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# Nanotechnologies in Diagnostics and Therapy of Oncological Diseases

**N. A. Brusentsov**

*Blokhin Cancer Research Center, Russian Academy of Medical Sciences, Kashirskoe sh. 24, Moscow, 115448 Russia*
*e-mail: brusentsov2005@yandex.ru*

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**Abstract**—Traditional pharmaceutics and therapies do not allow early and reliable diagnosis of malignant tumors and do not guarantee that a therapy optimal for each specific patient is prescribed. Traditional anticancer drugs and therapies fail to ensure full recovery of patients at late oncogenesis stages. Therefore, search for new anticancer drugs and development of their application strategies is in progress. Among recent findings we could mention magnetically controlled therapeutic and diagnostic nanopreparations (MNPs), techniques for tumor visualization and contrast agents for tumor diagnosis, as well as techniques for drug delivery to damaged cells and therapy [1–10].

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## INTRODUCTION

Properties of new preparations and their immobilization and activation in tumors and metastases are being studied and combinations of traditional pharmaceutics with magnetic preparations are being developed [2, 3, 5, 6, 8–20].

Magnetically controlled nanopreparations, which exhibit unique magnetic properties and are able to participate in biochemical interactions at the cellular and molecular levels, are used as starting components in the synthesis of contrast agents for magnetic resonance imaging [2, 3, 5, 13, 15]. Furthermore, they are also used as carriers for magnetic delivery, immobilization, and heating of traditional anticancer drugs in magnetic hyperthermia therapy [5–9, 10–18, 21–34]. Tumor targeting and immobilization of MNPs and their combinations with traditional preparations and photodynamic sensitizers [35–37] occur in an inhomogeneous constant external magnetic field [21, 23–28, 30–39].

About 1% of cells in tumor tissues are oncogenic cancer stem cells [8]. To suppress of their proliferation, drug combinations whose components affect the biomechanics of cell-cycle regulation systems, genetic apparatus of tumor cells, and cell-cycle progression and apoptosis control system are applied in polychemotherapy. Furthermore, MNPs directed to specific molecular targets are being developed [10, 20, 40–53].

Magnetohydrodynamic thermochemotherapy (MHTCT) is based on the following methods: magnetic resonance tomography (MRT) for early diagnosis [13, 50], magnetic fluid regional inductive hyperthermia (MFH) with electromagnetic heating of MNPs [7, 12–14, 49, 51–79], combined chemotherapy [32–34], organ-saving surgery [1], and radiation therapy [80–98]. The contents of preparations in animal tumor and organs during MHTCT were determined in real time by scanning animals on an electron detector [5, 10, 53, 99–104]. Clinical trials of the magnetic fluid regional inductive hyperthermia technique were conducted on patients with hepatic carcinomas, sarcomas, and metastases [54–58]. The results of treatment of prostate cancer were assessed by means of computer tomography [59–65]. Theragnostics (therapy + diagnostics) and MHTCT for defining an optimal therapy for a specific animal in real time are presented in [105–109].

The goal of the present publication is to review papers and patents in the following fields of nanotechnologies applied in experimental oncology:

– synthesis and testing of MNPs as diagnostic means and carriers for magnetic delivery, immobilization, and activation of traditional anticancer drugs in tumor;

– diagnosis, study, and suppression of oncogenesis, both spontaneous and induced by implanting tumors in experimental animals;

– clinical trials of new diagnostic and therapeutic nanopreparations and therapies.

### *Synthesis and Properties of Magnetically Controlled Therapeutic and Diagnostic Nanopreparations*

Magnetite nanoparticles ($Fe_3O_4$ and $\gamma$-$Fe_2O_3$) were used to synthesize dextran ferrite, citrate ferrite, Gd-substituted Mn–Zn ferrites, and $Mn_{1-x}Zn_x[Fe_{2-y}L_y]O$, which are candidates for hyperthermia applications [5, 10–14]. Conjugates containing drugs and nanopreparations controlled by external magnetic field, as well as preparations used as markers for cancer diagnostics were obtained. Trifunctional magnetic fluorescent bioconjugates capable of binding to cancerous cells by antibodies were synthesized from modules of nanoparticles, viruses, bacteria, and antibodies, bound to one of the two complementary proteins: barnase or barstar [10, 53]. Depending on the quantitative ratio of the modules, nanocomplexes with widely varied properties could be prepared. This made it possible to increase the local concentrations of diagnostic and therapeutic substances in the developing tumor, decrease the doses of therapeutic substances, and reduce their toxicity and enhance the therapeutic effect [5, 10, 53].

