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## INFLUENCE OF GLUCOSE ON CARTILAGE TISSUE STRUCTURE: PHYSICAL MECHANISM

*We propose a mechanism according to which the introduction of glucose into cartilage tissue changes the structure of this tissue. Cartilage tissue is considered to be a porous medium, where the role of solid component is played by collagen fibers and layers where proteoglycans are located. Collagen chains that connect the aforementioned objects are adsorbed on the surface of the fibers and the layer. The thermodynamic features of the links of adsorbed chains have been determined. It was shown that glucose molecules, being introduced into cartilage tissue, partially displace the adsorbed chains from the fiber surface. These chains cover cracks that may appear in the fiber under the action of external loads, which leads to a reduced rigidity of proteoglycan layers. A conclusion has been drawn that the introduction of glucose molecules enlarges the shear compliance of cartilage tissue. This conclusion is confirmed by experimental results obtained for a model system, gelatin gel, whose structure is considered to be similar to that of cartilage tissue.*

*Keywords:* cartilage tissue, glucose, shear compliance.

### 1. Introduction

It is known that cartilage tissue, together with the muscular system, plays the supporting-mechanical and protective role in the body [1]. In the publications dealing with this issue, considerable attention is paid to the study of the mechanical properties of cartilage tissue (see, for example, review [2]). A necessity in physical models that would describe a relation between the structure and mechanical properties of cartilage tissue was emphasized [3, 4].

In the course of the human body's vital activity under the influence of various external and internal factors, cartilage tissue can undergo a certain damage. One of the common diseases associated with the cartilage tissue damage is osteoarthritis [5]. When treating this disease, intra-articular injections of vari-

ous drugs, including those containing glucose, are applied [5–9].

The results of clinical studies performed in the cited works are usually discussed from the medical and biological viewpoints. The novelty of this article consists in that it considers the properties of cartilage tissue from a physical viewpoint. Namely, a physical mechanism is proposed that is responsible for the influence of glucose injections on the cartilage tissue structure. The paper is an ideological continuation of our works [10–12] on the physics of cartilage tissue.

### 2. Adsorbed Chains in Cartilage Tissue Structure

In works [11, 12], we proposed a structural model of cartilage tissue. The logic that brought us to this model is as follows. It is noted in the literature (see, e.g., [13]) that, according to certain features, cartilage tissue can be attributed to hydrogels. There is also a widespread view of hydrogel as a porous medium, whose pores are filled with water (see, e.g., [14]). Thus, we may consider cartilage tissue as a porous medium and use the ideas developed in the theory of porous media to describe its struc-

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ture (see, e.g., [15]). The starting point of this theory is the thesis of ideal frameworks based on either rods or plates. Both frameworks are cubic lattices, but in the former case the lattice is formed by rods, which are located at the edges of elementary cells (cubes), whereas, in the latter case, the lattice is formed by plates, which are the faces of elementary cells.

In works [11, 12], the concept of an ideal framework for cartilage tissue was introduced. It was called the rod-plate framework. Such a framework is a lattice formed by rods oriented in the same direction and connected by a set of infinite parallel plates (layers).

It is known (see, e.g., [1, 2]) that the main components of cartilage tissue are collagen, proteoglycans, and water. It is also known that collagen chains in cartilage tissue form fibers [1]. Accordingly, the rods in an ideal framework are identified with collagen fibers, and the layers contain proteoglycans. There are holes in the layers through which the fibers pass. Collagen chains that do not enter the fibers' content connect the fibers and the layers (see Fig. 1, where a cross-section along the fiber axis is shown).

As can be seen from Fig. 1, the collagen chains consist of two sections: a longitudinal one, which is connected to the fiber surface and directed along the fiber axis, and a transverse one, which is connected to the layer surface and directed along the fiber radius  $R$ . We will call such chains adsorbed. The points separating both areas of the adsorbed chains are located on a circle with radius  $R$ .

Let us denote, by  $\mathbf{S}$ , the compliance tensor of cartilage tissue with an ideal framework. The components of this tensor  $S_{ik}$  will be determined in the system of principal axes. As was shown in work [11], the following inequalities hold:

$$S_{44} \gg S_{ik} \quad (i, k \neq 4). \tag{1}$$

It was also shown in work [11] that the value of the component  $S_{44}$  is determined by the bending stiffness of the layers.

