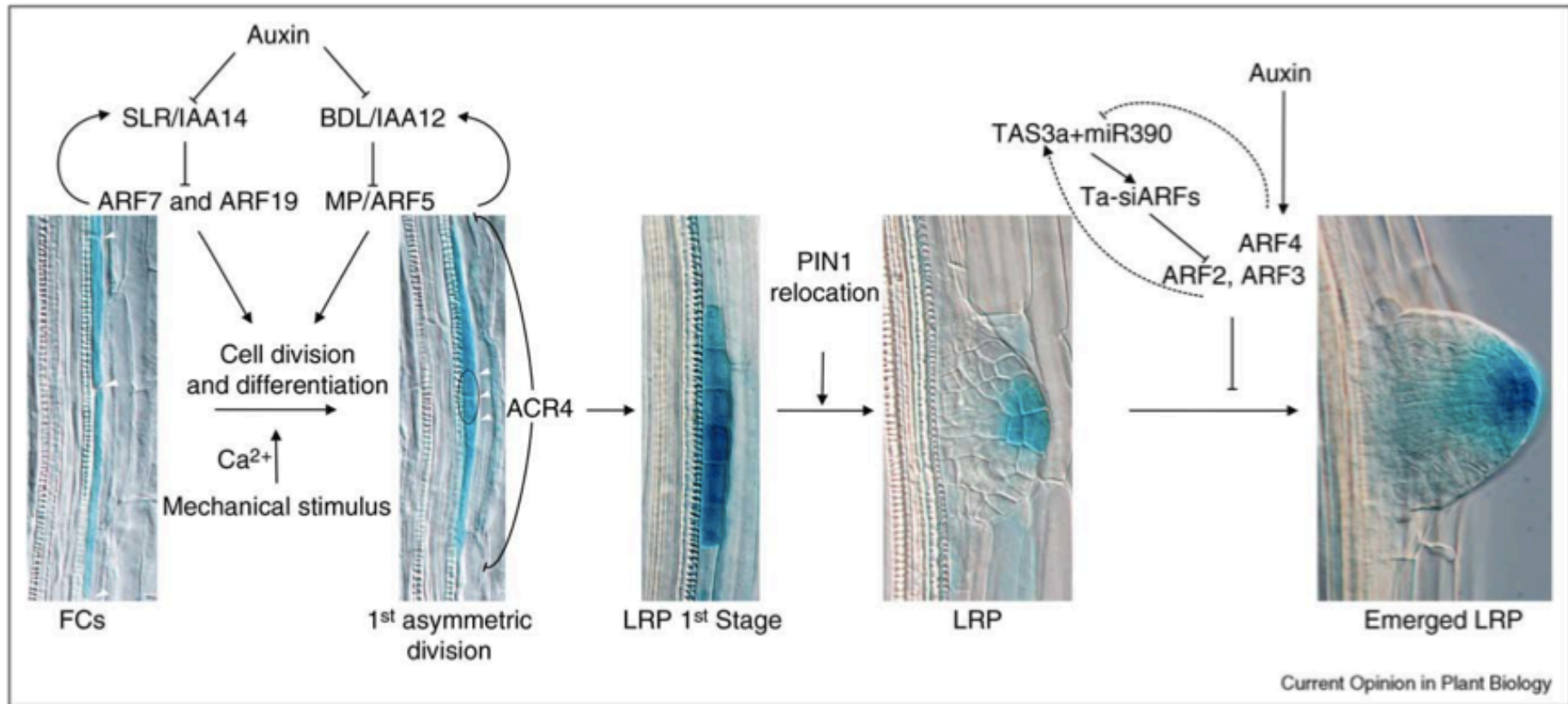


Auxin regulate apical dominance



The growing apical bud inhibits the growth of lateral buds
This is regulated by auxin

Auxin promote the formation lateral/adventitious root

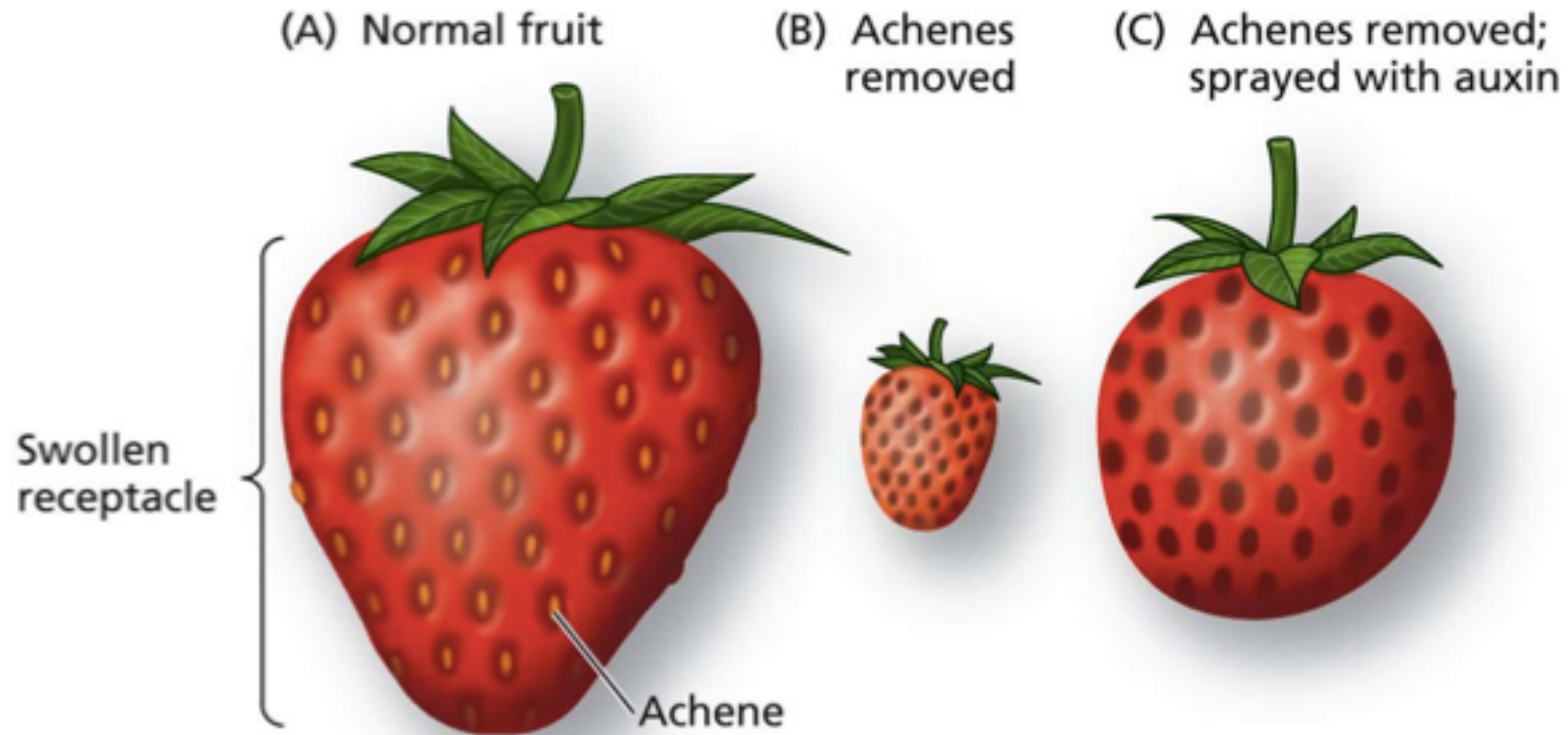


Transgenic Arabidopsis with DR5::GUS

At the 1st stage: auxin is located in ground meristem from which lateral root primordium initiate.

Then auxin is translocated toward the root tip of LR

Auxin promote fruit development



Achenes produce auxin
that regulate receptacle develop into fruit

GIBERRELLIN: discovery

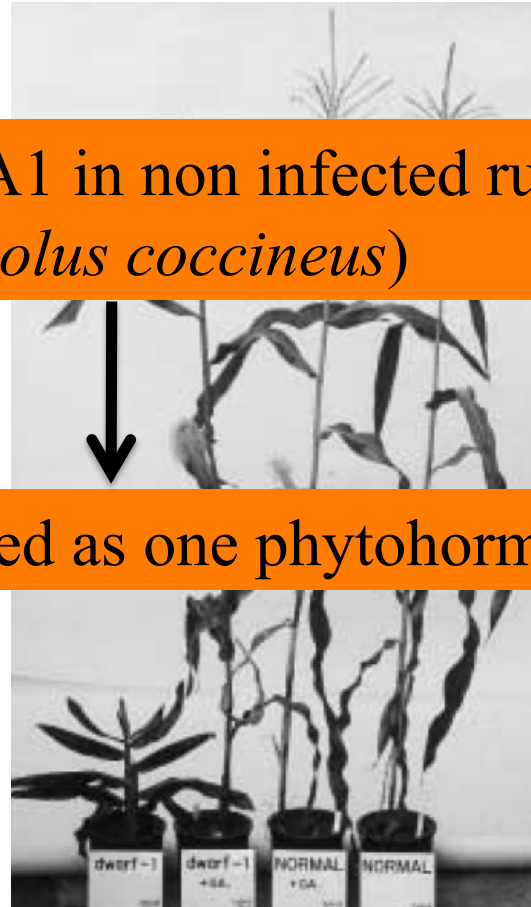


1958: Isolation of GA1 in non infected runner bean (*Phaseolus coccineus*)

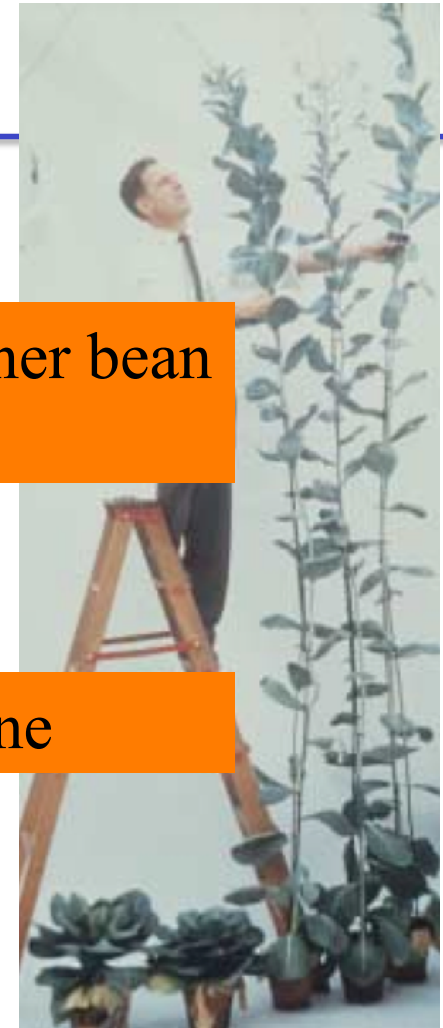
1930: Bakanae disease caused by *Giberella fujikuroi*. tall stem, less grain production

GA is considered as one phytohormone

1950: Identification Giberrellin A (GA) in culture filtrate
Separation in to 3 molecules
GA1, GA2 and GA3

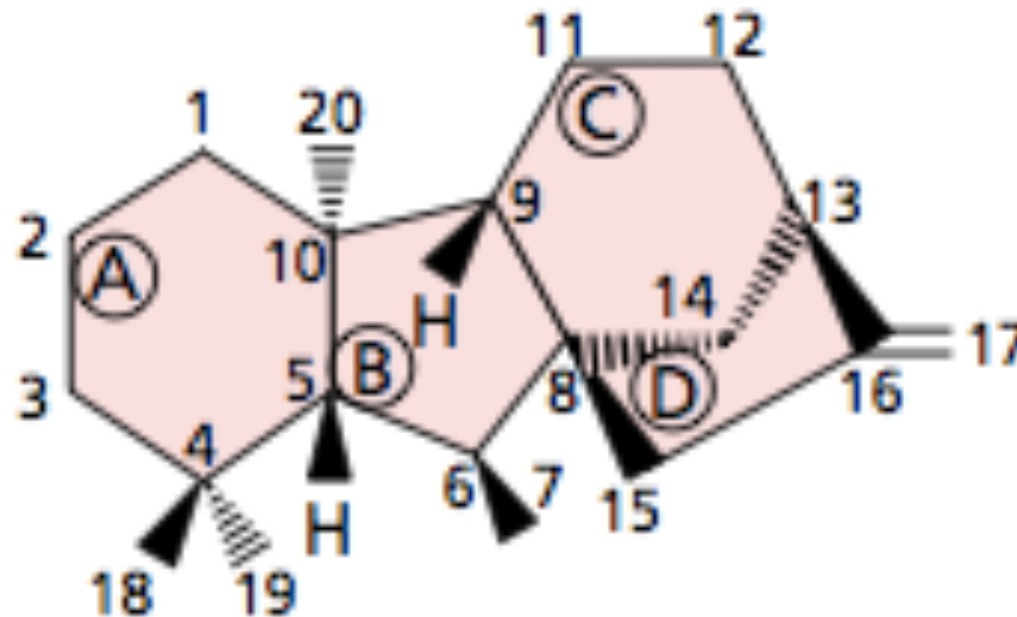


GA1 stimulate stem elongation in the dwarf mutant



GA3 induce internodes growth and flowering in cabbage

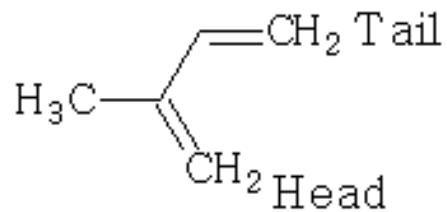
GIBERRELLIN: chemical structure



GAs are diterpenoid, formed from 4 isoprenoid unit
Each unit consisting of 5 carbons.

