Influence of laser radiation on carbon nanotubes for the formation of frame materials in bioelectronics

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Abstract. Single-walled carbon nanotubes (SWCNT) nanoframes have been created in the form of structured films on a silicon substrate as well as in the bulk of biopolymers of albumin, collagen, and chitosan. Biopolymers were required to create multilayer, electrically conductive bioelectronic structures for reconstructing the layers of the heart. For this, a laser setup was used based on a pulsed fiber ytterbium laser with a wavelength of 1064 nm and a scanning system. Liquid dispersions of SWCNT in ethanol and aqueous dispersions of biopolymers were applied onto a substrate by layer spraying. Then they were irradiated with laser radiation. The effect of the binding of SWCNTs and their bundles to each other under the action of laser radiation on a silicon substrate is demonstrated. Using SEM and TEM, the formation of “T”, “X” and “Y” shaped joints in films is demonstrated. The mechanical characteristics of structured films by laser have improved. The hardness of films with nanoframe after laser exposure increases more than 6 times compared to the original SWCNT film. The specific electrical conductivity of films with nanoframe after laser exposure increases more than 7 times. The specific electrical conductivity of nanoframe in biopolymer matrices varies in the range 0.6 - 12.4 S/m, depending on the type of biopolymer. These values exceed electrical conductivity of heart myocardium. The highest roughness is shown for the lower layer of chitosan and SWCNT, and the smallest for the upper layer of albumin and SWCNT of the bioelectronic structure. Using confocal microscopy, the possibility of the formation of a cellular structure under the action of laser radiation on an aqueous biopolymer dispersion of SWCNT has been demonstrated. The cellular structure, electrical conductivity and nanoframe from SWCNT promoted better vital functions of heart cells - cardiomyocytes.

1. Introduction
Carbon nanotubes are unique material with excellent mechanical, electrical, optical, thermal and magnetic properties. They are solid-state carbon nanostructures with a crystalline structure and a tubular shape, the diameter of which is much smaller than the length [1]. It is known that carbon nanotubes (CNT) are widely used for designing nanoelectronic components, in particular, for flexible sensory systems [2].

Single-walled carbon nanotubes (SWCNT) are used to produce efficient field-effect transistors with a channel width ~10 nm, which can be built into nanoelectronic systems [3]. Recently, it has been determined that use of nanostructures from bonded carbon nanoparticles is effective. These nanostructures include SWCNT hybrid fibers bonded along their entire length by graphene sheets [4]. Such hybrid nanostructures have been used to create efficient electric accumulators. In past two...
decades, CNT has been used to create electronic emitters. However, due to chaotic arrangement of nanotubes along the entire length in disordered systems, as well as insufficiently structured CNT ends in vertically oriented arrays, developments do not reach large-scale application. To solve this problem, laser technology for structuring vertically oriented CNT arrays has been developed. Laser action allows to straighten nanotubes at their ends, as well as to bind CNTs located next to each other. This leads to decrease in work and increase in current strength with decreasing voltage [5].

Unique electronic properties of CNTs have made it possible to achieve great success in creation of bioelectronic devices. On the basis of multi-walled and single-walled CNTs, bioreversible neural electrodes for electrocorticography and measurements of subdermal encephalogram [6], strain-sensitive elements for implantable alternators [7], ultrasensitive endocardial pressure sensors [8] and biochips for detecting biomolecules [9] have been created.

Bioelectronic devices are most useful for diagnosing and repairing electrically conductive tissues such as peripheral nerves, spinal cord and brain, as well as heart muscle (myocardium) [10-12].

In this work, we investigated nanoframe materials made of carbon nanotubes potentially suitable for the creation of promising materials for bioelectronics. It was found that structured nanotubes better support the adhesion and proliferation of heart cells as compared to randomly oriented [13]. To ensure biocompatibility and bioresorbability of structures, they must contain natural components such as proteins and amino sugars [14]. Addition of single-walled carbon nanotubes (SWCNT) to composition will make it possible to create nanoframe structures on their basis under the action of laser radiation [15-17]. Mechanism of nanoframe formation is associated with presence of defects on the surface of nanotubes that arise during their synthesis. These defects are places where nanotubes are bound to each other [18].

