193 EPIDERMAL GROWTH FACTOR (EGF): BINDING CHARACTERISTICS AND GROWTH RESPONSE OF NORMAL AND VIRALLY TRANSFORMED MOUSE FIBROBLASTS. Carmen Brutico and Roger Ladda, Penn State Univ Coll Med, MS Hershey Med Ctr, Dept Ped, Hershey, PA.

EGF, a polypeptide hormone isolated from male mouse submaxillary glands, is a potent stimulator of cell division and may largely replace serum for the induction of cell division. Since different serum fractions stimulate growth of normal Swiss 3T3 and transformed SV_{40}3T3 fibroblasts and suggest that each cell type may have its own peculiar responsiveness to specific growth promoting factors, we studied the response of 3T3 and SV_{40}3T3 to EGF. Normal 3T3 required permissive amounts of serum (0.25%) for maximal response to EGF (initiation of DNA synthesis, clonal/mass culture growth). Half-maximal DNA stimulation occurred at 0.20 nM with 3T3. EGF did not significantly enhance the growth of the SV_{40}3T3. Comparative binding studies of 3T3 and SV_{40}3T3 using ^{125}I-EGF at 24 and 37°C showed half-maximal saturation of receptor sites at 0.36 nM EGF; average dissociation rate was 0.44 x 10^{-4} sec^{-1}. Maximal binding occurred between pH 6.8-7.6. Unlabeled EGF competed with ^{125}I-EGF for receptor sites while fibroblastic growth factor and insulin did not. Scatchard analysis of the binding data revealed single high affinity binding sites with an observed Kd of 10^{-9} for both cell types. 3T3 and SV_{40}3T3 have essentially the same number of receptors on a per cell basis, but SV_{40}3T3 were smaller and had more receptor sites per μ^2 of cell surface. Transformation of Swiss 3T3 is accompanied by an altered responsiveness to EGF and a change in surface density of EGF receptors.

194 ONTOGENY OF RENAL β-AMINO ACID TRANSPORT IN RAT KIDNEY CORTEX. Russell W. Chesney and Diane K. Jax The University of Wisconsin Center for Health Sciences, Children's Hospital, Dept. of Pediatrics, Madison, Wisc.

Taurine (T), a β-amino acid characteristic of rodent urine, is taken up across the antiluminal membrane into adult cortex slices by 2 uptake processes that require oxidative metabolism and the presence of sodium. This non-metabolizable compound is not incorporated into TCA precipitable material (<.01%) and thus is an ideal agent to directly examine transport. It can be used to explore the ontogeny of peritubular transport processes. Uptake of T is slower in newborn (N.B.), 2 week and 4 week old rat cortex slices than in the adult at 0.1, 1.0 and 20 mM T, but faster in NB at 0.01 mM T. Moreover, NB tissue is far more capable of T uptake in the presence of N_2 (anoxia). Concentration-dependent uptake studies at all ages reveal at least 2 uptake processes: a high-affinity site with Km_1=0.1 to 0.5 mM and a low-affinity site with Km_2=12 to 15 mM. However maximal uptake at both sites proceeds adult > 4 week > 2 week > NB and initial uptake rate is greater in adult slices. β-alanine shares both uptake sites at all ages and both sites are Na-dependent. Efflux of T from preloaded slices is slower in NB, 2 week and 4 week slices as a further indication of the ontogenic maturation of peritubular transport processes with aging.

These studies provide a setting against which such manipulations as dietary alteration or the use of hormonal agents can be used to attempt to augment the rate of uptake and efflux of T in immature slices.

195 RELATION OF THYMIDYLATE SYNTHETASE (TS) ACTIVITY TO DNA ACCUMULATION RATE IN DEVELOPING RAT CEREBELLUM: EFFECT OF HYPER- AND HYPOTHYROIDISM. Brian R. Clark and Morton E. Weichsel, Jr., UCLA Sch. of Med., Harbor Gen. Hosp. Depts of Psychiatry and Pediatrics, Torrance, Calif.

TS, an enzyme of the pathway by which uridylate (an RNA precursor) is converted to thymidylate (a DNA precursor), is elevated in cells undergoing replicative DNA synthesis.

We measured TS activity and DNA content in developing cerebellum of normal, thyroxine-treated, and propylthiouracil-treated neonatal rats. An expression for cerebellar DNA accumulation rate (DAR) was derived by differentiation of the equation obtained using the method of least squares to fit a sigmoid curve ($y=A/1+be^{-Cc}$) to cerebellar DNA content versus age.

Hyperthyroidism induced by thyroxine significantly increased both TS activity and DAR above control values on days 2-7, and significantly decreased both measurements below control on days 9-15. The coefficient of correlation between TS activity and DAR was 0.96 for control and hyperthyroid pups.

Hypothyroidism induced by administration of propylthiouracil (PTU) to nursing dams significantly decreased both TS activity and DAR below control values by age 5 days and significantly increased both measurements above control on days 15-21. The coefficient of correlation between TS activity and DAR was 0.98 for pups of both control and PTU-treated dams.

