ULTRASTRUCTURE OF SYNOVIAL MEMBRANE IN OSTEO-ARTHRITIS

BY

S. ROY

Department of Pathology, University of Sheffield

Light microscopy of the synovium in osteo-arthritis reveals either no abnormality or only mild non-specific inflammatory changes in the synovium (Waine, 1958; Cooper, 1964) which may or may not be accompanied by hyperplasia of the synovial cells (Wilkinson and Jones, 1963).

It was considered worthwhile to study the ultrastructure of synovial membrane in osteo-arthritis to elucidate any alteration in its fine structure and to compare the changes with those found in other disease conditions.

Material and Methods

Synovial membrane was collected at operation from the knee joint of eight cases of osteo-arthritis during amputation or surgical exploration of the knee. These were divided into Grades I-IV according to the degree of fibrillation of articular cartilage as suggested by Collins (1949). There were two cases in each of the four groups. Six of eight patients had a history of painful knee of 1 to 5 months' duration without any definite pre-operative diagnosis. Osteo-arthritis was found to be present in these cases at surgical exploration. In the remaining two cases amputation was performed for gangrene of the lower third of the leg and foot; there was no history of any disease relating to the knee joints in these 2 cases and osteo-arthritic fibrillation was found only on opening the joint. Synovial membrane was removed before the actual amputation was done.

The tissue collected was immediately fixed in cold buffered osmium tetroxide (Palade, 1952) and processed according to the method described before (Ghadially and Roy, 1966). Ultrathin sections were cut with the Porter Blum microtome, mounted on copper grids, and examined under the A.E.I. E.M.6 using an accelerating voltage of 50 or 70 kV. An adjacent piece of synovium from every case was fixed in 10 per cent. formalin for routine light microscopic examination.

Results

Light Microscopy

Synovial membrane appeared normal in three and showed a mild lymphocytic infiltration in four cases (Fig. 1). Focal proliferation of synovial cells and patchy areas of moderate or marked lymphocytic infiltration were seen in only one instance.

Fig. 1.—Light microscopic appearance of synovial membrane in osteo-arthritis. Grade III × 310.
Electron Microscopy

The majority of the synovial lining cells contained both Golgi and rough endoplasmic reticulum (RER) and could hence be classified as intermediate cells (Roy, Ghadially, and Crane, 1966). Pure Type A cells containing prominent Golgi with scanty RER were infrequent, and Golgi apparatus in most of the cells was rather small and, unlike the normal synovial cells, the vesicles constituting most of the Golgi apparatus were smaller in size (Figs 2 and 9). There was also a paucity of smooth-walled vesicles in the cytoplasmic matrix.

Endoplasmic Reticulum.—Dilatation of the cisternae of the RER was a prominent feature in all the cases studied. Fig. 2 shows three synovial cells with a clear-cut though mild degree of cisternal dilation. A more severe example is demonstrated in Fig. 3 (opposite). Here the cisternae are grossly dilated and peninsular or papilliferous projections of the RER extended into these grossly dilated spaces. In most instances the cisternae were filled with an electron lucent material containing a scanty precipitate of finely granular or fibrillary material. Dilatation of the cisternae was very

Fig. 2.—Low-power view of synovial membrane in osteo-arthritis, showing dilatation of cisternae (C) of the rough endoplasmic reticulum. Note small size of Golgi apparatus (G). Grade IV × 14,000.
marked in six of the eight cases. There was however no strict correlation between the severity of this change and severity of the disease. Thus one case of Grade I osteo-arthritis showed marked dilatation of the RER cisternae.

Lysosomes and Cytolysomes.—The synovial cells were frequently seen to contain electron dense lysosome-like bodies. At times many such lysosomal bodies could be seen in a single cell (Fig. 4, overleaf).

They were single membrane bound and contained markedly electron dense granules of variable sizes.

Single or double membrane bound bodies containing membranes and granules could sometimes be demonstrated (Fig. 5, overleaf). These can be interpreted as cytolysomes arising in an area undergoing focal cytoplasmic degeneration.

Increase in lysosomal bodies and cytolysomes was clear cut in the six more severe cases (Grade II to IV) of osteo-arthritis, while in both Grade I cases the increase was minimal.
Fig. 4.—Synovial cell containing many lysosomes (L). Grade IV × 18,750.

Fig. 5.—Synovial cell, showing many cytolysomes (C). These contain membranous (M) and granular (G) material within them. The nucleus (N) of the synovial cell is seen. Grade IV × 25,300.
Mitochondria.—In many cells mitochondria were similar to those seen in normal synovium, while in others mitochondria abnormality was quite clearly seen. Commoner abnormalities were mal-orientated cristae, curled up to form arches or circles (Fig. 6).

Swelling of mitochondria with shortening of cristae (Fig. 7) and giant mitochondria (Fig. 8) were less frequently encountered.

