mations of dioxyadenine content of urinary sediment and close attention to parameters of renal function; in both patients toxicity necessitated reduction in dose or addition of allopurinol. Erythrocyte 5-phosphoribosyl-1-pyrophosphate which was normally elevated (40–70 mμmol/ml) as a consequence of PRT deficiency in these patients, was reduced to 5–15 mμmol/ml during adenine treatment, but not to within the normal range (1–5 mμmol/ml). The high uric acid excretion in both was unchanged. Despite treatment of the younger child for seven months, he developed spasticity, motor retardation and early self-mutilation. The older child also showed no improvement in neurologic dysfunction. We conclude that adenine is of no therapeutic benefit, and is potentially toxic, in treatment of patients with the Lesch-Nyhan Syndrome.

69 *Aspects of Lipid Synthesis Unique to Brown Fat.* Robert E. Greenberg and Charlotte Sumida, Dept. of Ped., Stanford Univ. Sch. of Med., Palo Alto

Chemical thermogenesis, necessary for extra-uterine adaptation, is partly mediated through oxidation of fatty acids in brown fat, requiring a rapid rate of triglyceride turnover. Previous studies from our laboratory indicated that brown fat contains glycerokinase, making it possible for brown fat to reutilize glycerol. Triglyceride synthesis in brown fat, thus, may not be completely dependent on glucose transport. The role of glycerol in triglyceride synthesis in brown fat has been further studied, using both *in vitro* and *in vivo* techniques.

Explants of brown fat were incubated for periods up to 3 days, with either glucose or glycerol as substrate. Insulin was added at varying times, as were tracer quantities of acetate C^{14}, glucose C^{14} or glycerol C^{14}. Glycerol promoted a greater incorporation of acetate C^{14} into mono-, di- and triglycerides of brown fat than did glucose. Incorporation of glycerol C^{14} and glucose C^{14} into lipids of brown fat were both increased by insulin. IP injection of glycerol-2-H^3 into newborn rats was followed by much greater incorporation into lipids of brown as compared to white fat. Net triglyceride synthesis was not demonstrable *in vitro* regardless of substrate or insulin, suggesting marked enhancement of triglyceride turnover by insulin. Increased turnover rate is also indicated by greater conversion of acetate C^{14} to $C^{14}O_2$ in the presence of insulin.

These results suggest the following: (1) Glycerol can be utilized by brown fat as substrate for lipid synthesis; (2) transport of glycerol in brown fat is subject to regulation by insulin; and (3) the anti-lipolytic effect of insulin is not demonstrable in the *in vitro* system used in these studies.

70 *Neonatal Fat Metabolism: Developmental Aspects in Isolated Human Adipose Tissue Cells.* Milan Novak and Ellen F. Monkus, Univ. of Miami Sch. of Med., Dept. of Ped., Miami, FL (introduced by William W. Cleveland).

The *in vitro* metabolism of a suspension of adipose cells, prepared by collagenase disintegration of 5 to 20 mg samples of subcutaneous (white) adipose tissue, was studied in normal newborns 6 h to 6 days of age in comparison with normal adults. In order to relate results to cell number they were calculated in terms of DNA content. Glycerol release was elevated during the first day of life and then decreased below the level found in adults. Glycerol release was less activated by nor-epinephrine in the neonate. In the adult the free fatty acid (FFA) release was consistent with glycerol release (molar ratio of FFA/glycerol of about 3); in the neonate FFA release was much less which suggests that FFA was either being re-esterified or partially oxidized. Oxygen consumption was essentially the same in the adipocytes of neonates and adults as was its activation by nor-epinephrine.

Previous studies using intact adipose tissue fragments indicated an increased glycerol release and also an increased oxygen consumption in young neonates; comparisons were made on a basis of wet weight. The present study not only confirms the previous findings but also shows the increased glycerol release in young neonates to be a property of the individual adipose cell. On the other hand the increased oxygen consumption of neonatal adipose tissue is probably a function of the increased cellularity.

71 *Effect of Folic Acid on Amino Acid Metabolism in Pyridoxine Unresponsive Homocystinurics.* Grant Morrow III, Deborah Meltzer and Lewis A. Barness, Dept. of Ped. Hosp. Univ. of Pa., Univ. of Pa. Sch. of Med., Philadelphia, Pa.

Plasma folate levels were measured in 3 homocystinurics and found to be < 1 ng/ml (normal 5–10 ng/ml). All were on unrestricted, constant protein diets while 2 (A and B) were taking anticonvulsants for seizure control. Patient A had anemia (8.0 G% hemoglobin) and macrocytosis that responded to folic acid. B had macrocytosis but no anemia while C had neither. Plasma folate levels ranged from 14–20 ng/ml after therapy. Within 3 months of discontinuing folic acid in B and C their blood folates had fallen to 2.0 and 2.6 ng/ml. Plasma B_{12} levels and excretion of methylmalonate were normal.

Plasma and urine amino acids responded to folic acid as noted in the table:

	Plasma μM/ml		
	Homocystine	Methionine	Glycine
A	0.067 (0.037)*	0.027 (0.047)	0.235 (0.315)
B	0.206 (0.193)	0.090 (0.439)	0.292 (0.350)
C	0.118 (0.108)	0.176 (0.218)	0.260 (0.246)
	Urine μM/day		
	Homocystine	Methionine	Glycine
A	650 (350)	47 (74)	1298 (2740)
B	471 (658)	111 (314)	2030 (4180)
C	1420 (1410)	123 (144)	1470 (1690)

* Post-folate values in parenthesis.

Methylation of homocystine to methionine requires folic acid. Many homocystinurics may increase methylation to methionine thereby requiring more folate and increasing glycine as a result of demethylation of serine. Some homocystinurics may require long-term folic acid supplementation (A and B) whereas others are unresponsive either biochemically or hematologically (C). (Supported in part by USPHS grants AM-02231 and HD-04837.)

72 *Homocystinuria: Biosynthesis of Cystathionine and Homolanthionine.* Gerald Gaull, Yoshiro Wada, Karmela Schneidman, David Rassin, Harris Tallan and John Sturman, Dept. of Ped. Res., N.Y.S. Inst. Basic Res. Ment. Retard. and Depts. Ped. and Ophthal., Mt. Sinai Hosp. Med. Sch., N.Y.C. (introduced by Horace Hodes).