The ability of dextran ferrite and citrate ferrite to break through the brain–blood barrier was studied; testing was performed on mice and rats. Before injection of nanopreparations, animals were anesthesized with chloral hydrate (300 mg/kg). Ten male rats (body weight 250–300 g) were divided into 2 equal groups. A vertical cut was made in the neck area, soft tissues were pulled out from both sides of the cut, and the common carotid artery (CCA) was released. The CCA was ligated, after which the first-group animals were injected with 0.1 mL of a 5% dextran ferrite solution into the left CCA, and the second-group animals were injected with 0.2 mL of a 5% citrate ferrite solution into the right CCA. No heart rhythm and respiration disorders were observed.

After intravenous injection of dextran ferrite and citrate ferrite, the concentrations of these magnetic nanoparticles in brain, liver, spleen, kidney, lungs, and heart were determined by scanning the animals on an electron detector [5, 10, 53, 99–105]. Scanning on a Bruker BIOSPEC 70/30 USR biospectrotomograph provided evidence for the presence of nanoparticles in brain, liver, spleen, and kidney. Moreover, it was found that part of the injected preparations underwent endocytosis and retained endothelial cells of the CCA and other vessels for a long time. The 20th min after injection and every week over the course of 51 days, animals were scanned by means of MRT. The carotid arteries with dextran ferrite in walls were not visualized in the resulting tomograms (Fig. 1) [20].

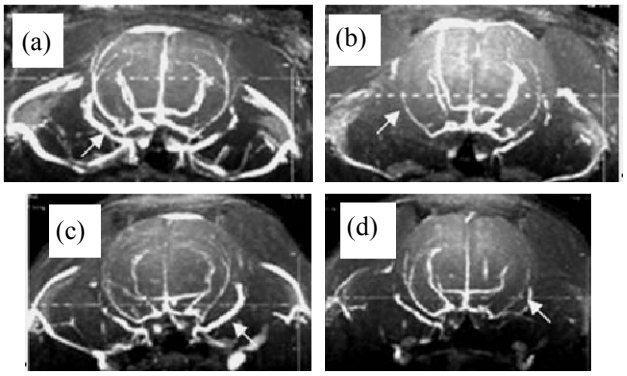


**Fig. 1.** Magnetic resonance angiograms of female Wistar rats prior to and after injection of magnetically controlled nanopreparations. Animal heads were periodically scanned over a period of 51 days: (a) prior to dextran ferrite injection (arrow); (b) 51 day after injection of 0.2 ml of 0.5% dextrane ferrite into the right common carotid artery (arrow); (c) prior to citrate ferrite injection; (d) 51 day after injection of 0.2 ml of 1.0% citrate ferrite into left common carotid artery (arrow) [20].

After intravascular injection of magnetic nanoparticles loaded with traditional anticancer drugs, endocytosis of the nanoparticles by vascular endothelial cells was observed during MHTCT. Preparations accumulated in vessel walls caused their occlusion and destroyed the vascular system [20, 98–108].

Analysis of the MRT images established that the therapy of cancer by intravenous injection of magnetic nanoparticles loaded with chemotherapeutic drugs did not lead to positive results [5, 20]. In this case, the nanopreparations are diluted by blood and entrapped (by more than 85%) by the reticuloendothelial system.

### *Delivery of Magnetically Controlled Nanopreparations to Body Tissues*

Delivery of MNPs is a physical method allowing their targeting, whatever they are loaded with drugs or not, to cell, tumor, organ, or body part [2–8, 87–92]. Passive and active targeted transport is possible. In the passive transport, the final distribution of magnetic particles is determined by their physical properties. In the active transport, the preparation binds with antigens by surface receptors of tumor cells.

Magnetic delivery of nanoparticles to tumor was performed by means of an inhomogeneous constant external magnetic field; therewith, active targeting transport was realized. For concentration and immobilization of loaded nanopreparations in tumor, magnetic bandages were used [5, 91, 92]. Treatment was completed by local inductive or magnetic hyperthermia with delayed graduated drug release, or by β-irradiation from delivered isotopes [14–18, 23–27, 87–98].

