



IIT ROORKEE



NPTEL ONLINE
CERTIFICATION COURSE

Molecular Genetics of Plant Development-I

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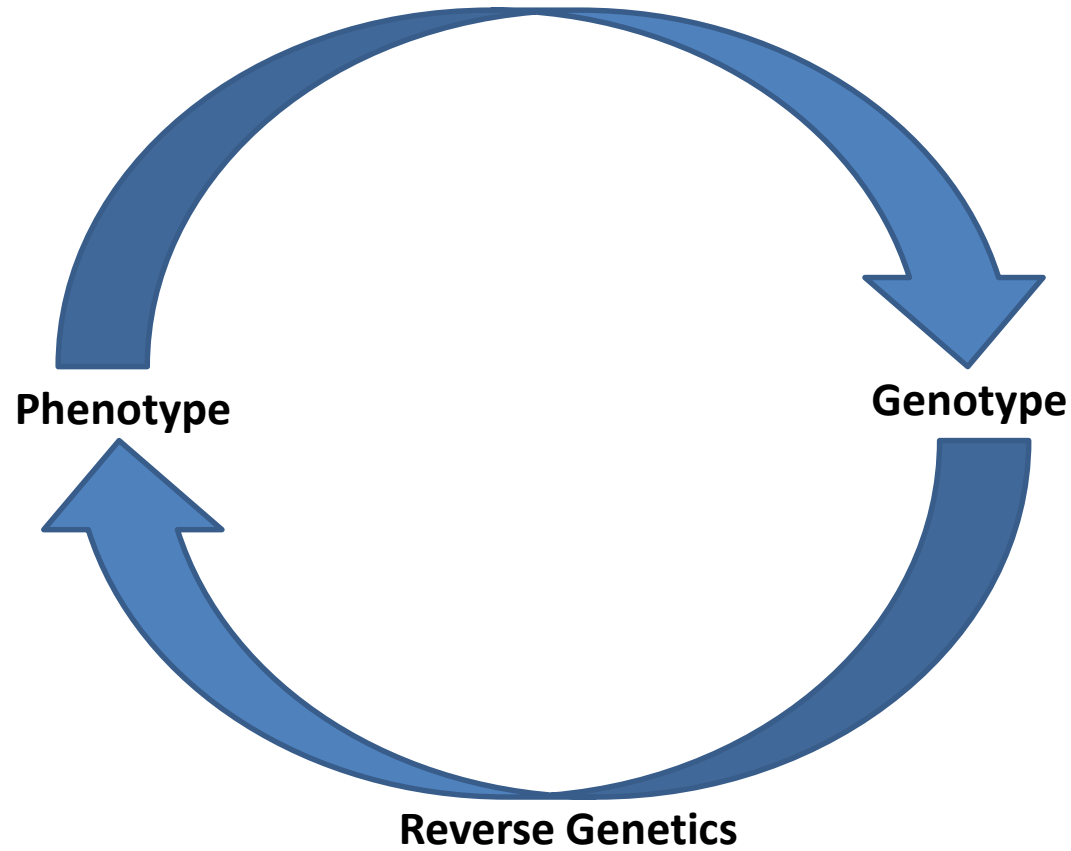
Indian Institute of Technology, Roorkee



Approaches to the study of plant development

- **Molecular Genetics:** Studying how genes and genome control various developmental processes in the plants
- **Forward Genetics:** An approach to determine genetic basis of a phenotype (Phenotype to Genotype)
- **Reverse Genetics:** An approach to study function of a gene (Genotype to Phenotype)

Forward Genetics



Approaches to the study of plant development

- **Forward Genetics:** An approach to determine genetic basis of a phenotype (Phenotype to Genotype)
 - *Identifying desired developmental phenotype (mutant)*
 - *Analyzing inheritance pattern of the phenotype (dominant/recessive)*
 - *Mapping genetic locus associated with the phenotype (gene mapping)*
 - *Validation of the genotype-phenotype association (complementation)*



