Some Effects of Nanohyperthermia on the Basis of Lanthanum-Strontium Manganites (La,Sr)MnO₃ Nanoparticles as Heating Mediators

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Abstract

The aim of the study was to synthesize (La,Sr)MnO₃ nanoparticles as heating mediator and evaluate the ability of nanohyperthermia to affect ascitic forms of cancer as well tumor cells resistant to cytostatic. Nanoparticles have been synthesized by sol-gel method and magnetic fluid on the basis of nanoparticles was prepared. It is heated under alternating magnetic field (300kHz, 7.7kA/m) and able to heat tumor cells. Nanohyperthermia is able to enhance the sensitivity of cisplatin resistant tumor cells (MCF-7Cp) to cytostatic action: about 30% of resistant tumor cells could overcome the resistance to cisplatin. Antitumor effect of nanohyperthermia was confirmed by the absence of ascitic fluid in peritoneal cavity of mice with lymphocytic leukaemia P-388 (ascitic form). It may be explained by the ability of nanoparticles to penetrate into the tumor cells and to cause the phenomenon so named as "intracellular hyperthermia" as it was demonstrated by electron microscopy. Applying of nanohyperthermia using (La,Sr)MnO₃ nanoparticles may be considered as the promising way to help in approaches to the problem of tumor cell resistance as alternative to the conventional chemotherapies. Nanohyperthermic intraperitoneal chemotherapy may be helpful within various multimodality strategies for prevention and treatment peritoneal carcinomatosis or malignant ascites.

Keywords: (La, Sr)MnO₃ nanoparticles; magnetic fluid; nanohyperthermia; drug resistance, ascitic fluid, electron microscopy

Introduction

Cancer is one of the most undiscovered and unsolved problems of modern medicine. In spite of slow improvement of standard techniques based on surgery, chemotherapy, radiation or their combination, there are many attempts to apply different types of alternative therapy concepts, for instance, different types of hyperthermia (HT). HT of human malignant tumor is used in the number of countries in the combined antitumor therapy and demonstrates the significant modifying effect that results in the improvement of patient's survival [1-4]. HT is used for cancer treatment by selective heating the tumor tissues up to 43 – 45°C. This treatment exploits the fact that tumors are more sensitive to heat than normal tissues. Tumor cell vitality is reduced already at 39°C and sharply suppressed under 40-42°C while healthy tissue remains vital under 44-46°C [5,6]. A large body of medical evidence shows that when hyperthermia is used in combination with radiation therapy or chemotherapy there is a dramatic improvement in response rates [7,8]. However, HT application is partially limited by the tumor localization in particular the treatment of patients with tumor of internal organs. Currently used devices for tumor hyperthermia operating at different frequencies of the electromagnetic field do

not allow reaching of homogenous tumor heating, especially the deep-seating one as well as ascitic forms of cancer [9]. Therefore, there is increasing interest in the development of novel and safe materials that exhibit the desired heating effects. Nanotechnology exploited in the magnetic hyperthermia by means of applying magnetic nanoparticles (NPs) allows solving such important problem. Nanoscience has witnessed spectacular progress in the past years. Among the various nano systems under active investigation in the biological and pharmaceutical sector, magnetic nanoparticles are possibly at the forefront because of the range of their potential applications: drug delivery systems, magnetic resonance imaging contrast imaging, magnetic separation, magneto-fluids, and hyperthermia applications [10].

Local induced HT with magnetic NPs having magnetic properties (nanohyperthermia, NH), which are able to be heated under an external alternating magnetic field (AMF) and to heat the surrounding tumor tissues, without changing of their properties, offers much ampler scope for the application of HT in the therapy of malignant neoplasms [11-13]. Magnetic NPs which generate

heating in an AMF were applied *in vitro* experiments and in animal models of head and neck cancer, malignant glioma, prostate cancer, malignant melanoma and lymphoma [14-19]. It was shown the effective application of magnetic NPs in combination with AMF in the experiment with transplanted tumors both in the regime of thermal ablation or combined with chemotherapy [20-21] as well as clinical trials for prostate cancer and brain tumors [22,23].

