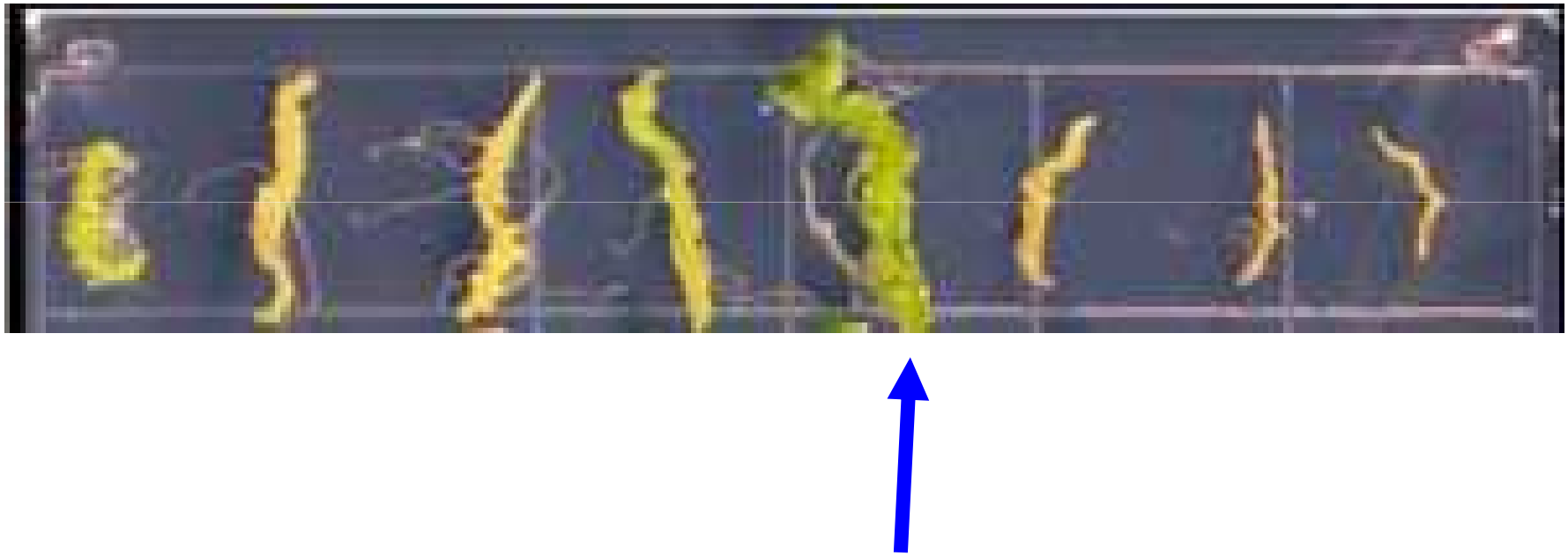
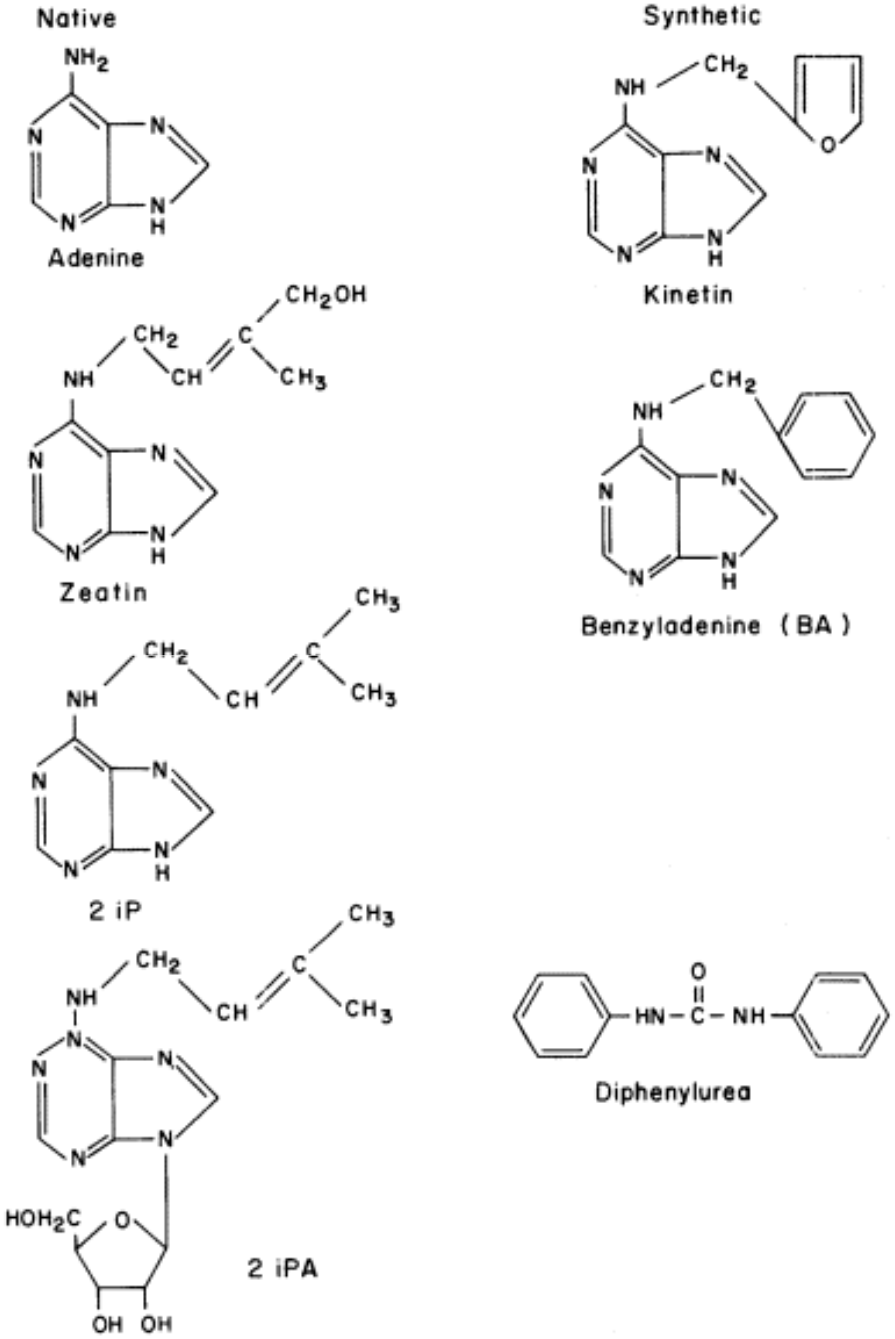
Role of cytokinin in plant development

Cytokinin – substance crucial for sustain of cell proliferation



Cytokinins


Native
Adenine
Zeatin
2 iP
2 iPA
Synthetic
Kinetin
Benzyladenine (BA)
Diphenylurea

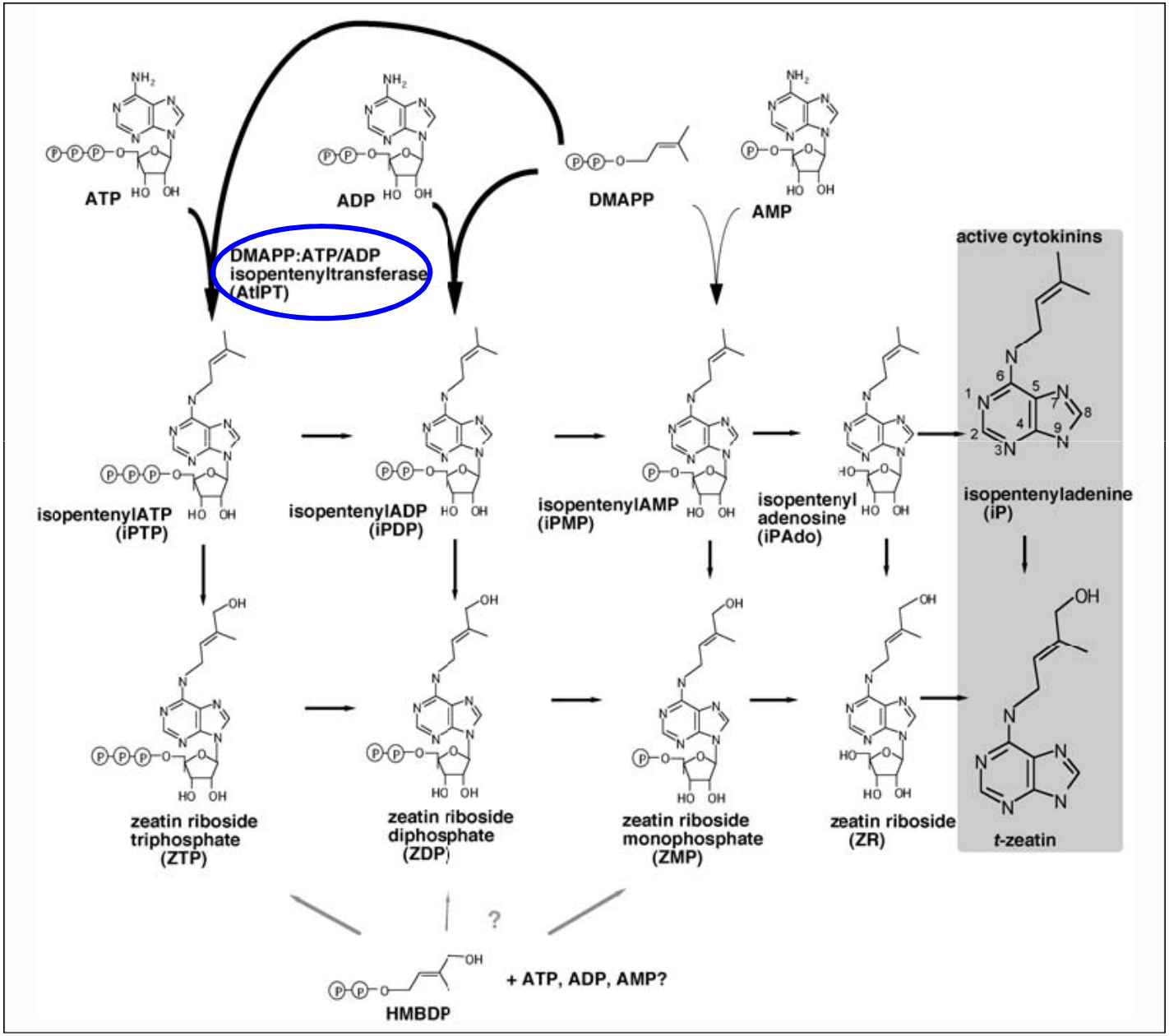
CYTOKININ - Metabolism

Synthesis – *IPT* genes

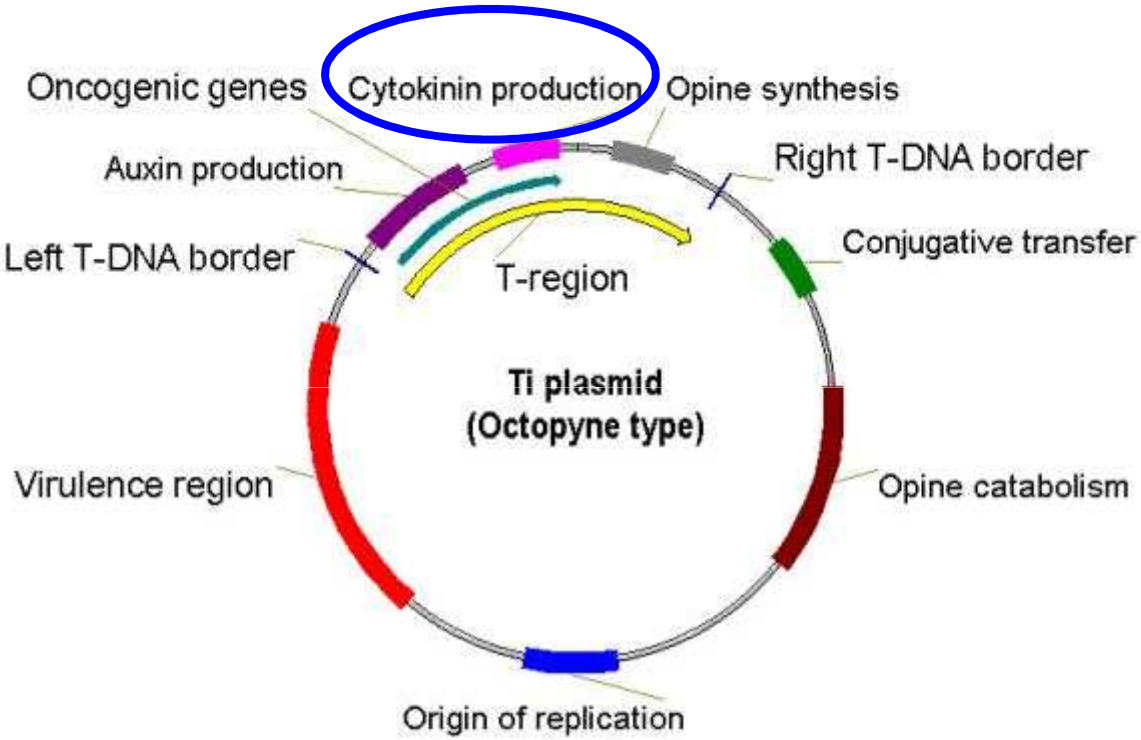
Degradation – CK-oxidase

Conjugation

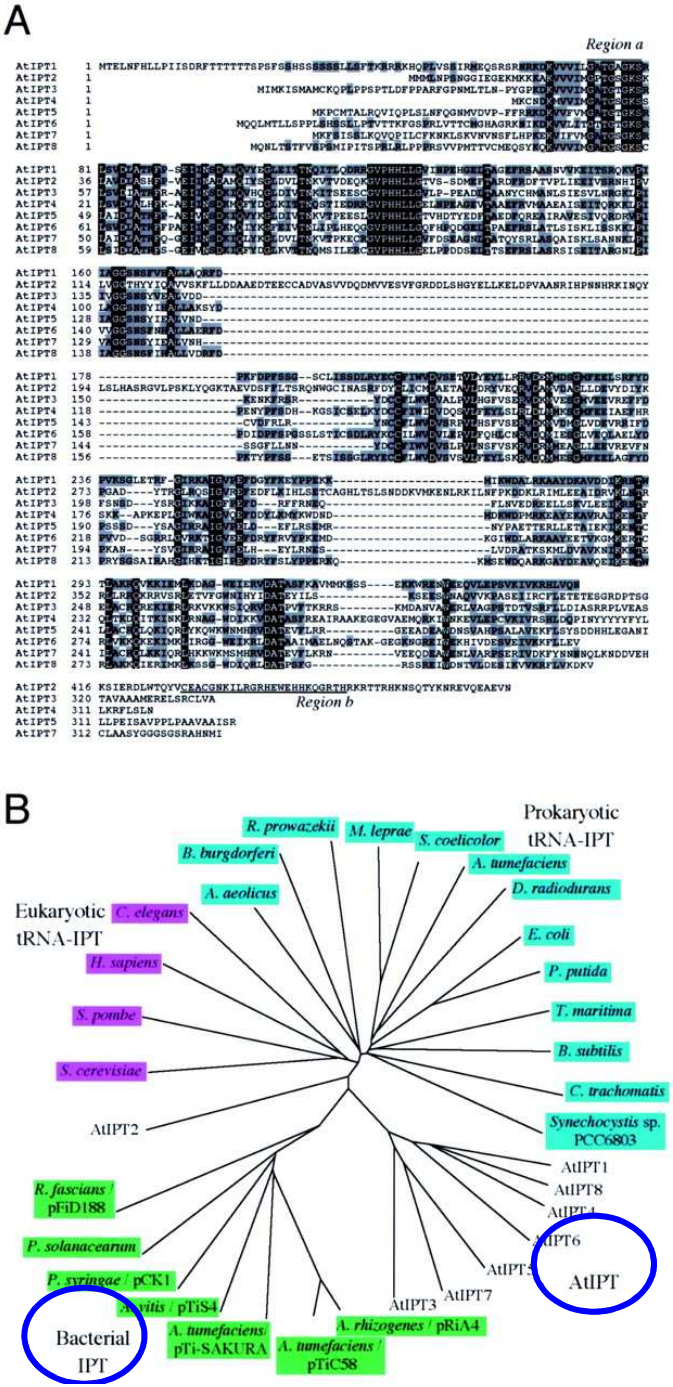
Cytokinin biosynthesis

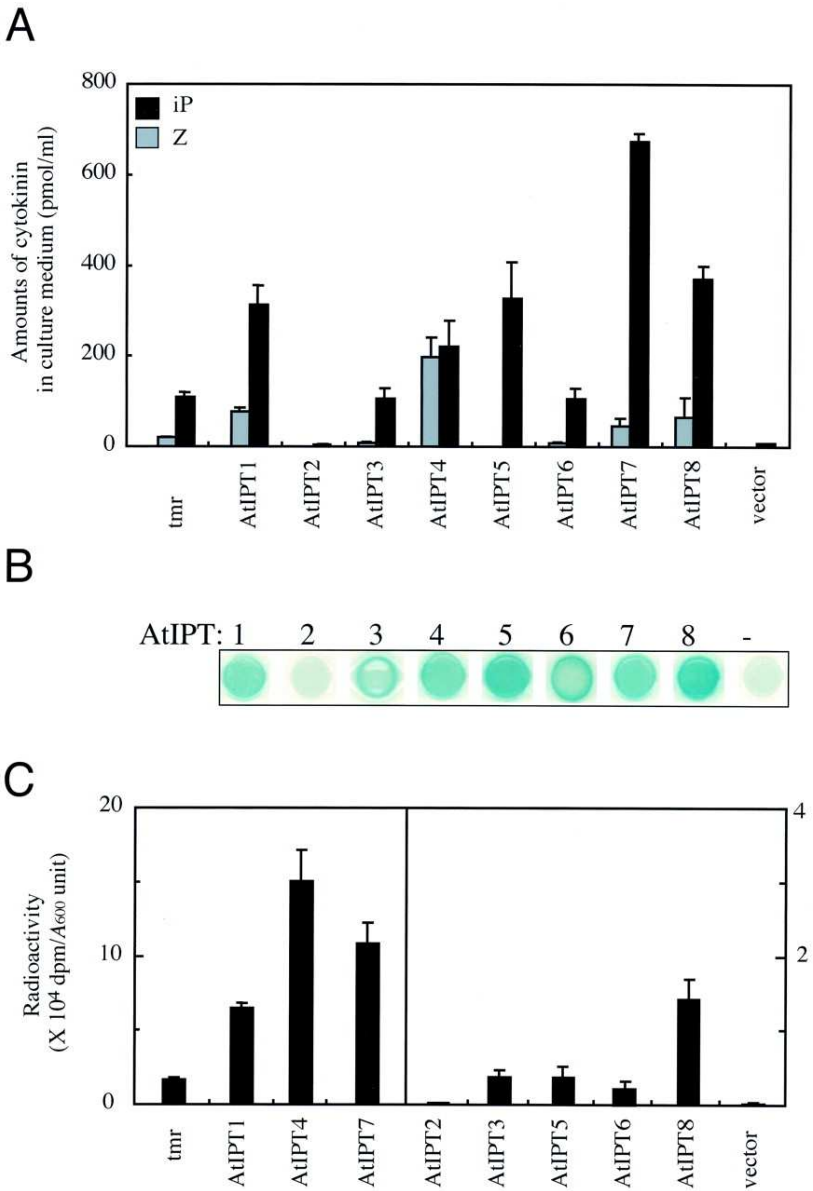


Agrobacterium tumefaciens – IPT gene for cytokinin biosynthesis



Isolation of Arabidopsis *IPT*s





Arabidopsis *IPT*s (8)

Differential expression patterns

Differently responsive promotors
(cytokinin, auxin, nitrate, combinations)

mutants, overexpression ???

