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Molecular Characterization of Transgenic “Osome” and “Seoul Baechu” (*Brassica rapa*) using a Bromelain Gene Coding Cysteine Protease

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Objectives

We are interested in the roles of protease in plants. Particularly with respect to the protection of crops from fungal, bacterial and virus attack. We have tried to make transformation of “Osome” and “Seoul Baechu” (*Brassica rapa*) with the bromelain gene coding cysteine protease from pineapple and investigated gene expression and genetic analysis in T0 & T1 generations.

Materials and Methods

1. Material : “Osome”, “Seoul Baechu” (*Brassica rapa*)
2. Methods : RT-PCR cloning, Sequence, Vector construction , PCR analysis, Southern blot and Northern blot in T0 & T1 generations

Results and Discussion

The recombinant DNA using gene coding cysteine protease from pineapple have constructed with the 35S promoter, to be transformed into “Osome”, “Seoul Baechu” (*Brassica rapa*) were co-cultivated with *Agrobacterium tumefaciens* LBA4404 strains containing, npt II gene and bromelain gene coding cysteine protease from pineapple transformation. Through initial selection of regenerated explants by culturing on a hygromycin containing MS medium, multiple shoots were obtained after 2 months of culture. For a complementary step of selection, putative transgenic shoots were obtained to 1/2 MS basal medium supplemented with 500mg/L hygromycin. The selected shoots were confirmed by PCR and Southern blot analysis. Northern blot showed that transcripts of bromelain gene were detected in the various tissues of transgenic T0&T1 generations.