Pengembangan Teknologi Pemuliaan Tanaman untuk Masa Depan*

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CV

Academic Qualification

- Oct '07 – March '08: Postdoc Fellow, Nat. Inst of Agrobiol Sci, Tsukuba, Japan
- Oct 2004 – Sept 2007: PhD course, Tokyo Univ. of Agric. & Tech, Japan
- Sept 2001 – Jan 2003: MSc in Crop Breeding, Wageningen University, NL
- Sept 1991 - August 1996: BSc in Plant Breeding, Universitas Padjadjaran, IDN

Field expertise:

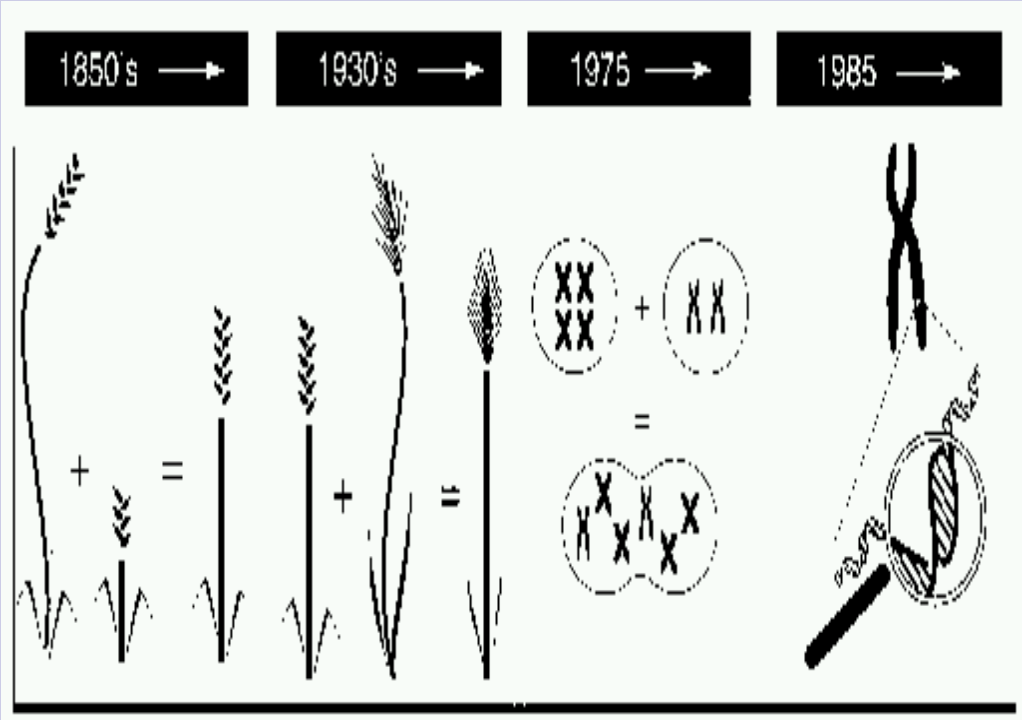
- *in vitro* culture, gene isolation and cloning, gene expression analysis, gene transfer/transgenic tech, biosafety assessment

Plant Breeding ?

- A science that applied to manipulate gene/DNA sequence or chromosome for improving existing cultivars and to develop new/elite cultivars with regard to human needs.
- Dynamic and sustainable efforts.
- Multidiscipline: genetics, agronomy, botany, physiology, phytopathology, entomology, molecular genetics, biochemistry, statistics, and bioinformatics.

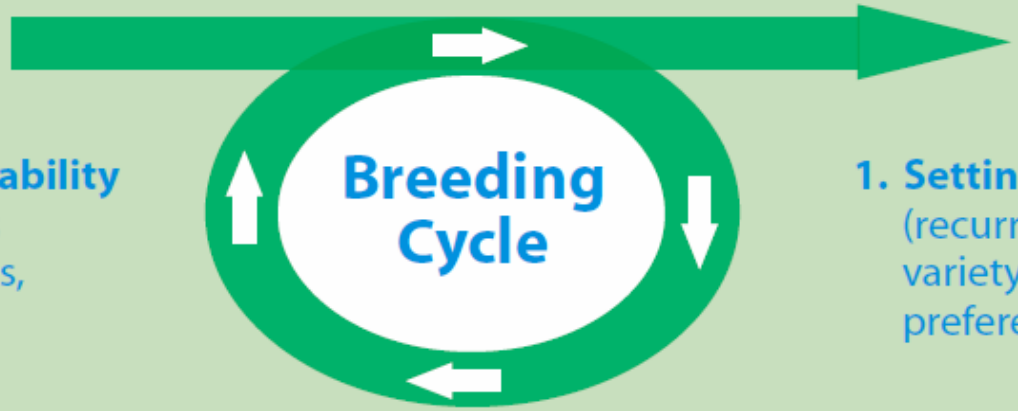
History

- 1850s Cross pollination between varieties of the same species, e.g. wheat
- 1930s Hybridisation between different species and genus e.g. wheat + rye = triticale
- 1975 Cell fusion to overcome species barriers by combining individual plant cells
- 1985 DNA technology - modification of crop characteristics at the level of individual genes.



Plant Breeding cycle

5. Variety adoption and diffusion
strengthen farmer systems, information exchange, overcome bottlenecks



4. Testing for adaptability
testing of varieties and advanced lines, farmer's managed testing, group evaluations

1. Setting breeding goals
(recurrent activity) variety type, farmer preferences, user needs

3. Narrowing down variability
farmer's selection, on-station selection, variety rehabilitation

2. Generating variability
new farmer varieties, modern varieties, segregating lines, farmer crosses, on-station

Plant breeding technology

- Conventional: hybridization (intra, inter, wide hybridization), selection, mutation/polyploidization.

- Modern tech: *in vitro* manipulation, molecular marker, recombinant DNA/ genomic tech(gene: isolation, cloning & expression), genetic transformation→ plant biotechnology.

In vitro technique

- Germplasm preservation;
- Micropropagation for virus elimination;
- Embryo rescue;
- Protoplast fusion & Cybridization (exchange mtDNA/cpDNA);
- Cellular selection for tolerance to salinity, pest & disease, herbicide, etc;
- Somaclonal variants;
- Production of haploid or double haploids via microspore/anther culture.

