International Journal of Engineering, Science and Metallurgy

(General article)

Emerging Implications of Nanotechnology on Cancer Diagnostics and Therapeutics

Sumita Thakur[a]*, Gaurav Aggarwal[a] and Pankaj Bhatia[a]

[a]Department of Biotechnology, Manav Bharti University, Solan-173229, India.

Accepted 11 July 2012, Available online 1Sept 2012

Abstract

Nanotechnology is multidisciplinary field that involves the design and engineering of objects <500 nanometers (nm) in size. The National Cancer Institute has recognized that nanotechnology offers an extraordinary, paradigm-changing opportunity to make significant advances in cancer diagnosis and treatment. In the last several decades, nanotechnology has been studied and developed primarily for use in novel drug-delivery systems (e.g. liposomes, gelatin nanoparticles, micelles). A recent explosion in engineering and technology has led to 1) the development of many new nanoscale platforms, including quantum dots, nanoshells, gold nanoparticles, paramagnetic nanoparticles, and carbon nanotubes, and 2) improvements in traditional, lipid-based nanoscale platforms. The emerging implications of these platforms for advances in cancer diagnostics and therapeutics form the basis of this review. A widespread understanding of these new technologies is important, because they currently are being integrated into the clinical practice of oncology.

Keywords: *Cancer, Nanotechnology*

1. Introduction

Nanotechnology is the exciting multidisciplinary field that involves the design and engineering of nanoobjects or nanotools <500 nanometers (nm) in size [1]. "Nano" refers to the scale of objects measured in nanometers (nm) (i.e., 1 billionth of a meter). Cancer nanotechnology seeks to characterize the interaction of nanoscale devices with cellular and molecular components specifically related to cancer diagnosis and therapy. The potential of cancer nanotechnology lies in the ability to engineer vehicles with unique therapeutic properties that, because of their small size, can penetrate tumors deeply with a high-level specificity. The National Cancer Institute has recognized this and has documented that nanotechnology offers an extraordinary, paradigm-changing opportunity to make significant breakthroughs in cancer diagnosis and treatment [2].

Over the last 2 decades, a variety of nanoscale vehicles, including gelatin [3, 4] ceramic [5] liposomes [6] and micelles [7] have been under development for therapeutic use. Preclinical models using these nanoscale tools have been documented well and have been reviewed elsewhere [6]. Nanoparticle-based drug-delivery systems offer the potential to optimize drug delivery while reducing drug or drug-carrier side effects [8]. Nanoparticle-based drug-delivery systems continue to evolve and are the subject of ongoing trials in clinical oncology. For instance, ABI-007, a 130-nm particle form of paclitaxel, has been studied in advanced breast cancer [9, 10] and other nonhematologic malignancies [11, 12].

Nanotechnology also is progressing rapidly with regard to in vivo imaging and therapeutics [1] This progress very likely will have important implications for management of the cancer patient in the near future. Recent improvements in engineering at the nanoscale level have lead to the development of a variety of new, novel nanoscale platforms (quantum dots, nanoshells, gold nanoparticles, paramagnetic nanoparticles, carbon nanotubes), which currently are under development and investigation (Table 1). These nanotools have been used for a wide variety of applications, from the detection of apoptosis through magnetic resonance imaging (MRI) [13] to sentinel lymph node mapping [14] to photothermal ablation of tumors [15]. In addition, previously developed nanoscale platforms, such as liposomes and micelles, have been modified and improved. The broad scope of these reports illustrates the versatility of nanotools and their potential wide-scale impact on improvements in cancer diagnostics and therapeutics. The emerging roles of these new platforms

*Corresponding author's email: sumitathakur159@gmail.com

for cancer imaging and therapeutic are the focus of this review.

Table 1 Uses of Nanoparticles Outlined in this Review

Type	Reported cancer-related applications
Lipid-based vehicles	Imaging; drug delivery
Quantum dots	Imaging
Nanoshells	Imaging; photothermal ablation; drug delivery
Gold particles	Imaging; photothermal ablation; radiation sensitizer; drug delivery
Paramagnetic particles	Imaging; magnetic field drug targeting
Carbon nanotubes	Imaging; photothermal ablation; drug delivery

Considerations on the In Vivo Use of Nanoparticles

Two modalities have been used to target nanoparticles to tumor sites, active and passive targeting. Active targeting involves linking ligands to nanoparticles that are tumor-specific [16, 17]. Several groups have reported the use of antibody-conjugated nanoparticles to localize cell surface proteins like c-erbB2 [18] epidermal growth factor receptor (EGFR) [19] and CA-125.20 Akermann *et al.* used quantum dots conjugated to peptides that were specific for either blood or lymphatic vessels to demonstrate specific targeting of vessels [16].

