

## Immunohistochemical study of the distribution of fibronectin in some zones of the meniscus

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The aim of our study was to trace out the distribution of the fibronectin in some different zones of the meniscus. The investigation was performed upon menisci of the knee joints of Wistar rats. A light microscopy, transmission electron microscopy and immunohistochemistry to demonstrate the fibronectin were used. We observed that fibronectin consisted of medium-sized granules located densely among themselves mainly in a row on the plasmalemma of cells in the SSZ. In TSZ the fibronectin consisted of fine and medium sized granules located not only on the cell membrane, but also near to the cells in the territorial matrix. Our observation revealed that in the deeper TPZ the fibronectin consisted by small granules situated in the territorial matrix of the cells.

*Key words:* meniscus, fibronectin, chondroblasts, fibroblasts, collagen.

### Introduction

The fibronectin with the chondronectin and the ancorin is one of the adhesive glycoproteins in cartilage. They act as “cellular glue” [2, 8]. Very little has been written in the literature concerning the distribution of fibronectin in different areas of the meniscus [4, 5]. For this reason we aimed to investigate the presence and distribution of fibronectin in some areas of the meniscus.

### Material and Methods

The materials of the investigation were menisci of the knee joint of 15 Wistar rats of both sexes, aged between 60 and 120 days, weighing about 250 g each. The animals were treated under the European Convention working with experimental animals. The fixation was carried out by glutaraldehyde and formaldehyde. Permanent histological preparations were obtained after appropriate procedures. They have been colored with HE and AZAN. Light microscopy (HE and AZAN), transmission electron microscopy (TEM) and immunohistochemistry to demonstrate the fibronectin were performed. We traced out the distribution of the fibronectin in some zones of the menisci.

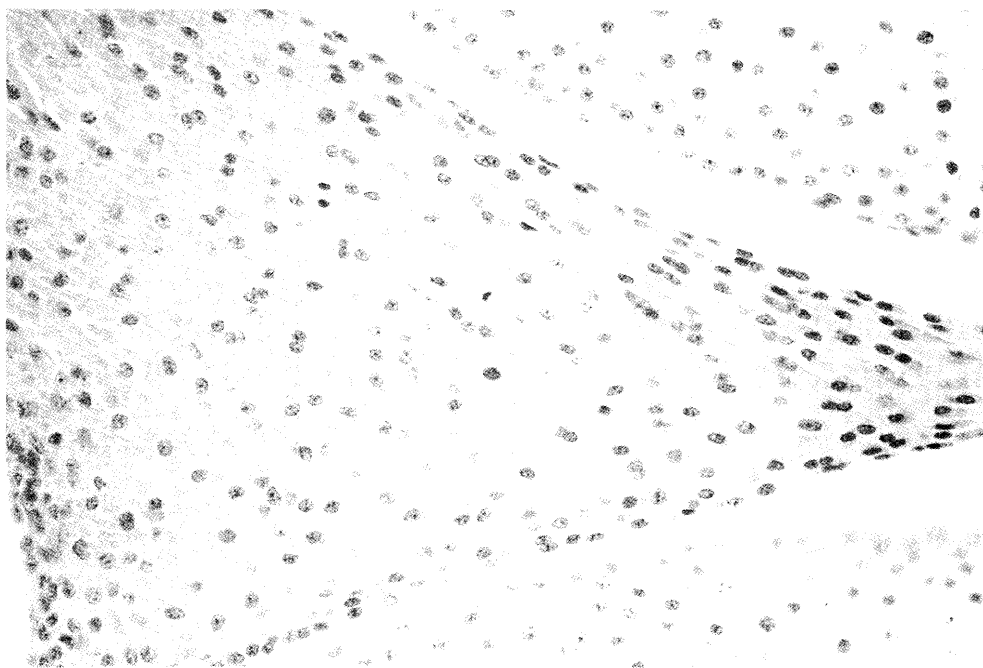


Fig.1. The Superficial Sliding Zone consists of 2 to 4 layers of elongated fibroblasts that have a longitudinal axis parallel to the articular cleft. The Transitional Sliding Zone is placed just below SSZ. It was composed of elliptical or oval cells with the character of chondroblasts and intercellular matrix represented by mixed network of collagen fibers type I and II. Staining – AZAN; X – 200.