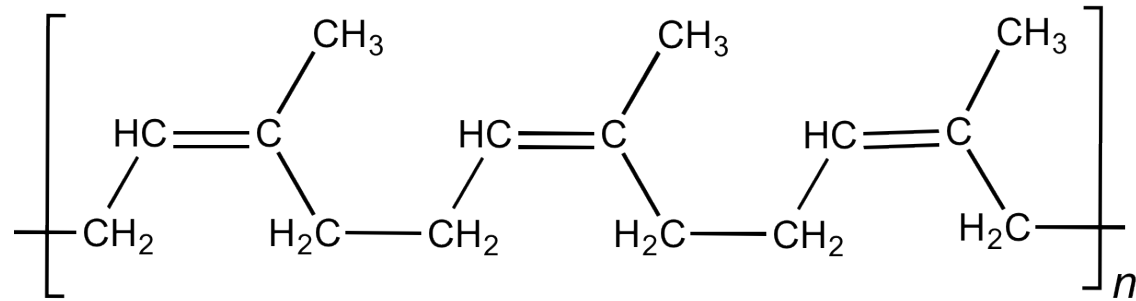
GIBERRELLIN: chemical structure

Isoprene (C₅ H₈)

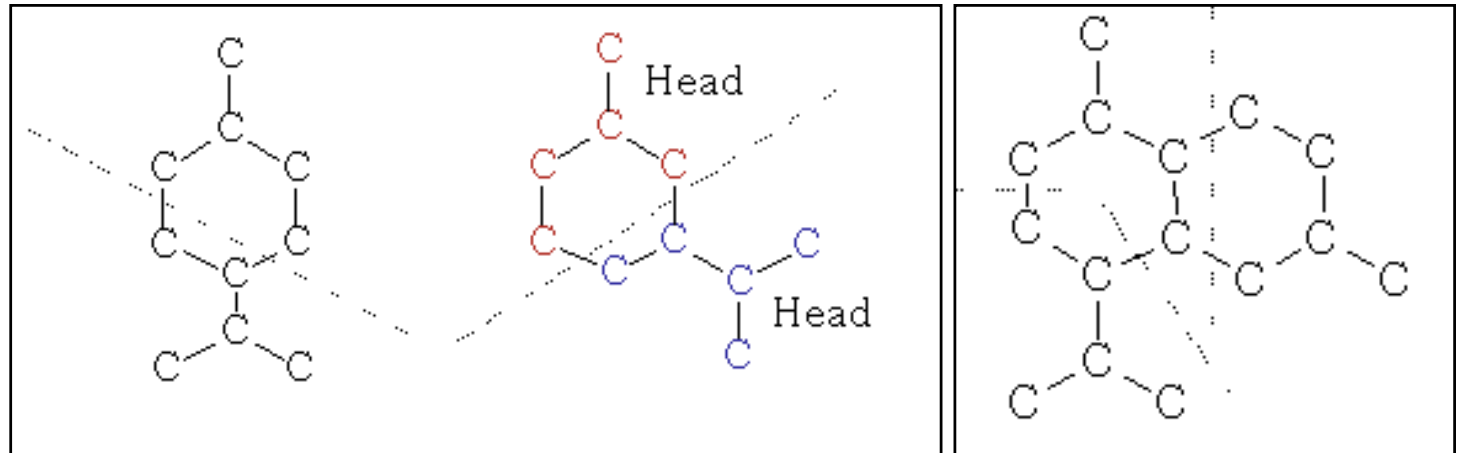


Terpene = (C₅ H₈)_n

- n = 1: Hemiterpenes
- n = 2: Monoterpenes
- n = 3: Sesquiterpenes
- n = 4: Diterpenes



Polyisoprene_linear form

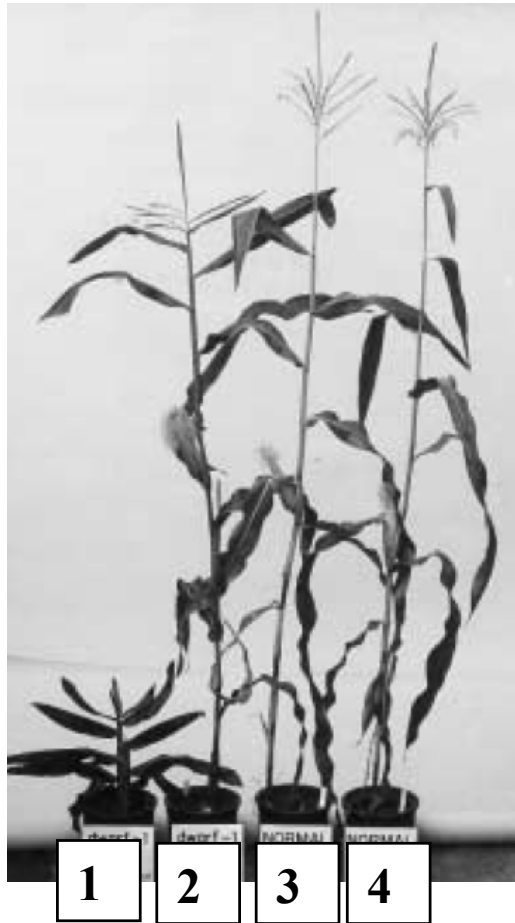


Polyisoprene_ring form

GIBERRELLIN: Biological function

- Stimulate stem and root growth
- Promote seed germination
- Promote transition from juvenile to adult phase
- Influence floral initiation
- Promote pollen development and tube growth
- Promote fruit set and parthenocarpy

GA stimulate stem and root growth



- 1: Dwarf mutant
- 2: Dwarf mutant + GA1
- 3: WT
- 4: WT + GA1

Stimulate stem elongation

- dramatic for **dwarf mutant**
 - no (a little) effect on tall (WT) plant
 - Dramatic for **rosette plant**
- EX: Grass family.



- 1: Cabbage in shortday time
- 2: Cabbage in shortday time + GA3

GA action depends on Auxin action

Auxin vs GA

Auxins	Gibberellin
Auxin promotes growth in shoot segments.	Gibberllin promotes growth in intact shoot.
There is little effect on leaf growth.	Leaf growth is enhanced.
Auxin has no effect on genetically dwarf plants.	It enhances internode growth in genetically dwarf plants.
It causes apical dominance.	Apical dominance is not affected.
It does not cause bolting in rosette plants and root crops.	It bring about elongation of stem or bolting in rosette plants and root crops.
It has no influence on the requirement of vernalisation.	Gibberllin can replace the requirement of vernalisation in most plants.
It has no effect on the flowering of long day plants.	It can replace the requirement of long photoperiods in long day plants.

Auxin vs GA

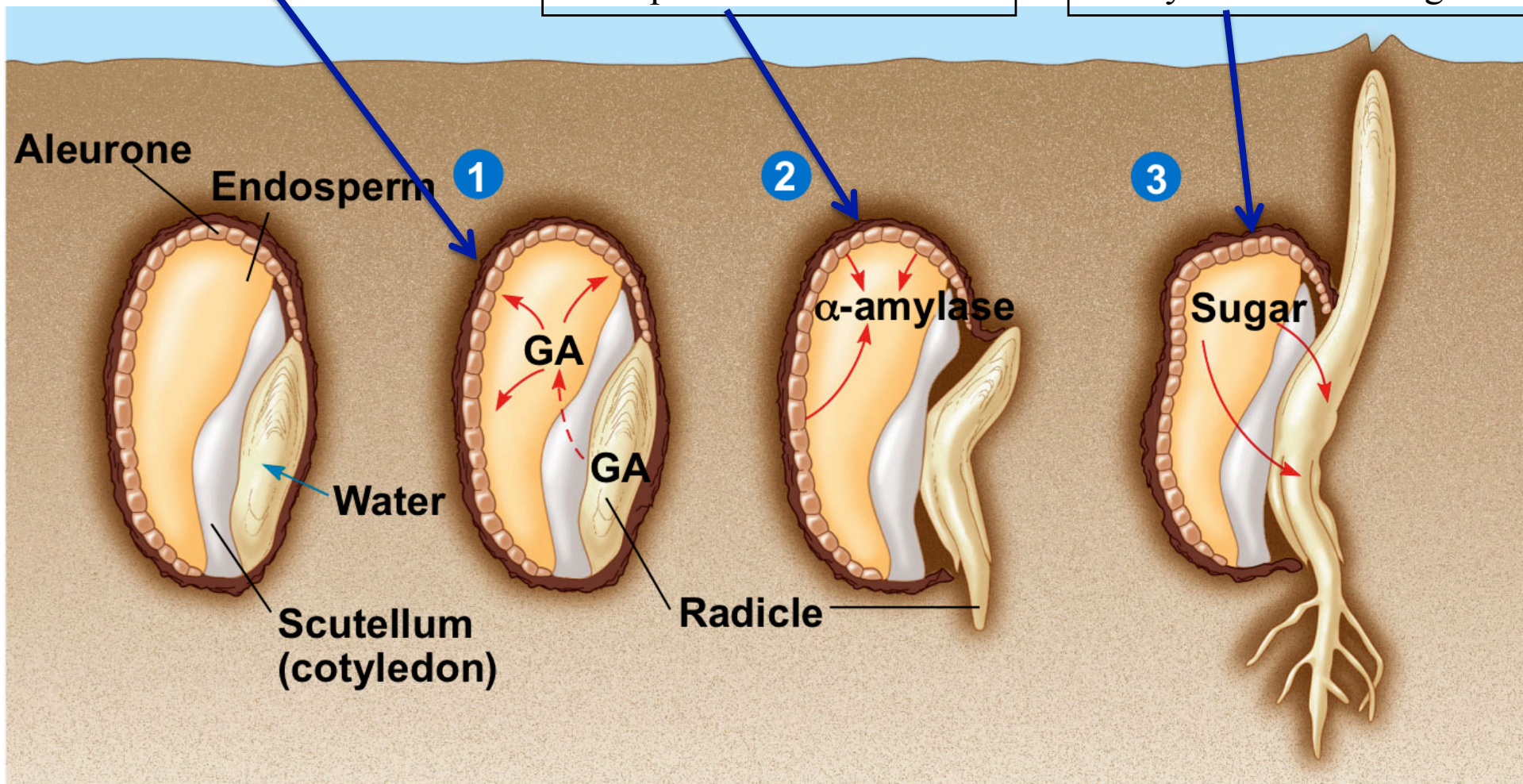
Auxins	Gibberellin
It is essential for the growth and formation of callus.	It does not influence formation and growth of callus.
It promotes rooting on cuttings.	It has no effect on rooting of cuttings.
It does not break dormancy.	It helps in breaking dormancy.
Auxin transport is polar.	It shows channel transport in different directions.
Root growth is promoted by low concentration of auxin and inhibited by its normal concentration.	Gibbellin has no specific effect on root growth.
It does not produce hydrolysing enzymes for mobilizing food reserve during seed germination.	Gibbellin produces hydrolysing enzymes for solubilisation of food reserve during seed germination.

GA promote seed germination

1. After a seed imbibe water, the embryo release GA as a signal to the aleurone,

2. Aleurone responds by synthesizing and secreting digestive enzyme that hydrolyze store nutrients in endosperm

3. Sugar and other nutrient absorbed from endosperm by cotyledon are consumed during growth of the embryo into a seedling



GA regulate the transition from juvenile to adult phase



Giant sequoia

Largest tree (97m height, 8m diameter)
Oldest plant (3500 years old)
Flowering at 70 – 100 years old
2 years for flower maturing