Such NPs must meet a number of requirements for successful application to be weakly agglomerated and single-domain ones (the size of single-domain NPs for La-Sr manganites is < 70 nm) superparamagnetic properties, biocompatibility and have the ability to be heated under the action of an AMF to certain temperature [24,25]. In this process, ferromagnetic NPs should be injected into the tumor with further heating in AMF. One of the most important problems of magnetic nanohyperthermia is temperature control inside the tumor. Overheating may damage normal tissues near the tumor, whereas tumor may not be destroyed when insufficient heat lower than 42°C is generated. It difficult to monitor non-invasively the temperature of deep-sited tumors inside a patient's body in real time.

At present majority of investigations are focused on magnetic iron oxides Fe₃O₄ (magnetite) and γ-Fe₂O₃ (maghemite) which have been proved to be well tolerated by the human body and may be used for the tumor heating [26-28]. Being in ferromagnetic state magnetic NPs are heated through absorbed magnetic energy and their heating ability is virtually absent in paramagnetic state. Therefore, to use of Fe₃O₄ NPs for nanohyperthermia has a number of disadvantages and the main one is high Curie temperature value (approx. 580°C) which is a cause of overheating of healthy tissues surrounding the tumor [29].

Heterosubstituted manganite's of lanthanum-strontium (La, Sr)MnO₃ with perovskite structure became an alternative object of intense scientific attention because they may be used in HT for heating of tumors, including deep-seated ones. The samples of (La, Sr) MnO₃ manganite's were obtained by some researchers, and their ability to be heated in the AMF was shown [30-34]. They crystallize in a perovskite structure and have a number of advantages over Fe₃O₄ and γ-Fe₂O₃ point before Curie temperature of (La, Sr) MnO₃ is in a range of 20-90°C [35]. It allows to create effective HT inducers on the basis of ferromagnetic NPs with heating up to 43-44°C and maintaining this temperature automatically that eliminates overheating of surrounding healthy tissues and their destruction [36]. It was shown that NPs are nontoxic and possess of high biocompatibility [30,37,38]. Moreover it was demonstrated the efficacy of magnetic fluid (MF) based on NPs of (La, Sr)MnO₃ manganites as an effective inductor of nanohyperthermia in experiments with solid rodent tumors [25].

As the parameters of these nanomaterials significantly depend on the method of their obtaining the aim of this study was synthesis of (La, Sr) MnO₃ NPs manganite's and estimate whether NH based on synthesized NPs possesses the ability to affect ascitic forms of cancer as well as enhance the sensitivity of cisplatin resistant tumour cells (MCF-7Cp) to cytostatic action.

Materials and Methods

Synthesis of (La, Sr)MnO₃ NPs manganite's

Ferromagnetic NPs of (La,Sr)MnO₃ were synthesized by means of method that provided high homogeneity of obtained product [39,40]. Aqueous solutions of metal salts La (NO₃)₃, Sr (NO₃)₂, Mn (NO₃)₂ were used as starting materials. Solutions of salts were mixed in equivalent ratios with further addition of citric acid (CA) and ethylene glycol (EG) as gel-forming agents. Obtained mixture was heated to 80°C with stirring to provide polyesterification reaction and obtaining polymer gel. Amorphous product was formed after pyrolysis of obtained gel at 200°C. Additional heat treatment at 600 – 800°C for 2 hours in air was performed to obtained crystalline (La, Sr) MnO₃ NPs. Obtained NPs were studied by method of X-ray diffraction (XRD) using diffractometer DRON-4 (CuKα radiation).

Size and morphology of NPs were analyzed by transmission electron microscope (TEM) JEOLJEM-1230. Particles size distribution was calculated according TEM-images data using Image Tool 3 and Origin Pro 8.5 SR1 software packages as described in [41].