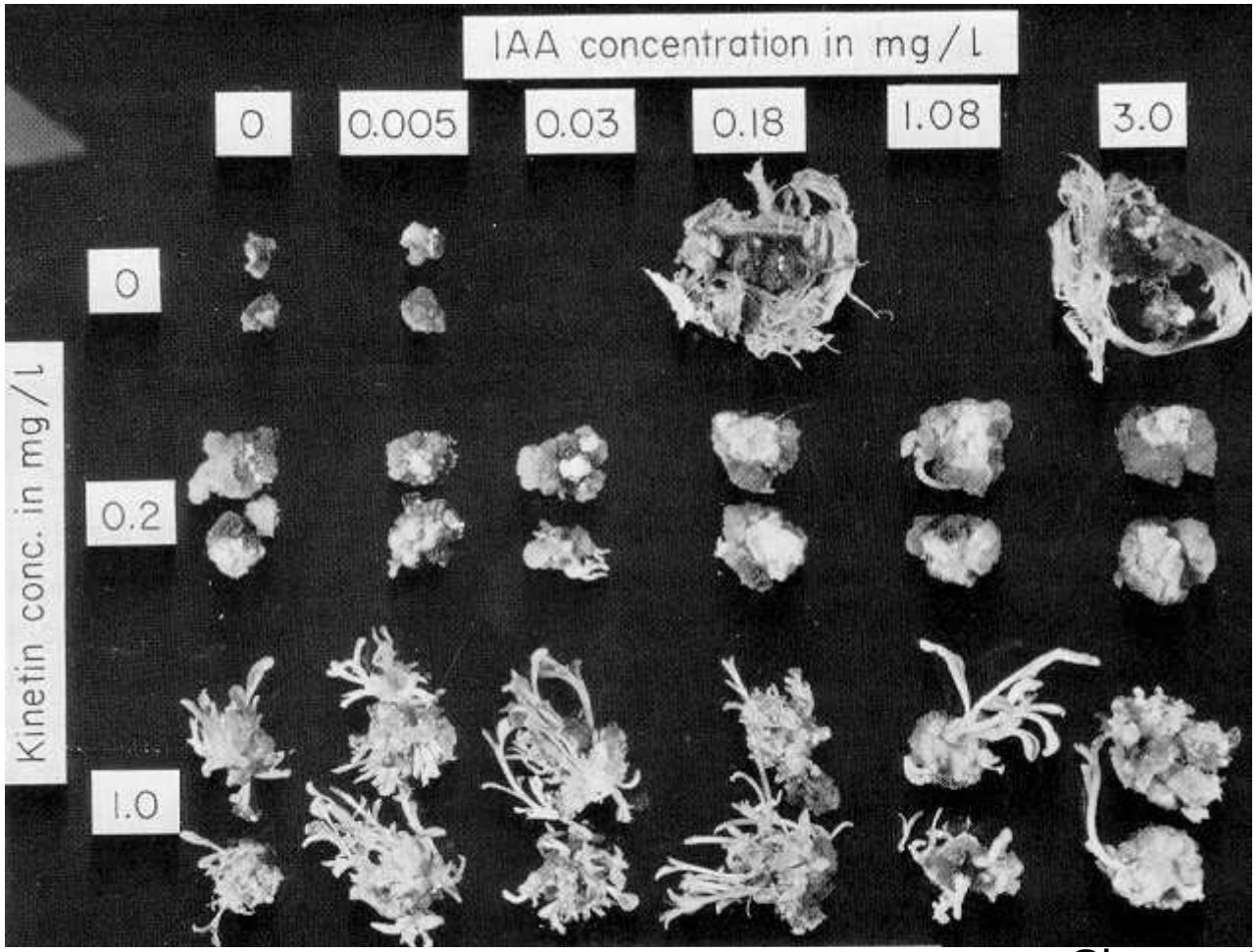
Kakimoto 2004

Cytokinin –
role in plant development

Auxin and cytokinin

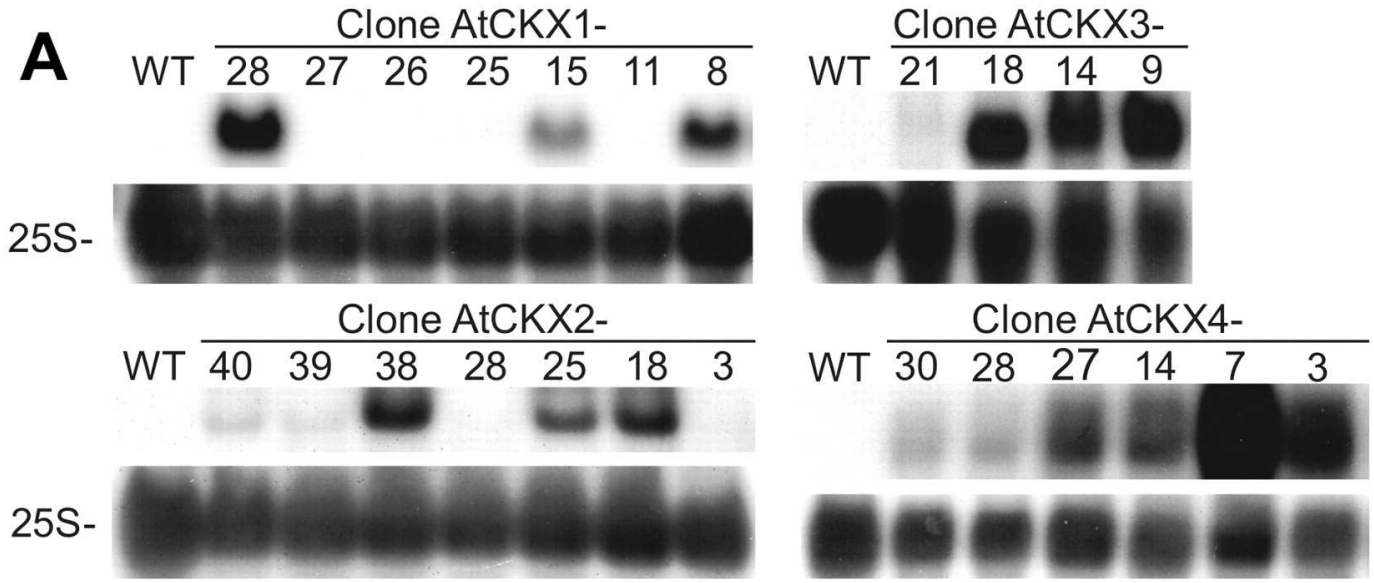
roots

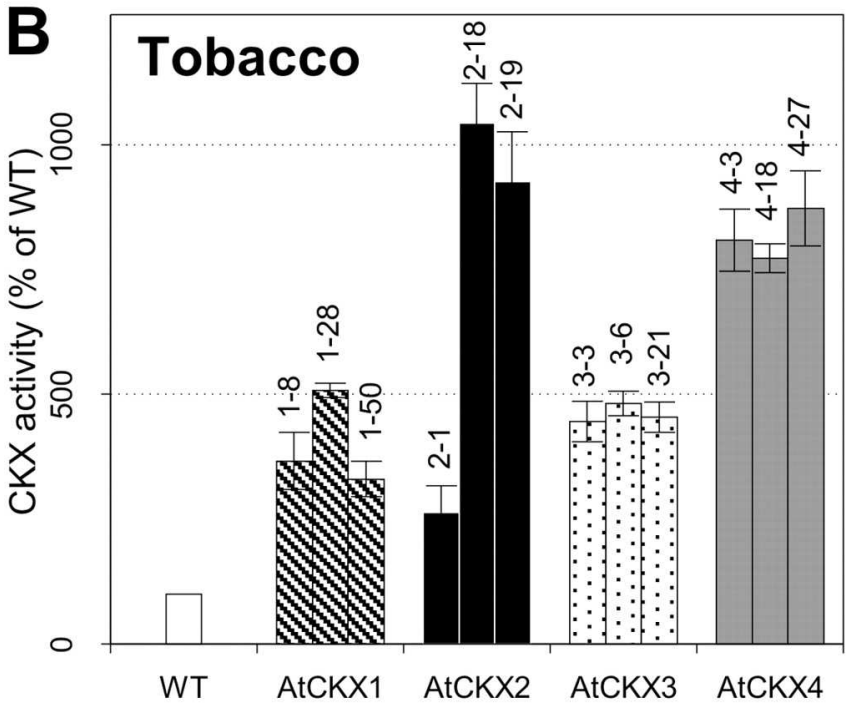
shoots

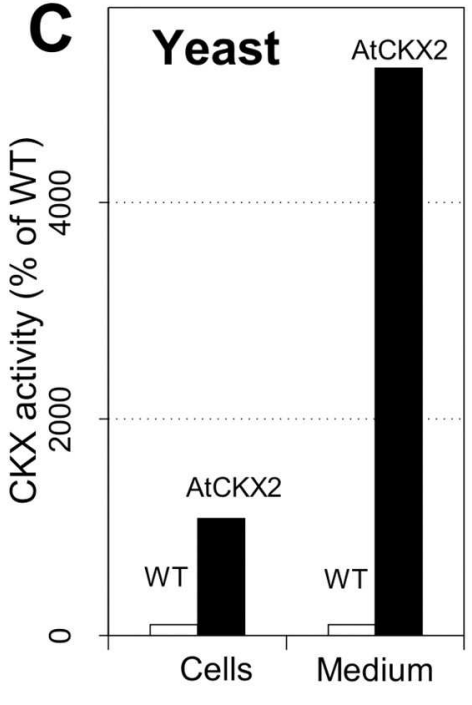


Skoog *et* Miller, 1957

Isolation of CK-oxidase (AtCKX)

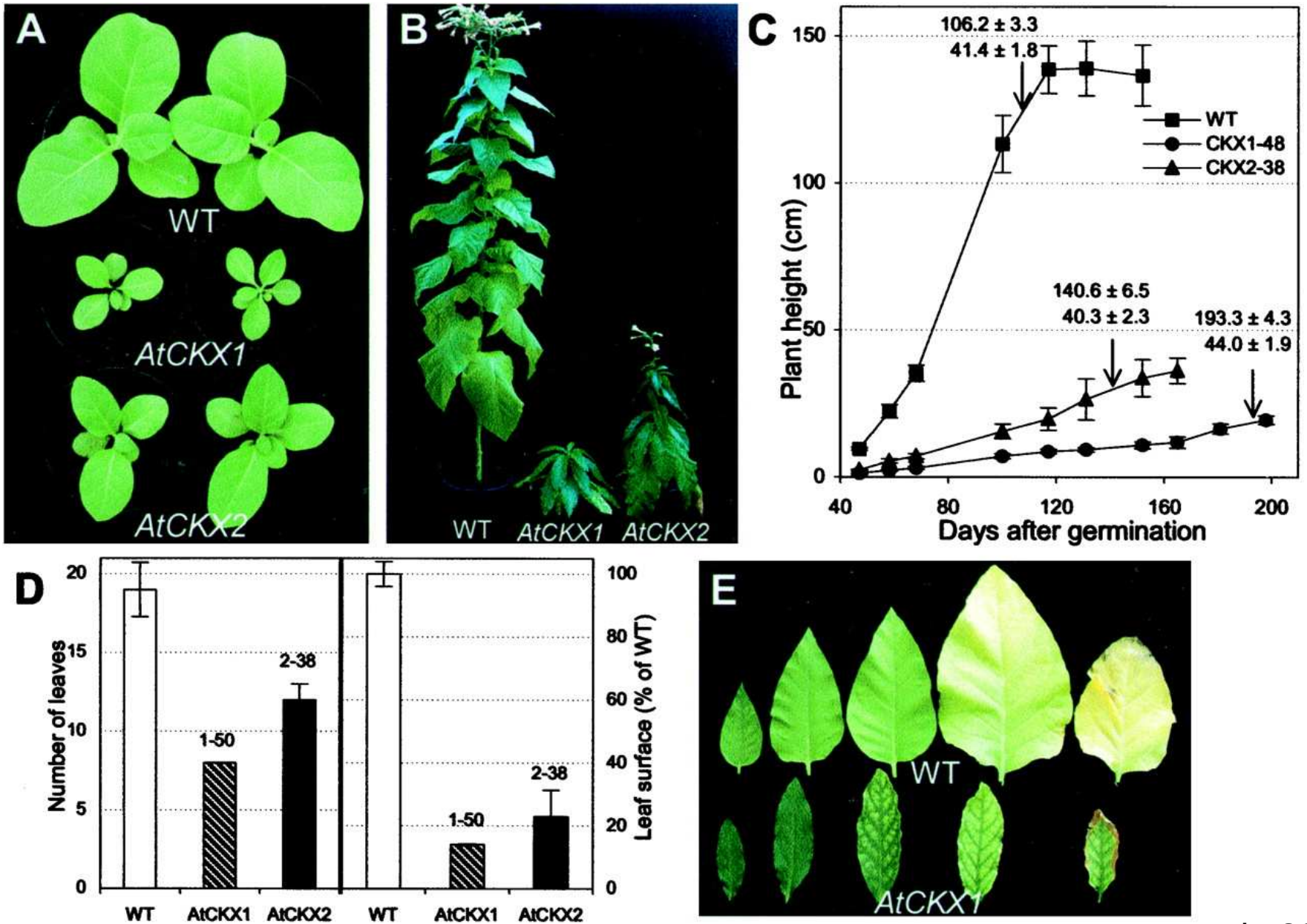






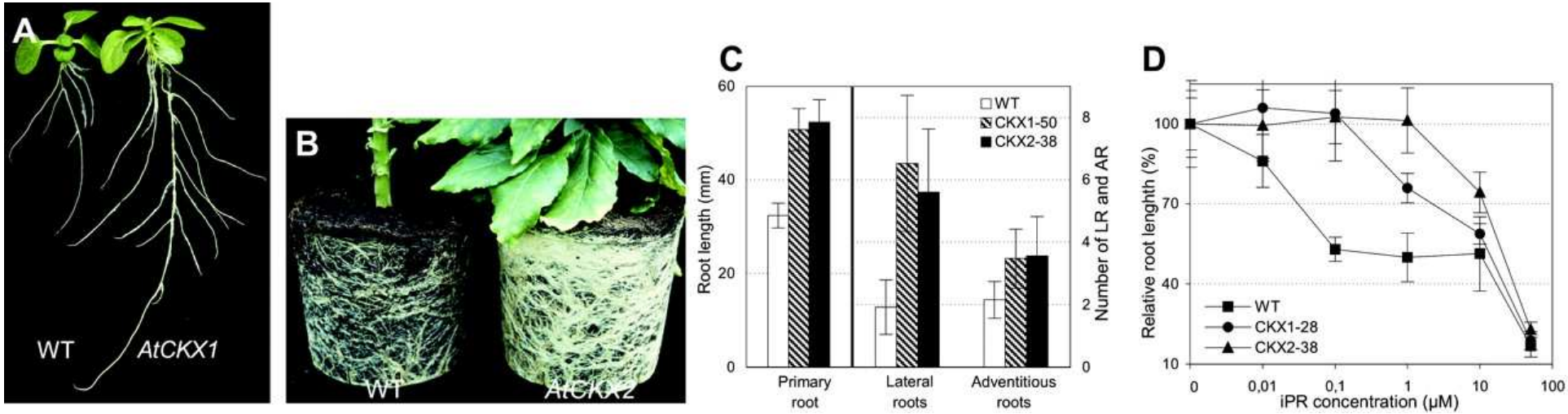
Werner et al., 2001

*AtCKX*s overexpression in tobacco



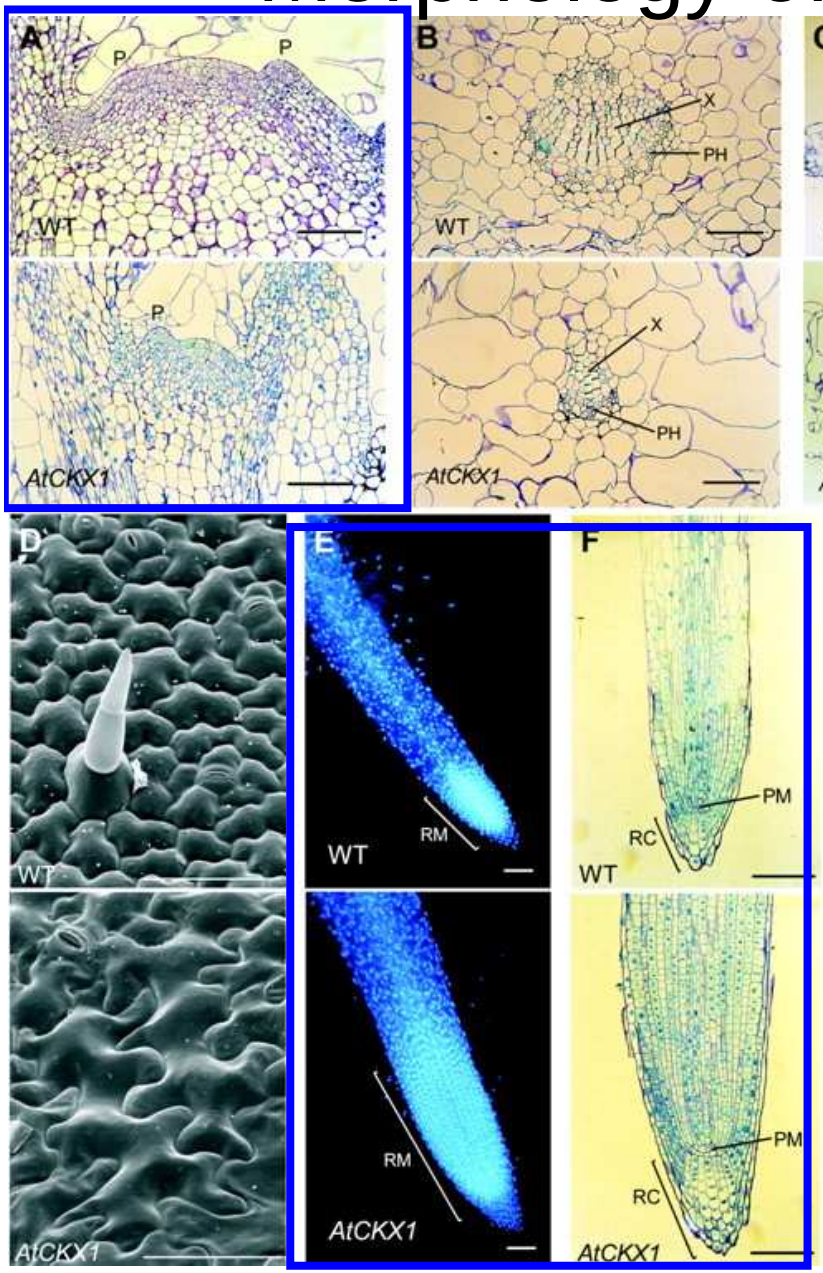
Werner et al., 2001

Effect of AtCKX on tobacco root



Werner et al., 2001

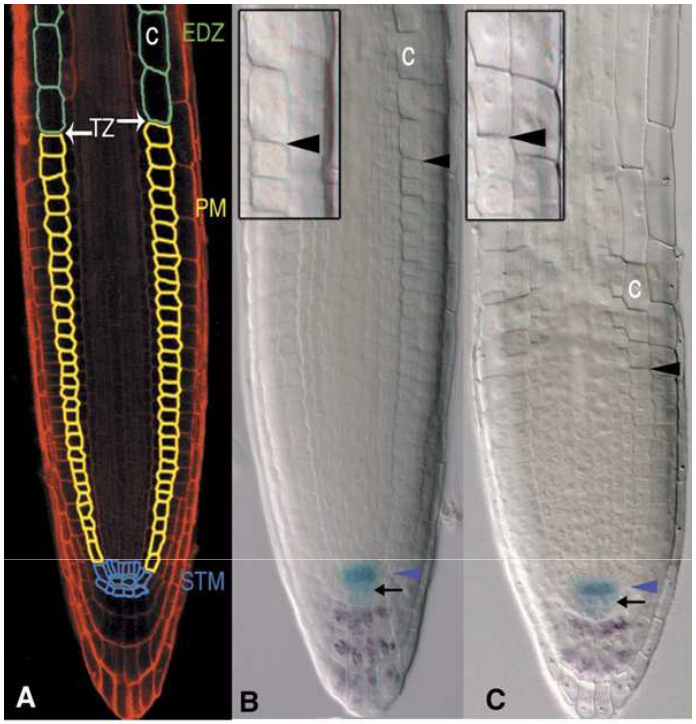
Morphology of AtCKX tobacco plants

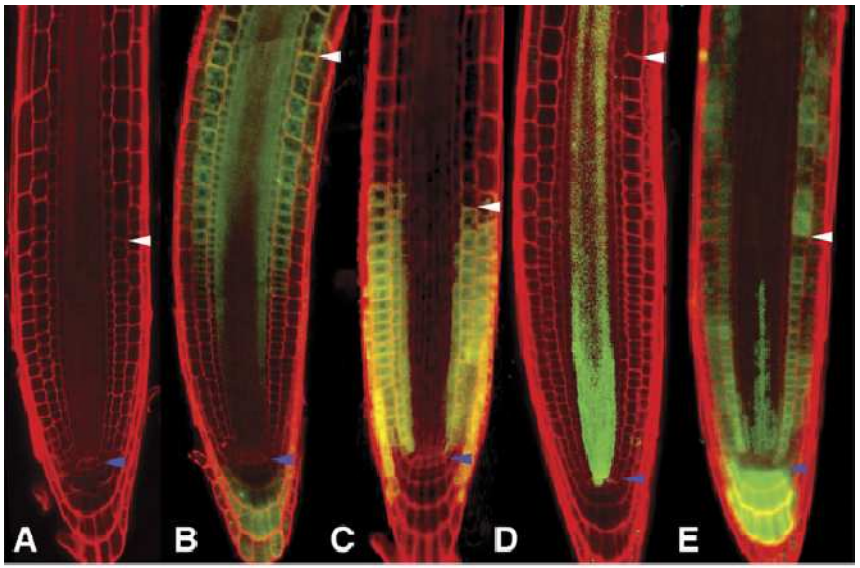


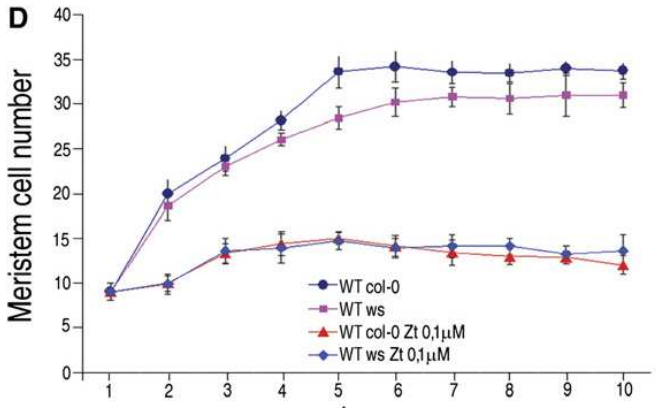
CK in shoot – positive regulator
CK in root – negative regulator

