cytoplasm numerous mitochondria and granules, these are often filled with very dark inclusions (figure 2). In the area where the cilium is found projecting, the plasma membrane folds into 12–13 projections (figure 3) and a cylindrical structure surrounding a portion of the cilium outside the cell surface is observed (figure 4).

Filaments can also be seen on the cell surface, and these appear to project in the coelomic space and to be continuous with the outer leaflet of the unit membrane (figure 5). Below and lining the epithelial layer (figure 6) are found the basal lamina 1 and a connective tissue layer containing collagen fibres. Fibroblasts immediately follow this basal lamina 1 layer. The next layer is made up of muscle fibres and neurons followed by a second basal lamina, a connective tissue layer and a third basal lamina

(the thickness of the laminae 1, 2, and 3 are, approximately and respectively, 300, 200 and 400 nm).

The germinal layer that follows the third basal lamina is composed of cells undergoing spermiogenesis. The mitochondria of early spermatids are round and quite typical and appear as a unique, conspicuous, mitochondrion in late spermatids. Also, the distal centriole is found in the articular fossa. The proximal centriole is also seen and its open side faces the nucleus. It is thus apparent from these observations that the ultrastructure of sea urchin testes shows no obviously important differences with the fine structure of testes reported for other organisms, and that the glutaraldehyde-paraformaldehyde mixture appears to be the most useful fixative for these preparations.

Morphological evidence of a polypeptide-like secretory function of the B cells in the mouse synovial membrane¹

G. Linck, M. E. Stoeckel, A. Petrovic² and A. Porte

Université Louis Pasteur, Institut de Physiologie, Section Sciences, F-67000 Strasbourg (France), 6 January 1977

Summary. In the synovial membrane of the mouse, morphological features associated with active secretion are unusually well developed in cells immediately subjacent to the lining layer (in the position of B cells), comparable to those of cells known to elaborate polypeptides.

6

The intimal lining of the synovial membrane is usually described as comprising 2 cell types, designated A and B cells, although distinctive morphological characteristics of these 2 types are not well defined 3-8. The A cells form the lining layer of the synovial cavity. B cells, which lie under the A cells, have a more highly developed rough endoplasmic reticulum, which suggests that they may secrete proteins 4. Our electronmicroscopic observations

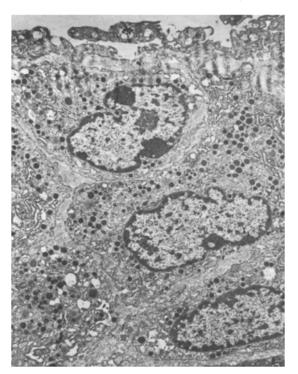


Fig. 1. Cluster of B cells with dense secretion granules under the epithelioïd layer lining the synovial cavity. $\times 6000$.

of synovial membranes in the mouse have revealed that, in this species, ultrastructural evidence of polypeptide synthesis is striking in cells occupying the position of B cells. Cells which show such morphological specialization characteristic of cells primarily involved in polypeptide secretion have not, to our knowledge, been described in any other type of connective tissue.

Methods. The phalangeal, metatarso-phalangeal and knee joints were taken from female and male mice after intracardiac perfusion with 5% glutaraldehyde solution (phosphate buffer 0.1 M, pH 7.4). After 24 h in the same fixative, followed by 5–8 days decalcification in fixative with 0.1 M EDTA, they were divided into sagittal sections, post-fixed in 2% osmic acid and embedded in araldite-epon. Semi-thin sections (1 μm) were colored using several techniques for histological studies. Thin sections were contrasted with uranyl acetate and lead citrate.

Results and Discussion. Under the layer of interdigitated processes of A cells which forms the lining of the synovial cavity, there are many cells, clustered in islets or dispersed, which are characterized by an abundance of small, dense cytoplasmic granules. These cells were never observed in direct contact with the synovial cavity. They are separated from the fibrous stroma by a basal lamina which does not extend into the islets. Although they sometimes present an epithelioid arrangement, individual cells are always separated by a narrow connective space

- 1 This work was supported by grant No 74.1.133.4, Institut National de la Santé et de la Recherche Médicale, France.
- A. P., Institut de Physiologie, Section Médecine, F-67085 Strasbourg Cédex, France.
- 3 E. Langer and F. Huth, Z. Zellforsch. 51, 545 (1960).
- 4 P. Barland, A. B. Novikoff and D. Hamerman, J. Cell Biol. 14, 207 (1962).
- J. C. Wyllie, R. H. More and M. D. Haust, Lab. Invest. 13, 1254 (1964).
- F. N. Ghadially and S. Roy, Ann. rheum. Dis. 25, 318 (1966).
- 7 S. Roy and F. N. Ghadially, Ann. rheum. Dis. 26, 26 (1967).
- 8 P. R. Krey and A. S. Cohen, Arthritis Rheum. 16, 324 (1973).

filled with the fine collagen fibrils. This fine collagenous material is also interposed between these cells and the lining layer, and it contrasts sharply with the fibrous bundles of the surrounding stroma. Fenestrated capillaries are often observed near these dense granule-containing cells.