Preparing of magnetic fluid (MF)

MF based on (La, Sr)MnO₃ NPs was prepared using 0.1% aqueous agarose solution as the stabilizer to study the heating efficiency (specific loss power, SLP) of these NPs in AMF. SLP measurements were performed using magnetic coil, which provided AMF with frequencies of 100 – 400 kHz and magnitude of 3.2 – 9.3 kA/m. Heating temperature of MF was measured by fiber optic thermocouple OPTOCON (Weidman GMBH).

Human breast cancer cell lines and mice C57Bl/6

Human breast cancer MCF-7S and MCF-7Cp cell lines, sensitive and resistant to ciplatin (Cp), respectively, were used for testing of NPs of (La, Sr) MnO₃ *in vitro* Cells were provided by the Bank of human and animal tissues lines at R.E. Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology of NAS of Ukraine (IEPOR). *In vivo* studies were provided with total number of 42 mice C57Bl/6 (20-23g of b.w., male) IEPOR bread, bearing lymphocytic leukemia P-388 (ascitic form); all experiments with animals had been approved by the regional animal ethics committee.

MTT test

MTT assay with 3-[4,5, dimethylthiasol-2-1]-2,5-diphenyltetrasolium bromide (Sigma,USA) was used for the evaluation of cytotoxic action of substances in cell culture [42].

Electron microscopy

Cell microstructure was investigated by means of electron microscopy. The cells were treated according to [43]. The ultrathin sections prepared on LKB-8800 ultratome and contrasted by uranyl acetate and lead citrate were examined by JEM-100B electron microscope (Japan) with 60 kV acceleration voltage with further specimen photography. Heating of MF in experiment *in vitro* and *in vivo* was provided under the external alternating magnetic field (AMF) (300 kHz, 7.7 kA/m).

Statistical analysis

T-test used to analyze the cumulative results in 95% confidence interval, at p<0.05 significance level.

Results

Synthesis of (La,Sr)MnO₃ NPs by sol-gel method and preparation of MF.

(La,Sr)MnO₃ NPs are single-phased, crystallize in the perovskite structure (Figure 1a). According to TEM data (Figure 1b), obtained NPs of (La,Sr) MnO₃ are weakly agglomerated and their average sizes are in the range of 30 – 40 nm.

Figure 2 represents the results of studying the dependences of heating temperature of MF with various concentrations of (La,Sr) MnO₃ NPs on time of the AMF action (f = 300 kHz, H = 7.7 kA/m). Dependence of temperature vs time for MF based on Fe₃O₄ NPs obtained via criochemical method is shown for comparison [44]. As it can be observed from the curves, the heating temperature of (La, Sr)MnO₃ NPs goes to saturation and becomes constant after certain time independently on their concentration in MF. At the same time, Fe₃O₄ NPs heat up uncontrolled to high temperature.

Figure 1: XRD pattern (a), TEM-image of (La,Sr)MnO₃ NPs (b) and their size distribution (c).

Figure 2: Dependences of heating temperature vs time for MF based on NPs: 1 – Fe₃O₄, C = 10 mg/ml; 2 – (La,Sr)MnO₃, C = 50 mg/ml; 3 – (La,Sr)MnO₃, C = 40 mg/ml; 4 – (La,Sr)MnO₃, C = 30 mg/ml; 5 – (La,Sr)MnO₃, C = 20 mg/ml; 6 – (La,Sr)MnO₃, C = 10 mg/ml; 7 – (La,Sr)MnO₃, C = 5 mg/ml.

Cytotoxicity of (La, Sr)MnO₃ NPs.

