Ishii Research Collaborative Group
Molecular Genetics Laboratory

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Goal

The transcriptional control is a key step for development, stress response, and various diseases of human being. Recent progress in the study of the mechanism of cellular proliferation has shown that signals for proliferation or differentiation are transferred into nuclei through receptors and other mediators. However, the mechanism of signal transduction remains elusive. Especially, it is unknown how transcription is regulated via change of nuclear architecture and chromatin structure. Through the use of molecular biology and biochemistry techniques and, in some cases, employing whole animal systems, we are investigating the mechanism of transcriptional control.

Activities

1. Nuclear oncogene product as a transcriptional regulator
2. Gene expression extracellular stimuli
3. Genetic study of transcription using Drosophila

Members

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Shunsuke ISHII, Ph. D. (1982. 4 ∽)

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Specific aim

1) Nuclear oncogene product as a transcriptional regulator

We have demonstrated that c-Myb activity is negatively regulated by corepressors including Ski. Removal or mutation of the negative regulatory domain (NRD) in the C-terminal half of c-Myb leads to increased trans-activation and oncogenic activation. We found that corepressor TIF1β directly binds to the NRD and negatively regulates c-Myb-dependent trans-activation. In addition, three corepressors (Ski, N-CoR, and mSin3A) bind to the DNA-binding domain of c-Myb together with TIF1β and recruit the HDAC complex to c-Myb. Ski competes with CBP for binding to c-Myb, indicating that the selection of coactivators and corepressors is a key event in c-Myb-dependent transcription. Mutations or deletion of the NRD of c-Myb and mutations found in the DNA-binding domain of v-Myb decrease its interaction with these corepressors and weaken the corepressor-induced negative regulation of Myb activity. These observations have conceptual implications for understanding how the nuclear oncogene is activated.

2) Gene expression regulation by extracellular stimuli

The biologically relevant molecular mechanisms that regulate c-Myb activity remain unclear. We have shown that c-Myb is phosphorylated and degraded by Wnt-1 signal via the pathway involving TAK1 (TGF-beta-activated kinase), HIPK2 (homeodomain-interacting protein kinase 2), and NLK (Nemo-like kinase). Wnt-1 signal causes the nuclear entry of TAK1, which then activates HIPK2 and the mitogen-activated protein (MAP) kinase-like kinase NLK. NLK binds directly to c-Myb together with HIPK2, which results in the phosphorylation of c-Myb at multiple sites, followed by its ubiquitination and proteasome-dependent degradation. The down-regulation of Myb by Wnt-1 signal may play an important role in a variety of developmental steps.

3) Genetic study of transcription factor using Drosophila

We have analyzed the role of Drosophila Myb (dMyb) for cell cycle regulation in eye imaginal disc. Our results indicate that dMyb is involved in G2/M progression by inducing the transcription of Cyclin B gene. We observed that over-expression of activated form of dMyb in eye imaginal disc, which lacks the C-terminal region, leads to rough eye phenotype. This system will be useful to identify the modulator of dMyb.

To search for the modulators that regulate the transcriptional regulation by the myb nuclear oncogene product (Myb), we have established the genetic screening system using Drosophila. Over expression of the C-truncated Drosophila Myb (dMyb) in the eye imaginal disc induced the rough eye phenotype. By introducing various mutations, we have observed the rough eye phenotype can be suppressed but in some cases enhanced. By using this change of rough eye phenotype, we have screened the Myb modifiers. We have conformed that Drosophila has more than 10 dMyb modifiers.

Bench for Drosophila experiments

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Publications

Original Papers (*Peer reviewed Journal)

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