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Agrobacterium mediated transfer of cry 2 gene to Ixora odorata

Ixora is a woody ornamental plant, which belongs to the family Rubiaceae. It is especially popular in the export sector. However, Lepidopteran pests are the major constraint in the production of Ixora species. The main objective of this study was to develop insect resistant Ixora odorata plants by transferring cry 2 gene from *Bacillus thuringiensis* via *Agrobacterium tumefaciens* vector system.

Ixora leaves were used as explants and cultured on half-strength Murashige and Skog (MS) medium containing 2,4 D (2mg/l) for callus induction. Then *Ixora* calli and leaf disks were co-cultivated separately with *Agrobacterium* strain AGL I(H) carrying recombinant pBI 121 with cry 2 gene from *Bacillus thuringiensis* strain 6e. pBI 121 consists of the selectable marker gene NPT II (Kanamycin resistance gene) and the reporter gene GUS (Glucuronidase). Five hundred milligrams per liter of cefotaxime controlled *Agrobacterium* successfully. The calli and leaf disks were sub-cultured to remove excess cefotaxime.

The time taken for callus induction was longer in co-cultivated leaf disks than normal leaf disks. Therefore *Ixora* calli were better than *Ixora* leaf disks for gene transformation. Kanamycin cannot be used as a selectable marker due to natural kanamycin resistance of *Ixora odorata* plants. Histochemical GUS assay and dot blot analysis were used for the confirmation of gene transformation. Thirty three percent of the co-cultivated calli were dot blot positive. Co-cultivated *Ixora* calli were cultured on half-strength MS medium supplemented with 2mg/kinetin for shoot induction.