●254 EFFECT OF CIMETIDINE AND ENDOTOXIN ON LUNG MICROSOMAL P450 ACTIVITY IN OXYGEN-TOXIC LAMBS. Thomas A. Hazinski, Patricia J. Hicks, Margaret L. France. Vanderbilt University School of Medicine, Department of Pediatrics, Nashville, TN.

One enzymatic source of free oxygen radical production in lung cells is the cytochrome P450 system. We have reported that a single dose of either endotoxin (E) or cimetidine (C), both inhibitors of hepatic P450 activity, significantly reduces pulmonary oxygen toxicity in lambs. To determine if the *in vivo* reduction of lung injury could be linked to lung P450 activity, microsomes were isolated from whole blood-free lung, and P450 activity was measured *in vitro* in 13 oxygen-toxic lambs and in 3 similarly instrumented lambs who breathed air for 3 - 5 days. P450 activity was estimated as the demethylation of d-benzphetamine with subsequent generation of formaldehyde. Results shown below (x ± 1 SD, * indicates p<.05 vs air, †indicates p<.05 vs O_2 alone.):

Group	Lung P450 Activity (nmoles CH_2O/10 min/mg protein)	Survival Time (h)
Air (n=3)	.87 ± .22	- - -
O_2 alone (n=5)	1.46 ± .37*	83 ± 11
O_2 + E (n=4)	.65 ± .15* †	131 ± 10 †
O_2 + C (n=4)	.35 ± .05* †	145 ± 6 †

The results indicate that in lamb lung 1) hyperoxia increases P450 activity, and 2) both cimetidine and endotoxin block the increase in P450 activity due to oxygen exposure. These studies lend further support to the hypothesis that P450-mediated oxygen metabolism may be important in the pathogenesis of pulmonary oxygen toxicity in lambs.

255 CORTISOL, ACTH β -ENDORPHIN: DIURNAL RHYTHMS OF INFANTS IN A NEONATAL INTENSIVE CARE UNIT. K.W. Hindmarsh, K. Sankaran, V.A. Laxdal, L.Tan, D.J. Christensen. Perinatal Research Laboratory, Newborn Services, University of Saskatchewan Saskatoon, Saskatchewan, Canada.

To determine whether a diurnal rhythm exists in neonates admitted to neonatal intensive care units (NICU) where there is continuous artificial lighting and periodic nursing and medical care exists, Plasma cortisol, ACTH and β-endorphin (BED) concentrations were measured in two groups of infants and another group of adult human volunteers. As expected, a diurnal rhythm is seen in adults. Plasma concentrations found during morning and (afternoon) hours were: cortisol - 475.0 ± 204.2 (255.0 ± 75.2) nmoles /L; ACTH - 10.9 ± 3.4 (13.0 ± 4.6) pg/mL; β-ED -22.1 ± 9.5 (16.8 ± 7.2) pg/mL. A diurnal rhythm was also observed for ACTH and cortisol in neonates (3-4 days postnatally):cortisol - 340 ± 194.5 (228.8 ± 101.0) nmoles/L; ACTH - 6.3 ± 4.2 (8.8 ± 5.5) pg/mL. Although there was not a statistically significant difference between morning and afternoon β -ED levels in this group, the afternoon levels like the adults were lower: 24.2 ± 13.0 (19 ± 9.8) pg/mL. There was no significant difference between the concentrations of any of the three substances in infants who were severely stressed clinically: cortisol - 320 ± 230.6 (406.4 ± 227.4) nmoles/L; ACTH - 7.0 ± 3.1 (8.0 ± 4.7) pg/mL; β- ED - 37.1 ± 19.4 (45.6 ± 35.7) pg/mL. It would appear that a diurnal rhythm exists in neonates within the first few days of postnatal life and that the continuous lighting, medical and nursing interventions do not seem to interfere with this rhythm. Severe stress seems to override this rhythm.

