DEFECT IN 5'-DEOXYADENOSYLCOBALAMIN SYNTHESIZING ENZYME IN METHYLMALONICACIDEMIA. Maurice J.Mahoney, Anita C.Hart and Leon E. Rosenberg (Intr. by Joseph B.Warshaw). Yale Univ., Depts. of Human Genetics and Pediatrics, New Haven.

Inherited methylmalonicacidemia (MM-emia) due to deficient activity of methylmalonyl-CoA mutase may be caused by an abnormal mutase apoenzyme or by reduced holoenzyme activity secondary to impaired synthesis of the mutase-requiring coenzyme, 5'-deoxyadenosylcobalamin (Ado-Cbl). We studied Ado-Cbl synthesis in 5 fibroblast lines from unrelated patients who have MM-emia due to defective cobalamin (vitamin B_{12}) metabolism. Their whole cells in culture fail to synthesize Ado-Cbl but make normal amounts of the other cobalamin coenzyme, methylcobalamin.

After breaking the cells to define the steps in Ado-Cbl synthesis, we assayed the final step which combines the reduction of cob(II)alamin to cob(I)alamin and the adenosylation of cob(I)alamin. Subcellular fractionation studies localized this activity to mitochondria. In crude broken cell extracts, enzyme activity was deficient or absent in 3 of the 5 mutant cell lines (4,0,0 pg Ado-Cbl/mg protein/30 min;control lines, 23 ± 8). The other two MM-emia lines had normal activities (23 and 17). These results establish deficiency of Ado-Cbl synthesizing enzyme as one cause of abnormal cobalamin metabolism and MM-emia. The two MM-emia lines that had normal enzyme activity suggest there is further heterogeneity within the disorders of cobalamin metabolism as well as in the MM-emia phenotype.

MORQUIO'S SYNDROME: A DEFICIENCY OF CHONDROITIN SULFATE N-ACETYLHEXOSAMINE SULFATE SULFATASE. Reuben Matalon, Bradley Arbogast and Albert Dorfman. Univ. of Chicago, Dept. of Pediatrics, Chicago, Illinois.

Morquio's syndrome, an autosomal recessive disease exhibiting severe skeletal deformities and platyspondyly, is characterized by excretion of keratan sulfate and chondroitin sulfate. Extracts of cultured skin fibroblasts were found to contain normal levels of arylsulfatases A, B and C, β-N-acetylhexosaminidase, β-glucuronidase and β-galactosidase. Incubation of extracts of Morquio fibroblasts with [^{35}S]chondroitin sulfate (prepared from embryonic chick epiphyses) resulted in release of 2% of the $^{35}SO_4$ in contrast to the release of 10-15% by extracts of normal, Hurler, Hunter or Sanfilippo A fibroblasts. Similar results were obtained when a heptasaccharide, GalNAc $^{35}SO_4$-(GlcUA-GalNAc $^{35}SO_4)_3$, prepared from chondroitin sulfate, was incubated with lysosomal preparations from cultured fibroblasts. These findings indicate that Morquio's syndrome is due to a deficiency of a specific N-acetylhexosamine sulfate sulfatase. On the basis of the structures of the compounds excreted in Morquio's disease, it is likely that the deficient enzyme hydrolyzes 6-0-sulfate linkages in glycosaminoglycans. Supported by USPHS Grant Nos. AM-05996, HD-04583, AM-05589-06 and 2-MOl RR-00305-08.

IS CYSTINE ESSENTIAL FOR PREMATURE INFANTS? John I. Malone, John S. Curran, and Lewis A. Barness. Univ. of S. Fla. and Tampa General Hospital, Dept. of Pediatrics, Tampa 33620.

Growth in premature infants (weight 1000-2000 gms, 26-32 wks gestation) fed a commercially available low cystine formula was not significantly different from those fed a formulation containing twice the dietary cystine. The formulas were identical with the exception of protein quality; the low cystine formula contained 13mg/100ml and the high cystine formula 24mg/100ml.

Formula		13 mg/100 ml	24 mg/100 ml
Weight change (6 wks)		187 gm/wk ± 62	157 gm/wk ± 56
Head Circumf. (6 wks)		.80 cm/wk ± .13	.68 cm/wk ± .17
Length (6 wks)		1.0 cm/wk ± .12	.75 cm/wk ± .15
Component	Time (wks)	13 mg/100 ml	24 mg/100 ml
1. Serum Cystine	3	.056 μM/ml ± .01	.08 μM/ml ± .038
	6	.024 μM/ml ± .02	.038 μM/ml ± .009
	9	.028 μM/ml ± .018	.058 μM/ml ± .018
	12	.036 μM/ml ± .01	.038 μM/ml ± .016
2. RBC	1	28 mg% ± 4.9	35.6 mg% ± 3.5
	2	29.4 mg% ± 4.6	29.2 mg% ± 5.6
	3	26.2 mg% ± 3.3	25.2 mg% ± 1.0

There was no significant difference in growth rate of infants, in the serum concentration or the tissue levels (glutathione) of cystine in the two groups. Cystathionase activity in the liver has been reported to be absent in premature infants. These results indicate that the low cystine formula contains adequate cystine for normal growth of premature infants & provides normal serum & tissue levels of this amino acid.

