F311 The structure-function study of the *YDR1* gene in the yeast *Saccharomuyces cerevisiae*

Hyun-Sook Yoo*, Hyung-Ryun Kim and Jong Gil Na
Department of Biology, Kunsan National University

The *YDR1* gene in yeast is the counterpart of hDr1, a transcriptional repressor. The *YDR1* is an essential gene and its overexpression is deleterious to the cells. hDr1 can replace yDr1 in yeast. The *ydr1* alleles were created by *in vitro* mutagenesisand replaced plasmid-borne *YDR1* gene in a plasmid shuttle strain YMH196 where the genomic *YDR1* was knocked out. DNA sequence analysis revealed the nature of the mutation. Four *ydr1* alleles were FOA-sensitive, suggesting that the Ydr1 in the allele is not functional. Four *ydr1* alleles conferred pleiotropic phenotypes. Three of four FOA-sensitive strains resulted from nonsense mutations which are R34 (*ydr1-4*), S50 (*ydr1-5*), and R118 (*ydr1-2*). The *ydr1-7* allele (L48P) showed cold-sensitive phenotype (unable to grow at 16℃) and The three *ydr1* alleles conferred temperature sensitive phenotype at 37℃ (E35K, *ydr1-3* ; S40F, *ydr1-1* ; E67L, *ydr1-6* ; Q96L, *ydr1-8*). The five base substitutions positioned within the histone-fold motif in the *YDR1* gene, indicating that the histone-fold motif is important for the Ydr1 function. The *ydr1* alleles obtained in this project is being used to generate revertants to uncover suppressor genes involved in transcription. These results will help us understand the regulation of eukaryotic transcription.

F312 The Nickel Resistance Determinant Cloned from the Enterobacterium *Klebsiella oxytoca* : Expression, Regulation and Physical Map

Min Won Kang*, Hans Günter Schlegel[1], Ho Sa Lee
Department of Biology, Kyung Hee University
[1]Institut für Mikrobiologie, Universität Göttingen, Federal Republic of Germany

Klebsiella oxytoca strain CCUG 15788, isolated from a mineral oil emulsion tank in Göteborg, Sweden, was found to be nickel resistant. Nickel resistance is an inducible property in *K. oxytoca*. A 4.3 kb *Hin*dIII fragment was cloned from genomic DNA of *K. oxytoca*. With 4.3 kb *Hin*dIII fragment as a biotinylated DNA probe it was shown by DNA-DNA hybridization that the nickel resistance determinant residues on the chromosome of *K. oxytoca* and not on its circular plasmid pKO1 or linear plasmid pKO2. No homologies were detected when the nickel resistance determinants of other well known nickel resistant bacteria, such as *A. eutrophus* CH34 or *A. denitrificans* 4a-2, were used as target DNA. *K. oxytoca* strain 58 is E. coli JM109 pBluescript :: KoH2.29(-14). For further study, we described the physical map and to determine the DNA sequence of nickel resistance determinant, we constructed various subclones.