†1213 LOSS OF APPETITE IN UREA CYCLE DISORDERS (UCD): THERAPEUTIC APPROACHES - Susan Hyman, Mark L. Batshaw, Joseph Coyle, Cynthia Porter and Claude Bachmann. Dept. of Pediat., Psych. and Kennedy Inst., Johns Hopkins Med. Inst. Balto. and Dept. of Clin. Chem. Berne.

Hyperammonemic animals have decreased food intake and increased serotonin (5-HT) and its metabolite 5-hydroxy indoleacetic acid (HIAA) in brain (Life Sci. 33: 2417, 1983). We have treated 2 anorectic children with UCD with precursor restriction and antiserotonergic drugs. A 7 yo. with argininosuccinic aciduria refused all oral feedings and had elevated levels of HIAA in CSF (78ng/ml). She responded to restriction of dietary tryptophan, Trp (7 vs. 34 mg/kg/d) with the onset of spontaneous food intake. She was then treated with the 5-HT receptor blocker cyproheptadine (Cyp), 0.3 mg/kg/d, while receiving 14 mg/kg/d Trp. Food intake during a 5 min. free feeding condition was increased under both of the low Trp conditions as compared to the high Trp condition. However, % of total daily calories consumed during free feeding, a measure of appetite, was higher in the low Trp + Cyp condition, 12.6 + 2.8%, than in the low Trp condition alone, 8.2 + 1.1%, $p < .01$ (df=11) suggesting a synergistic effect of Trp restriction and Cyp. Pizotifen, another 5-HT receptor blocker reduced feeding time in a 7 mo. citrullinemic child from 180 to 130 min/d, p=.02 at a dose of 0.11 mg/kg/d and to 105 min/d at 0.17 mg/kg/d. Decreasing oral Trp from 29 to 15 mg/kg/d resulted in further improvement. In addition to good metabolic control, restriction of 5-HT precursors combined with antiserotonergic drugs may be useful in the treatment of appetite problems in UCD.

1214 PROPERTIES OF RED BLOOD CELLS (RBC) FROM FETUSES OF DIABETIC RATS. John D. Johnson and Sandra Trissell, University of New Mexico School of Medicine, Department of Pediatrics, Albuquerque, NM.

Hyperbilirubinemia in human infants of diabetic mothers (IDM) can be explained in part by elevated bilirubin production as assessed by endogenous carbon monoxide excretion (Veco). We have explored the hypothesis that elevated Veco in IDM may result from abnormal RBC deformability and decreased life span by studying properties of RBC from fetuses of diabetic rats (FDM).

Wistar rats were made diabetic by i.v. injection of 40 mg/kg streptozotocin, mated and offspring studied at 21-days gestation. FDM were hyperglycemic and had elevated glycosylated hemoglobin vs. controls (C), but comparable serum insulin.

Using polycarbonate filters with 5 μm pore dia., filtration ratio (ratio of filtration time of red cell suspension to buffer) was markedly increased in FDM (6.60 ± 1.15 for FDM vs. 18.4 ± 0.10 for C, $p < .001$). RBC survival was determined by cross-transfusion of ^{51}Cr-labeled fetal RBC to normal adult recipients:

		Percent injected ^{51}Cr in circulation			
	N	1 hr.	4 hr.	24 hr.	t1/2 (hr.)
Controls	10	88.8 ± 2.0	71.1 ± 3.7	26.3 ± 3.3	13.2 ± 1.3
FDM	10	81.6 ± 1.6	56.8 ± 1.7	15.5 ± 1.7	9.5 ± 0.7
p value		.012	.002	.009	.018

We conclude that RBC from FDM are less deformable and have decreased survival vs. C. Since serum insulin is equivalent in FDM and C, these abnormalities must be secondary to hyperglycemia and/or its concomitant effects and may be the result of non-enzymatic glycosylation of critical RBC membrane proteins.

1215 LYSOSOMAL EFFLUX OF FREE SIALIC ACID: IMPLICATIONS FOR SALLA DISEASE. Adam J. Jonas and M. Helen Huls, (sponsored by F.H. Morriss), Dept. Ped., Univ. Texas Medical School, Houston, Tx 77030

Salla disease is characterized biochemically by greatly increased levels of free sialic acid in lysosomes. In order to study the metabolism of free sialic acid (N-acetyl neuraminic acid, NANA) in lysosomes, highly purified lysosomes were prepared from rat liver by Percoll gradient centrifugation. These lysosomes were loaded with sialic acid by incubation in 250 mM ^{14}C-NANA. Lysosomal loading was both temperature and time dependent with optimal conditions obtained at 37° C for 20 minutes. Lysosomes treated in this manner and then subjected to freeze/thawing lost both 90% of their sialic acid content and 90% of their β-hexosaminidase activity. The lysosomal sialic acid thus appeared to be soluble rather than protein bound. Lysosomes containing 5,500 pmol sialic acid/μmol/min β-hexosaminidase activity lost 204 pmol/min or 37% of their sialic acid content over 10 minutes at 25° C without evidence of lysosomal breakage. Loss of sialic acid slowed markedly after 10 minutes to 40 pmol/min. Addition of 2 mM $Mgcl_2$/ATP to the lysosomes resulted in a 50% increase in the loss of sialic acid during a 10 minute incubation without change in lysosomal integrity as measured by β-hexosaminidase activity. These studies suggest that lysosomes may contain a transport system for the efflux of free sialic acid. Accumulation of free sialic acid in Salla disease would be consistent with defective lysosomal efflux of sialic acid.