2. Preparation of SWCNT films

Thin SWCNT films were deposited to the substrates by spraying method. SWCNT were synthesized by the electric arc method, carboxylated, and were delivered in the form of a paste with a nanotubes content of ~ 2.5 wt.%. The length of nanotubes was in the range 0.3–0.8 μm, and the average diameter was 1.4–1.6 nm.

![Figure 1](image1.png)

**Figure 1.** Cuvette with SWCNT dispersion (a) and setup (b), used for deposition of SWCNT films on substrates: tube with dispersion (1), spraying system (2), camera (3), heating stage (4), sample (5).

Dispersion of SWCNT in ethanol was prepared. For this, required number of nanotubes was calculated and a solvent was added. Ultrasound was used to disperse and separate the SWCNTs bundles in the dispersion. Processing in submersible homogenizer and in ultrasonic bath was carried out at a temperature of 30–37 °C for 5 minutes both. Nanotubes concentration was 0.01 mg/ml. The resulting dispersion was applied to pre-treated silicon oxide (SiO₂) substrate. Substrate was treated with acetone, ethanol, and distilled water in an ultrasonic bath for 5 minutes each to improve the
adhesion of nanotubes. Also, the substrates were treated with ultraviolet light for 20 minutes. Films deposition was carried out using setup, shown in Figure 1. Dispersion from tube (1) was fed into the spraying system (2), which applied dispersion according to a given program. Camera (3) recorded application process. In the process of deposition, stage (4) with the sample (5) was heated, which made it possible to dry the layer of SWCNT dispersion after each pass. Thus, 2000 layers of SWCNT dispersion were deposited on silicon substrates.

3. SWCNT nanoframes

SWCNT film image is presented in Figure 2a. Also structure of SWCNT film was studied the by scanning electron microscopy (SEM) (Figure 2b). Sample structure was studied using a FEI Helios NanoLab 650 scanning electron microscope. Accelerating voltage of electron column was 2 kV, and the current of the electron probe was 21 pA.

![Image](image1.png)

**Figure 2.** Sample of SWCNT film on the SiO$_2$ substrate (a), SEM image of SWCNT film (b).

In image with x240000 magnification, SWCNT bundles are clearly visible. The diameter of bundles reaches 40 nm. This indicates that action of solvent ethanol is not sufficient for complete mixing and separation of nanotubes.

Samples with nanotubes were processed using a system based on an ytterbium fiber laser with a radiation wavelength of 1064 nm. To create nanoframe materials, it is necessary to form structures with a given area. Since laser had a pulsed type of radiation, a uniform pattern of sample surface irradiation was created in accordance with radiation parameters: duration of one pulse 100 ns, pulse frequency 30 kHz. Figure 3 shows diagram of laser system in which laser beam trajectory (4) on SWCNT film deposited on a silicon substrate (5) is a square 1.25x1.25 mm in size, consisting of individual laser pulses with a spot diameter of 35 μm. Distance between the centers of spots is 17 μm. Positioning of laser beam on the sample surface with nanotubes was carried out using a system of two rotating mirrors – a galvanometric scanner (2). Beam speed was 500 mm/s. Collecting lens (3) was used to focus the beam.

To achieve the effect of nanowelding and to study the effect of laser radiation power on the mechanical properties of nanotubes, SWCNT film was processed with power value 1.2 W.

As a result of high intensity laser treatment, the effect of nanowelding of SWCNTs was achieved. The picture of the silicon substrate with a film of SWCNT visible square region formed by laser radiation (Figure 4a). SEM image demonstrate the formation of SWCNTs network after processing of film (Figure 4b-d). Such network consists of welded individual nanotubes and SWCNT bundles. In places of cross and perpendicular SWCNT joints, “T” shaped joints (Figure 4b), “X” and “Y” shaped joints were formed (Figure 4b).
Figure 3. Scheme of laser system, used for nanowelding of SWCNT: 1 – pulsed laser, 2 – scanner mirrors, 3 – condensing lens, 4 – laser irradiation pattern, 5 – substrate with nanotubes.