### 3. Thermodynamics of Adsorbed Chains

The free energy  $F$  of an adsorbed chain is represented as the sum

$$F = F' + F'', \tag{2}$$

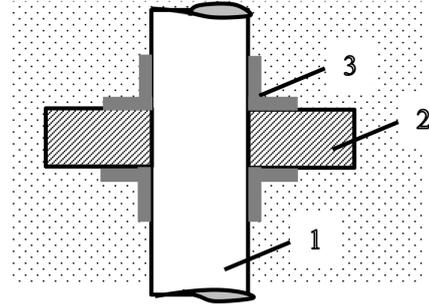


Fig. 1. Schematic diagram of fiber (1) connection with proteoglycan layer (2) using adsorbed chains (3)

where  $F'$  and  $F''$  are the free energies of the transverse and longitudinal sections, respectively. These summands are described by the well-known formulas (see, for example, [16])

$$F' = E' + k_B T \sum_J \ln \left( 2 \sinh \frac{\hbar \omega'_J}{2k_B T} \right), \tag{3}$$

$$F'' = E'' + k_B T \sum_J \ln \left( 2 \sinh \frac{\hbar \omega''_J}{2k_B T} \right), \tag{4}$$

where  $T$  is the temperature,  $k_B$  is the Boltzmann constant,  $E'$  and  $E''$  are the energies of the indicated sections at  $T = 0$ ,  $\omega'_J$  and  $\omega''_J$  are their natural vibration frequencies,  $\hbar = h/(2\pi)$ , and  $h$  is Planck's constant.

Let us denote the length of the adsorbed chain by  $l$ , the cross-section length by  $\chi$ , and the energy of bond breaking between the links of different collagen chains by  $\varepsilon'$ . Accordingly, we have the following formulas:

$$E' = \varepsilon' \frac{\chi}{a}, \tag{5}$$

$$E'' = \varepsilon'' \frac{l - \chi}{a}, \tag{6}$$

where  $a$  is the link size.

To determine the features in the thermodynamics of adsorbed chains, we will use the Debye approximation. The sections of the adsorbed chain will be considered as rods on an elastic base.

It is known (see, e.g., [17]) that three types of waves can propagate in a rod: one longitudinal and two bending waves. The stiffness of the chain along the axis substantially exceeds its bending stiffness. Therefore, the vibration frequencies of bending waves are considerably lower than the vibration frequencies of

the longitudinal wave. Therefore, we neglect the contributions of the latter to the free energy. Then the equation of motion for a rod on an elastic base looks like

$$-\rho a^2 \frac{\partial^2 u}{\partial t^2} = D \frac{\partial^4 u}{\partial x^4} + \alpha u, \quad (7)$$

where  $\rho$  is the rod density,  $u$  is the transverse displacement,  $t$  is the time,  $D$  is the rod bending stiffness,  $x$  is the coordinate axis directed along the undeformed rod, and  $\alpha$  is the elasticity coefficient of the base.

Considering the ends of the rods to be supported on an elastic base, we have the following formulas for the natural rod vibrations:

$$u' = \text{const} \sin k'x \exp(-i\omega't), \quad (8)$$

$$u'' = \text{const} \sin k''x \exp(-i\omega''t). \quad (9)$$

Substituting them into Eq. (7), we obtain the following equalities for the natural frequencies:

$$\omega' = \left[ \frac{D'}{\rho a^2} (k')^4 + \frac{\alpha'}{\rho a^2} \right]^{1/2}, \quad (10)$$

$$\omega'' = \left[ \frac{D}{\rho a^2} (k'')^4 + \frac{\alpha''}{\rho a^2} \right]^{1/2}. \quad (11)$$

In formulas (8)–(11), the quantities primed one time (two times) are related to the transverse (longitudinal) section. The values for the wave numbers are

$$k'_J = \frac{\pi J}{x} \left( J = 1, 2, \dots, \frac{x}{a} \right), \quad (12)$$

$$k''_J = \frac{\pi J}{l-x} \left( J = 1, 2, \dots, \frac{l-x}{a} \right). \quad (13)$$