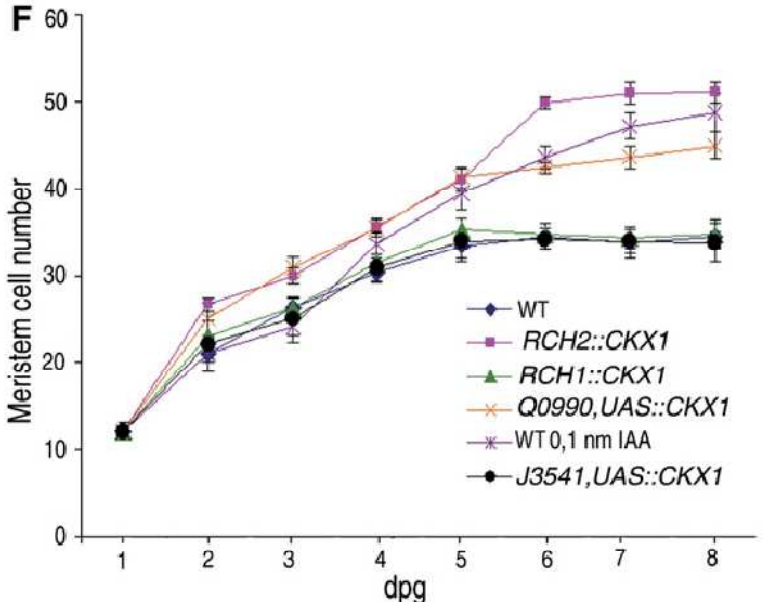
Werner et al., 2001

Cytokinin – root meristem development



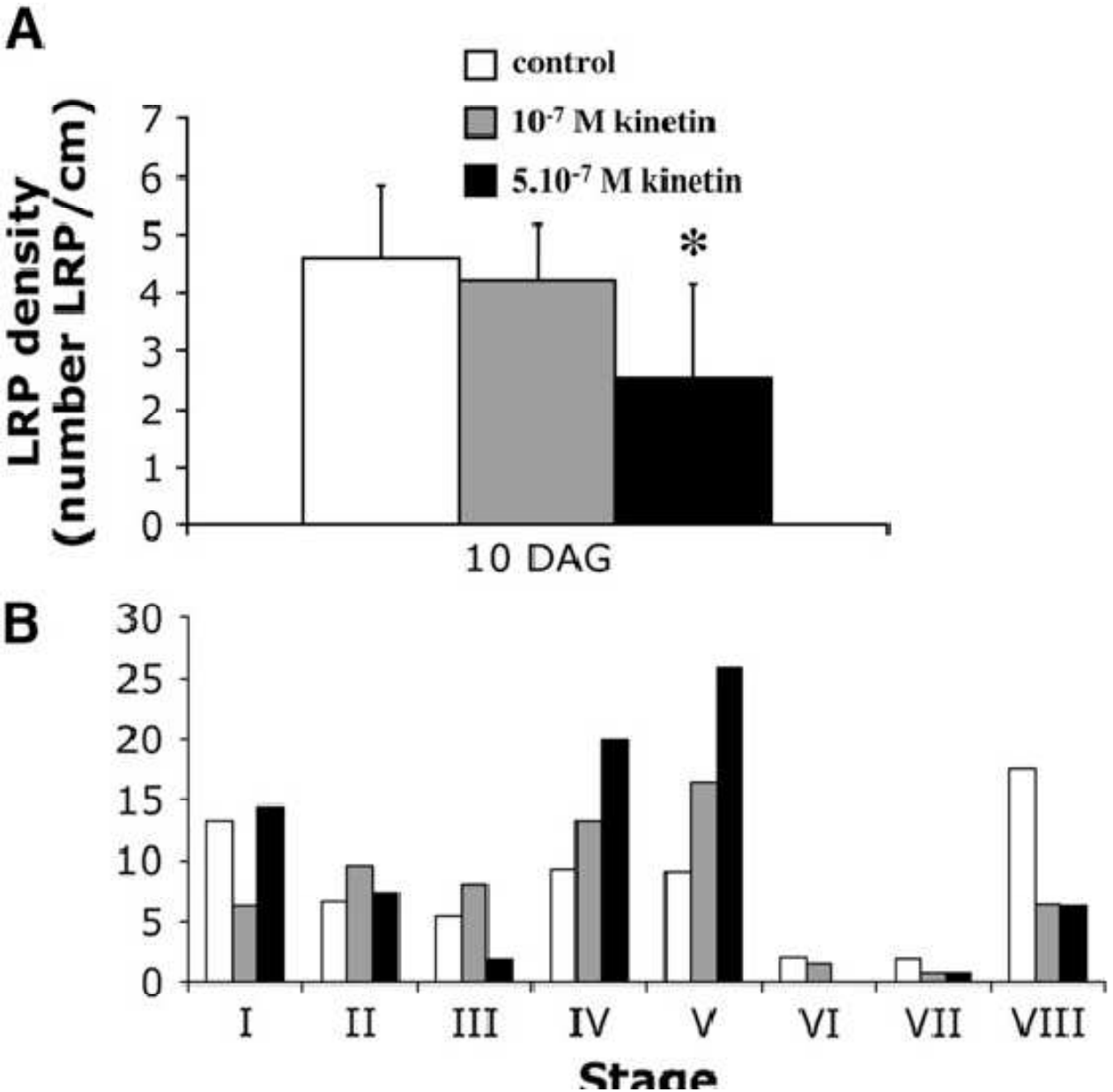






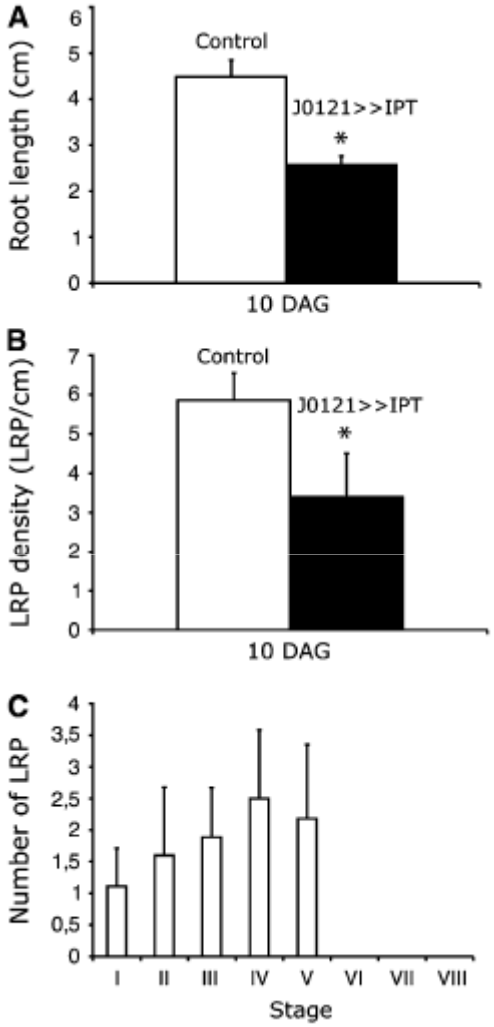
Dello Ooio et al., 2007

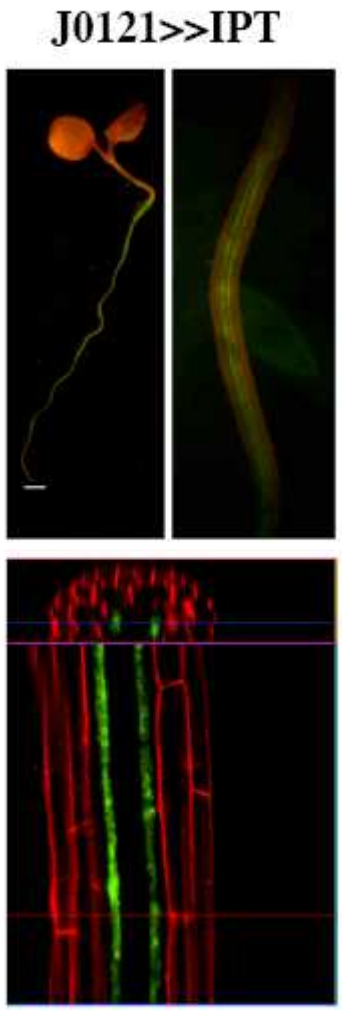
Cytokinin - lateral root organogenesis

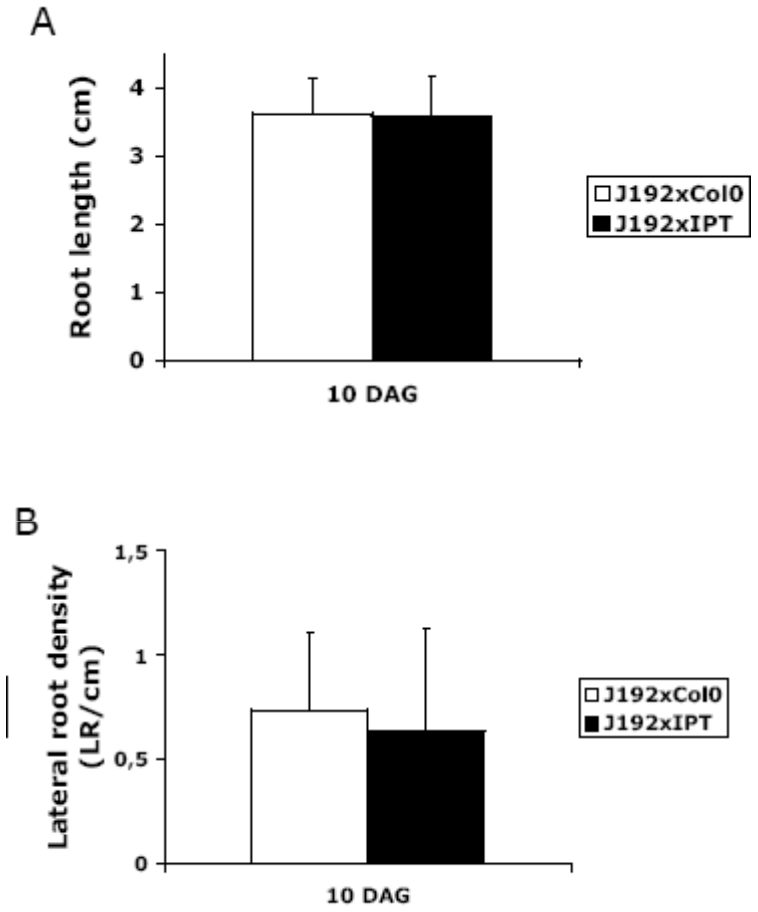


Laplaze et al., 2007

Cytokinin effect is stage specific



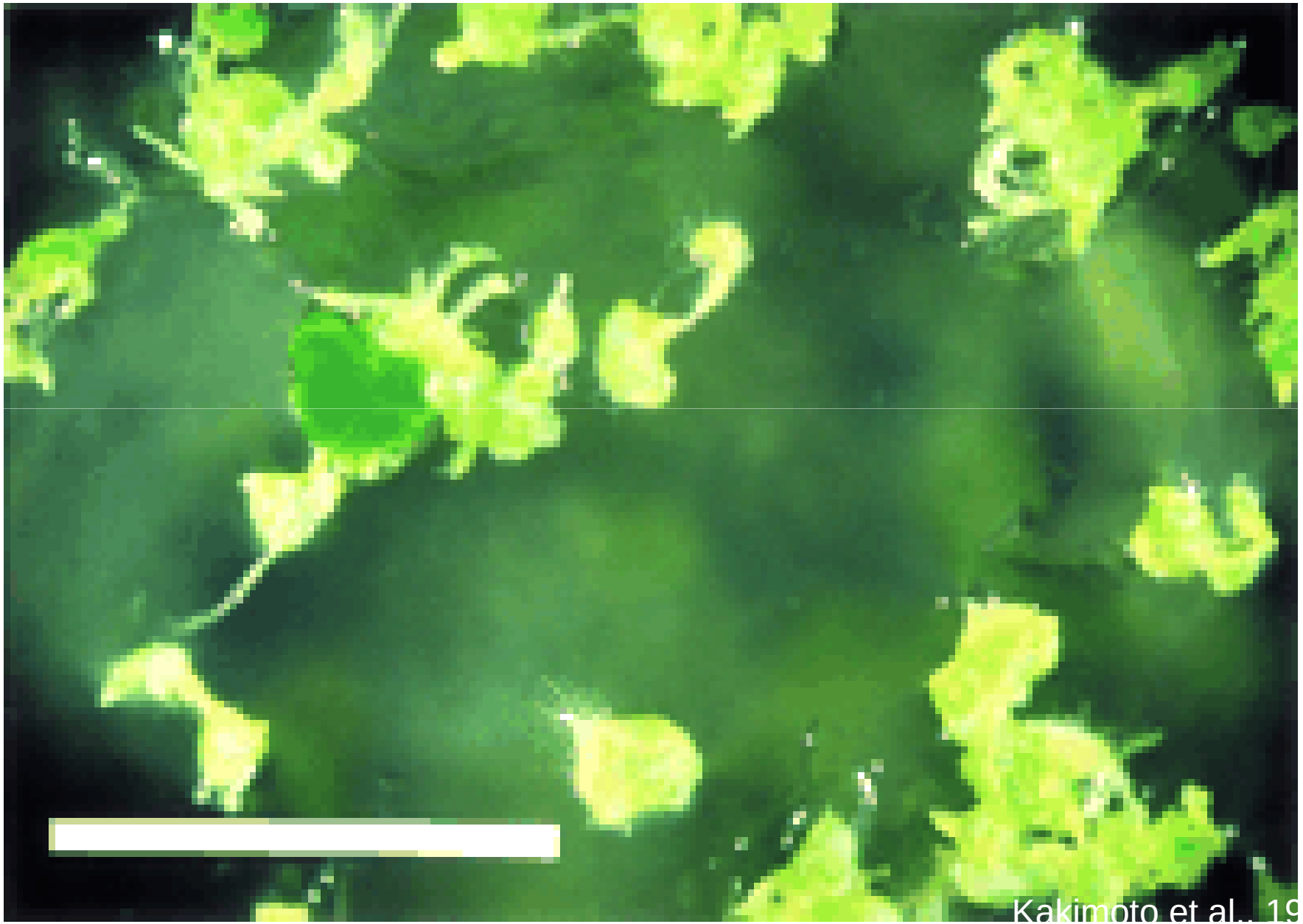