Figure 4. Image of a silicon substrate with a SWCNT film (a), SEM image of SWCNT nanoframes as a result of ytterbium laser processing at 1064 nm and TEM images with highlighted areas of nanotube and nanotube bundles junctions formed by laser radiation (c, d).

It is known that high temperatures contribute to destruction of SWCNTs and C–C bonds with subsequent formation of new bonds [5–8]. High-intensity laser radiation heats nanotubes to high temperatures around 1800°C. This contributes to formation of welded SWCNTs networks. In addition to the effect of nanowelding, new chemical bonds are formed on the contact surfaces of heated nanotubes. This is evidenced by presence of small particles on the surface of the nanotubes in Figure 4.

Formation of “T” shaped joint occurs upon the formation of atomic bonds at the end of single walled carbon nanotube with atoms in the places of defects in nanotube wall. “X”-shaped connection of two SWCNTs is formed when nanotubes are crossed at the moment of laser irradiation. “Y” connection is formed by connecting of three ends of SWCNT. Also, creation of contacts between
nanotubes is possible due to amorphous carbon formation between nanotubes under the action of laser radiation.

3.1. Electrical conductivity of nanoframes
Since insulated nanotubes have high electrical conductivity, a nanoframe made from such nanotubes must exhibit high electrical conductivity values. To measure electrical conductivity of obtained SWCNT films, four-probe contact conductivity measurement method was used. Squares were scratched inside initial and laser treated areas. Results of measuring the electrical conductivity of the SWCNT film are presented in Table 1.

| Specific electrical conductivity (initial film), S/m | Laser power, W | Specific electrical conductivity (laser irradiated film), S/m |
|---------------------------------------------------|----------------|----------------------------------------------------------|
| 15                                                | 1.0            | 18                                                       |
|                                                  | 1.2            | 112                                                      |
|                                                  | 1.4            | 27                                                       |

Based on obtained data, it can be concluded that initial SWCNT film had conductivity value of 18-98 S/m. Treatment with laser power 1.2 W led to conductivity value increase up to 112 S/m, which corresponds to increase in electrical conductivity more than 7 times compared to the initial SWCNT film. This effect of electrical conductivity increasing indicates the formation of nanoframe media from SWCNTs, which is confirmed by data obtained by the SEM method.

3.2. Hardness of nanoframes
To study the effect of laser radiation power on the mechanical properties of the sample, experimental hardness tests were carried out. To obtain more accurate hardness estimate, minimum of 5 measurements were performed on the surface of each film section. Applied load was 60 mN.

Hardness measurements were performed on initial and laser irradiated areas of SWCNT film. Hardness measurements results depending on the power of laser radiation are shown in Table 2.

Based on the obtained data, it can be seen that sample area, irradiated with power 1.2 W has improved hardness. This can be explained by the fact that in this area effect of nanowelding between nanotubes was achieved, which led to an increase in hardness. However, with a further increase in power, a decrease in hardness was observed, which can be justified by the fact that a high laser power leads to sublimation of nanotubes. Therefore, the area irradiated with power 1.4 W has the lowest hardness.

| Hardness (initial film), GPa | Laser power, W | Hardness (laser irradiated film), GPa |
|------------------------------|----------------|--------------------------------------|
| 0.178                        | 1.0            | 0.226                                |
|                              | 1.2            | 1.117                                |
|                              | 1.4            | 0.453                                |
4. Electrically conductive implants with SWCNT nanoframes for restoration of heart tissues