Targeted delivery of magnetic nanoparticles loaded with chemotherapeutics with minimal systemic side effects is the first element of MHTCT [5, 53, 73, 99, 100, 103, 109]. The main advantages of targeted transport of loaded MNPs over traditional techniques include higher doses of drugs delivered and retained in tumor and their lower distribution to healthy tissues and the reticuloendothelial system, as well as reduced side effects [2–7, 16–20, 87–108].

### *Improvement of the Contrast and Resolution of Magnetic Resonance Images by Combinations of Negative and Positive Contrast Magnetic Resonance Agents*

One of the challenging problems of diagnostics with the use of traditional magnetic resonance imaging (MRI) contrast agents (MW ≤ 10 kDa, hydrodynamic molecular diameter ~3.3 nm) is associated with nonselective delivery and retention of the tomographic contrast agent in the proliferation area, as well as nonspecific and short-term effects [13].

Magnevist is a systemic diagnostic contrast agent for MRT is efficient as little as 30 min after administration. The contrasting time can be prolonged by increasing the dose of magnevist, which generally entails toxic effects.

To improve the MRT image contrast and resolution, proton signals of healthy tissues, which interfere with the revealing of early pathological changes, are suppressed. The suppression of signals of tumor-surrounding tissues by means of magnetically controlled nanopreparations allows visualization and sizing of neoplasms not resorting to complicated mathematical approaches to recognition of overlapping images [13, 68].

In the research aimed at improving the contrast, resolution, and brightness of images of the damaged area, obtained using MRI contrast agents and prolonging their residence time, we injected in animals intravenously one after another 0.1–0.2 mL of a 0.5–5% sol of dextran ferrite and citrate ferrite nanoparticles (6–90 nm) in a dose of no more 2.92 mg Fe/kg body weight and magnevist (4–10 μL) and then performed $T_1$-and $T_2$-weighted gradient echo MRT imaging of the animal body. Therewith, dextran ferrite (or citrate ferrite) was injected from 6 min to 40 h and magnevist 4–6 min before MRT (Fig. 2) [5, 13, 68, 84]. Tumors, metastases, and invasion margins of tumor cells into healthy tissues were diagnosed by visual inspection of the resulting images.

Optimization of diagnostics and therapy by targeted delivery of MNPs to tumors, concentration and immobilization of nanopreparations, as well as selective noninvasive visualization of tumor tissues before and after physicochemical and therapeutic action on tumor tissues was the goal such preparations were developed and the main advantages they offer over traditional MRT contrast agents and traditional antitumor drugs. The use of optimized diagnostics and therapy in experimental oncology is exemplified by magnetohydrodynamic thermochemotherapy [2–7, 16–20, 91–108].

The improved contrast of the MRT images of biological tissues in vivo, provided by dextran ferrite and citrate ferrite, allows one to visualize the invasion of malignant cells into healthy tissue and metastases of internal organs (which was established in experiments on mice and rats). These results were confirmed by visual and hystopathological examinations. The hyspopathological observation of metastases gave grounds for essential corrections in the intensity and volume of therapy.

To map tumor-feeding vessels, intravenous injections of MRI contrast agents, for example, magnevist (Bayer Schering Pharma, Germany), were used [15]. The MRT images were used to determine the size of tumor-feeding vessels, their localization and blood feeling, as well as to optimize the procedure of