Factors Affecting *in vitro* Culture

a. Growth Media

– Minerals, Growth factors, Carbon source, Hormones

b. Environmental Factors

– Light, Temperature, Photoperiod, Sterility,

c. Explant Source

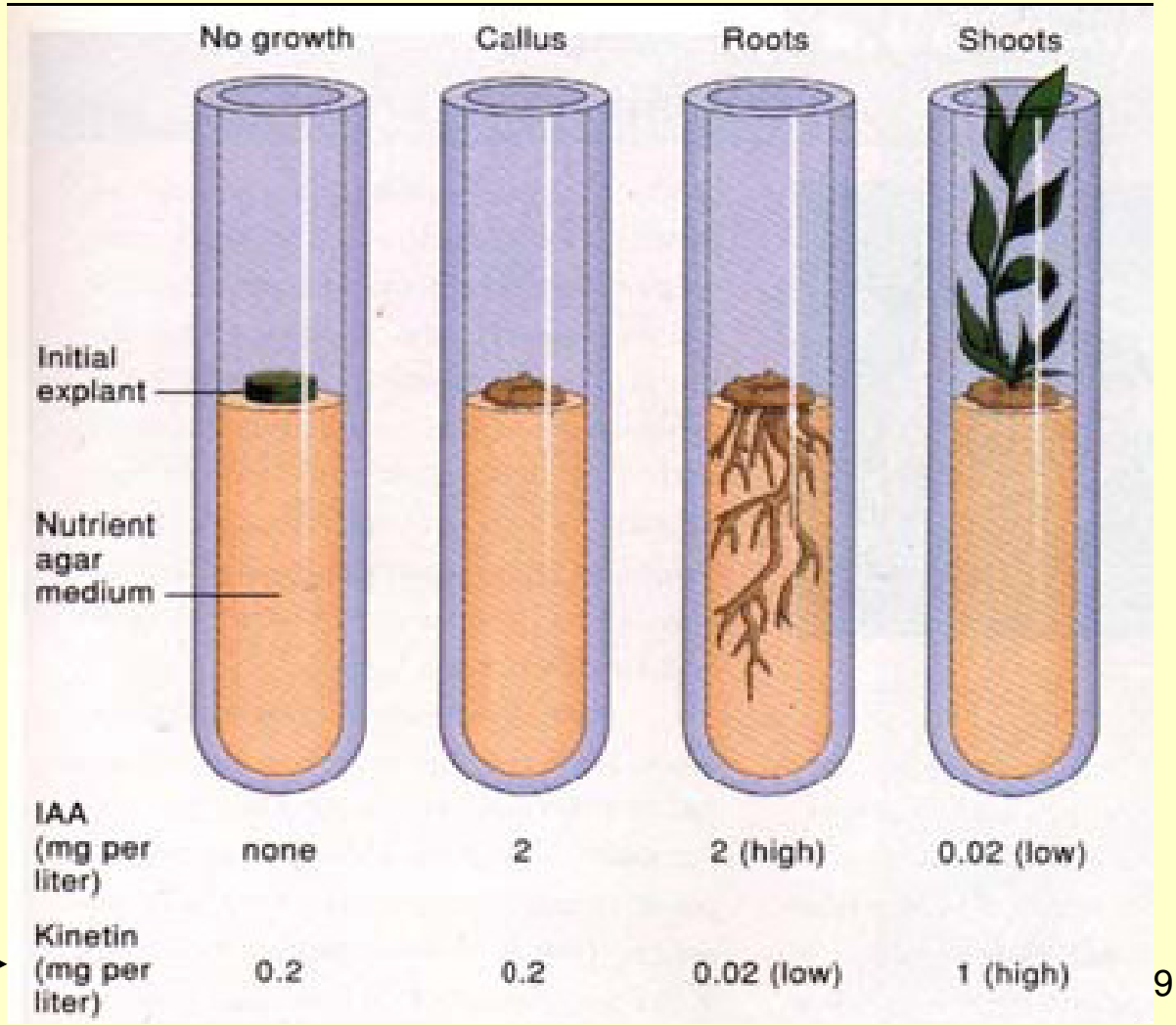
– Usually, the younger the better

d. Genetics

– Different species show differences in amenability to tissue culture

– In many cases, different genotypes within a species will have variable responses to tissue culture;