Giant sequoia 14-week-old

Spray GA₃ about 8 weeks earlier

GA regulate the transition from juvenile to adult phase



White Spruce WT

25 m height in height
Flowering at 12-30 years old



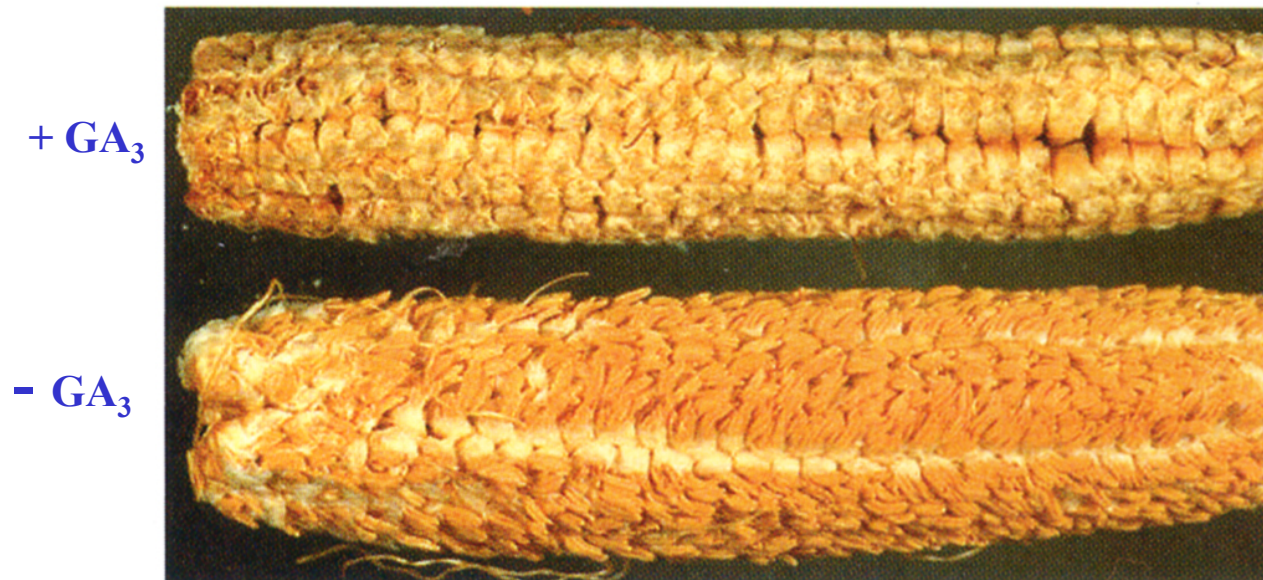
White spruce

**Stem injected the
previous summer
with GA₄/GA₇
mixture in aqueous
ethanol**

006 Sl

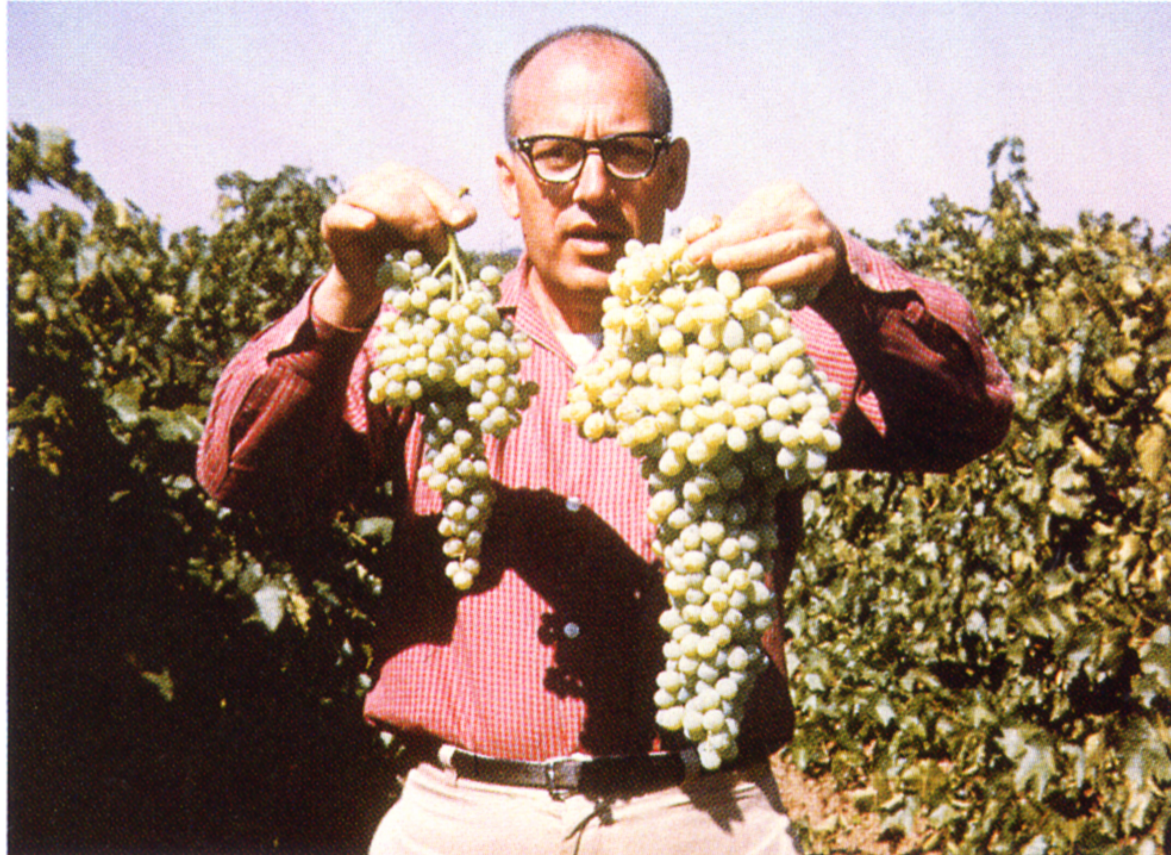
GA promote pollen development and tube growth influence floral initiation and sex determination

Tassel + GA → female (pistillate) induction,
suppress stamen (anther) development



GA-deficient mutant

GA promote fruit set and parthenocarpy

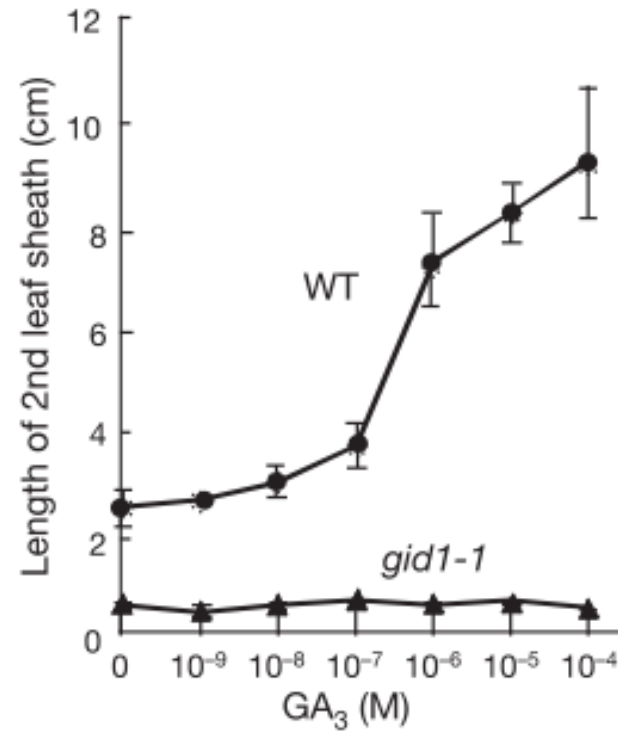


GA induces growing in Thompson Seedless grapes

GA signaling pathways



Gid1: GA-insensitive dwarf1



GA₃-induced elongation of the second leaf sheath

GID1 is GA receptor

Figure 20.14 Structure of the GA₃-GID1a-DELLA complex

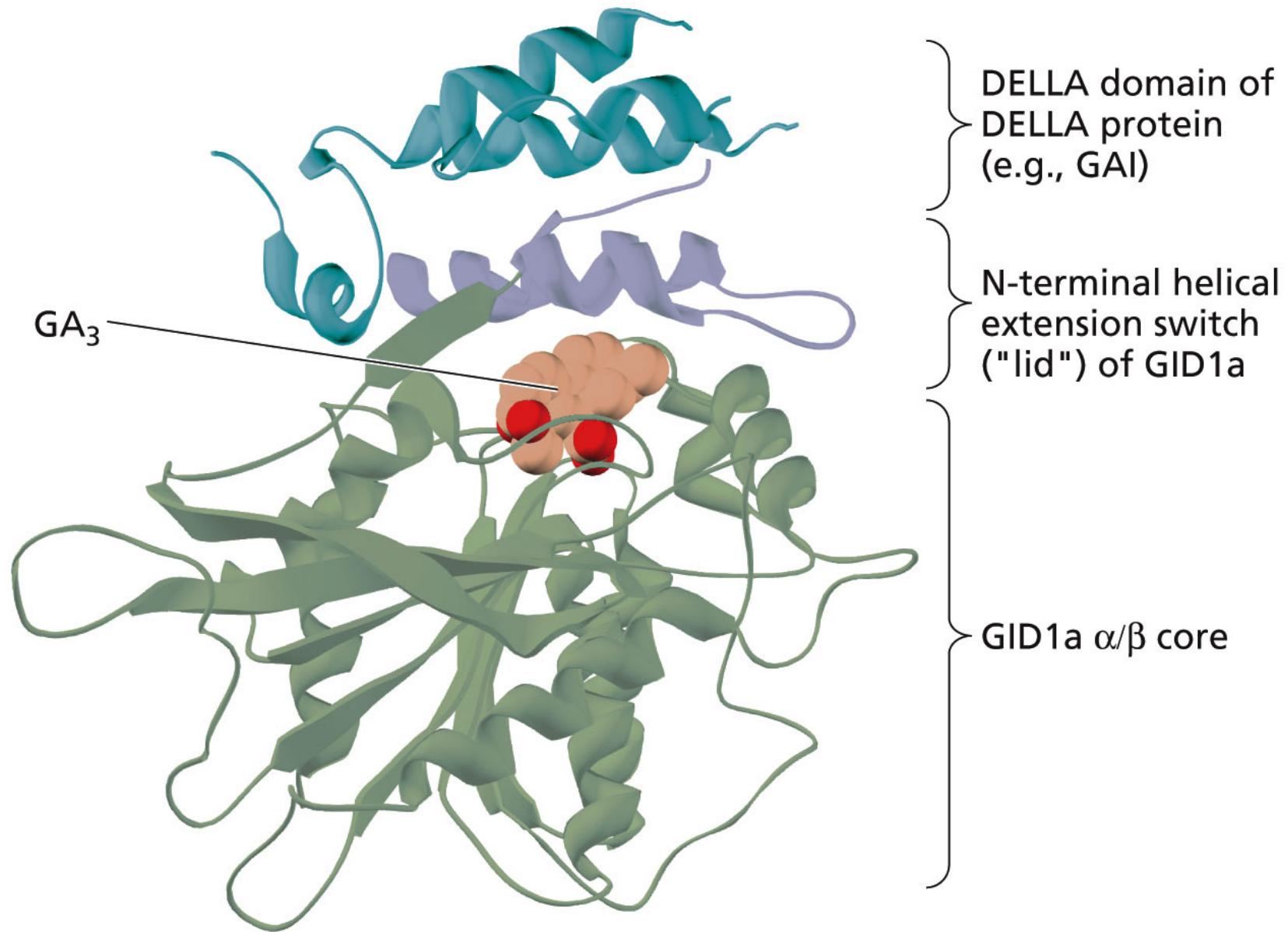
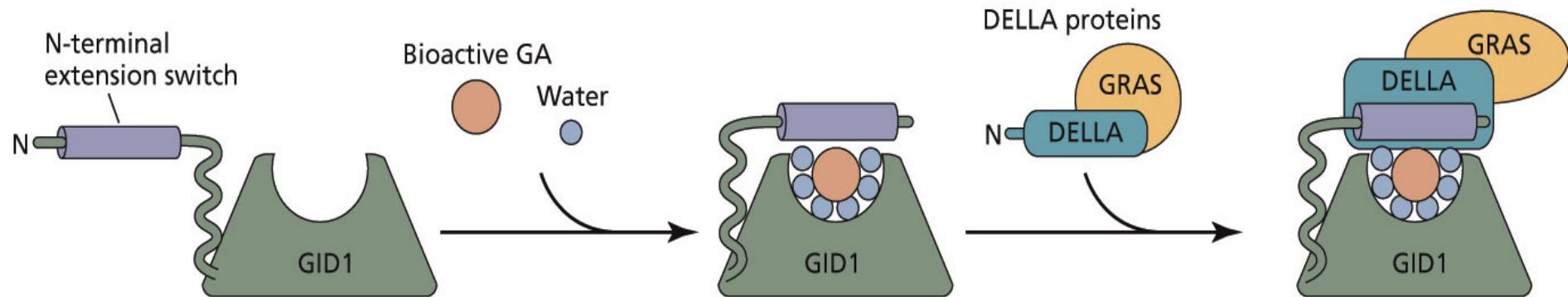


Figure 20.15 Model of GA-induced change in the GID1 protein

1. GID1 is an allosteric protein. Binding GA causes a conformational change that leads to the extension switch closing like a lid.

2. Binding to the GA-GID1 complex causes a conformational change in the N-terminal DELLA domain of a DELLA protein.

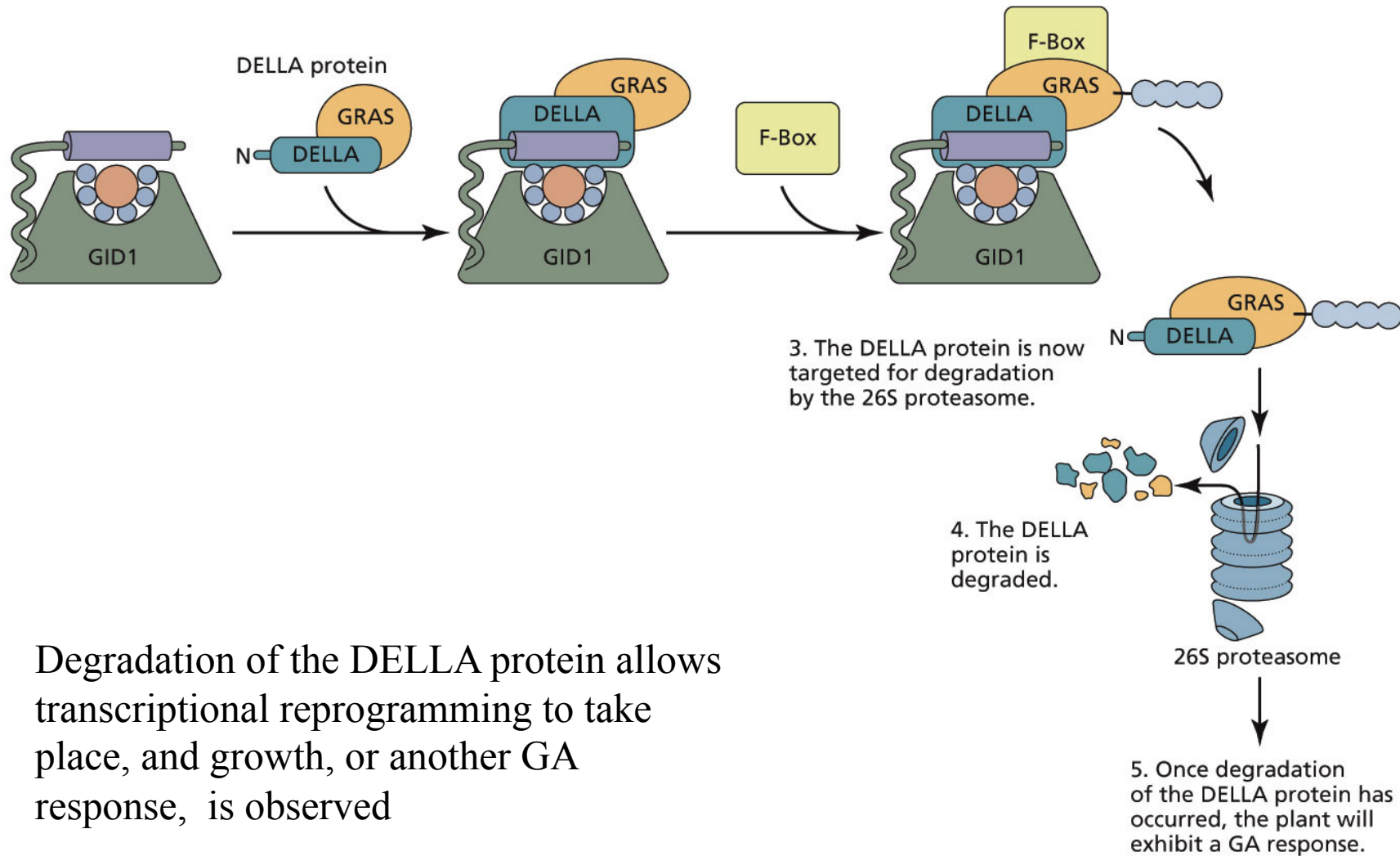
3. Changes in the DELLA domain may also induce a conformational change in the GRAS domain.