Passive targeting of nanoparticles takes advantage of the inherent size of nanoparticles and the unique properties of tumor vasculature [21, 22]. In contrast to normal endothelium, tumor vessels are lined by a simple layer of endothelium with few pericytes and smooth muscle cells [23]. Tumor blood vessels are distinct from normal vessels, in that the endothelial cells in tumors possess wide fenestrations, ranging from 200 nm to 1.2 μm [23–25]. The large pore sizes allow the passage of nanoparticles into the extravascular spaces and accumulation of nanoparticles inside tumors. Gao *et al.* demonstrated this property in a murine prostate cancer model by successfully localizing and visualizing unlabeled quantum dots at the tumor site [21].

Nanoscale objects with hydrophobic surfaces administered in vivo are taken up primarily by the reticuloendothelial system (RES) [26, 27]. This property limits the circulation time of systemically administered nanoscale objects and may hinder their intended application.3 Coating nanoparticles with hydrophilic molecules, such as polyethylene glycol (PEG), is a commonly employed strategy to overcome rapid reticuloendothelial system uptake [3, 27]. PEG modification does not appear to hinder other biological properties [16].

Lipid-based vehicles

Liposomes, micelles, and polymersomes are nanoscale lipid-based vehicles. Preclinical and clinical models using these nanoscale platforms have been documented well over the past several decades [6]. These lipid-based vehicles have been used primarily for increasing the solubility of hydrophobic chemotherapeutics and for limiting drug toxicity. However, several problems exist with these traditional vehicles, including nonspecific uptake by the RES, rapid clearance, and instability, all of which have limited the therapeutic potential of these vehicles [6, 28].

Novel preparations of these compounds recently have been developed with the objective of overcoming some of those limitations. Liposomes coated with PEG, so-called stealth liposomes, have increased bioavailability significantly because of reduced, nonspecific RES uptake [28–30]. Liposomes constructed with novel lipid polymers have resulted in significantly increased membrane stability and bioavailability [28, 31]. Micelles and liposomes coated with tumor-specific antibodies have been used for tumor targeting [32]. Liposomes that recently were synthesized with self-hydrolysable lipids may allow for time-controlled release of drugs [33]. Acoustically active liposomes that contain small amounts of air, which originally were used as air ultrasound contrast agents, have been synthesized that can package chemotherapeutics and release them in response to an ultrasound frequency [34].

Quantum dots

Quantum dots are novel semiconductor nanocrystals with broad potential for use in various applications in the research, management, and treatment of cancer [35, 36]. Quantum dots owe their fluorescence emission to electron excitation [37]. They are composed of an inorganic elemental core (e.g., cadmium, mercury) with a surrounding metal shell and have an intrinsic fluorescence emission spectra wavelength between 400 nm and 2000 nm, depending on their size and composition.16 Quantum dots possess unique optical properties that not only allow them to be tunable to discreet narrow frequencies but also are an order of magnitude more resistant to photobleaching than their organic fluorophore counterparts [38]. Dubertret *et al.* [39] demonstrated this by following fluorescent quantum dot-injected Xenopus embryos through embryogenesis. After 80 minutes of constant illumination, those investigators observed

complete photobleaching of dextran-labeled controls. In sharp contrast, there was no change in signal intensity in quantum dot fluorescent-labeled embryos. In addition, quantum dots can be prepared that are excited by a single light source but emit light or color at different wavelengths, allowing for independent labeling and identification of numerous biologic targets [21, 40]. For example, Gao et al. were able to localize 3 different quantum dots with a single light source after subcutaneously injecting them into different areas of a mouse [21].

Because of their composition of heavy metals and previous reports of cytotoxicity, the potential use of quantum dots in humans may be limited [42, 43, 46]. Uncoated or nonpolymer-protected quantum dots are unstable when exposed to ultraviolet (UV) radiation and have been shown to release toxic cadmium. [46]. Modification of quantum dots (i.e., PEGylation and micelle encapsulation) may limit the release of toxic metals in response to UV radiation [21, 39, 44, 47].

Nanoshells

Nanoshells (appproximately 10–300 nm in dimension) are composed of a dielectric core, usually silica, surrounded by a thin metal shell, typically gold [15, 48]. The optical properties of nanoshells are different from quantum dots. Nanoshells rely on the plasmon-mediated conversion of electrical energy into light [37]. Similar to quantum dots, nanoshells have the ability to be tunable optically and have emission/absorption properties that range from theUV to the infrared [37]. Nanoshells are attractive because they offer imaging and potential therapeutic properties similar to those of quantum dots without the potential for heavy metal toxicity. A potential limitation of nanoshells is their relatively large size compared with quantum dots [37].