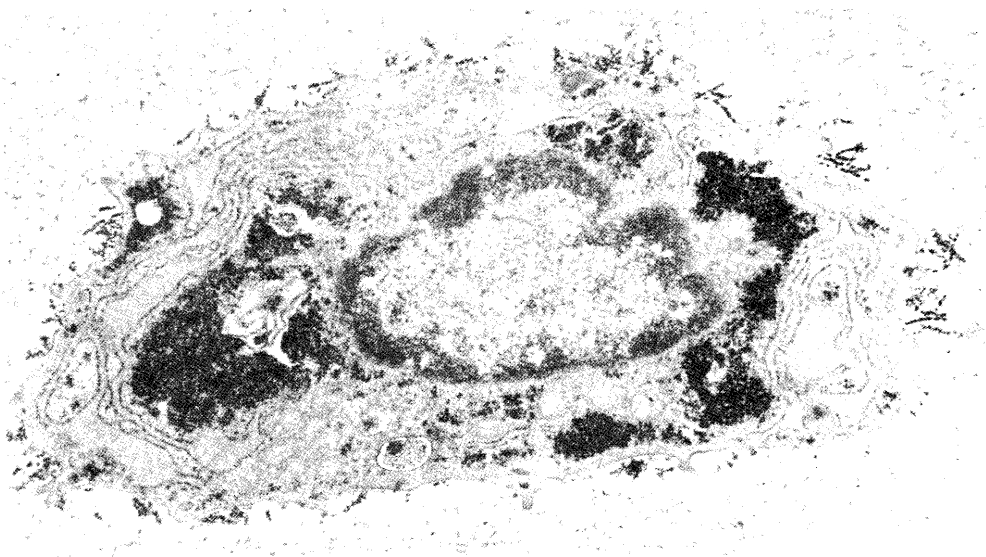


Fig. 2. Immunohistochemical study showed that the fibronectin is represented by medium-sized granules located densely among themselves mainly in a row on the plasmalemma of cells of the SSZ. TEM with immunohistochemistry for fibronectin; X – 11500

