

F029

### Studies of Biological Function of DksA in *Streptomyces coelicolor* M1600.

Eun Sook Kim\* and Kye Joon Lee

School of Biological Sciences, Seoul National University

The *E. coli* *dksA* (*DnaK* suppressor A) gene was first isolated as a multicopy suppressor of the temperature sensitivity of *dnaK* mutants. Deletion and overexpression of *dksA* have pleiotropic effects, including defects in chaperonin function, gene expression, cell division, amino acid biosynthesis, quorum sensing, and virulence in various bacteria. Since then, DksA is identified to amplify function of ppGpp by binding RNAP recently.

In our lab, we found DksA homologue from *Streptomyces coelicolor*. The C-terminus of *S. coelicolor* DksA showed 28.8% identity in comparison with *E. coli* DksA. *E. coli* DksA has a coiled coil with highly conserved Asp residues. *S. coelicolor* DksA also has this conserved Asp residues. We began to study about role of *S. coelicolor* DksA through analyzing structure of *S. coelicolor* DksA. In company with this research, we are constructing *S. coelicolor*  $\Delta$ *dksA* by PCR targeting and making anti-DksA to investigate physiological function of DksA *in vivo*.

F031

### Study of *Streptomyces coelicolor* Extracellular Proteases and Protease Inhibitors Involved in Differentiation and Antibiotics Production

Dae Wi Kim\*, Ki Chan Yoon, and Kye Joon Lee

School of Biological Science, Seoul National University

*Streptomyces coelicolor* is soil bacteria producing diverse secondary metabolites. In nature, diverse organic component consists of insoluble polymers accessible only to organisms having suitable extracellular enzymes and systems for their regulation. In *S. coelicolor* genome, more than 60 extracellular proteases were reported. In this study, diverse extracellular proteases were chosen and disrupted by PCR-targeting methods. Morphological differentiation and fermentation profiles of each mutant strain were analyzed. No remarkable changes were observed except for SCO5913 (trypsin) defected strain. This strain showed *whi* phenotype in MS media and also showed high trypsin inhibitor activity. STI (Trypsin inhibitor, SCO0762) is *bltA*-AdpA dependent protein. STI defected strain was constructed and phenotype, fermentation profile were analyzed. STI defected strain showed high antibiotics production. There are lots of proteases in *S. coelicolor*, so it is important to find key proteases, inhibitors involved in differentiation and antibiotics production. Characterized SCO5913 and STI might be key extracellular proteins involved in differentiation and secondary metabolites production.

F030

### The Effect of (p)ppGpp on Antibiotic Production in *RelA* and *Arsh* mutants of *Streptomyces coelicolor* M1600

Yong-Ku Ryu\* and Kye Joon Lee

School of Biological Sciences, Seoul National University

In *Escherichia coli*, ppGpp is made from ATP and GTP by RelA (a ribosomal-bound ppGpp synthetase) and SpoT. RelA synthesizes ppGpp when a non-aminoacylated tRNA binds to the acceptor site of translating ribosome (for a review, see Cashel *et al.*, 1996). ppGpp is also synthesized by SpoT when phosphate is depleted on culture media, but the amount of ppGpp is very small. The main role of SpoT was limited to hydrolyzing ppGpp. In *Streptomyces coelicolor* M1600, there are two genes, RelA and Rsh (*relA*/*spoT* homologue). To know function of *relA* and *rsh* in M1600, the disruption of *relA* and *rsh* was done by PCR-targeting and confirmed by PCR and Southern Hybridization. Carbon, phosphate, and amino acid-limited culture were carried out and the production of ppGpp was measured using HPLC. Batch culture of M1600 was carried out and the concentration of glucose, DCW, NH<sub>3</sub>, phosphate, actinorhodin (Act), undecylprodigiosin

F032

### Molecular Epidemiology of Endogenous Retroviruses in Broiler Eggs in Korea

You-Jung Kim

Veterinary Medical Research Center, College of Veterinary Medicine, Chonnam National University

Endogenous retroviruses (ERV) have been found in all eukaryotes. These proviruses are supposed to result from germline retrovirus infection. In chicken, ERVs including EAV, ART-CH, ev, and ev/J have been reported. To evaluate endogenous retrovirus infection in meat type chicken in Korea, sequencing analysis of nucleotide and reduced amino acid of env gene for ev and ev/J, a part of transmembrane and long terminal region for EAV, and gag-related region for ART-CH was performed with embryonated eggs from three grand parent breeders by amplified products of PCR specific for each ERV. By PCR, all kinds of ERV were amplified in all breeders examined. Phylogenetic analysis of reduced amino acid revealed Korean ev ERV was grouped together but distinct from the other known ERV. Phylogenetically, however, nucleotide substitution of Korean EAV, ART-CH and ev/J was scattered within the other known each ERV, respectively. From these results, Korean major broiler breeders were infected with every known ERV. In addition, Korean ev ERV might have different evolutionary pathway.