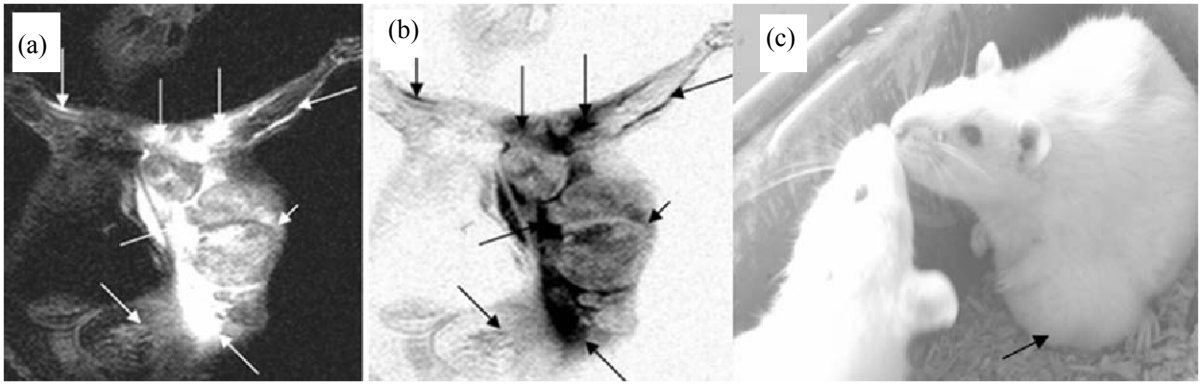


**Fig. 2.** Images of spontaneous breast carcinoma in Wistar rats: (a, b) dextrane ferrite- or citrate ferrite-enhanced MRT images at $T_2$-weighed echo gradient (500/15) scanning; (a) positive, primary tumor 6–12 min after systemic injection of nanopreparation (short arrow), tumor cell proliferation, invasion, and metastases in healthy tissues (long arrows); (b) negative of the same layer; (c) spontaneous breast carcinoma in female Wistar rat (arrow) [13, 84].

injection of MNPs. Figure 3 demonstrate the immobilization of MNPs in tumor tissue [5, 15, 68, 84].

The understanding of the molecular biologic mechanisms and principles of action of MRI contrast agents and MNPs in MHTCT allowed us to improve the procedure of early diagnosis and to enhance the therapeutic effect, which revealed itself in complete remission of tumors and increase of lifespan [5, 12–15, 41–43, 45–50, 54–65, 70–74, 76–90, 98–109].

*Combined Polychemotherapy*

Drug combinations which exhibit synergistic cytotoxicity are delivered to and activated in tumor cells by means of molecular targets for diagnostic recognition of tumor cells [10, 53]. The components of such combinations affect the biomechanics of cell-cycle regulation systems, genetic apparatus of tumor cells, and cell-cycle progression and apoptosis control system. The application of drug combinations in clinical oncology allows development of personalized combined polychemotherapy. To this end, certain combination of traditional anticancer drugs, which are introduced in strict sequence with angiogenesis inhibitors. Timely introduction of such drugs results in normalization and improvement of the functioning of the genetic apparatus of cancer cells and apoptosis. Therapeutic mistakes may entail severe disorders in the genetic apparatus of cancer cells and enhancement of oncogenesis [8, 20].

It was shown that high-dose personalized polychemotherapy led to complete destruction of cells with severe genetic disorders without considerable toxicity enhancement and development of drug resistance, and, even though complete remissions could be reached at doses reduced to 20%, part of cancer stem cells survived, which led to relapses. Whew prescribing neoangiogenesis inhibitors, the stage of the process and the changeability and viability of cancer stem cells were taken into account. For a

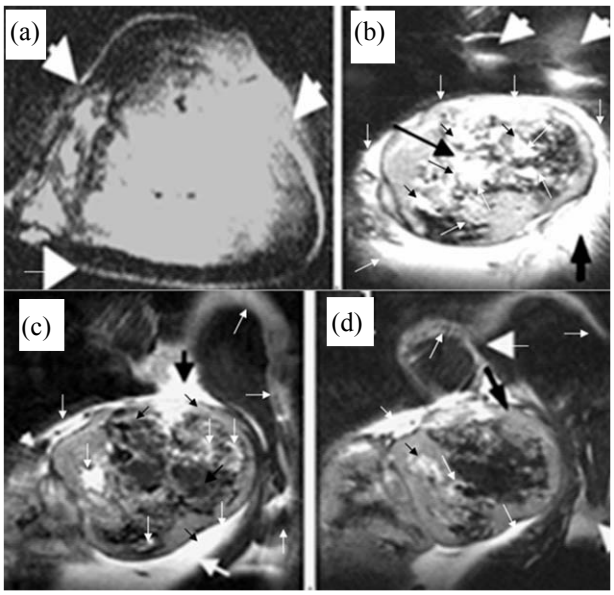


**Fig. 3.** MRT images of breast adenocarcinoma Ca 755 in C57Bl/6j mice: (a) tumor sac: tumor-feeding blood vessels (arrows); (b) two tumor-feeding vessels (white thick arrows), functioning blood vessels (black arrows); (c) surface tumor-feeding vessel enters the sac (small black arrow) and vessels (small white arrows); (d) magnevist-enriched blood coming from tumor-feeding vessels (thick white arrows) to the sac (black arrow) and functioning blood vessels (small arrows) [15].