Unlike the collagen fiber links, the arrangement of proteoglycan chains is disordered by definition, which leads to the inequality

$$\varepsilon' > \varepsilon''. \quad (14)$$

The coefficients of base elasticity are evaluated using the formulas

$$\alpha' = \frac{|\varepsilon'|}{a^3}, \quad (15)$$

$$\alpha'' = \frac{|\varepsilon''|}{a^3}. \quad (16)$$

By definition, the energies  $\varepsilon'$  and  $\varepsilon''$  are negative. According to expression (14), we have the inequality

$$|\varepsilon'| < |\varepsilon''|, \quad (17)$$

and comparing it with equalities (15)–(16), we obtain

$$\alpha' < \alpha''. \quad (18)$$

As can be seen from the aforesaid, the free energy of an adsorbed chain depends on the length cross-sectional  $x$ ,

$$F = F(x). \quad (19)$$

Let the cross-section be absent at a certain time moment, i.e.,  $x = 0$ . According to inequality (14), with the appearance of this cross-section and the growth of its length, the free energy  $F$  should increase. But at the same time, as follows from expressions (10)–(13) and (18), the frequency value decreases, which, according to formulas (2) and (3), is accompanied by a simultaneous decrease of  $F$ .

As a result of both those factors, an equilibrium state of the adsorbed chain is established, which corresponds to the minimum free energy value  $F(x_0)$ . This is a state for which the cross-section length is equal to  $x_0$ .

#### 4. Adsorbed Chains under Glucose Action

It is known (see, e.g., [1]) that a proteoglycan chain is branched, and its side chains are polysaccharides. The vast majority of the proteoglycan chain links are the links of polysaccharide side chains. So, we may assume that the links of the transverse section in the adsorbed chain form bonds only with the links of the polysaccharide chain. It is believed that glucose injections should contribute to the treatment of cartilage tissue damage. Let us consider a possible mechanism of such a treatment.

Let the damage be a crack (Fig. 2, *a*) that arose in the fiber, for example, under the action of an external load. When injected, glucose enters the pores of cartilage tissue and gains a possibility to be adsorbed on the surface of supramolecular formations in cartilage tissue. The side chains of polysaccharide chains that compose the proteoglycan chain are structurally similar to a glucose molecule. Therefore, it is most probable that the glucose molecules will bind

just with these links. Let us denote the breaking energy of such a bond as  $\varepsilon'_g$ . Since the formation of the described bond is more probable than the formation of a bond between a polysaccharide chain link and a collagen chain link, the following inequality is valid:

$$\varepsilon'_g < \varepsilon'. \quad (20)$$

Therefore, the appearance of glucose molecules near the transverse section of the adsorbed chain leads to that some polysaccharide chain links break the bonds through which they were previously connected to the links of the transverse section and bind with glucose molecules (Fig. 2, b).

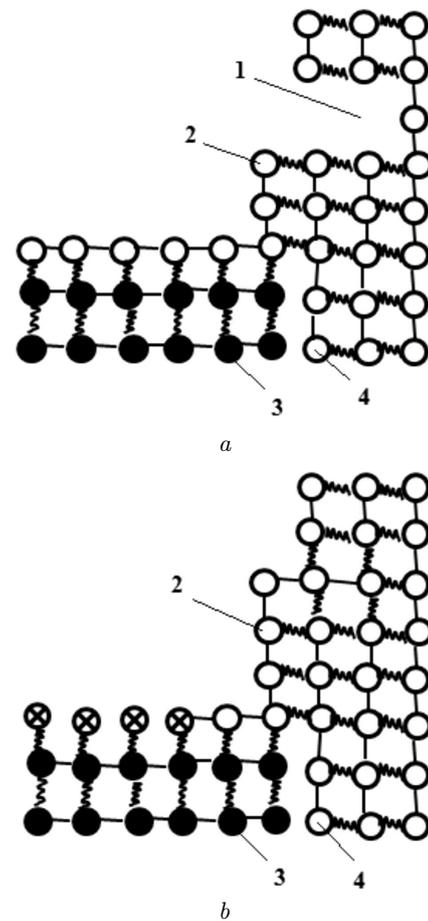
Glucose molecules displace the transverse section from the surface of the proteoglycan layer, and the equilibrium length of this section becomes equal to  $x'_0 < x_0$ . This length is achieved owing to the diffusion of the adsorbed chain along the proteoglycan layer and fiber surfaces. Finally, the described chain turns out in the crack. The formation of bonds between the adsorbed chain and the crack walls substantially reduces the free energy  $F$  of this chain, which enhances the stability of the new equilibrium state of the adsorbed chain.

