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In-vivo Studies of Ultrasound-activated Drug-loaded Porous Silicon Nanoparticles for Cancer Therapy Application

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Abstract

It is investigated the therapeutic efficacy of combined action of ultrasound and porous silicon nanoparticles loaded with anticancer drug doxorubicin by using an experimental cancer model of lung Lewis carcinoma in vivo. Time dependences of growth of the primary tumor with introduced nanoparticles and without them, as well as the life span of mice after exposure to therapeutic ultrasound with intensity of 1W/cm^2 and frequency of 1 MHz were studied. The obtained results show the effectiveness of inhibiting the growth of primary tumor site, as well as slowing the process of metastasis, in the case of combined action of ultrasound and drug-loaded porous silicon nanoparticles that indicates the prospect of latter as sonosensitizers and nanocontainers for the delivery and controlled release of drugs in sonodynamic therapy of malignant tumors.

Keywords: silicon nanoparticles, nanocontainers, medical ultrasound, sonodynamic therapy, sonosensitizers

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1. Introduction

Sonodynamic therapy (SDT), which is one of the modern approaches to the treatment of malignant tumors, is based on an application of ultrasound irradiation (USI) with relatively low intensity, whose action is locally enhanced by using special substances, i.e. sonosensitizers [1]. When nanoparticles (NPs) are used as sonosensitizers, the therapeutic efficacy of USI occurs due to heating effect (hyperthermia), acoustic cavitation and sonochemical reactions of generation of active radicals nearby NPs [1, 2]. Among various biomedical nanomaterials silicon nanoparticles (SiNPs) are low toxic and biodegradable [3, 4]. Developed internal surface of porous SiNPs is favorable for

a substantial reduction of the threshold of acoustic cavitation that is promising for SDT [5]. Nanometer size particles can accumulate in tumor tissues in a natural way, penetrating through the pores in the blood vessels of malignant tumors, the size of which ranges from 100 to 300 nm. To deliver drug-loaded SiNPs into the cells the former can be exposed to USI with pressure amplitude exceeding the threshold of inertial cavitation. Additionally, drug-loaded SiNPs covered with thermally sensitive polymer can be used for controlled drug release under external heating [6], which can be also induced by therapeutic USI [7]. Our work is aimed to in-vivo studies of porous SiNPs as sonosensitizers for SDT and nanocontainers for anticancer drug, whose release can be triggered by therapeutic USI.

2. Materials and methods

The study of biological tolerance and biological efficiency of SiNPs were performed in mice with well-developed transplantable LLC tumors in the later stages of their growth. We use nanoparticles of mesoporous silicon (MPSi) prepared by standard electrochemical method [3-5] and non-porous nanocrystalline SiNPs (NCSi) formed by plasma-assisted fragmentation of polycrystalline Si powder [8]. An evaluation of the growth kinetics of primary site tumor with introduced nanoparticles and assessment of the life expectancy of animals with grafted tumors were carried out with introduced nanoparticles of both types.

All animal experiments were carried out in accordance with the principles of working with laboratory animals (NIH Rules No. 85-23, revised in 1985) and the European Convention for the Protection of Animals used for Experimental and other scientific purposes (Strasbourg, 18.III.1986, protocol ETS 170). In the experiments, linear mice (C57Bl / 6, BDF1) were used, which were transplanted with Lewis lung tumor (LLC). The tumor appeared spontaneously, as carcinoma of the lungs of C57B1 / 6 mice in 1951. It is transplanted for 12-14 days of growth, the average life expectancy of animals is 28 ± 2 days. Hematogenically metastasizes to the lungs in virtually 100% of cases.