Measurement of cell viability was conducted by MTT assay. MTT cellular assay is based on the fact that only cells that are alive can convert a non-toxic MTT reagent to molecule that shows absorbance at 492 nm. The study was provided with human breast cancer MCF-7S cell line. For estimation of the cell viability against nanoparticles, cells were seeded at 1x10⁴ cell/100 μl in 96-well plates and preincubated for 24 h. After MF was introduced into culture medium at different concentrations ranging as 6.2, 12.5, 25, 50, 100, 200 and 400 μg/ml. Rates of cell survival were estimated after 48 h and 72h of incubation with MF. The cells were stained by MTT to evaluate cytotoxic and antiproliferative effects of MF. It was found that cell staining by the conventional dye, in particular Sulforhodamine B resulted in the appearance of significant errors due to background staining of nanoparticles of MF.

Minimal cytotoxic dose IC₁₀ (the most sensitive cells in the population died, but not more than 10% of the cells); mean cytotoxic dose MCD – IC₅₀, where the 50% of the cells died; absolute cytotoxic dose IC₁₀₀ (minimal dose that causes the death of 100% of the cells). Obtained data concerning the cytotoxicity of MF are presented in Table 1.

IC ₁₀₀ (μg/ml)	IC ₅₀ (μg/ml)	IC ₁₀ (μg/ml)
<400.0	≈ 220.0 – 240.0	≤ 25.0

Table 1: Cytotoxicity (IC) of MF based on (La,Sr)MnO₃ NPs.

It was determined that the incubation of tumor cells (human breast cancer MCF-7S cell line) with MF at a dose of 400 μg/ml during 48 h was resulted in the death of 90% of cells, whereas doses between 100 and 200 μg/ml induced the death of not less than 40% of the cells. It was evaluated that IC₁₀₀ and IC₅₀ of MF are 400 and 220-240 μg/ml, respectively, and minimal cytotoxic dose is 25.0 μg/ml, that may be considered as therapeutic one.

Electron microscopy studies of the effects of MF based on (La, Sr) MnO₃ NPs.

Investigation of microstructural morphology of the human breast cancer MCF-7S cells cultured in DMEM with MF of (La,Sr) MnO₃ manganite in increasing doses NPs, that are more higher than IC₁₀ namely in a doses of 50.0 μg/ml and 100 μg/ml has been provided. It was shown that NPs of (La,Sr)MnO₃ manganite are able to penetrate into tumour cells and accumulate in intracellular structures, in particular in lysosomes and vacuoles (Figure 3). NPs penetrate into the cells through endocytosis and exit through exocytosis (Figure 4), that may result in the death of tumor cells, partly through apoptosis (Figure 5 and 6).

Primary and secondary lysosomes, Golgi complex, cytoskeletal system, i.e., organelles bound up with synthesis of enzymes for intercellular and extracellular decomposition of foreign material may be considered as reactive organelles that reply to nanoparticles as to the damage factor. The appearance of organelles that are responsible for cellular defense from damage factor started to be observed at a small dose of MF (25 μg/ml). But the irreversible alterations of ul-

Figure 3: MCF-7S cells. Polymorphism of vacuoles and channels of endoplasmic reticulum, phagosomes with nanoparticles. Dose of MF is 50 µg/ml. (x20000);
1 - polymorphism of vacuoles; 2 - channels of endoplasmic reticulum; 3 - phagosomes with nanoparticles; 4 - nucleus, 5 - nucleolus.

Figure 4: MCF-7S cells. Exo- and endocytosis. Accumulation of nanoparticles in the intercellular space. Dose of MF is 50 µg/ml. (x20000);
1 - endocytosis; 2 - exocytosis; 3 - accumulation of nanoparticles in the intercellular space.

Figure 5: MCF-7S cells. Death of cell. Exit of phagosomic vacuoles into intercellular space. Dose of MF is 50 µg/ml. (x10000);
1 - exit of phagosomic vacuoles into intercellular space; 2 - vacuoles; 3 - intercellular space.

Figure 6: MCF-7S cells. Death of the cell. Exocytosis, empty cytoplasm, soft net of cytoskeletal fibrils. Dose of MF is 100 µg/ml. (x20000);
1 - nucleus; 2 - cytoplasm; 3 - exocytosis.

trastructural morphology of cells (mitochondria swelling, appearance of phagosomes with different size that keep digested material, and splitting of nucleus membrane) were observed after administration of (La, Sr) MnO₃ manganite NPs at a high doses (more than 50 µg/ml).