It has been demonstrated that nanoshells may be potentially attractive vehicles for in vivo and in vitro imaging. Coating the gold surfaces of nanoshells with polyethylene glycol improved their bioavailability and their in vivo circulating half-life [45, 49, 50]. Nanoshells have been used in vivo as a contrast agent for imaging with optical coherence tomography [51] and photoacoustic tomography [49] (Table 2). Wu *et al.* were able to localize and image 20-nm gold nanoshells loaded into gel phantoms through diffuse optical tomography [52]. Nanoshells may be conjugated to immunoparticles to allow targeting to tumor-specific molecules. Loo et al. targeted HER-2 on in vitro cultures by using HER-2 antibody-conjugated nanoshells that were prepared to scatter light in the NIR spectra using optical coherence tomography [50].

Nanoshells may be constructed as simultaneous carriers of different antitumor agents. Sengupta et al. demonstrated increased in vivo antitumor activity from novel nanoshells that contained combrestatin (an antiangiogenic factor) and doxorubicin [53] and demonstrated the temporal release of these agents, which allowed for vascular breakdown (action of combrestatin) and sequestration/concentration of doxorubicin within the tumor milieu. This enhanced the antitumor activity of these agents significantly compared with combrestatin or doxorubicin alone.

Table 2 Imaging Modalities

Type	Description
Diffuse optical tomography	Near infrared-based imaging that uses body water, deoxygenated hemoglobin, and oxygenated hemoglobin to create a tomographic map of tissue
Photoacoustic tomography	Uses short-pulse lasers to generate ultrasound waves that can be used to create a tomographic map of tissue
Optical coherence tomography	Uses short laser pulses over a broad range of frequencies to generate an interference pattern that maps tissue tomographically

The ability of specifically engineered nanoshells to act as photoabsorbers with resultant heat generation has powerful potential therapeutic implications for the use of nanoshells in photothermal ablation. In vivo ablation of tumors using nanoshells that havebeen delivered either intratumorally or systemically and exposed to powerful NIR light has been reported [15, 48, 54].

Gold nanoparticles

Colloidal gold nanoparticles are another attractive platform for cancer diagnosis and therapy [17]. Gold nanoparticles are attractive because gold has been approved and used for treatment of human disease (e.g. rheumatoid arthritis) [55]. Gold nanoparticles also are attractive because they are relatively easy to synthesize [17].

Gold nanoparticles have been used as contrast agents in vitro based on their ability to scatter visible light [56].

Sokolov *et al*. successfully used gold nanoparticles conjugated to EGFR antibodies to label cervical biopsies for identification of precancerous lesions [56]. Photoacoustic tomography has been used to image gold nanoparticles to a depth of 6 cm in experiments using gelatin phantoms [57]. Based on this property, photoacoustic tomography may be useful for in vivo imaging of gold nanoparticles.

Gold nanoparticles also have been used as a platform for novel experimental cancer therapy. In a subcutaneous model of colon cancer, it was demonstrated that systemically delivered gold nanoparticles (size, approximately 33 nm) conjugated to tumor necrosis factor (TNF) accumulated in tumors [17]. Mice that were treated with TNF-conjugated gold nanoparticles had improved survival compared with mice that were treated with native TNF alone [17]. Gold particles also have been used to enhance sensitivity to external beam radiation [58]. Systemically administered gold nanoparticles (size, 1.9 nm) accumulated in a murine subcutaneous tumor model and greatly enhanced local X-ray therapy and overall survival compared with mice that received radiation alone.

Gold nanocages, a new type of gold nanoparticle, recently have been described [59]. These particles have been used to detect c-erbB2 and EGFR in in vitro assays by using NIR and optical coherence tomography [59]. Gold nanocages may be constructed to generate heat in response to NIR light and, thus, also may have a potential application in photothermal ablation [59].

Paramagnetic nanoparticles

Nanosized contrast agents are under development to improve the utility of MRI and computed tomography (CT) in imaging cancer. Superparamagnetic iron oxide contrast agents consisting of 50-nm to 100-nm particles were developed initially in the late 1980s [60]. These contrast agents are attractive because they have much greater magnetic susceptibility than traditional MR contrast agents, such as gadolinium. Such particles have rapid hepatic uptake after intravenous administration, which makes them useful for the characterization of hepatic tumors [60]. Ultrasmall, superparamagnetic iron oxide contrast agents consisting of 5-nm to 10-nm particles subsequently were characterized that had more widespread tissue distribution (because of their smaller size), allowing uptake in lymph nodes and bone marrow [61]. Ultrasmall, superparamagnetic iron oxide nanoparticles have been used clinically in humans to characterize lymph node status in patients with breast cancer [62], lung cancer [63], prostate cancer [64], endometrial cancer [65] and cervical cancer [65]. It has been shown experimentally that gadolinium-containing nanoparticles coated with folate or PEG accumulate in tumors [66]. A nanoscale paramagnetic contrast agent, G6, recently was demonstrated as useful for lymphatic mapping in a murine model [67].

