

Executive Summary of the Major Research Project Entitled “Generating insect’s resistance cowpea plants” sanctioned to Dr. Darshna Chaudhary, Assistant Professor, Centre for Biotechnology, MDU, Rohtak by University Grant Commission, New Delhi,

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A simple and reproducible *in vitro* plant regeneration system was developed by using 4-d-old cotyledonary node explants. *Agrobacterium*- mediated genetic transformation system of cowpea for production of fertile stably transformed plants was developed. The different parameters affecting *Agrobacterium*- mediated gene transfer in 4-d-old cotyledonary node explants were optimized by using a reporter gene *uidA* through *A. tumefaciens* strain EHA105 (pCAMBIA2301) for transient GUS expression. The optimal conc. of kanamycin for selection of transformed plants was also identified. The kanamycin at 85mg/l was found most effective in Pusa Komal genotype. The effect of phenolic compound acetosyringone and thiol compounds like L-cysteine (5.0mM) and dithiothreitol (1.5mM) in bacterial inoculation and co-cultivation medium were also standardized. The frequency of transient GUS expression was approximately 100% in sonicated cotyledonary node explants inoculated with *Agrobacterium* 10^6 cells/ml for 25 min and co-cultivated for 3 d at 25°C in co-culture medium having acetosyringone (100 µM) and thiol compounds like L-cysteine (5.0mM) and dithiothreitol (1.5mM) at pH-5. The addition of thiol compounds and sonication of explants had positive effect on GUS expression. For each variable in the experiment, 28 explants were used and each experiment was repeated thrice. By using this method, putative insect resistant transgenic plants were developed by using *Agrobacterium tumefaciens* strain EHA105 carrying a binary vector pBinAR-*cry2Aa* and established in soil and analyzed by PCR for the presence of *nptII* gene and *cry* gene. Molecular analysis by Southern and Western blotting is under progress.