

Assignment of human thymidine kinase gene locus to chromosome 17 by identification of its distinctive quinacrine-fluorescence in man/mouse somatic hybrid cells. ORLANDO J. MILLER, PENELOPE W. ALLDERDICE, DOROTHY A. MILLER, WILLIAM R. BREG, and BARBARA R. MIGEON. *Coll. of Phys. and Surg., Columbia Univ., N. Y., N. Y., Yale Univ. Sch. of Med., New Haven, Conn., and Johns Hopkins Univ. Sch. of Med., Baltimore, Md.*

Human chromosomes are preferentially eliminated from man/mouse somatic hybrid cells. Two groups of workers obtained viable hybrids by mixing human cells with a murine cell line lacking thymidine kinase and growing the mixture of cells in a selective medium in which cell survival required the presence of this enzyme. The hybrid cells usually contained a single human submetacentric chromosome, whose size and shape suggested that it was a member of the E-group, either chromosome 17 or 18. This chromosome presumably carries the human thymidine kinase locus.

Chromosomes of the E-group can be readily identified in human cells by their distinctive quinacrine-fluorescence patterns. By applying this technique to metaphase figures from one of the hybrid cell lines studied by Migeon and Miller (*Science* 162, 1005, 1968), we have found that the submetacentric chromosome which is present has the characteristic quinacrine-fluorescence pattern of a human chromosome 17.

Human chromosome identification by differential staining. PATRICE R. CHERNAY, LILLIAN Y. F. HSU, and KURT HIRSCHHORN. *Mt. Sinai Sch. Med., City Univ. N. Y., N. Y.*

We have employed the differential staining technique of F. Arrighi and T. C. Hsu (1971) to identify individual human chromosomes. After pretreatment with RNAase, the DNA is denatured with NaOH, renatured with saline citrate buffer and then stained with Giemsa. Our studies have shown that in addition to the most distinct densely staining area of the distal 2/3 of the long arm of the Y chromosome, the Nos. 1, 3, 9, 11, 16 and 17 chromosomes carry a densely staining area on the long arm adjacent to the centromere, most noticeable in No. 1; No. 18 has a densely staining area on the short arm, close to the centromere. Studies of patients with trisomy 21 and trisomy 13 demonstrated that chromosomes No. 21 and 13 are identifiable. The three No. 21 chromosomes showed densely stained centromeres and two Nos. 22 showed lightly stained centromeres. The three No. 13 chromosomes in trisomy 13 had dense staining at the centromeres and on the long arms in comparison to those of Nos. 14 or 15. The basis of this differential staining technique is that renatured DNA appears better able to combine with stain than partially denatured DNA. It is apparently the repetitive DNA associated with constitutive heterochromatin which renatures most rapidly and is stained most densely. This method may be very useful in identification of structural as well as numerical chromosomal aberrations.

The origin of some bone marrow fibroblasts. KURT HIRSCHHORN, JEAN HENTEL, and JESSICA W. GRANT. *Mount Sinai Sch. of Med., City Univ. New York, N. Y., N. Y.*

An attempt was made to determine the origin of bone marrow fibroblasts which almost always appear when bone marrow aspirates and explants are grown on solid surfaces in tissue culture. Bone marrow aspirates from two individuals with chronic myelogenous leukemia demonstrating the Ph-1 chromosome and from an individual with acute leukemia demonstrating the trisomic C-group karyotype served as sources of the fibroblasts. These

were analyzed for the presence of the marker chromosomes found in the leukemic cells of these patients. Over half of the dividing fibroblasts demonstrated the marker chromosomes. This positive finding indicates that at least some bone marrow fibroblasts are derived from hemopoietic stem cells. These cells should, therefore, prove useful in the study of cellular differentiation.

The value of fluorescence microscopy in studying abnormalities of G group chromosomes. LESTER WEISS and MARILYN DULLY. *Henry Ford Hosp., Detroit, Mich.*

Caspersson et al demonstrated that chromosomes stained with quinacrine mustard and examined under ultraviolet light had distinctive patterns of fluorescence. This technique has been used to study the G group chromosomes from 25 individuals.

The pattern of fluorescence of the G group chromosomes from 10 normal individuals was determined. The very bright fluorescence on the distal end of long arm of the Y chromosome, as described by others, was apparent. The 4 autosomes could be separated into 2 distinct pairs. One pair had a broad band of fluorescence encompassing 2/3 of the proximal long arm. The second pair had a small area of increased fluorescence in the region of the centromere and short arm. The trisomic chromosome in 9 patients with Down's syndrome was the one with a broad band of fluorescence on the proximal 2/3 of the long arm. Chromosome #21 is smaller than chromosome #22. In a family with a G group marker, the fluorescence technique made identification of the marker chromosome, as #21, possible.

Chromosomes from 2 phenotypic males with XX sex chromosomes were examined. The brightly fluorescent region of the long arm of a Y chromosome was not found translocated on any part of the genome. These data plus morphologic considerations indicate that if any Y material were present, it could only be short arm DNA. This is further evidence for male determinants being located on the short arm of the Y chromosome.

Quinacrine mustard staining and UV microscopy is a new technique that enables us to identify specific chromosomes and regions within chromosomes.

## CARDIOLOGY

Assessment of systemic and pulmonary baroreceptor function in intact and unanesthetized fetal and newborn lambs. ELLIOT SHINEBOURNE, EERO VAPAAVUORI, ROBERT WILLIAMS, MICHAEL HEYMANN, and ABRAHAM RUDOLPH. *Cardiovas. Res. Inst., Univ. of California, San Francisco, Calif.*

Baroreceptor responses have been observed in exteriorized fetal lambs, but there have been no quantitative studies of changes with maturation in fetuses in utero. In 9 fetal and 3 newborn lambs an inflatable balloon catheter was passed from the femoral artery into the descending aorta. In 7 other fetuses inflatable balloons were placed around the pulmonary artery (PA) or the aortic isthmus. Vinyl catheters were positioned in a brachial or carotid artery, and catheters and ECG leads were exteriorized. Arterial pH, PCO<sub>2</sub> and PO<sub>2</sub> were normal in all studies. Reflex bradycardia in response to blood pressure elevation by balloon inflation was measured repeatedly for several weeks. Baroreceptor sensitivity was expressed as the regression coefficient of the beat-to-beat relationship between systolic (SP) and pulse (PP) pressure, and the subsequent R-R interval (R-R) or heart rate (HR). In over 100 observations we found: (1) elevation of systemic but not main PA pressure elicits reflex bradycardia; (2) reflex bradycardia could be elicited in all animals but was frequently absent in fetuses less