Efforts currently are ongoing to develop biologically targeted nanoparticle contrast agents for cancer imaging with MRI. HER-2/*neu* is a tyrosine kinase that is expressed in many breast cancers. Breast cancer-specific, superparamagnetic nanoparticles conjugated with antibodies to HER-2/*neu* have been used to image HER-2/*neu*-positive breast cancer cells in vitro with MRI [68]. Many breast cancer cells express receptors for luteinizing hormone-releasing hormone (LHRH). Leuschner et al. recently demonstrated the in vivo detection of breast cancer cells using LHRH-conjugated, superparamagnetic nanoparticles [69]. The antigen $\beta_v\beta_3$ is expressed on neovascular endothelial cells, and tumor neovasculature has been imaged in vivo by using paramagnetic nanoparticles conjugated to a peptidomimetic antagonist of $\alpha_v\beta_3$ [70]. Telomerase is expressed in cancer cells, which gives them limitless replicative activity. The construction of nanoparticles capable of changing their magnetic state after annealing with telomerase-synthesized sequence TTAGGG has allowed the detection of telomerase activity by MRI in an experimental model [71].

The ability to monitor apoptosis in vivo may represent a method for monitoring response to cancer therapy. Ligands that are specific for apoptosis (e.g., the C2 domain of synaptotagmin and annexin 5) have been conjugated to iron oxide nanoparticles and, experimentally, have demonstrated an ability to bind apoptotic cells *in vitro* and *in vivo* [13, 72].

The magnetic properties of paramagnetic nanoparticles have been used in an effort to concentrate drug delivery to tumors. In 1 such report, investigators used magnetic fields in an effort to concentrate methotrexate-conjugated nanoparticles in tumor implanted in the hind limb of rabbits to focus greater amounts of the drug in desired areas while being able to administer lower concentrations systemically or intraarterially [73]. Although systemic administration of the novel drug conjugate did not result in any efficacy above controls, femoral artery administration with magnetic field concentration resulted in remission of the tumor at a concentration of 50% of the normally administered, systemic, chemotherapeutic dose [73]. Methotrexate alone at 50% of the standard, systemic dose or methotrexate-conjugated nanoparticles without magnetic field administration in the intrafemoral artery resulted in no tumor regression or remission [73]. This nanoparticle delivery system also was translated into a Phase I clinical trial conducted by Lubbe et al. that demonstrated patient tolerance of therapy with no magnetic field-associated toxicities and with the successful localization of nanoparticles to tumors in patients [74].

Nanoscale CT Contrast Agents

The interest in nanoscale CT contrast agents stems from the ubiquitous nature of CT already in clinical use. Most work on nanoscale contrast agents for CTimaging has focused on lymphatic mapping [75–77]. Specific imaging of diseased lymph nodes has been demonstrated [76, 77]. It is believed that the signal attenuation observed in these cancerous lymph nodes is because of architectural disruption of the lymph node and replacement of host RES by tumor cells. To our knowledge, there have been 2 clinical trials to date that assessed the safety and efficacy of a novel nanoscale liposomal contrast agent for liver CT imaging; however, because of the presence of mild-to-moderate adverse effects (i.e., chills, nausea/emesis) in a significant percentage of patients, the agent has not been developed further for clinical use [78–80].

Carbon Nanotubes

Carbon nanotubes are carbon cylinders composed of benzene rings, and their use in a variety of applications from molecular electronics to energy storage has been documented well in the literature [81]. Their use in biologic applications is evolving. For example, carbon nanotubes have been used as gene therapy delivery vectors [81–83]. Nanotubes can be made in different sizes and, at small sizes, have been shown to be internalized intracellularly through endocytosis.82 Recently, investigators have functionalized nanotubes for biologic applications by adsorbing different molecules and antigens to their surface so that they specifically may target tumor cells. Kam et al. used folic acid (FA) and fluorescent tag-conjugated, single-walled carbon nanotubes (SWNTs), a type of nanotube, specifically to target HeLa cells, in vitro [83]. HeLa cells, which are FA receptor-rich cells, actively endocytosed these SWNTs and were identified through the fluorescent tag under confocal microscopy [83]. Furthermore, SWNTs were engineered to absorb NIR light and subsequently were used for photothermal ablation applications in vitro [83]. SWNTs coupled to paramagnetic gadolinium particles are being developed for use as a high-performance MRI contrast agent [84].

