D124

CeST-BP, a Single Stranded Telomere Binding Protein in the Nematode *C. elegans*

Su Young Yi* and Junho Lee
Department of Biology, Yonsei University, Seoul

Telomeres are multifunctional elements that shield chromosome ends from degradation and end-to-end fusions, prevent activation of DNA damage checkpoints, and modulates the maintenance of telomeric DNA by telomerase. Telomeric DNA has been found to interact with proteins in many organisms. We wanted to identify and elucidate functions of telomere binding proteins in the nematode *C. elegans*. Telomeric repeat of *C. elegans* consists of a long stretch of (TTAGGC)n. We have identified a protein (CeST-BP) in *C. elegans* embryonic nuclear extract that specifically binds single-stranded telomere sequences by gel mobility shift assay. CeST-BP did not efficiently bind with repeated RNA sequences of UUAGGC, thus we concluded that CeST-BP bound specifically with DNA only. CeST-BP was sensitive to salt concentrations, and insensitive to RNase treatment. We found that CeST-BP needs at least 2 repeats of (TTAGGC) sequences for binding. More specifically, we found that the core nucleotides of 5′ GCTTAGG 3′ within the $(TTAGGC)_2$ were essential for binding and that two non-specific nucleotides should reside in front of the core binding site for efficient binding. The size of CeST-BP was found to be about 40 KD in South-Western hybridization. We plan to identify CeST-BP by purification using affinity chromatography and MALDI-TOF protein sequence analysis. Subsequently, studies of the functions of CeST-BP in vivo will follow.

D125

Characterization of the Regulatory Elements Required for Neuron-specific Expression of SNAP-25 in the Nematode

Soon Baek Hwang* and Junho Lee
Dept. Biology, Yonsei University, Seoul

SNAP-25 is a presynaptic protein involved in synaptic vesicle docking and fusion. We previously showed that SNAP-25 is exclusively expressed in the nervous system of the nematode. We wanted to characterize the regulatory elements that are required for this tissue-specific expression. We determined the whole sequence of the 5′ upstream region including the relatively large first intron from the YAC Y22F5 sequence which covers the cosmid C29F1 which was previously shown to contain the full length of the SNAP-25 gene. We screened a genomic library of *C. briggsae* by southern hybridization and obtained a fosmid clone named G01P23 that contained the full genomic sequence of the *C. briggsae* homolog of SNAP-25. We also obtained the entire sequence of the fosmid from the genome center. As transformation of G01P23 into the *C. elegans* SNAP-25 mutants complements the mutant phenotypes, we concluded that specific transcription factors of C. elegans may be able to bind the cis-acting elements in the *C. briggsae* SNAP-25 gene, and that the cis-acting elements may be conserved in these two species. By both examining serially-deleted 5′ upstream regions and the first intron of SNAP-25 and examining the sequences conserved both in *C. elegans* and *C. briggsae* SNAP-25 genes, we were able to identify the regulatory elements required for neuron-specific expression of SNAP-25. We defined two sequence motifs in the promoter region and two cis-acting regulatory motifs in the first intron. The -1076~-1064 region of 5′ upstream was required in motor neurons of the body region and the -207~-197 region was required in amphid and phasmid

neurons. A 200 bp patch of the first intron may act in pharyngeal neurons and another 300 bp patch may act in mechanosensory neurons. We expect that each motif confers binding site for transcription activator(s) in different subsets of neuron cells, which may differ in cell lineage and function.

D126

Interaction of Rbm, a Male Infertility Protein, with hnRNP K Suggests Its Function in mRNA Processing during Spermatogenesis.

Euisu Kim[1*], Sung Key Jang[2], Kyungjin Kim[1] and Kunsoo Rhee[1]

School of Biological Sciences, Seoul National University, Seoul 151-742[1] and Department of Life Science, Pohang University of Science and Technology, Pohang, Kyungbuk 790-784[2]

Microdeletions in the *AZFb* are strongly associated with male infertility. Multiple copies of the *Rbm* genes are located in the *AZFb* region on the Y chromosome. *Rbm* expression is restricted to the testis, especially the nuclei of the male germ cells. To understand biological functions of Rbm, we tried to identify proteins that interact with the Rbm protein. When we carried out yeast two-hybrid screening with a Rbm protein as a bait, we were able to observe a positive interaction with hnRNP K. Interacting domains of Rbm and hnRNP K were defined in yeast using several truncated mutants of both genes. Since the hnRNP K protein has multi-functions as a docking platform for controlling gene expression at the post-transcriptional level, we propose that Rbm plays an important role in spermatogenesis by controlling gene expression at the post-transcriptional level.

D127

Subcellular Localization of Nek2 Suggests Its Dual Functions as a Cell Cycle Regulator

Jun Youl Choi*, Yongha Kim, Yeon-Tae Jeong and Kunsoo Rhee

Seoul National University, School of Biological Sciences, Seoul 151-742

Nek2 is a mammalian protein kinase, structurally homologous to *Aspergillus* NIMA. Since NIMA functions as a cell cycle regulator by controlling chromatin condensation, it was proposed that Nek2 may also play a similar role as a cell cycle regulator in association with chromatin. On the contrary, it was reported that Nek2 can associate with centrosome, playing a key role in centrosomal behaviors during mitosis. To define biological functions of Nek2, we determined expression pattern and subcellualr localization of endogenous Nek2 in the ovarian follicular cells as well as of the exogenous Nek2 mutants in NIH3T3 cells. The results revealed that Nek2 was expressed in a cell cycle-specific manner, in that the Nek2 protein was present abundantly in S/G2 phase of the cell cycle. The cell-cycle dependence of Nek2 expression may be regulated, at least in part, with proteolytic mechanisms. The Nek2 protein was localized both in the nucleus and in the cytoplasm. In the nucleus, Nek2 appeared to associate with mitotic chromatin. These results suggest that Nek2 may have dual functions related to the chromatin condensation and the centrosome cycle.

D128

Postnatal Changes of the Steroidogenic Acute Regulatory Protein mRNA Expression in the Rat Brain

김현준, 박창환, 강상수*, 최완성

Department of Anatomy, College of Medicine,