Hence, according to the proposed mechanism, the role of glucose molecules consists in “displacing” the adsorbed chain, making it diffuse toward the crack, and ultimately closing the latter.

### 5. Adsorbed Chains and Cartilage Tissue Compliance

Below, the matter concerns the verification of the proposed mechanism. As one can see from Fig. 2, when glucose is introduced, the adsorbed chain moves from the surface of the proteoglycan layer to the surface of the collagen fiber. This chain has a substantial rigidity. Therefore, its displacement from the layer surface should increase the compliance  $S_{44}$  of the latter. This fact suggests the following testing procedure for the proposed mechanism: it is necessary to measure the compliance  $S_{44}$  of cartilage tissue with introduced glucose and compare the obtained result with the compliance  $S_{44}$  of the tissue without glucose. If it turns out that the compliance  $S_{44}$  in the former case is larger, this fact will serve as an argument in favor of the proposed model.

As was already mentioned [see formula (1)],  $S_{44}$  is the compliance of an ideal framework. It is clear



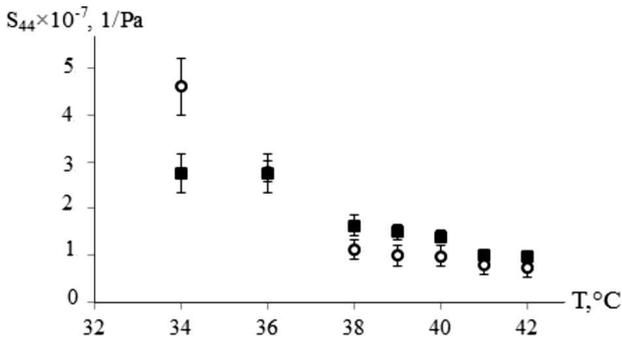
**Fig. 2.** Cartilage tissue damage (a) and its healing (b): crack (1), adsorbed chain (2), proteoglycan layer (3), collagen fiber (4), collagen fiber links (o), polysaccharide chain links (•), glucose molecules (⊗), intramolecular bonds (—), intermolecular bonds (zigzag)

that a real framework differs from the ideal one. In work [11], a model of cartilage tissue with a real framework was proposed. In this model, the parameter  $S_{44}$  acts as a local shear compliance, which is related to the shear modulus  $G$  of cartilage tissue by the formula

$$S_{44} = \frac{5}{2} G^{-1}. \quad (21)$$

### 6. Experimental Part

As was already mentioned, it is common to classify cartilage tissue to hydrogels. Logically, this means that the model of an ideal rod-plate framework proposed for cartilage tissue should remain valid for other



**Fig. 3.** Temperature dependences of shear compliance  $S_{44}$ : 12% gelatin hydrogel (○), 12% gelatin hydrogel with glucose (■)

representatives of the class of hydrogels. This circumstance opens certain possibilities for studying cartilage tissue under various conditions using model systems.

The central point of this paper is the behavior of adsorbed chains of cartilage tissue. These are collagen chains. Therefore, in the cartilage tissue model, the adsorbed chains should also be collagen. This condition is satisfied by gelatin hydrogel, which consists exclusively of collagen chains. Accordingly, these same chains form fibers, which makes the selected model related to cartilage tissue. These chains form layers in the model system, which distinguishes the latter from cartilage tissue, where the layers contain proteoglycans. Since the matter concerns hydrogel, there must exist a polymer network in the latter [18]. In cartilage tissue, the network structure is formed by layers, where the network is formed by proteoglycan chains. Logically, similar layers must exist in the model system as well. But now the network is formed by collagen chains.

When preparing specimens, we used edible gelatin bloom 200 (France). Specimens of two types were used. They were prepared as follows.

*Type I.* Gelatin was added to distilled water with a temperature above 65 °C (according to work [14]), and a solution with a concentration of 12% was made. The resulting “water-gelatin” liquid system was permanently stirred to ensure its homogeneity. Then, a 20% dextrose (D-glucose) solution was added to the solution, which was cooled down to 40 °C (this is a temperature at which the system is a structure of flexible single coils [15]). The amount of the specimen was determined according to the

dose given in work [5] and corresponded to 0.5 ml of 20% dextrose (D-glucose) per 100 ml of gelatin solution. The 40% aqueous solution of glucose (1 ml of the solution contained 400 mg of glucose monohydrate) produced at the pharmaceutical company “Darnytsia” was mixed with bidistillate in the ratio 1:2. Cylindrical polyethylene cuvettes were filled with the prepared liquid system cooled down to room temperature and kept for one day at a temperature of 18–20 °C. Afterward, the shear modulus of the liquid was measured.