Mitochondrial abnormality was regularly seen in more advanced cases of osteo-arthritis (Grade II to IV) but was infrequent in Grade I.
Fine Fibrillary and Granular Material in Cytoplasm.—In three cases belonging to Grade II, III, and IV respectively, many synovial cells showed an abundant amount of very pale finely granular or fibrillary cytoplasm (Fig. 9). The organelles in such a cell appeared to be fewer than those present in normal synovial cells and were widely separated from each other. The fibrillar material in these cells was very loosely arranged. They, unlike the fine filaments present in rheumatoid arthritis (Ghadially and Roy, 1967), were shorter, thinner, and irregularly arranged in the cytoplasm.

Vascular Endothelium.—In two cases many of the endothelial cells also showed pale granular and fine fibrillar cytoplasmic matrix with separation of organelles (Fig. 10). Loose irregularly-arranged fine filaments were again quite unlike those described in vascular endothelial cells of rheumatoid synovium (Ghadially and Roy, 1967).

Rod-shaped bodies morphologically similar to those observed by us in normal synovium and described in the synovia of rheumatoid arthritis (Highton, Caughey, and Rayns, 1966; Ghadially and Roy, 1967) were seen in some of the endothelial cells in all eight cases (Figs 10 and 11, opposite).

Fig. 9.—Two synovial cells (S), showing very pale granular and fine fibrillar cytoplasmic matrix. Note small size of Golgi apparatus (G). Grade II × 37,000.
Lipid.—Lipid droplets of medium density were only occasionally seen in synovial cells in osteo-arthritis (Fig. 12, overleaf).

Discussion

Dilatation of Endoplasmic Reticulum

A most striking and common abnormality found in the osteo-arthritis synovia was increase in the RER (eight cases) and marked dilatation of its cisternae (six cases). Frequently dilatation was very marked, causing disorganization of the cisternae (Fig. 3). The fact that marked dilatation was seen in one of the two cases of Grade I osteo-arthritis indicated that this abnormality was an early manifestation of osteo-arthritic change in synovial membrane. However, the relationship of this change to the disease process itself is difficult to understand.

The cause of this abnormality is also obscure. Whether the enormous dilation of RER cisternae...
indicates increased protein synthesis, as suggested by Sorenson (1964), in myeloma cells or is a degenerative change, as described in cells under anoxic conditions (Sulkin and Sulkin, 1965), is not clear from this morphological study. With normal or increased vascularity seen in most of the osteoarthritic synovia, it is unlikely that anoxia will play any part in causing damage to the synovial cell. In normal synovial fluid 2 per cent. protein is firmly bound to protein-hyaluronate complex (Sandson and Hamerman, 1962). There is evidence that this protein in hyaluronate-protein complex is not a normal serum protein (Blau, Janis, Hamerman, and Sandson, 1965), and is probably formed by synovial cells (Roy and others, 1966). In rheumatoid arthritis, this bound protein increases to 10 per cent. (Hamerman and Sandson, 1963) and the synovial cells show morphological evidence of increased protein synthesis (Ghadially and Roy, 1967). In osteo-arthritis the total protein content of synovial fluid increases (Ropes and Bauer, 1953). It would be interesting to know whether there is any increase in bound protein in this condition or not.

**Lysosomes and Cytolysomes**

The appearance of large number of lysosomes and cytolysomes in synovial cells was also quite striking. Although they were not as numerous as in rheumatoid synovium (Ghadially and Roy, 1967), they were more frequent than in normal or in traumatic arthritis (Roy and others, 1966). It is interesting to note that increase in such bodies was obvious in the more severe cases of osteo-arthritis (Grade II-IV) while there was no unequivocal increase in Grade I. It is therefore likely that increase in lysosomal bodies in synovial cells in osteo-arthritis is a secondary change.

The presence of cytolysomes in areas undergoing focal cytoplasmic degeneration is seen in many other situations (Novikoff and Essner, 1962; Hruban, Spargo, Swift, Wissler, and Kleinfeld, 1963; Parry and Ghadially, 1965). Thus it appears that similar bodies may occur in cells as a result of various forms of cell injury caused by a variety of agents and are non-specific. This change has been described in synovial cells in rheumatoid arthritis (Barland,
Novikoff, and Hamerman, 1964; Wyllie, Haust, and More, 1966; Ghadially and Roy, 1967) and has been observed by us also in traumatic arthritis (Roy and others, 1966) and in experimental haemarthrosis (Roy and Ghadially, 1966). It is therefore obvious that this abnormality in synovial cells seen in various arthritic conditions is non-specific and secondary to some noxious injury. The nature of the noxious agent in osteo-arthritis is obscure. In this condition another explanation for increase in lysosomal bodies may also be considered. As a result of cartilage erosion some mucopolysaccharides from the cartilage reach the synovial fluid and may be taken up by the phagocytic synovial cells. As a response of non-specific nature, the synovial cells will then form more of these lysosomal bodies to deal with the ingested extraneous material. This explanation is consistent with the idea recently put forward by Hamerman (1966).