Approaches to the study of plant development

➤ Types of Developmental mutants:

- *Meristem maintenance mutants*
- *Organ identity and patterning mutants*
- *Organ shape and size mutants*
- *Developmental phase transition mutants*



➤ Identifying desired developmental phenotype: Natural variation

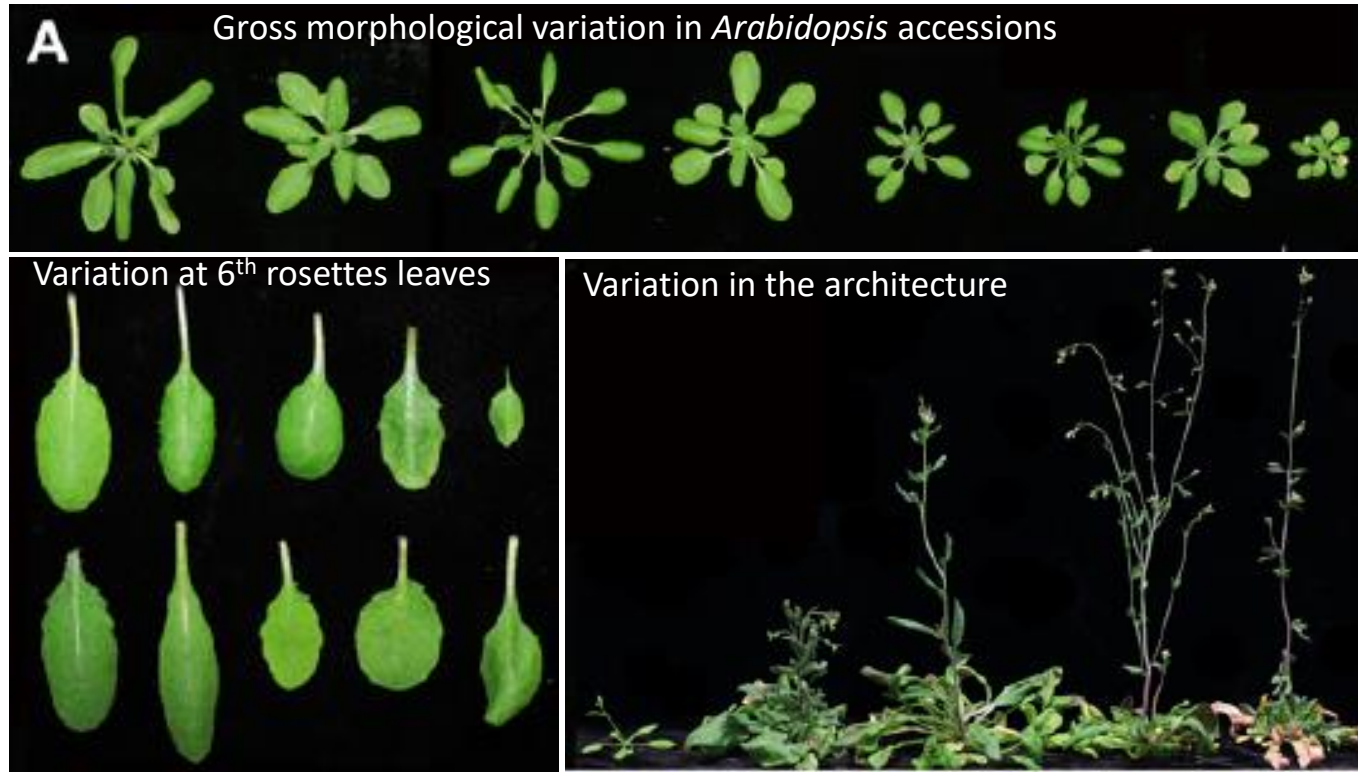
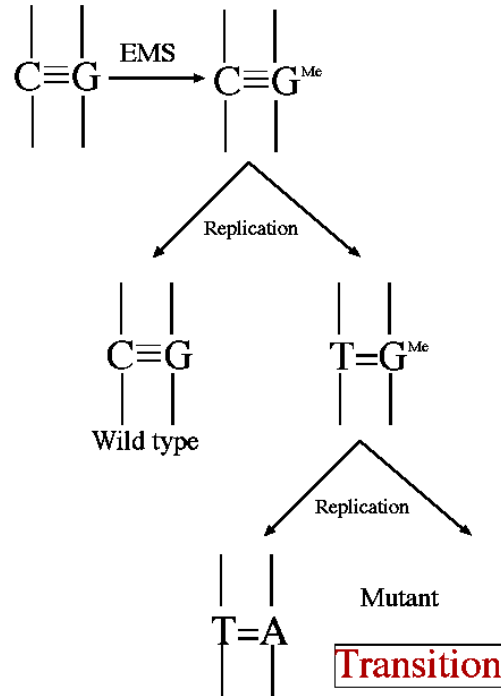


