Quinacrine Fluorescence of the Human *Y* Chromosome

RECENT observations[1-4] of specific localized fluorescence in *Y* chromosomes stained with quinacrine mustard and quinacrine hydrochloride have provided a new parameter for the study of abnormal *Y* chromosomes. We have re-examined regularly prepared chromosome and buccal smear slides of at least 100 cells from two patients with a small *Y* chromosome[5] and from patients with normal *Y* chromosomes. We used a modification of techniques described earlier[1-4]. We stained for 5 min in 0.5% quinacrine hydrochloride (Winthrop); washed in distilled water; differentiated for 3 min in citric acid–phosphate buffer, *p*H 5.5; washed and mounted slides in phosphate buffer, *p*H 7.4, and sealed with nail polish. Preparations were examined with a Leitz Ortholux microscope with a mercury arc lamp, standard fluorescence filters, and dark-field illumination, and were photographed with Ektachrome high speed film. Fluorescence persisted for several days. Full details of the technique will be published later.

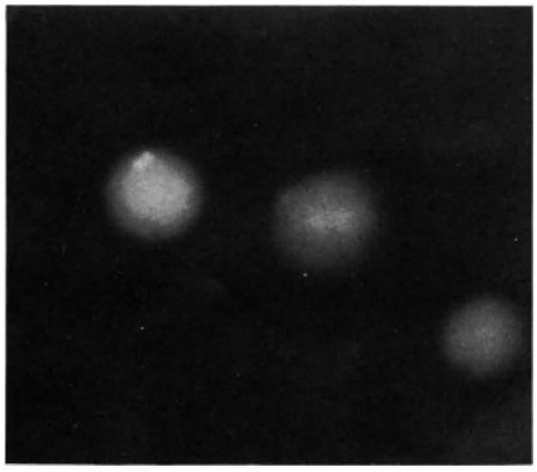

Fig. 1 Interphase nucleus showing two adjacent fluorescent bodies.

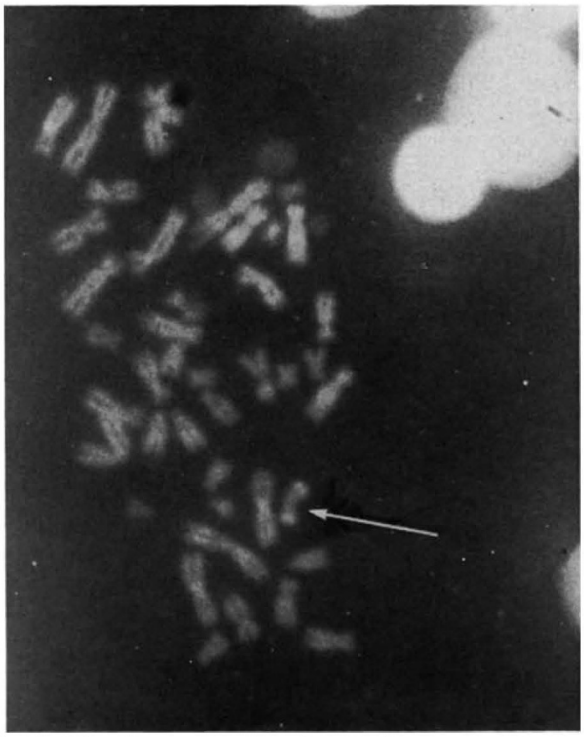

Fig. 2 Metaphase spread showing two fluorescent regions on the dicentric *Y* chromosome.

The distal portion of the long arm of the normal *Y* chromosome fluoresced clearly and distinctly in metaphase plates and a corresponding fluorescent particle was present in many interphase nuclei, but no comparable fluorescence was detected in preparations from patients with a small *Y* chromosome. This may be because of deletion of the heterochromatic region in the long arm.

We have also investigated a patient with a dicentric chromosome, and found two adjacent fluorescent spots in interphase nuclei (Fig. 1), and could see two fluorescent spots on both the distal ends of the dicentric *Y* chromosome (Fig. 2). It will be interesting to study patients with an iso-chromosome of the long arm of *Y* with this technique. One could expect to see two fluorescent regions in these patients.

We have previously shown that fertility and other sexual characteristics are normal in men with a small *Y* chromosome[5]. The observation described here indicates that maleness is not dependent on the presence of a fluorescent region in the long arm of the *Y* chromosome. Caution may therefore have to be exercised in using the quinacrine fluorescence technique for determination of the *Y* chromosome[6].

We thank Mrs Ruth Classon and Miss Betsy Hill for technical assistance. We also thank Drs Hope Punnett and Kistenmacher, Children's Hospital, Philadelphia, for allowing us to study the slides of their patient with the dicentric *Y* chromosome. This work was supported in part by a grant from the US Public Health Service.

DIGAMBER S. BORGAONKAR

Division of Medical Genetics, Department of Medicine,

DAVID H. HOLLANDER

Division of Cytopathology, Department of Pathology, Johns Hopkins University, School of Medicine, Baltimore, Maryland 21205

Received July 9; revised November 11, 1970.

1 Zech, L., *Exp. Cell. Res.*, **58**, 463 (1969).
2 Caspersson, T., Zech, L., Modest, E. J., Foley, G. E., Wagh, U., and Simonsson, E., *Exp. Cell. Res.*, **58**, 141 (1969).
3 Pearson, P. L., Bobrow, M., and Vosa, C. G., *Nature*, **226**, 78 (1970).
4 George, K. P., *Nature*, **226**, 80 (1970).
5 Borgaonkar, D. S., McKusick, V. A., Herr, H. M., de los Cobos, L., and Yoder, O. C., *Ann. Genet.*, **12**, 262 (1969).
6 *Nature*, **226**, 897 (1970).

Use of the *Y* Chromosome in Prenatal Sex Determination

REPEATED observations of the cellular composition of human endocervical mucus from the earliest weeks of pregnancy, using various staining techniques, have revealed not only cells of maternal origin, but also chorionic cells which have been shed into the fairly static cervical mucous plug.

I have used the fluorescein dye test[1-4] on smears taken with a cotton swab from the mid-cervical mucus of thirty patients in the first, second and third trimesters of pregnancy. In eighteen cases quinacrine hydrochloride revealed in a proportion of the interphase chorionic nuclei a single fluorescent spot, indicative of *Y* chromosome material, and thus of a male conceptus. In the other twelve patients smears developed no fluorescent spot, indicative of a female conceptus. Six of the eighteen have now been delivered of boys and four of the twelve have produced girls. These observations are being extended.

LANDRUM B. SHETTLES

The Presbyterian Hospital, 662 West 168th Street, New York 10032

Received September 28, 1970.