- **246** A STUDY OF THE STRUCTURAL AND BIOCHEMICAL DEVELOPMENT OF HUMAN FETAL ISLETS OF LANGERHANS. Hy Goldman, Ines Wong and Yogesh C. Patel (Spon. by C.R. Scriver) McGill University-Montreal Children's Hospital Research Institute & Fraser Labs., Royal Victoria Hospital, Montreal, Quebec, Canada
- Variably-sized, histologically intact islets were isolated from 14 human fetal pancreases (12.0-19.5 wks. gestation) and grouped according to gestation age, region of the pancreas (splenic and duodenal) and diameter sizes. The mean number of cells/islet, based on DNA/islet was $\sim 30,000$ ($>300\mu\text{m}$ diameter), $\sim 11,000$ ($200-300\mu\text{m}$), $\sim 3,500$ ($100-200\mu\text{m}$) and $\sim 2,000$ ($<100\mu\text{m}$). In both duodenal and splenic islets and all sizes the mean concentration of insulin (IRI) and glucagon (IRG) and somatostatin (IRS), expressed as pg or pg equivalents of insulin, glucagon and somatostatin per islet simultaneously increased significantly in three distinct step-wise developmental tiers: (1) 12.0-14.5 wks, (2) 15.0-18.5 wks, (3) 19.0-19.5 wks. In contrast, pancreatic polypeptide (IRPP) concentration did not. At all fetal ages and islet sizes, content of IRI, IRG and IRS was significantly greater than IRPP in splenic islets while IRPP was greater in duodenal islets. Mean hormone ratios at all ages and sizes indicate IRI content is significantly greater than IRG and IRPP but not IRS. In summary, a precise pattern of multiphasic hormone development of IRI, IRG and IRS as well as the relative importance of IRS-containing D cells has been uncovered in human endocrine pancreas during fetal ages 12.0-19.5 wks.

- **247** RELATIONSHIP OF BINDING AND METABOLIC DISPOSITION TO INTERNALIZATION OF INSULIN. Robert E. Greenberg and William F. Howell, University of New Mexico School of Medicine, Department of Pediatrics, Albuquerque
- Previous studies by others have demonstrated internalization and lysosomal degradation of insulin in isolated cells, following binding to membrane receptors. In the present studies, the perfused rat liver was used to study the relationship between internalization, binding and metabolic disposition of ^{125}I insulin. Washed, human RBC in 4% BSA and KRB were used in open perfusion of liver in situ in weanling rats. After ten minutes of perfusion, 1×10^6 CPM insulin 125 were added to the pre-hepatic line. Samples were serially collected for determination of total, TCA soluble and precipitable counts.
- At a perfusate concentration of 250 $\mu\text{U/ml}$, 35% of labelled insulin is bound to liver. During the subsequent 20 minutes of perfusion, approximately 30% of bound insulin re-appears in the perfusate as intact (TCA precipitable) insulin. Less than 1% of total counts are present in liver after 20 minutes. Addition of methylamine (10 mM), which inhibits clustering of hormone-receptor complexes, increased the amount of ^{125}I insulin bound, and reduced amount degraded. Lysosomotropic agents (chloroquine, colchicine, and NH_4Cl) had no effect on the amount of insulin bound but markedly inhibited ($> 60\%$) the amount degraded.
- These studies indicate that: 1) greater than one half of bound insulin is internalized; 2) insulin binding to receptors is independent of rates of lysosomal degradation; 3) insulin bound to hepatic membranes may be, in part, released intact to the peripheral circulation.

- 248** DEVELOPMENTAL CHANGES IN FATTY ACID OXIDATION IN NEWBORN GUINEA PIG HEART. Daniel E. Hale, Charles A. Stanley, Lester Baker, University Pennsylvania School of Medicine, Children's Hospital of Philadelphia, Department of Pediatrics, Philadelphia. Fatty acids (FA) are important substrates for the myocardium and liver in adult mammals. Developmental changes have been demonstrated in FA oxidation in newborn rat and guinea pig liver. The purpose of this study was to determine if similar changes occur in guinea pig heart. In vitro rates of FA oxidation were determined using homogenates of heart from near term fetuses delivered by C-section, from newborn pups, and adult guinea pigs. Rates of ^{14}C CO $_2$ production from [$1-^{14}\text{C}$] palmitate (0.75mM) at 35°C over 30 minutes are expressed as $\mu\text{M/hr/gm}$ tissue.

	FETUS	1 DAY	5 DAY	ADULT
Mean $\pm$ SEM	1.12 $\pm$.03	2.50 $\pm$.36	3.12 $\pm$.27	3.11 $\pm$.13
(N)	(5)	(4)	(4)	(4)

At 6 hours of age, 2 spontaneously born guinea pigs had FA oxidation rates of 1.79 and 1.09 $\mu\text{M/hr/gm}$. These findings indicate that in the newborn guinea pig heart there is a 3 fold increase in the capacity to oxidize FA. This developmental change occurs post-natally. The sites of limitation and regulatory mechanisms are yet to be determined. These findings may have implications in the nutrition of premature infants.