Image source:
Weigel D. (2012)
Plant Physiol., 158,
2-22.

➤ *Identifying desired developmental phenotypes: Mutagenesis*

- Chemical mutagens
 - **Ethyl methanesulfonate (EMS)**: G/C to A/T substitution
- Physical Mutagens
 - Electromagnetic radiation: **γ rays**, X rays, UV light
- Biological Mutagens
 - *Agrobacterium*-mediated insertion: ***T-DNA, Transposon***

➤ Identifying desired developmental phenotypes: EMS-Mutagenesis



➤ Identifying desired developmental phenotypes: EMS-Mutagenesis

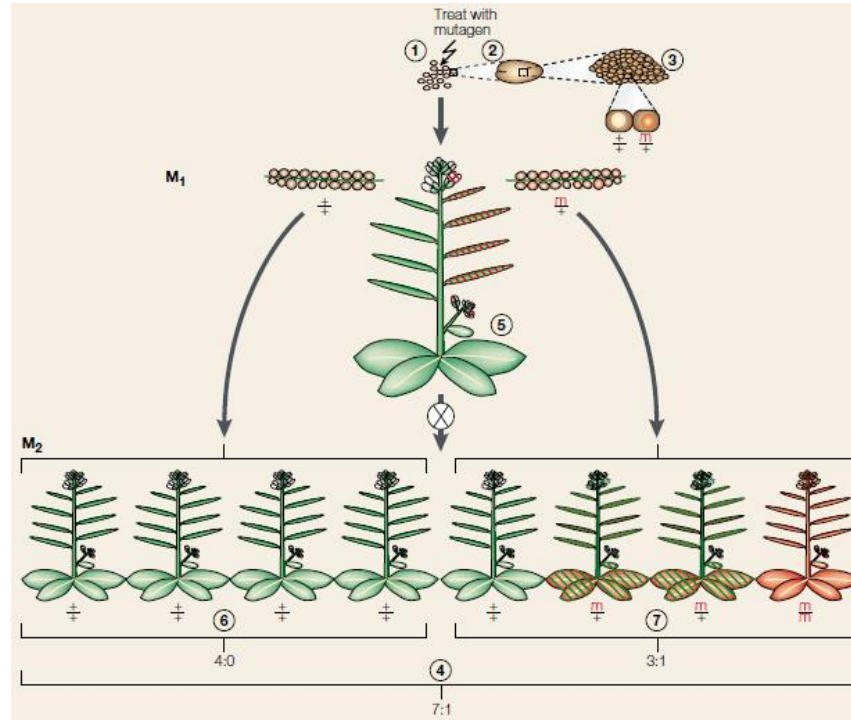


Image source:
Page & Grossniklaus(2002)
Nat Rev Genet., 3, 124-26.