†256 EXAMINATION OF THE "SURFACE TENSION" OF NEWBORN RAT SKIN: METHOD AND EFFECTS OF EPIDERMAL GROWTH FACTOR (EGF) Steven B. Hoath, William Pickens, Diane Sneller (spon. by Jeffrey Whitsett) U. Cincinnati Med. Ctr. Dept. of Pediatrics, Cincinnati, Ohio.

In mammals, the declination of hair follicles suggests an asymmetrically aligned tension in the skin during development. We hypothesized that skin, as an elastic body, would exhibit retraction in vitro proportional to the tension in vivo. We tested this hypothesis in the newborn rat during the time of hair follicle morphogenesis and examined the effect of a known epidermal morphogen, EGF. Circular 12mm sections of dorsal skin from 12 control and 12 EGF-treated (500 ng/g BW) newborn rats were allowed to assume figures of equilibrium on prewetted polystyrene plates prior to culture in serum free Waymouth's media for 24h. Initial axial dimensions for the circular sections were:

	Sagittal (mm)	Transverse (mm)	
Control (day 0)	8.1 ± 0.2	12.0 ± 0.2	Mean ± SE
4h p EGF Rx	10.0 ± 0.3*	11.6 ± 0.2	*p < .001

Sagittal retraction was not inhibited by neuromuscular blocking agents (pancuronium, curare) or by sodium azide. All sections became circular after 24h in culture. Ellipse formation was not present in skin sections from 7-day-old pups.

Conclusions: 1) Newborn rat skin exhibits an endogenous tension in the direction predicted by the cephalo-caudal angulation of the developing hair follicles; 2) EGF treatment decreases skin tension as measured by the retraction assay described; 3) the tension-generating element in skin is not the hypodermal skeletal muscle; 4) this phenomenon is lost during adaptation to in vitro life and is limited to the neonatal period.

257 IN VITRO FORMATION OF SAGITTAL HELICES FROM NEWBORN RAT SKIN: CHIRALITY, ONTOGENY, AND LINEAR DIMENSIONS Steven B. Hoath, William Pickens, Diane Sneller (Spon. Jeffrey Whitsett) U. of Cincinnati Med. Ctr., Dept. of Pediatrics, Cincinnati, Ohio.

Organ culture explants adapt to conditions in vitro by forming "figures of equilibrium" with their environment (conformations of minimal surface energy). We propose that explant "morphogenesis" during in vitro adaptation is a function of in vivo tissue organization and lines of tension. In this report, we used a McIlwain tissue chopper to prepare 1mm x 20mm strips of newborn rat dorsal skin and compared the figures of equilibrium obtained from perpendicularly oriented sections. Transversely cut strips remained extended or formed gentle "u's" in culture. Sagittal strips, however, rapidly formed helices of opposite handedness in serum free Waymouth's media. Measurements of 18 sagittal strips from newborn (day 0) pups are shown:

Anatomic Site	# Coils (360°)	Handedness of helix	Transverse diameter of end coil Cephalic end (mm)	Caudal end
L side	3.5 ± 0.2	L (9/9)	3.1 ± 0.2	2.1 ± 0.2*
R side	3.1 ± 0.1	R (9/9)	3.3 ± 0.2	1.9 ± 0.1*

* p <0.01, cephalic versus caudal, mean + SE (all values)

The number of coils was stable for 24h in culture. In 3-day-old rat pups, mean coil number was 2.5 + 0.2 (N=30).

Conclusions: 1) In the newborn rat, sagittal skin strips form helices in vitro which retain their in vivo chirality; 2) sagittal helices are conical rather than cylindrical with the tightest coil located at the caudal end; 3) helix formation is stable for 24h in vitro; 4) the number of coils formed per sagittal centimeter of skin decreases rapidly after birth.