Conclusion

In conclusion, the integration of nanotechnology into cancer diagnostics and therapeutics is a rapidly advancing field, and there is a need for wide understanding of these emerging concepts. The development of new nanoscale platforms offers great potential for improvements in the care of cancer patients in the near future. Areas of greatest clinical impact likely include novel, targeted drug-delivery vehicles, molecularly targeted contrast agents for cancer imaging, targeted thermal tumor ablation, and magnetic field targeting of tumors. Because nanotechnology is a rapidly progressing field, future advances in nanotechnology research and development likely will be associated with the further development of novel, high-impact approaches to cancer diagnosis and treatment.

References

1 Ferrari M. Cancer nanotechnology: opportunities and challenges. Nat Rev Cancer. 2005; 5: 161–171.
2 Exploring nanotechnology and cancer. Available at URL:http://nanocancer.gov/resource center/nano critical.asp.
3 Kaul G, Amiji M. Biodistribution and targeting potential of poly(ethylene glycol)-modified gelatin nanoparticles in subcutaneous murine tumor model. J Drug Target. 2004; 12(9–10): 585–591.
4 Kaul G, Amiji M. Tumor-targeted gene delivery using poly (ethylene glycol)-modified gelatin nanoparticles: in vitro and in vivo studies. Pharm Res. 2005; 22: 951–961.
5 Cherian AK, Rana AC, Jain SK. Self-assembled carbohydrate-stabilized ceramic nanoparticles for the parenteral delivery of insulin. Drug Dev Ind Pharm. 2000; 26: 459–463.
6 Sahoo SK, Labhasetwar V. Nanotech approaches to drug delivery and imaging. Drug Discov Today. 2003; 8: 1112–1120.
7 Torchilin VP. Targeted polymeric micelles for delivery of poorly soluble drugs. Cell Mol Life Sci. 2004; 61(19–20): 2549–2559.
8 Jain KK. Nanotechnology-based drug delivery for cancer. Technol Cancer Res Treat. 2005; 4: 407–416.
9 Ibrahim NK, Samuels B, Page R, et al. Multicenter Phase II trial of ABI-007, an albumin-bound paclitaxel, in women with metastatic breast cancer. J Clin Oncol. 2005; 23: 6019–6026.
10 Gradishar WJ, Tjulandin S, Davidson N, et al. Phase III trial of nanoparticle albumin-bound paclitaxel compared with polyethylated castor oil-based paclitaxel in women with breast cancer. J Clin Oncol. 2005; 23: 7794–7803.
11 Damascelli B, Cantu G, Mattavelli F, et al. Intraarterial chemotherapy with polyoxyethylated castor oil free paclitaxel, incorporated in albumin nanoparticles (ABI-007): Phase II study of patients with squamous cell carcinoma of the head and neck and anal canal: preliminary evidence of clinical activity. Cancer. 2001; 92: 2592–2602.
12 Nyman DW, Campbell KJ, Hersh E, et al. Phase I and pharmacokinetics trial of ABI-007, a novel nanoparticle formulation of paclitaxel in patients with advanced nonhematologic malignancies. J Clin Oncol. 2005; 23: 7785–7793.
13 Schellenberger EA, Bogdanov AJr., Hogemann D, Tait J, Weissleder R, Josephson L. Annexin V-CLIO: a nanoparticle for detecting apoptosis by MRI. Mol Imaging. 2002; 1: 102–107.
14 Parungo CP, Ohnishi S, De Grand AM, et al. In vivo optical imaging of pleural space drainage to lymph nodes of prognostic significance. Ann Surg Oncol. 2004; 11: 1085–1092.
15 Hirsch LR, Stafford RJ, Bankson JA, et al. Nanoshell-mediated near-infrared thermal therapy of tumors under magnetic resonance guidance. Proc Natl Acad Sci USA. 2003; 100: 13549–13554.
16 Akerman ME, Chan WC, Laakkonen P, Bhatia SN, Ruoslahti E. Nanocrystal targeting in vivo. Proc Natl Acad Sci USA. 2002; 99: 12617–12621.