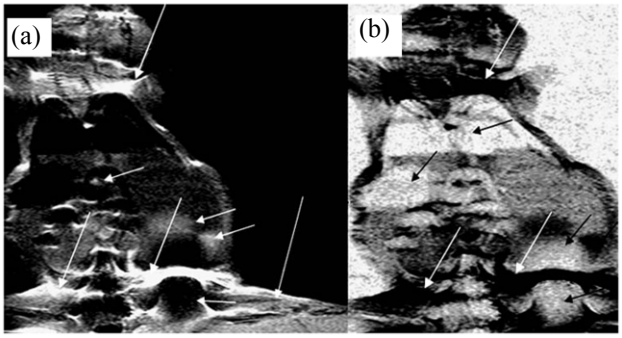


**Fig. 4.** Longitudinal MRT images of relapsed epidermoid Lewis lung carcinoma more than 600 $mm^3$ in volume, implanted in a C57Bl/6j mouse right hip muscle, enhanced with a negative contrast agent, at longitudinal *T*2-weighed echo gradient (500/15) scanning: (a) positive, 1 h after intravenous injection of dextran ferrite and citrate ferrite with additional injection of magnevist: secondary round tumors (short arrows) and chelate-like neoplasm with a shellfish-tailed body (long white arrows); (b) negative: tumor cell proliferation and invasion into internal organs (long white arrows) and metastases (black arrows) [95].

maximum anticancer effect, the choice of drugs was based on the biochemical, molecular, and pharmacokinetic mechanisms of their interactions. The administration of cytokines favors restoration of bone marrow functions lost as a result of inadequate chemotherapy and ensures safety of high-dose personalized polychemotherapy. The rate of complete remissions of solid tumors correlates with dose intensity (mg/$m^2$/week). It was established that intensified filgrastim or pegfilgrastim polychemotherapy at late stages of cancers of different localizations gives better results than standard therapy schemes [32–34].

*Cancer Thermochemotherapy*

Combinations of surgical, radiation, as well as chemo-, physio-, and biotherapeutic methods with other methods of cancer diagnosis and therapy are finding growing application in clinical oncology [1]. Chemotherapy [32–34], radiotherapy and thermoradiotherapy [44, 48, 76–83, 85, 86], and also photodynamic therapy are combined with hyperthermia [5, 35–37, 43, 45]. Regional electromagnetic hyperthermy induced by magnetically controlledми nanopreparations in alternating magnetic fields is one of the efficient techniques [7–10, 12, 13, 15, 20–30, 44–49, 64–68].

At whole-body hyperthermia the body temperature of the patient is raised using water, air, or electromagnetic field to +42.4°C [4, 38]. At that temperature many cells survive [38, 48, 82–86]. Hyperthermia in combination with reduced metabolism, varied pH, and hyperglycemia leads to weakening of cancer cells and much enhances the anticancer effect during combined radiation and drug therapy [4, 5, 38, 75, 83, 85, 86]. Before the patient's body was warmed up to +43°C, a complex of drugs, including glucose and high doses of Urotropine are injected with simultaneous deep narcosis with head cooling. Patients with generalized tumor growth take a session of whole-body hyperthermia 2.5–5.0 h long [4, 38]. As a result, the survival rate of patients increases by 10−30% compared to control groups [38, 82].

There are two methods of local electromagnetic heating of cancer tissues, used in regional hyperthermia [4, 38–41, 42, 48, 55–67, 70, 77, 78–86]: (1) direct heating (volume or induction) at frequencies of >1 MHz, when the patient's body functions as a part of the electromagnetic contour, and different body tissues warm up to different temperatures, depending on their electric properties; (2) indirect heating of cancer tissues at frequencies of 0.05–1.00 MHz, where the heating elements are ferromagnetic implants (special needs, catheters, capsules, grains, microspheres, nanoparticles) that absorb electromagnetic energy at these frequencies and transform it into heat to warm up cancer tissues, as well as traditional anticancer drugs.

Direct heating contributed little into tissue warm-up at the frequencies 0.05–1.00 MHz. The usual temperature exposure regime for regional therapy of external tumors includes maintaining temperatures at 42–46°C inside tumor from 30 min to 2 h [4, 64–67].

Hyperthermia induces primary reversible effects in cells and tissues [48, 49, 52, 77, 81–86]. Without additional therapy hyperthermia is of poor efficiency when used without other therapies. A few minutes after the hyperthermia session, tumor cells express heat shock proteins which protect the cells from thermal stress, thereby enhancing proliferation, invasions, and metastasis (Fig. 4) [86].

