● 756 DEFICIENCY OF MUSCLE CYTOCHROME C OXIDASE (CO) IN LEIGH'S DISEASE. George E. Hoganson, Dennis J. Paulson, Raymond Chun, Robert L. Sufit, Austin L. Shug. University of Wisconsin Medical School, Center for Health Sciences, Departments of Genetics, Pediatrics and Neurology, Madison (Spon. by Russell Chesney)

A 3y/o male presented with a progressive neurologic disorder characterized by hypotonia, ophthalmoplegia and ataxia. Postmortum studies at 4 1/2y revealed Leigh's disease. Biochemical findings included elevation of blood lactate (L) (ranging from 1.9 to 7.0mM, normal mean ± SD 1.0±0.4mM), pyruvate (P) (0.11 to 0.25, normal 0.11±0.03mM) and L/P ratio (14 to 35, normal 10 to 18). β-OH-butyrate (BOH), acetoacetate and L were intermittently present in urine. Plasma amino acids were normal. CSF L (4.7mM) and P (0.24mM) were elevated. Increased urinary excretion of glycine and alanine was present. No increase in blood L or P was noted after IV glucose (100mg/kg). During an 18 hr fast blood BOH increased to 2000μM (normal 91±81μM, n=4) with little change in blood L, P, or glucose levels. Skeletal muscle biopsy (premortum) showed absent CO on histochemical staining. Muscle mitochondria exhibited decreased rates of State 3 respiration; 47 natoms O_2/min/mg protein with pyruvate/malate and 63 natoms O_2/min/mg protein with succinate/rotenone (reported normal rates 147 and 158 natoms O_2/min/mg protein, respectively). Muscle cytochrome spectrum obtained from the difference in absorption of reduced and oxidized forms revealed an absence of the 605 nM peak corresponding to CO. This case confirms the findings of Willems JL, et al (Ped 60:850, 1977) indicating that a subgroup of patients with Leigh's encephalomyelopathy have CO deficiency.

● 757 LACK OF HETEROGENEITY OF RESIDUAL ISOVALERYL-COA DEHYDROGENASE ACTIVITIES IN FIBROBLASTS FROM PATIENTS WITH ISOVALERIC ACIDEMIA. David B. Hyman and Kay Tanaka, Yale Univ. Sch. of Med., Dept. of Human Genetics, New Haven, CT (Spon. by Leon E. Rosenberg)

Fibroblasts from patients with isovaleric acidemia (IVA), a disorder of leucine catabolism, have deficient activity of isovaleryl-CoA dehydrogenase (IVDH), as demonstrated by Rhead and Tanaka (PNAS 77:580, 1980) with a tritium release assay using [2,3-^{3}H]isovaleryl-CoA as substrate. They found 12% of control activity using mitochondria isolated from 5 IVA lines. However, in crude homogenates of IVA cells they measured 50-60% of normal activity, while [2-^{14}C]leucine oxidation by the intact cells was 1-2% of normal. While this demonstrated a specific deficiency of mitochondrial IVDH activity in IVA cells, the magnitude of residual dehydrogenating activity could not be accurately determined. To resolve this problem, we improved the assay by using (methylenecyclopropyl)acetyl-CoA, an inhibitor of IVDH and several other acyl-CoA dehydrogenases, in paired assays, to determine the amount of tritium non-specifically released from the substrate. With this improved assay IVDH in crude cell homogenates of all nine fibroblast lines tested from patients with IVA had less than 3.5% residual activity (range 0 to 0.67 pmole/mg/min vs. 19.4 ± 8.0 for controls), and clinical severity did not correlate with residual activity. The uniformly low residual activities do not imply the existence of only one type of mutant allele, but do suggest that differences in clinical severity are not related to the magnitude of the residual IVDH activity.

● 758 SUBTLE ANTIGENIC DIFFERENCES BETWEEN NORMAL AND MUTANT ARYLSULFATASE B (ASB) DETECTED BY MONOCLONAL ANTIBODIES. S. Percy Ivy, Pamela Shirley, Miriam G. Blitzer, Emmanuel Shapira. The Hayward Genetics Center. Tulane University School of Medicine, New Orleans.

Normal ASB and the mutant enzyme in Maroteaux-Lamy syndrome (MLS) have been shown to be immunologically indistinguishable when compared using rabbit anti-ASB antiserum. In the present study a monoclonal antibody library was obtained by fusion of spleen cells from mice immunized with purified ASB and X63NS1/1-Ag-4 (NS1) mice myeloma cells. Seventy-six hybridoma lines with ASB antibody specificity were obtained, of which 64 were subcloned and further studied. Of these, 56 reacted with both control and mutant ASB. Five reacted only with the active form of ASB and not with either the mutant form or with partially denatured (enzymatically inactive) control ASB. Three reacted only with heat denatured or urea treated control ASB and with the mutant enzyme but not with the active form. The specificity of these antibodies toward ASB was documented using Western blotting. Several of these hybridoma lines were injected into Pristane sensitized mice, and the specific monoclonal antibodies were purified from the ascites fluid. A solid phase radioimmunoassay was developed using IV-16-8 antibody (reacts with control and mutant ASB and has high binding affinity to polyvinyl chloride), ^{125}I-labeled V-4-7 (reacts with both mutant and control ASB), and ^{125}I-labeled V-18-8 (reacts specifically with active ASB). Using this assay the ratio between active and inactive ASB was determined in liver, fibroblast and leukocyte homogenates from controls, homozygotes and heterozygotes for MLS.

