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Optomechanical tests of hydrated biological tissues subjected to laser shaping

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Abstract**. The mechanical properties of a matrix are studied upon changing the size and shape of biological tissues during dehydration caused by weak laser-induced heating. The cartilage deformation, dehydration dynamics, and hydraulic conductivity are measured upon laser heating. The hydrated state and the shape of samples of separated fascias and cartilaginous tissues were controlled by using computer-aided processing of tissue images in polarised light.**

Keywords: *optomechanics, laser, cartilaginous tissue, fascias, plastic surgery.*

1. Introduction

Many diseases of diarthroidal joints are caused by pathological changes in the connective articulation tissue. Its physiological state, size, and mechanical properties depend mainly on the state of the interstitial fluid in the extracellular matrix [1, 2].

Among all the connective tissues, the cartilage has the most hydrated intercellular matrix. It is also known [2] that this tissue is poorly curable due to poor reparation of the cartilaginous surface. At present procedures for improving the cartilage recovery are being developed [3]. They are based on the administration of hormonal drugs into the cartilage defect and the replacement of damaged sites by implants of the artificial cartilaginous tissue grown from autological hondrocytes [4]. However, as a rule, these implants are rectangular [5]. The mechanical change in the cartilage shape produces internal stresses which return the cartilage to its initial shape.

The laser shaping of tissues [6] can be used as the final thermal treatment phase in the fabrication of implants of the specified shape. In addition, the local laser processing is used in some new medical technologies to change the size and shape of surfaces. Thus, the laser hyperthermia of the skin [7] is related to a change in the mechanics of tissue fibres, while the nonablative keratocorrecton is based on the macroscopic changes of mechanical stresses in the eye produced by laser irradiation of the sclera and cornea [8]. The mechanical properties (the elasticity, hydraulic conductivity, and shape change) of such tissues modified by laser radiation are inadequately studied at present [9].

The aim of this paper is to study the mechanical properties of hydrated biological tissues subjected to laser heating and to estimate the influence of thermal dehydration on their shape and size.

2. Materials and methods

We studied by optical methods the dynamics of the shape and geometrical dimensions, mechanical rigidity, compressibility, and hydraulic conductivity of biological tissues heated by a 1.56-μm cw Er-doped fibre laser. The tissues under study were fresh and lyophilic articulation cartilages and degreased fascias of the zygomatic part of two-year old pigs.

Cartilaginous discs of diameter 3 mm and thickness 0.7 – 0.9 mm were cut out of the subchondrial region of a bone for studies. Disc samples of diameter 2 – 4 mm and thickness 0.5 – 1 mm were prepared from flat cartilages with the help of a sampler. Tissue samples of length 2 cm, width 1 cm, and thickness 0.5 mm were cut out of fascias. The samples were stored before tests in a physiological solution at temperature 2 – 4 °C.

The deformation bending of cartilages was tested by using the setup (Fig. 1) equipped with a hemispherical transparent indenter, which was employed to perform laser irradiation of samples and measure the deformation bending.

The mechanical (static) tests gave the dependence of the bending S on the specified pressure drop Δp in cartilages during laser irradiation and in the absence of irradiation (Fig. 2). The statistic deformations of discs were produced due to the hydrostatic pressure drop obtained on the sample tissue by evacuating the volume under the sample. Evacuation was performed by a 20-mL syringe connected by a rigid tube of diameter d to the cavity under the sample. The dynamic mechanical characteristics of samples (elastic moduli and internal friction) were studied by the methods of free and induced oscillations. Variations in the dynamic mechanical characteristics of tissues during laser heating were controlled by the opto-acoustic method [10].

Figure 3 shows the cross section of cartilaginous tissue samples before and after the mechanical load and laser irradiation.

The hydraulic conductivity and elasticity of cartilaginous tissues exposed to laser radiation were studied under quasi-static conditions of penetration of the physiological fluid

