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Comparative Ultrastructural Analysis of the Articular Cartilage in Overloading and in Big Temperature Deviations

N. Vidinov, K. Vidinov, M. Kalniev, D. Krastev, N. Krastev

Department of Anatomy, Medical University, Sofia

We had compared the results from our experiments on Wistar rats undergoing overloading on the tredbahn and experiments on other Wistar rats whose limbs had been subsequently put into cold water- 1-2 degrees and then into hot water 60 degrees. We played special attention to the articular cartilage as this is a particular structure with a specific metabolism and the chondrocytes comprising it are not connected with a vascular system. From the results of the light and electron microscopy of the two animal groups it becomes clear that the wave-like undulating bordering line between the articular cartilage and the bone — the so called tide mark, is significantly thinner, the proteoglycans in the territorial matrix were significantly and evidently different from those in the interteritorial matrix. The computer analysis of the globular subunits in the extracellular matrix of the articular cartilage of the first group showed that in cases of overloading an increase of the big subunits in contrast to the small and average once was found. The same thing was observed with the globular subunits in the extracellular matrix of the articular cartilage in animals converged to sharp temperature changes.

Key words: articular cartilage, ultrastructure, proteoglycans, globular subunit, computer analysis.

Introduction

The state of the articular cartilage in different conditions, such as overloading [7, 8] is always expressed by variety of symptoms such as: stiffness, limited mobility and pains. These symptoms are also seen in some individuals during rapid temperature changes, which make these individuals believe that they have a certain meteorological abilities in predicting the whether. Previous research on the structure of the cartilage in these conditions shows that it is an extremely sensitive structure, giving quick changes evident on the electron microscopical examinations [1, 6, 8]. It was our aim to compare the results from our experiments on Wistar rats undergoing overloading on the tredbahn and experiments on other Wistar rats which limbs had been subsequently put into cold water- 1-2 degrees and then into hot water 60 degrees. We paid special attention to the articular cartilage as this is a particular structure with a specific metabolism and the chondrocytes comprising it are not connected with a vascular system.

Material and Methods

Material taken from 36 "Wistar" rats aged 8-12 months were employed. The 1st group of animals was investigated under the condition of intensive movement (animals were put in the tredbahn for 1 hour every 10 days), and the 2nd group were investigated under the condition of high temperature differences (their right legs were put in the water with temperature 1 degrees C and 60 degrees C). Material for investigation from intensive zone of articular cartilage was taken. Light microscopical investigations were performed after the methods of Masson. Electron microscope study was performed also for demonstrating proteoglycan complexes of the intercellular cartilage matrix after the method of Shepard and Mitchel (1976 (5), using Safranin O for the purpose.

Results

Comparing the results from the light microscopy of the two animal groups it becomes clear that the wave-like undulating bordering line between the articular cartilage and the bone — the so called tide mark — is significantly thinner. It is easily distinguished as well stained band, which outer border is relatively flat and the inner one is largely indented. Looking closely and investigating specifically this structure we found out that it is comprised mainly of chondroblasts with hypertrophy and have an oval shape and relatively big size (Fig.1). In the extracellular matrix a fine network

