Barrier around Synovial Cells *in vitro*

In the course of experiments designed to study the interaction of lymphocytes with synovial cells grown *in vitro*[1], we have found an optically clear zone beyond the cytoplasm of the synovial cells which appears to prevent contact between the two types of cell. This zone (Fig. 1) may extend laterally at least 20μ beyond the cell borders, and, judging from the appearances after a denser inoculum of blood cells, also extends in the vertical plane. The barrier can be more conveniently demonstrated with washed red blood cells. It is generated within a few hours of the attachment and spreading of synovial cells from a trypsin-dispersed cell suspension in culture medium, and persists at least 5 days. The zone has also been shown by adding a suspension of indian ink or molybdenum disulphide (average particle size 0·5μ), but within 12–20 h these substances pass through and appear within the cell boundaries. The phenomenon has consistently been found in several strains of human synovial cells isolated in this laboratory, but has been much less pronounced in a strain of human embryonic fibroblasts. Preliminary studies with bacterial hyaluronidase suggest that the barrier is largely composed of hyaluronic acid (Fig. 2).

Direct contact appears to be essential for the demonstration of specific reactions between tissue cells and sensitized lymphocytes[2–4]. The intensity of these reactions has been attributed to the proportion of immunologically competent lymphocytes[5] and to the antigenic pattern of the target cell[6]. It would appear that the pericellular zone might, in some cell strains at least, provide a further important determinant of such reactions.

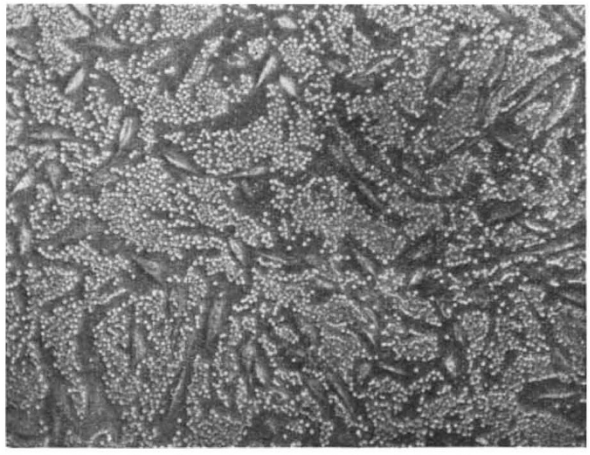

Fig. 1. Replicate culture of synovial cells inoculated with a suspension of washed human red blood cells (group O).

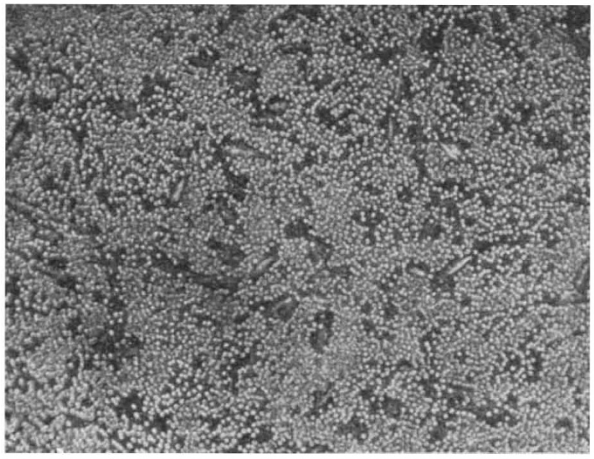

Fig. 2. As in Fig. 1, but the culture was treated with bacterial hyaluronidase (10 T.R.U./ml.).

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[1] Fraser, J. R. E., and McCall, J. H., *Ann. Rheum. Dis.*, **24**, 351 (1965).
[2] Rosenau, W., and Moon, H. D., *J. Nat. Cancer Inst.*, **27**, 471 (1961).
[3] Koprowski, H., and Fernandes, M. V., *J. Exp. Med.*, **116**, 467 (1962).
[4] Wilson, D. B., *J. Cell. Comp. Physiol.*, **62**, 273 (1963).
[5] Wilson, D. B., *J. Exp. Med.*, **122**, 143 (1965).
[6] Möller, E., *Science*, **147**, 873 (1965).

Gene Activity dependent on Chromosome Synapsis in the Polytene Chromosomes of *Drosophila melanogaster*

One of the more intensively studied phenomena used in investigating the control of gene activity in higher organisms is the "puffing" of the polytene chromosomes of Diptera, a phenomenon which has been shown to be an expression of gene activity[1]. The changing pattern of puffs during larval and prepupal development in *Chironomus* and *Drosophila* is influenced, to a certain extent, by the hormone ecdysone[2,3]. There is evidence that the effect of this hormone is indirect, acting by way of a control of the ionic composition of the cell and nuclear sap[4–6].

Genetically determined variation in puffing activity has been analysed in only a few instances[7–9]. I have recently found in *Drosophila melanogaster* that the laboratory "wild" stock Oregon-*R* and Thoday's selection line *vg*6 differ in some aspects of their pattern of puffing activity during the late third instar larval and prepupal periods. One aspect of these differences is that in *vg*6 I find a puff at 64*C* (notation of P. N. Bridges[10]) on the left arm of the third chromosome, whereas a puff at this location has never been found in the Oregon stock, nor in a number of other wild and laboratory stocks examined. Neither is it found in the Berkeley wild stock of the sibling species of *D. simulans*.

In the *F*1 hybrid between Oregon and *vg*6 the puff at 64*C* can be seen on both the homologous chromosomes when there is full synapsis at this region, but on only one of the homologues when there is asynapsis. The size of the puff in synapsed hybrids is never as large as that seen in the *vg*6 parent stock. The frequency of asynapsis in these *F*1 animals is very low (it was only seen about ten times in many hundred nuclei examined). The Payne inversion covers the relevant region of chromosome 3 and in order to extend the evidence use was made of the considerable asynapsis that occurs in the inversion heterozygotes. In Oregon/Payne *F*1 larvae no puff at 64*C* was ever found, regardless of the synaptic condition of the homologous polytene chromosomes (Figs. 1 and 2). In the *vg*6/Payne *F*1, however, both homologues were puffed when they were synapsed (Fig. 4), but only the homologue from the *vg*6 parent (that is, the homologue showing a standard banding sequence) puffed when the homologues were unpaired over the critical region (Fig. 3).

This situation is quite unlike that found by other workers[11–15] in puff heterozygotes in chironomids and *Sciara*. These authors found, in such heterozygotes, that only one of the homologous polytene chromosomes was puffed at a particular locus, despite complete chromosome synapsis. On the other hand, Mechelke[16] has described two strains of *Acricotopus lucidus* only one of which has a particular Balbiani ring. In the heterozygote between them a small puff is found, which is continuous