➤ Identifying desired developmental phenotypes: EMS-Mutagenesis

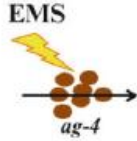


Image source:
Vaten et al. (2011)
Dev. Cell, 21, 1144-55.

➤ Identifying desired developmental phenotypes: EMS-Mutagenesis



ag-4: Indeterminate floral meristem; no carpel, stamen present



ag-4 hua1 hua2: stamens → petals



hua1 hua2: No obvious phenotype



hua1 hua2 hen1: Loss of floral determinacy; stamens → petals



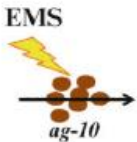
hua1 hua2 ago10: Loss of floral determinacy



hua1 hua2 ag-10: Loss of floral determinacy; stamens → petals



ag-10: Similar to wild type



ag-10 ago10: Loss of floral determinacy



ag-10 clf: Loss of floral determinacy

Identifying enhancers and suppressors



Image source:
Dinh et al. (2014)
Methods Mol. Biol.,
1110, 127-56.

Image source:
Wang et al. (2009)
Plant Cell, 21, 3781-91.

Approaches to the study of plant development

➤ *Identifying desired developmental phenotypes: Gamma Mutagenesis*

- Gamma radiated mutant screen in faba bean

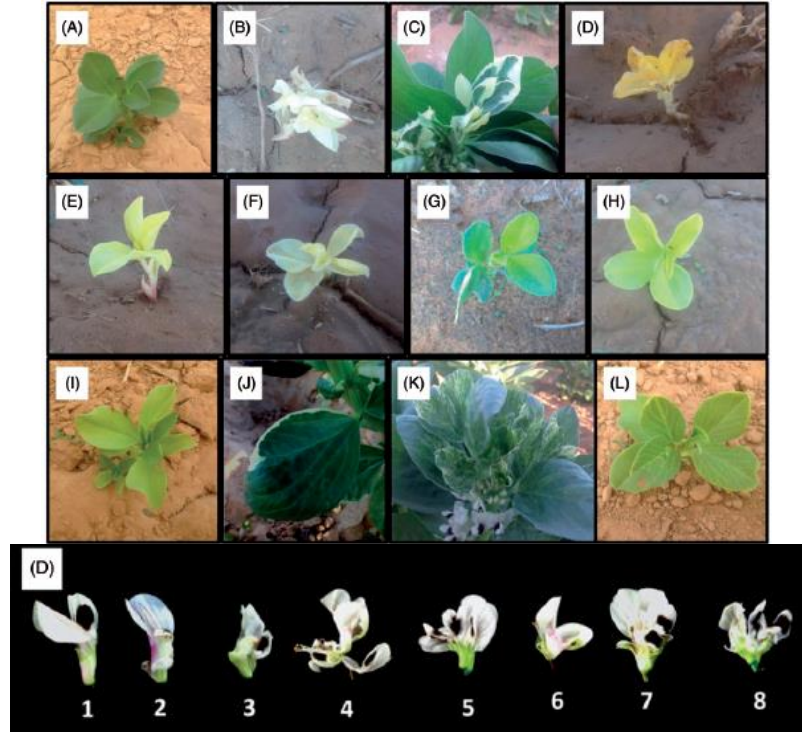


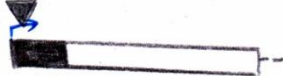
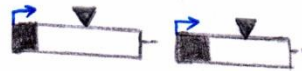
Image source:
Nurmansyah et al. (2018)
Int. J. Rad. Biol., 94, 174-85.