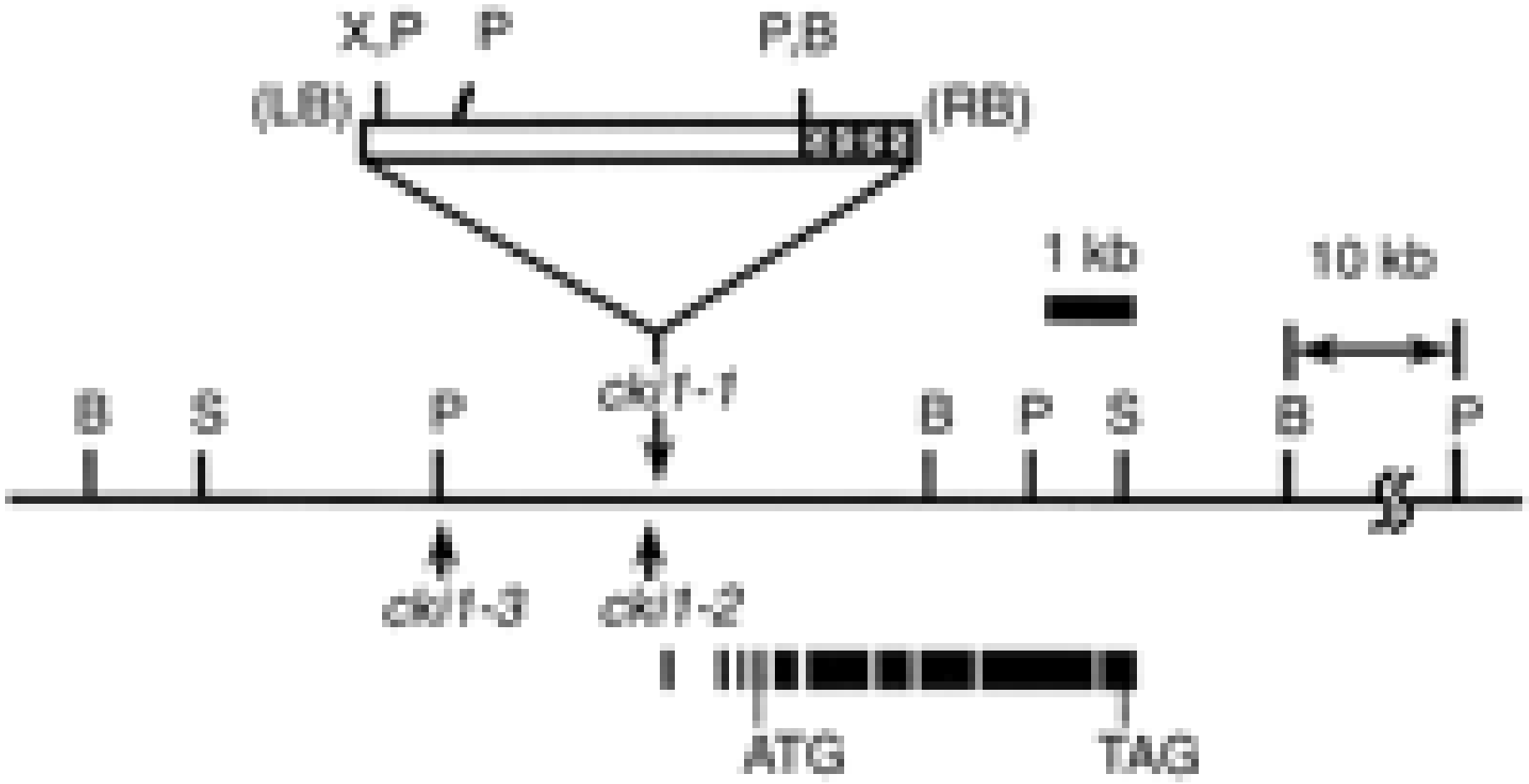
Laplaze et al., 2007

Cytokinin
–signal perception and transduction

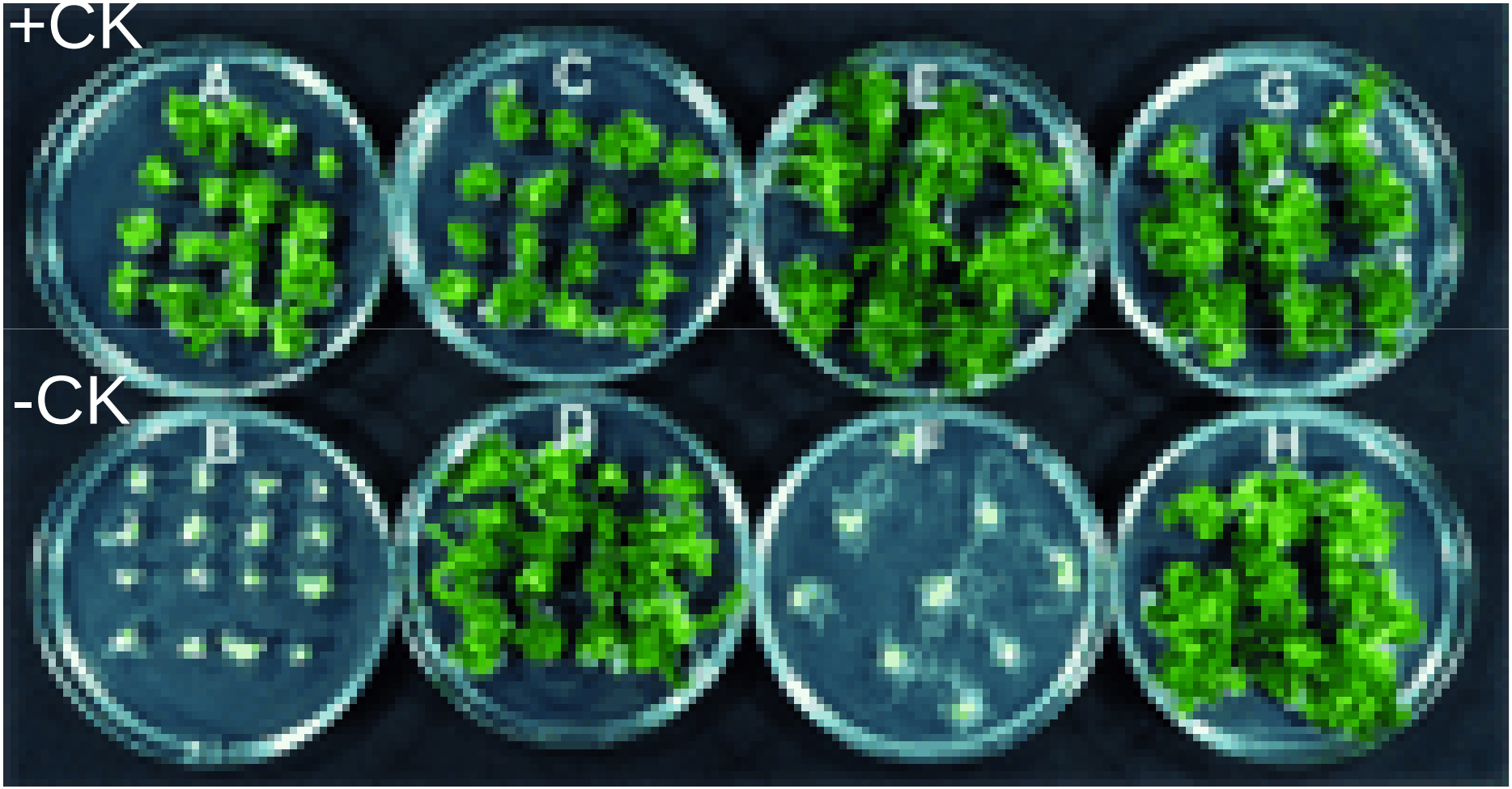
Isolation of CK independent (*cki1*) mutant



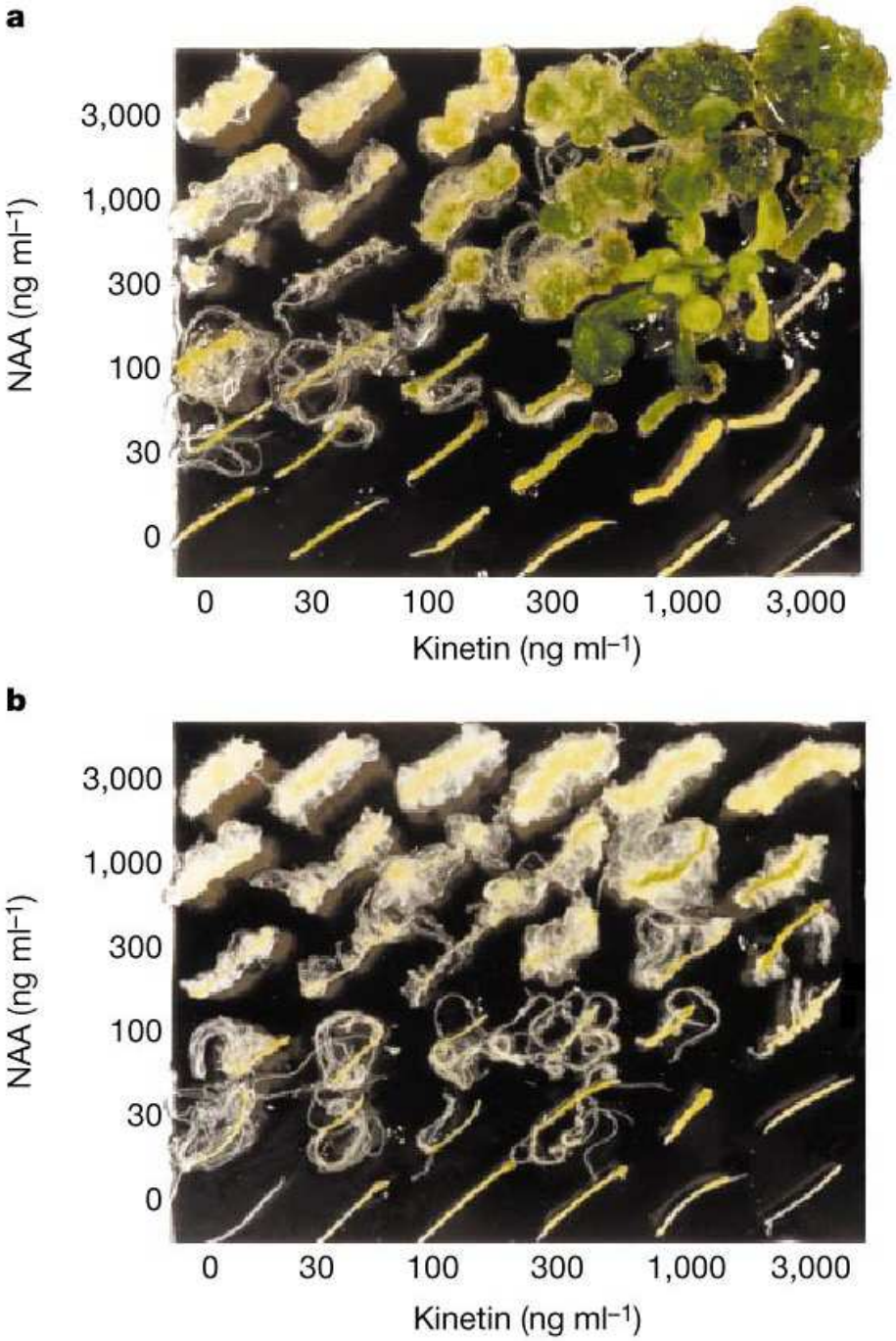
Identification of *CKI1* gene



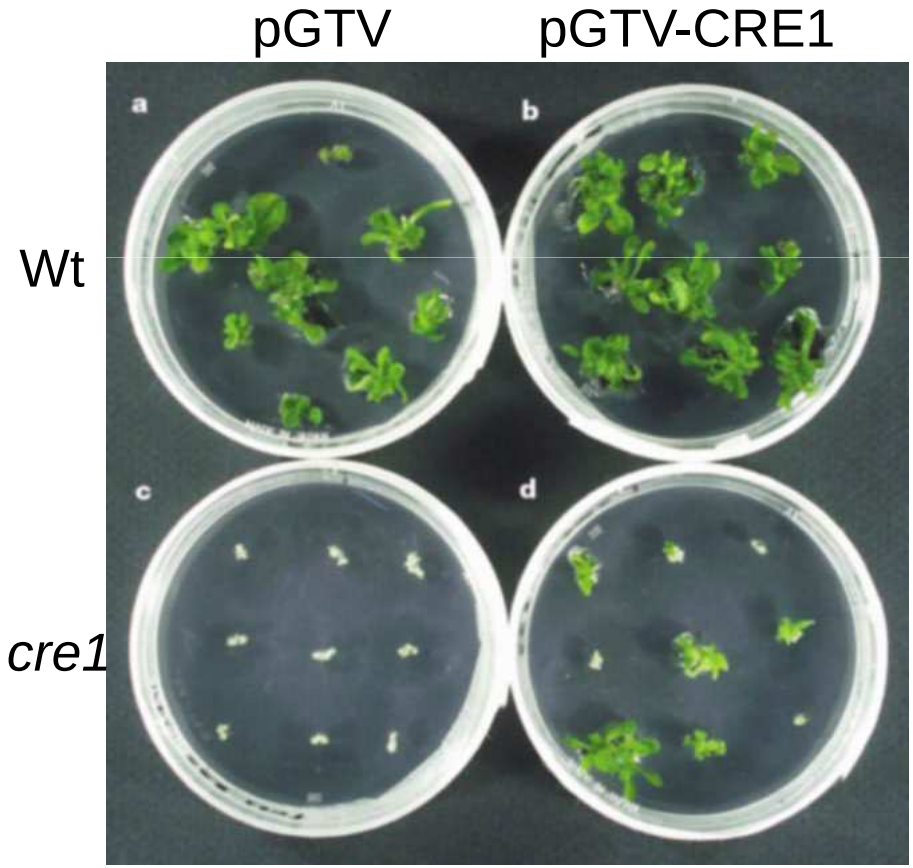
Verification - *35S::CKI1* transgene



Kakimoto et al., 1996

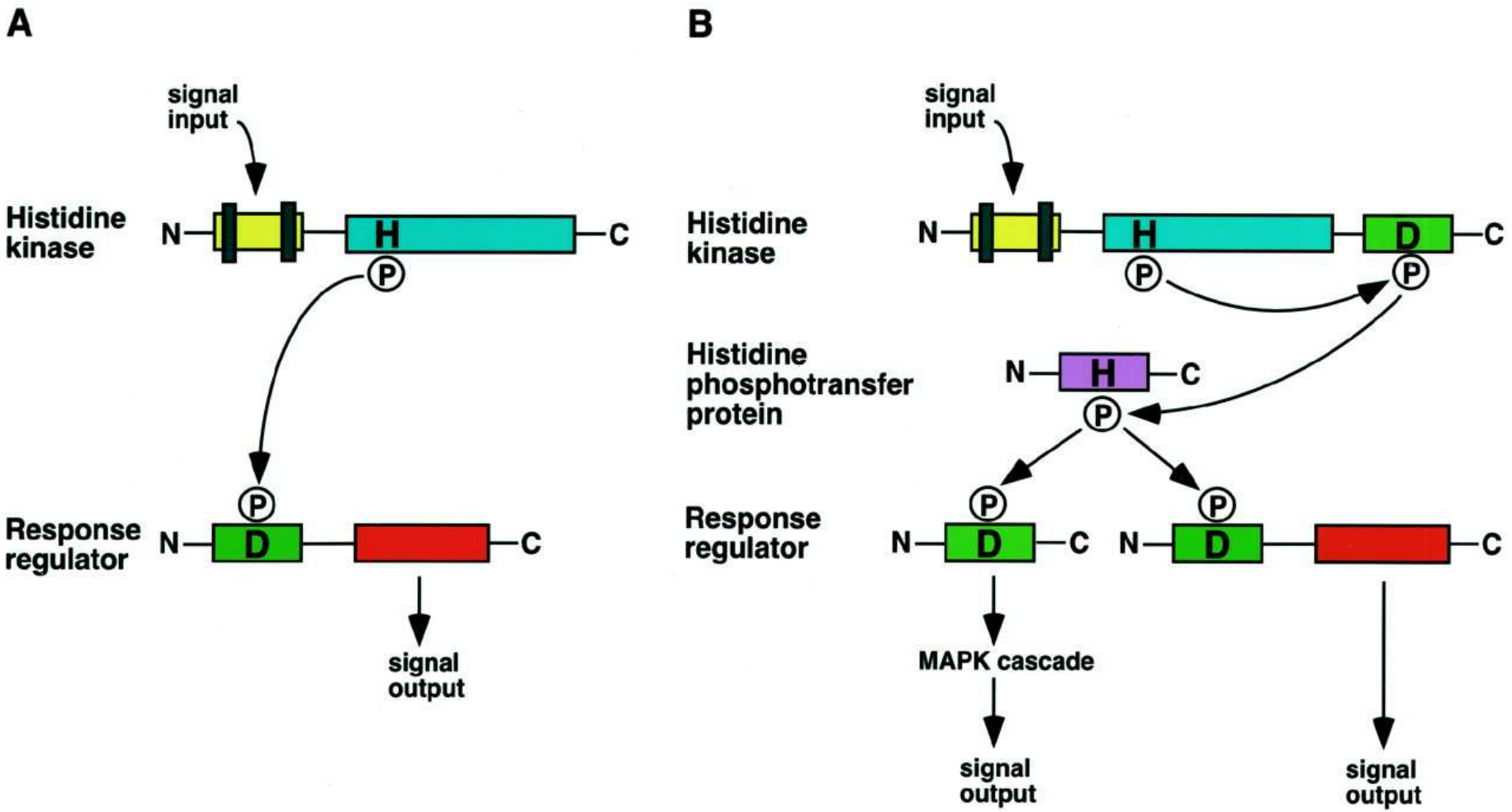


Next strike
- CK response mutant (***cre1***)

