**D109** Identification of Homeobox Family Gene, Nkx-2.1, Messenger RNA in the Rat Brain Regions Related to Optic Input

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Nkx-2.1 is a DNA binding protein of mammalian NK family homeobox gene and transcription factor for thyroglobulin and lung surfactant protein gene expression. Nkx-2.1 is also involved in the fetal diencephalon formation. The present study aimed at identification of Nkx-2.1 gene expression in the rat optic system and pineal gland. Four-week old male rats were housed under a normal light and dark cycle or continuous light condition for 2 weeks and sacrificed at 1200h (light group) and 2400h (dark group). RNase protection assay showed the Nkx-2.1 mRNA in the rat optic track, pineal gland and retina. Nkx-2.1-like protein was determined by electrophoretic mobility shift assay using double strand oligomer probe containing a putative Nkx-2.1 binding domain. In situ hybridization histochemistry localized Nkx-2.1 mRNA signals in the optic chiasma, suprachiasmatic nucleus and supraoptic nucleus. Nkx-2.1 mRNA signal at night was stronger as compared to day time. Continuous light exposure clearly decreased Nkx-2.1 mRNA signals. Taken together, these data suggest that Nkx-2.1 play a regulatory role of rat optic system during day-night cycle.

**D110** Expression of Nel Gene in the Rat Hypothalamus during Prepubertal and Pubertal Development as Determined by Differential Display PCR

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Using estrogen sterilized rat (ESR) model and differential display PCR (ddPCR), we have identified more than 50 genes whose expression was changed in the rat hypothalamus in response to neonatal treatment of estrogen. One of differentially displayed genes was identified as a nel homologue. Nel was first identified in chick embryo as a brain specific new gene encoding a protein with EGF-like repeats and nel-like genes were cloned in mouse and human brain. We cloned 2.7 Kb nel cDNA from rat hypothalamic cDNA library using ddPCR product as probe and determined the base sequence. Rat nel homologue was 76% and 85% identical in cDNA and amino acid sequences with chick counterpart. Hypothalamic RNA was extracted from neonatal, 6 day-, 28 day-, 31 day- and 40-day old male and female rats and analyzed by Northern blot hybridization. One band of 3.7 Kb mRNA was detected in all stage hypothalami. Nel mRNA was markedly increased in the prepubertal 28 day-old female rat hypothalamus. This increase of mRNA was clearly reduced by neonatal treatment of estrogen. In the male rat hypothalamus, neither prepubertal increase of nel mRNA nor effect of neonatal treatment of estrogen was observed. These data suggests that nel play a role in the puberty initiation and sexual differentiation of hypothalamus.