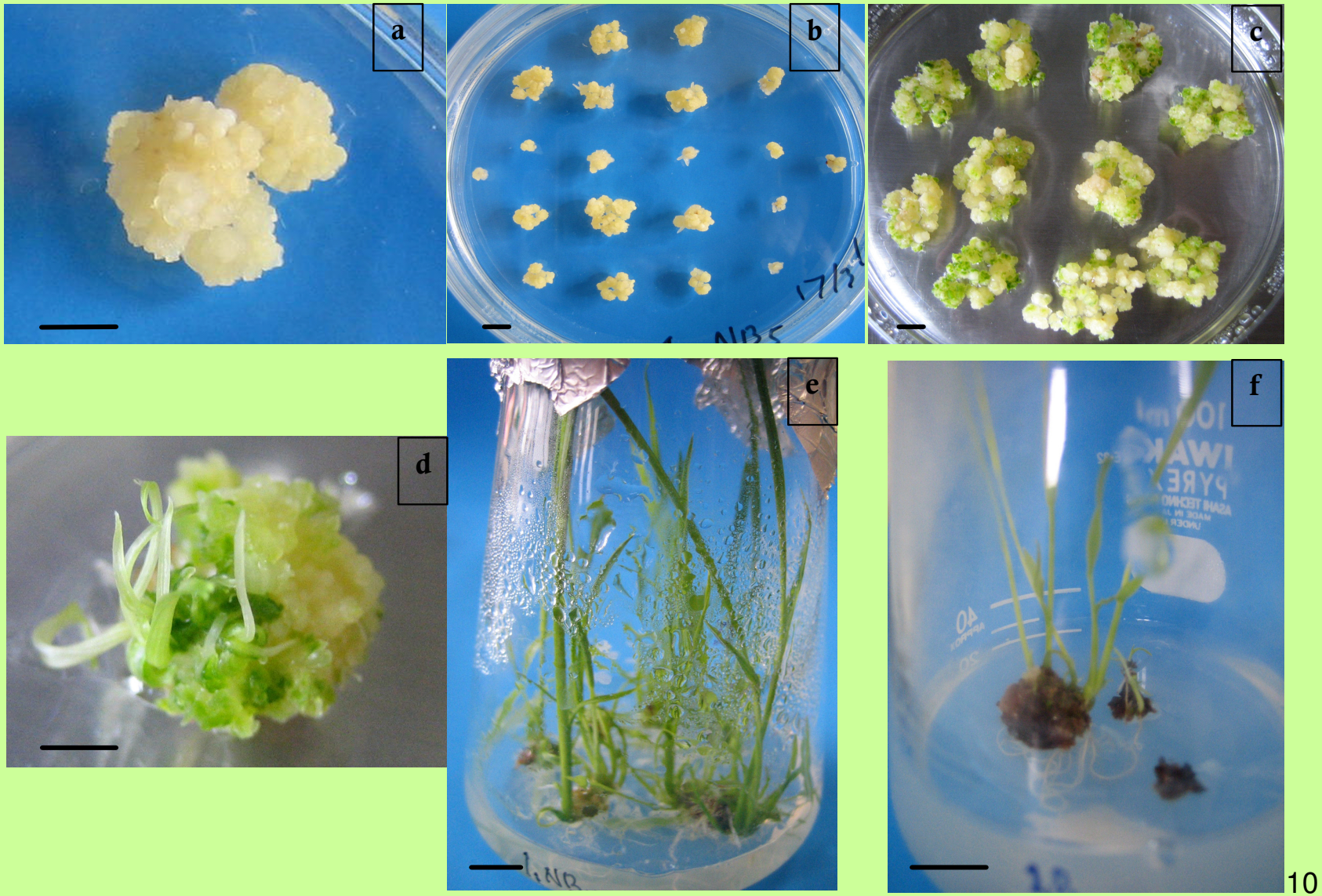
Hormonal control: organogenesis



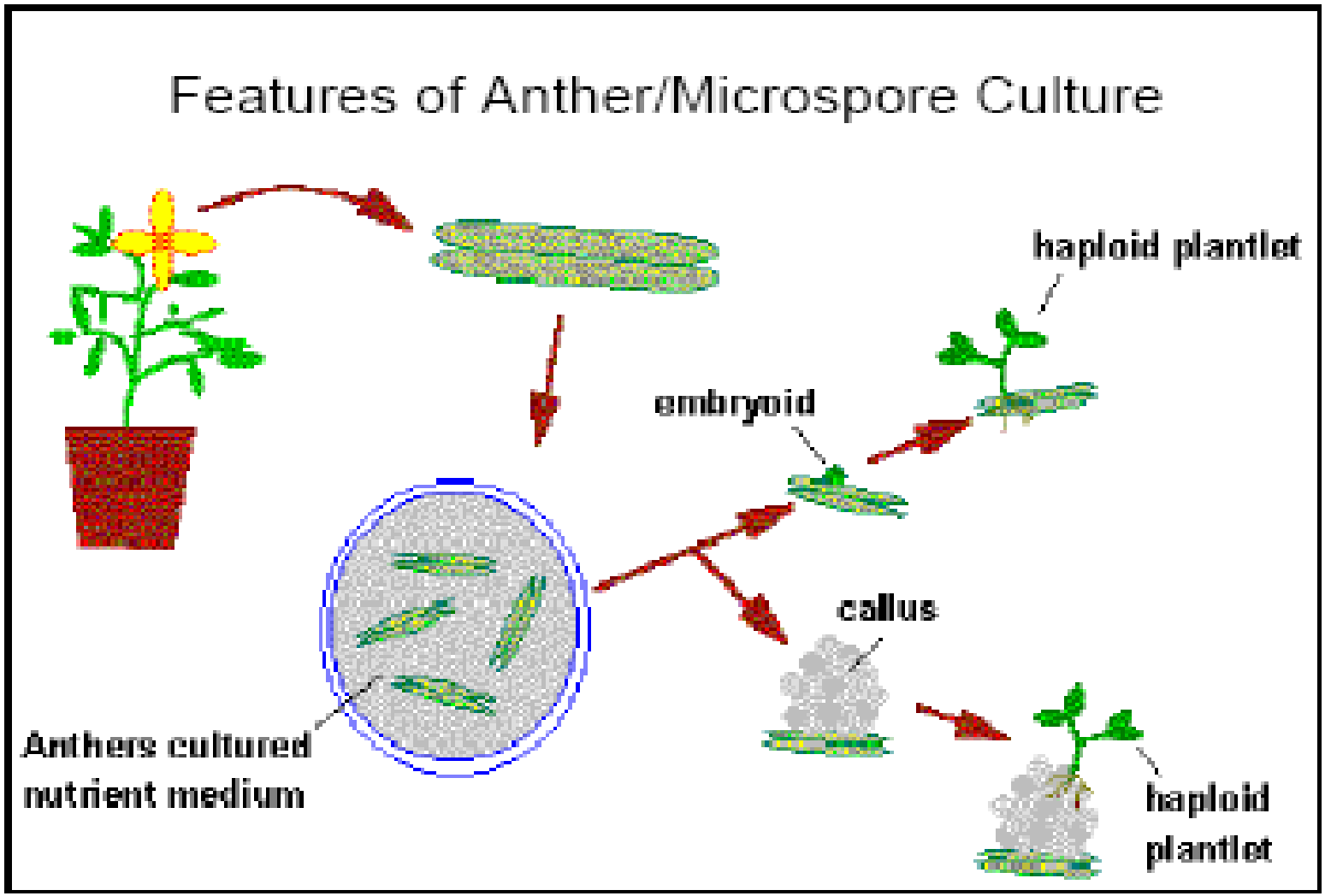
Auxin →

Cytokinin →

From callus induction to plant regeneration



Anther/microspore culture



Molecular marker

- Molecule that used to mark gene/ specific DNA sequence
- DNA-based marker:

  - RFLP (restriction fragment length polymorphism)

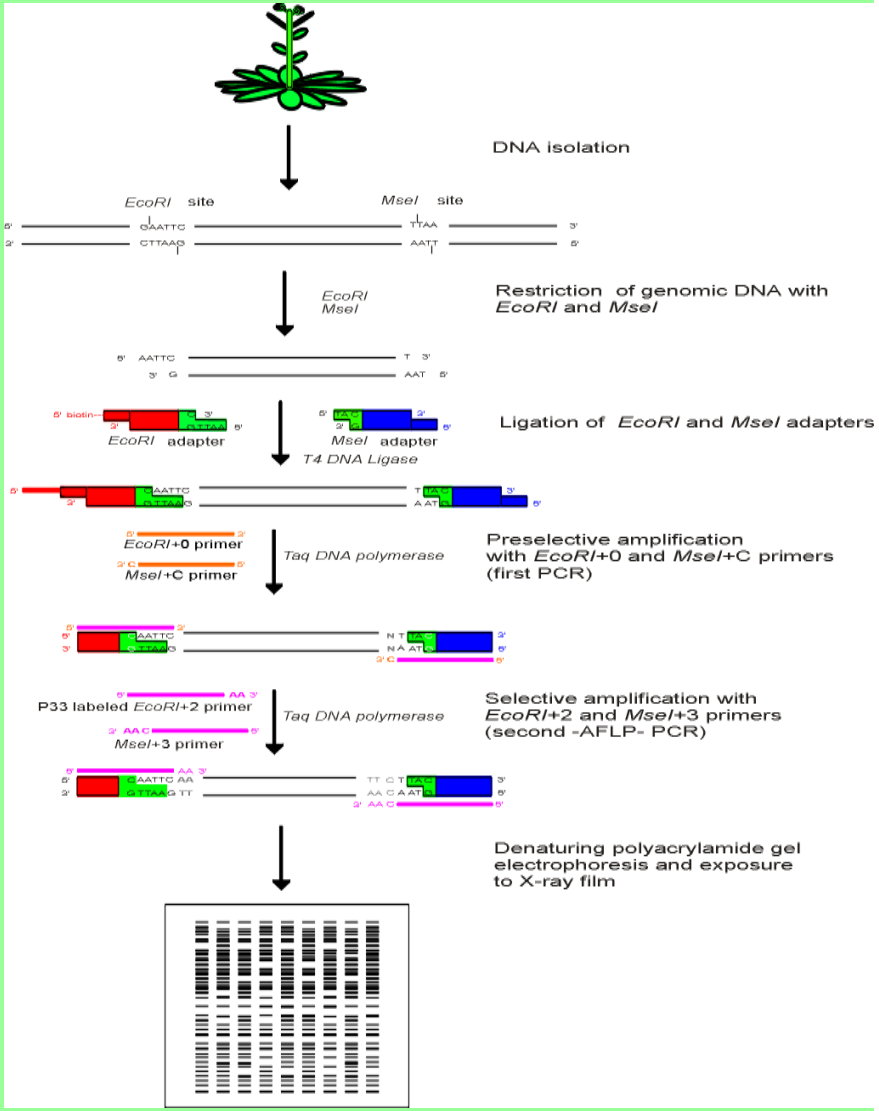
  - RAPD (random amplified fragment polymorphic DNA)

  - AFLP (amplified fragment length polymorphism)

  - VNTR (variable number of tandem repeat loci)

  - SSR (simple sequence repeats)/STR (short tandem repeat)

  - SNP (Single Nucleotide Polymorphism), etc



Future direction in molecular marker

- Further development of molecular marker that generates high polymorphism,

- Involvement of genome information from relatives and database worldwide for development of new marker.