Transplantation of a solid tumor was carried out with a homogenate of tumor tissue in a sterile solution of medium 199. The donor animals were sacrificed and pieces of the tumor were excised without necrotic sites and ground. The resulting tumor mass was diluted with medium 199 and administered to mice intramuscularly in 0.2-0.4 medium 199 for cell culture. The LLC suspensions were intramuscularly injected into the left hip muscle of male C57BL mice. After reaching a tumor volume of 210 ± 30 mm^3 on the 8th day after inoculation, suspensions of SiNPs (1 mg/mL) were

injected intratumoraly (5 injections of 0.1 mL. The time interval after the administration of nanoparticles and ultrasound was 15 min. The ultrasound was performed using a MedTeco standard medical device at a frequency of 1 MHz with an intensity of 1 W /cm^2 for 6 minutes. Before the impact of USI, the tumor zone was depilated. To improve the contact between the head of the radiator and the tumor zone, a standard medical gel was used for ultrasonographic ultrasonography (Mediagel Geltek, Russia). In order to control the temperature dynamics of the tumor growth during USI the following approaches were used: (i) functional microwave thermometry before and after performing the ultrasonic action; (ii) contact temperature sensor of the digital medical precision thermometer directly during USI.

The control group of mice did not receive nanoparticles and no USI was performed. The results were statistically processed using Student's t-test with a reliability of 0.95.

3. Results and discussion

In the case intratumorally injected MPSi NPs the biological effect of therapeutic importance was evaluated by analyzing the life expectancy increase and by the criterion of inhibition of growth of tumors. The average life expectancy of laboratory animals of the experimental group was 18% more than in the group of intact control (mice with inoculated tumor LLC is not subjected to any exposure) as it is shown in Fig.1. A study of the kinetics of tumor growth showed that in mice of the experimental group, the primary site of the tumor grew slowly than in the control group throughout the observation period. The difference in the percentage of inhibition of growth of tumors reached 31% to 27 day from the beginning of the development of tumors (see Fig.2).

Our experiments performed with nanoparticles of non-porous structure (NCSi) showed that the average duration of mice lifetime of the experimental group was 16% more than in the control group, and the difference in the percentage of inhibition of the tumor was about 26%. It was found that intravenously administered NCSi nanoparticles did not induce any immediate or remote toxic effects of therapeutic significance.

In order to evaluate an effect of USI the following groups of animals were investigated:

1. Mice subjected to the action of ultrasonic radiation with the previously entered intratumorally MPSi NPs.
2. Mice subjected to the action of ultrasonic radiation with the previously entered intratumorally MPSi NPS loaded with doxorubicin.

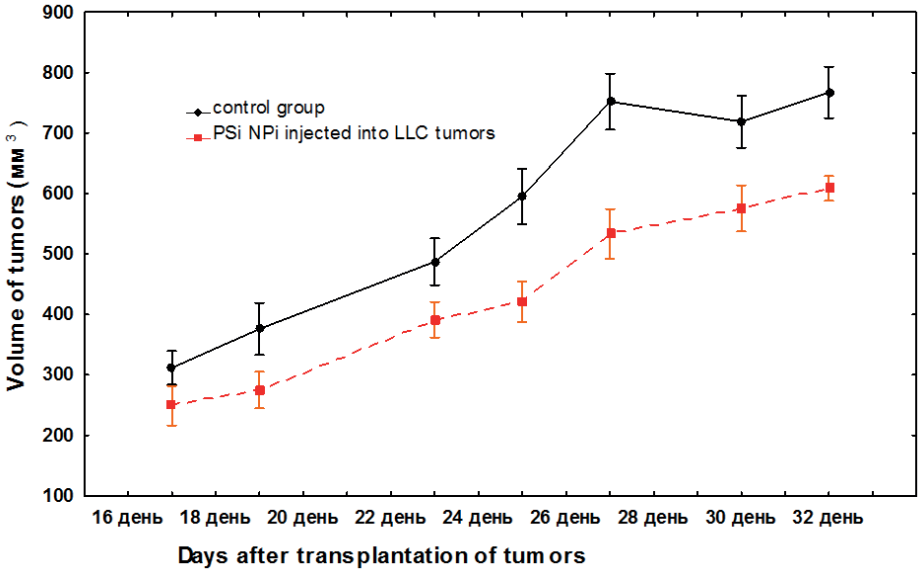

Figure 1: Tumor growth kinetics for the mice group with injected MPSi NPs and for the control one.

