Benha University **Faculty of Agriculture**

Agricultural Biotechnology Program

4th level / 1st Semester 2019- 2020

Agronomic Biotechnology **Model Answer**

The first questions (30 marks)

Answer **"three only"** from the following points:

1) Fill in the blanks

a) Carotene source of vitamin a

b) callus

c) totypotency

d) explant--- root – shoot – flower or callus

e) sub culture

f) Agrobacterium tumefuciens

2) What are types of tissue culture? Write short notes about advantage and disadvantages of tissue culture.

Types of tissue culture

1. Organized culture:

   - The culture of whole or parts of a plant. The characteristics and organizational structure of a plant or organ is maintained.
   - Axillary bud culture
   - Terminal bud culture
   - Seed culture
   - Embryo culture
   - Ovary culture
   - Pollen culture

3) What are the most famous methods of gene transfer? Write short note on only method of them.

Gene transfer strategies:

*Agro bacterium

*Direct DNA uptake like gene gun or Micro injection

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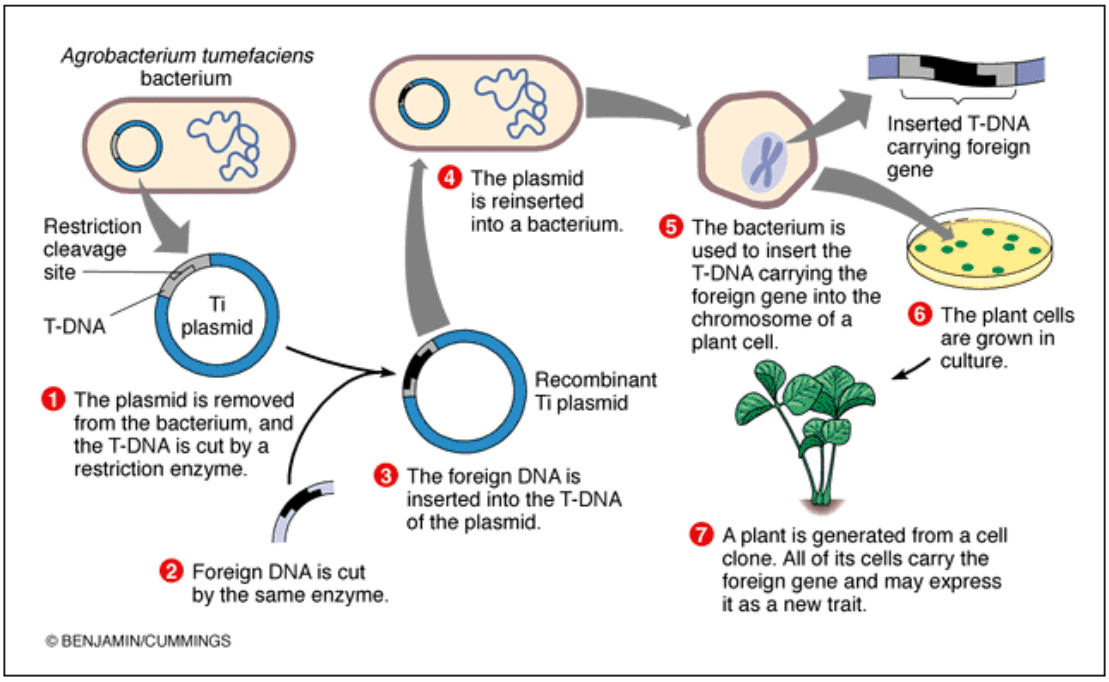
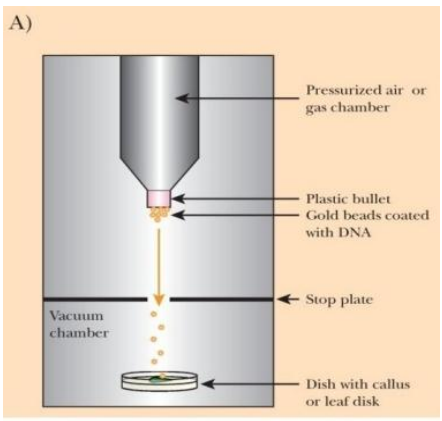


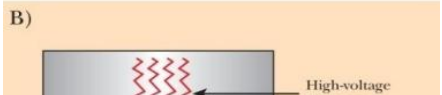
Summary of process illustrated in the diagram (above):

- The agrobacterium cell contains a bacterial chromosome and a Tumor inducing plasmid- "**Ti Plasmid**".
- The Ti plasmid is removed from the agrobacterium cell and a **restriction enzyme** cleaves the **T-DNA** restriction site.
- Next foreign DNA, which is also cleaved by the same enzyme, is inserted into the T DNA at the site that was **cleavage site**.
- The modified plasmid is then reinserted in the agrobacterium and the bacterium inserts the TDNA, which now carries a foreign gene into the plant cell.
- The plant cell is then cultured and results in a new plant that has the foreign DNA trait.

