E323 Isolation, Characterization and Overexpression of the *Candida* Gene, CGR1, Encoding Glutathione Reductase.

Yong-Un Baek*, Yeon-Ran Kim and Sa-Ouk Kang
Laboratory of Biophysics, Department of Microbiology, College of Natural Sciences, and Research Center for Molecular Microbiology, Seoul National University

Using degenerate oligodeoxyribonucleotides based on the highly conserved amino acid region of known glutathione reductase(GR) amino acid sequences, the *Candida* gene encoding glutathione reductase(GR), CGR1, was isolated using PCR followed by screening of a *Candida* genomic DNA library. Sequence analysis of the cloned DNA revealed that the enzyme was consisted of 461 amino acid residues and was homologous to the enzymes of *Saccharomyces cerevisiae*, *Schizosaccharomyces pombe*, sharing similarities to the pyridine nucleotide-disulfide oxidoreductase family of flavoenzymes. *Candida* cells transformed with a multicopy plasmid pRC2312 containing the genomic clone overproduced GR activity twofold. It showed the yeast-to-mycelial transition during overnight starvation without inducer, which implicate the potential involvement of GR in the regulation of the dimorphism in *Candida albicans*.

E324 Expression and Purification of Recombinant Toxin Shock Syndrome Toxin I

Choi Young Sil * · *Lee Chong Sam*
Dept. of Biology, College of Natural Sciences, Sungshin Women's Univ.

Toxic-shock-syndrome toxin I (TSSTI), an exotoxin produced by *staphyloccous aureus*, has been closely associated with the pathogenesis of toxic shock syndrome. We report here a strategy for the high level expression and the simplified purification of TSST I. It has subcloned the coding region of TSST I into a pRSET-B vector contained an inducible T7 promoter sequence and TSST protein in host strain (*Escherichia coli* JM109) were expressed. The recombinant TSST I protein contained 6 squential histidine residues(Histag) at its N-terminals was isolated and purified by using the nickel-agarose-affinity resin effectively. Histag-TSST I(H-TSST I)was further purified by utilizing a Ni-NTA column. Through this course 80~120μg of highly purified H-TSST I can be consistently obtained per 50mℓ of culture. It was confirmed by RPLA-SET assay whether H-TSST I was given the effect by Histag or the presence of biological activity.