

CYTOLOGY

Chromosomal Constitution of Human Endometrium

It is generally assumed that there is a diploid number of chromosomes in nearly all human somatic cells. That this is not true for the endometrium was first reported by Timonen¹ in 1950. He found chromosome numbers ranging from 4 to 104 in 1,000 cells of human endometrium. Manna² observed similar aneuploidy in this tissue. Sachs³, on the other hand, found no variation from the normal diploid number in either human or rat endometrium. Walker and Boothroyd⁴, although they found chromosome counts ranging from 42 to 56, believed that the apparent aneuploidy was caused by the methods used, and suggested that Timonen's results were similar artefacts produced by the technique. Since all these investigators used squash preparations without previous hypotonic treatment, it is quite possible that in some cases accurate counts were not possible or that fragmentation occurred. Furthermore, the human diploid chromosome number was, at that time, believed to be 48, and this erroneous belief may also have biased the results obtained. Although new methods were introduced into human cytogenetics and the quality of the preparations was greatly improved, the question of aneuploidy in the endometrium has remained unanswered. In 1958, Tjio and Puck⁵ reported that they had found only diploid and some tetraploid cells in endometrium grown in tissue culture. Takemura⁶, however, found that of 109 cells only 51.4 per cent had 46 chromosomes, the chromosome counts in the others ranging from 33 to 95.

The purpose of our investigation was to establish the chromosomal pattern of the human endometrium as a part of a study concerned with reproductive disorders. The endometrium was obtained by biopsy from 28 women of child-bearing age, including seven normals, 14 with a history of sterility and seven with a history of repeated abortions. Of these, seven represented the proliferative and 21 the secretory phase of the menstrual cycle. The tissue was cultured *in vitro* after a modification of the method described by Chu and Giles⁷. In most cases primary cultures were used for chromosome preparations. Colchicine in a final concentration of 2–3 µg/ml. was added to the cultures 16 h before harvesting. The cultures were then trypsinized, and the cells were treated with 20 per cent Hanks's balanced salt solution in distilled water for 20 min, fixed, allowed to air-dry on slides, and stained with dilute Giemsa stain. All metaphase spreads of adequate technical quality were counted, and every attempt was made to exclude broken cells.

Chromosomes were counted in a total of 449 mitoses, with the counts ranging from 17 to 103 chromosomes per cell. 119 of the cells counted had the normal number of 46 chromosomes. The distribution of the counts is given in Table 1. Aneuploidy was found in every culture. No chromosomes with abnormal morphology were observed.

Table 1. DISTRIBUTION OF CHROMOSOME COUNTS IN 449 CELLS OF HUMAN ENDOMETRIUM

Chromosomes/Cell	No. of cells
14–18	2
19–23	2
24–28	10
29–33	14
34–38	48
39–43	117
44–48	208
49–53	20
54–58	4
59–63	5
64–68	2
69–73	3
74–78	4
79–83	4
84–88	4
89–93	—
94–98	1
99–103	1

These results support the findings of high degrees of aneuploidy in the human endometrium. The chromosome counts are similar to those obtained by the earlier workers, all of whom used the direct squash method without tissue culture in their investigations. This indicates that the conditions of *in vitro* growth, as used in this work, do not significantly alter the chromosome complement or result in selective growth of cells with a given karyotype. Similarly, the only difference between our preparations from primary cultures and sub-cultures has been a higher mitotic rate in the latter.

It seems clear that there is aneuploidy in the human endometrium, and that aneuploidy is, indeed, normal for this particular tissue. Since aneuploidy is believed to be the result of certain abnormalities of either meiosis or mitosis, and since endometrium is a very rapidly growing tissue, it is likely that the high degree of aneuploidy observed in the endometrium reflects in some way the normal growth and development of the tissue itself. If this is true then the degree of aneuploidy would be expected to vary with the menstrual cycle. Our preliminary results suggest that this is, indeed, the case.

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⁶ Takemura, T., *J. Jap. Obstet. Gynec. Soc.*, **7**, 300 (1930).
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SOIL SCIENCE

Influence of Ionic Phosphate Diffusion in the Soil on the Growth of *Hormidium flaccidum*

A KNOWLEDGE of the ionic mobility of the essential plant nutrients in the soil is of considerable importance in determining the method and effect of particular fertilizer programmes in the eventual crop growth. Scott Russell and Newbould¹ and Schofield² have indicated the importance of this and the difficulties which lie behind the investigation of it. Calcium and phosphate would, for example, directly enter into exchange reactions in the soil so that the rate of ionic mobility would become limited by the degree of this interaction. Newbould¹ showed that calcium could reach a state of equilibrium in the exchange system of the soil within seven days; Fried and Shapiro³ elaborated the factors affecting the mobility of ions by demonstrating that both the water movement in the soil, and the addition of inert material to the soil, would accelerate the diffusion of ions. Most of this work has been carried out using labelled isotopes. However, Scott Russell¹ has indicated that, even using these techniques, the knowledge of the mechanisms of diffusion was still meagre.

During an investigation of a new method of soil bio-assay using algae I found⁴ that *Hormidium flaccidum*, when impregnated into nutrient-deficient soils, would give, by way of its growth, indication of the distance and of the rate at which nutrient ions may diffuse in the soil.

In the method used, 10 g of dried, sieved soil with a known phosphate deficiency was added to a sterile standard bacteriological dish. An inoculum containing 5×10^6 cells/ml. of *Hormidium flaccidum* was mixed with the soil until it became saturated to a mud-like paste. To encourage algal growth throughout the soil, the soil was then incubated at $25^\circ \pm 1^\circ \text{C}$ for 7 days at an even