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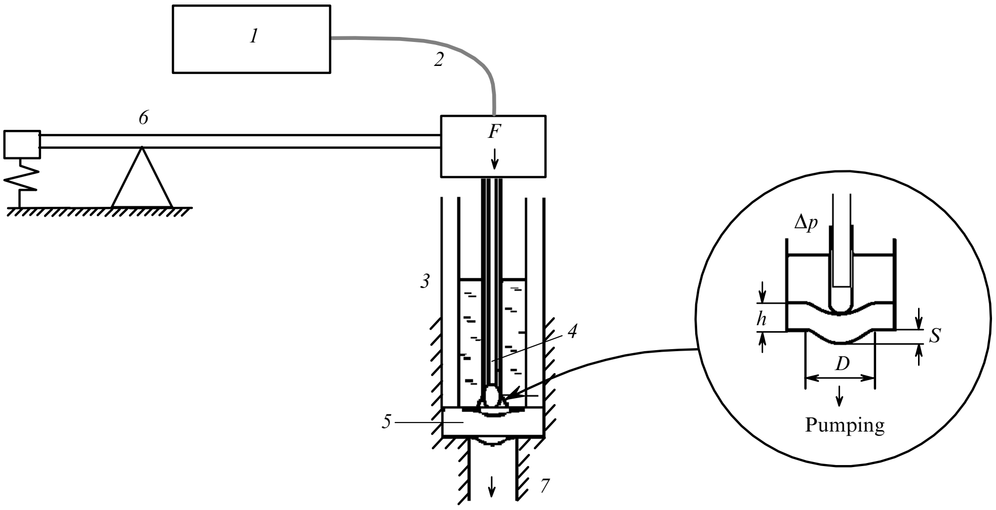

Figure 1. Scheme of the device for formation of a tissue and measuring static deformations of cartilaginous discs irradiated by a Er-doped fibre laser: (*1*) laser; (*2*) optical fibre; (*3*) vessel with a physiological solution; (*4*) transparent indenter with a fibre; (*5*) cartilage; (*6*) variable load; (*7*) pumping. The inset shows the scheme for measuring deformations: (h) cartilaginous disc thickness; (D) disc diameter; (S) sample bending; (Δp) pressure drop.

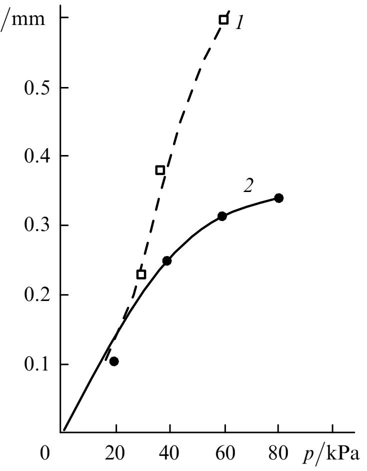

Figure 2. Pressure dependences of the cartilage deformation obtained in deformation bending tests upon laser irradiation (*1*) and in the absence of irradiation (*2*).

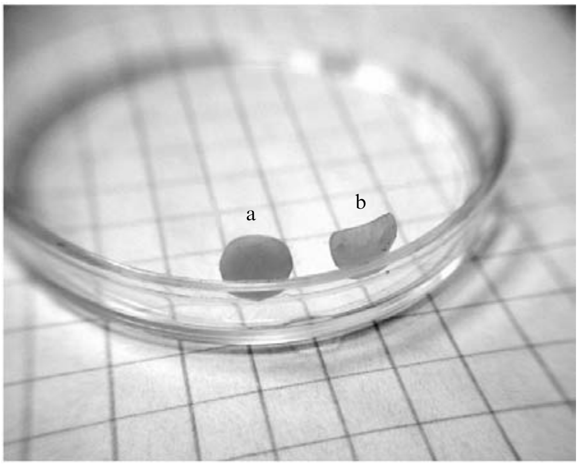

Figure 3. View of a cartilage sample before (a) and after (b) laser irradiation.

through a tissue layer at a constant hydrostatic pressure drop and a constant laser radiation power $P = 2.5$ W.

The mechanical stretching of fascias was performed under the action of a constant force 0.2–0.4 N. The stretching deformation of fascias during laser irradiation was controlled with a video system allowing the recording of a sequence of frames in a PC memory. The variation dynamics of the tissue dimensions was studied by the computer-aided processing of the images of irradiated samples.

We measured the degree of depolarisation of initially linearly polarised polychromatic radiation from a source transmitted through a cartilaginous tissue sample in liquid heated by the laser (Fig. 4). Cartilaginous tissues were imaged in the CCD camera plane by an optical system forming an optical beam with polarisation controlled by polarisers. The temperature of cartilaginous tissues was measured during their heating and the video images of samples in polarised light with crossed polarisers were stored in the PC memory.

3. Experimental results

The hydraulic conductivity coefficient in our experiments was $(1.3 - 1.5) \times 10^{-15}$ m^4 N^{-1} s^{-1} for radiation-free samples, which is consistent with the results obtained in [2] $(1.5 \times 10^{-15}$ m^4 N^{-1} s$^{-1})$, and 20×10^{-15} m^4 N^{-1} s^{-1} for irradiated samples. According to the Darsy equation, the hydraulic conductivity coefficient $K = hL/(\Delta pt)$ is determined by measuring the column of liquid of height L pumped through a sample of diameter D at a pressure of Δp (Fig. 1a) during time t.

