DEVELOPMENT OF Agrobacterium rhizogenes MEDIATED TRANSFORMATION PROTOCOL FOR INDICA RICE

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Agronomical important genes in rice have been identified based on whole-genome and transcriptome sequencing. Among those, number of candidate genes associated with root architecture, physiological functions and biotic and abiotic interactions are predicted. Functional characterization and confirming their functions are essential in developing gene-specific molecular markers for rice breeding. Nevertheless, such efforts are limited due to less efficiency of available rice genetic transformation systems. Therefore, the current project focused on developing an Agrobacterium rhizogenes mediated root transformation protocol for rice, result in chimeric plants with transgenic roots and non-transgenic shoots. The Yellow Fluorescence Protein (YFP) reporter gene and Indica rice variety Bg250 were picked as the choice of gene and the rice genotype, respectively. A three-factor factorial experiment was designed to test the effect of explant type, bacteria inoculation method and presence of sugar in the co-cultivation media on transformation efficiency, defined as the percentage of explant expressing YFP. Either a 24 h old liquid or solid A. rhizogenes strain, MSU 440 harbouring pBIN-YFP vector was used to inoculate freshly wounded two-days old seedlings at either at root tip or root shoot junction. Inoculated plants were placed on MS medium either with 3% sugar or without sugar and maintained at 15 °C for 14 days. Then the plants were transferred to the same media combinations with Cefotaxime (at 300 mgL⁻¹) and maintained at 24 °C at 16 h light at 85 µmolm⁻²s⁻¹ light intensity and 8 h dark. After seven weeks, root shoot junction explants, bacteria inoculated by liquid method, co-cultivated and maintained in MS medium supplemented with sugar resulted 41% YFP expressing calli and 21% YFP expressing roots.

Keywords: Agrobacterium rhizogenes, Bg250, Indica rice, Plant transformation, Yellow Fluorescence Protein (YFP).