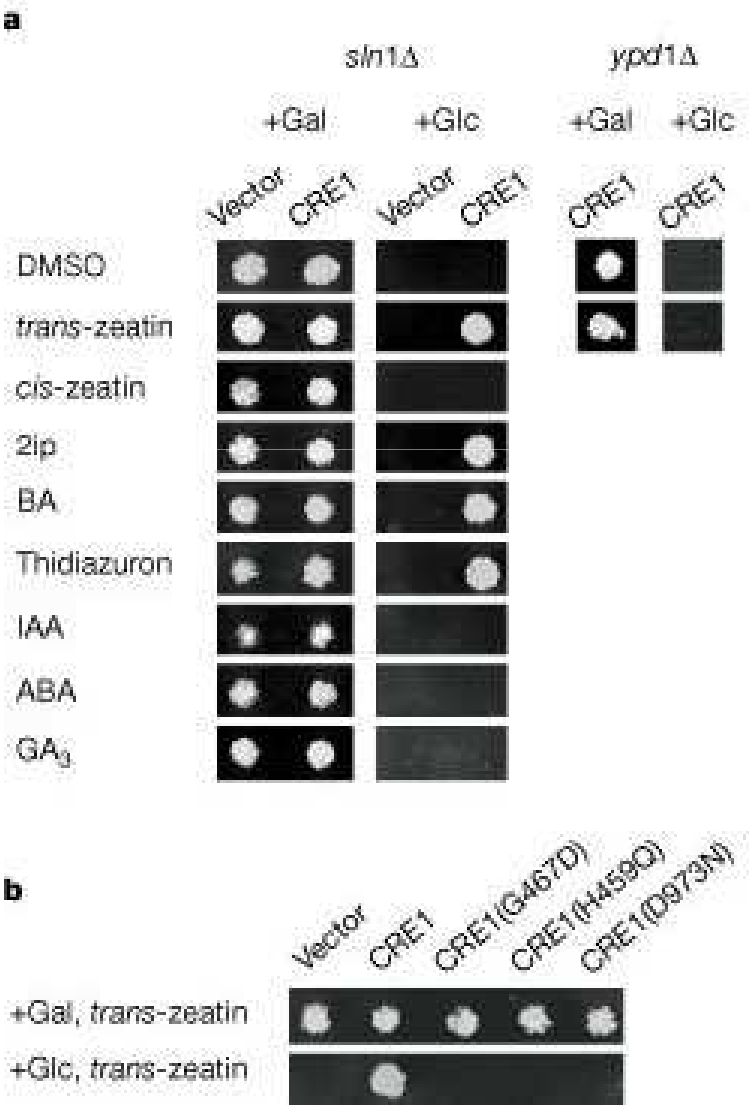


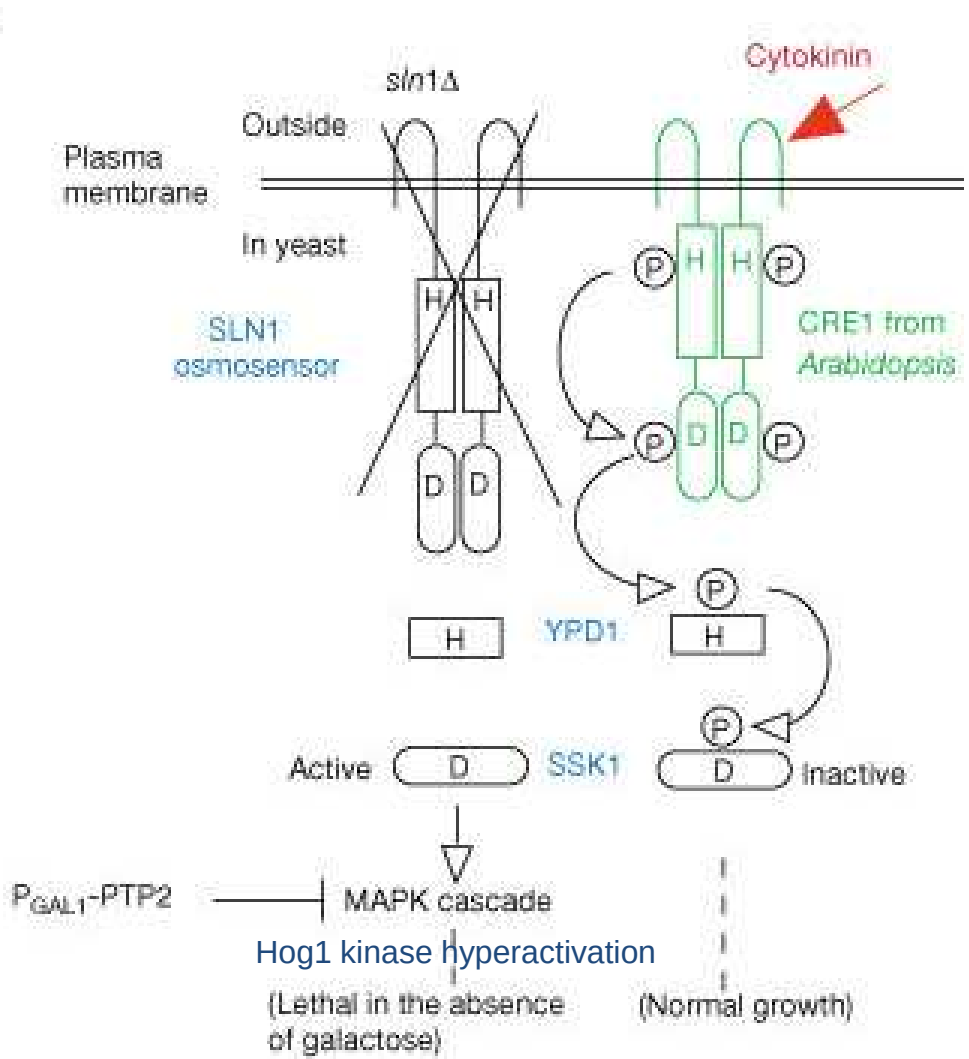
Inoue et al., 2001

His kinase transduction pathway



Piece of genius - complementation





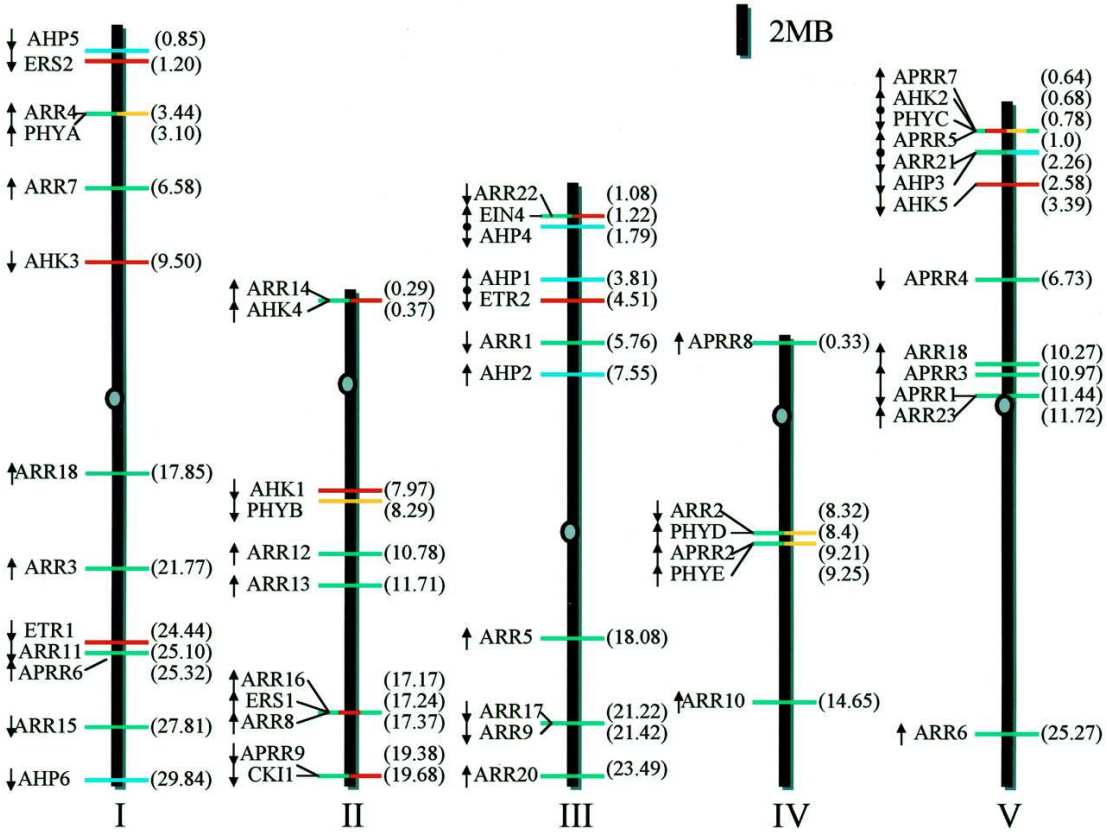
Inoue et al., 2001

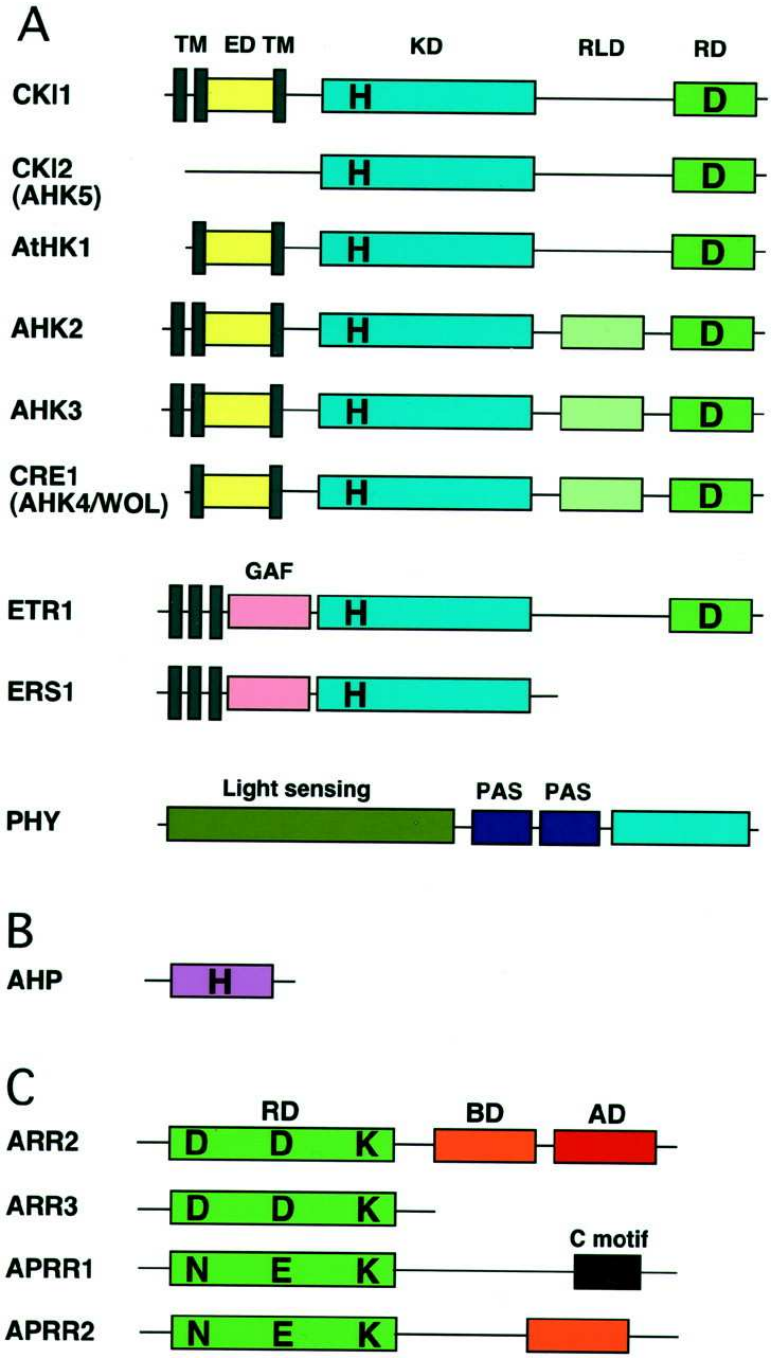
Cytokinin receptors – what else?

3 CRE1 homologous proteins (AHKs)
multiple mutant phenotypes – additive
(not lethal)

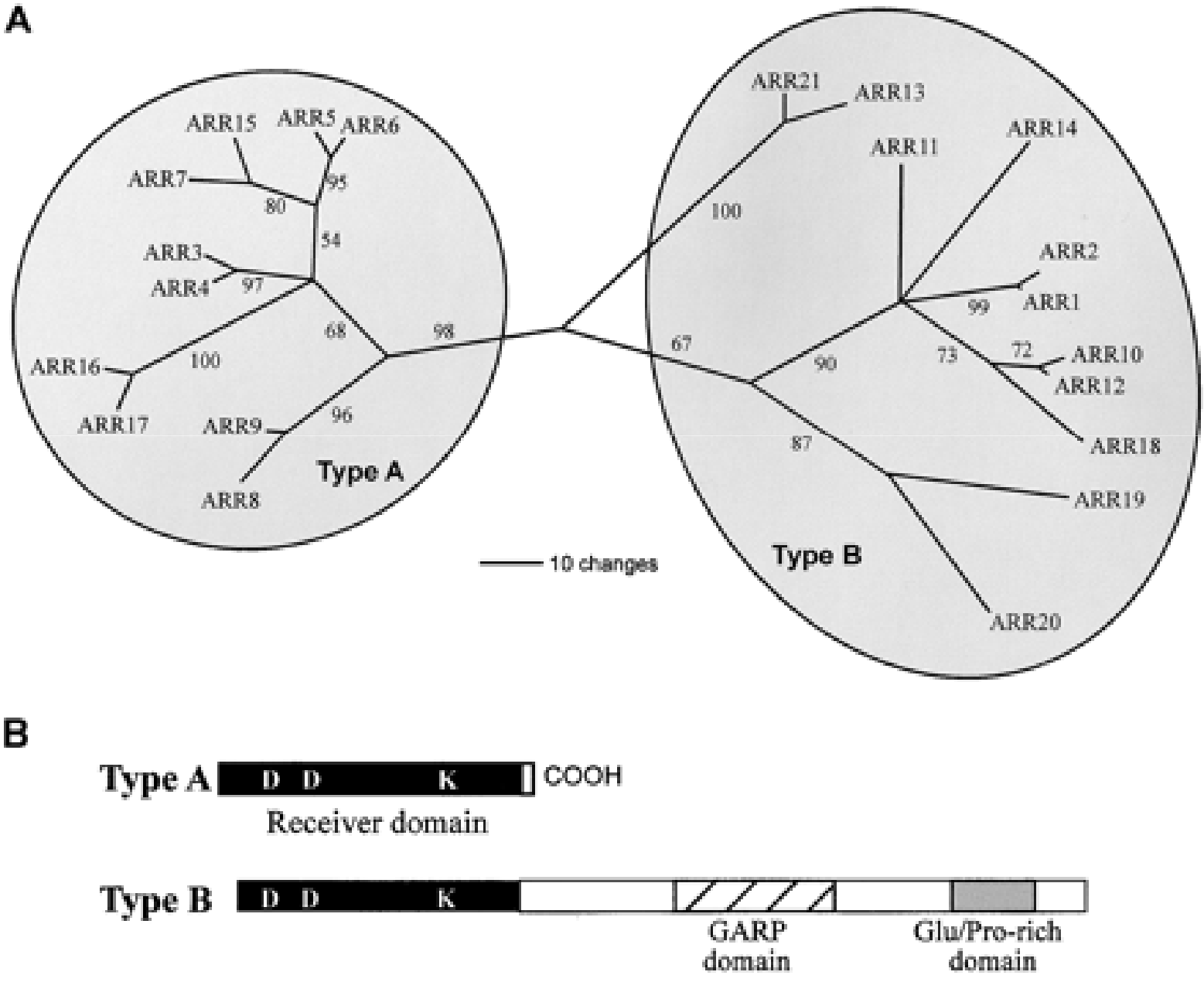
What does CKI1?

His kinase pathway components in Arabidopsis

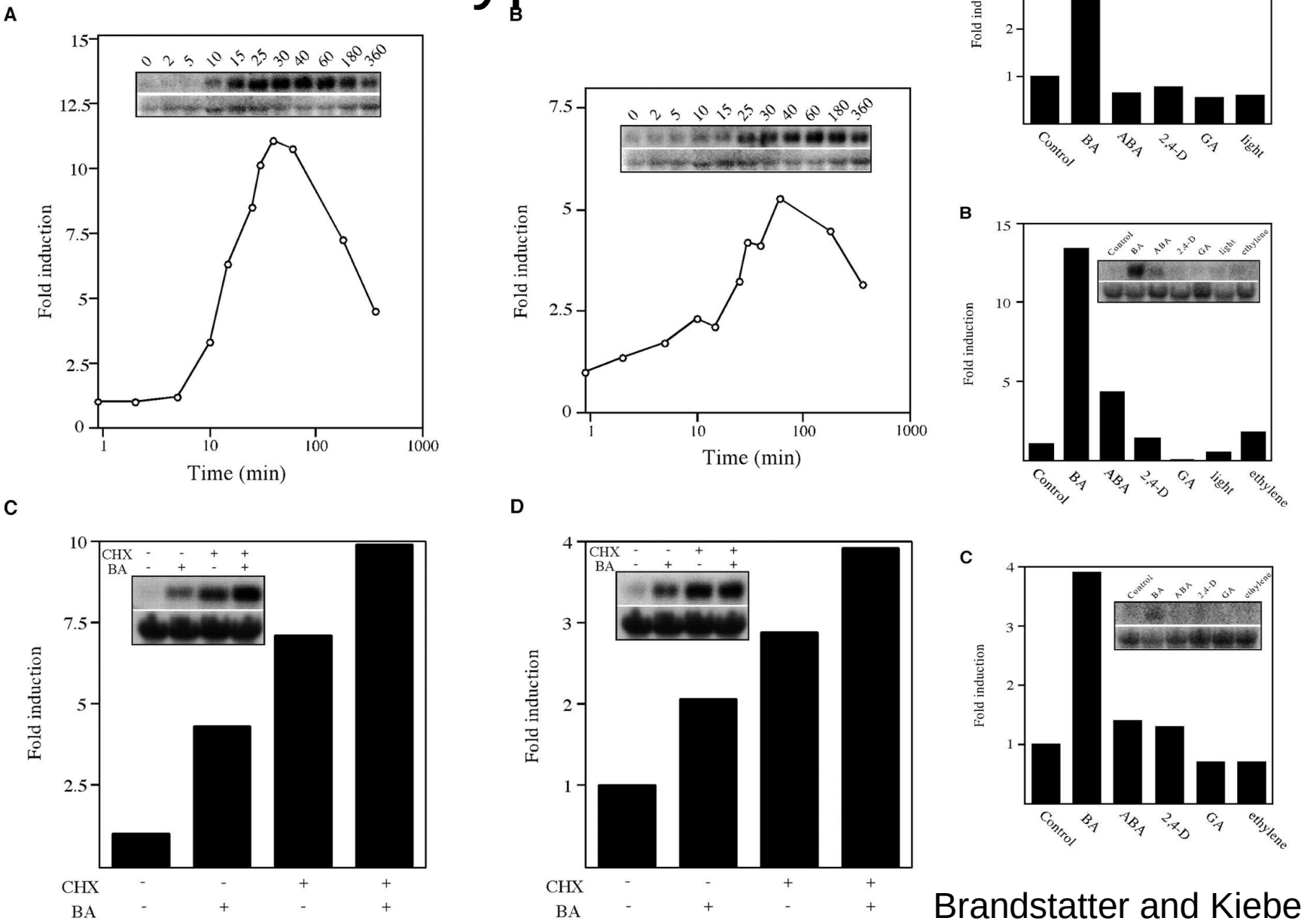




Response Regulators in *Arabidopsis*

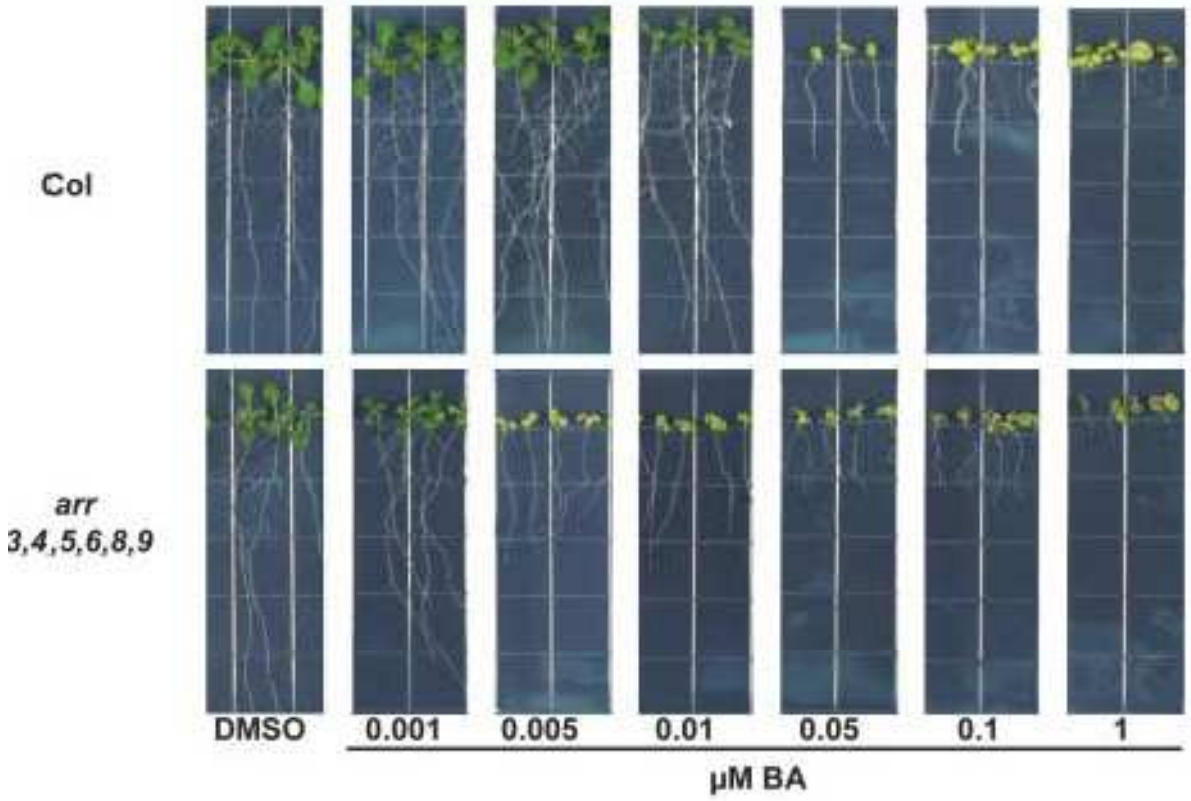


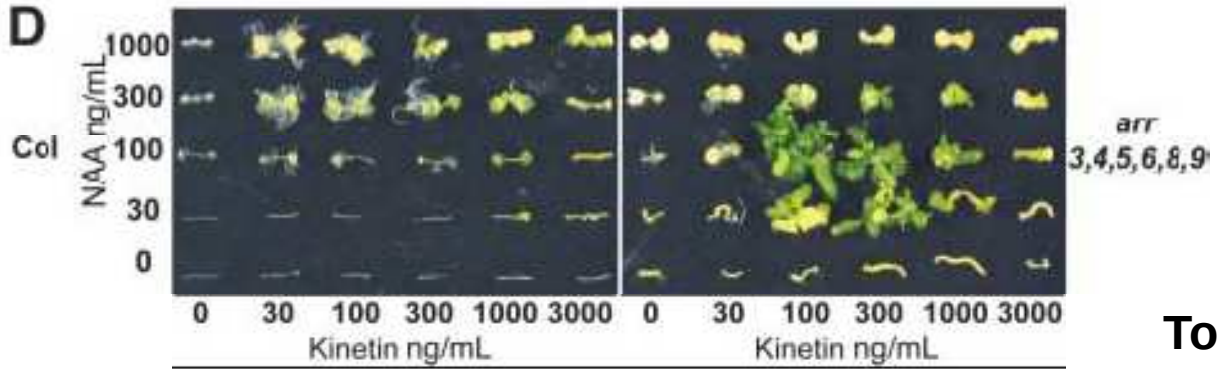
CK responsive genes – *ARR* type A



Brandstatter and Kieber, 1999

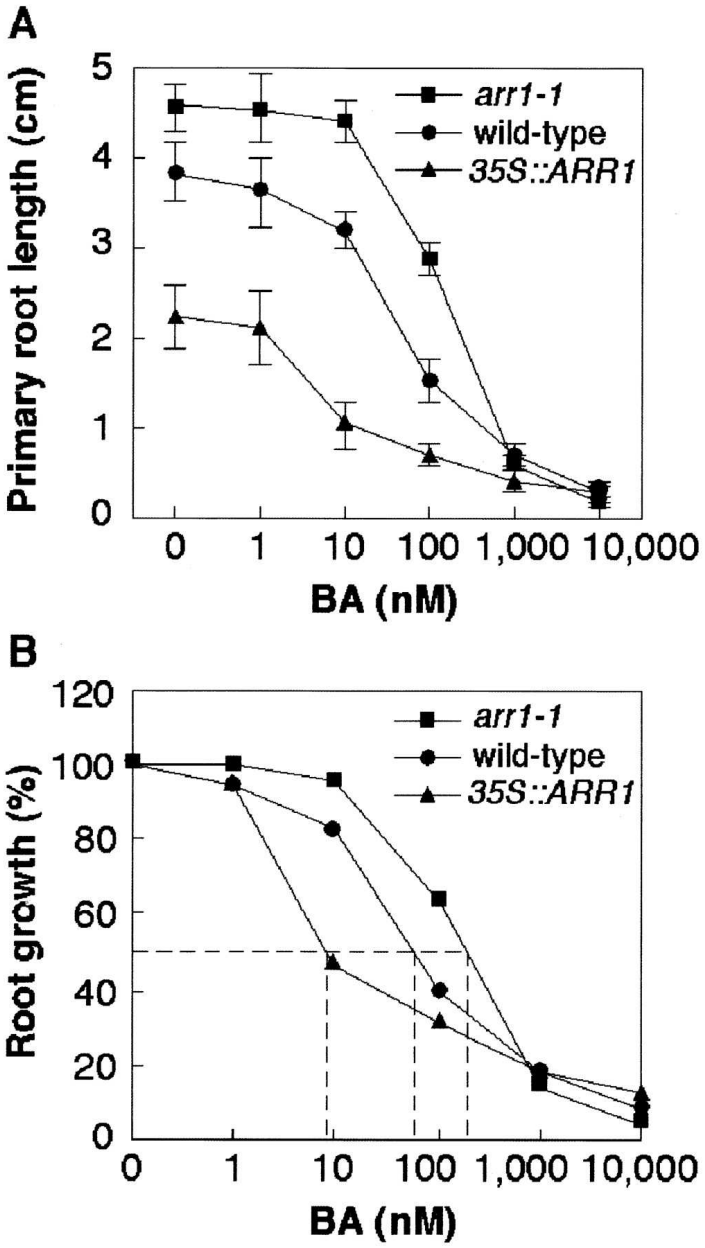
Response regulator ARR type A-negative regulators of cytokinin signalling

