511 AN X/AUTOSOME TRANSLOCATION IN A GIRL WITH DUCHENNE MUSCULAR DYSTROPHY (DMD): EVIDENCE FOR DMD GENE LOCALIZATION. R.M. Greenstein, M.P. Reardon, Peds.; T.S. Chan, Physiol. UConn Hlth. Ctr., Farmington, Conn.

A 16 year old girl, an only child, was found to have Duchenne Muscular Dystrophy (DMD) on the basis of clinical history and course, elevated CPK and aldolase levels, typical EMG and muscle biopsy histology consistent with a dystrophy picture. A chromosome analysis from her skin and blood revealed a reciprocal X/11 translocation: 46,Xt(X;11) (Xqter→Xp21::11q13→11qter) (11pter →11q13::Xp21→Xpter), i.e., the breakpoint occurred in the X short arm at band p21; there is no evidence for mosaicism. The father's chromosomes were normal (46,XY). The mother's study revealed 46,XX, with 20% breakage and clinically she displayed untreated polycythema vera of perhaps 4 years duration. Autoradiography of the subject's blood cells indicated non-random inactivation of the normal X. The subject exhibited normal 1° and 2° sexual development, including menses. Repeated CPK levels in the family suggests that the mother is not a DMD carrier. Additional genetic markers were determined, including PGK, Xg^a, G6PD and color vision. The occurrence of DMD in this girl may be explained by either the "uncovering" of a spontaneous mutant DMD gene on the translocated X chromosome, while the normal X is selectively inactivated; or that the break in Xp21 has caused a mutation of the normal locus leading to a null gene condition. The latter explanation would localize a regulatory or structural DMD gene to the region of the Xp21 band. Future efforts to identify the DMD gene product may find this X/11 translocation cell line useful.

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512 AN IN VITRO MODEL FOR THE STUDY OF TRANSFORMATION: TEMPERATURE-SENSITIVE (Ts) MUTANT. Phyllis Guinivan and Roger Ladda, Penn State Univ Coll Med, M S Hershey Med Ctr, Dept Anat & Ped, Hershey, PA.

Ts systems provide an opportunity to induce a cell with a defined genotype to express either the transformed or normal phenotype by simply manipulating temperature. Rat kidney fibroblasts transformed by a Ts derivative of the Kirsten murine sarcoma virus grow on agar or a preexisting monolayer at the permissive temperature (32°C) but are unable to grow under these conditions at the nonpermissive temperature (39°C). The purpose of this study was to determine the suitability of this model to study processes accompanying "phenotypic transformation" (normal ⇌ transformed). Growth characteristics (clonal/mass culture), cell size, cAMP levels, and surface morphology were periodically monitored following the manipulation of temperature from 39°C ⇌32°C. Ts, normal and transformed control cells grew slower at 32°C, but attained a saturation density equivalent to cells grown at 39°C. Cell size was inversely related to the growth rate; each cell type was smallest during the period of maximal growth rate and became larger as division slowed and cells reached confluency. cAMP levels were also inversely related to growth rate; all cells growing at similar rates had similar cAMP levels. Scanning EM showed that cells maintained at 32°C became spindle-shaped and "piled up" at low density and low serum concentration while cells at 39°C formed flat, confluent monolayers and required greater serum concentration for growth. This system may provide the means to distinguish cellular changes due to transformation from those common to "normal" cells with similar growth rates.

513 POSSIBILITY OF PRENATAL DIAGNOSIS OF DUCHENNE MUSCULAR DYSTROPHY (DMD). Florence P. Haseltine, Maurice J. Mahoney, John C. Hobbins, and C. Thomas Caskey. Depts. Human Genetics and Obstetrics and Gynecology, Yale Univ. Sch. Med., and Baylor College of Medicine, New Haven, CT and Houston, TX (Spon: Leon Rosenberg)

DMD is an X-linked recessive disease which we have not been able to diagnose prenatally. Creatine phosphokinase (CPK) activity is markedly elevated in the plasma of affected infants postnatally and may be similarly elevated in early fetal life. We are testing this hypothesis by measuring plasma CPK at 18-20 weeks gestation. Plasma samples are obtained by placental venipuncture during fetoscopy or by placental aspiration from preabortion fetuses or fetuses at risk for hemoglobinopathies. When the sample contains amniotic fluid and maternal blood, we determine their contribution to the total CPK activity and calculate the fetal plasma CPK. Our results from 12 pregnancies give a range of 40-140 mU/ml (adult normal 5-85) and less than 8 for amniotic fluid. Plasma samples after prostaglandin abortion in 9 fetuses had CPK activities from 60-380. We have monitored one pregnancy of a possible DMD carrier; the fetal CPK was 52 and the pregnancy is continuing. Determination of CPK activities in the fetuses of obligate carriers will establish the validity of this diagnostic approach.

