F001

ATP-binding Motifs Play Key Roles in Krp1p, Kinesin-related Protein 1, Function for Bi-polar Growth Control in Fission Yeast.

DongKeun Rhee[1*], BonA Cho[1], and HyongBai Kim[2]
[1]*School of Life Sciences and Biotechnology, Korea University,* [2]*Department of Bioinformatics, Korea University*

Kinesin is a microtubule-based motor protein with various functions related to the cell growth and division. It has been reported that Krp1p, kinesin-related protein 1, which belongs to the kinesin heavy chain superfamily, localizes on microtubules and may play an important role in cytokinesis However, the function of Krp1p has not been fully elucidated In this study, we overexpressed an intact form and three different mutant forms of Krp1p in fission yeast constructed by site-directed mutagenesis in two ATP-binding motifs As a functional consequence, a point mutation of ATP-binding domain 1 (G89E) in Krp1p reversed the effect of Krp1p overexpression in fission yeast, whereas the specific mutation in ATP-binding domain 2 (G238E) resulted in the altered cell polarity In conclusion, these results suggest that krp1p is involved in regulation of cell-polarized growth through ATP-binding motifs in fission yeast.

F002

A Leucine-zipper Domain in Krp1p, Kinesin-related Protein1, Is Involved in Chromosome Decondensation in Fission Yeast.

BonA Cho[1*], DongKeun Rhee[1], and HyongBai Kim[2]
[1]*School of Life Sciences and Biotechnology, Korea University,* [2]*Department of Bioinformatics, Korea University*

Kinesin is required for spatial organization of microtubules and chromosomes in the mitotic spindle, microtubule stabilization, and chromosome-microtubule interactions Moreover, the leucine zipper-like motif in nuclear kinesins is known to be involved in localization on the spindle pole of kinesin and DNA-binding In this study, we overexpressed an intact form and a mutant form of Krp1p in fission yeast constructed by truncation of the leucine zipper-like motif (LZiP). We observed hyper-extended microtubules and the aberrant nuclear shape in Krp1p-overexpressed fission yeast The truncation of the leucine zipper-like domain (LZiP) at the C-terminal of Krp1p showed a normal nuclear division, so that cell division was completed, though there was a little delay. These suggest that Krp1p is involved in microtubule polymerization in cooperation with other microtubule regulatory proteins and the LZiP motif of Krp1p might be involved in cell-division mechanism.

F003

Characterization of the Activity of Human Cytomegalovirus IE1 Protein to Desumoylate PML In Vitro

Heejung Kang*, Eun-Tae Kim, Hye-Ra Lee, and Jin-Hyun Ahn
Department of Molecular Cell Biology, Sungkyunkwan University School of Medicine

In human cytomegalovirus infection, the immediate-early IE1 protein disrupts the subnuclear structures known as PML oncogenic domains by causing loss of sumoylation of PML, and this is believed to be required for efficient viral growth at low multiplicity of infection. We studied how IE1 ablates sumoylation of PML in vitro In sumoylation reactions, either in vitro translated PML or bacterially purified GST-IE1 was sumoylated When PML was sumoylated first and IE1 was added later, IE1 was not sumoylated but the level of PML sumoylation was reduced by IE1, suggesting that IE1 is able to cause desumoylation of PML without competing for SUMO-1 To improve the assay system, GST-IE1 was prepared from insect cells and cell lysate-free sumoylated forms of PML were prepared from E.coli When sumoylated PML produced in E.coli was used, GST-IE1 (from either bacteria or insect cells) did not desumoylate PML whereas GST-SuPr-1, a SUMO-specific protease, efficiently did so Thus, the results suggest that IE1 does not have the intrinsic desumoylation activity against PML in vitro but may cooperate with cellular components to induce desumoylation of PML in vivo

F004

The *Aspergillus nidulans silG* Gene Functions in Repression of Sexual Development in Response to Light

Hyo-Jeong Kim[1*], Kap-Hoon Han[2], and Dong-Min Han[3]
[1]*Institute of Basic Natural Science, Wonkwang University,* [2]*Bio-Med Research Center, Pai Chai University,* [3]*Division Life Science, Wonkwang University*

The *silG* gene presented in here was identified as a multi-copy suppressor of one of the *snd* mutations(suppressors of nsdD). Multi-copy of *silG* blocked cleistothecia development in an SND mutant The *silG* gene is predicted to encode a 703 aa polypeptide with three C2H2 zinc finger DNA-binding domains at the C-terminus The *silG* null mutant produced a high number of cleistothecia even under the visible light, which normally inhibits sexual development However, high osmolarity or poor carbon sources blocked sexual development of the *silG* deletion mutant, suggesting that SilG may play a specific role in negative regulation of sexual development in response to light Further supporting this hypothesis, over-expression of *silG* resulted in a great reduction of sexual development in dark, which preferentially enhance sexual development in wild type Accumulation of *silG* mRNA undulated throughout the lifecycle in a certain recurring pattern Interestingly, *silG* mRNA levels sharply elevated upon exposure to light and the response requires the functional *A nidulans veA* gene.

[Supported by grants from KRF]