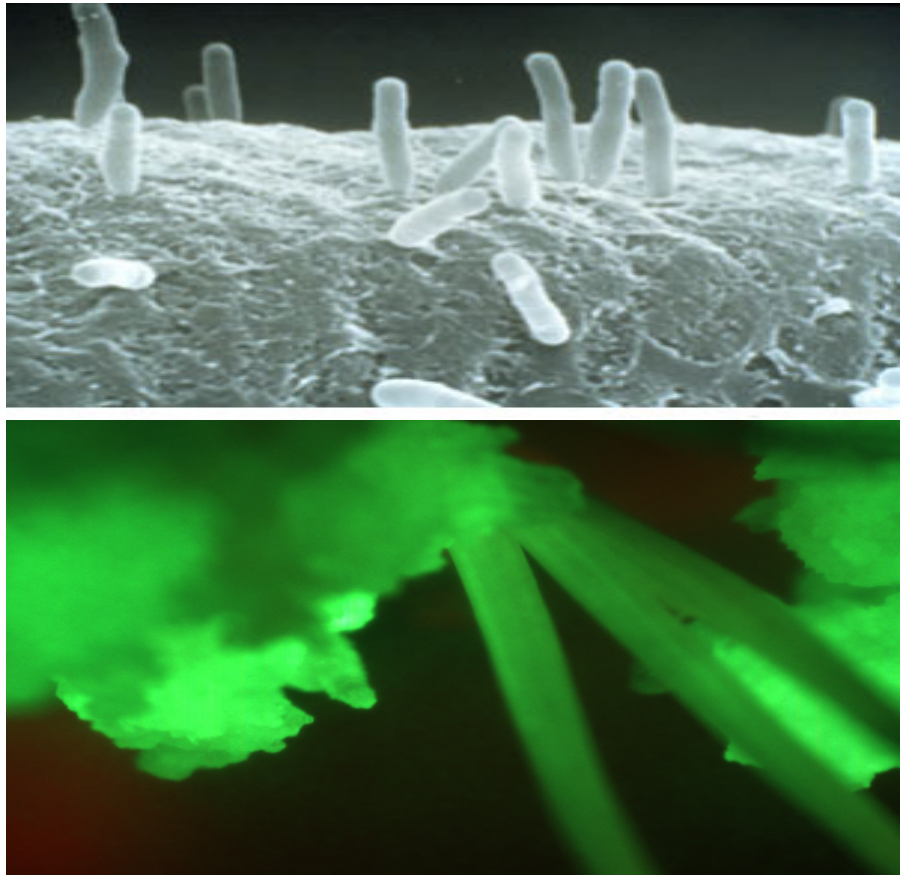
Genetic Transformation/ Gene Transfer

- **Herbicide resistant plants** (glyphosate, sulfonylureas, basta).
- **Virus resistant plants** (viral coat protein genes).
- **Insect resistant plants** (Bt genes; trypsin inhibitors).
- **Alteration of seed storage proteins and lipid biosynthesis.**
- **Micronutrient fortification of foods**
- **Antisense technology for the suppression of gene expression** (e.g., induction of cms trait by barnase/barstar genes).
- **Production of secondary metabolites through transformed roots via *Agrobacterium rhizogenes*.**
- **Studies on gene function via transient gene expression.**
- **Creation of mutants by transposable elements.**
- **Bioremediation by the use of transgenic plants for heavy metal tolerance.**
- **Studies on the process of morphogenesis.**

Mode of delivery

- **1.Transfer of DNA via vector**
- a. *Agrobacterium*-mediated
  - floral dip
  - wounded explants
  - cultured cells