➤ *Identifying desired developmental phenotypes:* Biological Mutagenesis

- **T-DNA mutagenesis:** *Agrobacterium*-mediated T-DNA insertion
- **Transposon mutagenesis:** *Ac/Ds* or *Tos17* transposon system
- **Activation Tagging mutagenesis:** Random insertion of multiple enhancers

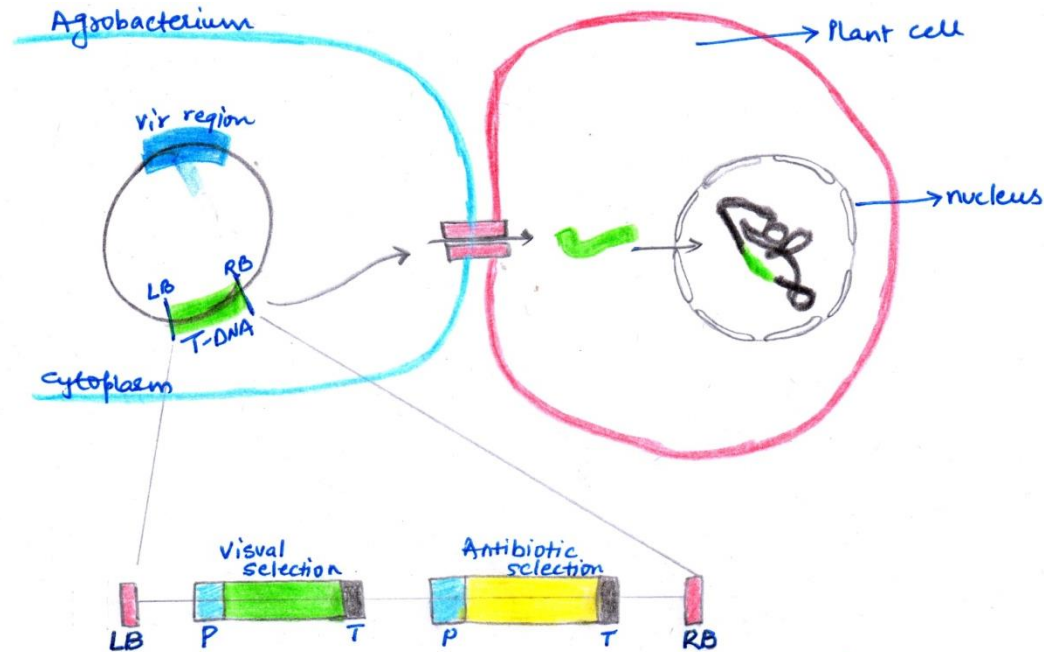


➤ Identifying desired developmental phenotypes: Insertional-Mutagenesis

<u>Name</u>	<u>Location</u>	<u>Result</u>
 knock out	Coding region/ promoter	null
 knock down	promoter/3' UTR	reduced expression
 knock on	promoter	increased expression
 Knock - Knock	Coding region/ promoter	multiple Koz in one plant

