crossing-over. Alternatively, a single site on only one of the two non-sister chromatids may initiate crossing-over. Models to explain crossing-over in terms of repair or error correction have been proposed by Whitehouse (1963) and by Holliday (1964) and events at stage two may well follow one or other of these schemes.

SUMMARY

1. A comparison between the recombination frequencies in zygosporangia irradiated during both stage one (preleptotene) and stage two (late zygotene-pachytene) and those irradiated in stage one only, suggests that irradiation at stage one does not inhibit chromosome pairing.

2. The results provide indirect support for the two-step model of crossing-over (Lawrence, 1967*a*, *b*).

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SPLIT AND ENLARGED SATELLITES IN MAN

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1. INTRODUCTION

ALL the five pairs of acrocentric chromosomes in man are satellited (Ferguson-Smith and Handmaker, 1961; German, 1964). Peculiarities in the size and structure of satellites have been reviewed by De La Chapelle

(1963) and been found subsequently in clinically normal and abnormal individuals by Handmaker (1963), Jacobson *et al.* (1964) and Bartfield and Morrill (1965).

In the course of our study of chromosome complements in normal as well as abnormal individuals in India, a certain peculiarity, hitherto unreported, in the structure of the satellite has been encountered in a normal male individual.

2. MATERIAL AND METHOD

The observations were made on preparations from cultures of whole blood obtained from finger pricks. The blood was added into a universal container containing 5 ml. of Parker 199, 1 ml. of AB plasma and 0.15 ml. of Difco Phytohaemagglutinin (M). The same procedure was followed in subsequent cultures of leucocytes from the same person on different dates.

TABLE 1

The incidence of quadripartite satellites in a sample of 64 cells

Cell No.	Chromosomes with split satellites	
	Group D	Group G
1	—	1
2	—	1
3	2	—
4	1	—
5	1	—
6	1	—
7	—	2
8	—	1
9	—	1
10	—	1
11	—	1

3. RESULTS

Two types of peculiarities in the structure of the satellites in the acrocentric chromosomes of both the groups D and G have been noticed.

1. *Split satellites.* Some of the acrocentric chromosomes in a few cells have quadripartite satellites instead of the usual bipartite condition (plate I, 1 and 2). Each satellite-sector is separately connected by means of a thin strand with the chromatid of the short arm of the chromosome. The split nature of the satellites and their stalks is more distinct in the acrocentrics of the G group. When two acrocentrics belonging to the same group carry split satellites in the same cell, it is rather difficult to say whether they are homologous or not.

Eleven out of 62 metaphase cells in the first culture had split satellites (table 1). In two cells, however, one acrocentric of the G group had three satellites (plate I, 3). Such a condition may be due to either the fusion of two satellites in a 4-part structure or the non-splitting of one of the two normal satellites.

2. *Enlarged satellites.* Acrocentrics of groups D and G are also found with enlarged satellites (plate I, 4 and 5). They stain deeply, sometimes more deeply than the main body of the chromosomes. Often these satellites are bent as if the stalks were unable to support their weight (plate I, 5).

Further, two satellites of different sizes may be present in the same chromosome (plate I, 4). Plate I, 6 shows a chromosome with only one satellite. Seventeen of the 62 metaphase cells in the first culture have enlarged satellites, in 22 cells they are not conspicuous and in the rest satellite size is normal (table 2).

Three further cultures were subsequently studied under similar conditions. In the second culture only 5 good metaphase cells were obtained. In one cell one chromosome of the G group had split satellites and in the same cell, one chromosome of the D group had enlarged satellites. In the third

TABLE 2
The incidence of enlarged satellites in a sample of 64 cells

Cell No.	Satellite size					
	Two large		One large and one normal		One large only	
	Group D	Group G	Group D	Group G	Group D	Group G
1	1	—	—	—	1	—
2	2	—	—	—	—	—
3	1	1	—	—	—	—
4	1	—	—	—	—	—
5	1	—	—	—	—	—
6	—	—	—	—	1	—
7	—	—	—	—	1	—
8	—	—	—	—	1	1
9	3	—	—	—	—	—
10	4	2	—	—	—	—
11	2	—	1	—	—	—
12	—	—	—	—	1	1
13	1	—	—	—	2	—
14	1	—	—	—	—	—
15	1	—	2	—	—	—
16	2	1	—	—	—	—
17	—	—	—	—	3	1

culture out of 42 metaphase cells, 5 had normal satellites and the remaining 37 cells had no conspicuous satellites. In a sample of 52 cells from the fourth culture, the satellites were clear and normal in 25 and in the remaining 27 cells they were not conspicuous. Usually in our preparations the satellites are clear in most of the cells and of the size shown in Plate I, 7.

4. DISCUSSION

Since these observations could not be repeated in subsequent cultures, the abnormalities must have been due to undefined conditions in the first culture. Even so the observation raises the important issue of the number of chromatids in a chromosome and the variability in the structure of the satellites.

Although there are many claims for the presence of subchromatids in animal and plant chromosomes, there has been no previous evidence for their presence in human chromosomes. But the split stalks and satellites reported here may be an indication of a half-chromatid organisation.

Enlarged satellites occur but not universally in Marfan's syndrome (Tjio *et al.*, 1960; Handmaker, 1963). They are also found in normal

- 1.—Chromosome of the G group with four satellites.
- 2.—Chromosome of the D group with four satellites.
- 3.—Chromosome of the G group with three satellites.
- 4.—Chromosome of the D group with one large and one small satellites.
- 5.—Chromosome of the D group with two large satellites.
- 6.—Chromosome of the D group with one large satellite.
- 7.—Chromosomes with normal satellites.

individuals (see De La Chapelle, 1963). The present investigation shows that it may also occur sporadically due to unknown causes under apparently well controlled culture conditions.

5. SUMMARY

1. In a leucocyte culture from a normal male, some of the acrocentric chromosomes of groups D and G have quadripartite satellites with four separate stalks.

2. Some of the acrocentric chromosomes of both groups also have enlarged satellites. The structure of the satellites varies in different cultures made from the blood of the same individual.

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THERMO-ELECTRIC COOLING FOR MAKING PERMANENT CYTOLOGICAL PREPARATIONS *

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THE rapid freezing of cytological preparations for the removal of coverslips is widely recognised as quick, reliable and superior to the traditional cytological procedures. However, the necessity for either "dry ice" (Conger and Fairchild, 1953) or liquid carbon dioxide (Bowen, 1955) to produce the low temperature required has, on occasions, restricted the use of the technique.

* Demonstrated at the Second Oxford Chromosome Conference, held at the Botany School and Magdalen College, Oxford, 5th to 8th September 1967.