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Cloning of potato *SBgLR* gene and rice *Glb* promoter to produce lysine-rich rice

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Rice which is the staple food in Sri Lanka is deficient in lysine which is one of the eight essential amino acids. As a result, protein deficiency disorders are particularly severe where rice is the major staple food. Genetic engineering is one of the possible ways to resolve this nutritional problem through enhancing the total lysine and total protein content in rice. The objective of this study was to increase both the lysine and total protein content of the rice seeds by introducing a pollen-specific *Solanum tuberosum* genomic lysine-rich (*SBgLR*) gene from potato (*Solanum tuberosum* L.) into rice seed under the control of the rice seed specific globulin (*Glb*) promoter.

Total RNA from potato pollen was reverse transcribed with oligo dT primers and PCR amplified using *SBgLR* gene-specific primers. The amplified product (~ 650bp) was purified and cloned in pCR®2.1-TOPO® vector. Recombinants were screened by colony PCR and putative clones were subjected to restriction digestion with *Eco* RI. A fragment of the expected size (~ 650bp) was observed. The clone was sequenced and analyzed using a sequence analysis program. A search of the non-redundant nucleic acid database at National Center for Biotechnology Information (NCBI) revealed significant similarity (95%) to a previously cloned sequence (Acc. No. AY377987.1) which had been used to design *SBgLR* gene-specific primers. The deduced amino acid sequence of the *SBgLR* protein in this study displayed a high degree of similarity (94%) to the deduced amino acid sequence of the *SBgLR* sequence cloned previously.

In order to clone the *Glb* promoter, DNA was isolated from rice leaves (*Oryza sativa indica*) and was amplified by PCR using gene-specific primers for the *Glb* promoter. The amplified product (~ 1000bp) was cloned in pCR®2.1-TOPO® vector. The colonies were screened by a rapid screen method and colony PCR to identify the recombinant clones. Recombinant clones were also subjected to restriction digestion with *Eco* RI. A fragment of expected size (1000bp) was observed confirming the cloning *Glb* promoter.

At present, work is in progress to clone the *SBgLR* gene under the control of the *Glb* promoter in pCAMBIA for *Agrobacterium* mediated transformation to rice.

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