211 GAS CHROMATOGRAPHIC-MASS SPECTROMETRIC-COMPUTER (GC-MS-COMP) ANALYSIS OF LOW MOLECULAR WEIGHT (LMW) ORGANICS IN AMNIOTIC FLUID. Claudia R. Schuth, Betty J. Dowty, Fred A. Korndorffer, John L. Laseter (Spon. by Richard L. Fowler). LSU School of Med., Dept. of Ped. & OB-Gyn; and Univ of N.O., Center for Bio-Organic Studies, New Orleans.

Seventeen samples of amniotic fluid were analyzed by GC-MS-COMP techniques to establish a normal profile of LMW volatile organic compounds. All specimens were obtained by amniocentesis done for evaluation of fetal lung maturity. Of the 15 women undergoing amniocentesis prior to C-section, 11 had normal pregnancies, 2 had a history of hypertension, 1 a history of epilepsy (on medication) and 1 (not on medication) had CA-in-situ of the cervix and a history of one convulsive episode. The GC profiles were qualitatively reproducible in all samples except the 2 from the women with convulsions. These 2 showed an extra, still unidentified, GC peak not seen in any other sample. Of the 13 pregnancies with similar GC profiles, all infants were apparently normal at birth except for an Rh-affected infant. Of the 2 pregnancies with the extra GC peak, one infant died of a hypoplastic left heart syndrome, the other was apparently normal. Mass spectral data thus far has tentatively identified acetone, dichloromethane, hexane, methylcyclopropane, toluene. These data demonstrate the reproducibility of a "normal profile" of LMW volatile organics in amniotic fluid. The possibility exists that abnormalities in these compounds may be associated with certain disease states. The potential of the GC-MS-COMP methodology as a future clinical diagnostic tool appears worthy of further investigation.

212 POTASSIUM STIMULATION OF ALDOSTERONE IN THE NEWBORN LAMB. Sharon R. Siegel (Spon. by Delbert A. Fisher) Fetal Maternal Research Laboratories, UCLA-Harbor General Hospital, Torrance, CA

Plasma renin activity (PRA) and aldosterone levels are dependent on daily sodium balance and have been significantly correlated with urinary sodium concentrations and blood pressure in the adult and in newborn infants. Potassium is an important stimulus for aldosterone secretion in the adult but it is not known whether potassium stimulates aldosterone in the newborn. The purpose of this study is to determine the effects of potassium on aldosterone and PRA levels in the newborn lamb. Five newborn lambs 5-10 days of age were infused with KCl 2 mEq/Kg in 20 ml of isotonic saline over 15 min. Blood samples were drawn at 5, 15, 30 and 60 min. after the start of the infusion for measurements of plasma aldosterone and PRA by RIA and serum sodium and potassium by flame photometry. The serum potassium levels increased from a M and SEM baseline of 4.6 $\pm$ .005 mEq/L to 6.4 $\pm$ 0.4 ($p<.05$) at 15 min. and 6.4 $\pm$ 0.2 ($p<.05$) at 30 min. decreasing to 5.6 $\pm$ 0.3 at 60 min after starting the potassium infusion. Plasma aldosterone levels increased from a (M and SEM; ng/dl) baseline of 5.3 $\pm$ 0.4 to 11 $\pm$ 1.1 at 15 min. ($p<.01$), 12.8 $\pm$ 2.3 at 30 min. ($p<.05$) and 19.5 $\pm$ 7.2 at 60 min. There were no changes in PRA, Hct, serum Na, or blood pressure during the 60 min. study.

These results show that potassium is capable of stimulating aldosterone in the newborn lamb, while not depressing PRA.

213 FUROSEMIDE STIMULATION OF THE RENIN-ANGIOTENSIN-VASOPRESSIN SYSTEM IN THE FETAL LAMB. Sharon R. Siegel, Richard E. Weitzman, and Delbert A. Fisher, Fetal-Maternal Research Laboratories, UCLA-Harbor Gen. Hosp. Torrance, CA

We have shown previously that furosemide (FU) stimulates secretion of renin and arginine vasopressin (AVP) in the newborn lamb. Nephrectomy abolished the plasma renin activity (PRA) and AVP responses, and saralasin acetate, an angiotensin II inhibitor, blocked the AVP response, suggesting that the AVP stimulation by FU is mediated by angiotensin II. The present study was designed to assess whether FU influences the renin-angiotensin-AVP system in the fetus. Seven fetal lambs 120-142 days gestational age and 6 newborn lambs were studied (term = 145 days). FU (2 mg/kg estimated fetal weight) was infused over 1-2 min. Blood samples were drawn at 8, 20, 35, and 65 min. post infusion. In the fetal lambs the M and SEM PRA (ng/ml/hr) increased from 10.0 $\pm$ 3.3 to 12.5 $\pm$ 3.6 at 8 min. ($p<.05$), 16.8 $\pm$ 4.4 at 20 min. ($p<.01$) and 24.3 $\pm$ 5.1 ($p<.01$) at 35 min. post FU. In the newborn lambs PRA increased from a base of 16.7 $\pm$ 5 to 41.8 $\pm$ 6 ($p<.01$) 20 min post FU and remained high. The M and SEM plasma AVP (µU/ml) increased 30 min. after PRA from a baseline of 2.1 $\pm$ 0.5 to 5.7 $\pm$ 1.8 ($p<.05$) 35 min. and 6.1 $\pm$ 1.4 ($p<.05$) 65 min. post FU in the fetal lambs. In the newborns AVP increased from a baseline of 2.7 $\pm$ 0.5 to 9.9 $\pm$ 3.4 ($p<.05$) 35 min and 13.9 $\pm$ 4.4 ($p<.05$) 65 min post FU. Plasma sodium, Hct, and osmolality did not change. Conclusions: 1) FU stimulates the renin-angiotensin-AVP system in the fetal lamb after 120 days gestation; 2) fetal responsiveness is less than in the newborn animal.