Short note Particle Bombardment Technology (Gene gun)

- Works with all types of plants.
- DNA is carried on microscopic metal particle.
- Fired by a gun into plant tissue.



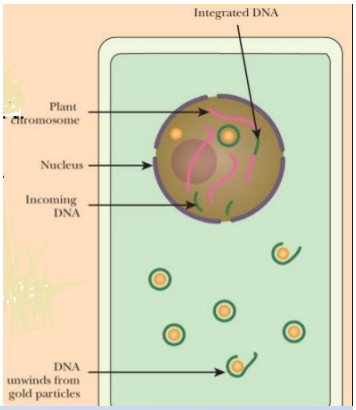


Method

- DNA coated on microscopic gold beeds.
- Beeds are placed at the end of a plastic bullet.
- Blast of helium used to project them.
- Plastic meshwork stop is used to stop the bullet.

- Alternative method is by strong electrical discharge.
- Amount of penetration into tissue can be changed.
- Beeds enter the cytoplasm or nucleus of the cell.

- DNA is free and recombine with chromosomal DNA.
- Leaf transferred to selection media for cell to grow.
- Transformed plants are regenerated using tissue culture techniques.
- Screened for gene of interest.



DNA Carried on Microscopic Gold Particles Can Integrate into Plant Chromosomes
After penetrating the cell, the DNA unwinds from around the gold carrier particle. Some of the DNA enters the nucleus and is successful in integrating into the plant chromosomes.

Short note on Vectorless Gene Transfer

Gene transfer can be effected by certain means that do not use vectors. These include

a. Electroporation: Temporary holes are formed in the plasma membrane of the host cell by applying a high voltage. These pores permit entry of foreign DNA.

b. Chemical mediated genetic transformation some chemicals, such as polyethylene glycol, help foreign DNA to enter the host cell.

c. Micro-injection: The host cell is immobilized by applying a mild suction with a blunt pipette. The foreign gene is then injected with a micro-injection needle.

4) **What are the most famous examples Which indicates the success of genetic engineering technology in the field of crops**
5) **Golden rice**
6) **Rice rich of iron**
7) **Corn resistance to insects Bt gene**
8) **Cotton resistance to insects Bt gene**
9) **Golden rice**

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The second questions (30 marks)

Answer "**three only**" from the following points:

1) What are the most application of tissue culture in crop improvement?

Application of tissue culture in crop improvement
¬Newer molecular and cellular technologies have a broad significant impact on crop improvement
¬The various application of tissue culture approaches to crop improvement are-
Breeding and biotechnology
¬Wide hybridization
¬Haploids
¬Soma clonal variation
¬Micropropagation
¬Synthetic seeds
¬Pathogene radication
¬Germ plasm preservation
¬Plant breeding separated in to two activities
¬ Manipulating gene ¬ Plant evaluation
¬Controlled pollination of plants lead to specific crosses result in new generation performed better in the yield
¬Tissue–culture techniques is having significant impact in manipulating genetic diversity
-Wide hybridization

2) Summarize the applications of molecular marker for crop improvement.

Giv1. DNA fingerprinting for varietal identification
2. Germplasm evaluation
3. Phylogenetic and evolutionary studies
4. Development of saturated maps
5. Gene tagging
6. Marker assisted selection
7. Comparative mapping
8. Construction of linkage maps and QTL mapping
9. Map-based gene cloning
10. Molecular markers in heterosis breeding

3) e the scientific term (without explanation) of the following abbreviations:

AFLP – PCR – SSR – RAPD – MAS

AFLP : Amplified Fragment Length Polymorphism

PCR: Polymerase Chain Reaction

ISSR: Inter Simple Sequence Repeats

SSR: Simple Sequence Repeats

RAPD: Random Amplified Polymorphic DNA MAS: Marker- assisted selection

4) Explain the steps of PCR?

The three steps of PCR are:

1. Denaturation: Unwinding the double helix by heating to 95 degrees Celsius for 30 seconds.
2. Annealing: Priming the DNA by cooling the test tube to 50 degrees Celsius for 30 seconds.
3. Extension: Adding on complementary nucleotides and reheating to 72 degrees Celsius for 60 seconds.

5) Why molecular marker systems are better than "classical" markers in detecting variation?

Molecular Technology	traditional Breeding
1-short time process 2- easily did not need power 3- Effective in crop improvement. 4- not need to make artificial environment for screening genotypes 5- Possibility of detect gene in any period of plant life	1-Long process 2-Lot of man power 3- Limited possibility of improved traits. 4- need to evaluate genotypes under stress condition 5- there were a specific for showing the variation

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