258 ALLOCATION OF SYSTEMIC GLUCOSE PRODUCTION TO CEREBRAL GLUCOSE UTILIZATION AS A FUNCTION OF INTRAUTERINE CANINE GROWTH. M Huang, R Kliegman, C Trindade, D Kall, K Voelker. Case Western Res.Univ, Rainbow Babies & Childrens Hosp, Dept.Peds, Cleve,OH

Hypoglycemia in intrauterine growth retardation may be due to an imbalance between glucose production (GP) and cerebral glucose needs. We investigated relationships between GP, cerebral glucose uptake (CMRG), brain and body weight in 16 fasted 3 hr old term pups weighing 263 g (170-345 g). Brain wt 6.1±0.3 g was 2.3% of body wt, while liver wt 11.8±1.5 g was 4.4% of body wt. Brain wt:liver wt ratio was 0.66 for the entire group. Mean pH 7.23, pCO_2 48.4, pO_2 90.0, blood glucose 3.36 mM (1.45-10.77mM), steady state GP 49.6 umol/kg/min, cerebral blood flow (CBF) 0.83 ml/g/min, CMRG 0.60 umol/g/min, and cerebral O_2 uptake ($CMRO_2$) 1.76 umol/g/min. The brain utilized 36.6±7.9% of total body GP. To test the effect of growth status at term upon brain glucose utilization, we analyzed these data as a function of birth wt. Both brain and liver wt increased as a function of body wt. However, brain:liver (r=-0.6, p<0.02) and brain:body (r= -0.63, p<0.01) ratios were greatest in smaller pups. Although CBF increased, $CMRO_2$, CMRG, and GP did not correlate with body wt. However, the % of GP used by the brain increased exponentially with body wt (r=0.50, p<0.05).
CONCLUSION: In small pups a smaller % of GP was used by brain despite the fact that the brain represents a larger % of body wt. These data suggest that smaller pups do not use a disproportionately larger % of GP to maintain their normal rates of cerebral glucose utilization. Within the range of wt there was no imbalance between GP and CMRG.

259 RELATIONSHIP OF SYSTEMIC GLUCOSE PRODUCTION TO CEREBRAL GLUCOSE UTILIZATION IN NEWBORN DOGS. M Huang, R Kliegman, K Voelker, D Kall, K Chau. Case Western Reserve Univ, Dept.Peds., Cleve, OH

To determine the magnitude of steady-state systemic glucose production (GP) used by the brain in 16 fasted 3 hr old term pups, we measured GP with a primed constant infusion of tracer [6-^{3}H] glucose; and cerebral uptake of glucose (CMRG) and O_2 ($CMRO_2$) taken as the product of arteriovenous difference of substrate and cerebral blood flow (CBF), as measured by the FICK principle with ^{14}C-antipyrine. The following static parameters were:

Body Wt	263.3±13.8g	Brain:body wt	2.32±0.11%
Brain Wt	6.08±0.28g	Glucose	3.36±0.63mM; range 1.45-10.77
Lactate	1.04±0.15mM	GP	49.6±11.0 µm/kg/min
pH	7.23±0.01	$CMRO_2$	1.76±0.37 µm/g/min
pCO_2	48.4±1.33	CMRG	0.60±0.15 µm/g/min
pO_2	90.0±7.0	CBF	0.83±0.10 ml/g/min

Oxygen/glucose index was 52.9±9.17%. Glucose extracted by the brain (A-V/A) was 27.6±4.1%. 36.6% of systemic GP was accounted for by brain uptake. The brain utilized 11.42±1.6 µm/kg/min of glucose provided by GP. Within the range of glucose levels, glucose did not correlate with CBF, CMRG or $CMRO_2$. CMRG was not related to GP. In contrast A-V/A decreased as an exponential function of glucose (r=-0.51, p<0.05). Furthermore, percent of GP used by the brain was an inverse function of GP (r=-0.71, p<0.001). IN CONCLUSION: although the brain is 2.3% of body wt, it uses 36% of GP. Secondly, within this range of glucose, CMRG is not affected and may be maintained by increased extraction at the lower end of blood glucose spectrum. Mean±S.E.