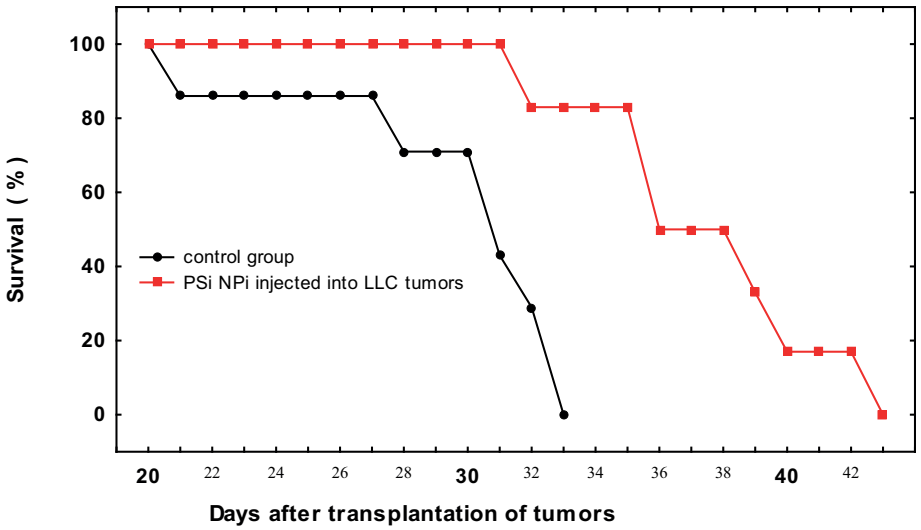

Figure 2: The survival rate of mice with injected MPSi NPs and for the control group.

3. Mice with pre-entered intratumorally MPSi NPS loaded with doxorubicin without USI.

Animals of the first group exhibited a significant biological effect of MPSi NPs as sonosensitizers. The average life expectancy of animals in this group increased by 23% relative to the control group and the growth inhibition of tumors reached 47% in maximum (Figs. 3 and 4). MPSi NPS loaded with doxorubicin led to a stronger effect of the ultrasound treatment. The growth inhibition of tumors was 60% at maximum (Fig. 5).The average life expectancy of animals in this group increased by 30% relative to the

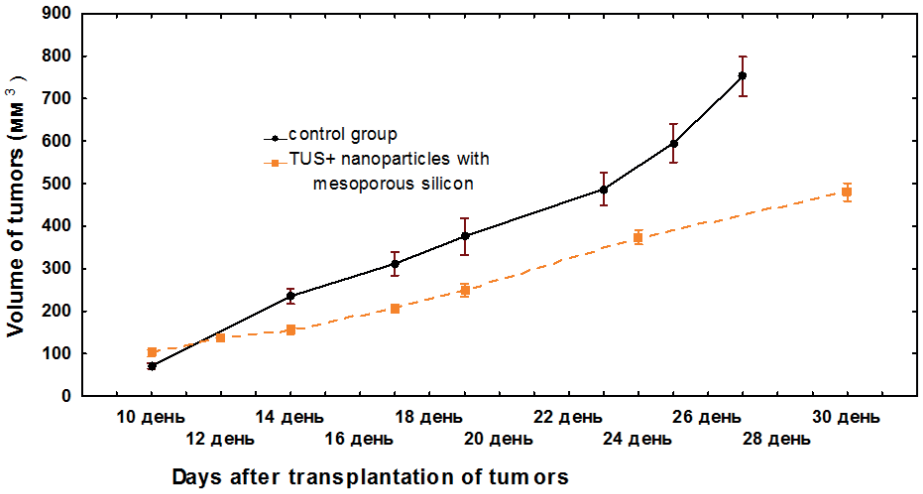

Figure 3: Tumor growth kinetics for the mice with injected MPSi NPs followed by USI and for the control group.

