

An Ultracentrifugal Analysis of Synovial Fluid*

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SUMMARY

Synovial effusions aspirated from patients with rheumatoid disease or osteoarthritis or following trauma have been fractionated by equilibrium sedimentation in caesium chloride density gradients. The relative concentrations of hyaluronate and chondroitin sulphate have been quantitated and compared to the respective concentrations of these substances in normal ox synovial fluid. Chondroitin sulphate concentration was significantly raised in rheumatoid arthritis and osteoarthritis; there was a less marked drop in hyaluronate concentration. Cartilage damage appears to be associated with high concentrations of chondroitin sulphate in the synovial fluid.

S. Afr. Med. J., **45**, 1205 (1971).

The presence of small quantities of chondroitin sulphate in synovial fluid has been confirmed by Silpananta *et al.*¹ As a result of the marked difference in density between hyaluronate and chondroitin sulphate, it was possible to separate and quantitate the two polysaccharides. Separation was achieved by equilibrium sedimentation in a caesium chloride density gradient.

The chondroitin sulphate in synovial fluid can only have originated in articular cartilage. Mankin² has shown the half-life of cartilage protein-polysaccharide to be of the order of 8 days; chondrocytes continually replace matrix constituents which diffuse out into the synovial fluid or are enzymatically degraded. It has been suggested elsewhere³ that the integrity of the hyaluronate molecule and its con-

centration may be important in resisting the rapid diffusion of protein-polysaccharide from cartilage, and that one might expect higher than normal concentrations of chondroitin sulphate in dilute synovial effusions. Protein-polysaccharides might also be lost from articular cartilage as a result of the action of proteolytic enzymes released into rheumatoid joints.

The purpose of this study was to determine the relative concentrations of hyaluronate and chondroitin sulphate in synovial effusions.

MATERIALS AND METHODS

Synovial fluid was aspirated from the knee joints of 26 patients with synovial effusions: 14 patients had rheumatoid arthritis without significant radiological features of joint destruction; 6 had rheumatoid arthritis with joint destruction; 4 had osteoarthritis; 1 had a post-traumatic effusion and 1 had a torn meniscus. The specimens were centrifuged at $22\,000 \times G$ for 30 minutes to remove particulate matter before being stored at $-15^{\circ}C$. When sufficient specimens had been collected they were thawed and duplicate analyses performed. Total uronic acid analyses⁴ were performed on aliquots of each specimen. Equilibrium sedimentation of each specimen was carried out as described by Silpananta *et al.*¹ with the following modifications. Ultracentrifuge tubes (2 ml) in appropriate adaptors were used in a Spinco type 65 angle rotor. Synovial fluid (0.15 ml) was added to 2.35 ml caesium chloride solution and dialysed to give a final density of 1.67 g/ml. A 2-ml aliquot of the solution was used for the analysis. At the

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end of the run, the tube contents were fractionated as described, and the uronic acid content of each fraction was quantitated. Synovial fluids from 3 apparently normal oxen were obtained, for comparison.

TABLE I. EQUILIBRIUM SEDIMENTATION OF SYNOVIAL EFFUSIONS IN CAESIUM CHLORIDE DENSITY GRADIENTS*

Pathology	Chondroitin sulphate	Hyaluronate
Rheumatoid arthritis	85.8	44.6
Rheumatoid arthritis with joint destruction	145.0	17.6
Osteoarthritis	129.6	50.6
Post-traumatic effusion	12.0	23.8
Torn meniscus	112.8	54.8
Normal (ox)	18.0	85.6

* The mean concentration of hyaluronate and chondroitin sulphate in the effusions are represented as micrograms per 0.1 ml of synovial fluid.

RESULTS

The average concentration of chondroitin sulphate in ox synovial fluid is 18.0 $\mu\text{g}/0.1$ ml and that of hyaluronate 85.6 $\mu\text{g}/0.1$ ml. The mean concentration of chondroitin sulphate in synovial fluids taken from rheumatoid joints without appreciable radiological evidence of joint destruction was 85.8 μg . In the group of rheumatoid joints with significant radiological changes the mean concentration was 145.0 μg , and in osteoarthritic joints 129.6 μg . The

post-traumatic effusion contained 12.0 $\mu\text{g}/0.1$ ml and the fluid taken at meniscectomy 112.8 $\mu\text{g}/0.1$ ml respectively. The mean concentration of hyaluronate in the effusions were all lower than in ox synovial fluid.

CONCLUSIONS

Those effusions associated with joint damage have the highest concentrations of chondroitin sulphate. Even a torn meniscus appears to shed a considerable quantity of chondroitin sulphate into the synovial fluid. The lowest concentrations of hyaluronate are found in those patients with rheumatoid arthritis and significant joint destruction; the effusion in these joints contained the most chondroitin sulphate. This suggests that a fall in hyaluronate concentration may permit a more rapid diffusion of chondroitin sulphate from articular cartilage. Other possible explanations are that severely damaged joints may be unable to produce sufficient hyaluronate, or that the high concentration of chondroitin sulphate in the synovial fluid may resist the diffusion of hyaluronate into the joint.

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