➤ Identifying desired developmental phenotypes: Insertional-Mutagenesis

- T-DNA mutagenesis: *Agrobacterium*-mediated T-DNA insertion



➤ Identifying desired developmental phenotypes: T-DNA Mutagenesis

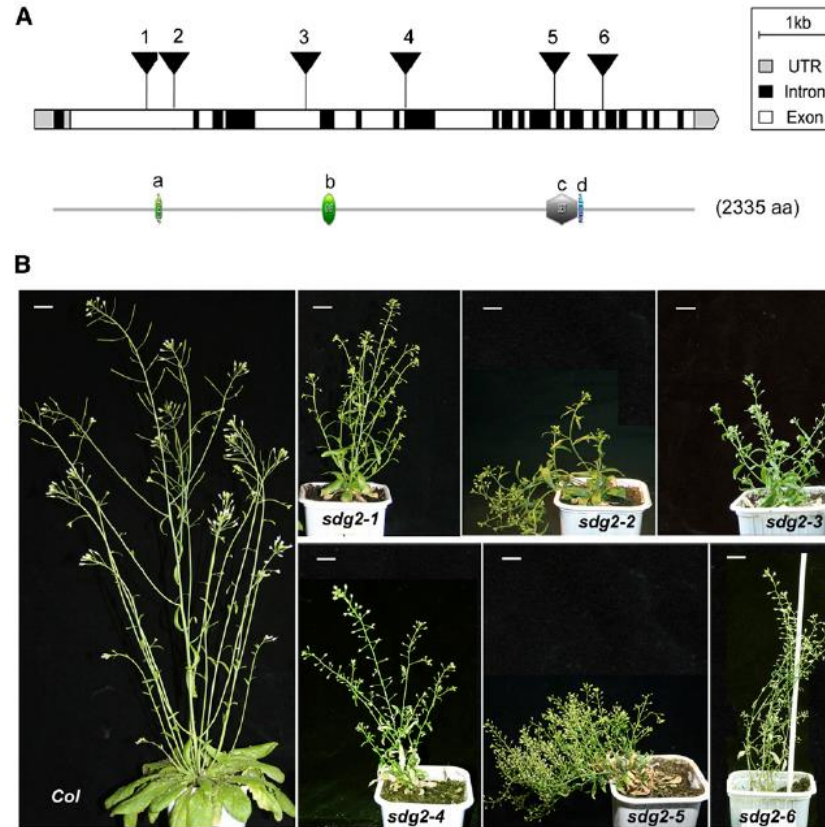
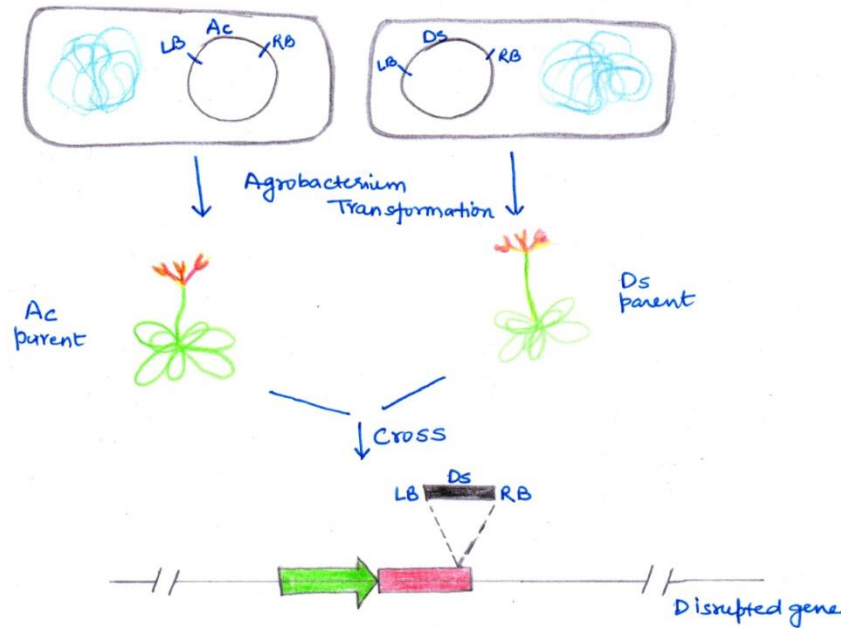


Image source:
Berr et al. (2010)
Plant Cell, 22,
3232-48.

