

## ABSTRACT

### Agrobacterium Mediated Transformation of *Anthurium andreanum* Towards Engineering Resistance to Bacterial Blight.

Morexa M. Martin

The objective of the project was to investigate the various aspects that are involved in engineering resistance to bacterial blight in anthurium. It involved:

- (1) – The evaluation of the various protocols which are available for *in vitro* propagation, to determine which of the widely used protocols, best facilitates *in vitro* response on a range of cultivars and is suitable for regeneration of transformants. Of the inoculation methods investigated, petiole inoculation of *in situ* plants was found to be the most appropriate. However,
- (2) - The development of an efficient protocol for agrobacterium mediated transformation, and did not seem appropriate for determining levels of resistance. Alternative strategies for screening are discussed.
- (3) - Investigation of the bacterial pathogen in order to propose a suitable gene(s) for use in agrobacterium mediated transformation, as well as a suitable screening technique to evaluate potential transformants.

The results indicated that though there was genotype to medium interaction at initiation, a modification of Pierik's (1976) protocol is the most appropriate for

*in vitro* propagation of the cultivars which were investigated. It was also found that shoots and leaves from *in vitro* produced plants can also be induced to produce callus and eventually plantlets when this protocol is used. There was no evidence of genotype to medium interaction during the proliferation stage of micropropagation, however, it was found that the extent of proliferation was in fact genotype specific.

A modification of the protocol which was proposed by Chen and Kuehnle (1996) for transformation of *Anthurium andreaeanum* was found to be less efficient than modifications of the grape protocol proposed by Perl *et al.* (1996), as well as the protocol which was proposed by Charity pers. comm. (1997), for transformation of etiolated internodes, shoots/cuttings and callus.

Several pathogenic isolates were characterised at the CMI as saprophytic *Pseudomonas* species. Of the inoculation methods investigated, petiole inoculation of in situ plants was found to be the most appropriate. However, resistance did not appear to be at the level of petiole and leaf, and varied with plant age and leaf age and did not seem appropriate for determining levels of resistance. Alternative strategies for screening are discussed.

**Keywords:** agrobacterium mediated transformation, *Agrobacterium tumefaciens*, *Anthurium andreaeanum*, bacterial blight, biotechnology, disease resistance, *in vitro* propagation, *Pseudomonas*, *Acidovorax*.

Finally I will like to thank God for the health, strength, opportunities, special friends and family which he provided as my valuable support system.