  b. viral vector



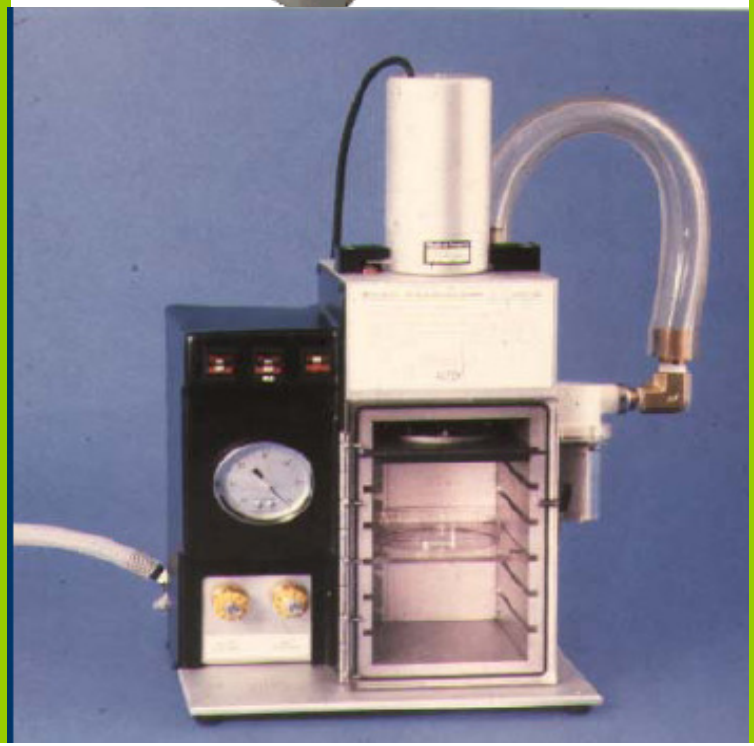
Mode of delivery

2. Direct transfer

- Protoplast PEG method
- Protoplast electroporation

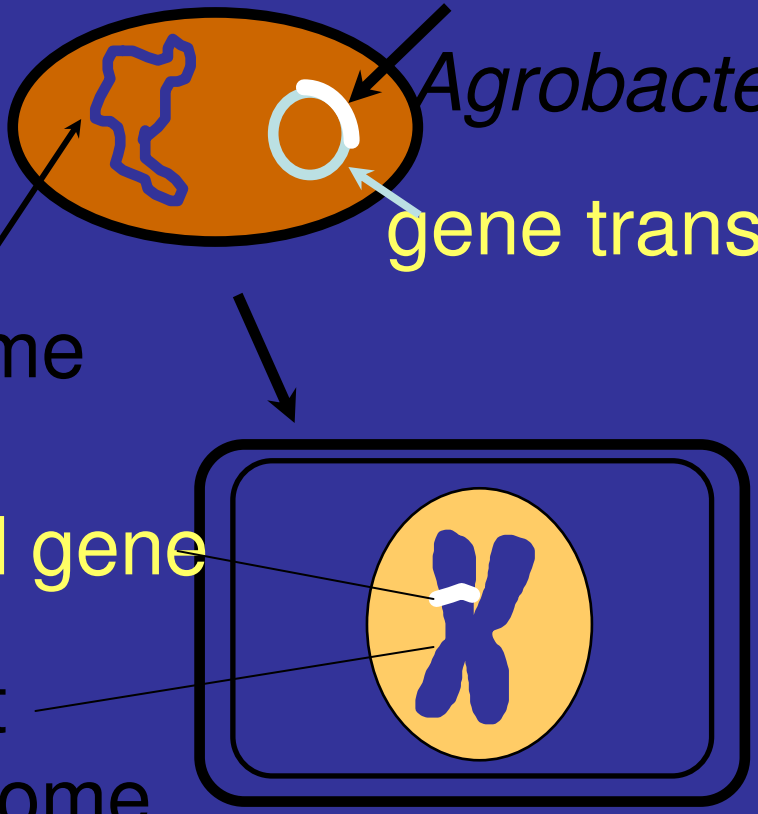
3. Transfer of DNA via carrier

- Protoplast microinjection
- Particle bombardment



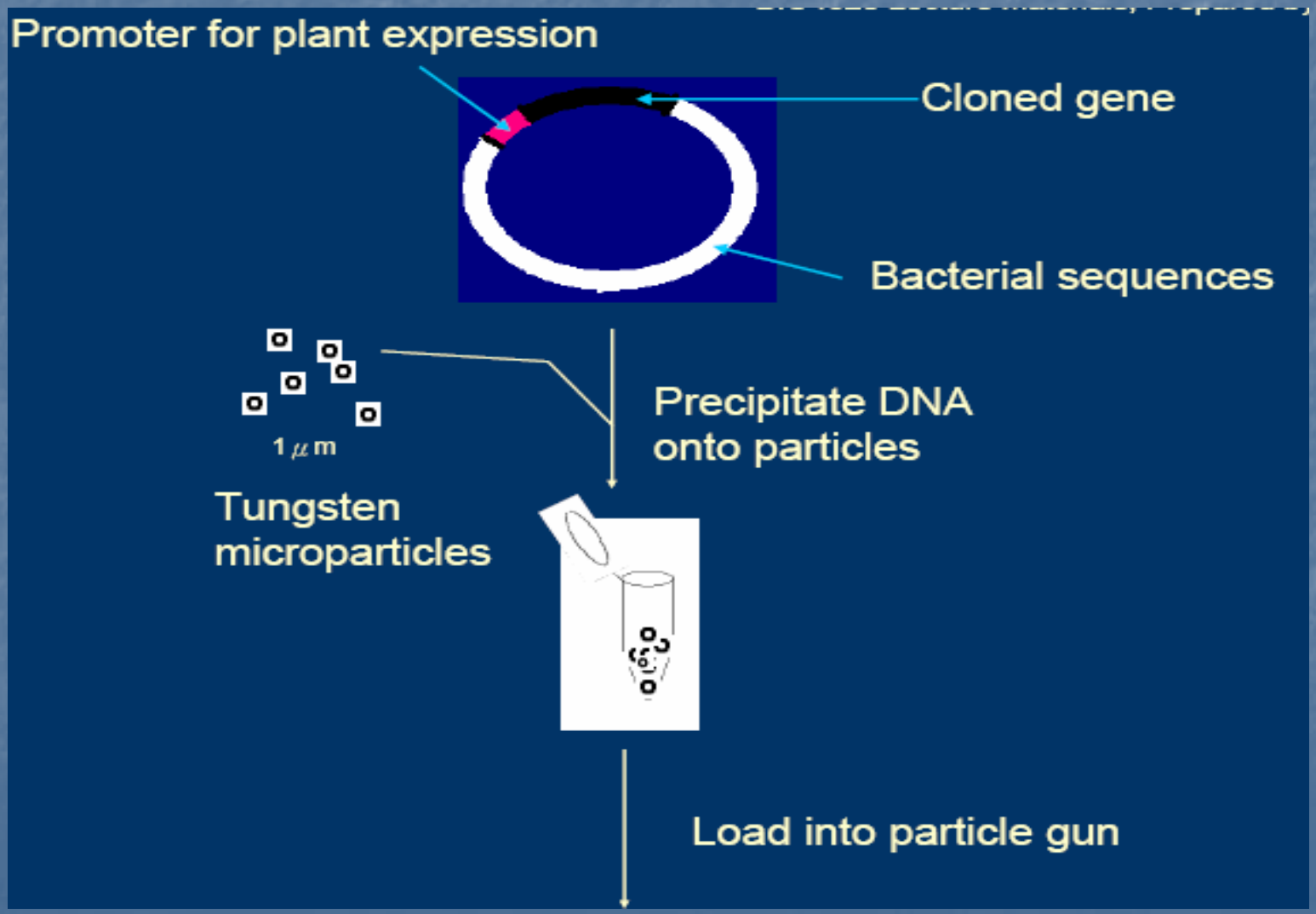
Agrobacterium method

disarmed T-DNA (contains transgene)