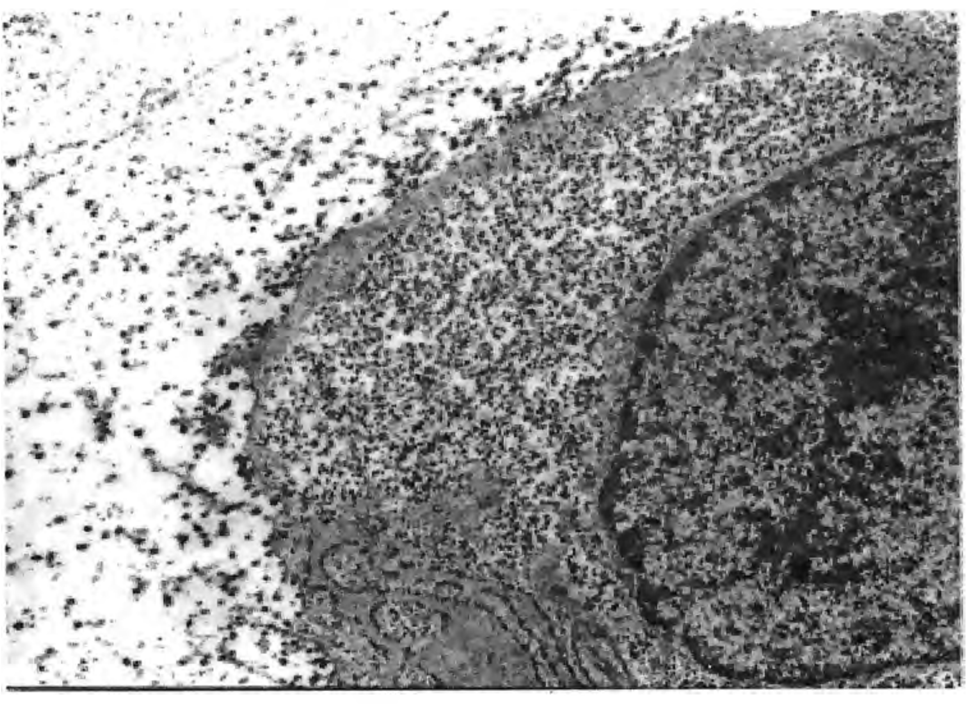

Fig.1. We found out that the tide mark is comprised mainly of chondroblasts with hypertrophy and have an oval shape and relatively big size (× 18 000)

of collagen bundles and proteoglycan complexes, equally distributed in the territorial and interterritorial matrix was observed. When examining the chemical structure of the proteoglycan complexes more closely, we established that it is composed of one core protein in the center, connecting proteins and glucosaminoglican chains. During prolongated and intensive overloading (10 days with 1 hour each day) on the tredbahn changes in the surface layer of the articular cartilage were detected. Indentations and roughness of the articular cartilage as well as disappearance of Lamina splendes were also found. In the tide mark changes in the carbohydrates metabolism are observed both during overloading and in drastic temperature changes. At first we depicted glycogen deposition in the chondroblasts and in the same time increase of the proteoglycan concentration in the territorial matrix of the cells. The intensity of the staining and the structure of the proteoglycans in the territorial matrix were significantly and evidently different from those in the interteritorial matrix. The computer analysis of the globular subunits in the extracellular matrix of the articular cartilage of the first group showed that in cases of overloading an increase of the big subunits in contrast to the small and average once was found. The same thing was observed with the globular subunits in the extracellular matrix of the articular cartilage in animals converged to sharp temperature changes. In those case a bigger clustering of the proteoglycan complexes was also typical (Fig. 2). When staining the slides for collagen and proteoglycans we see a fine network in which the proteoglycans serve as bridge structures between the collagen fibers, keeping them in a certain distance from one another (Fig. 3). As a result of the overloading and the temperature abnormalities this particular network is slightly damaged and disturbed, causing the proteoglycan bridges to disappear and then the collagen fibers stick to one another.

