TRANSGENIC OIL PALM MEDIATED BY Agrobacterium

tumefaciens

DAYANG IZAWATI ABANG MASLI; AHMAD PARVEEZ GHULAM KADIR and ABDUL MASANI MAT YUNUS



MPOB INFORMATION SERIES • ISSN 1511-7871 • JUNE 2008

MPOB TT No. 401

enetic engineering is a powerful tool to manipulate the genome of an organism for it to produce a new trait(s) or to change its existing trait(s). Plant genetic engineering involves procedures like gene isolation, characterization, modification and, eventually, introduction and expression of the gene in the transgenic plant. It covers the fields of tissue culture, cell biology, molecular biology and plant gene transfer (plant transformation). Plant transformation is now a core research tool in modifying the genetic traits for improving plants. Basically, transformation is merely the transfer of a gene(s) into a plant cell and then regenerating the

transgenic cell into a plant. The ultimate objective is to regenerate a normal fertile plant with the new trait(s).

Agrobacterium – MEDIATED TRANSFORMATION

There are several ways to introduce foreign genes into plants, for example, *Agrobacterium*-mediated transfer, particle bombardment, polyethyleneglycol transfer, electroporation and microinjection. *Agrobacterium tumefaciens* mediation has become the most popular method, followed by particle bombardment (*Figure 1*). *Agrobacterium* has been used to introduce traits like herbicide tolerance, and virus and disease resistance.

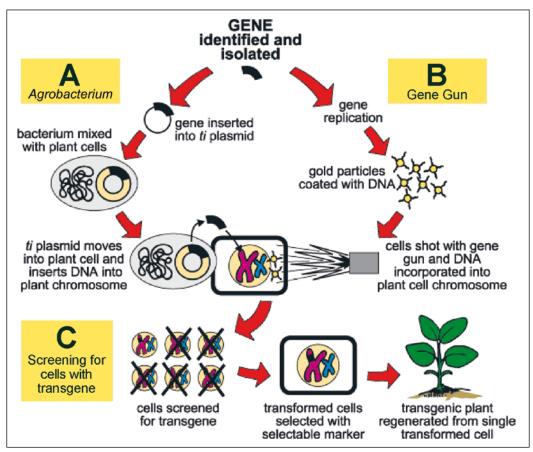


Figure 1. Plant transformation mediated by Agrobacterium and bombardment.





CONCEPT OF Agrobacterium TRANSFORMATION

The gene transfer (or transformation) process starts with recognition and attachment of *Agrobacterium* to the host plant. Phenolic compounds produced by the plant will activate the *vir* genes in *Agrobacterium* which will lead to the transfer of T-DNA (the contained gene of interest) from the bacterium to the plant. The T-DNA will then integrate into the plant cell genome and the transformed cells selected using the inserted selectable marker gene (*Figure 2*).

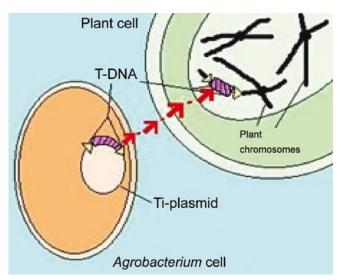


Figure 2. Agrobacterium-mediated transformation process.

OIL PALM TRANSFORMATION

Embryogenic callus is used as the target tissue in *Agrobacterium* transformation of oil palm. Preand post-treatments of the callus are carried out with the transformation process (*Figure 3*). After co-cultivation, the oil palm embryogenic calli are exposed to the herbicide Basta to screen for resistant embryogenic calli, which are then regenerated into whole plants (*Figure 4*). The transgenic status is confirmed by molecular analyses (*Figure 5*) and leaf painting (*Figure 6*).

ADVANTAGES OF Agrobacterium MEDIATED TRANSFORMATION

This method is technically simple and cheap. It reduces the required copy number of the transgene, leading to fewer problems with transgene cosuppression, gene silencing, rearrangement and instability. In addition, it is a single-cell transformation system which would eliminate the formation of chimaeric plants, which occur

more frequently with direct transformation. Furthermore, the size of foreign DNA that can be carried by the transformation vector is unlimited. The transformants are mitotically and meiotically stable.