Hyperthermia affects the activity of regulatory proteins and cyclin-dependent kinase enzymes. This entails cell- cycle disorders up to apoptosis [52, 53, 62, 79, 83, 84, 88]. Hyperthermia induces denaturation of cell surface receptors of tumor cells, and the latter are better recognized by the immune system of the host and attacked by killer cells. The effect of hyperthermia on tumor cells *in vivo* is accompanied by changes in the microvasculature, blood circulation, and oxygen energy [35, 36, 77].

The combined effect of radiotherapy and hyperthermia on intracellular biomolecules is as-sociated with heating which induces polyfunctional reparative processes after radiation damage. If hyperthermia is performed before or after radiotherapy-induced damage, without accounting for the time factor, combined tumor therapy is less efficient. Combining hyperthermia with radio- and chemo-therapy with account for the time factor improves the results of radio- and pharmacotherapy [6, 85, 86] and results in a more than 74 % tumor regression [81, 84]. Regional hyperthermia, when experimental animal tissues warmed up above +47°C, caused thermal ablation accompanied by acute necrosis, coagulation, and, in the case of long-term exposure, carbonization of tissues. Such heating is impermissible in clinical hyperthermia because of systemic complications (hypertension, heart attack, etc.) [4, 48, 58, 70, 71, 76–81].

Over the past years new methods have become the practice of experimental oncology, including magnetic fluid regional induction hyperthermia [4–10, 12–21, 88] and magnetohydrodynamic thermochemotherapy [5, 16, 17]. The development and realization of these methods were based:

– on the methods of synthesis of magnetically controlled carriers [2, 3, 7, 11–15];

– diagnostic and therapeutic devices [4, 5, 8, 99–104];

– composite magnetically controlled antitumor agents containing traditional antitumor drugs [3, 6, 9, 11, 14, 15, 18, 26, 30];

– negative MRI contrast agents for combined diagnostics and therapy (theragnostics) of oncological diseases [25–27, 66–70, 93–99, 106–109].

Dextran ferrite and citrate ferrite [94–99] which are capable of transforming the electromagnetic energy into heat [4, 18] were used as magnetic carriers of antitumor agents.

Devices for regional hypertherapy are designed for external or intratissue operation. In experimental therapy, the whole-body heating of mammals is performed using radiofrequency (10–100 MHz) and microwave (>300 MHz) hyperthermia devices [4, 64, 83].

A BSD-2000 device equipped with a SIGMA 60 ring applicator, the electromagnetic field is generated by four pairs of antennas. Body temperature is monitored by thermistors, fiber optic sensors, or thermocouples, which are inserted into catheters 1.4–1.8 mm in diameter; the temperature probes are implanted surgically or placed on the skin [4, 78, 79].

The parts of tumor tissue, which are surrounded by vessels dissipate more heat than other tumor parts, and this circumstance complicates regional therapy and has an adverse impact on the therapeutic effect. The perfusion of liver, lung, and kidney tumors does not allow their temperature to be raised to 44–46°C and maintained at this level for a long time. The edge effects between the bone and muscle tissues, are a reason why glioblastomas are still being treated by regional hypertherapy including craniotomy.

The combined effect of regional hypertherapy and hyperthermia on intracellular biomolecules is associated with heating which induce polyfunctional reparative processes after radiation damage. Such a combined effect is weaker if heating is performed not accounting for the temperature interval between these therapies and chemotherapy before and after disorders caused by regional hypertherapy [9]. The minimal temperature of +44°C, which is required for the therapy of tumors located beneath a thick fat layer is quite difficult to reach by means of traditional devices for regional hypertherapy. In such cases, combining the three above-mentioned therapies potentiates the antitumor effects and leads to remission.

At the same time, a number of problems in the MFH technology are still to be solved. These problems are associated with the following circumstances. Blood perfusion though tumor tissue, which is enhanced by this type of hyperthermia, accelerates clearance of magnetic nano-particles; as a result, the tumor rapidly empties and does not warm up. As magnetic carriers, there were used magnetite nanoparticles ($Fe_3O_4$, Curie tem-perature $T_C$ = 585 C, and $\gamma$-$Fe_2O_3$), coated with dextran (dextran ferrite) [7, 9, 11–15, 21–30, 41–43] or citrate ions (citrate ferrite), because no magnetic nanocarriers with $T_C$ = 43–46°C have still been developed [93–98]. Magnetic fluid regional hyperthermia leaves some areas on the surface of large vessels inside the tumor and at its edges, as well as in the tumor sac, with temperatures below 45–46°C, which allows tumor stem cells to survive and induce tumor progression. The distribution of nanoparticles is not proportional to the thermal convection of tumor tissues. The temperature monitoring by means of an invasive fluorooptic method (catethers with sensors implanted in tumor tissue) may cause tumor dissemination and metastasis. Temperature control inside

tumor during the MFH session ("electromagnetic generator turn-on and turn-off") is accompanied by changes in alternating magnetic field parameters. No information on how each MFH session affects the tumor effect on the tumor is available. Conclusion on the results of treatment is drawn at the end of the full course, not accounting for the results of individual MFH sessions.