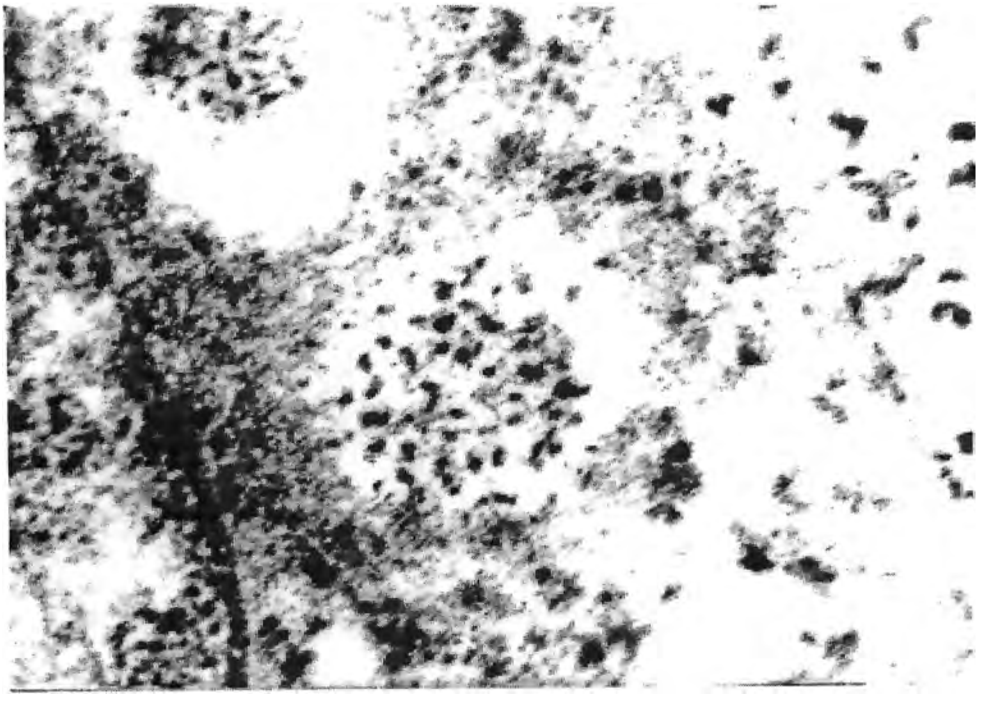

Fig. 2. In thise case a bigger clustering of the proteoglycan complexes was also typical (× 23 000)

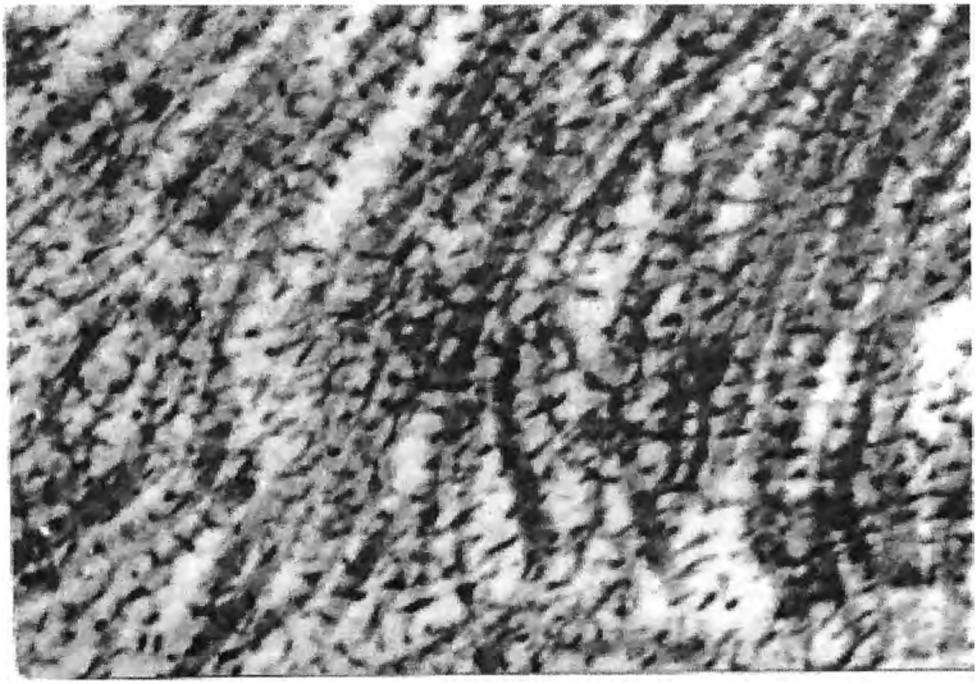

Fig. 3. Staining for collagen and proteoglycans shows a fine network in which the proteoglycans serve as bridge structures between the collagen fibers, keeping them in a certain distance from one another (× 48000)

Discussion

The results from the comparative analysis show that there are similarities in the fine changes in the matrix of the articular cartilage in sharp temperature changes and overloading of the articular apparatus. They are mainly expressed in the processes of increased anaerobe glycolisis in the transitional zone cartilage-bone called tide mark [9]. Morphologically that is visualized by glycogen deposition in the chondroblasts and increase of the proteoglycan synthesis [4]. At the same time we established that the produced proteoglycans are composed of bigger globular subunits and tend to merge with one another. Alterations in the character of the collagenogenesis also take place. It can be assumed that the alterations in the cartilage metabolism lead to the hindered elimination of the waste products by the synovial membrane and to the clinical signs of articular discomfort. With the same mechanism we can explain the changes in the cartilage due to sharp change of the climate and mainly sharp temperature changes.

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