GA signaling pathways

1. Upon binding of a DELLA protein to the GA-GID1 complex, a change in the conformation of the GRAS domain is thought to facilitate its binding to the F-box component of an SCF complex.

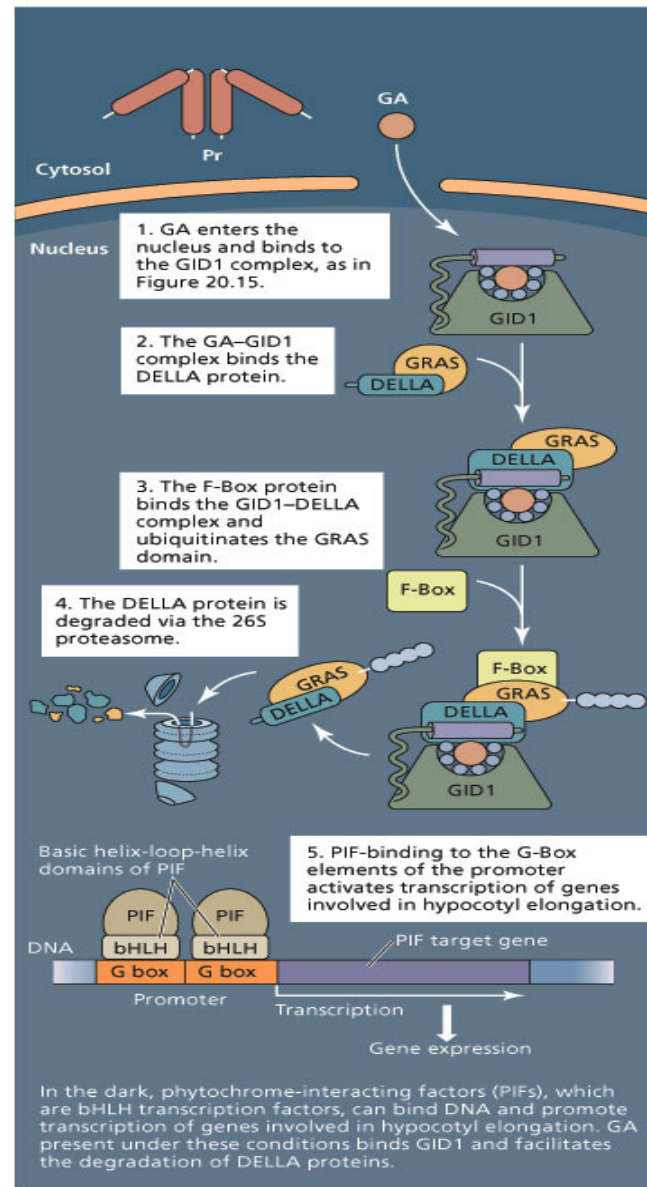
2. The F-box protein polyubiquitinates the GRAS domain of the DELLA protein.



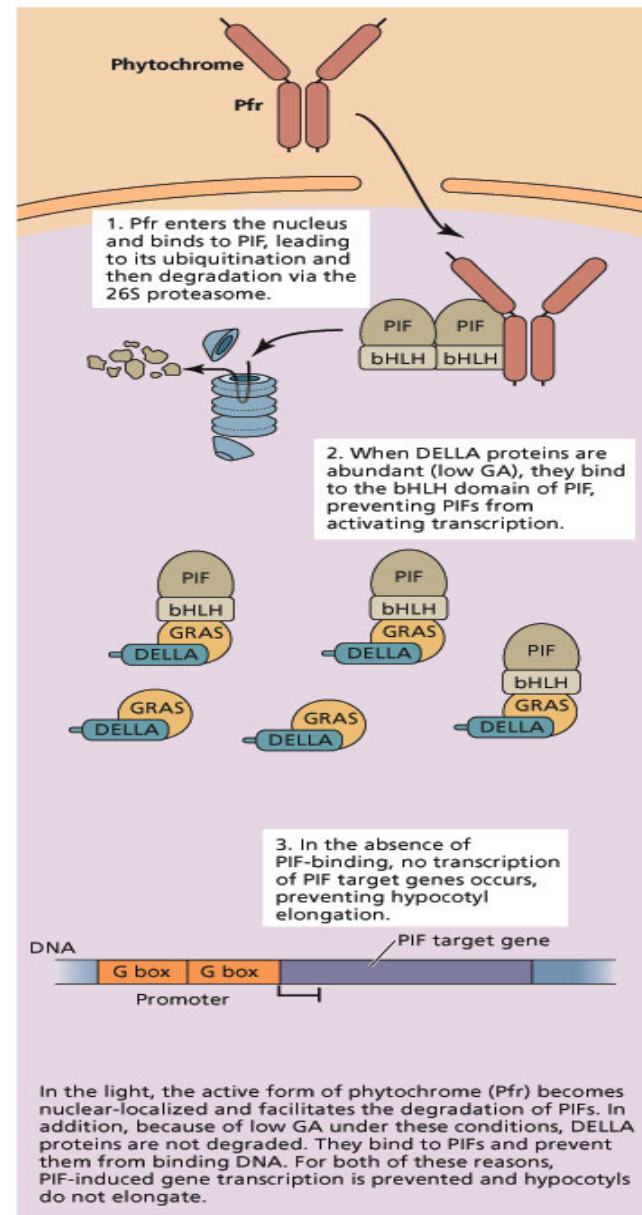
Degradation of the DELLA protein allows transcriptional reprogramming to take place, and growth, or another GA response, is observed

How does GA stimulate stem elongation?

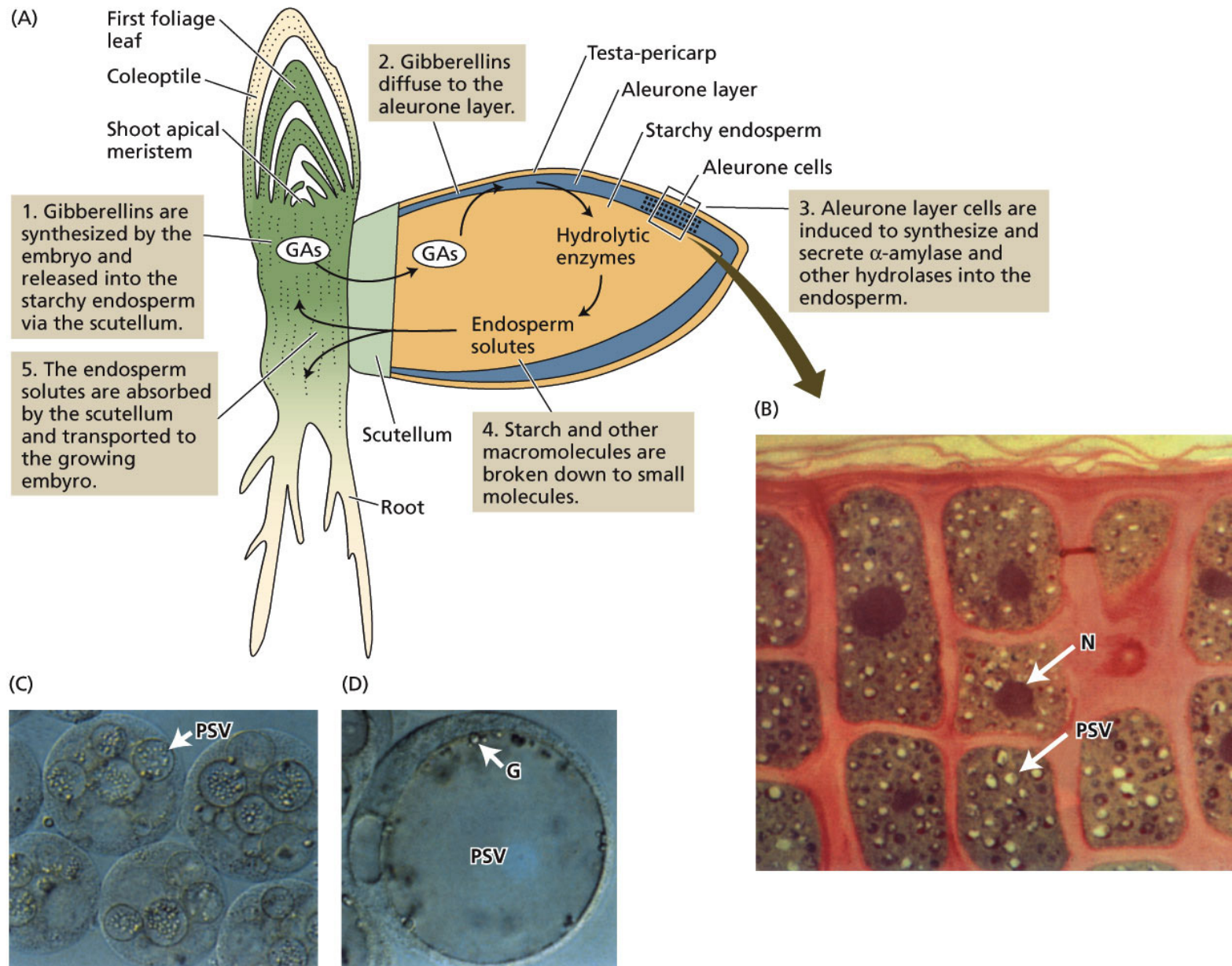
(A) Dark/high GA → long hypocotyl



(B) Light/low GA → short hypocotyl



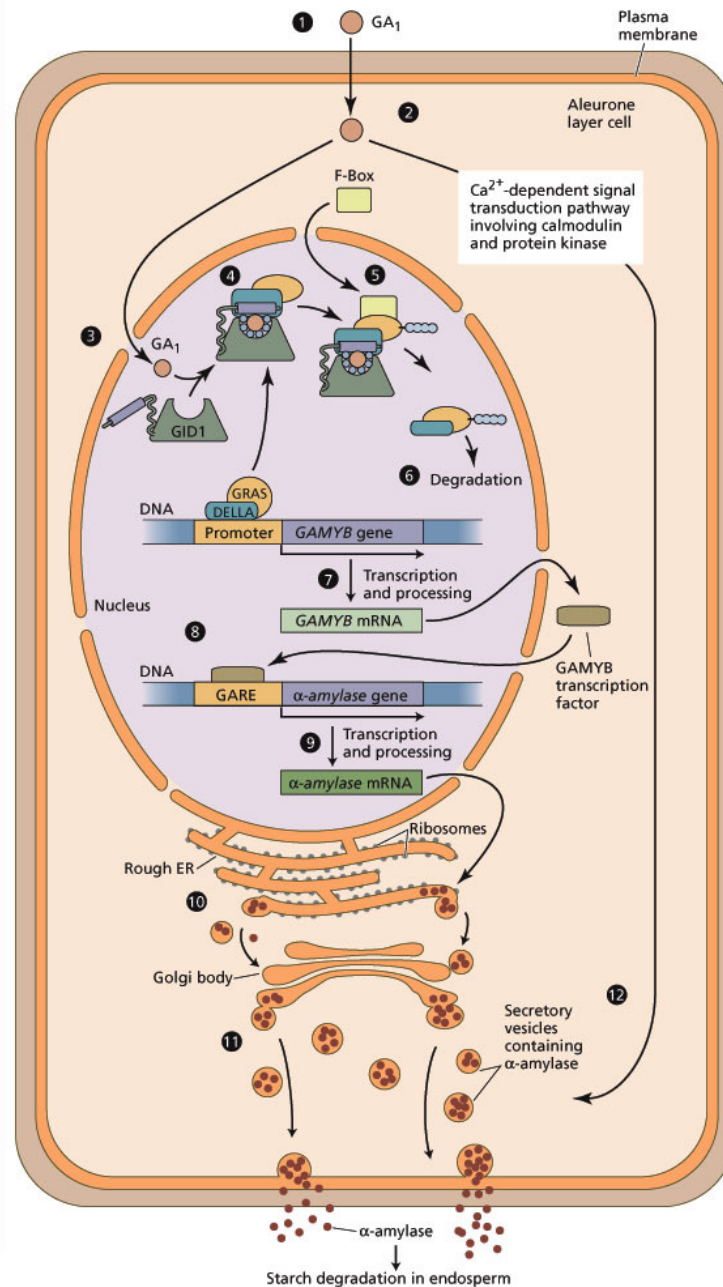
How does GA promote seed germination?