The most part of the above problems could be solved by the development of MNPs with a high specific electromagnetic absorption and Curie temperatures of 44–46°C. Such nanopreparations should automatically maintain the tumor temperature in the preset range for indefinite time. The MNPs with $T_C$ temperatures close to 45°C, synthesized by the present time, have a low specific electromagnetic absorption, and they fail to warm up to 44–46°C within 30 min [5, 11–15, 93–98].

*Magnetohydrodynamic Thermochemotherapy*

Magnetohydrodynamic thermochemotherapy of malignant tumors is based on radiotherapy and organ-saving surgery [1, 78–82], chemotherapy [32–34], and MFH [7, 12–14]. The basic principles of these methods are largely valid for the MHTCT of malignant tumors [93–98]. The earlier the tumor is diagnosed, the smaller its volume, and the earlier it has been removed, the more probable is recovery. It was shown that under MHTCT (frequency 0.88 MHz, induction 9.3 kA/m, power 0.15 kW) the temperature of skin over tumor is +44°C, and the temperature in tumor center reaches +46°C; cells in this tumor region are necrotized. However, after 3 therapeutic sessions, histological cuts of tumors revealed live stem cells at the periphery of tumors, on the surface of tumor sac and large vessels, as well as inside tumors. The survived stem cells formed tumors in subcutaneous tissue and regional lymph nodes.

After nine MHTCT sessions no live tumor stem cells were observed in histological cuts. Intratumoral injection of a dextran ferrite sol containing alkeran, followed by concentration of MNPs in tumor tissues by means of magnetic bandages [91, 92] and inductive heating led to complete regression of solid lymphocytic leukemia P388 tumors in 30% of female $BDF_1$ mice and increased their average lifespan by 180% at initial tumor volumes of 40–50 $mm^3$ [12–15]. Under the same conditions, when the initial tumor volumes were 400–500 $mm^3$, complete tumor remission was observed only in 10% of female $BDF_1$ mice [5, 12–15, 94–97].

Magnetite nanoparticles were delivered to tumor by means of a supersensitive electron navigation system which allowed one to treat tumors located deep in the brain or in the vicinity of brain parts responsible for speech and motor functions [60–65, 99–104]. Depending on tumor volume, to ensure uniform distribution MNPs, they were injected into tumor tissues which were arbitrarily divided into sectors [12–15, 21–30, 39–42, 94–99]. After repeated dextran ferrite injections, optimal MHTCT conditions were determined for each animal, considering the contents of dextran ferrite in tumor and reticuloendothelial cells [5, 98, 99]. To this end, scanning of the animals was performed [5, 100–105].

Unlike MFH, where no concentration, distribution, and immobilization of MNPs are formally envisioned, in MHTCT, the inhomogeneous constant magnetic field gradient was amplified by means of magnetic bandages [91, 92] with the aim to concentrate and immobilize MNPs in tumor tissues before heating [96–98].

In experiments on rabbits with implanted VX-2 carcinoma (volume 3500 $mm^3$), ferromagnetic nanoparticles containing the anticancer drug mitoxantrone were injected into the great femoral artery in a constant magnetic field. As a result, complete regression of tumors and lack of systemic toxicity were observed [52].

Dextran ferrite in combination with cisplatin and melphalan was applied at early stages of oncogenesis in ferrimagnetohydrodynamic thermochemotherapy in magnetic field (0.88 MHz, 7.3 kA/m, 0.15 kW, hyperthermia at 46°C for 30 min). Regression of 30% of P388 tumors with volumes of 30 $mm^3$ prior to metastasis in BDF1 mice, and the lifespan increased by up to 290%. The treatment by the same procedure of animals with 500-$mm^3$ metastasizing tumors, using citrate ferrite, increased the lifespan by 180% [5]. The high survival rate of experimental animals was provided by hyperthermia in combination with chemotherapy and photosensitization of tumor tissues [35–37, 40, 41, 65], as well as by MHTCT [86–90].