214 EFFECT OF LIGHT ON THE PERFUSED GUNN RAT LIVER, T. R. C. Sisson, B. Granati, R. Sonawane, T. Fiorentino. (Spon. by A. DiGeorge) Temple University School of Medicine, Department of Pediatrics, Philadelphia.

The livers of 6 Gunn rats (200 gm.), 3 jj and 3 Jj, were perfused for 1 hr. with std. KBR soln. to which bilirubin in conc. of 20 mg/dl was added to 4. One jj and 1 Jj were perfused in the dark, 2 each in blue fluorescent light (420-470 nm, 18 $\mu w/cm^2/nm$). Bilirubin was extracted in CCl_4 from half of each liver, the other half analyzed for activity of: cytochrome P450, b5, p-nitrophenol glucuronidation, benzo(a)pyrine and analine hydroxylation, aminopyrine demethylation. All livers perfused with KBR-bili. or KBR alone under light had little or no bilirubin at end of perfusion. Those perfused in the dark had significant amounts of bilirubin. Assays of both jj and Jj rat livers showed a marked increase in activity of all 6 enzymes, 40+ percent. As all rats were female, no intersex variable was encountered.

Determination of bilirubin conc. of outflow KBR-bili. perfusate showed a small but significant decline q5 min. during perfusion. Bilirubin extract from livers of both jj and Jj rats in the dark was in high conc.

We conclude that visible light penetrates through the intact rat liver, directly photodegrading bilirubin, possibly enhancing its uptake. These data further indicate that visible light (420-470 nm) induces hepatic cell enzyme activity.

The possibility exists that visible light irradiation, as in phototherapy, will enhance drug metabolism by hepatic cell enzyme induction.

215 GUANOSINE 3':5'- CYCLIC MONOPHOSPHATE (cGMP) IN BROWN ADIPOSE TISSUE OF DEVELOPING RATS. Josef P. Skala, Peter Hahn and Brian L. Knight. University of British Columbia, Department of Pediatrics, Vancouver, B.C., Canada, and M.R.C.Lipid Metabolism Unit, Hammersmith Hospital, London, U.K.

Variety of cell functions,e.g. neuronal excitation,transport, secretion, proliferation and differentiation, are believed to be influenced by cGMP, a unique component in the complex network of biological regulations. To study its possible involvement in the regulation of both the function and the ontogenic development of brown fat (a heat generating organ most important neonatally) we have assayed the steady-state levels of cGMP in tissue extracts. Between 20 and 60 pmol/g w.w. (1 to 5 % of the cAMP levels) were found. Highest concentrations were observed perinatally and were followed by a progressive decline with age. Cold stress resulted in a significant but temporary increase in cGMP levels,as did NE administration to one-month-old animals. Chemical sympathectomy by 6-hydroxydopamine decreased the"resting"levels of cGMP in the tissue. Injections of insulin and of glucocorticoids to suckling rats increased the level of the nucleotide in brown fat;the time dependent bi-phasic profile observed did not correlate with that of cAMP.The protein kinase(s) system of brown fat from fetal and newborn rats showed 10 times higher affinity for cGMP than that of older animals. Further experiments will be required before a function of cGMP in brown fat can be proposed ; nevertheless its presence,its responsiveness to physiological and pharmacological stimuli,and the presence of its target system - a cGMP-dependent protein kinase - have been established.

216 PROTEIN KINASES IN BROWN ADIPOSE TISSUE OF DEVELOPING RATS. SEPARATION, SUBSTRATE SPECIFICITIES AND CHANGES IN INDIVIDUAL ACTIVITIES DURING PERINATAL DEVELOPMENT Josef P. Skala & Brian L. Knight,University of British Columbia, Department of Pediatrics, Vancouver,B.C.,Canada, and M.R.C.Lipid Metabolism Unit, Hammersmith Hospital, London, U.K.

Brown adipose tissue of the rat undergoes a very rapid process of functional and morphological maturation perinatally,reaches a peak in its differentiation and functional capacity 1 to 2 weeks after birth and its involution commences at 3 to 4 weeks of age. The regulation of the tissue function, i.e. heat production, and of its maturation, are hormone-mediated and are associated with the phosphorylation and de-phosphorylation of specific proteins and enzymes. The diversity of the protein kinase system, and yet its specificity to carry out phosphorylations associated with a particular function, is reflected in that nine activities could be separated from the tissue soluble extracts on polyacrylamide. Five of the enzymes were histone kinases,two preferred phosvitin or casein, and arginine-rich histone and protamine were rapidly phosphorylated by the remaining two. Cyclic AMP stimulated the phosphorylation of histones but not that of the other proteins. The pattern of activity changes during development was different with different protein substrates, as was the relative abundance of each individual enzyme. It is speculated that there are three classes of protein kinases in brown fat: a) those related to the tissue's function,e.g. lipolysis and glycogenolysis ; b) those related to proliferation and cell division ; c) those related to the tissue's differentiation, e.g. enzyme inductions.