4.1. Samples for cardiovascular tissues restoration
The components of the dispersion for fabrication of electrically conductive cardiovascular structures were single-walled carbon nanotubes with high purity, taken with concentration of 0.01 wt.% in the paste form. Albumin (25 wt.%) was used in lyophilized form, collagen (1 wt.%) was used as suspension, and chitosan (2 wt.%) was used as powder. Three dispersions of carbon nanotubes in tridistilled water solution of proteins albumin, collagen and aminosugar chitosan were prepared. Then the dispersions were evaporated by laser radiation in layers. The result was composite structures with carbon nanoframe in rubber-like state. Electrical conductivity of such samples decreased in comparison with nanoframe (Table 1), but was still high (0.6 - 12.4 S/m). These values exceeded electrical conductivity of heart myocardium.

4.2. Confocal microscopy
The investigation of the surface of the samples was carried out with laser confocal microscope Lasertech VL 2000 DX. The wavelengths of the laser radiation source used for investigation were 405 nm, 488 nm, 561 nm. The layer structure was formed by mesh of about 50 µm in size, which corresponded to the given trajectory of the laser during formation (Figure 5). The mesh can be formed based on the geometric parameters of the cells to be seeded. When selecting the geometric characteristics of the mesh and cells, it will be possible to achieve their location in the lower parts of the mesh and the binding between the biopolymer molecules and the cell membranes.

![Figure 5. Confocal microscopy images of layers with SWCNTs and a – albumin, b – collagen, c – chitosan.](image)

4.3. Atomic force microscopy studies
The study of each of the structure layers was carried out by atomic force microscopy. As a result of the experiment, it is possible to obtain the relief of the investigated surface with a high resolution. The measurements were carried out on a Bruker atomic force microscope by the semicontact method in the Peak Force mode. Areas of 5 to 8 µm were examined in detail (Figure 6).

The bottom layer, consisting of chitosan and carbon nanotubes is the roughest, and the top layer of albumin with nanotubes is the smoothest according to data obtained with AFM. The presence of irregularities on the surface positively affects the cell adhesion to the material. So, roughness of the lower layers is a positive feature, because nutrients and growth factors can be difficultly delivered to the bottom. And smoothness of the upper layers is associated with a decrease of damage to blood elements.
4.4. In vitro investigations

To assess the effect of the samples on the proliferation of heart cells - cardiomyocytes - they were incubated with the samples in CO$_2$ atmosphere at a temperature of 37°C for 72 hours. After that, the samples were stained with ethidium bromide. Fluorescent images of cells were obtained using an Olympus BX43 microscope (Figure 7).

![Images of cardiomyocytes on samples: a, b – experimental, c – control (cover glass).](image)

In the case of cardiomyocyte cells, there are groups of cells (up to 5–6 cells) located nearby. On the coverslip, which is a control, the cells are at a greater distance from each other. Cardiomyocytes in a native heart are capable of transmitting electrical impulses and contracting synchronously; therefore, the close arrangement of cells favours the formation of gap junctions between them and providing of the contractile function of the newly formed cardiac tissue.

5. Conclusion

SWCNT nanoframes have been created in the form of structured films on a silicon substrate as well as in the bulk of biopolymers of albumin, collagen, and chitosan. For this, a laser setup was used based on a pulsed fiber ytterbium laser with a wavelength of 1064 nm and a scanning system. The effect of the binding of SWCNTs and their bundles to each other under the action of laser radiation on a silicon substrate is demonstrated. The formation of “T”, “X” and “Y” shaped joints in films is demonstrated. The mechanical characteristics of structured films by laser have improved. The hardness of films with nanoframe after laser exposure increases more than 6 times compared to the original SWCNT film. The specific electrical conductivity of films with nanoframe after laser exposure increases more than 7 times. The specific electrical conductivity of nanoframe in biopolymer matrices varies in the range 0.6 - 12.4 S/m, depending on the type of biopolymer. The highest roughness is shown for the lower layer of chitosan and SWCNT, and the smallest for the upper layer of albumin and SWCNT of the bioelectronic structure. The possibility of the formation of a cellular structure under the action of laser
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