It has allowed to conclude that NPs of MF in high doses may damage intracellular structures, in particular protein-synthesizing ones, and it can be suggested that it mediates probably their cytotoxic effect.

Experiments *In vitro*

Studies were performed in the system *in vitro* on two cell lines of human invasive breast ductal carcinoma, which are sensitive to cytostatics – MCF-7/S and resistant to cisplatin – MCF-7/Cp [45]. All cells were grown in Dulbecco’s Modified Eagles Medium (Sigma), supplemented by recombinant human insulin (0.01 mg/ml) and 10% FBS. All cultures were grown in humidified atmosphere with 5% CO₂ at 37°C. Variation of this line resistant to cisplatin (MCF-7/Cp) was obtained by growing the cells in culture medium by addition of the rising concentrations of Cp. Studied cells were analyzed in order to determine the level of their resistance with MTT vitality test every two months. Level of resistance of MCF-7/Cp cells was 4.0 at the time of investigation.

Reseeding cells was injected (the cells were dissociated by means of 1.0 mM EDTA) after formation of the dense monolayer on the cell’s substrate (the 3rd day of the growth). To estimate the cell viability in NH experiment, cells were seeded at 20×10³ cell/tube in 100 µl of DMEM supplemented with 10% serum and antibiotics with preincubation for 24 h. MF was further introduced into the culture medium at two-doses: 12.5 µg/ml and 25 µg/ml.

There were the following experimental groups:

- Sensitive cells MCF-7S; application of AMF for 30 min;
- Sensitive cells MCF-7S were treated by MF (12.5 µg/ml) with application of AMF for 30 min;
- Sensitive cells MCF-7S were treated by MF (25 µg/ml) with application of AMF for 30 min;
- Resistant cells MCF-7Cp; application of AMF for 30 min;
- Resistant cells MCF-7Cp were treated by MF (12.5 µg/ml) with application of AMF for 30 min;
- Resistant cells MCF-7Cp were treated by MF (25 µg/ml) with application of AMF for 30 min;
- Untreated cells (control).

Tubes with cells and NPs were placed into the center of the coil of AMF generator (300 kHz, 7.7 kA/m). Duration of NH was 30 min.

Temperature of culture medium with tumor cells and with MF was increased in average by 4.0°C up to 30 min under the action of AMF (300 kHz, 7.7 kA/m) since the concentration of ferromagnetic NPs in both doses in culture medium (37°C) of the samples was not great. At the same time, temperature of culture medium with tumor cells only was not changed under the action of AMF. After the heat-

ing cells were cultivated under the standard conditions during 48 h and then the rate of cell survival was estimated by counting the viable cells. Obtained results are presented in Table 2.

Cell line	Experimental groups			
	control	AMF (30 min)	MF(12.5 µg/ml) +AMF(30 min)	MF(25 µg/ml)+ AMF(30 min)
	Number of viable cells, (%)			
MCF-7S	100%	100%	62%	57%
MCF-7Cp	100%	106%*	72,4%	64%

Table 2: Survival of MCF-7S and MCF-7Cp tumor cells in 48 h after heating.

*the increase of the cell number was observed.

It was shown that the cultivation of MCF-7S tumor cells (human breast cancer, cisplatin sensitive) as well MCF-Cp (cisplatin resistant) even with low dose of MF (12.5 µg/ml) under AMF (300 kHz, 7.7 kA/m) during 30 min resulted in the decrease of the number of viable cells up to 62% and 72% in 48 h, respectively. Under NH conditions about 30% of resistant tumor cells could overcome the resistance and became unviable. It is relevant that cytostatic effect of NH was observed both for cisplatin sensitive and resistant tumor cells. This result demonstrates the realization cytostatic action of NH on tumor cells independently from tumor cell sensitivity to cytostatics, in particular to cisplatin.

