



612/E2

Development of transgenic rice plants with lysine rich protein coding gene

W W P Rodrigo,^{1*} H H K Achala,¹ K G W W Bandara,¹
W T G S L Withana,¹ N V Chandrasekharan² and S G Senaratne³

¹Biotechnology Unit, Industrial Technology Institute, 363, Bauddhaloka Mawatha,
Colombo 07

²Department of Chemistry, Faculty of Science, University of Colombo, Colombo 03

³Vocational Training Authority of Sri Lanka, 354/2, "Nipunatha Piyasa",
Elvitigala Mawatha, Colombo 05

Even though it is the staple food of more than half the world's population, rice is not considered a rich source of protein. One of the essential amino acids, lysine is low in rice seed. Hence, this study was aimed at increasing both the lysine and total protein content in rice seeds by introducing the pollen-specific lysine-rich protein encoding gene (*SBgLR*) from potato (*Solanum tuberosum*) into indica rice (*Oryza sativa* L.) seed under the control of the rice seed-specific globulin promoter. Total Ribonucleic Acid (RNA) from potato pollen grains was extracted and the produced cDNA and *SBgLR* gene was amplified by Polymerase Chain Reaction (PCR) using *SBgLR* gene-specific primers. The isolated rice genomic Deoxy Ribonucleic Acid (DNA) from rice variety Bg 94-1 was subjected to PCR to amplify the promoter sequence of the globulin gene. The amplified promoter region was cloned into pGEM[®]-T Easy vector, and then into pCAMBIA1391Z vector. Recombinants were selected and sequenced. The *SBgLR* gene containing recombinant vector (pCR[®]2.1-TOPO-*SBgLR*), previously cloned in our laboratory was restriction digested with *Bam*H1 and *Eco*R1 enzymes, and cloned into the corresponding sites of pCAMBIA1391Z-*Glb* vector construct. Electro competent cells of *Agrobacterium* strain GV3101 was transformed with the recombinant construct (pCAMBIA1391Z-*Glb-SBgLR*). Bg 94-1 rice seed derived calli, and active rice embryos were used for transformation. The embryo transformation method proved less time consuming and more effective in producing transgenic rice plants. PCR analysis of regenerated transformed plants indicated the presence of the *SBgLR* gene.

Keywords: *Agrobacterium*, lysine, transgenic rice, transformation

Acknowledgement: TG11/53 from the Sri Lanka Treasury