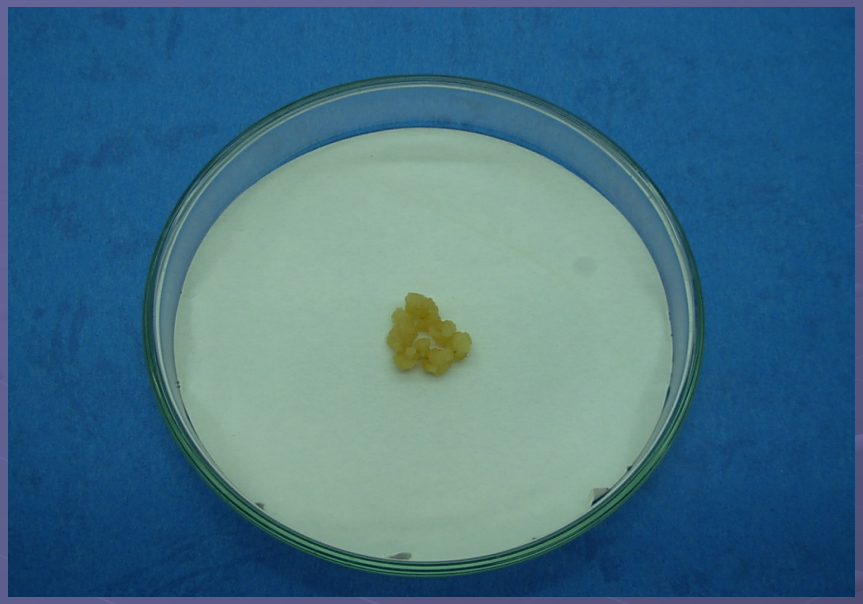


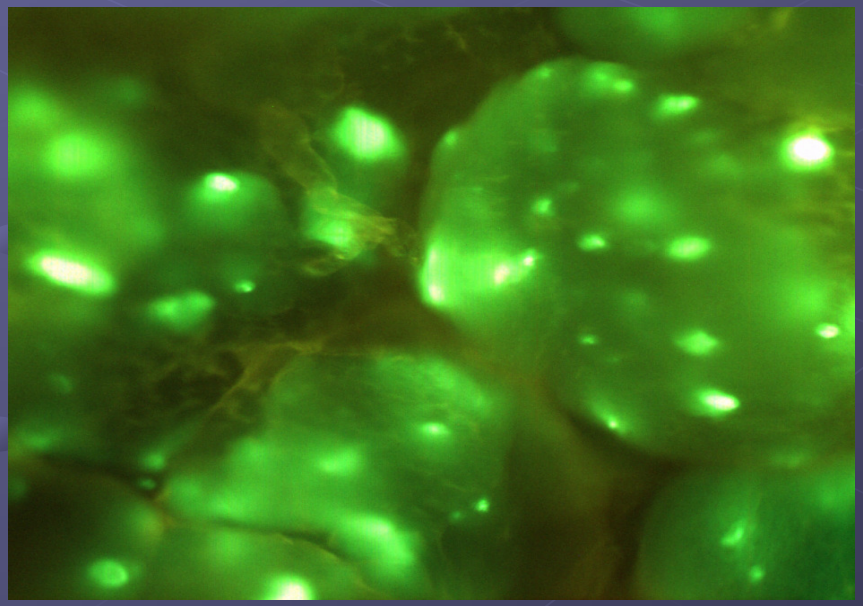
Source: Monsanto

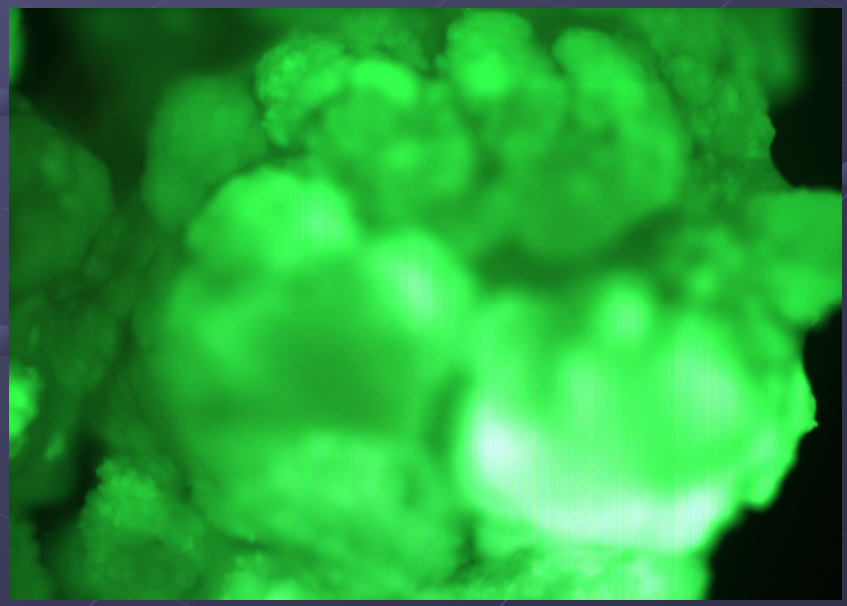
Particle bombardment: principle



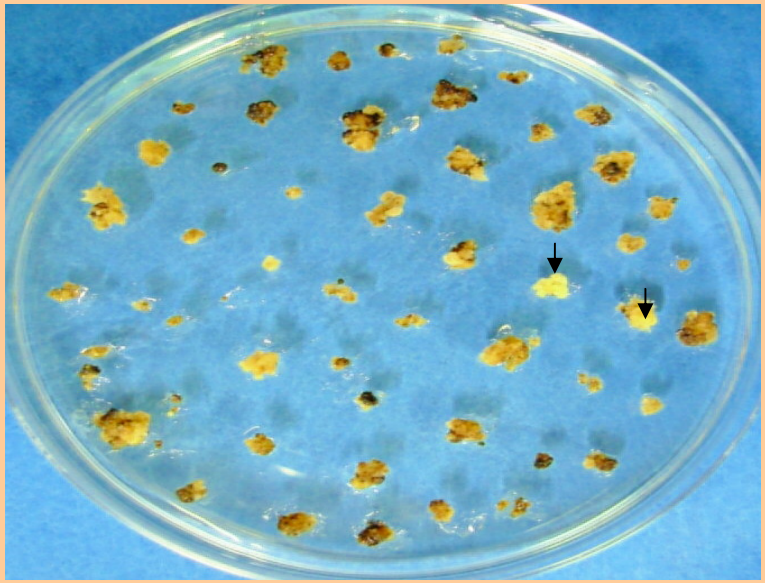
Helios Gene Gun

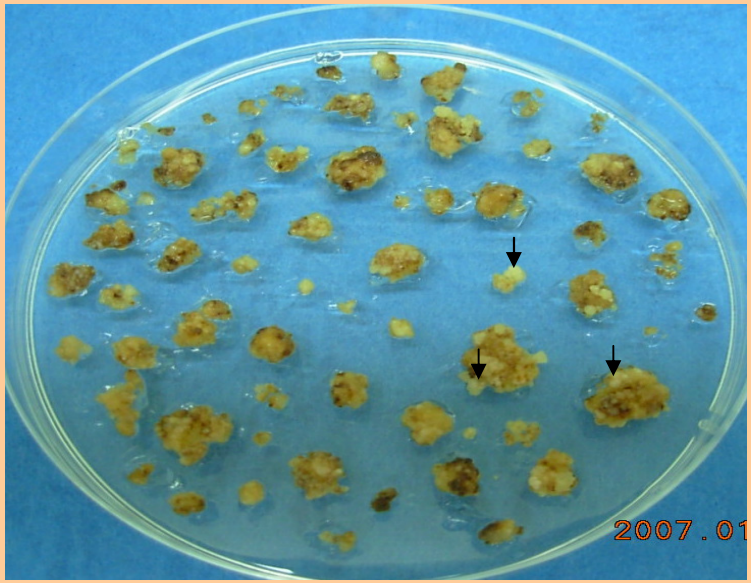




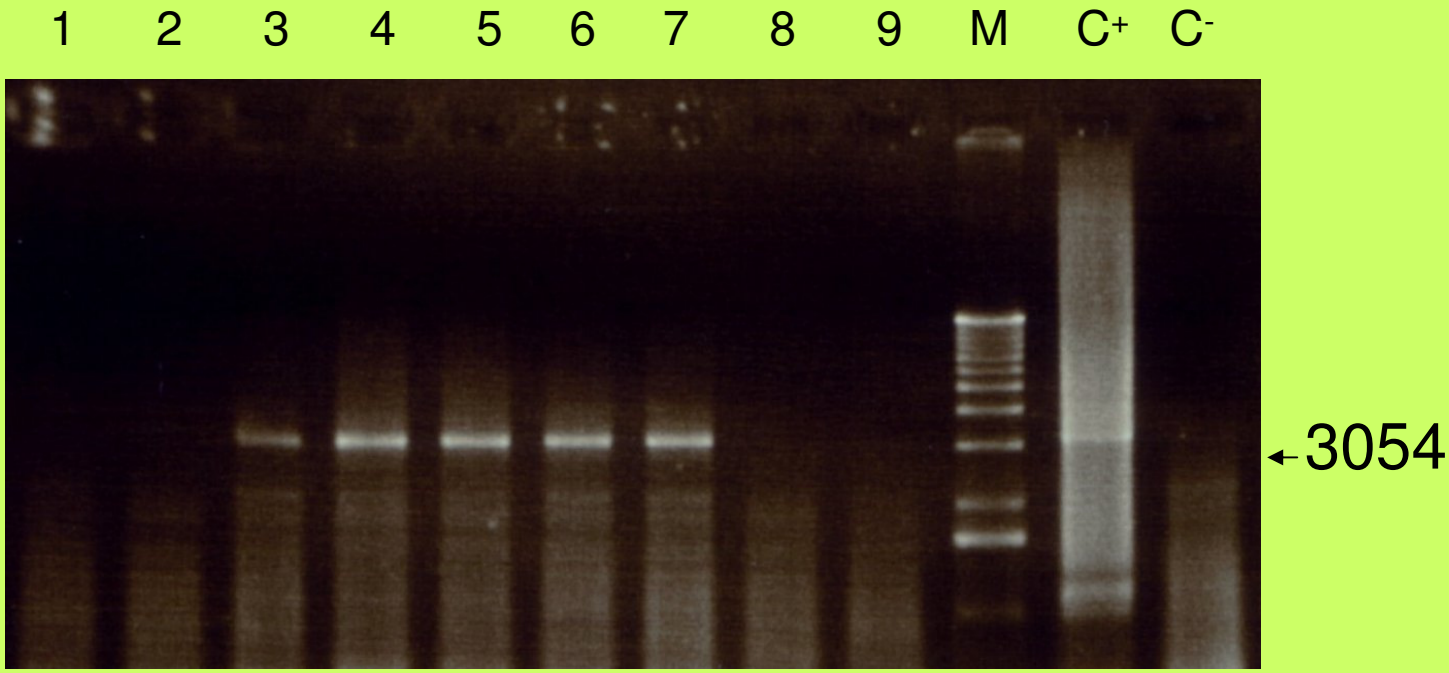


Experiment: production of transgenic rice

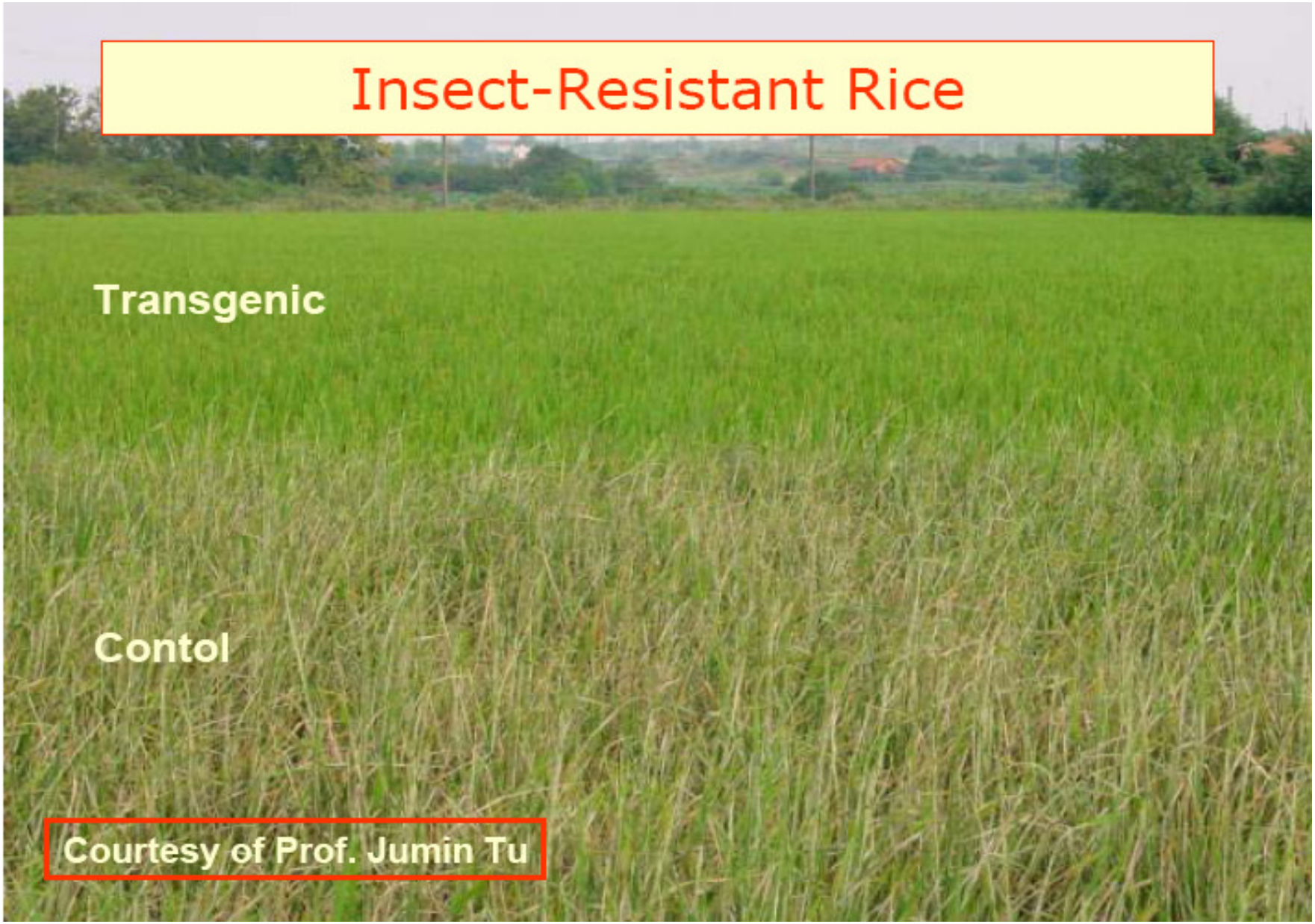




Detection of transgene in transgenic rice

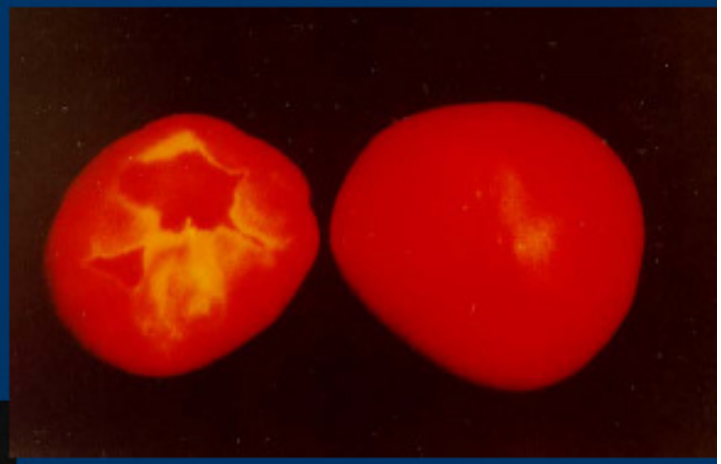


Insect-Resistant Rice



Virus Resistant Tomato

Courtesy of Prof. Samuel Sun

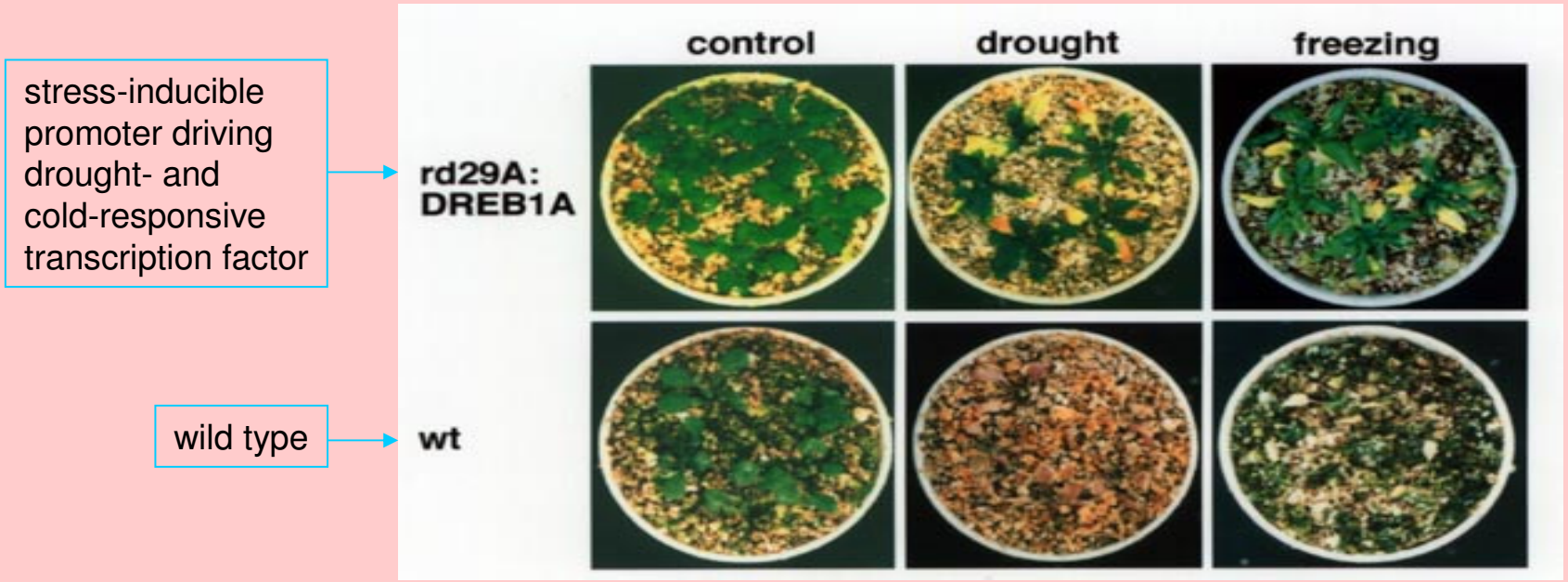


Regular Tomato

Genetically Engineered Virus Resistant Tomato

What are the Uses of Transgenic Plants?

- Research
  - Largest number of transgenic plants are currently created for research purposes
    - Knock-outs, over-expression, modified proteins



K. Yamaguchi-Shinozaki, JIRCAS, Japan