Observed appearance of sensitivity to cisplatin in resistant tumor cells due to HT with MF that may be a way to decrease the level of resistance or even a way for overcoming drug resistance.

Experiments *In vivo*

The influence of MF based on (La,Sr)MnO₃ manganite on the tumor growth of transplanted tumor leukemia P-388 (ascitic form)

was tested. Leukemia P-388 was transplanted intraperitoneally (i.p.) by means of usual technique using 5.0x10⁶ cells per mouse.

Mice were divided in to the following groups: 1) control mice – untreated; 2) mice, treated with i.p administration of MF only; 3) three groups of mice exposed to action of AMF during 10 min, 15 min, 20 min, respectively, and 4) mice with combined treatment (administration of MF supplemented with application of AMF within 20 min). MF was injected i.p. in to mice at a dose of 250 mg/kg of body weight. As it was determined earlier in experiments *in vivo*, MF at doses up to 300 mg/kg on body weight as a i.p injections are not toxic [46].

Treatments were performed once on the first day after tumor transplantation. To provide AMF, mice were placed into the net-hammed in such way that body part of mouse from sternum to the groin was located within AMF.

It was determined that the administration of MF into the peritoneal cavity of P-388 leukemia-bearing mice followed by influence of AMF (300 kHz, 7.7 kA/m) on the abdomen of mice for 20 min has resulted in the absence of ascitic fluid in peritoneum of mice on the 8-9th day after tumor transplantation. The revision of peritoneum cavity has not detected the ascitic fluid (Figure not presented).

Leukemia-bearing mice which were treated by administration of MF only or by application of an AMF alone during 10, 15 or 20 min, respectively, had ascitic fluid in the peritoneum in volume range almost the same as no treated mice had. It has to be noted that the separate AMF or MF being apply alone did not affect the volume of ascitic fluid in mice. Obtained results are presented in Table 3 and Figure 7. It is important to note that AMF in used regime did not stimulate tumor growth.

Index	Control Group	MF(i.p.),	AMF (duration in min)			MF +AMF (duration 20 min)
			10	15	20	
Change in the weight of mice (%)	+ 3.35	+ 2.3	+1.75	– 1.3	– 1.7	– 7.5*
Volume of ascitic fluid (ml)	2.45±0.28 (1.8-3.2)	1.9±0.54 (0.8-2.5)	2.0±0.34 (1.6-2.5)	2.3±0,27 (2.2-2.8)	2.1±0,25 (1.9-2.4)	Ascitic fluid was absent

Table 3: Body weight of P-388 leukemia-bearing mice and volume of ascitic fluid after combined action of MF and AMF.

- The body weight loss less than 10% is considered as nontoxic for animals [47].

Figure 7: Mouse with leukemia P-388 (ascitic form) with ascitic fluid in the peritoneum.

The 7th day after transplantation. Separate both AMF or MF being apply alone did not affect ascitic fluid volume in mice. The peritoneal cavity is visibly increased indicating the presence of ascitic fluid in the peritoneum.

It may be summarized that MF injected i.p. into the mice with ascitic form of leukemia P-388 under the action of AMF resulted in the thermal effect on the tumor cells that perish under such conditions. It should be noted that this experiment shown the possibility to use NH for treatment patients with ascitic fluid in the peritoneum caused by tumor growth.

Discussion

We used sol-gel method for synthesis of single-domain weakly agglomerated NPs based on (La, Sr) MnO₃ (Figure 1). MF based on obtained NPs are heated up effectively to constant temperature under the action of AMF that is provided by presence of phase transition from ferromagnetic to paramagnetic state in this range of temperatures. NPs are heated up under the action of AMF when they are in ferromagnetic state only (Figure 2). It is possible to effect on the SLP value of NPs by changing their concentration in MF (Table 4). Application of MF based on manganites (La, Sr) MnO₃ NPs excluded the overheating of healthy tissues in NH process in contrast to MF based on Fe₃O₄ NPs, which are characterized by high Curie temperature (~580°C) and respectively is heated up uncontrolledly to high temperatures (Figure 2).