➤ Identifying desired developmental phenotypes: Insertional-Mutagenesis

- Transposon mutagenesis: *Ac/Ds* transposon system



➤ *Identifying desired developmental phenotypes: Insertional-Mutagenesis*

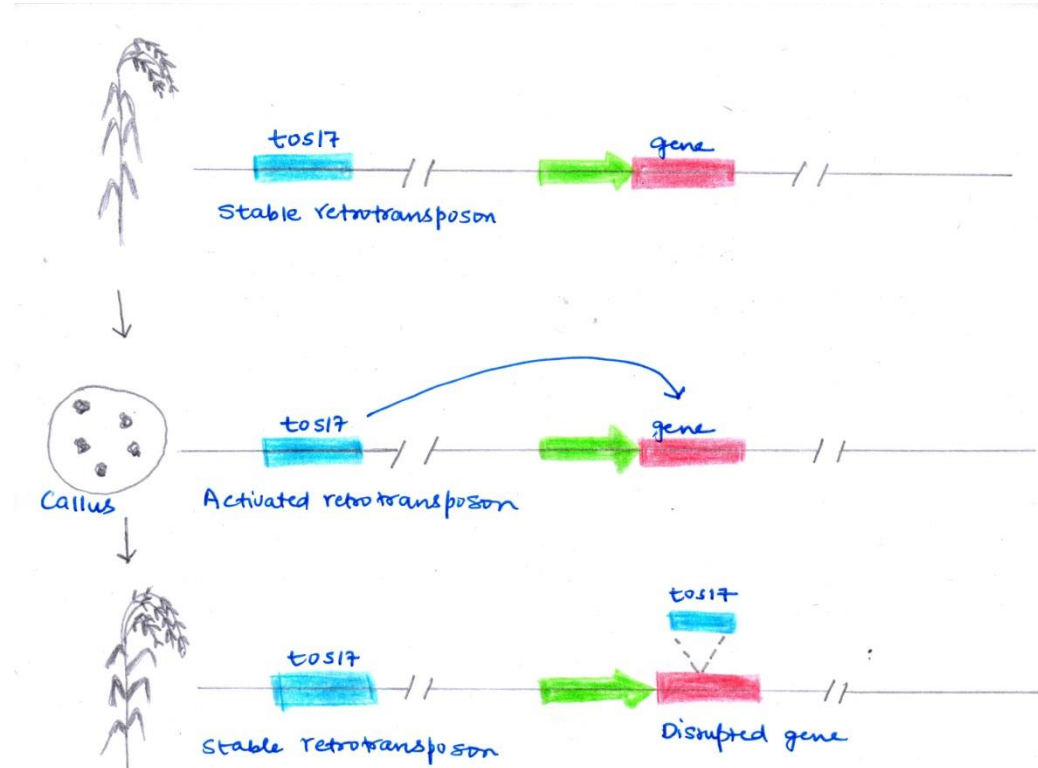
- Transposon mutagenesis: *Ac/Ds* transposon system



Image source:
Strable et al. (2017)
Plant Cell, 29,
1622-41.

➤ Identifying desired developmental phenotypes: Insertional-Mutagenesis

- Transposon mutagenesis:
(Tos17 retrotransposon)



➤ Identifying desired developmental phenotypes: Insertional-Mutagenesis

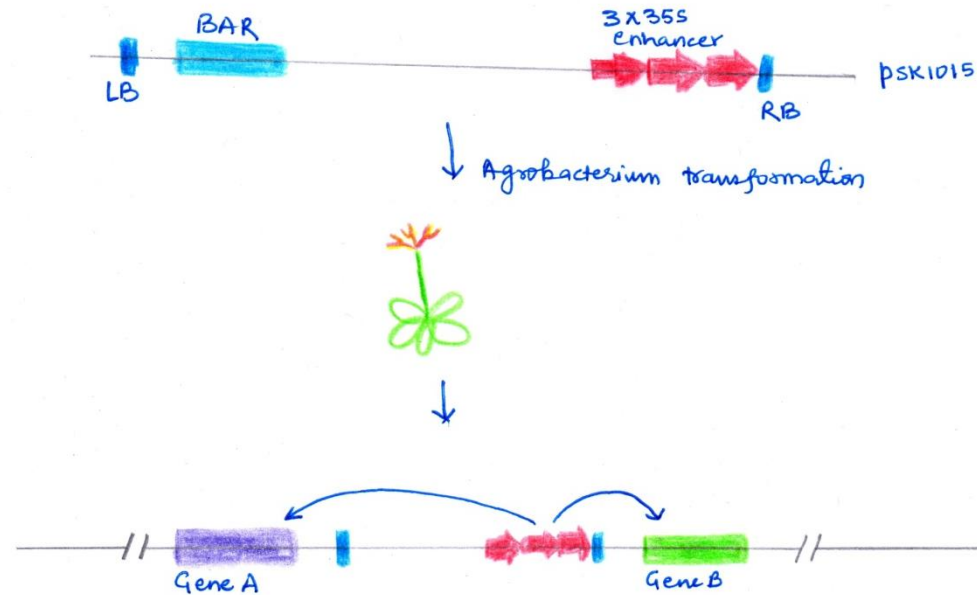
- Transposon mutagenesis: *Tos17* (retrotransposon)