PLANT PHYSIOLOGY, 5e, Figure 20.21

How does GA promote seed germination?

1. GA₁ from the embryo enters an aleurone cell.
2. Once inside the cell, GA₁ may initiate a calcium-calmodulin-dependent pathway necessary for α-amylase secretion.
3. GA₁ binds to GID1 (a soluble GA receptor) in the nucleus.
4. Upon binding GA₁, the GID1 receptor undergoes an allosteric change that facilitates its binding to a DELLA protein.
5. Once the DELLA protein has bound the GA₁-GID1 complex, an F-box protein (part of an SCF complex) is now able to polyubiquitinate the GRAS domain of the DELLA protein.
6. The polyubiquitinated DELLA protein is degraded by the 26S proteasome.
7. Once the DELLA protein has been degraded, transcription of an early gene is activated. (GAMYB is shown, in this model, as an early gene, although there is evidence that transcriptional regulation of other early genes may occur first.) The mRNA for GAMYB is translated in the cytosol.
8. The newly synthesized GAMYB transcription factor enters the nucleus and binds the promoters of α-amylase and genes encoding other hydrolytic enzymes.
9. Transcription of these genes is activated.
10. α-amylase and other hydrolases are synthesized on the rough ER, processed, and packaged into secretion vesicles by the Golgi body.
11. Proteins are secreted by exocytosis.
12. The secretory pathway requires GA stimulation of the calcium-calmodulin-dependent pathway.



CYTOKININ: discovery

1934: Medium (mineral, vitamin, sucrose) + No hormone:

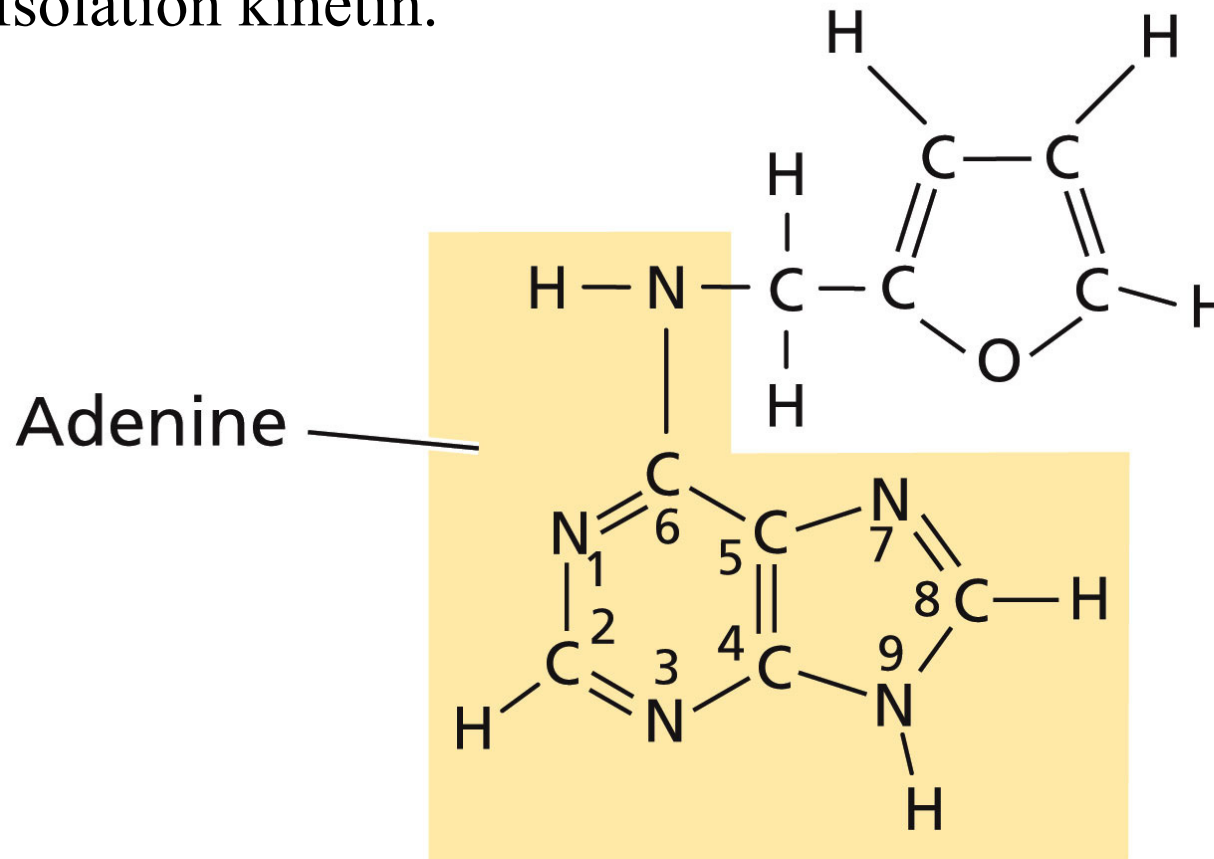
- Root can be grown
- Shoot can not be grown

1948: Medium + coconut milk: shoot can be grown

CYTOKININ: discovery

1955: autoclaved herring sperm DNA stimulate cell division;

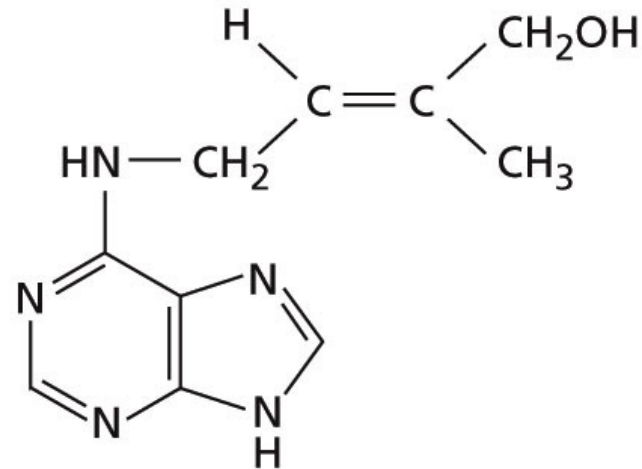
Isolation kinetin.



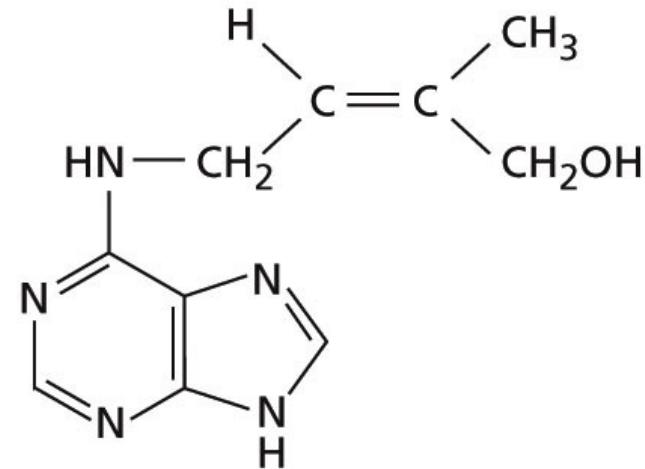
Kinetin

CYTOKININ: discovery

1961: First naturally occurring cytokinin found in plants (immature endosperm maize), later called zeatin



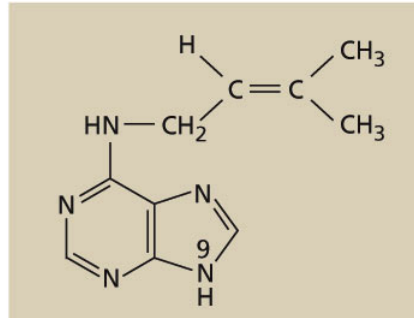
trans-Zeatin



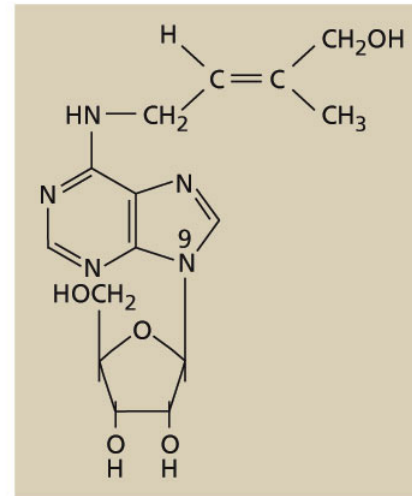
cis-Zeatin

6-(4-Hydroxy-3-methylbut-2-enylamino)purine

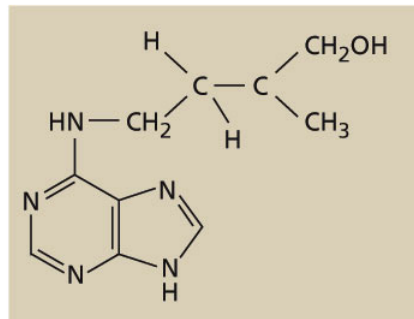
Figure 21.2 Structures of other aminopurines that are active as cytokinins



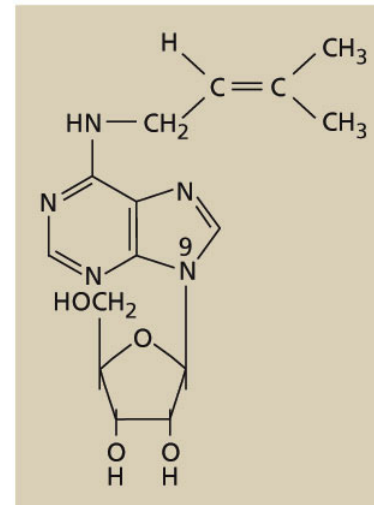
N⁶-(D²-Isopentenyl)-adenine (iP)



Ribosylzeatin (zeatin riboside)



Dihydrozeatin (DZ)



N⁶-(Δ²-Isopentenyl)adenosine ([9R]iP)

CYTOKININ: biosynthesis

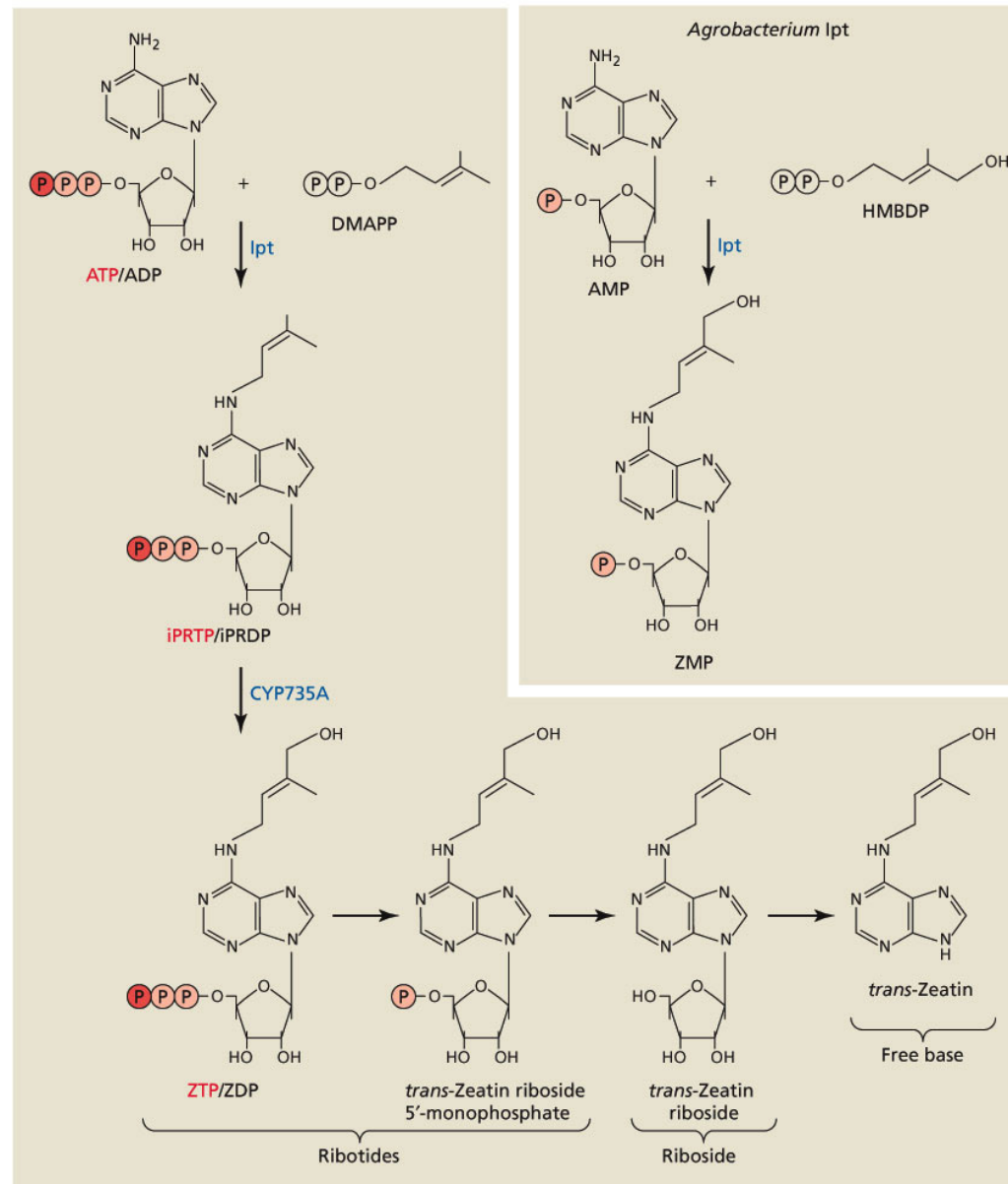
- Generated mostly in the **root apical meristems** but also found in:

- Root cap cells
- Ovules
- Phloem cells
- Leaf axils
- Tips of young inflorescences
- Fruit
- Seeds

Other organisms make cytokinins to influence the plant for their own benefit

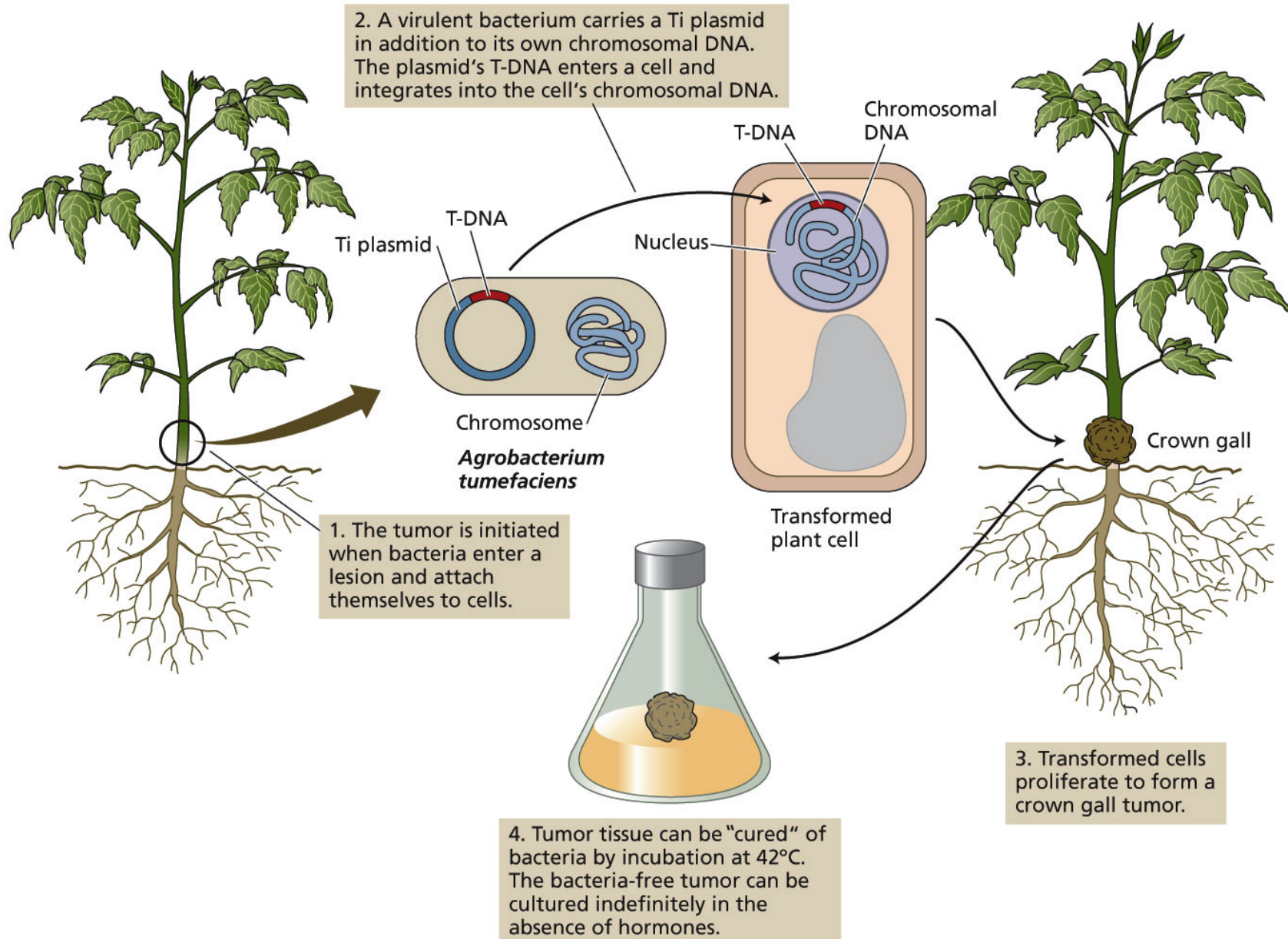
- Bacteria
- Fungi
- Insects
- Nematodes

CYTOKININ: biosynthesis pathway



PLANT PHYSIOLOGY, 5e, Figure 21.5

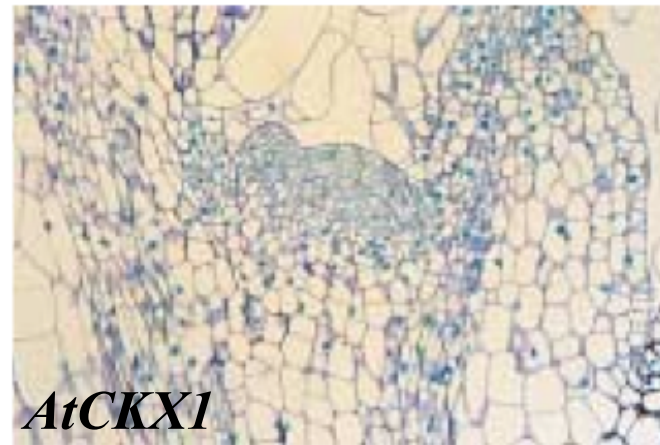
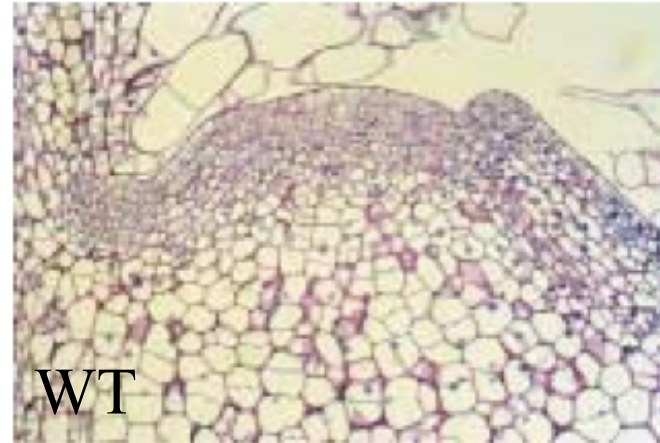
CYTOKININ: biosynthesis in tumor