The mechanical deformation bending tests of cartilages gave the dependence o deformations on the load up to 100 kPa (Fig. 2) for irradiated and radiation-free samples. For $\Delta p = 25$ kPa, the maximum bending was $S = 0.24$ mm.

Mechanical deformation bending tests of laser-irradiated cartilages showed that the bending deformation of cartilaginous discs increased when the indenter was pressed in the tissue during laser irradiation (Fig. 2). This suggests that the tissue is softened in the laser-irradiated region. According to the estimates performed by expressions for the membrane

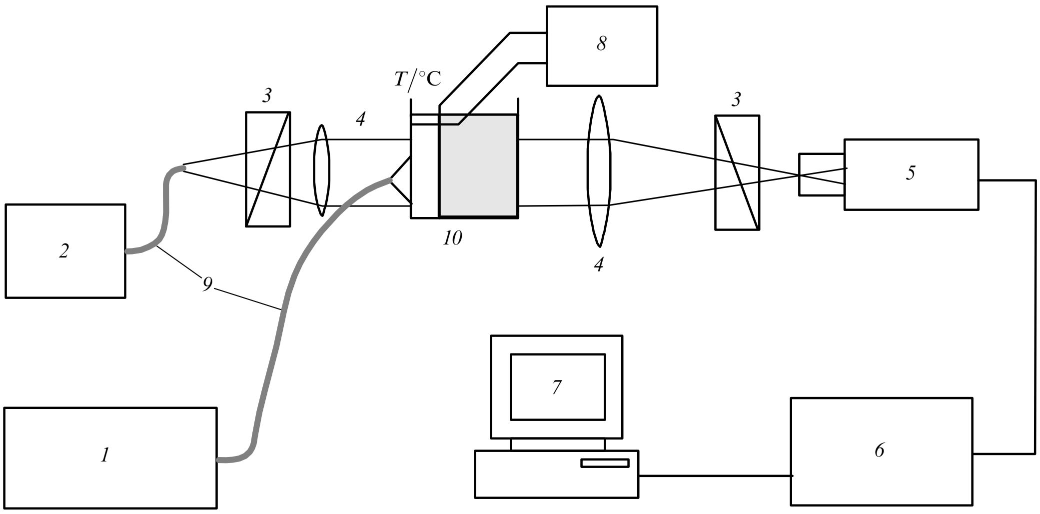

Figure 4. Scheme of the optical setup for studying birefringence in hydrated biological tissues heated by laser radiation: (*1*) laser; (*2*) radiation source; (*3*) polarisers; (*4*) optical system; (*5*) CCD camera; (*6*) ADC; (*7*) computer; (*8*) thermocouple; (*9*) optical fibres; (*10*) cartilaginous tissue in liquid.

bending [11], the elastic modulus of a tissue changes by an order of magnitude after its irradiation (the elastic modulus before and after irradiation is 0.2 and 0.021 MPa, respectively).

The processing of video images of fascias obtained upon laser heating showed that the maximum shortening of the length (up to 30 %) was achieved at low laser radiation intensities (Fig. 5). Note that after the laser was switched off, the initial length was partially recovered (by ~10 % of the laser-induced deformation).

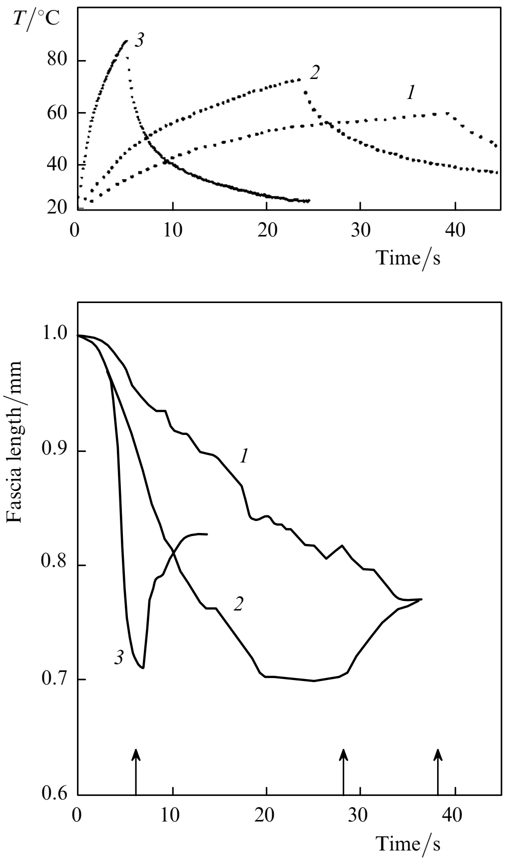

Figure 5. Changes in the length and temperature of the fascial tissue heated by radiation from a Er fibre laser in the case of a slow heating (the radiation intensity is $I = 2$ W cm^{-2}) (*1*), moderate heating (5 W cm^{-2}) (*2*), and rapid heating (10 W cm^{-2}) (*3*) (the arrows show the moments of the maximum contraction of the fascia and laser switching off).