The close correlation between cerebellar TS and DAR in control, hyperthyroid, and hypothyroid animals supports the possibility that TS may be critical in the control of cell replication rate in developing rat cerebellum.

196 EMBRYONIC RENAL INJURY: A POSSIBLE FACTOR IN FETAL MALNUTRITION. Jackson J.W. Clemmons (Spons. by J. Lucey), University of Vermont College of Medicine, Department of Pathology, Burlington, Vermont.

Chick embryo metabolism was monitored in a radiorespirometer where O_2 consumption, CO_2 production and $^{14}CO_2$ from ^{14}C-labeled proline, glutamate, aspartate and arginine was measured continuously during development. At nine days of development chemically dissimilar nephrotoxins (spermine, $Na_2Cr_2O_7$, ethylene glycol monoethyl ether) were inoculated into the air sac which resulted in a temporary change in respiratory quotient from 0.71 to 0.85. At 18 days injured embryos consumed oxygen equal to that of a 14 day embryo and were 30-35% smaller in weight. Embryos with renal injury but matched with controls in terms of O_2 utilization had a profound decrease in ability to metabolize the $^{14}C_2$-labeled substrates. Decreased ability to metabolize ^{14}C-labeled proline was associated with decreased collagen formation. At 18 days there was pulmonary hypoplasia with sparce mesenchyme maturation but little residual evidence of renal injury that could be observed at ten days. It is suggested that renal injury during early fetal development may alter renal gluconeogenesis in addition to interfering with pathways essential for collagen formation, thereby contributing to fetal malnutrition and pulmonary hypoplasia. Supported by N.I.H. Grant HD08864.

197 POSTNATAL CHANGES OF ACTIVITY OF ACID LIPASE AND NONSPECIFIC NEUTRAL ESTERASE IN RAT LIVER. Paul M. Coates, Spencer A. Brown, Jocelyn Jumawan and Otakar Koldovsky (Spon. by Jean A. Cortner). The Children's Hospital of Philadelphia, Joseph Stokes, Jr. Research Institute Phila.

Hydrolysis of the 4-methylumbelliferyl (4-MU) esters, oleate and nonanoate, was studied in homogenates of liver from suckling and adult rats. In both age groups, the pH optimum for 4-MU oleate hydrolysis was 4.0, whereas 4-MU nonanoate was hydrolyzed maximally at pH 6.5-7.0. Acid lipase (4-MU oleate, pH 4.0) was resistant to inhibition by the organophosphate, diethyl-p-nitrophenyl phosphate (E600), while nonspecific neutral esterase (4-MU nonanoate, pH 7.0) was sensitive to inhibition. Acid lipase and nonspecific neutral esterase differed substantially in the patterns of their postnatal development in the liver. Acid lipase activity was high during the suckling period and decreased after the third week of life to adult levels which were 50% of suckling levels. Nonspecific neutral esterase increased markedly with age, and adults attained levels 10 times those of sucklings. These experiments together with our concomitant findings of high acid lipase in the small intestine of suckling rats suggest the important metabolic role of acid lipase during the suckling period when fat intake is high. (Supported in part by NIH grants HD 08536 and HL 18723-01).

198 EFFECT OF INTRAUTERINE HYPOXIA ON MITOCHONDRIAL RESPIRATORY ACTIVITY OF THE FETAL LAMB. Maria Delivoria-Papadopoulos, Leena Mela, James D. Ferguson, and Leonard D. Miller. University of Pennsylvania School of Medicine, Departments of Physiology, Pediatrics, & Surgery. Philadelphia, PA

Fetal brain and heart mitochondrial respiratory activity during maternal hypotension was studied in 12 fetal lambs of 120-130 days gestation. Control data were obtained from 16 lambs of comparable gestation. All 28 ewes had low spinal anesthesia. Maternal hypotension was sustained by continuous infusion of trimetaphan camsylate for 60-90 min. Maternal and fetal blood gases were measured prior to and during hypotension. The respiratory activity of isolated brain and heart mitochondria were standardized per mole of cytochrome oxidase determined from the same preparations. In control fetuses brain and heart mitochondrial respiratory activity was 1037 moles of O_2 utilized per mole of cytochrome($a + a_3$) per min. and 1949 moles of O_2 utilized per mole of cytochrome($a + a_3$) per min., respectively. During maternal hypotension fetal blood oxygen tension decreased by a mean of 12 mm Hg from baseline values. Brain and heart mitochondrial respiratory activity decreased to a range of 228-442 moles of O_2/mole($a + a_3$) per min. and 312-400 moles of O_2/mole($a + a_3$) per min., respectively, a significant drop in the metabolic activity(75% in brain and 85% in heart) of the mitochondria. In previous studies we have shown that acute hypoxia does not damage mitochondria when tissue perfusion is well-preserved. The present results suggest that deterioration of fetal mitochondrial respiratory activity is due to a significantly decreased oxygen availability, presumably produced by placental hypoperfusion.