*Type II.* The specimens were prepared similarly to the samples of type I, but without adding D-glucose. The shear modulus was measured using a torsion pendulum according to the method described, e.g., in work [19].

The measurements were carried out within a temperature interval of 34–42 °C, which corresponds to the temperature interval of the human body functioning.

The local shear compliance  $S_{44}$  was calculated using formula (21). The calculation results are presented in Fig. 3, where the white circles demonstrate the experimental results for 12% gelatin hydrogel, and the gray squares do the same for 12% gelatin hydrogel with the addition of glucose.

According to the applied structural model, the deformation of cartilage tissue is a consequence of the deformation of the layers connecting the collagen fibers via the adsorbed collagen chains. As was mentioned above, these layers have a network structure. It is known (see, e.g., [18]) that under an external loading action, the network compliance should increase at low temperatures and decrease at high temperatures, which is in agreement with the experimental dependence shown in Fig. 3.

As one can see from this figure, another prediction of the theoretical model also agrees with the experiment; namely, the compliance grows as glucose is introduced into the examined systems. According to Fig. 3, this occurs within the temperature interval 36 °C <  $T$  < 42 °C.

## 7. Conclusions

The influence of glucose on the cartilage tissue structure has been determined in the framework of a model where cartilage tissue is considered as a porous medium with a rod-plate framework. The rods are collagen fibers. The plates (layers) are formed by

swollen proteoglycan chains. The axes of the fibers are directed perpendicularly to the layer surface. The fibers are connected to the layers by collagen chains adsorbed on their surface. Such chains consist of two sections: the longitudinal section is located on the fiber surface, and the transverse one on the layer surface.

The therapeutic effect of glucose injections is provided by the following mechanism. Let cartilage tissue be damaged by an external load. Namely, let a crack emerge on the fiber surface beyond the longitudinal section of the adsorbed chain. During the injection, glucose molecules enter the pores of cartilage tissue, whence they diffuse into the proteoglycan layers. The transverse section is held on the layer surface by the bonds that exist between the links of this section in the adsorbed collagen chain and the links of polysaccharide subchains that are part of the proteoglycan chain. When glucose molecules appear in the layer, these bonds get broken, because, from a thermodynamic point of view, it becomes advantageous for the links of polysaccharide chains to form bonds with glucose molecules, which have a structure similar to these links.

The breaking of the mentioned bonds is accompanied by an increase in the free energy of the adsorbed chain. In its attempt to reduce the free energy, the adsorbed chain reduces the length of the transverse section with the broken bonds. As a result of the diffusion, the links that were previously a part of the transverse section reach the fiber surface and increase the longitudinal section length. The adsorbed chain partially “crawls” onto the fiber surface. By the moving diffusively, the adsorbed chains reach the crack, integrate into its walls, close the crack, and, thereby, eliminate the damage.

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ВПЛИВ ГЛЮКОЗИ  
НА СТРУКТУРУ ХРЯЦОВОЇ  
ТКАНИНИ: ФІЗИЧНИЙ МЕХАНІЗМ

Пропонується механізм, завдяки якому введення в хрящову тканину глюкози призводить до зміни структури цієї тка-

нини. Хрящова тканина розглядається як пористе середовище, в якому роль твердої компоненти відіграють колагенові волокна та шари, де розташовуються протеоглікани. На поверхні волокон та шарів адсорбуються колагенові ланцюги, що з'єднують згадані об'єкти. Визначено термодинамічні особливості адсорбованих ланцюгів ланки. Показано, що введені в хрящову тканину молекули глюкози частково витісняють адсорбовані ланцюги з поверхні волокон. Ці ланцюги закривають тріщини, які можуть з'являтися у волокні під дією зовнішніх навантажень, що призводить до

зменшення жорсткості протеогліканових шарів. Зроблено висновок про те, що введення молекул глюкози збільшує зсувну податливість хрящової тканини. Цей факт підтверджується результатами експерименту, виконаного на модельній системі – желатиновому гелі, структура якого вважається подібною до структури хрящової тканини.

*Ключові слова:* хрящова тканина, глюкоза, зсувна податливість.