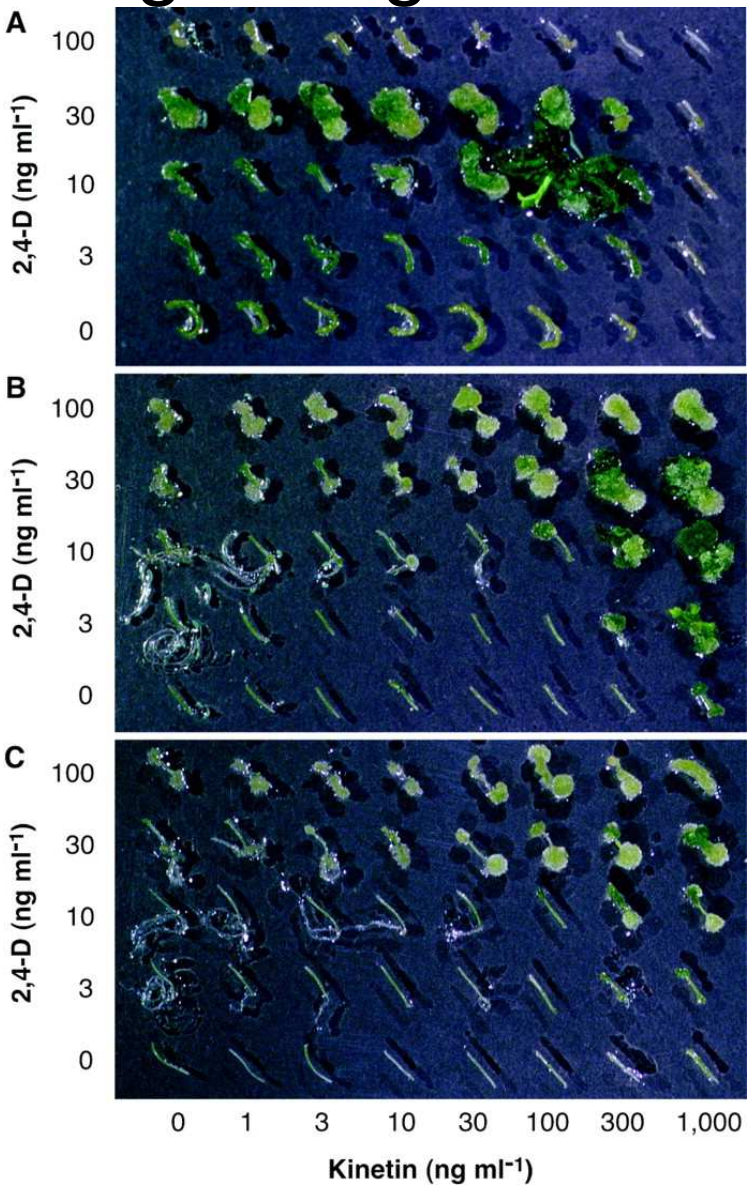




To et al., 2004

Phenotypes of *arr* type B positive regulator of CK signalling





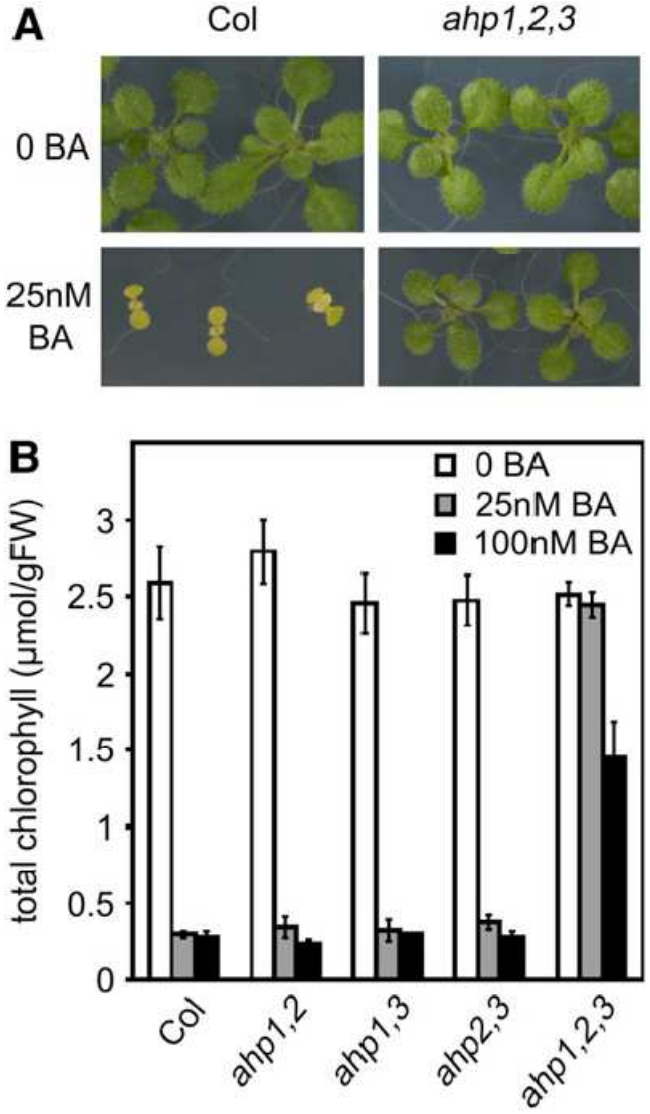
35S::ARR1

wt

arr1-1

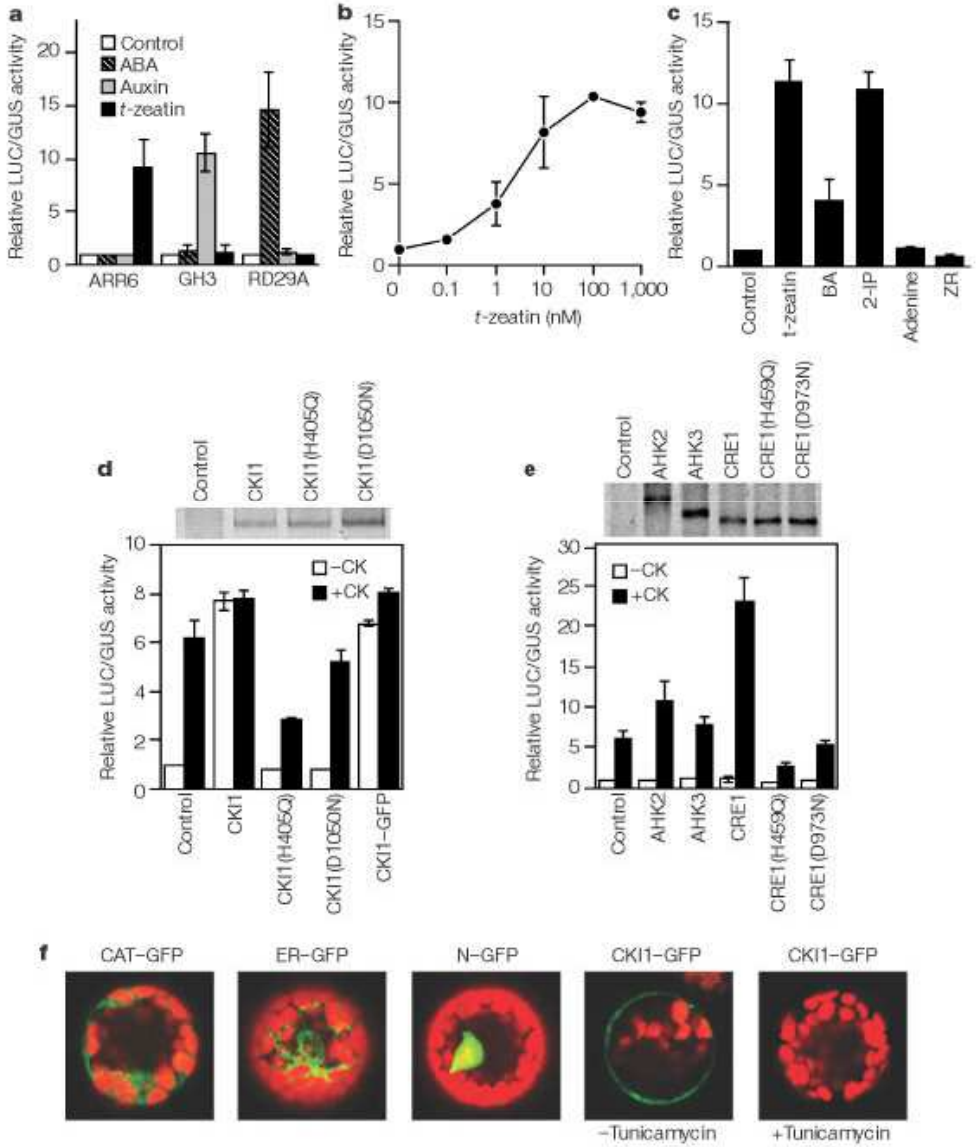
Sakai et al., 2001

AHPs mediate transfer of cytokinin signal between cytoplasm and nucleus

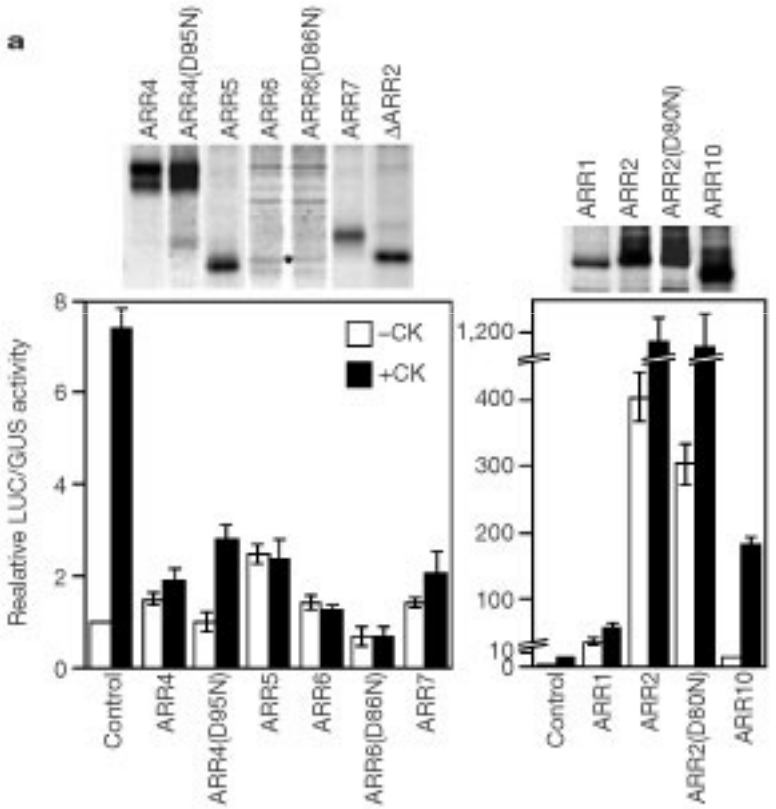


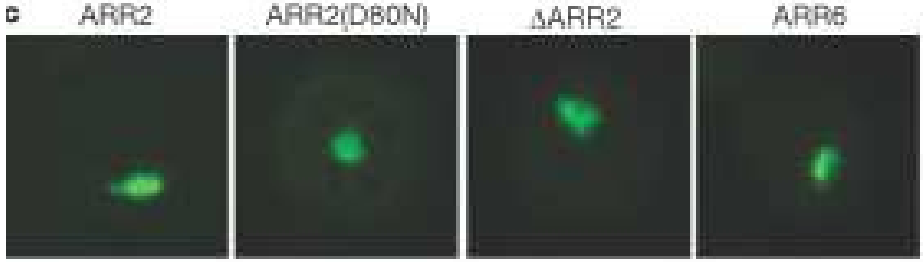
Hutchison et al.,2006

Games with protoplasts

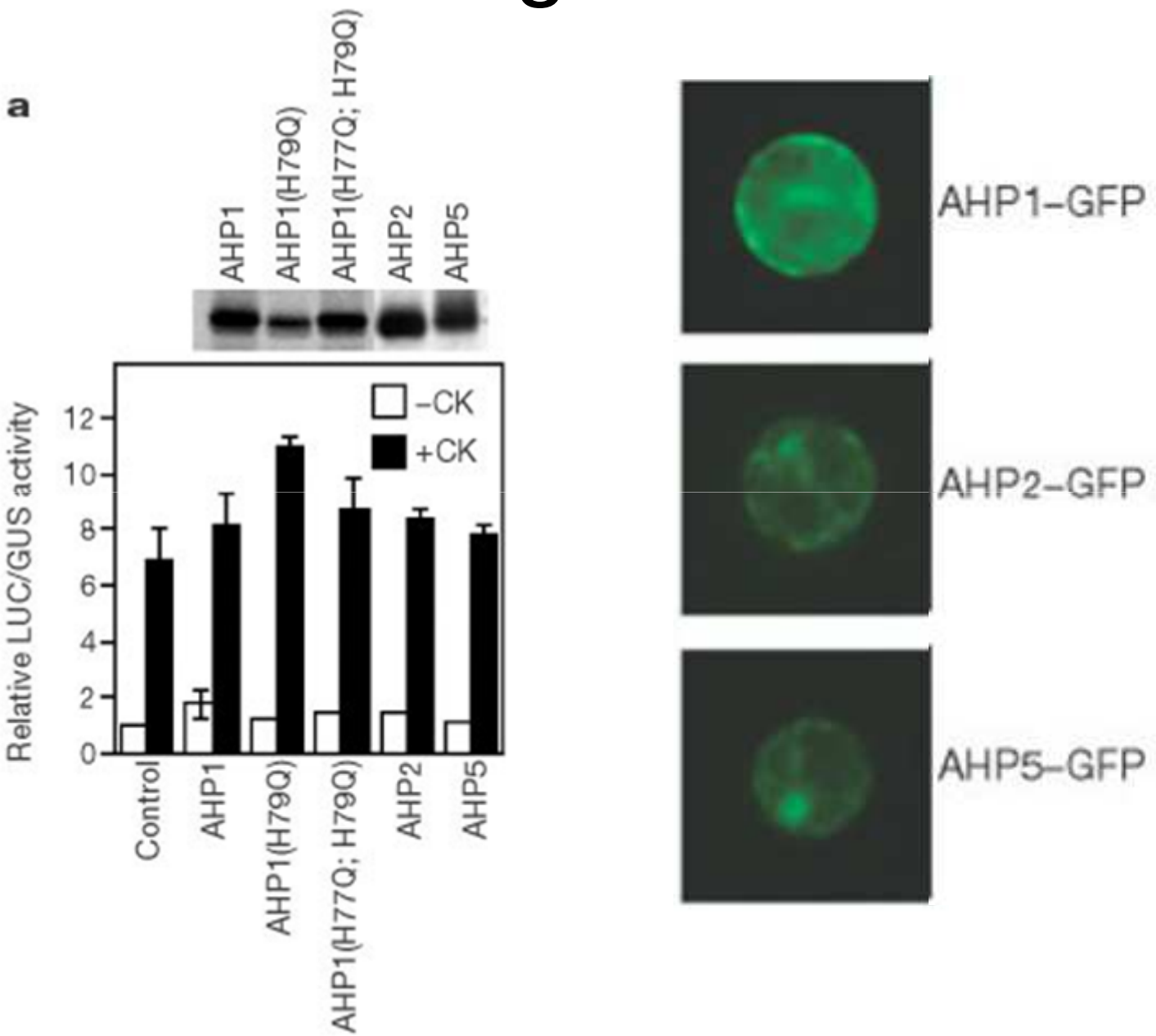


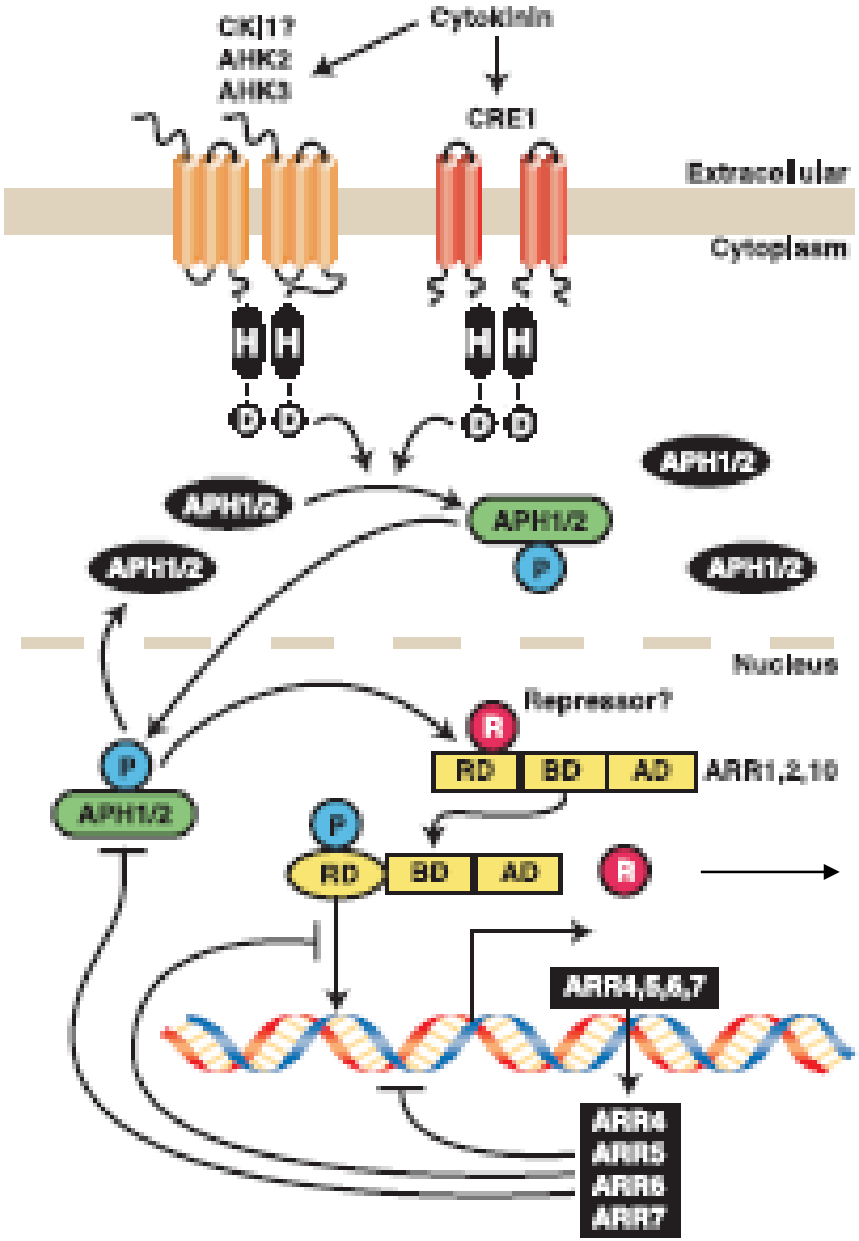
Opposite effects of two classes of ARRs on CK signalling





