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The Polycomb Group Protein Pcl2 Interacts with YY1, and Both Proteins Regulate Neural Tissue Development in *Xenopus* Embryos

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Polycomb group (PcG) proteins are involved in the heritable stable expression of a hox genes. A novel gene, *Xenopus Polycomblike 2 (XPcl2)*, encodes a protein similar to *Drosophila Polycomblike*. *XPcl2* mRNA is present both maternally and zygotically. *XPcl2* is highly expressed in the anterior-dorsal region of *Xenopus* following the neurula stage. Overexpression of *XPcl2* disturbs the development of the anterior. Here we show that Yin Yang 1 (YY1), a homolog of the *Droshophila* PcG protein Pleiohomeotic (Pho), interacts with the *Xenopus* PcG protein XPcl2. As a multifunctional transcriptional factor YY1 is reported that acts as an activator, repressor, or inhibitor of transcription of numerous cellular and viral genes and controls expression of developmentally regulated genes. *Xenopus*YY1 (XYY1) protein was localized in the central nervous system (CNS), particularly anterior neural tube of tailbud stage embryos. To study the interaction of both XPcl2 and XYY1, co-injection was perfomed using XPcl2 antisense phosphorothioate oligonucleotide (s-oligo) and XYY1 antisense s-oligo. And examined the expression of target genes including neural marker genes. We will present the interaction of the XPcl2 and XYY1 in the development of anterior neural tissue.

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Identification and Characterization of Inorganic Pyrophosphatase in *C. elegans*

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Hydrolysis of inorganic pyrophosphate is catalyzed by an enzyme inorganic pyrophosphatase (PPase). It has been proposed that catalysis of PPase thermodynamically driving force for a number of important biosynthetic reactions. The enzyme is also shown to be essential for viability of yeast and bacteria. It has been shown that the level of PPase is significantly altered in samples of gastric cancer patients, which was identified by two-dimensional PAGE at the Gyung Sang proteome research center. It is also reported that PPase is increased in adenocarcinomas. Therefore, in order to understandthe association of this PPase with gastric cancer we characterized the homologue of PPase gene in *C. elegans*.The *C. elegans* genome database revealed the presence of a predicted PPase gene (we termed *ppa-1*) which is located on cosmid C47E12.4 (LG IV). It shows about 53% amino acid identity with human PPase. The *ppa-1*is strongly expressed in the intestine and hypodermal cells. Recombinantly expressed enzyme exhibits specific activity of catalyzing hydrolysis of PPi to Pi *in vitro*. A deletion mutant of *ppa-1*was isolated by TMP/UV mutagenesis. The deletion mutant revealed developmental arrest phenotype at L2 stage, which suggests that PPase is essential for larval development in *C.elegans*.

E111

Role of Munc18-1 in Glutamate Release from the Pubertal Rat Hypothalamus

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Munc18-1, a mammalian homologue of the unc18 gene, has been identified as an essential component of the synaptic vesicle fusion protein complex. Onset of female puberty follows a series of prepubertal cellular and molecular events including changes of synaptic plasticity, synthetic and releasing activity and gene expression. In the present study, we used RNase protection assay to determine the expression of Munc18-1 mRNA in the female rat hypothalamus during juvenile and the pubertal process. Female rats were assigned -- based on uterine weights, the presence and abundance of uterine fluid, and their vaginal patency -- to one of the following: anestrus, early proestrus, late proestrus, estrus, and diestrus. Munc18-1 mRNA level significantly increases at the anestrus of puberty when is the first phase of female puberty onset. To determine the effect of estrogen onthe change in Munc18-1 gene expression, we measured hypothalamic Munc18-1 mRNA levels in the ovariectomized and estrogen-treated rats. Estrogen induced a significant increase in Munc18-1 mRNA level in the medial basal hypothalamus (MBH). Furthermore, Munc18-1 synthesis inhibition by treatment of antisense oligomer significantly decreased glutamate release from incubated MBH fragments. The present results suggest that Munc18-1 plays an essential role in the regulation glutamate release under estrogen feedback environment.

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RCN-1(Regulator of Calcineurin 1) Inhibits Mutiple Function of Calcineurin

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Calcipressins area conserved family of calcineurin binding proteins that inhibit the Ca2+/calmodulin-dependent protein phophatase calcineurin. They have shown to play a role in the negative feedback regulation of calcineurin and have been implicated in several diseases such as Down syndrome, Alzheimer's disease, and cardiac hypertrophy. In this study, we have characterized *rcn-1* (regulator of calcineurin 1), *C. elegans* homologue of Down Syndrome Critical Region 1 (DSCR-1). *rcn-1*is expressed in hypodermal cells, nerve cords and various neurons, vulva epithelial and muscle cells, marginal cells of the pharynx, and structures of the male tail. RCN-1 specifically binds to calcineurin A from *C. elegans* lysate in a calcium-dependent manner, and effectively inhibits bovine calcineurin phosphatase activity dose-dependently confirming its conserved role asa calcipressin. Overexpression of RCN-1 in N2 caused calcineurin-deficient phenotypes such as small body size, cuticle defects, fertility defects, slow growth, and serotonin-resistant egg-laying defects. Where as overexpression of RCN-1 in calcineurin gain-of-function mutant restored normal phenotypes. These results demonstrate an effective and specific inhibition of calcineurin in vitro as well as in vivo by RCN-1.