(05-1-77)

Cloning and Functional Analysis of Tuber-Specific Promoter for Lipoxygenase in Potato (Solanum tuberosum L.)

Soon Hwa Kwak, Tae Young Chung, Moosik Kwon

Department of Genetic Engineering, Sungkyunkwan University, Suwon 440-746, Korea

Objectives

Potato (Solanum tuberosum L.) is one of the most important crops worldwide. In development of transgenic potatoes, tuber-specific promoter is very important for effective gene expression, protein targeting, and plant growth. In this study, tuber-specific gene expression of lipoxygenase1 (lox1) was investigated, and 5'-flanking region of the gene was cloned and analyzed.

Materials and Methods

Total genomic DNA was isolated from potato (Solanum tuberosum L. cv. Atlantic) by CTAB method. Gene-specific primers and partial gene fragment of lox1 were used for amplification of DNA fragment, and 5'-flanking region of lox1 gene was cloned using Genome Walker kit (Clonetech Co., USA) and was analyzed.

Results and Discussion

According to the results of RNA gel blot analysis by Kolomiets et al. (The Plant Cell 13:613-26, 2001), lox1 transcripts were detected only in underground organs, not in leaves, stems of flowers, and the highest levels of mRNA occurred in actively growing tubers. A portion of 5'-flanking region (1.5kb) of lox1 was cloned and sequenced. Within this region several putative cis-acting elements were identified. 1,285-1,289 and 1,409-1,414 regions of cloned DNA fragment are predictied as CAAT-boxes, and 1,498-1,504 region is predicted as a TATA-box by homology search in sequence database (PlantCARE: http://intra.pasb.ugent.be:8080/PlantCARE/). Putative promoter sequence and putative transcription start site are in 1,530-1,551 region of the clone. To confirm the promoter activity and essential sequence motif, the proximal 1.5kb and progressively 5'-deleted fragments were fused with a β-glucuronidase (GUS) reporter gene. The tissue-specific expression patterns will be investigated using transgenic potato. Development of tuber-specific promoter will be useful to control the foreign gene expression in potato.

*Corresponding author : Moosik Kwan, Tel : 031-290-7861, E-mail : mskwon@skku.edu