CYTOKININ: promote shoot growth



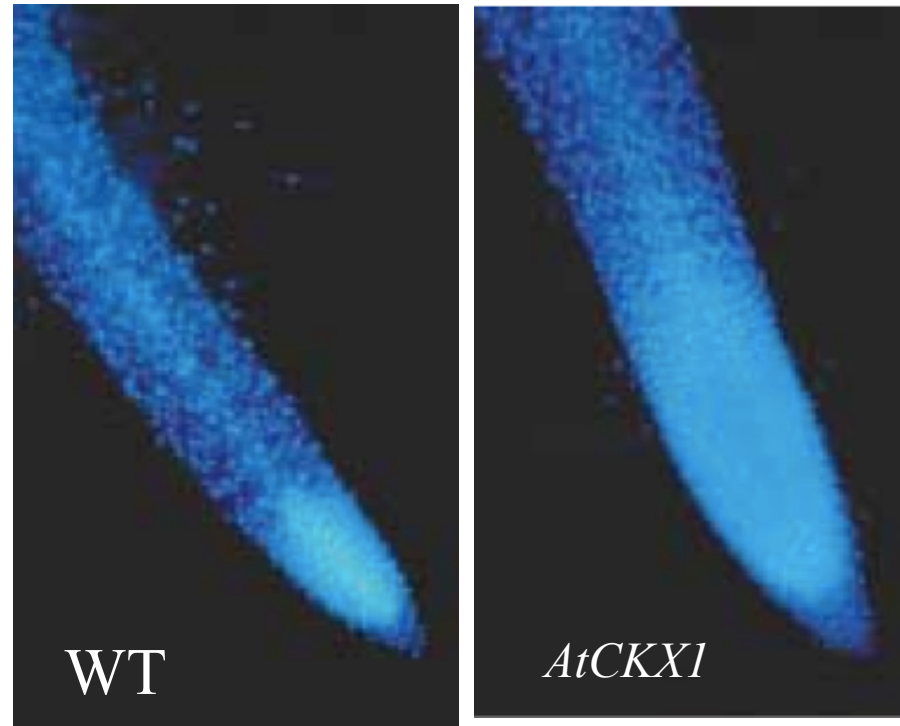
Longitudinal section through SAM



AtCKX1,
AtCKX2:
overexpress
of cytokinin
oxidase

Cytokinin promote shoot growth by **increasing cell poliferation in SAM**

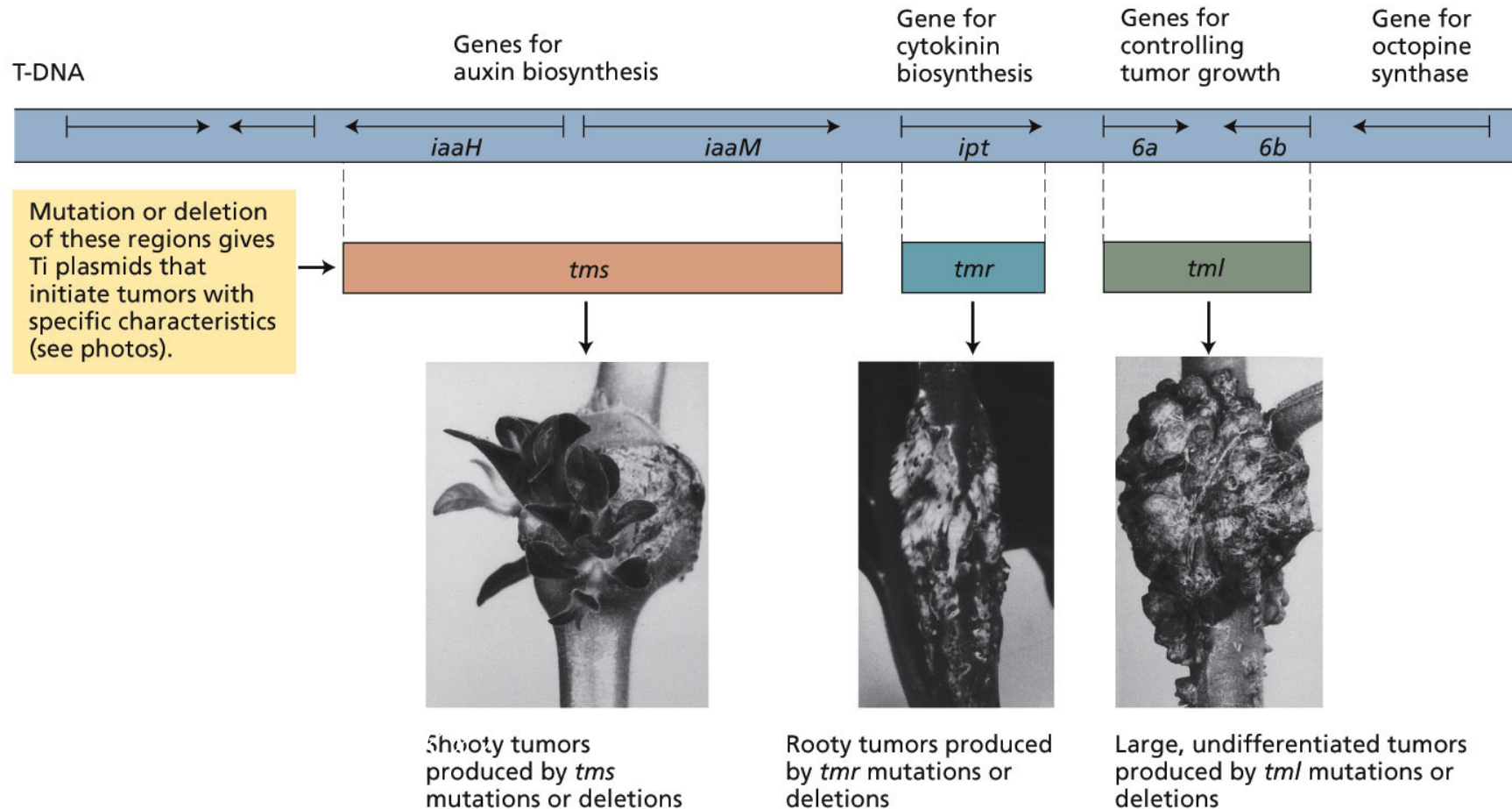
CYTOKININ: inhibit root growth



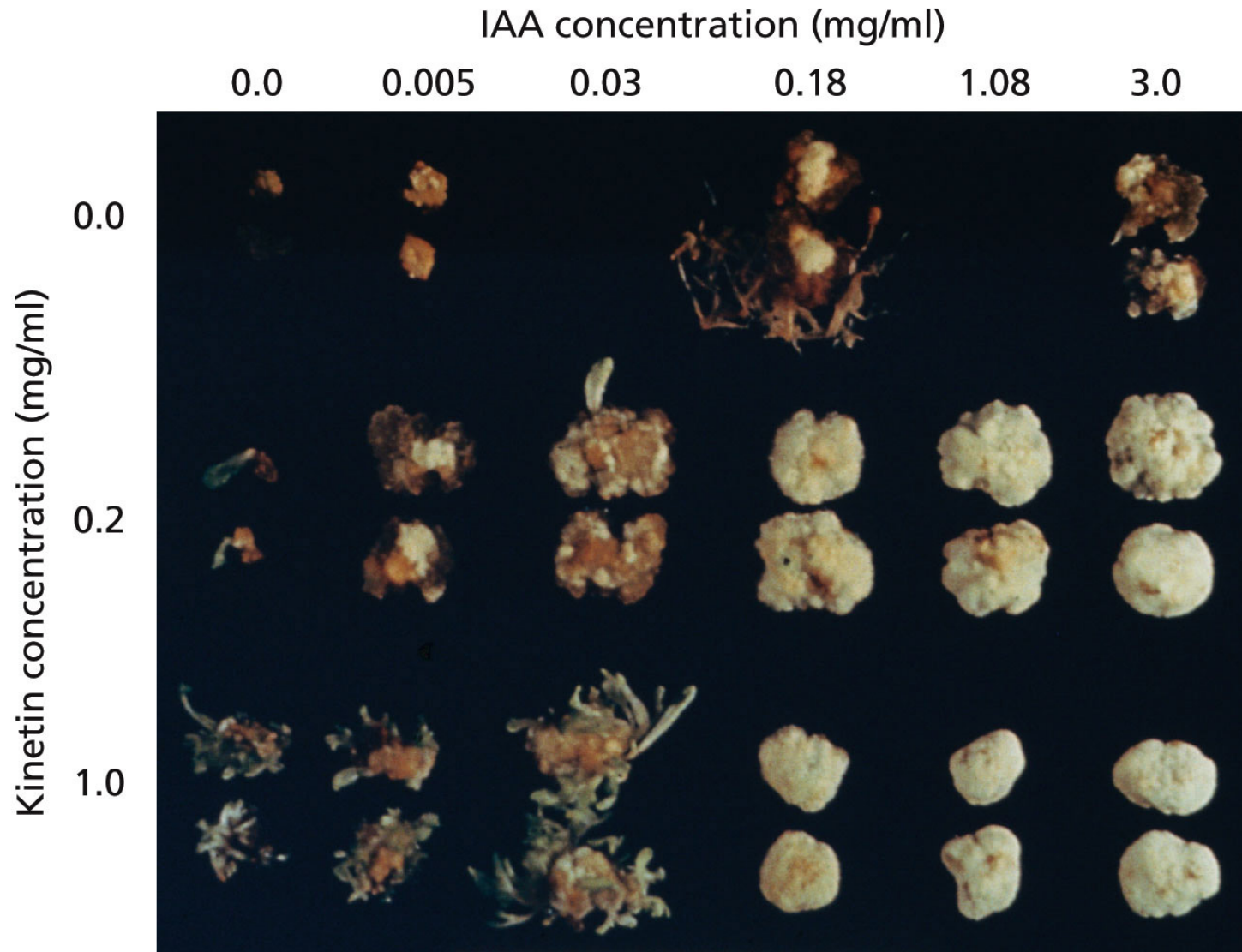
DAPI (4,6 diamindino-2-phenylindole) staining

Cytokinin inhibit root growth by **promoting the exit of cells from RAM**

Auxin/cytokinin ratio regulates morphogenesis in cultured tissues



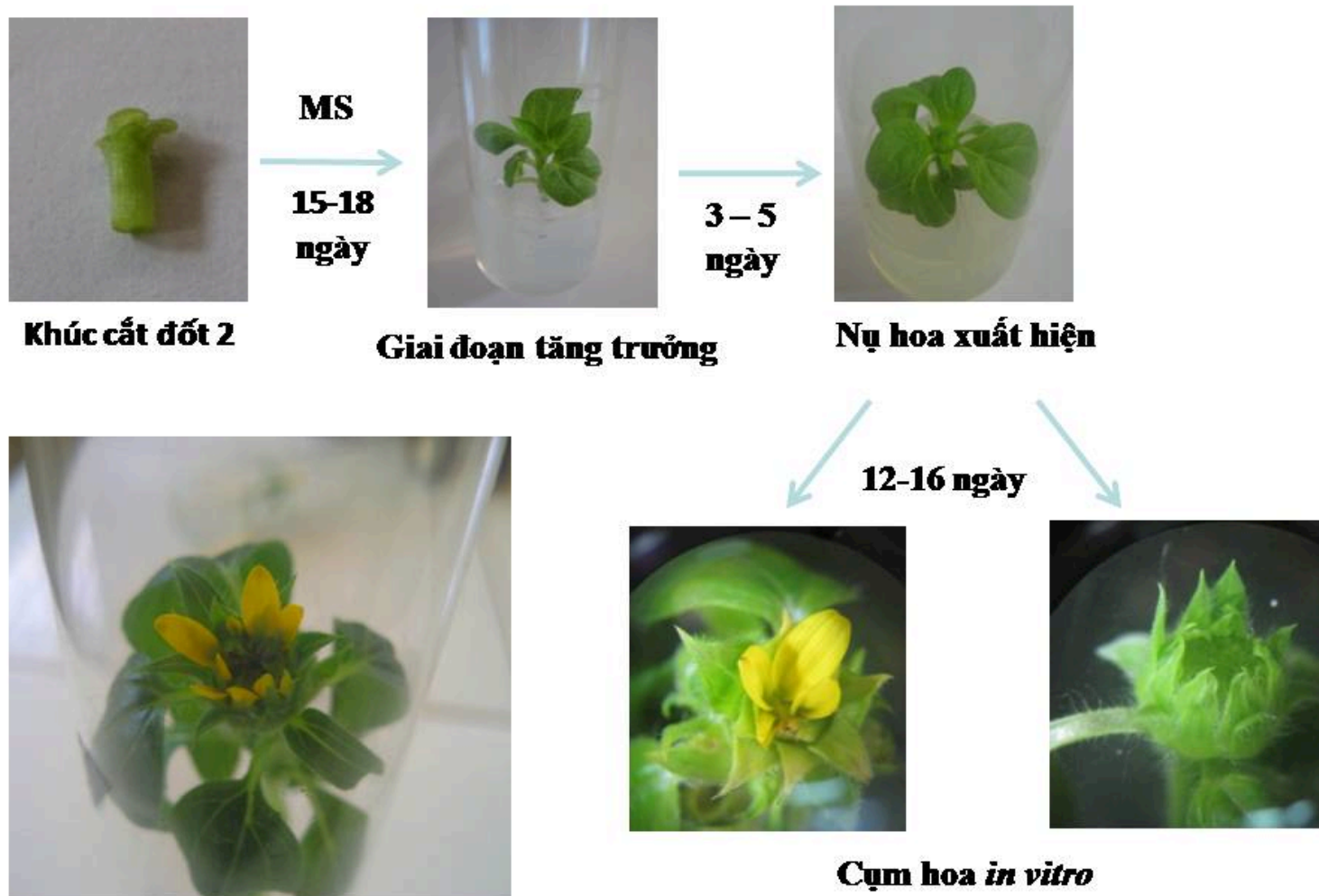
Auxin/cytokinin ratio regulates morphogenesis in cultured tissues



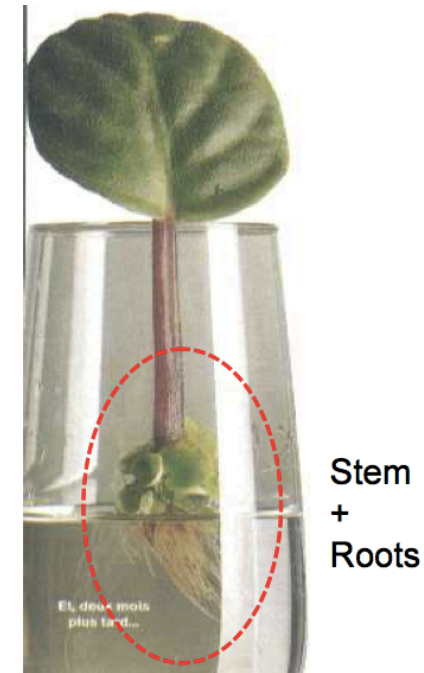
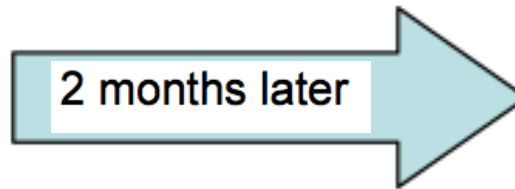
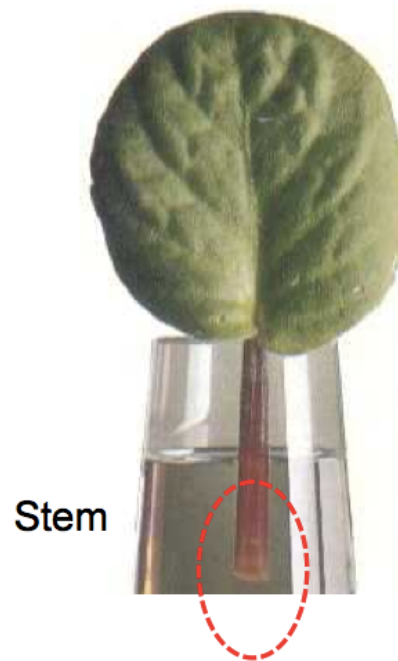
PLANT PHYSIOLOGY, 5e, Figure 21.16

Auxin/cytokinin ratio regulates morphogenesis in cultured tissues: case studies

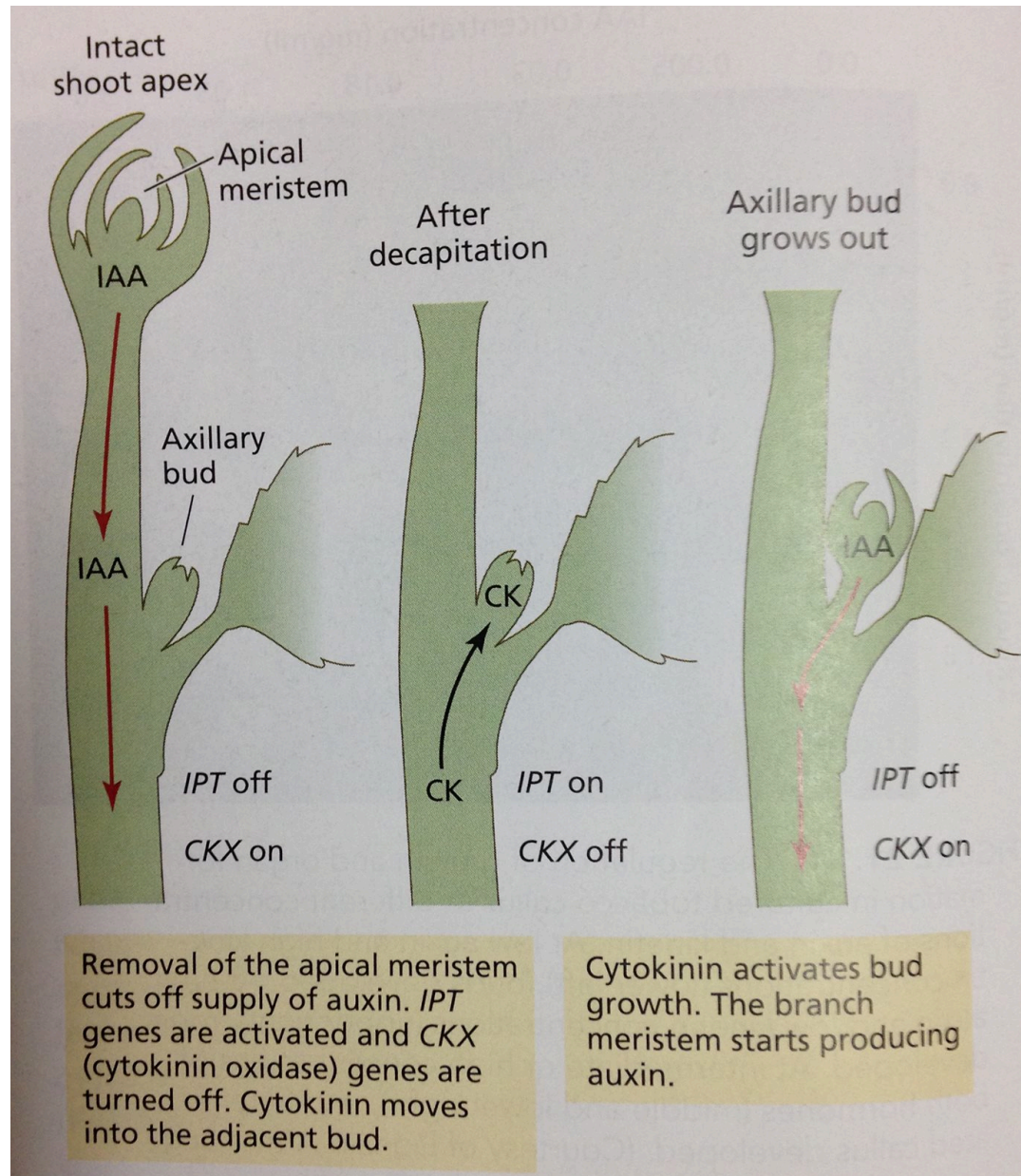
Sự ra hoa Hướng Dương trong ống nghiệm



Auxin/cytokinin ratio regulates morphogenesis in cultured tissues: case studies



CYTOKININ: modify the apical dominance



Cytokinin interact with auxin to modify apical dominance and promote the lateral bud growth