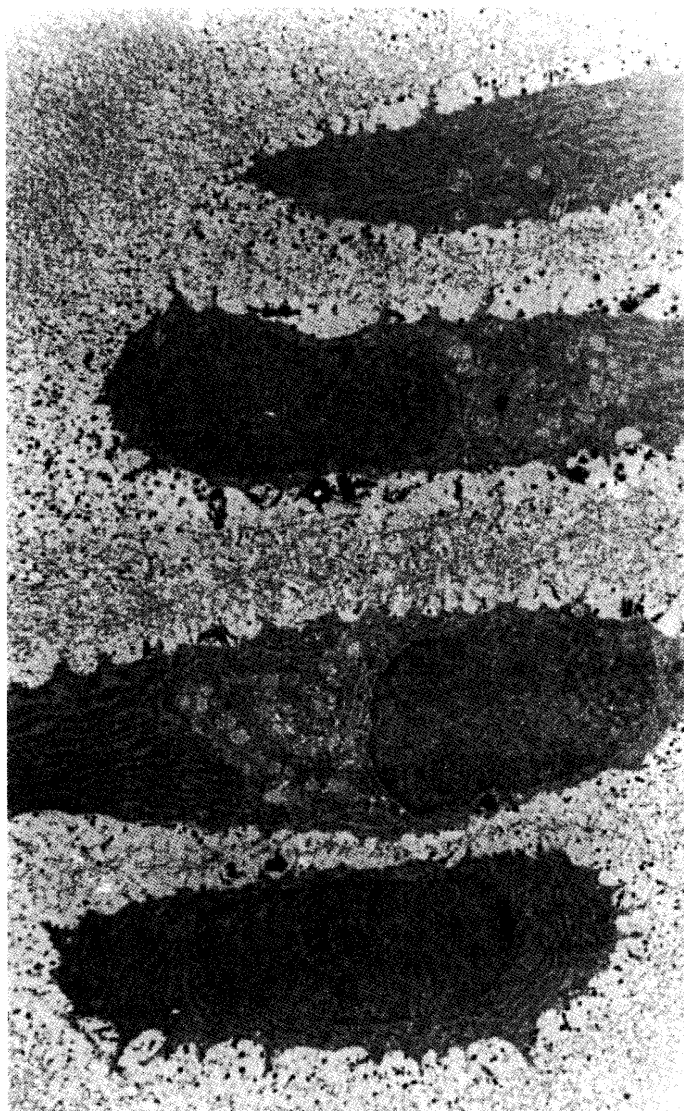


Fig. 3. Fine and medium sized granular fibronectin located near and on the cell membrane of TSZ. TEM with immunohistochemistry for fibronectin; X - 6000

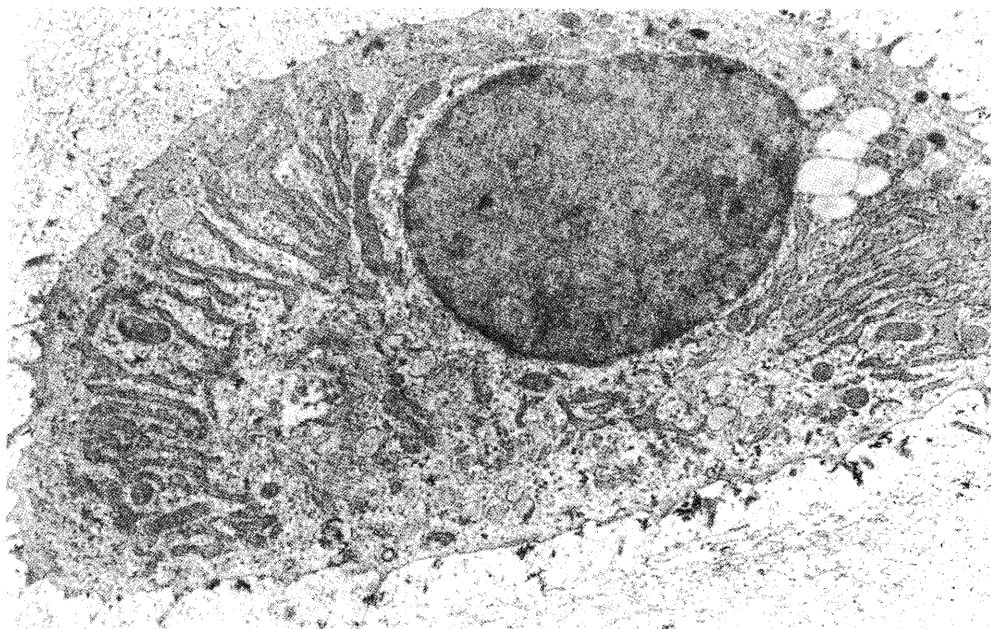


Fig. 4. Electron image showing the transitional pressure zone (TPZ) situated deeper in the meniscus. It is composed of chondroblasts with well-developed GER and Golgi complex. TEM; X – 8000

