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In situ Transient expression Assay of The Modified *E. coli* β -Glucuronidase

문은표
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The β-glucuronidase (GUS) gene from *E. coli* has been used as a reporter for gene fusion studies in various organisms. Especially in plants, it has been extensively used to examine the promoter strength and the differential expression of many genes. However, it can't be used as a reporter for secretion and targeting of proteins since it loses its activity by the glycosylation in the ER. To overcome this problem, I have destroyed the glycosylation site in the GUS gene by changing an amino acid using site-directed mutagenesis. Three different amino acids were substituted for the asparagine residue at the glycosylation site. *In situ* transient expression assay revealed that the GUS substituted with serine retains the activivy even when sent to ER.

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Analysis on Genes Related to Antituberculosis Drugs by Single Strand Conformation Polymorphism(SSCP)

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Many genes encoding targets of antituberculosis drug or related to drug resistance were elucidated at recent. There are several reports to detect antituberculosis drug resistant strain using SSCP which permits discrimination of point mutation. Some genes, *rpoB, katG* and *inhA* related to susceptibility to rifampicin(RMP), isoniazid(INH) and ethionamide(ETH) respectively were screened by SSCP using 15% acrylamide gel and silver staining. RMP resistant strains were also screened by Line Probe Assay(LiPA). Only 16(44.4%) out of 36 RMP resistant strains(MIC ≥ 40 μg/㎖ in the Lowenstein Jensen media) could be discriminated by SSCP, while 34 strains by LiPA. As codon 463 mutation of *katG* gene was expected as a maker of INH resistant, we screened this mutation in INH sensitive strains. Mutation was found at this codon in 21(75%) out of 28 INH sensitive strains(MIC ≥ 0.2 μg/㎖). We also screened ETH resistant strains(MIC ≥ 20 μg/㎖) and found new mutatation site at codon 21 in addition to known site of codon 94 in the *inhA* gene.