759 PYRUVATE DEHYDROGENASE COMPLEX DEFICIENCY: A NOVEL PRESENTATION. K. Johnston, S. Packman, C. Newth, M. Patel, K-F. Sheu and G. Heldt. University of California San Francisco, Department of Pediatrics and Case Western Reserve University, Department of Pediatrics, Cleveland.

We report a central hypoventilation syndrome (CHS) in a boy with pyruvate dehydrogenase complex (PDHC) deficiency. Dephosphorylated PDHC was assayed in disrupted fibroblasts after pretreatment with dichloroacetate (DCA). Maximal specific activity of activated patient PDHC was 10-30% of controls, and was not increased by alterations in concentrations of pyruvate or cofactors. Normalization of plasma lactate by a high lipid diet did not prevent slow neurologic decline, with intermittent ataxia, episodic weakness, psychomotor retardation, ophthalmoplegia and retinal pigment epithelial changes. A CHS was documented by radiologic, electrophysiologic and pulmonary function criteria. Theophylline, progesterone and ritalin neither altered ventilatory response to CO_2 nor permitted weaning from the ventilator. In contrast peripheral chemoreceptor stimulants (doxapram; Almitrine) effected an acute doubling of minute ventilation with decreases in p_aCO_2. Longer-term response to Alimtrine was equivocal. In conclusion, measurement of disrupted fibroblast PDHC following DCA activation is an accurate assay for PDHC deficiency. PDHC deficiency must be considered in the differential diagnosis of CHS; to our knowledge, this is the first report of such an association. Finally, trial of a peripheral chemoreceptor agonist is warranted in the management of CHS.

760 PRENATAL DIAGNOSIS OF A DE NOVO COMPLEX CHROMOSOME REARRANGEMENT AND POSTNATAL CONFIRMATION IN ABORTUS WITH INTRAUTERINE GROWTH RETARDATION. Hyon J. Kim, Virginia Bogosian, Mary Ann Perle, and Alba Greco. (Spon. by Kurt Hirschhorn.) Mount Sinai School of Medicine, Beth Israel Medical Center, Department of Pediatrics and New York University School of Medicine, Department of Pathology, New York, N.Y.

A complex chromosome rearrangement, apparently a balanced translocation involving #4, #6, #15 and #16 chromosomes, was found in cultured cells of amniotic fluid from a 32-year-old primigravida who requested amniocentesis for prenatal diagnosis because of a family history of mental retardation. Chromosome analysis of peripheral blood from both parents were normal. The couple was counseled for the prenatal diagnosis of this de novo complex translocation and, subsequently, they decided to terminate the pregnancy. Postmortem examination revealed a 23-week fetus with intrauterine growth retardation and some facial dysmorphism. The identical chromosome rearrangement was found in cultured fibroblasts from skin and cord obtained from the abortus.

Eleven reported cases of complex chromosome rearrangements were reviewed, 6 were found to be de novo in origin and all were ascertained because of an abnormal phenotype. To our knowledge, this is the first case ever reported as a prenatal diagnosis involving 4 or more chromosomes. There are clinical and counseling implications of prenatal diagnosis of an apparently balanced, but de novo complex chromosome rearrangement.

761 DUPLICATION SYNDROME OF THE DISTAL SEGMENT OF 4q (4q25→ 4qter). Hyon J. Kim, Mary Ann Perle, Deborah Vine, Wallace B. Lehman (Spon. by Kurt Hirschhorn.) Mount Sinai School of Medicine, Beth Israel Medical Center, Department of Pediatrics and Hospital for Joint Diseases-Orthopaedic Institute, Department of Orthopaedics, New York, N.Y.

A dysmorphic newborn male infant with multiple congenital anomalies was found to have a structurally abnormal karyotype of 46,XY,8p+. Parental chromosome studies revealed normal karyotypes. Application of high resolution banding studies (G and R) on extended chromosome preparations of this patient enabled us to identify this de novo unbalanced karyotype as 46,XY,-8,+der(8),t(8;4)(p23;q25). This indicates that the patient is trisomic for the distal segment of 4q (4q25→4qter) and possibly monosomic for the very tip of the short arm of #8.

Review of 16 reported cases of partial trisomy 4q resulting from various chromosome rearrangements showed that, in spite of different monosomic involvements in each case, a recognizable pattern of malformations, including characteristic facial features, leads to clinical identification of duplication syndrome of distal segment of 4q.

Identification of de novo chromosome rearrangement and recognition of specific duplication syndrome as a clinical entity in the newborn with multiple congenital anomalies will enable us to provide genetic counseling with appropriate anticipatory guidance to parents and better management and care for such patients.