Mitochondria

Abnormalities in mitochondria like shortening and malorientiation of cristae and the presence of giant mitochondria as seen in osteo-arthritis have also been observed in rheumatoid synovium (Barland and others, 1964; Ghadially and Roy, 1967). It seems likely that these mitochondrial abnormalities are also non-specific and may be caused by the agent responsible for focal cytoplasmic degeneration and increase in number of lysosomes and cytolysomes. It is worth noting here that mitochondrial abnormality was frequently seen only in severe cases of osteo-arthritis and hence again it is more likely to be a secondary manifestation.

Cellular "Oedema"

Another abnormality observed in the synovial cells in osteo-arthritis was abnormal retention of water. This was manifested by marked electron lucency of the cytoplasm with wide separation of organelles in some of the cases. This cytoplasmic change was marked in three cases and was also present to some extent in others. The mechanism of this phenomenon is obscure. Two possible explanations may be considered: abnormality in membrane permeability, and intracellular damage. The latter may cause destruction of the cell organelles and break-down of cytoplasmic substance increasing osmotic tension inside the cell and helping to retain more water. Changes seen in the endoplasmic reticulum may be a part of this intracellular disturbance. Whatever the mechanism, the relationship of this change to the disease process is difficult to understand.

Golgi Apparatus

The importance of Golgi apparatus in synthesis of acid mucopolysaccharides in synovial cells (Roy and Ghadially, 1967) and in chondrocytes (Godman and Lane, 1964) has been demonstrated. Smooth-walled cytoplasmic vesicles are considered as the mode of transport of such material from the Golgi region to the surface of the cell. Paucity of the Golgi apparatus, as judged by their infrequent presence and small size of the stacks and vesicles constituting them, in osteo-arthritis probably indicates reduced synthesis of hyaluronic acid by synovial cells. This is consistent with the reduced amount of hyaluronic acid in synovial fluid from osteo-arthritis joints (Decker, McGuickin, McKenzie, and Slocumb, 1959; Castor, Prince, and Hazelton, 1966). It is not clear whether this change in synovial cells is primary or is a result of osteo-arthritis.

If the reduction of Golgi apparatus is considered as a primary lesion in osteo-arthritis, it may explain, at least partly, the development of cartilage erosion in osteo-arthritis, for articular cartilage drains its nutrition from synovial fluid (Barnett, Davies, and MacConaill, 1961).

Rod-shaped Bodies

Highton and others (1966) described rod-shaped bodies in vascular endothelial cells of rheumatoid arthritis and suggested that these could represent an infectious agent of aetiological significance. They claimed that these bodies differed from those described by Weibel and Palade (1964) in normal vascular endothelium. Rod-shaped bodies morphologically similar to those described by Weibel and Palade (1964) and Highton and others (1966) were seen in the present study in synovial endothelial cells in all eight cases of osteo-arthritis. We have seen similar bodies also in vascular endothelial cells of normal, rheumatoid, and traumatic arthritic synovia.

In view of their presence in normal and in different diseased synovia, it is unlikely that they are specific agents of any disease of the synovial membrane.

Summary

Synovial membranes from eight cases of osteo-arthritis were studied by electron microscopy. Increase in rough endoplasmic reticulum with frequent dilatations of their cisternae was a common abnormality found in synovial cells. There was often an associated reduction in size and number of the Golgi apparatus and of smooth-walled cytoplasmic vesicles.
Other abnormalities were increase in lysosomal bodies, alteration in mitochondrial structure, and intracellular "edema". Most of these changes were marked in advanced osteo-arthritis and hence considered as secondary manifestations of the disease.

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REFERENCES


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L’ultrastructure de la membrane synoviale dans l’ostéo-arthrose

Résumé

On étudia la synoviale par microscopie électronique dans huit cas d’ostéoarthrose. Parmi les anomalies des cellules synoviales on trouva habituellement une augmentation du réticulum endoplasmique grossier avec des dilatations fréquentes des cisternae. Souvent il y eut aussi une diminution de la taille et du nombre des appareils de Golgi et des vésicules cytoplasmiques aux parois lisses.

Parmi d’autres anomalies on nota une augmentation des corps lysosomiques, un altération de la structure mitochondrial et un “œdème” intracellulaire. La plupart de ces modifications était accusée dans l’ostéoartrhose évolutée et, par conséquent, considérée comme manifestation secondaire de la maladie.

La ultrastructura de la membrana sinovial en la osteoartrosis

SUMARIO

Se estudió en ocho casos de osteoartrosis la sinovia bajo microscopio electrónico. La anomalía común encontrada en las células sinoviales fue un aumento del retículo endoplásmico grosor con dilatación frecuente de sus cisternae. Con esto se asociaba a menudo una reducción del tamaño y del número de los aparatos de Golgi y de las vesículas citoplásmicas con paredes lisas.

Se notaron también otras anormalías, tales como una aumentación de los cuerpos lisosómicos, una alteración de la estructura mitocondrial y un “edema” intracelular. La mayoría de estas alteraciones se destacaba en la osteoartrosis adelantada, de manera que estas se pueden considerar como manifestaciones secundarias de la enfermedad.