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PATHS OF NUTRITION IN ARTICULAR CARTILAGE AND INTERVERTEBRAL DISCS

By

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Ingelmark & Sääf (1), Ingelmark & Ekholm (2), Holmdahl & Ingelmark (3), Ekholm & Ingelmark (4) and Ekholm (5) have studied articular cartilage and its physiology in rabbits and have found it probable that both bone marrow and joint cavity are of importance for the nutrition of articular cartilage. In connection with other, as yet not published investigations, the opportunity of studying the penetration of a substance, sodium-3-oxypyrene-5,8,10-tri-sulphonate (Bayer), into articular cartilage is offered. This substance has a yellow-green fluorescence in ultra-violet light. The spreading of the substance in the intervertebral discs has also been studied. The fluorochrome was earlier used by Strugger (6) on plants.

MATERIAL

4 rabbits, 4 weeks old.

Sodium-3-oxypyrene-5,8,10-tri-sulphonate, 5 % solution in 0.9 % sodium chloride. pH of solution 5.3.

Reichert Lux U V fluorescence microscope with a Philora 500 W super high pressure mercury lamp.

METHOD

50 mg of sodium-3-oxypyrene-5,8,10-tri-sulphonate was injected into an ear vein of 2 animals. The animals seemed quite unaffected by the injection. They were killed with a blow on the back of the neck *about* 30 seconds and 3½ minutes respectively, after the injection. A small piece of the articular cartilage of one of the lateral tibial condyles and

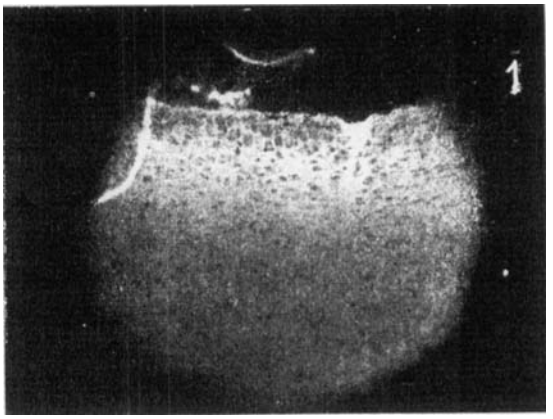

Fig. 1 a.

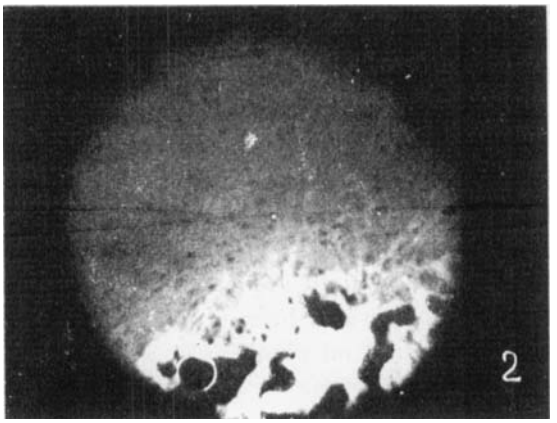

Fig. 1 b.

Articular cartilage from lat. tibial condyle of a rabbit killed 3½ min. after injection of 50 mg fluorochrome. Rather weak fluorescence near the joint cavity and around the epiphysis core. Strong fluorescence in bone.
1 = articular surface.
2 = epiphysis.

also one intervertebral disc with contiguous fragments of bone were immediately removed, cut with the freezing-microtome in 10 μ thick sections without fixation, mounted in paraffin on non-fluorescent slides, and photographed in the fluorescence microscope. Two other rabbits were given 100 mg of the fluorochrome intravenally and were killed 15 resp. 30 seconds later. The spine with the nearest musculature was immediately removed and frozen in ethanol + CO_2-ice. The pre-

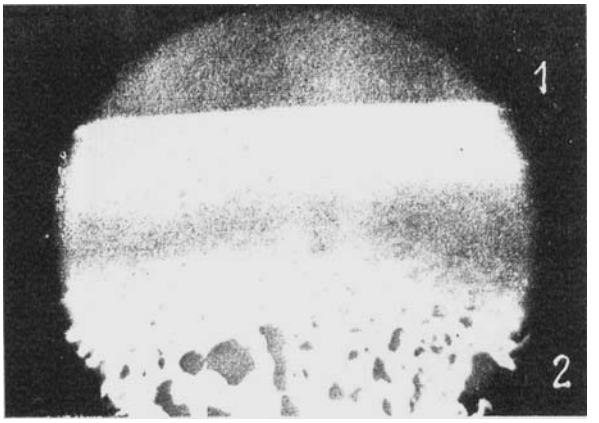

Fig. 2.

The same articular cartilage of a rabbit killed 30 sec. after injection of 50 mg fluorochrome. Rather strong fluorescence both near the joint cavity and near the epiphysis core; hardly any fluorescence centrally in the cartilage. Strong fluorescence in bone.
1 = articular surface.
2 = epiphysis.

parations were planed at −15° C until suitable parts of the intervertebral discs appeared. The preparations, still frozen, were photographed in ultra-violet light.

RESULTS

A. *Articular cartilage.* (Fig. 1–2).

1) The fluorochrome appeared to reach the articular cartilage very rapidly.
2) 30 sec. after the injection strong yellow-green fluorescence was seen in the articular cartilage, partly in an area immediately beneath the articular surface, partly around the epiphysis core while the part between these areas did not show any definite fluorescence.
3) Articular cartilage of the animal killed 3½ min. after the fluorochrome injection showed considerably weaker and more evenly distributed fluorescence than articular cartilage of the other animal killed 30 sec. after the injection.