†1216 ROLE OF PLASMA GLUCOSE IN THE REGULATION OF ENDOGENOUS GLUCOSE PRODUCTION IN THE HUMAN NEWBORN. Satish C. Kalhan, Anita Oliven, Katherine C. King and Carlos Lucero. Case Western Reserve University at Cleveland Metropolitan General Hospital, Div. of Pediatr. Metabolism, Cleveland, OH.

Although the endogenous glucose production rate (Ra) in the newborn human and animals is high compared with adults, their plasma glucose concentrations are low. The newborn frequently develop hyperglycemia in response to glucose infusions. Previous data have shown that glucose release by isolated liver from human fetus is regulated by glucose concentration. In the present study, the role of glucose in the regulation of endogenous glucose production was examined by infusing glucose at 2.6 - 4.6 mg/kg·min to 8 normal term, 5 preterm and 8 small-for-gestational age (SGA) infants. All infants were healthy and had no overt clinical problems. Ra was measured during basal state and during glucose infusion by tracer dilution using [6,6,$^2H_2$]glucose. (mean ± SD)

	Basal		Glucose Infusion	
	Ra	Glucose	Peak Glucose	Ra
	mg/kg·min	mg/dl	mg.dl	mg/kg·min
AGA	4.13 ± 0.22	61.3 ± 10.1	90.1 ± 12.9	1.07 ± 0.58
Preterm	3.49 ± 0.38	45.5 ± 14.1	81.4 ± 12.2	1.89 ± 0.57
SGA	4.25 ± 0.98	47.6 ± 5.8	66.1 ± 10.9	2.2 ± 1.12

There was a negative correlation between peak glucose (P_G) and Ra during glucose infusion ($r = -0.59$ $P = .006$). P_G was also related to the rate of glucose infusion. These data show that as in human adults, Ra is regulated by plasma glucose in the newborn infant.

1217 DEVELOPING RAT BRAIN (RB) BINDS MONOIODINATED 125 I-INSULIN ISOMERS SIMILAR TO OTHER EXTRAHEPATIC TARGET TISSUES. Michael S. Kappy, Derrel W. Clarke, Fred T. Boyd, Jr., and Mohan K. Raizada, U. of Florida College of Med., Depts. of Pediatrics and Physiology, Gainesville; and Bruce H. Frank, Lilly Research Laboratories, Indianapolis.

Considerable interest in the relationship between insulin and the brain has developed since Havrankova et al reported insulin and insulin receptors in RB in 1978. There is evidence that insulin may be synthesized in RB, and that the insulin/insulin receptor system in RB is functionally separate from that in the periphery. Recently, we documented that insulin stimulates protein and nucleic acid synthesis in glia from newborn RB, and established that the developing RB is a target tissue for insulin.

We studied the relative binding of monoiodinated insulin isomers (B26 and A14) to determine if membranes and cells from newborn RB showed relative binding characteristics which were similar to other extrahepatic insulin target tissues. RB membranes, neurons and glia bound B26 better than A14, whereas liver plasma membranes bound both equally. Competition-inhibition curves were generated using homologous 127 I-insulin isomers. Binding of B26 was greater than A14 at all concentrations. The relative maximal binding of B26 compared to A14 was 1.35.

Scatchard plots of the data were curvilinear, which is characteristic of other insulin target tissues. Receptor concentrations for each isomer were similar, but affinities for B26 were greater than for A14 at all points, suggesting that the entire differences in relative binding could be accounted for by differences in receptor affinity for the two isomers. This is in agreement with the findings in other insulin target tissues.

† 1218 SITES OF ACTION OF SOMATOMEDIN ON CHICK CHONDROCYTES. Stephen F. Kemp, J. Paul Frindik, Fu-Ju Ma, M. Joycelyn Elders. Department of Pediatrics, University of Arkansas for Medical Sciences, Little Rock, Arkansas.

Somatomedin C/insulin-like growth factor I (SM-C/IGF-I) is the major regulator of cartilage growth and glycosaminoglycan (GAG) biosynthesis. We have previously shown that stimulation of sulfate incorporation into GAG by SM is dependent upon protein synthesis, particularly synthesis of GAG protein core. Using a cultured chick chondrocyte system, we investigated the time necessary for stimulation by SM. Increased incorporation of thymidine into DNA occurred after a lag of 5 hours, sulfate into GAG after 1-2 hours, and leucine into protein and uridine into RNA within 15 minutes. Because stimulation of RNA and protein synthesis are both rapid, we studied whether the stimulation of GAG biosynthesis requires SM stimulation of RNA. Actinomycin D (5 ug/ml, which inhibits RNA synthesis without blocking SM stimulation of native GAG synthesis) and 4-methylumbelliferyl-B-D-xyloside (1 mM, which acts as an acceptor for initiation of GAG chains) were used for these studies. Addition of xyloside increased incorporation of sulfate into soluble GAG, but decreased incorporation into cell-bound GAG. The effect of SM was still present after addition of the xyloside with and without actinomycin D. We conclude that SM stimulation of protein and RNA synthesis occurs within minutes after addition of SM, and before stimulation of sulfation or DNA synthesis. This stimulation is independent of RNA synthesis, and is reflected by an increase in xyloside-initiated chains.