Image source:
Miyao et al. (2007)
Plant Mol Biol, 63,
625-35.

➤ *Identifying desired developmental phenotypes: Biological Mutagenesis*

- **Activation Tagging mutagenesis:** Random insertion of multiple enhancers



➤ Identifying desired developmental phenotypes: Biological Mutagenesis

- **Activation Tagging mutagenesis:** Random insertion of multiple enhancers

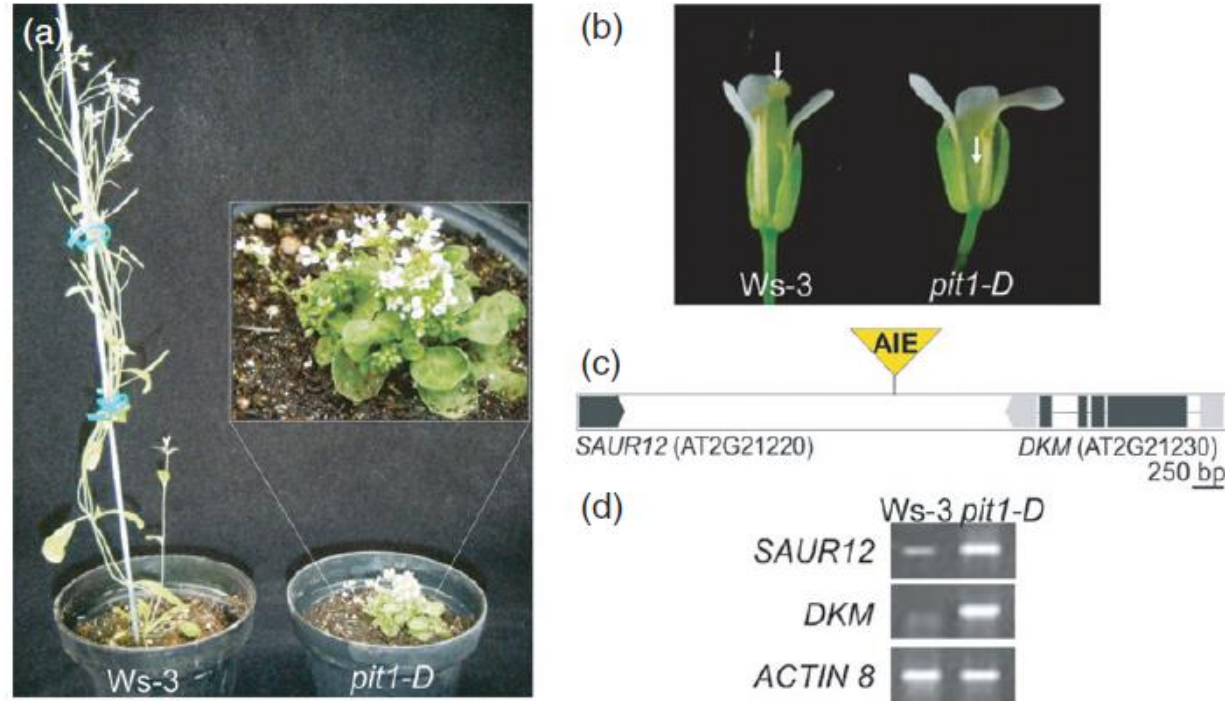


Image source:
Lozano-Sotomayor et al.
(2016) Plant Journal, 88,
437-51.