These cells, which correspond in position to the B cells described in other species, show, in the mouse, evidence of active secretory function. Their rough endoplasmic reticulum is well developed, as is the Golgi apparatus,

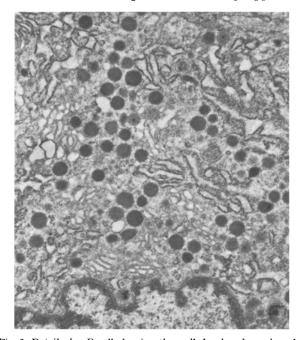


Fig. 2. Detail of a B cell showing the well developed rough endoplasmic reticulum, dense secretory granules and formation of granules by the Golgi complex. $\times 14,300$.

which shows classic images of elaboration of secretion granules: budding off of vesicles which then increase in density to give rise to dense secretory granules, with a mean diameter of 200 nm (max. 220 nm) (figures 1 and 2). It has not yet been possible clearly to visualize the secretory material on histological sections. At best, can one detect a fine intracytoplasmic granularity weakly colored with PAS on semi-thin sections. An absence of metachromasy with toluidine blue and a negative reaction with alcian blue pH 2.4 clearly distinguishes these cells from mastocytes, which are strongly colored by these 2 techniques.

Besides the elaboration of secretory granules, presumably proteins, these cells appear to have a fibroblastic activity, as evidenced by the abundance of fine collagen fibrils which lie around them and even within surface invaginations. The specific secretory activity of these connective tissue cells is probably responsible for the modification of their surrounding stroma. One can only conjuncture regarding the role of the elaborated material. It should be remembered, for example, that hyaluronic acid in synovial fluid is bound to a special protein, whose origin is yet undetermined 9, 10. Also a connective tissue activating peptide' has recently been isolated from synovial tissue 11. In addition, the possibility that these cells are involved in a hormonal regulatory mechanism should be kept open, considering the frequent proximity of B cells and underlying fenestrated capillaries. In any case, it is obvious that these cells play an important and probably specific role in the metabolism of the connective tissue associated with the synovial cavity. The synovial membrane of the mouse provides a particularly useful model for further investigations of function of joint connective tissues, utilizing histochemic and radioautographic techniques.

- 9 J. Sandson and D. Hamerman, J. clin. Invest. 41, 1817 (1962).
- M. J. How, J. W. Long and D. R. Stanworth, Biochim. biophys. Acta 194, 81 (1969).
- 11 C. W. Castor, Arthritis Rheum. 14, 41 (1971).

The effect of concanavalin-A on the reaggregation of cells dissociated from Xenopus laevis early embryos

M. Stanisstreet and J. L. Smith

Department of Zoology, University of Liverpool, Brownlow Street, P. O. Box 147, Liverpool L69 3BX (England), 24 January 1977

Summary. The effects of concanavalin-A on the reaggregation and sorting of cells from Xenopus laevis early embryos have been studied. The results suggest that at high concentrations, concanavalin-A can prevent reaggregation.

Concanavalin-A (Con-A) is a plant lectin which binds preferentially to α -methyl-mannoside and α -methyl-D-glucopyranoside carbohydrate residues¹, and thus it can be used to block these groups on cell surfaces. Such groups could play a part in the control of morphogenesis, since Con-A has been shown to affect a number of embryonic systems such as sea urchin embryos² and chick retinal cells³. The results of a number of recent experiments suggest that Con-A binding residues might have a role in amphibian early morphogenesis too. Embryos of Amblystoma maculatum cultured in Con-A show a slower rate of development than normal, and are blocked at gastrulation⁴. If embryos of Xenopus laevis are exposed to fluorescein-isothiocyanate-labelled Con-A to localize the binding sites, concentrations of label are seen

at the dorsal lip of the blastopore of the gastrula and on the neural folds of the neurula⁵, both regions associated with active morphogenetic movements. Fluoresceinlabelled Con-A has also been used to demonstrate a change in membrane properties of amphibian cells at gastrulation: isolated Rana pipiens blastula cells bind Con-A

- I. J. Goldstein, C. E. Hollerman and E. E. Smith, Biochemistry 4, 876 (1965).
- 2 R. Lallier, Exp. Cell Res. 72, 157 (1972).
- S. J. Kleinschuster and A. A. Moscona, Exp. Cell Res. 170, 297 (1972).
- 4 D. Moran, J. exp. Zool. 188, 361 (1974).
- 5 D. S. O'Dell, R. Tencer, A. Monroy and J. Brachet, Cell Different. 3, 193 (1974).