17 Paciotti GF, Myer L, Weinreich D, et al. Colloidal gold: a novel nanoparticle vector for tumor directed drug delivery. Drug Deliv. 2004; 11: 169–183.
18 Wu X, Liu H, Liu J, et al. Immunofluorescent labeling of cancer marker Her2 and other cellular targets with semiconductor quantum dots. Nat Biotechnol. 2003; 21: 41–46.
19 Nida DL, Rahman MS, Carlson KD, Richards-Kortum R, Follen M. Fluorescent nanocrystals for use in early cervical cancer detection. Gynecol Oncol. 2005; 99(3 Suppl 1): S89–S94.
20 Wang HZ, Wang HY, Liang RQ, Ruan KC. Detection of tumor marker CA125 in ovarian carcinoma using quantum dots. Acta Biochim Biophys Sin (Shanghai). 2004; 36: 681–686.
21 Gao X, Cui Y, Levenson RM, Chung LW, Nie S. In vivo cancer targeting and imaging with semiconductor quantum dots. Nat Biotechnol. 2004; 22: 969–976.
22 Vasir JK, Labhasetwar V. Targeted drug delivery in cancer therapy. Technol Cancer Res Treat. 2005; 4: 363–374.
23 Hobbs SK, Monsky WL, Yuan F, et al. Regulation of transport pathways in tumor vessels: role of tumor type and microenvironment. Proc Natl Acad Sci USA. 1998; 95: 4607–4612.
24 Maeda H, Fang J, Inutsuka T, Kitamoto Y. Vascular permeability enhancement in solid tumor: various factors, mechanisms involved and its implications. Int Immunopharmacol. 2003; 3: 319–328.
25 Dvorak HF. Leaky tumor vessels: consequences for tumor stroma generation and for solid tumor therapy. Prog Clin Biol Res. 1990; 354A: 317–330.
26 Brigger I, Dubernet C, Couvreur P. Nanoparticles in cancer therapy and diagnosis. Adv Drug Deliv Rev. 2002; 54: 631–651.
27 Otsuka H, Nagasaki Y, Kataoka K. PEGylated nanoparticles for biological and pharmaceutical applications. Adv Drug Deliv Rev. 2003; 55: 403–419.
28 Torchilin VP. Recent advances with liposomes as pharmaceutical carriers. Nat Rev Drug Discov. 2005; 4: 145–160.
29 Blume G, Cevc G. Liposomes for the sustained drug release in vivo. Biochim Biophys Acta. 1990; 1029: 91–97.
30 Portney NG, Ozkan M. Nano-oncology: drug delivery, imaging, and sensing. Anal Bioanal Chem. 2006; 384: 620–630.
31 Xu JP, Ji J, Chen WD, Shen JC. Novel biomimetic polymersomes as polymer therapeutics for drug delivery. J Control Release. 2005; 107: 502–512.
32 Torchilin VP, Lukyanov AN, Gao Z, Papahadjopoulos-Sternberg B. Immunomicelles: targeted pharmaceutical carriers for poorly soluble drugs. Proc Natl Acad Sci USA. 2003; 100: 6039–6044.
33 Ahmed F, Discher DE. Self-porating polymersomes of PEG-PLA and PEG-PCL: hydrolysis-triggered controlled release vesicles. J Control Release. 2004; 96: 37–53.
34 Huang SL, MacDonald RC. Acoustically active liposomes for drug encapsulation and ultrasound-triggered release. Biochim Biophys Acta. 2004; 1665(1–2): 134–141.
35 Bruchez MJr., Moronne M, Gin P, Weiss S, Alivisatos AP. Semiconductor nanocrystals as fluorescent biological labels. Science. 1998; 281: 2013–2016.
36 Seydel C. Quantum dots get wet. Science. 2003; 300: 80–81.
37 Alper J. Shining a light on cancer research. NCI Alliance for Nanotechnology in Cancer USA, 2005.