AHPs – signal to nucleus



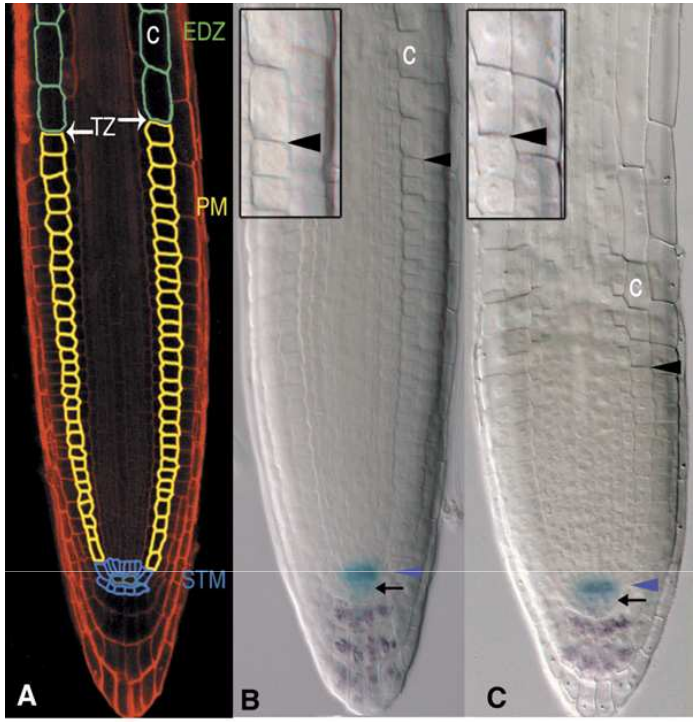


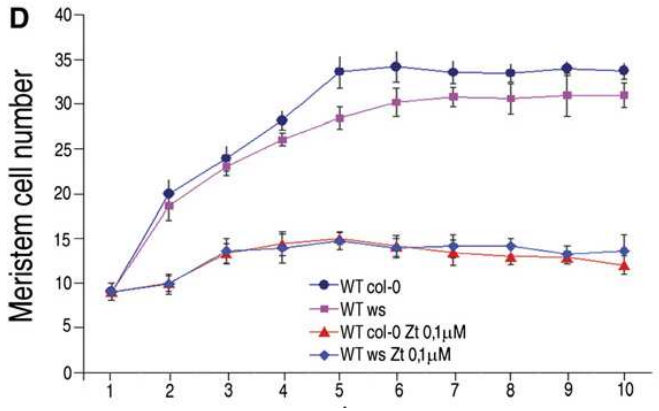
Hextuple of type A ARRs confirms role as negative regulators

e.g. Root elongation assay

Transcritpion
Cell division
Shoot formation
Delayed senescence
Vascular develppment

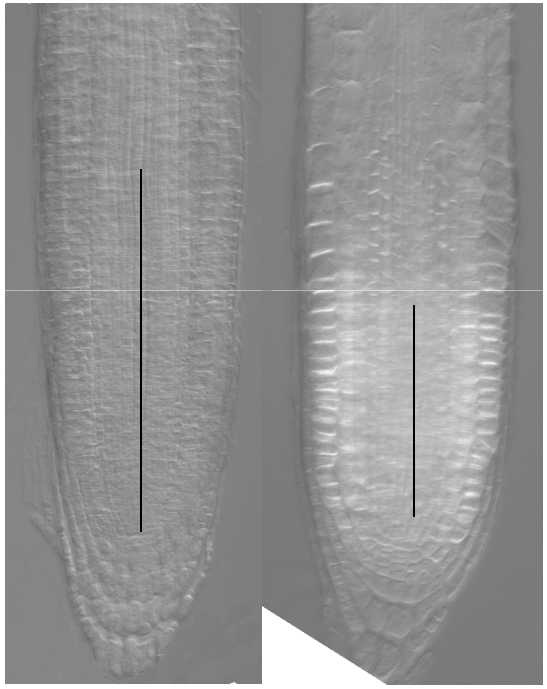
Cytokinin – root meristem development



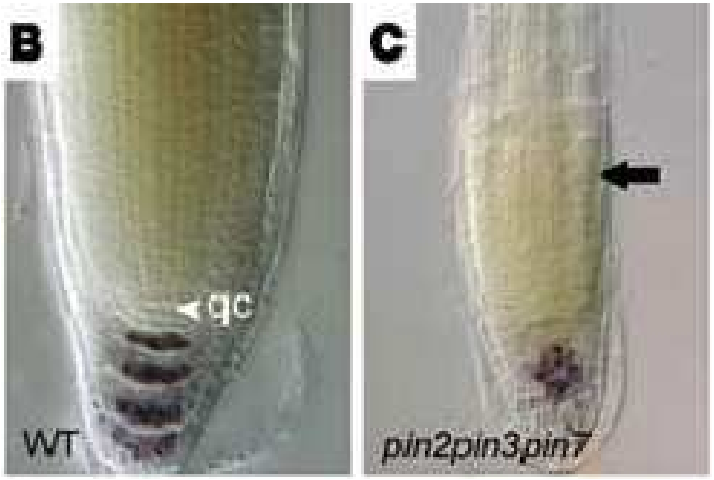


Dello Ioio et al., 2007

Auxin related mutants with short meristem



MS cytokinin

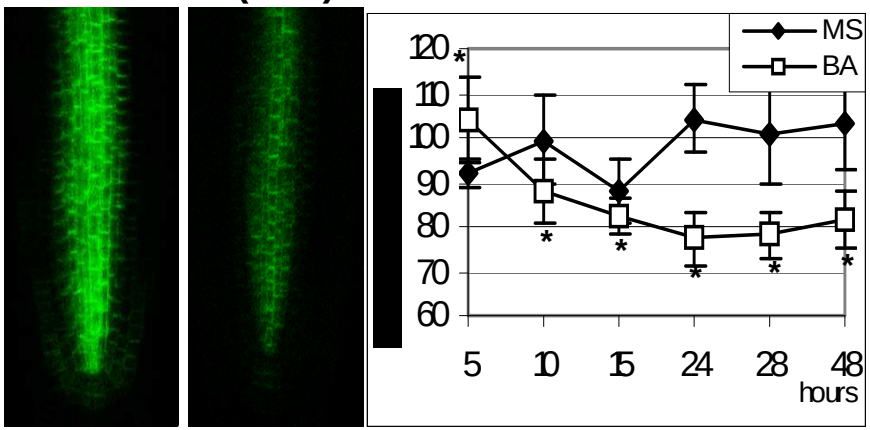


Blilou et al.,2005

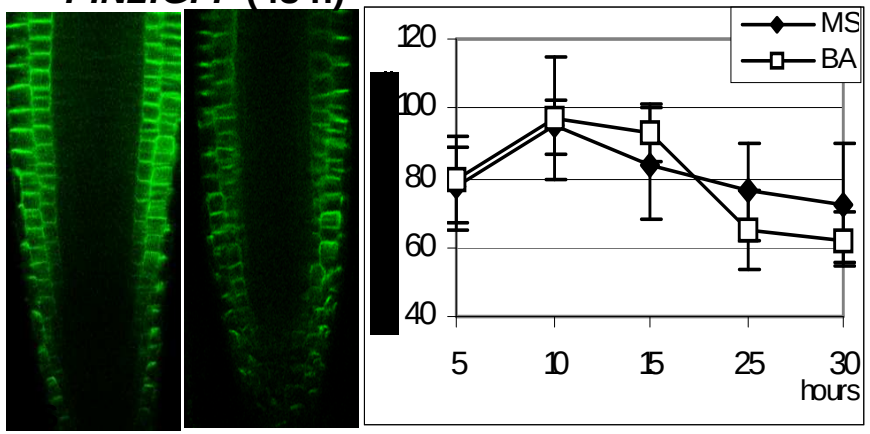
Auxin transport mutants

Cytokinin modulates auxin transport

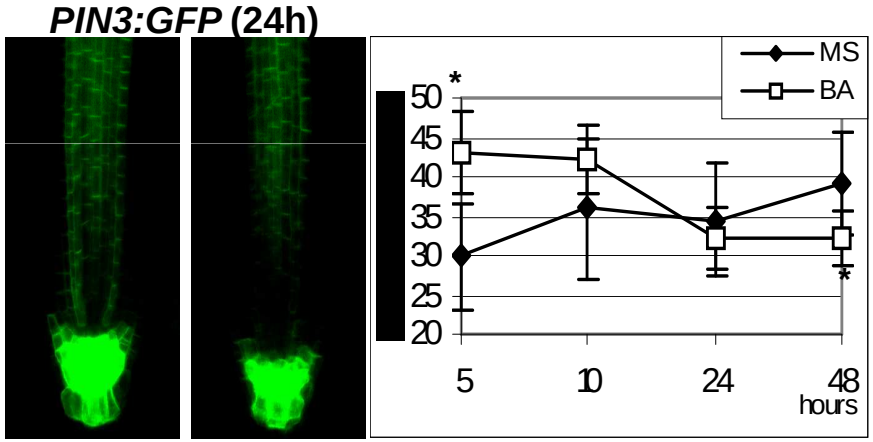
PIN1:GFP **(24 h)**



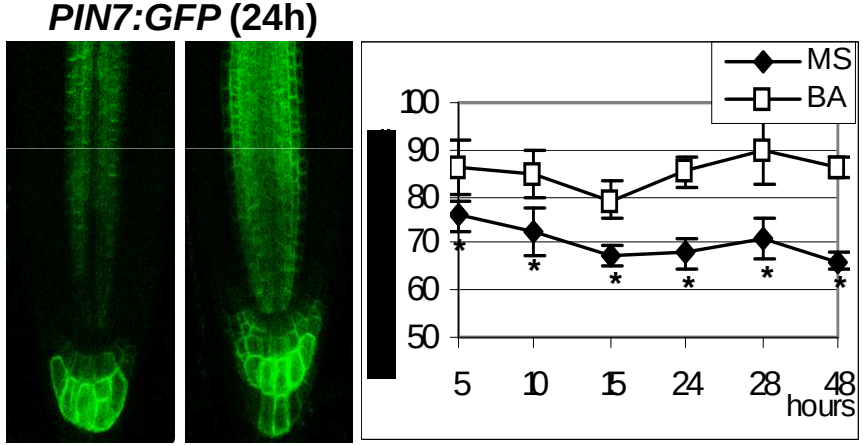
PIN2:GFP **(48 h)**



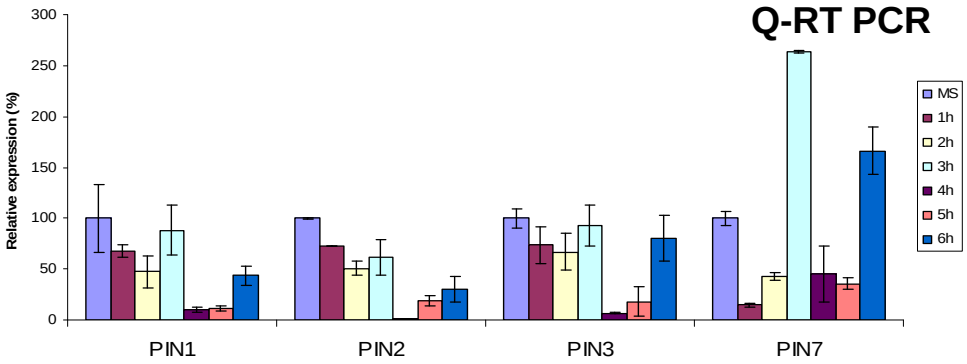
PIN3:GFP **(24h)**



PIN7:GFP **(24h)**



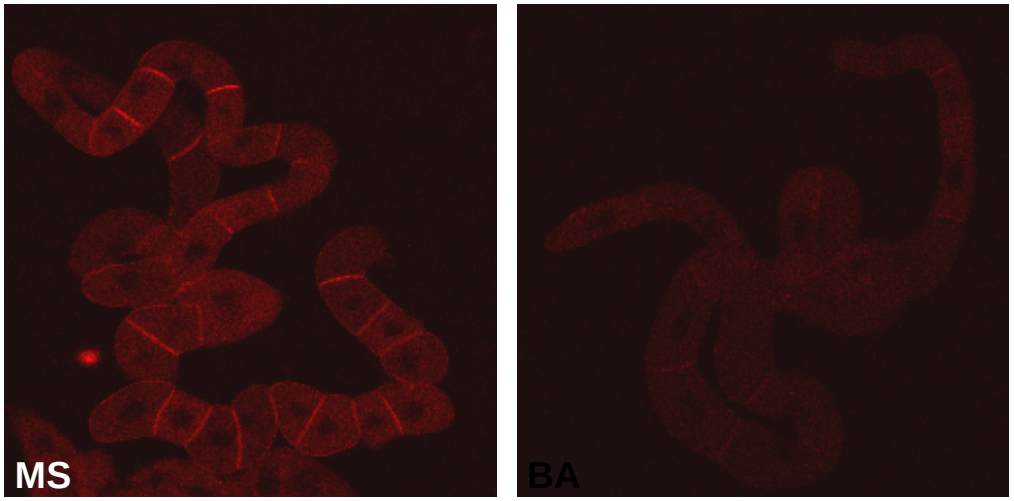
Q-RT PCR

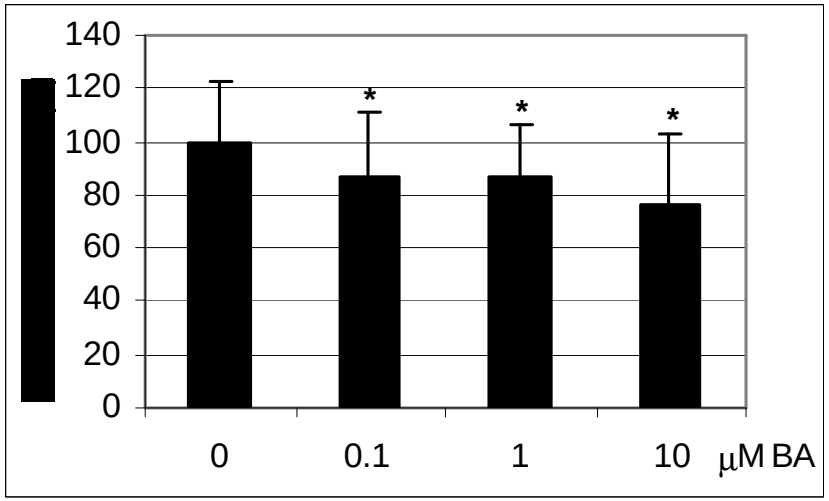


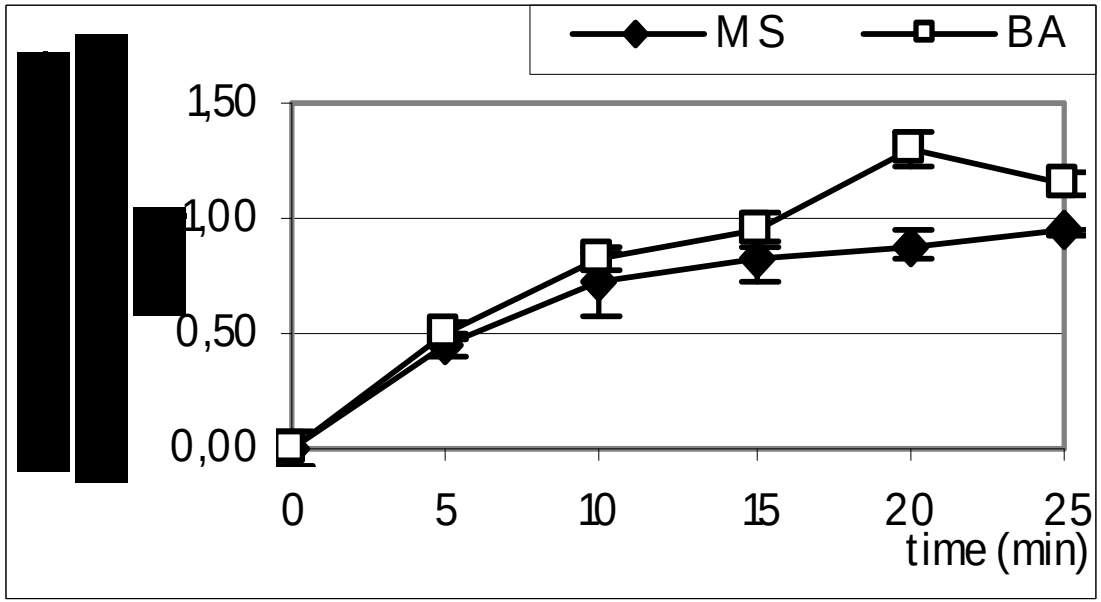
Ruzicka et al., 2009

Cytokinin reduces auxin efflux in tobacco BY2 cells

PIN1:RFP

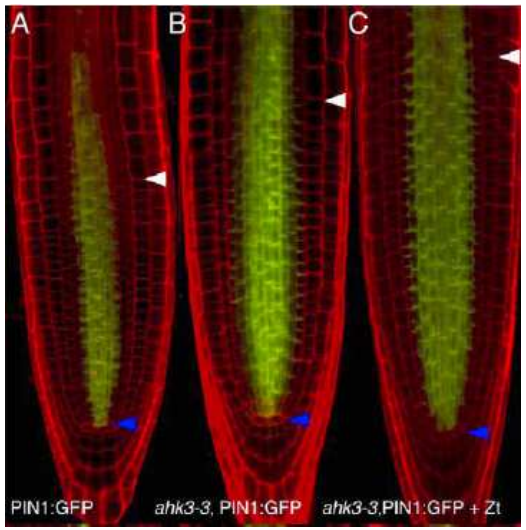




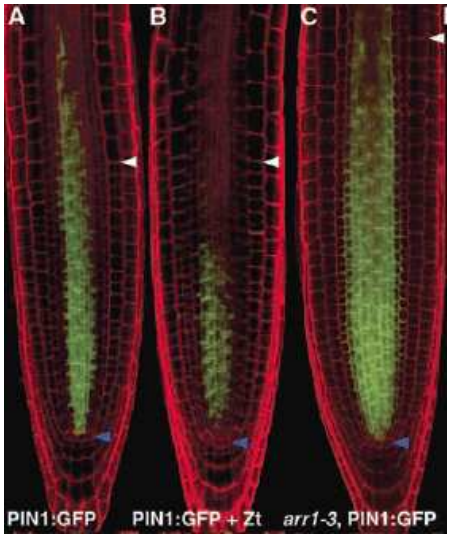


Ruzicka et al., 2009

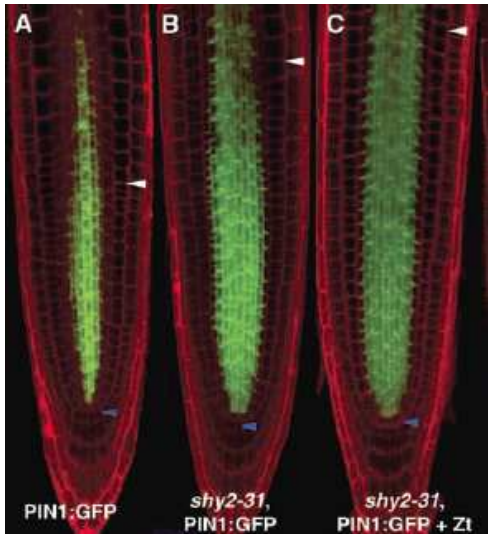
CK – auxin signalling pathways interaction to modulate auxin transport