CYTOKININ: delay leaf senescence

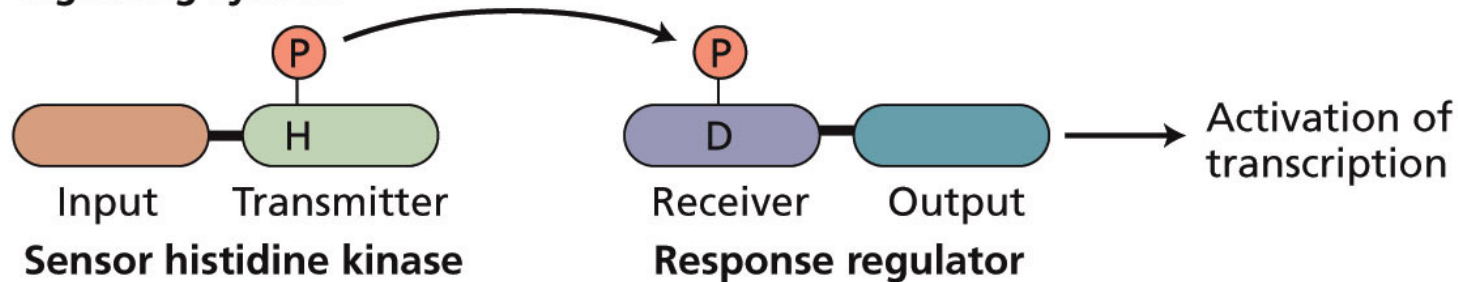


1: plant expressing *IPT* gene
2: WT

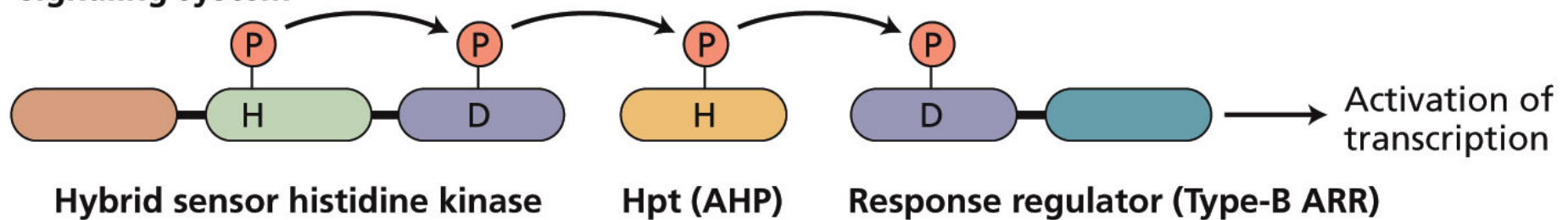
CYTOKININ signaling pathway

Simple versus phosphorelay types of two-component signaling systems

Simple two-component signaling system

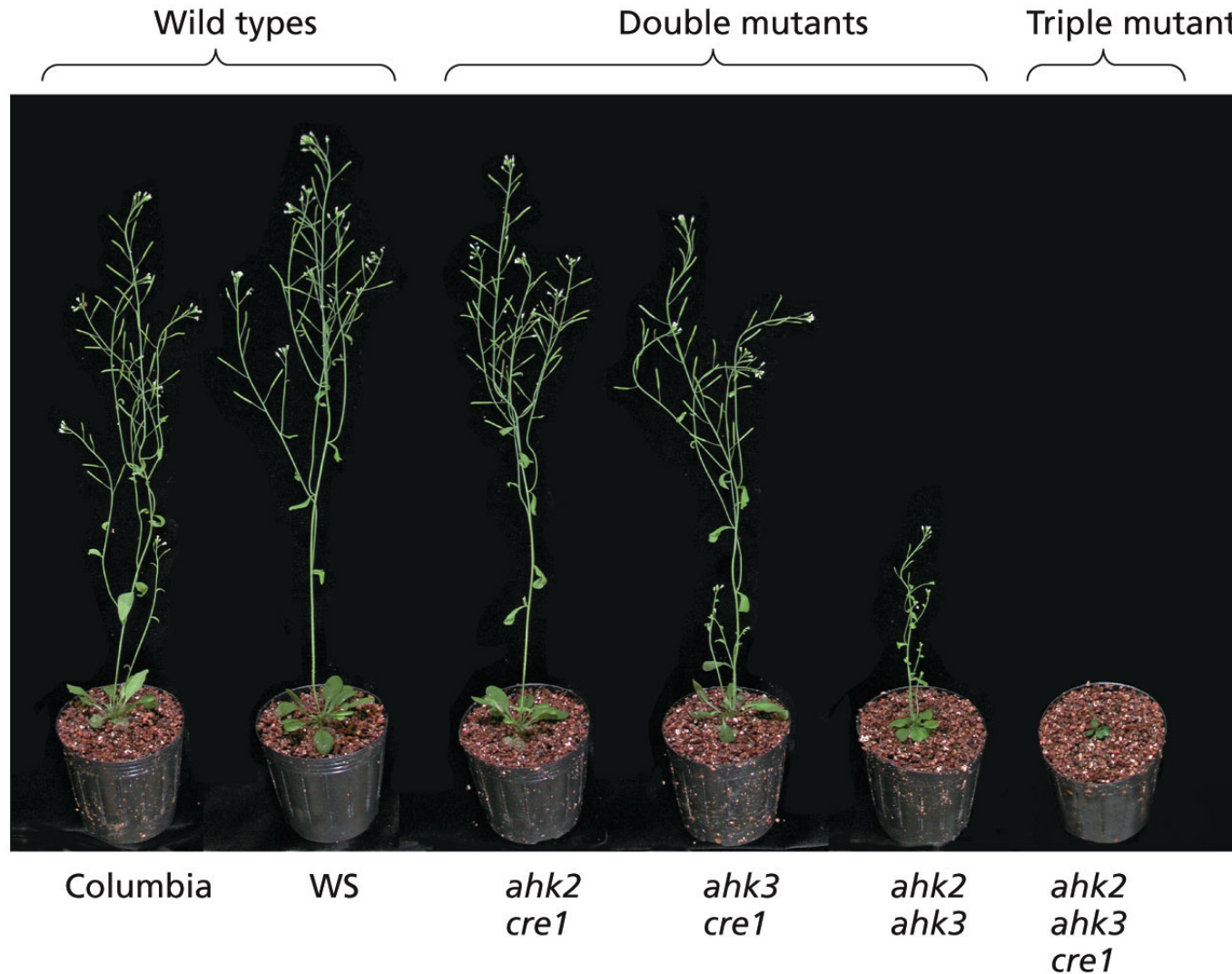


Phosphorelay two-component signaling system



CYTOKININ signaling pathway

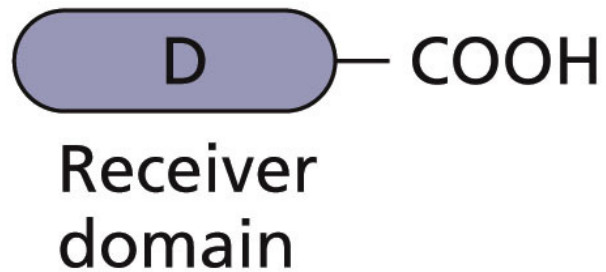
Phenotypes of Arabidopsis plants harboring mutations in the cytokinin receptors



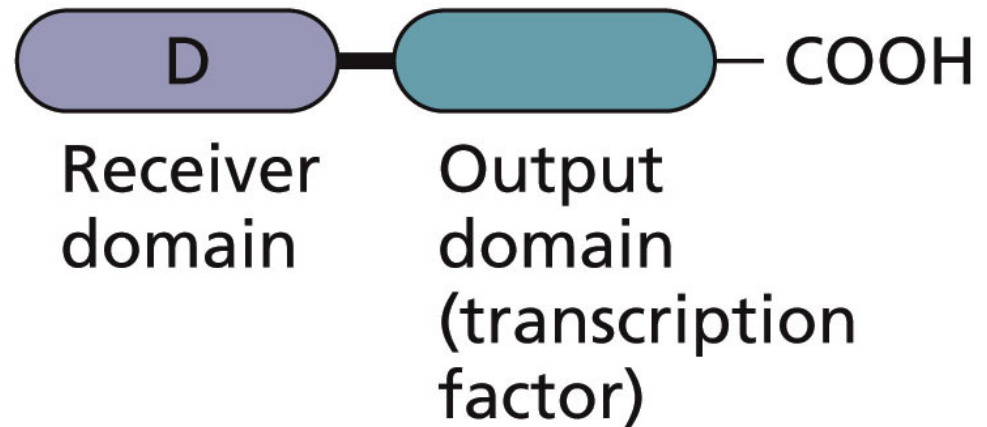
PLANT PHYSIOLOGY, 5e, Figure 21.7

CYTOKININ signaling pathway

Comparison of the structures of the type-A and type-B ARR1s



Type-A ARR1s



Type-B ARR1s

CYTOKININ signaling pathway

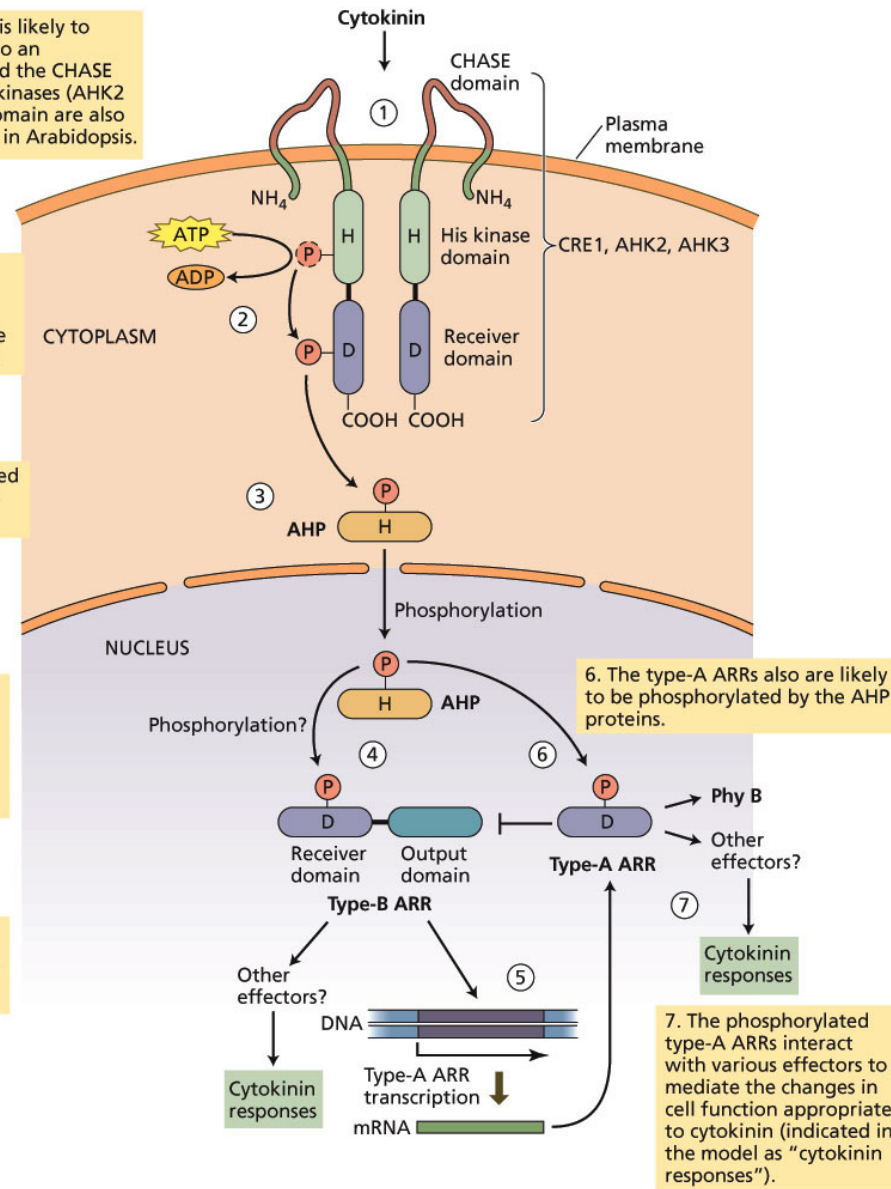
1. Cytokinin binds to CRE1, which is likely to occur as a dimer. Cytokinin binds to an extracellular portion of CRE1 called the CHASE domain. Two other hybrid sensor kinases (AHK2 and AHK3) containing a CHASE domain are also likely to act as cytokinin receptors in Arabidopsis.

2. Cytokinin binding to these receptors activates their histidine kinase activity. The phosphate is transferred to an aspartate residue (D) on the fused receiver domains.

3. The phosphate is then transferred to a conserved histidine present in an AHP protein.

4. The AHP proteins move into the nucleus, where they transfer the phosphate to an aspartate residue located within the receiver domain of type-A and type-B ARR.

5. The phosphorylation of the type-B ARR activates the output domain to induce transcription of genes encoding type-A ARRs.



PLANT PHYSIOLOGY, 5e, Figure 21.9