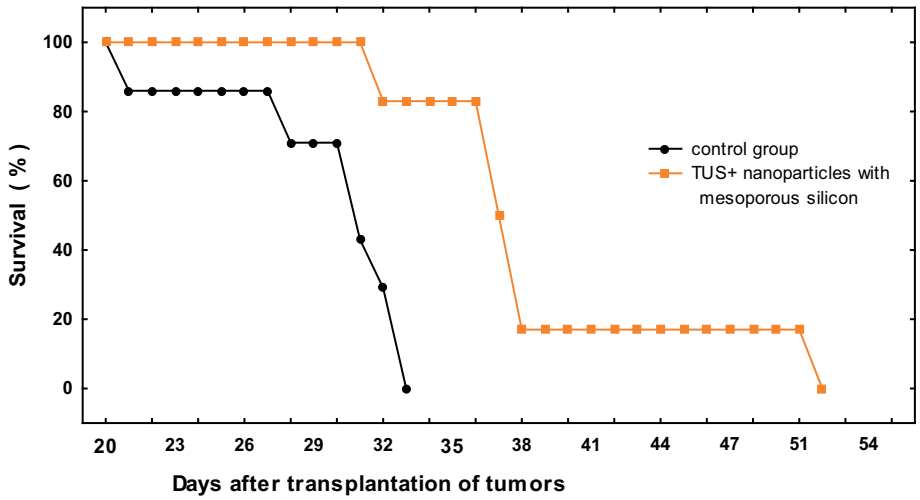

Figure 4: Survival rate of mice with introduced intratumorally MSi NPs followed by USI and the same for control group.

control group (Fig. 6). Thus, in contrast to the control group, the death of animals begins only in the later stages of cancer development. Animals of the third group (intratumoral introduction porous silicon nanoparticles loaded with doxorubicin without conducting ultrasonic treatment) observed growth inhibition of tumors by 49% and increase life expectancy by 24%.

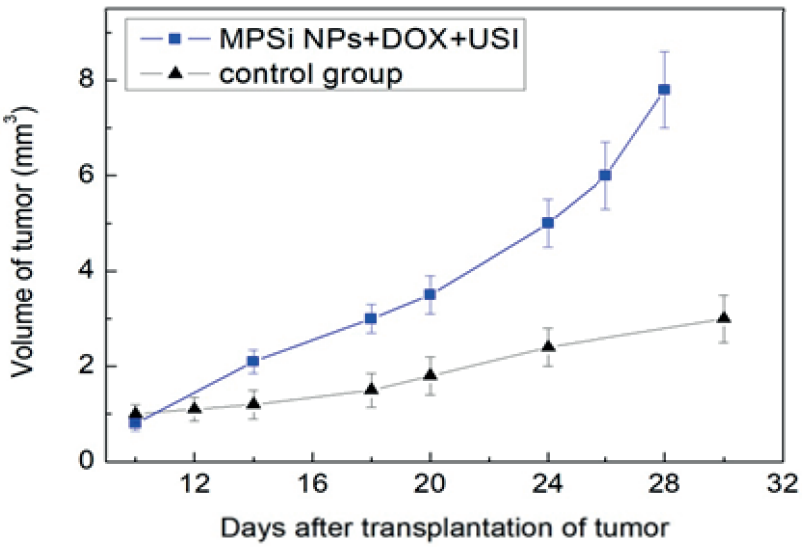

Figure 5: Kinetics of growth of tumors with doxorubicin-loaded MSi NPs followed by USI.

