Nootjaree Tudses 2009: The Transformation of *Green Fluorescent Protein* Gene into *Cryptocoryne* spp. *via Agrobacterium tumefaciens*. Master of Science (Agricultural Biotechnology), Major Field: Agricultural Biotechnology, Interdisciplinary Graduate Program. Thesis Advisor: Assistant Professor Sermsiri Chanprame, Ph.D. 134 pages.

The multiple shoots of 2 *Cryptocoryne* species, *C. affinis* and *C. wendtii* 'Brown' were induced from apical shoot explants for using as plant material for genetic transformation. The shoot explants were cultured for 5 days in MS liquid medium supplemented with 0.01 mg/l TDZ and 0.3 mg/l BA then transferred onto MS solid medium for 4 weeks in which, for *C. affinis,* 4 mg/l BA was added and for *C. wendtii* 'Brown', 1 mg/l BA was added. Thereafter, the procedures of gene transformation in *C. affinis* and *C. wendtii* 'Brown' using *Agrobacterium tumefaciens* were optimized. Two strains of *A. tumefaciens* harboured the pCAMBIA1304 which contained *β-glucuronidase* (*gus*) and *green fluorescent protein* (*mgfp5*) genes as reporter genes and *hygromycin phosphotransferase* (*hpt*) gene as a plant selectable marker gene were compared. The 5 mm long *in vitro* shoots wounded by cut in half lengthwise followed by 5 sec sonication were suitable the target tissues. The suitable stain of bacterial for *C. affinis* transformation was *A. tumefaciens* strain EHA105 and for *C. wendtii* 'Brown' was the strain AGL-1. *Agrobacterium* was also activated by100 μM acetosyringone during inoculation and co-cultivation steps. The optimal period for inoculation was 2 hr and for co-cultivation was 3 days. The bacteria was then eliminated from the explants followed by selected on multiple shoots induction media stated above and supplemented with 5-10 mg/l hygromycin and 300 mg/l cefotaxime for 6 weeks. The survived shoots were then cultured on the same medium but without any antibiotic. The number of 35 putative transformed shoots of *C. affinis* and 6 of *C. wendtii* 'Brown' were recovered. After subjected to PCR analysis of *gus, mgfp5* and *hpt* genes, the result revealed the difference in the presence of the transgenes in putative transformants which confirmed that the putative transformed shoots were chimeras. Latter, the putative transformed shoots were investigated for the expression of *mgfp5* gene using stereo microscope (excitation at 480 nm and emission at 510 nm) and only red and yellow fluorescence were observed but not the green fluorescence.

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