38 Qu L, Peng X. Control of photoluminescence properties of CdSe nanocrystals in growth. J Am Chem Soc. 2002; 124: 2049–2055.
39 Dubertret B, Skourides P, Norris DJ, Noireaux V, Brivanlou AH, Libchaber A. In vivo imaging of quantum dots encapsulated in phospholipid micelles. Science. 2002; 298: 1759–1762.
40 Voura EB, Jaiswal JK, Mattoussi H, Simon SM. Tracking metastatic tumor cell extravasation with quantum dot nanocrystals and fluorescence emission-scanning microscopy. Nat Med. 2004; 10: 993–998.
41 Frangioni JV. In vivo near-infrared fluorescence imaging. Curr Opin Chem Biol. 2003; 7: 626–634.
42 Parungo CP, Colson YL, Kim SW, et al. Sentinel lymph nodemapping of the pleural space. Chest. 2005; 127: 1799–1804.
43 Parungo CP, Ohnishi S, Kim SW, et al. Intraoperative identification of esophageal sentinel lymph nodes with near-infrared fluorescence imaging. J Thorac Cardiovasc Surg. 2005; 129: 844–850.
44 Stroh M, Zimmer JP, Duda DG, et al. Quantum dots spectrally distinguish multiple species within the tumor milieu in vivo. Nat Med. 2005; 11: 678–682.
45 Morgan NY, English S, Chen W, et al. Real time in vivo non-invasive optical imaging using near-infrared fluorescent quantum dots. Acad Radiol. 2005; 12: 313–323.
46 Derfus AM, Chan WC, Bhatia SN. Probing the cytotoxicity of semiconductor quantum dots. Nano Lett. 2004; 4: 11–18.
47 Gao X, Yang L, Petros JA, Marshall FF, Simons JW, Nie S. In vivo molecular and cellular imaging with quantum dots. Curr Opin Biotechnol. 2005; 16: 63–72.
48 Loo C, Lin A, Hirsch L, et al. Nanoshell-enabled photonics-based imaging and therapy of cancer. Technol Cancer Res Treat. 2004; 3: 33–40.
49 Wang Y, Xie X, Wang X, et al. Photoacoustic tomography of a nanoshell contrast agent in the in vivo rat brain. Nano Lett. 2004; 4: 1689–1692.
50 Loo C, Lowery A, Halas N, West J, Drezek R. Immunotargeted nanoshells for integrated cancer imaging and therapy. Nano Lett. 2005; 5: 709–711.
51 Barton J, Halas NJ, West J, Drezek R. Nanoshells as an optical coherence tomography contrast agent, Tuchin V, Izatt J, Fujifoto J, editors. Proc SPIE. 2004; 5316: 99–106.
52 Wu C, Liang X, Jiang H. Metal nanoshells as a contrast agent in near infra-red diffuse optical tomography. Opt Commun. 2005; 253(1–3): 214–221.
53 Sengupta S, Eavarone D, Capila I, et al. Temporal targeting of tumour cells and neovasculature with a nanoscale delivery system. Nature. 2005; 436: 568–572.
54 O'Neal DP, Hirsch LR, Halas NJ, Payne JD, West JL. Photo-thermal tumor ablation in mice using near infrared-absorbing nanoparticles. Cancer Lett. 2004; 209: 171–176.
55 Mottram PL. Past, present and future drug treatment for rheumatoid arthritis and systemic lupus erythematosus. Immunol Cell Biol. 2003; 81: 350–353.
56 Sokolov K, Follen M, Aaron J, et al. Real-time vital optical imaging of precancer using anti-epidermal growth factor receptor antibodies conjugated to gold nanoparticles. Cancer Res. 2003; 63: 1999–2004.
57 Copland JA, Eghtedari M, Popov VL, et al. Bioconjugated gold nanoparticles as a molecular based contrast agent:

implications for imaging of deep tumors using optoacoustic tomography. Mol Imaging Biol. 2004; 6: 341–349.
58 Hainfeld JF, Slatkin DN, Smilowitz HM. The use of gold nanoparticles to enhance radiotherapy in mice. Phys Med Biol. 2004; 49: N309–N315.
59 Chen J, Saeki F, Wiley BJ, et al. Gold nanocages: bioconjugation and their potential use as optical imaging contrast agents. Nano Lett. 2005; 5: 473–477.
60 Stark DD, Weissleder R, Elizondo G, et al. Superparamagnetic iron oxide: clinical application as a contrast agent for MR imaging of the liver. Radiology. 1988; 168: 297–301.
61 Weissleder R, Elizondo G, Wittenberg J, Rabito CA, Bengele HH, Josephson L. Ultrasmall superparamagnetic iron oxide: characterization of a new class of contrast agents for MR imaging. Radiology. 1990; 175: 489–493.
62 Michel SC, Keller TM, Frohlich JM, et al. Preoperative breast cancer staging: MR imaging of the axilla with ultrasmall superparamagnetic iron oxide enhancement. Radiology. 2002; 225: 527–536.
63 Nguyen BC, Stanford W, Thompson BH, et al. Multicenter clinical trial of ultrasmall superparamagnetic iron oxide in the evaluation of mediastinal lymph nodes in patients with primary lung carcinoma. J Magn Reson Imaging. 1999; 10: 468–473.
64 Harisinghani MG, Barentsz J, Hahn PF, et al. Noninvasive detection of clinically occult lymph-node metastases in prostate cancer. N Engl J Med. 2003; 348: 2491–2499.
65 Rockall AG, Sohaib SA, Harisinghani MG, et al. Diagnostic performance of nanoparticle-enhanced magnetic resonance imaging in the diagnosis of lymph node metastases in patients with endometrial and cervical cancer. J Clin Oncol. 2005; 23: 2813–2821.
66 Oyewumi MO, Yokel RA, Jay M, Coakley T, Mumper RJ. Comparison of cell uptake, biodistribution and tumor retention of folate-coated and PEG-coated gadolinium nanoparticles in tumor-bearing mice. J Control Release. 2004; 95: 613–626.
67 Kobayashi H, Kawamoto S, Sakai Y, et al. Lymphatic drainage imaging of breast cancer in mice by micro-magnetic resonance lymphangiography using a nano-size paramagnetic contrast agent. J Natl Cancer Inst. 2004; 96: 703–708.
68 Artemov D, Mori N, Okollie B, Bhujwalla ZM. MR molecular imaging of the Her-2/neu receptor in breast cancer cells using targeted iron oxide nanoparticles. Magn Reson Med. 2003; 49: 403–408.
69 Leuschner C, Kumar C, Urbina MO, et al. The use of ligandconjugated supraparamagnetic iron oxide nanoparticles for early detection of metastasis NSTI Nanotechnol. 2005; 1: 5–6.
70 Winter PM, Caruthers SD, Kassner A, et al. Molecular imaging of angiogenesis in nascent Vx-2 rabbit tumors using a novel alpha(nu)beta3-targeted nanoparticle and 1.5 Tesla magnetic resonance imaging. Cancer Res. 2003; 63: 5838–5843.
71 Grimm J, Perez JM, Josephson L, Weissleder R. Novel nanosensors for rapid analysis of telomerase activity. Cancer Res. 2004; 64: 639–643.
72 Zhao M, Beauregard DA, Loizou L, Davletov B, Brindle KM. Non-invasive detection of apoptosis using magnetic resonance imaging and a targeted contrast agent. Nat Med. 2001; 7: 1241–1244.
73 Alexiou C, Arnold W, Klein RJ, et al. Locoregional cancer treatment with magnetic drug targeting. Cancer Res. 2000; 60: 6641–6648.
74 Lubbe AS, Bergemann C, Riess H, et al. Clinical experiences with magnetic drug targeting: a Phase I study with 4′-epidoxorubicin in 14 patients with advanced solid tumors. Cancer Res. 1996; 56: 4686–4693.
75 Wisner ER, Katzberg RW, Koblik PD, et al. Iodinated nanoparticles for indirect computed tomography lymphography of the craniocervical and thoracic lymph nodes in normal dogs. Acad Radiol. 1994; 1: 377–384.
76 Wisner ER, Katzberg RW, Griffey SM, Haley PJ, Johnson DK, Vessey AR. Characterization of normal and cancerous lymph nodes on indirect computed tomography lymphographic studies after interstitial injection of iodinated nanoparticles. Acad Radiol. 1996; 3(Suppl 2): S257–S260.
77 Wisner ER, Katzberg RW, Link DP, et al. Indirect computed tomography lymphography using iodinated nanoparticles to detect cancerous lymph nodes in a cutaneous melanoma model. Acad Radiol. 1996; 3: 40–48.
78 Leander P, Hoglund P, Kloster Y, Borseth A. New liposomal liver-specific contrast agent for CT: first human phase I clinical trial assessing efficacy and safety. Acad Radiol. 1998; 5(Suppl 1): S6–S8.
79 Leander P, Hoglund P, Borseth A, Kloster Y, Berg A. A new liposomal liver-specific contrast agent for CT: first human Phase-I clinical trial assessing efficacy and safety. Eur Radiol. 2001; 11698–704.
80 Hoglund P, Leander P, Hustvedt SO, Kloster Y, Borseth A. Human pharmacokinetics and modeling of the concentration-attenuation relationship of a new liposomal liver-specific contrast agent for CT. Acad Radiol. 1998; 5(Suppl 1): S47–S48.
81 Bianco A, Kostarelos K, Partidos CD, Prato M. Biomedical applications of functionalised carbon nanotubes. Chem Commun (Camb). 2005: 571–577.
82 Kam NW, Dai H. Carbon nanotubes as intracellular protein transporters: generality and biological functionality. J Am Chem Soc. 2005; 127: 6021–6026.
83 Kam NW, O'Connell M, Wisdom JA, Dai H. Carbon nanotubes as multifunctional biological transporters and near-infrared agents for selective cancer cell destruction. Proc Natl Acad Sci USA. 2005; 102: 11600–111605.
84 Sitharaman B, Kissell KR, Hartman KB, et al. Superparamagnetic gadonanotubes are high-performance MRI contrast agents. Chem Commun (Camb). 2005: 3915–3917.