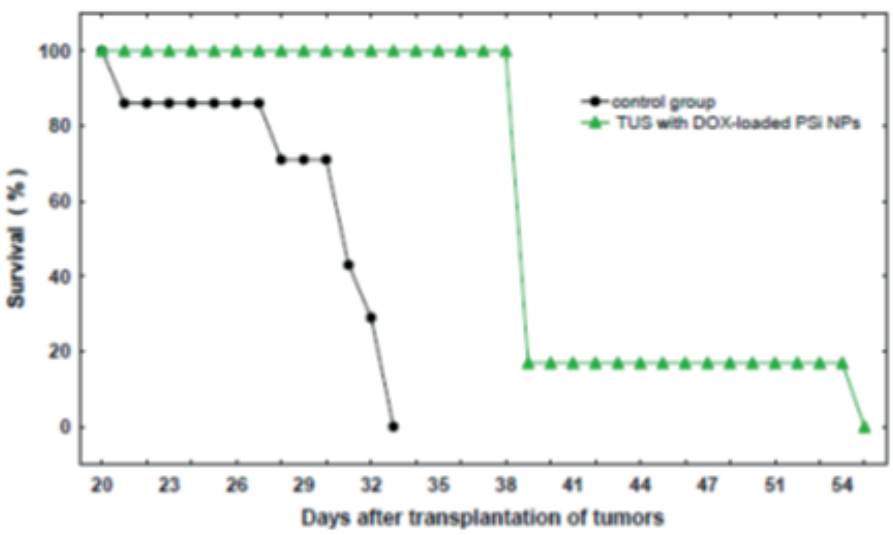

Figure 6: Survival rate of mice after USI with introduced intratumorally MSiNPs loaded with doxorubicin and for the control group.

4. Conclusions

The obtained results have revealed the biological tolerability of porous and non-porous silicon nanoparticles under interstitial or intravenous administrations. The most pronounced therapeutic effect was achieved when conducting sonodynamic therapy with the specified parameters in the case of porous silicon nanoparticles load with doxorubicin that demonstrates new opportunities for the use of biocompatible silicon nanoparticles in sonodynamic therapy of malignant tumors.

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References

[1] N. Yumita, N. Okuyama, K. Sasaki, S. Umemura. Sonodynamic therapy on chemically induced mammary tumor: pharmacokinetics, tissue distribution and sonodynamically induced antitumor effect of gallium-porphyrin complex ATX-70. Cancer Chemother. Pharmacol. V.60, 891-897 (2007).

[2] A. K. W. Wood, C. M. Sehgal. A review of low-intensity ultrasound for cancer therapy, Ultrasound in Med. and Biol., 41, 905–928 (2015).

[3] L T Canham. Nanoscale semiconducting silicon as a nutritional food additive. Nanothechnology, 18, 185704 (2007).

[4] A. D. Durnev, A. S. Solomina, N. O. Daugel-Dauge, A. K. Zhanataev, E. D. Shreder, E. P. Nemova, O. V. Shreder, V. A. Veligura, L. A. Osminkina, V. Yu. Timoshenko. Evaluation of genotoxicity and reproductive toxicity of silicon nanocrystals. Bull. Exp. Biol. Medic. 149, 445-449 (2010).

[5] A.P. Sviridov, L.A. Osminkina, A.L. Nikolaev, A.A. Kudryavtsev, A.N. Vasiliev, V.Yu. Timoshenko. Lowering of the cavitation threshold in aqueous suspensions of porous silicon nanoparticles for sonodynamic therapy applications, Appl. Phys. Lett. 107, 123107 (2015).

[6] K. Tamarov, W. Xu, L. Osminkina, S. Zinovyev, P. Soininen, A. Kudryavtsev, M. Gongalsky, A. Gaydarova, A. Närvänen, V. Timoshenko, V.-P. Lehto. Temperature responsive porous silicon nanoparticles for cancer therapy – spatiotemporal triggering through infrared and radiofrequency electromagnetic heating. J. Contr. Release, 241, 220-228 (2016).

[7] A.P. Sviridov, V.G. Andreev, E.M. Ivanova, L.A. Osminkina, K.P. Tamarov, V.Yu. Timoshenko. Porous silicon nanoparticles as sensitizers for ultrasonic hyperthermia, Appl. Phys. Lett., 103, 193110 (2013).

[8] A.A. Ischenko, G.V. Fetisov, L.A. Aslanov, Nanosilicon: Properties, Synthesis, Applications, Methods of Analysis and Control, Ch.5, CRC Press, Taylor& Francis Group, Boca Raton, London, New York, 2015.