287 NOCICEPTION: POSTNATAL DEVELOPMENT OF RESPONSIVENESS TO ANTI-CHOLINERGICS. C. T. Bennett and J. M. King. Neuropsychology Br., Experimental Medicine Div., Biomedical Laboratory, Chemical Systems Laboratory, APG, MD 21010.

Responses of rats (5, 10, 15, 20 and 25 days of age) to nociceptive stimuli were measured using the tail flick and hot plate tests. Following baseline measurements, the neonates received either saline (0.9%) or benactyzine (5.0 or 10 mg/kg) and tested 15 minutes later. In the hot plate test, 5 mg/kg benactyzine had no behavioral effect at any day tested. By 20 days of age, however, 10 mg/kg of benactyzine significantly reduced response latency in this procedure. In contrast to this is the effect of benactyzine on the tail flick behavior of the neonates. Beginning at 5 days of age, the low dose of benactyzine (5 mg/kg) significantly reduced tail flick latency. This effect was antagonized by physostigmine (0.3 mg/kg). The data clearly indicate a differential effect by dose, and by age, on the two tasks differing in motor complexity. These studies suggest that the functional development of the cholinergic system mediating nociception does not develop homogenously.

288 INTRACELLULAR LOCALIZATION OF ALPHA-FETOPROTEIN (αFP) IN THE DEVELOPING RAT BRAIN - AN IMMUNOCYTOCHEMICAL STUDY. Robert H. Bennett and Terence H. Williams. Dept. of Anatomy, University of Iowa, Iowa City, Iowa.

Alpha-fetoprotein (αFP) is a serum protein present temporarily in high concentrations in the serum of developing vertebrates, but absent in the adult. Molecules of αFP with specific estrogen binding properties exist in the developing rat brain and, on the basis of biochemical evidence, it has been hypothesized previously that αFP is present extracellularly in the developing brain. Whereas an appropriately timed action by estrogen on the developing rat brain appears to be both beneficial and necessary, it has been claimed that premature estrogenization may be deleterious, and that αFP serves to block this effect. The objective of this study is to localize αFP in developing brain to gain clues concerning its true role.

Thirteen, 15, 18 day fetal, 2 and 5 day postnatal and adult male and female Sprague-Dawley rats were perfused with Bouin's fixative and embedded in paraffin. Five micron sections were cut and processed for immunocytochemistry by the unlabeled antibody peroxidase-antiperoxidase technique.

Intracellular localization of αFP in the rat brain was noted in all animals aged from 15 days fetal to 5 days postnatal. Many recognizable neuronal cell groups contain αFP. These include some that have been shown to possess high affinity estrogen receptors. An interesting finding was discrete high intensity localization in some, but not all, of the cells lining the ventricles. Also, αFP was associated with meninges, choroid plexus, circumventricular organs and blood vessels. The localization of αFP in cells surrounding blood vessels in the brains of the 18 day fetal animals and the absence of similar staining in the 2 and 5 day postnatal animals suggested to us that maturation of the blood-brain barrier (BBB) prevents access of blood borne αFP into most brain areas. Persistent αFP localization observed in the circumventricular organs and choroid plexus up to postnatal day 5 can be explained by absence of the BBB in these areas. αFP was absent in the adult brain.

Two alternative hypotheses can be constructed to explain the observed immunocytochemical localization of αFP in developing rat brain:

(1) All neural cells may require αFP for some process associated with differentiation and/or migration.

(2) Only a particular population of cells, possibly estrogen sensitive cells, may require αFP.

Supported in part by an NSF predoctoral fellowship to R.H.B. and NIH grant NS11650 to T.H.W.

289 SOURCE OF NORADRENERGIC SPROUTING IN THE CEREBELLUM OF RATS TREATED NEONATALLY WITH 6-HYDROXYDOPAMINE. Ranbir K. Bhatnagar and Richard H. Schmidt*. Dept. of Pharmacol., Univ. of Iowa, Iowa City, IA 52242.

Treatment of neonatal rats with subcutaneous 6-hydroxydopamine (6-OHDA) is known to produce an extensive, permanent degeneration of noradrenergic (NE) fibers in the telencephalon, but results in a marked regenerative sprouting of NE terminals in the cerebellum. It is not known if the locus coeruleus is the source of all or any of these sprouted fibers, as other NE cell groups also appear to contribute to the cerebellar innervation. This study was conducted to determine the regional distribution of locus coeruleus innervation to the cerebellum in normal and 6-OHDA treated rats.

Female rat pups were injected subcutaneously with 100 mg/kg 6-OHDA or vehicle on postnatal days 1 and 2. When 100 days of age unilateral electrolytic lesions (2mA for 10 seconds) or sham implacements were made in the locus coeruleus. Two and three weeks later the cerebellum was removed and dissected into anterior, middle and posterior regions as described in the abstract by Schmidt and Bhatnagar. Each region was assayed for synaptosomal NE uptake, dopamine β-hydroxylase (DBH) and endogeneous NE.

Ipsilateral to the lesion in vehicle control rats NE levels in the parietal cortex were reduced to 15% of sham control, while on the contralateral side NE was reduced by less than 25%. This, as well as histology, indicates that the lesions were complete.

In the cerebella of sham-lesioned vehicle control rats the anterior region of cerebellum contained about 150% as much innervation as the other two regions. Neonatal 6-OHDA treatment caused increases in NE, DBH and NE uptake in all regions. The largest rease (60%) occurred in the posterior region while the smallest rease (20 to 30%) was found in the middle region.

In the vehicle control rats the lesion destroyed, in all cerebellar regions, 50 to 60% of the NE innervation of the ipsilateral side and 20 to 30% on the contralateral side. In rats treated natally with 6-OHDA the lesion destroyed 75 to 80% of the NE rvation on the ipsilateral side relative to sham-lesioned treated levels. No degeneration was detected on the side contralateral to the lesion. These data demonstrate that the locus coeruleus is the source of all 6-OHDA induced sprouting in the cerebellum and this sprouting is exclusively ipsilateral. The normal contralateral projection thus does not regenerate or following the initial neurotoxic effects of 6-OHDA.

rted by USPHS grant NS-12121)

290 DELAYED DISAPPEARANCE OF PLACING AFTER HEMISPHERECTOMY IN THE KITTEN. Joseph E. Bogen, Barry Campbell* and Morton Suzuki*, Rose-Loma Med. Grp., Los Angeles, CA 90026.

After sensorimotor corticectomy, frontal lobectomy or hemispherectomy there is an immediate and enduring loss of paw contact placing in the contralateral limb. A second ablation (of the remaining frontal lobe) results in immediate return of placing (Bogen & Campbell, Science 135:709-710, 1962) suggesting that the previous loss was due to unbalanced, tonic, ipsilateral inhibition. The cortical origin of this inhibition is indicated by subsequent experiments in which hemispherectomy of kittens 10-17 days old is followed by loss of contralateral paw contact placing only after a five week delay. During this delay, chin contact placing and visual placing appear, before subsiding gradually rather than abruptly as in the adult cat. This experiment exposes the ontogenesis of corticofugal influences on a behavior whose essential features appear to be organized subcortically.