- 249** FAILURE OF EARLY INFANT STIMULATION IN IMPROVING THE PROGNOSIS OF SEVERELY ASPHYXIATED NEONATES. R.G. Harper, B. Wilson, C.G. Sia, M. Olson, R. Berman, S. Lubrano (Spon. by F. Lifshitz). Cornell Univ. Med. Coll., North Shore Univ. Hosp., Depts. of Ped., Ob.-Gyn., and Neur., Manhasset, N.Y.
- The results of early stimulation in the treatment of severely asphyxiated neonates was evaluated. Eight patients with fetal distress (8/8) and perinatal asphyxia (8/8) who required resuscitation in the delivery room (8/8) with thick meconium in the trachea (7/8) and 1-min Apgar scores of ≤ 4 (7/8) were assessed at 8 mos-2 yrs of age. These patients had prolonged and difficult-to-control seizure activity (8/8) and required multiple anticonvulsant therapies (8/8) and respirator support (7/8) during the neonatal period. All received early sensory motor stimulation (i.e., aural, visual, tactile, oromotor, positioning, etc.) from a neurodevelopmental therapist within the first 13 mos of life for a minimum of 2 hrs/wk and the parents were instructed to give ongoing daily therapy. Despite early infant stimulation all infants (8/8) showed persistent psychomotor delay. Abnormal initial CTT's occurred in 4/8; abnormal followup CTT's in 6/6; abnormal EEG's in 7/8. Severely asphyxiated infants who receive early stimulation do not achieve normal development despite comprehensive intervention.

- **250** THYROXINE (T $_4$) ACCELERATES EPIDERMAL GROWTH FACTOR (EGF) PRODUCTION IN NEONATAL MOUSE SUBMAXILLARY GLAND (SMG). Steven Hoath, Jay Lakshmanan, Susan Scott and Delbert A. Fisher, UCLA School of Medicine, Harbor-UCLA Medical Center, Department of Pediatrics, Torrance, CA
- EGF is a 6045 MW protein found in highest concentration in mouse SMG. It is known to accelerate epidermal maturation in rodents, i.e., hair differentiation, incisor eruption and neonatal eyelid opening. Previously, we have described the normal, rapid, logarithmic increase in EGF concentration in mouse SMG during the first 60 days of life as well as an increase in adult SMG-EGF after T $_4$ treatment. In the present study we report the effect of T $_4$ on EGF production during development. Sixteen newborn albino mice were randomly allotted to 2 litters. Pups in 1 litter received T $_4$ from birth (1.0 μg T $_4$ daily); the other litter served as controls. Mice were sacrificed on postnatal day 17. The SMG were dissected, homogenized and assayed for EGF by a specific radioimmunoassay sensitive to 20 pg per tube. Eyelid opening in the T $_4$ treated animals was noted 48-72 hours prior to eyelid opening in the matched controls. No significant differences in mean body or SMG weights were noted between groups. EGF concentrations in control and T $_4$ treated SMG were 0.025 $\pm$.003 and 33.5 $\pm$ 4.1 ng/mg tissue, respectively (Mean $\pm$ S.E.). A similar mean increase of 1800 fold was observed when results were expressed as ng EGF/mg SMG protein. Conclusions: 1) T $_4$ markedly increases EGF concentration in the developing mouse SMG. 2) T $_4$ accelerates epidermal maturation in the developing mouse. Speculation: The epidermal maturational effects of T $_4$ may be mediated by EGF.

- 251** THE ACUTE EFFECT OF FETAL EXCHANGE TRANSFUSION (XT) WITH ADULT BLOOD ON FETAL OXYGENATION. Joseph Itskovitz, Boyd W. Goetzman, Abraham Rudolph, Univ. Calif., Cardiovasc. Res. Institute, San Francisco.
- Reducing hemoglobin (Hb) affinity for O $_2$ by exchanging fetal with adult blood has been reported to have no effect on O $_2$ consumption (VO $_2$) in fetal lambs studied acutely (J Appl Physiol 26:410, 1969). We performed 3 volume isovolumetric XT with maternal blood in 7 chronically instrumented fetal lambs. Umbilical blood flow (UBF) was measured by the microsphere method. Umbilical venous (UV) and descending aortic (UA) pH, blood gases, and O $_2$ content were measured before and one hour after XT. UV PO $_2$ increased from 28.6 $\pm$ 2.1 ($\bar{x} \pm \text{S.E.}$) to 36.0 $\pm$ 2.1 torr ($p < 0.05$) and UV O $_2$ saturation fell from 78.3 $\pm$ 3.9 to 40.0 $\pm$ 3.3% ($p < 0.001$). UA PO $_2$ and PCO $_2$ did not change significantly from the controls of 20.2 $\pm$ 1.0 and 43.0 $\pm$ 0.6 torr. pH fell from 7.38 $\pm$ 0.01 to 7.23 $\pm$ 0.04 ($p < 0.02$) and Hb O $_2$ saturation fell from 51.8 $\pm$ 4.0 to 18.1 $\pm$ 1.5% ($p < 0.001$). UBF decreased from 245 $\pm$ 14 to 153 $\pm$ 9 ml/min/kg ($p < 0.001$). Total O $_2$ delivery fell dramatically from 22.84 $\pm$ 0.90 to 7.21 $\pm$ 0.96 ml/min/kg ($p < 0.001$) and fetal VO $_2$ decreased from 7.40 $\pm$ 1.03 to 3.58 $\pm$ 0.58 ml/min/kg ($p < 0.01$). Thus, replacing fetal blood with adult blood having low Hb O $_2$ affinity significantly reduced fetal oxygenation. As a result of reduced Hb O $_2$ saturation of the UV blood and reduced UBF, fetal O $_2$ delivery falls and fetal VO $_2$ is compromised. Consequently, metabolic acidosis develops, further compromising the O $_2$ saturation of Hb in the placenta. (Supported by NIH grants HL 06285 and HL 24056.)