

## D111

### Effect of Estrogen on the Gestation Profiles in Gene Expression of Placental Lactogen I, II and Pit-1 in the Rat Placenta

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To investigate gestation profiles in gene expression of placental lactogen I (PLI), PLII and Pit-1, RNA was extracted from 12-day to 20-day placenta at 2 day intervals. Northern blots showed changes in gene expression of PLI, II and Pit-1. Messenger RNA size of PLI and II was changed during gestation. The amounts of mRNA of PLI, II and Pit-1 were increased along with gestation progress. To examine the effect of estrogen on the gene expression of PLI, II and Pit-1, pregnant female rats were ovariectomized (OVX) and daily injected with  $17\beta$ -estradiol (OVX + E). Ovariectomy markedly lowered the amount of PLI and II mRNA and shifted mRNA size from 1 kb to 1.3 kb and to 0.6 kb, respectively, but had no effect on the mRNA size of Pit-1. Estrogen injection reversed the effect of ovariectomy on the shift of mRNA size but not on the amount of mRNA. The size of major Pit-1 mRNA was shifted from 2.6 kb to 1.2 kb by OVX + E. Present results suggest that estrogen may play a pivotal role on the gene expression of PLI and II such as alternative RNA splicing, and Pit-1 may be involved in the gene expression of PLI and II by estrogen. (HRC-95-0104)

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### Metalloproteinases Activities during Urodele Limb Regeneration

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Degradation and remodeling of extracellular matrix(ECM) are key events in the patterning process of many developmental system. Matrix metalloproteinases(MMPs), a group of matrix degrading enzymes, are known to degrade various ECM components such as collagen, laminin, and fibronectin in human fibroblast and mouse blastocyst. In amphibian limb regenerate, MMP-9, trypsin-like protease, and chymotrypsin-like protease were found to be highly expressed during dedifferentiation period and retinoic acid(RA), an inducer of limb pattern duplication was found to evoke enhanced and prolonged dedifferentiation state. In the present study, we were interested to know changes of matrix degrading enzyme activities during urodele limb regeneration. For this purpose, we performed zymographic study using SDS-polyacrylamide gel impregnated with gelatin. In the normal regenerate of axolotl, *Ambystoma mexicanum*, three types of gelatinases(92, 86, 80KDa) were observed. Among those, 92KDa gelatinase activity was very high until 12 days after amputation, while 80KDa gelatinase activity appeared at 10 days after amputation. In the normal regenerate of Korean salamander, *Hynobius leechii*, three types of gelatinases(92, 86, 80KDa) were observed as in the case of axolotl. In this case, 92KDa gelatinase activity increased until 4 days after amputation and gradually decreased, while 80KDa gelatinase activity appeared at 4 days after amputation. Since EDTA and 1,10-phenanthroline, specific inhibitors of MMPs, caused the inhibition of these enzyme activities, 92, 86, 80KDa gelatinases are believed to be members of MMPs. With RA treatment, increased level of 92 and 80KDa gelatinase activities(10 days after amputation in axolotl, 4 days after amputation in Korean salamander) were maintained longer compared to control. These results indicate that MMPs are mediators of dedifferentiation process which is a crucial step in the regeneration process of amphibian limb.