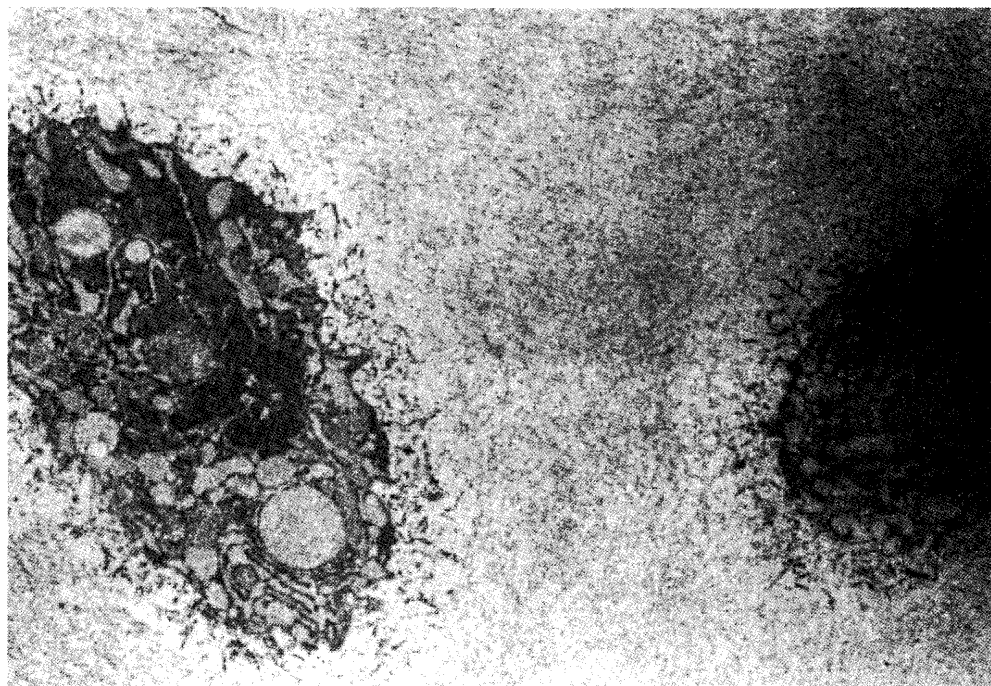


Fig. 5. Chondroblasts of TPZ. There were observed small granules of fibronectin in the territorial matrix. TEM with immunohistochemistry for fibronectin, X – 6000

## Results

The results from light microscopic examination in AZAN showed the location of the Superficial Sliding Zone and the Transitional Sliding Zone in the meniscus.

Immunohistochemical study of fibronectin in SSZ indicates the presence of reaction product in the territorial matrix directly on the plasmalemma of cells. These are medium-sized granules located densely together most often in a row on the plasmalemma (Fig. 2). Sometimes it can be observed much granules in two or more lines, but in certain sections may be missing.

Immunohistochemical study of fibronectin in TSZ showed a similar picture as in SSZ. We observed fine and medium sized granular palisading arranged on the cell membrane of chondrocytes (Fig. 3).

The transitional pressure zone is composed by chondroblasts. They usually had well-developed GER and Golgi complex.

The study of fibronectin showed a large amount of reaction product in the territorial matrix of SPZ and TPZ. The reactionary granules are located densely and wrong without forming layers (Fig. 5).

## Discussion

Unlike the chondronectin, which is available exclusively pericellular in the territorial matrix [2] and is associated with collagen type II and proteoglycan aggregates [7], our investigation shows that fibronectin is located both on the plasmalemma of cells and pericellular in the territorial matrix. Moreover, the fibronectin is placed in the territorial matrix of cells when the cells are chondroblasts [1, 3]. When the cells are fibroblasts as in SSZ the fibronectin is located upon plasmalemma itself moreover in a row. In TSZ the cells are chondroblasts [10] and the collagen is both type I and type II. The fibronectin in TSZ is situated both upon cell plasmalemma and in the territorial matrix. TPZ is composed by chondroblasts and collagen only type II [10]. The fibronectin in this zone is situated only in the territorial matrix around the cells but not on the plasmalemma of cells TPZ.

The distribution of the fibronectin is obviously related with the type of cells and collagen types in different zones of the meniscus. Cells are fibroblasts and the collagen is type I in SSZ [10]. Here the fibronectin is located only upon plasmalemma but not in the intercellular matrix. This fact confirms long-established concept, that fibronectin is a product of fibroblasts [9]. However its presence pericellular around chondroblasts shows that the fibronectin is also related with the chondrogenesis [6]. It is clear that further investigation about the distribution of the fibronectin in the other zones of the meniscus is necessary.

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List of the most used abbreviations:

AZAN – azocarmine aniline orange

GER – granular endoplasmic reticulum

HE – haematoxylin eosin

SPZ – superficial pressure zone

SSZ – superficial sliding zone

TEM – transmission electron microscopy

TPZ – transitional pressure zone

TSZ – transitional sliding zone