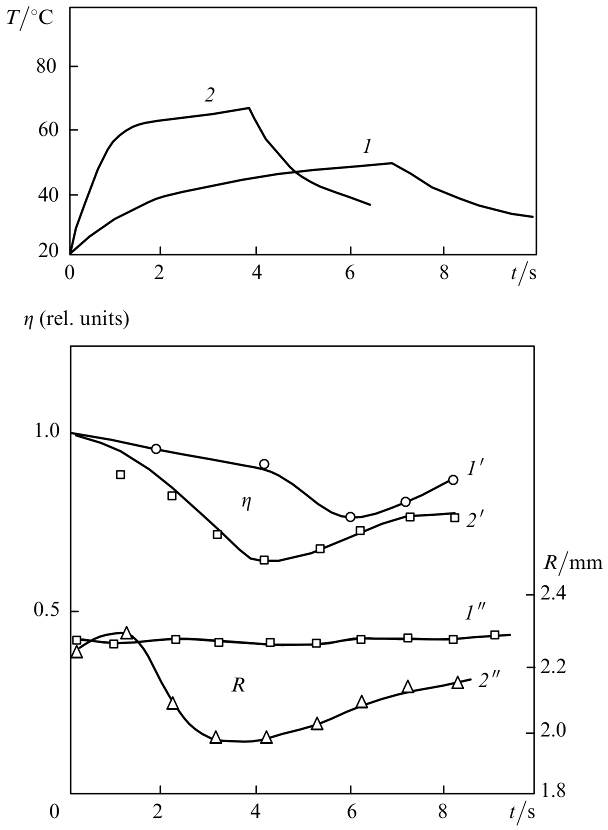

Figure 6. Changes in the temperature T (*1, 2*), the degree of polarisation η (*1′,2′*), and the cartilage size R (*1″,2″*) after the propagation of initially polarised polychromatic light through a thin cartilaginous tissue sample (disc of thickness 0.7 mm and diameter 1.25 mm) heated in a physiological solution by radiation from a Er fibre laser with intensities 100 (*1*) and 250 W cm^{-2} (*2*).

Figure 6 presents the results of processing of video images obtained in polarised light. The degree of depolarisation $(I_{\|} - I_{\perp})/(I_{\|} + I_{\perp})$ was calculated from the measured integrated intensities of the cross- and co-polarised components ($I_{\perp}$ and $I_{\|}$, respectively) of light transmitted through a sample. It was found that changes in the degree of polarisation were maximal when cartilages were heated up to 65 °C; in this case, samples experienced the maximum shortening. The change in the laser radiation intensity more than twice resulted in a decrease in the time interval from the beginning of heating to the moment of the maximum shortening of the sample size.

4. Discussion

The study of the hydraulic conductivity of flat cartilaginous discs has shown that their irradiation by a Er-doped fibre laser leads to the increase in their hydraulic conductivity coefficient by 20 – 30 times. It is known that the mechanical properties of cartilages are mainly determined by the state of water in the intercellular matrix [1]. The relaxation of stresses depends on the hydraulic conductivity of the tissue [2], and its change caused by laser heating suggests that the tissue structure has changed. The presence of microscopic pores in the tissue irradiated by a laser is confirmed by the results of atomic-force microscopy [12]. Thus, the change in the hydraulic conductivity of the tissue can be caused by the formation of channels in the tissue upon the appearance of the open porosity during laser-induced dehydration. The reduction of the tissue size can be caused by the partial denaturation of collagen and (or) the shrinkage of pores upon dehydration occurring during a prolong heating of the tissue.

5. Conclusions

Our experiments have shown that laser irradiation of hydrated biological tissues causes the dehydration of the matrix and short-term changes in a number of mechanical characteristics: the decrease in the elastic modulus of the tissue by an order of magnitude and the increase in the hydraulic permeability by 20 – 30 times; this is accompanied by changes in the tissue shape and size. The reduction of the tissue dimensions depends on the radiation intensity and achieves $\sim 20\,\%$ for fascias and $\sim 10\,\%$ for cartilages at $I \sim 10$ W cm^{-2}.

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