(05-1-32)

Analysis of *Cis*-elements in a Multiple Stress-inducible *SWPA4* Promoter from Sweetpotato

Sun-Hwa Ryu, Yun-Hee Kim, Suk-Yoon Kwon, Sang-Soo Kwak

Laboratory of Environmental Biotechnology, Korea Research Institute of Bioscience and Biotechnology (KRIBB), Oun-dong 52, Yusong-gu, Daejeon 305-806, Korea

Objectives

We have cloned 10 peroxidase (POD) cDNAs from suspension cultures of sweetpotato (*Ipomoea batatas*). Among them, *swpa4* was most highly induced by both abiotic and biotic stress, suggesting that this gene was regulated by a multiple stress-inducible promoter. In the previous study, *SWPA4* promoter was characterized in transgenic tobacco plants and cultured cells in terms of environmental stresses. In this study, we characterized *SWPA4* promoter in terms of its *cis*-elements by transient assay using tobacco BY-2 protoplasts.

Materials and Methods

1. Materials: Sweetpotato (*Ipomoea batata* L. Lam. cv. White Star), tobacco BY-2 suspension cells
2. Methods: Analysis of promoter activity by fluorometric GUS assay, transient assay using BY-2 protoplast

Results and Discussion

A *SWPA4* genomic clone consisted of 2,433 bp of 5'-upstream sequence from the translation start site, two introns (115 bp and 168 bp) and a 1,229 bp coding region. Sequence analysis reveals that *SWPA4* promoter contains putative binding sites for several stress related transcription factors including AP1, HSE, CARE, W-box and ERE. Interestingly, -433 bp promoter is sufficient to confer high expression upon stress treatment. Analysis of further 5' deletion mutants of -433 bp promoter showed that the removal of the 60 bp region between -237 and -177 bp reduced GUS activity to the level of CaMV 35S promoter and that the deletion of 5'-untranslated region (59 bp) caused a loss of GUS activity. The 5'-untranslated region was shown to enhance transient expression by placing it between the 35S minimal promoter and GUS reporter gene. The results suggest that the region downstream of -237 bp is sufficient and essential sequence for stress-inducible activity of the *SWPA4* promoter.

*Corresponding author : Sang-Soo Kwak, Tel : 042-860-4432, E-mail : sskwak@kribb.re.kr