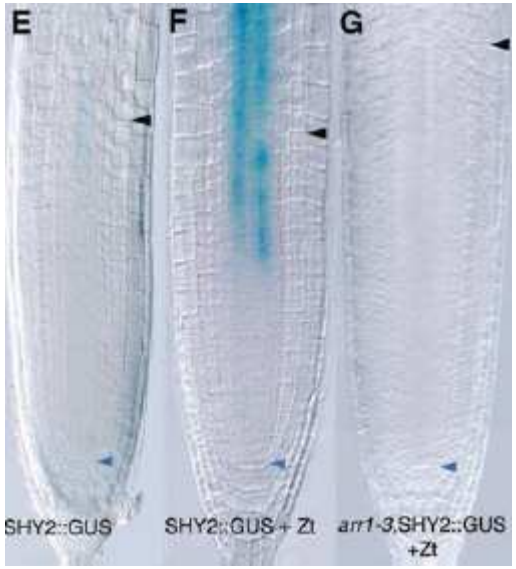
CK receptor



CK response regulator



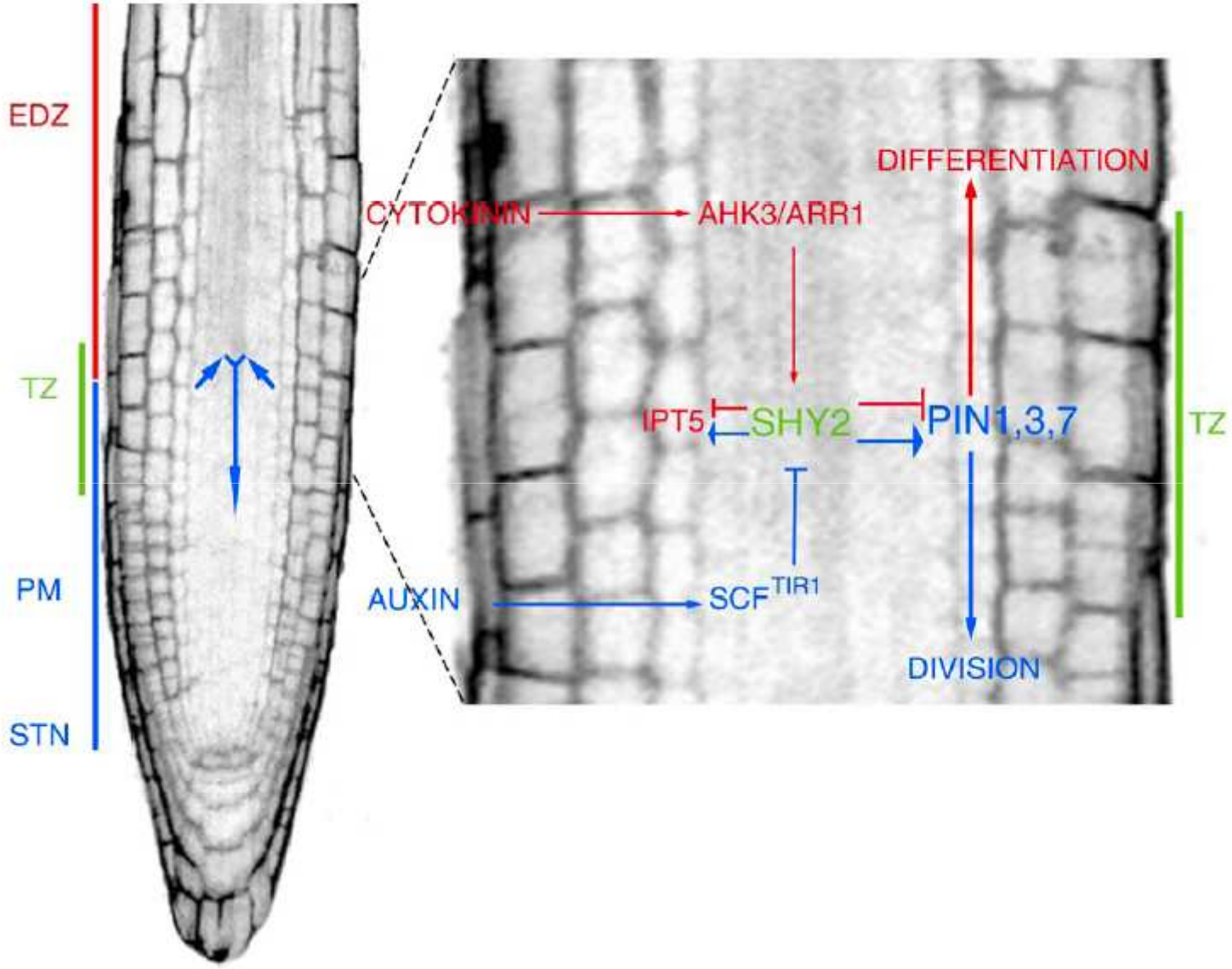
AUX/IAA negative regulator



Expression of IAA3/SHY2 is CK regulated

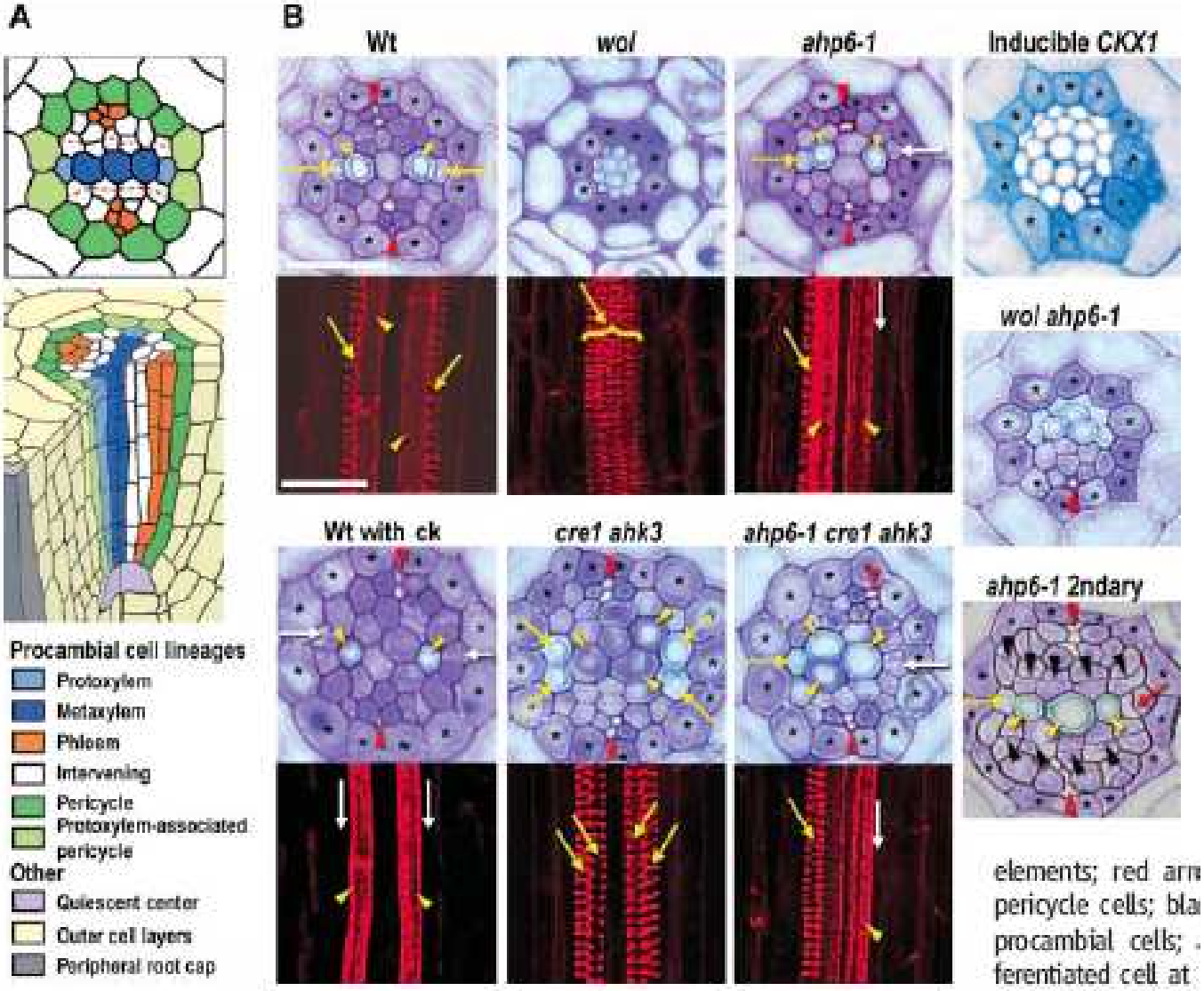
Dello Ioio et al., 2008

Model of cytokinin and auxin interaction in root meristem



Dello Ioio et al., 2008

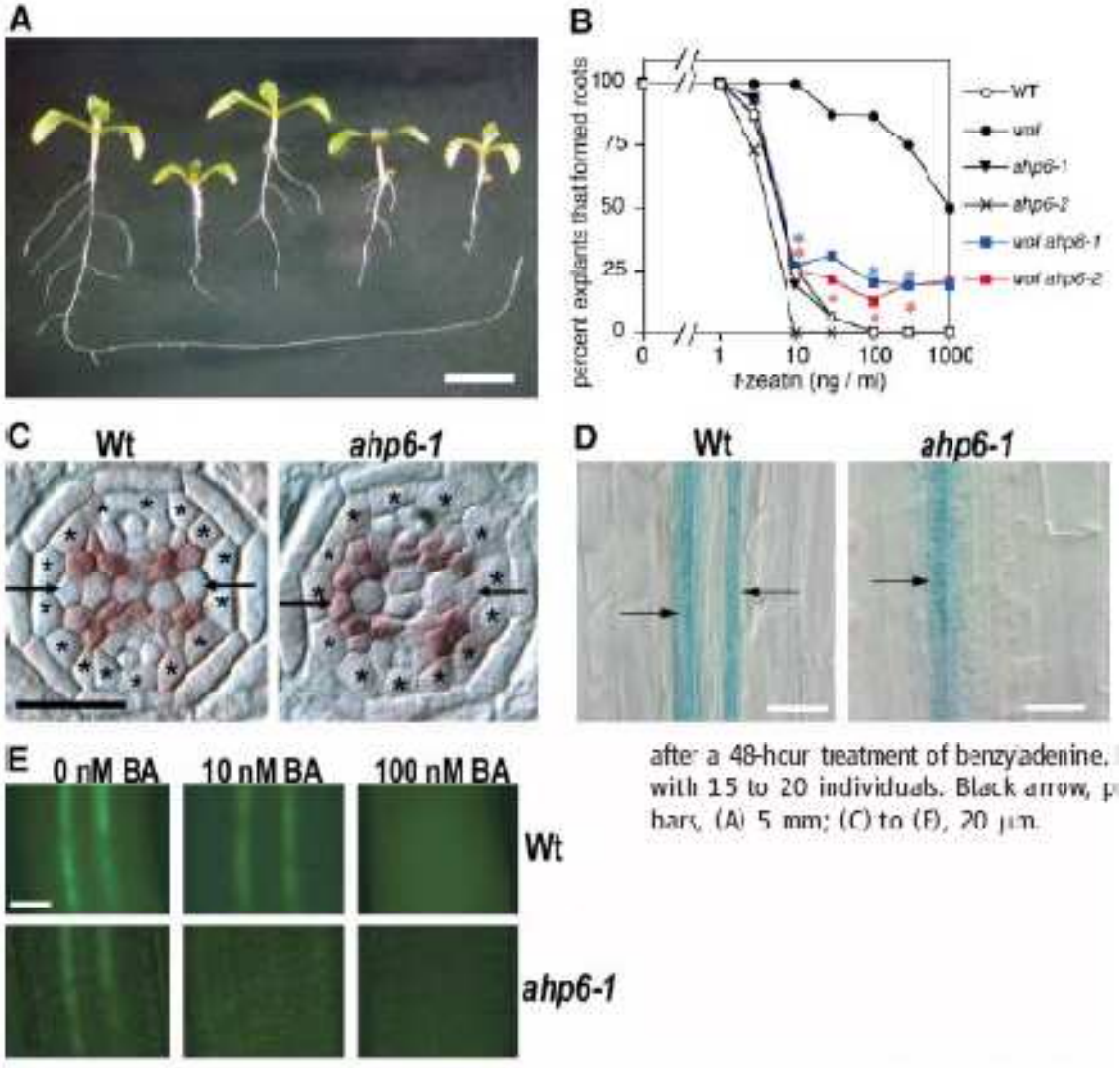
Cytokinin signalling regulates vascular development



elements; red arr
pericycle cells; bla
procambial cells; .
ferentiated cell at

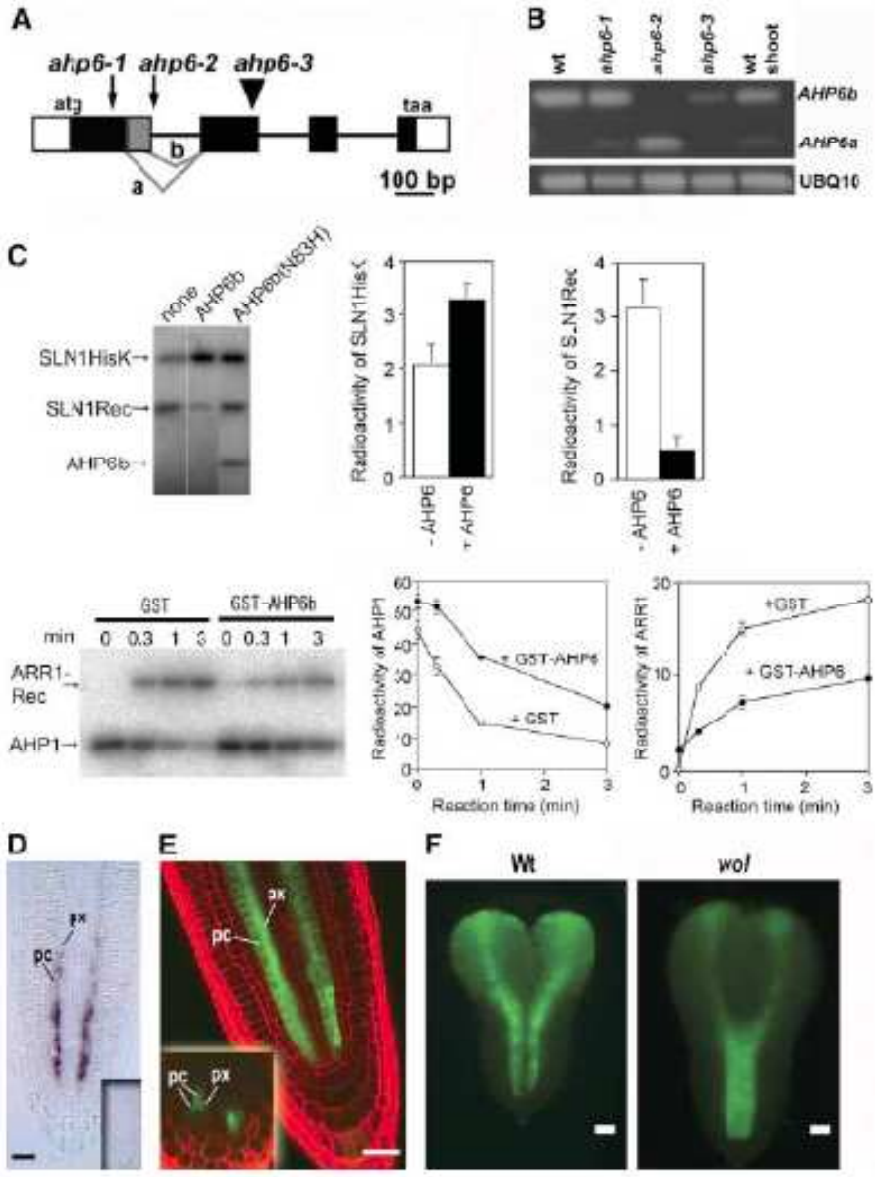
Mahonen et al., 2006

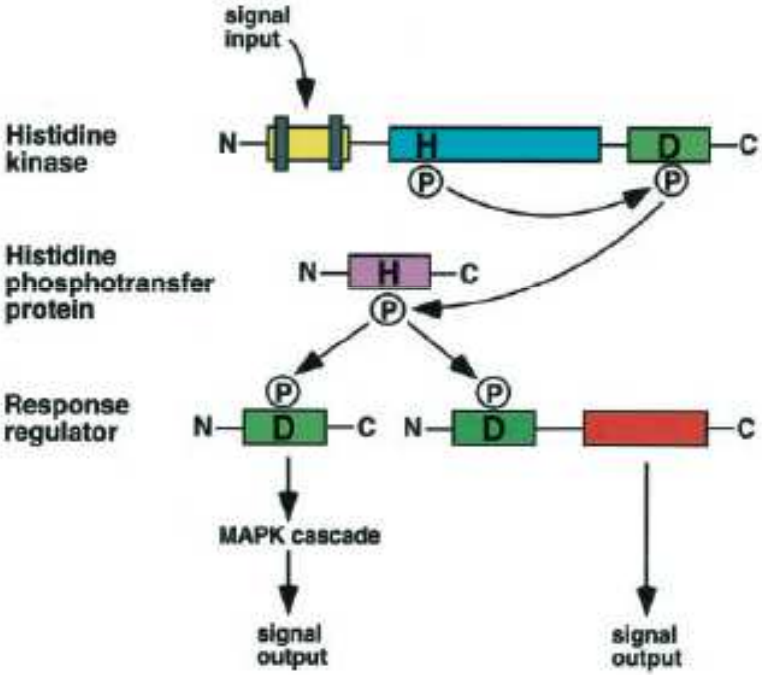
ahp6 suppress *wol* insensitivity to cytokinin



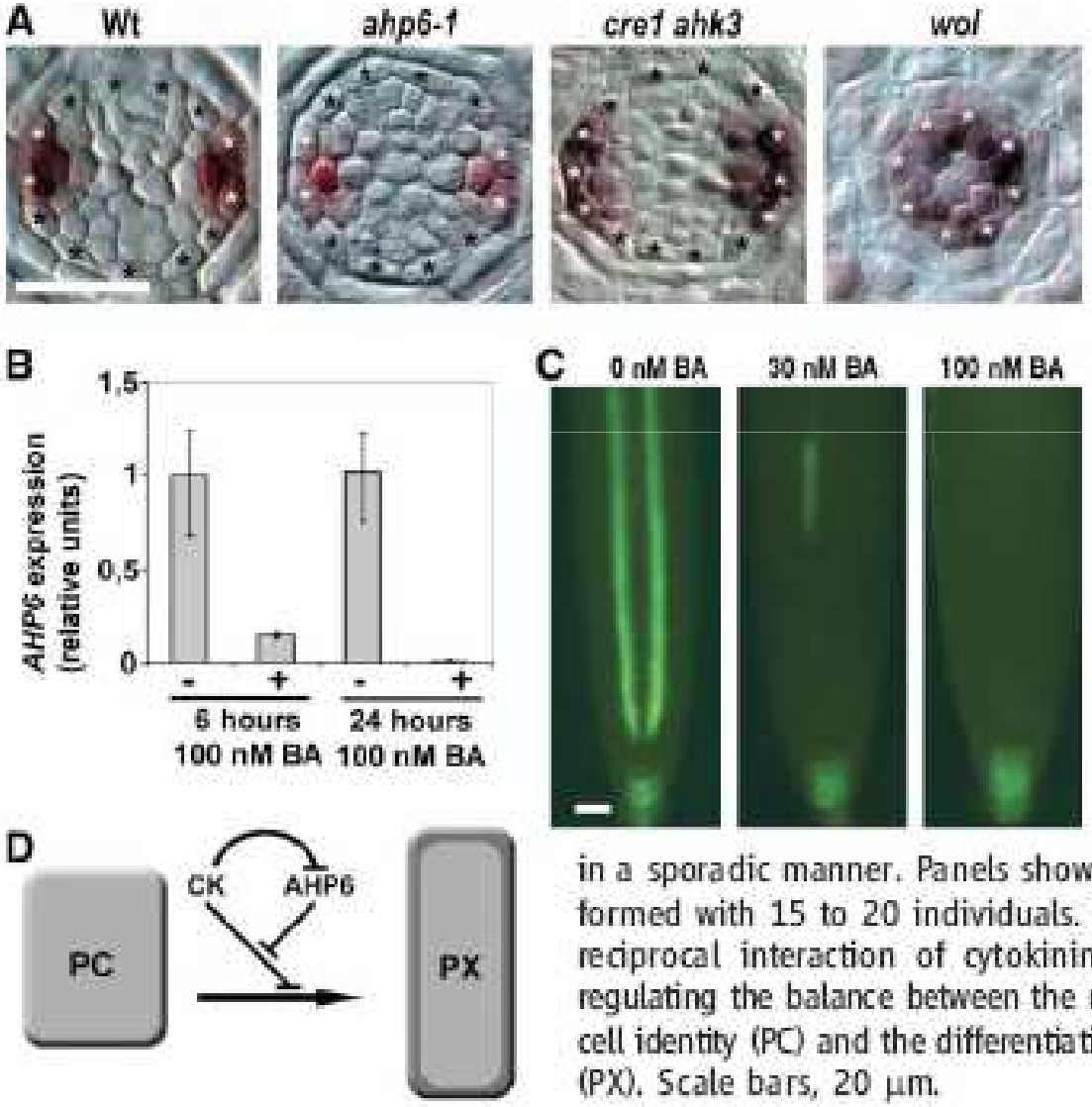
after a 48-hour treatment of benzyadenine.
with 15 to 20 individuals. Black arrow, p
bars, (A) 5 mm; (C) to (F), 20 μm.

AHP6 codes for histidine phosphotransfer protein 6





AHP6 is negative regulator of cytokinin signalling



in a sporadic manner. Panels shown
formed with 15 to 20 individuals. (
reciprocal interaction of cytokinin
regulating the balance between the m
cell identity (PC) and the differentiatio
(PX). Scale bars, 20 µm.