Ferrimagnetodynamic thermochemotherapy resulted in regression of adenocarcinoma 755 (volume ~45 $mm^3$) prior to metastasis in 40% of animals and lifespan increase of up to 280%. In the case of ~300-$mm^3$ tumor, the same therapy with aspiration of necrotic material and cyclophosphamide therapy of metastases

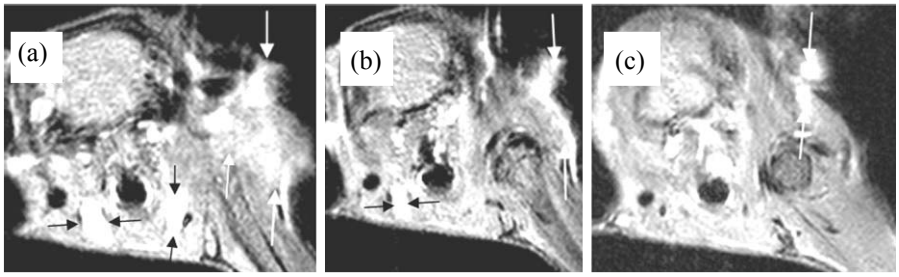


**Fig. 5.** $T_2$-Weighted MRT images of epidermoid Lewis lung carcinoma after megnetohydrodynamic thermochemotherapy in male C57Bl/6j mouse: (a) metastatic tumor at the neck lymph nodes, developed 90 days after completion of therapy prior to after-treatment (between arrows); (b) tumor regression after the first session of therapy (between arrows); (c) tumor regression after the third session of therapy (between arrows) [13, 15].

increased the lifespan by up to 200% [15, 84]. Regression of tumor with increasing number of MHTCT sessions in C57Bl/6j male mice with Lewis epidermoid lung carcinomawas obserbed by MRT (Fig. 5) [13, 15].

### *Clinical Trials of Nanotechnologies in Diagnostics and Therapy of Oncological Diseases*

Clinical trials of MFH were performed on patients with solid tumors, glioblastoma, primary hepatocellular carcinoma, prostate cancer, and liver carcinomas, sarcomas, and metastases [55–57, 59–65, 73, 76, 77, 88]. At phase I clinical trials of MNPs containing 4'-epidoxorubicin, hyperthermia therapy was given to 14 patients with solid tumors. During hyperthermia, a low toxicity of nanopreparations was noted. It was established that by the end of the course 50% of the preparation was present in liver [73, 88].

The FeRx company after phase 2−3 clinical trials of a MagneTarg drug delivery system (magnetic targeted carrier, MTC) in its advanced product MTC−doxorubicin on patients with primary hepatocellular carcinoma, which were given MFH therapy, provided evidence showing that the preparation is concentrated in the damaged part of liver by a magnet (8×2.5 cm) positioned at a distance of 15 cm from the patient's body surface [76, 77].

Phase II clinical trials of a combination of hyperthermia with radiotherapy were performed on 8 patients with liver carcinomas, sarcomas, and metastases. After targeting MNPs, the temperature inside tumor was increased to 43–50°C and maintained at this level for 60 min. The heating procedure was repeated 11 times. In 6 patients stabilization of hyperplasia was observed. The tumor volume did not increase, in 2 patients remission lasted from 9 to 14 months. The most difficult problems associated with MFH relate to nonuniform tissue distribution of MNPs on their local injection into tumor [60, 61, 77].

The targing transport of MNPs loaded with antibodies specific to breast cancer cells did not guarantee uniform nanoparticle distribution in tumor tissues, and inductive heating was still nonuniform [55–58].

The results of MFH therapy were assessed in patients with prostate cancer by computer tomography prior to and after heating of MNPs in magnetic field (100 kHz, 0–18 kA/m). Data processing using AMIRA software showed that nanoparticles concentrated in prostate all over the entire 6-week course. Efficient cooling of adjacent organs and tissues allowed hyperthermia to be performed without anesthesia. Invasive thermometry was performed in the first and sixth of the 6 sessions each 60-min long. The intraprostatic temperature varied from 40.0 to 48.5°C at 4.0–5.0 kA/m during the first session and from 39.4 to 42.5°C during the sixth session of hyperthermia. The resulting data made it possible to initiate phase I clinical trials of MFH [59].

## ACKNOWLEDGMENTS

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