INCREASED GLYCOGEN CONTENT IN SUBCUTANEOUS ADIPOSE TISSUE FROM NEWBORN OFFSPRING OF DIABETIC MOTHERS: DYNAMICS AND ULTRASTRUCTURE. Ellen Monkus, Hana Pribylova and Victoriano Pardo. (Intr. by William W. Cleveland). Dept. of Ped., Univ. of Miami Sch. of Med., Miami, Fla., Inst. for the Care of Mother and Child, Prague, and V.A. Hosp., Miami, Fla.

Glycogen content of adipose tissue in the human neonate falls rapidly in the first hours of life. Since little or no glucose-6-phosphatase is found in fat tissue (rats), presumably glycogen is utilized only in situ, both to support energy metabolism (ATP generation) and to furnish alpha-glycerophosphate for reesterification of fatty acids. Previous investigations showed increased lipolysis (glycerol release) in adipose tissue from offspring of diabetic mothers.

In this study glycogen content (mg/gm w.w.) was measured in adipose tissue samples from the subcutaneous fat pad of the buttocks in 30 infants of insulin dependent diabetic mothers (IDM) and in 34 infants of gestational diabetic mothers (IGDM) obtained during the first five days of life. Comparisons were made with 86 infants from normal gestations.

Glycogen content was significantly increased in fat tissue from both IDM and IGDM. The rapid rate of decrease seen in normal infants was not observed regardless of whether these infants were infused with glucose or not. Electron microscopic studies suggested a characteristic ultrastructure of adipose tissue glycogen deposits when increased amounts of glycogen are present. These results are consistent with the hyperinsulinism of IDM and IGDM.

FATTY ACIDS IN REYE'S SYNDROME. Peter Mamunes, George H. DeVries, Charles D. Miller, Ronald B. David, Dept. Ped.& Biochem., Med. Col. of Va., Richmond, Va. (Intr. by H.M. Maurer).

We have postulated (APS-SPR, Apr. '72) that short and/or medium chain fatty acid accumulation is important in the pathogenesis of RS, causing mitochondrial swelling, cerebral edema, hypoglycemia and fatty viscera. This study reports gas chromatographic quantitation of serum free fatty acids (C8-C18) from 7 patients with RS, 8 controls (C) and 3 patients with acute and chronic liver failure (LF). Mean total free fatty acids (FAs) were much higher in all RS patients (9.5-185 mg%) vs C (0.35-2.75 mg%) and LF (0.69-3.0 mg%). Of greater significance was the 13-1500 fold increase in caprylic acid (C8) in 5 RS patients (vs C or LF). Because the sera were obtained at different stages of the disease the normal C8 levels in 2 patients may have been due to the very mobile nature of serum FAs. One 10 mo. old with coma had a C8 of 170 mg% (95.5% of total FA) on admission. Four hrs. after treatment with I.V. glucose and insulin the C8=0.15 mg% (normal range=0-0.12 mg%), but her status had not changed, necessitating 2 exchange transfusions 12 hrs. apart. C8 before the 2nd transfusion was 12 mg%. She recovered without sequelae and 12 days later C8= 0.08 mg%. All RS cases had appreciable elevations of other medium chain FAs (as high as 1.9 mg% C10). Long chain FAs were also elevated but only in proportion to the total FA. LF patients had no significant FA elevations. These findings implicate C8 (and possibly other medium chain FAs) as an important toxin in RS.

MALIGNANT TYROSINEMIA WITH APPARENT DIETARY CURE. Grant Morrow III and Lewis A. Barness. Univ. of Ariz., Dept. of Ped., Tucson, Ariz. and Univ. of S. Fla., Dept. of Ped., Tampa, Fla.

Tyrosinemia when associated with hypermethioninemia results in chronic liver disease and/or death. We report a 3 y.o. tyrosinemic male who has no evidence of liver disease after early dietary therapy. L.W. was diagnosed shortly after birth because a previous sib died of liver failure at age 6 months with tyrosinemia and hypermethioninemia (plasma tyrosine 0.519 and methionine 0.656 uM/ml; normal ranges are tyrosine 0.045-0.099 and methionine 0.023-0.035). L.W.'s plasma tyrosine, normal at birth, became elevated on day 3 (0.222 uM/ml) and rose on a low protein diet to 0.822 by 5 weeks. Methionine was abnormal at 5 weeks (0.160 uM/ml). Sobee plus 3200-AB begun at 6 weeks corrected tyrosine values but methionine ranged from 0.310 to 1.550 until 6½ months of age when he was able to tolerate a normal, low protein diet. Serum albumin (0.96 G%), prothrombin (0), pro time (15%) at 2 months rose to normal (albumin 4.06 G% at 11 mos: and pro time 85% at 22 mos). Repeated renal phosphate and creatinine clearances were normal. He had no difficulty with a general anesthetic for a herniorrhaphy at 22 months. Psychomotor development is normal as are liver function studies at 3 years of age. Height, weight and head size fall in the normal percentiles.

Early dietary, tyrosine restriction can apparently be very effective in preventing liver disease in this disorder.