Concentration (La,Sr)MnO ₃ (mg/ml)	5	10	20	30	40	50
SLP (W/g)	23	24	26	28	32	40

Table 4: SLP values for MF based on manganite (La,Sr)MnO₃ NPs with various concentrations.

Demand of safety is prerequisite criterion for magnetic NPs: to be non-toxic and to be biocompatible for the consideration of clinical application [48]. It was determined the cytotoxicity of studied MF based on (La, Sr) MnO₃ NPs for the possibility to use MF as inductor for heating human breast cancer MCF-7S cells. It was evaluated IC₁₀₀ and IC₅₀ of MF and minimal cytotoxic dose was determined as 25 µg/ml. In vitro determination of toxicity of MF on cell lines may reveal preliminary information concerning the safety of the MF. These assays may have limited relevance, as they do not represent the real *in vivo* conditions. Taken into account the data of our previous studies concerning toxicity of MF based on (La, Sr) MnO₃ NPs in experiments *in vivo*, it can be concluded that such MF does not display any cytotoxicity: it was not observed any physical injuries or weight loss of animals, histostructural alterations of tissues both after i.p. or intra tumoral injections of MF [45]. It means that treatment is safe and well tolerated.

I was shown that developed MF on the basis of NPs is viable heating agent with extremely low cytotoxicity. MF is heated as well is able to heat the surrounding tissues under the AMF and may be considered as a potentially promising mediator of local hyperthermia. Significant antitumor effect of MF based on (La, Sr) MnO₃ NPs and AMF namely of NH was confirmed by the absence of ascitic fluid in peritoneal cavity of mice with lymphocytic leukaemia P-388 (ascitic form). It may be explained by the ability of NPs to penetrate into tumor cells and to cause the phenomenon so-called as “intracellular hyperthermia” as it was demonstrated by electron microscopy investigations. At the same time administration of MF alone into the tumour or application of AMF alone has not resulted into the changes in ascitic volume in peritoneal cavity.

Cancer drug resistance is one of the biggest challenges in clinical cancer treatment with anticancer drugs [49-51]. After repeat-

ed treatments, cancer cells that are initially vulnerable to certain drugs might stop to respond to them. In such cases the chemotherapy is rendered ineffective and either more higher drug dosages or new therapeutic agents are necessary to treatment continue. Appearance of sensitivity of tumor cells that are resistant to ciplatin (MCF-7Cp) under NH may be a way to decrease the level of tumor cell resistance in the treatment of patients with tumor recurrence.

Conclusion

It was synthesized NPs based on (La, Sr) MnO₃. On the basis of NPs MF is viable heating agent with extremely low cytotoxicity. MF may be considered as a potentially promising mediator of local nanohyperthermia. Significant antitumor effect of NH was confirmed by the absence of ascitic fluid in peritoneal cavity of mice with lymphocytic leukemia P-388 (ascitic form).

Nano hyperthermic intraperitoneal chemotherapy may be helpful within various multimodality strategies for prevention and treatment peritoneal carcinomatosis or malignant ascites.

It was shown that under NH resistant tumor cells may acquire sensitivity to cytostatics, in particular to cisplatin. In this aspect application of NPs may come to be considered as an innovative and promising alternative to the conventional tumor chemotherapies as a way for overcoming drug resistance showing enhanced therapeutic efficacy and reduced adverse side effects [52].

At present whole body HT [53], regional HT [54] or interstitial HT [55] as well as intraperitoneal hyperthermia [56] have been reported in clinical use. NH may be considered as one of the alternative methods: obtained results indicate the practicability to create the effective and safety method of NH for the treatment of human malignant tumors.

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Conflict of Interest

We declare that we have no conflict of interest.

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