B. *Intervertebral discs.* (Fig. 3–5).

1) The hyaline cartilage plate rapidly absorbs the fluorochrome. The contiguous part of the annulus fibrosus shows rather weak fluoroscence.

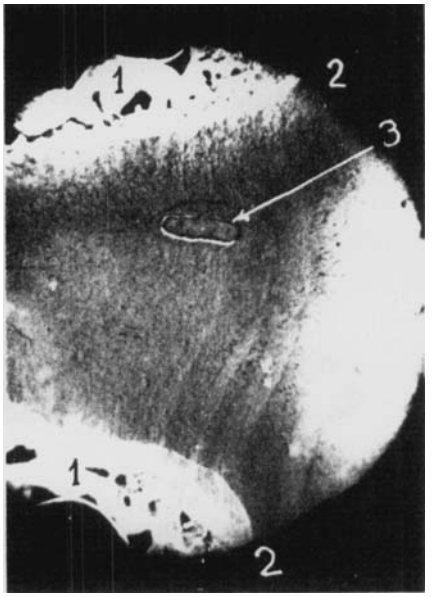

Fig. 3.

Periphery of intervertebral disc of a rabbit killed 30 sec. after injection of 50 mg fluorochrome. Parts of adjoining vertebrae with a thin layer of hyaline cartilage between bone and disc. Strong fluorescence peripherally to the disc, in hyaline cartilage and in bone. The fluorochrome appears to pass into the disc from the periphery and via the hyaline cartilage which forms a remarkably sharp border to the disc.

1 = vertebral body.
2 = hyaline cartilage plate.
3 = artifact in the film.

2) The annulus fibrosus has the greatest concentration of fluorochrome in its peripheral parts and the amount decreases gradually towards the nucleus pulposus. In the vicinity of this no definite fluorescence could be observed.

C. *Bone.* (Fig. 1 b, 2, 3).

The fluorochrome appears to have an appreciable affinity to bone tissue the fluorescence of which appears to be about equally strong 30 sec. and 3½ min. after the administration.

DISCUSSION

Sodium-3-oxypyrene-5,8,10-tri-sulphonate, even in rather large quantities, apparently does not hurt rabbits in experiments of short duration. It has a strong fluorescence.

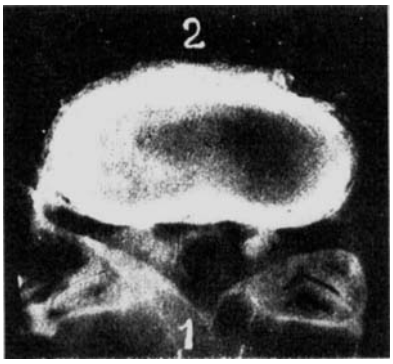

Fig. 4.

A somewhat oblique section of frozen lumbo-sacral disc of a rabbit killed 30 sec. after injection of 100 mg fluorochrome. In the left part of the disc the surface of the cut lies near the hyaline cartilage plate. The fluorescence is strongest in the peripheral parts of the disc.
1 = dorsally.
2 = ventrally.

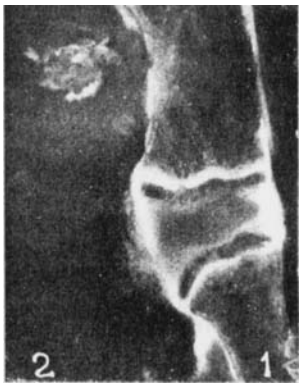

Fig. 5.

Sagittal cut of frozen lumbar disc of a rabbit killed 15 sec. after injection of 100 mg fluorochrome. The fluorescence is strongest dorsally. No definite fluorescence can be found in the nucleus pulposus.
1 = dorsally.
2 = ventrally.

As cartilage lacks capillaries it must be assumed that nutriment is supplied through extravascular circulation. The spread of the fluorochrome in articular cartilage and intervertebral discs in the experiments described above where a very short time has elapsed between the administration of the fluorochrome intravenally and the killing of the ani-

mal, should reasonably indicate from which direction liquid and substances dissolved in it penetrate into the cartilages mentioned, thus giving a rough idea of the nutrition paths. Postvital diffusion of the matter in the tissues has not been perceived in the microscopic cuts during an observation period of at least 20 min.; neither has it been possible to register such spreading when macro-photographing larger preparations at various times.

The examination of articular cartilage confirms the views put forward by other authors (Ingelmark, Sääf, Ekholm, Holmdahl).

Dr. J. G. Nordén has been responsible for the microphotographing and has, in many other ways, been of great help during this investigation.

SUMMARY

Through experiments with a fluorescent compound on rabbits, the penetration of the compound into intervertebral discs and joint cartilage is studied. The compound penetrated the intervertebral discs partly from the periphery and partly, although to a more limited degree, from the vertebral bodies. The compound penetrated the articular cartilage both from the articular surface and from the epiphysis.

RESUME

Dans des essais sur les lapins avec des matières fluorescentes, l'on a étudié la pénétration de la substance dans les disques intervertébraux et les cartilages articulaires.

La substance pénètre dans le disque intervertébral en partie de la périphérie et en partie, quoique dans une moindre proportion, du corps de la vertèbre. Dans le cartilage articulaire, la substance s'infiltre aussi bien de la surface articulaire que de l'épiphyse.

ZUSAMMENFASSUNG

Durch Versuche mit fluoreszierenden Stoffen wurde am Kaninchen das Eindringen des Stoffes in die Zwischenwirbelscheiben und den Gelenksknorpel untersucht. Der Stoff dringt teilweise von der Peripherie und teilweise, aber in minderem Ausmasse, von den Wirbelkörpern in die Zwischenwirbelscheiben ein. In den Gelenkknorpel dringt der Stoff sowohl von der Gelenkoberfläche als auch von der Epiphyse her, ein.

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