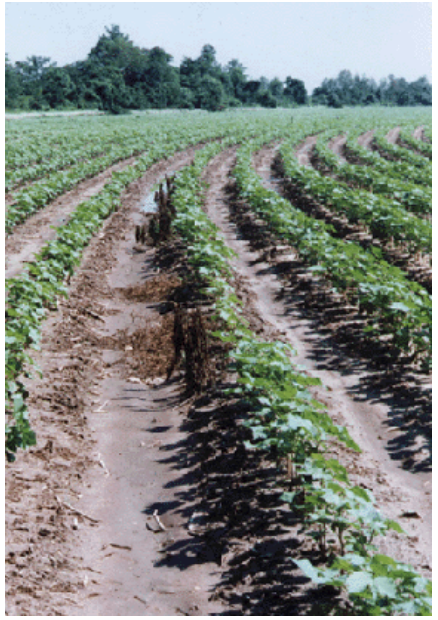
What are the Uses of Transgenic Plants?

- Commercial Applications
  - Altered agronomic traits
    - Disease/insect resistance
    - Virus resistance
    - Herbicide resistance
    - Salt/drought tolerance
    - Cold tolerance
    - Enhanced yields, other quantitative traits
    - Phytoremediation

time

2007

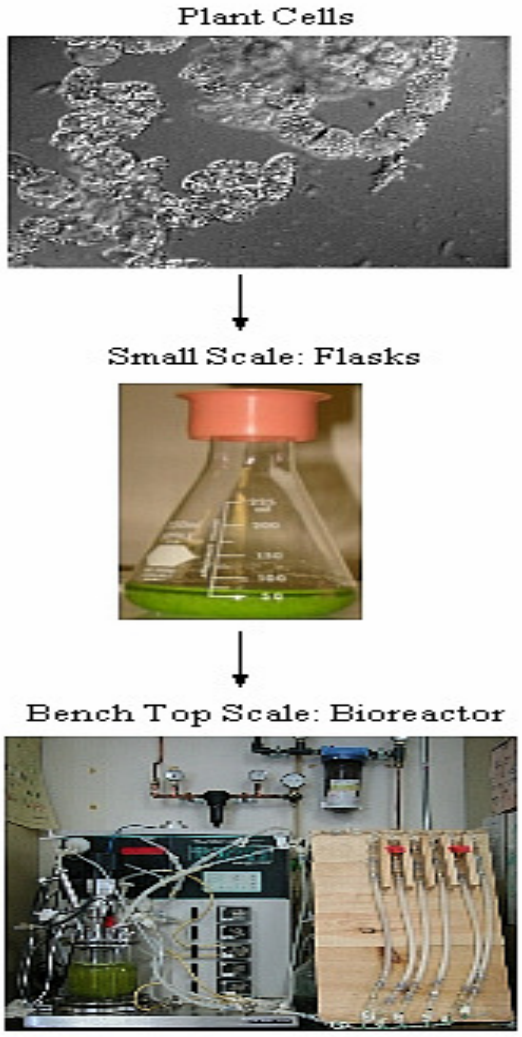
Application of Roundup herbicide



Field following application

Other uses of transgenic plants

- Bioreactors / Molecular farming
  - Therapeutic proteins
    - Human lactoferrin to treat iron deficiencies
    - Antibodies
  - Vaccine production
    - Antigen expression
      - HIV, HPV



Dow AgroSciences Achieves World's First Registration for Plant-Made Vaccines

Indianapolis, IN - January 31, 2006

Dow AgroSciences LLC, a wholly owned subsidiary of The Dow Chemical Company, (NYSE: DOW), announced today that it has received the world's first regulatory approval for a plant-made vaccine from the United States Department of Agriculture (USDA) Center for Veterinary Biologics. This approval represents an innovative milestone for the company and the industry...

Other uses of transgenic plants

- Functional foods (humans and livestock)
  - Today: Golden rice
    - Vitamin A enriched
  - Future directions:
    - Boosted antioxidants
    - Elevated content of specific minerals
    - Removal of food allergens, carcinogens

Greater public acceptance when the technology is shown to more greatly benefit consumers?

Future of transgenic technology

New techniques will improve efficiency and may resolve some health or environmental concerns.

- Insertion at specific points in the genome
- New marker genes to replace antibiotic resistance markers
- Better control of gene expression (only when and where needed)
- Transformation of chloroplasts rather than nuclei

Thank You !