Development of Transgenic Bananas with Improved Agricultural Traits

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Introduction

In Malaysia, bananas is a lucrative commercial crop and are planted on a commercial scale. The breeding of banana for improved agronomical traits or disease resistance by conventional methods has been limited mainly due to the long generation times, triploidy and sterility of most edible cultivars. The application of genetic engineering techniques to study banana improvement is an attractive alternative. The use of biolistic technique and *Agrobacterium tumefaciens transformation* are preferred methods to introduce foreign genes into bananas. The biolistics process is a method by which foreign substances are introduced into intact cells via high velocity micro projectiles. Corm slice asnd scalps are reported to be good target materials for transformation of bananas as they are easily regenerable. Our study reports a successful method of genetic transformation in local banana cultivars.

Materials and Methods

Callus tissue of banana was obtained from *in vitro* cultured plants. Scalps were obtained from proliferated shoot tips and embryogenic cell suspension was initiated from meristematic globules derived from the scalps. Scalps of 1.0x0.5 cm2 in size and embryogenic cell suspension of 0.1 to 0.5 ml were subjected to the bombardment procedure. Scalps were plated on MS medium supplemented with 10uM BAP and 1.0uM IAA and embryogenic cell suspension was plated on hormone free ½ MS medium for one week prior to bombardment. Bombardment was performed using the Biolistic PDS-1000/He system instrument. The bombardment parameters were; vacuum: 28 inches Hg, Target distance: 6,9 and 12cm; Helium pressure: 450, 900, 1100 and 1350 psi and particle size: 1.0 g.

The *Agrobacterium tumefacies*- mediated transformation system has been used and to improve the condition of *Agrobacterium* infection, various factors such co culture periods, strains and concentration of acetosyringone were examined by GUS transient and GFP assay.

Results and Discussion

The most effective helium pressure for scalp transformation were 1100 and 1350 psi with a target distance of 9 cm and a helium pressure of 900psi with a target distance of 6 cm was most efficient for the transformation of embryogenic cell suspension. The optimal condition for the transient expression of gfp-flourescence was found to be at 1100psi disk pressure and 12 cm target distance for callus and 6 cm for rhizome slices. The gfp-gene with green fluorescence color could be detected even after 3 months of bombardment, which is in regenerated rhizome slices. GFP can be used as a visual and non-destructive reporter; it provides the opportunity to follow growth and development of transformed cells. Stable expression of gene of interested was achieved via *Agrobacterium tumefaciens* transformation system with 30 minutes co culture period, super virulent binary strains and 100 uM concentration of acetosyringone based on histochemical GUS transient assay. Transformation of corm slices which contain numerous adventitious bud was particularly effective because these thin slices of tissue can be readily exposed to selective inhibition during plantlet formation, thus reducing possibilities of regenerating chimeric plants or non-transformed plants.

Conclusions

A stable transformation protocols in bananas has been developed. This technique is worth considering as a potential route to producing disease-resistant cultivars as resistant to Fusarium wilt disease, which is a major fungal disease in bananas in Malaysia.

Benefits from the study

Establishment of new banana varieties in the future those are of superior quality as dessert and processed fruit and have potential for commercialisation of superior elite varieties.

New techniques and technologies were developed in producing superior banana cultivars.

The study also accounted for a number of publications in national and international journals, and provided scope for post-graduate programs for human resource development.

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