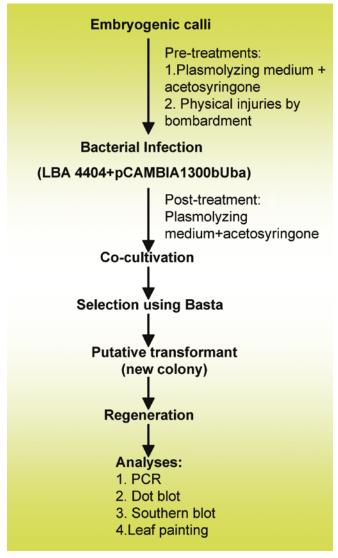


Figure 3. Flow chart of oil palm transformation process.

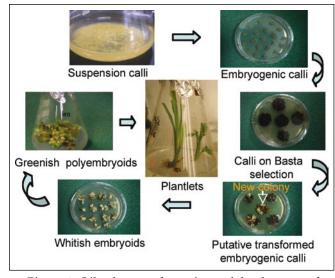


Figure 4. Oil palm transformation and development of transgenic oil palm.

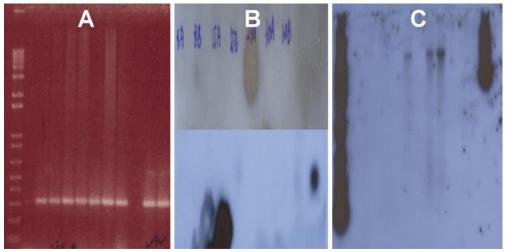


Figure 5. Molecular analyses of transgenic oil palm (A) PCR analysis (B) Dot blot (C) Southern blot.

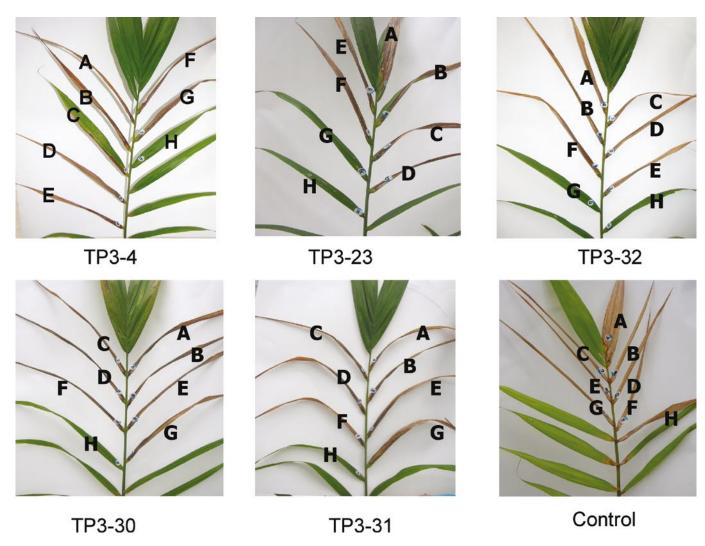


Figure 6. Leaf painting analysis: comparison between transformed and untransformed plants after exposure to Basta solutions of different concentrations. A: 1000 μg ml⁻¹, B: 500 μg ml⁻¹, C: 300 μg ml⁻¹, D: 250 μg ml⁻¹, E: 200 μg ml⁻¹, F: 150 μg ml⁻¹ G: 100 μg ml⁻¹ and H: 50 μg ml⁻¹. The lowest concentration (50 μg ml⁻¹) Basta did not kill any transgenic leaves as with the control leaves.

WHO SHOULD BENEFIT

The oil palm industry, especially tissue culturists and breeders. Using this method they can introgress desired trait(s) into oil palm. It will save 70% - 80% of the time from using conventional breeding.

INTELECTUAL PROPERTY

A patent has been filed for in Malaysia – *A Method for Producing Transgenic Plants* (PI 20071240).

For more information kindly contact:

Director-General MPOB P. O. Box 10620 50720 Kuala Lumpur, Malaysia. Tel: 03-87694400 Website: www.mpob.gov.my