514 CYTOLOGIC EVIDENCE FOR POSITION EFFECT IN BURKITT'S LYMPHOMA. Frederick Hecht, Barbara Kaiser McCaw. University of Oregon Health Sciences Center, Department of Pediatrics, Portland.

Background: Tumor cells differ from normal cells not by having new genes, but by showing inappropriate gene expression. Chromosome rearrangements in other organisms often alter gene expression by "position effect."

Findings: Burkitt's lymphoma cells have a consistent translocation from chromosome 8 to 14 (McCaw et al., Int. J. Cancer, in press). Densitometric analysis disclosed neither a gain or loss of genetic material. We then applied R- and Q- banding techniques and found that in Burkitt's the segment of 8 translocated to 14 stained paradoxically. For example, with acridine orange, the ends of 8 and 14 are red, indicative of denatured single-stranded DNA. However, in Burkitt's the segment of 8 translocated onto the 14 stains green, signifying native double-stranded DNA.

Implications: This is the first cytologic evidence for "position effect" in human cells. It provides a mechanism for inappropriate gene action in tumor cells.

515 BIOTIN-DEPENDENT PROPIONIC ACIDEMIA: A NEW VARIANT. Richard E. Hillman and Julian C. Williams. Washington Univ. Sch. Med., Dept. of Ped., St. Louis Children's Hospital, St. Louis.

Gompertz et al reported a case of biotin-dependent propionic acidemia and β-methyl crotonic acidemia now thought to have a defect in biotin ligase. We report a second biotin dependent case with only propionic acidemia.

The patient had pernicious vomiting and poor growth. Serum glycine and NH_3 were elevated on only single occasions. Serum propionate concentrations (PA) were 40 and 130 μM (norm <5). Urine tiglic acid and butanone were detectable. Clinical response to biotin (10 mg/day) and a low protein diet (1.2 gm/kg) was rapid. While on the diet biotin was stopped. PA rose in 48 hrs but did not peak for 6 weeks. (Day 0, 4.5 μM; day 5, 12 μM; day 7, 16 μM; day 46, 40 μM) During this period urine propionate excretion increased over 400 fold. In response to a minor infection, the child became acidotic, hypoglycemic, and comatose. She recovered with fluids and biotin and has remained normal since. PA returned toward normal. At no time were serum β-methyl crotonate or urine β-OH isovaleric or β-methylcrotonylglycine increased. In leukocytes, before receiving biotin, isoleucine oxidation was reduced; 4 wks off biotin propionyl CoA carboxylase was in our low normal range and stimulated 3 fold with biotin. In fibroblasts grown in biotin containing medium, propionyl CoA carboxylase was 26% of normal controls. β-Methyl crotonyl CoA carboxylase was normal in both leukocytes and fibroblasts.

These findings are most consistent with a biotin binding defect to propionyl CoA carboxylase.

516 THE NUMBER OF C-BANDS OF HUMAN ISOCHROMOSOME Xqi AND THEIR RELATIONSHIP TO 45,X MOSAICISM. Lillian Y.F. Hsu, Sophie Paciuc, Karen David, Ralph Moloshok and Kurt Hirschhorn, Dept. of Peds., Mt. Sinai Sch. of Med., NYC.

It is known that X isochromosome (Xqi) is frequently associated with 45,X mosaicism. Of 35 cases with Xqi collected by Ferguson-Smith, 23 cases were mosaic for 45,X/46,XXqi. Cases with 45,X/46,XXqi/47,XXqiXqi are also known. Apparently a structurally abnormal chromosome has a tendency for either anaphase lag or mitotic nondisjunction. With C-banding, two major types of human isochromosome X have been identified, namely Xqi with one C-band and Xqi with two C-bands. Since chromosomes with two centromeres are known to be unstable, it is possible that Xqi's which appear to have only one centromere but have two C-bands are more unstable during mitosis than Xqi with one C-band. We have studied six cases of Xqi. Three showed one C-band; all these were without 45,X mosaicism. The other three cases showed two C-bands and were mosaic (one 45,X/46,XXqi; two 45,X/46,XXqi/47,XXqiXqi-identical twins). Seven other cases of isochromosome Xqi have been reported with C-banding studies. Of these, four showed one C-band with XO mosaicism in two; three demonstrated two C-bands and all three were XO mosaics, thus in a total of 13 cases of Xqi studied with C-banding, seven showed one C-band with XO mosaicism in 2 cases, one of which showed no XO cells in skin fibroblasts; six Xqi's had two C-bands and all were mosaic for 45,X/46,XXqi or 45,X/46,XXqi/47,XXqiXqi. Thus it is evident that Xqi with two C-bands has a greater tendency for anaphase lag or mitotic non-disjunction leading to mosaicism.