Diagnostic and therapeutic applications of nanotechnology in cancer.

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PASS

Research paper
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Abstract

Nanotechnology is currently one of the most important chapters of medical research.
In this paper applications of nanotechnology in medicine and especially in cancer diagnosis and treatment will be discussed.
Cancer is one of the most frequent causes of morbidity and mortality in the UK. While the number of deaths from cardiovascular diseases is coming down over years it is not so obvious in the case of cancer, and it is assumed that in 10 – 20 years it is going to become the number one cause of death.
Current modalities of diagnosis and treatment of different diseases, and especially those applied for cancer, have some major limitations. These are connected with low sensitivity and specificity of diagnostic tools as well as toxicity of drugs and accidental damages to healthy tissues in the course of cancer therapy.
Recent developments in nanotechnology offer new opportunities coming alongside with utilization of nanoparticles for improved detection tests, targeted drug delivery and new modalities of treatment such as noninvasive thermal destruction of cancer cells.
All these developments will allow clinicians to offer patients more personalized cancer therapies. Treatment is going to be individualized to specific subtype of cancer. Specific treatments will be provided to patients who are likely to benefit from them and not to the ones, who would not respond, to avoid not justified severe side effects.
Potential side effects, particularly in the long-term use of nanoparticles are not fully recognized yet and further investigations will be required.

Introduction

Nanotechnology

Nanotechnology (Greek nano – "dwarf") was envisioned for the first time by physicist, laureate of Nobel Prize, Richard Feynman at his lecture "there is plenty of room in the bottom" in 1959.

It is defined as the study and use of structures between 1 and 100 nanometers in size; the scale of molecules like proteins and receptors antibodies.

The following illustration can help you comprehend how small 1 nanometer actually is. In reality it is 100,000 times smaller then the diameter of human hair.

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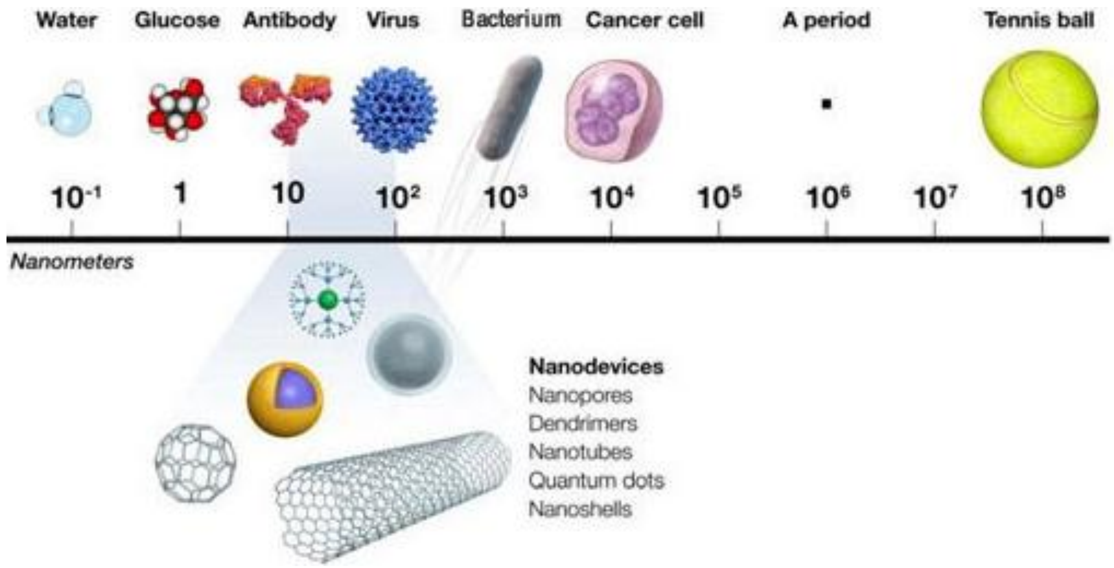

Fig.1

Scientists used to work with nanoparticles for centuries, but they were not able to see their structure until recent years, when microscopes capable of displaying structures as small as atom were developed. With better scientific understanding of processes on molecular level it was possible to create the smallest devices and use them to help in various designations.

The spectrum of applications of nanotechnology is huge, including electronics, optics, chemistry, information technology and biosciences.

Fig. 2 is summarizing all major fields nanoparticles are playing important role.

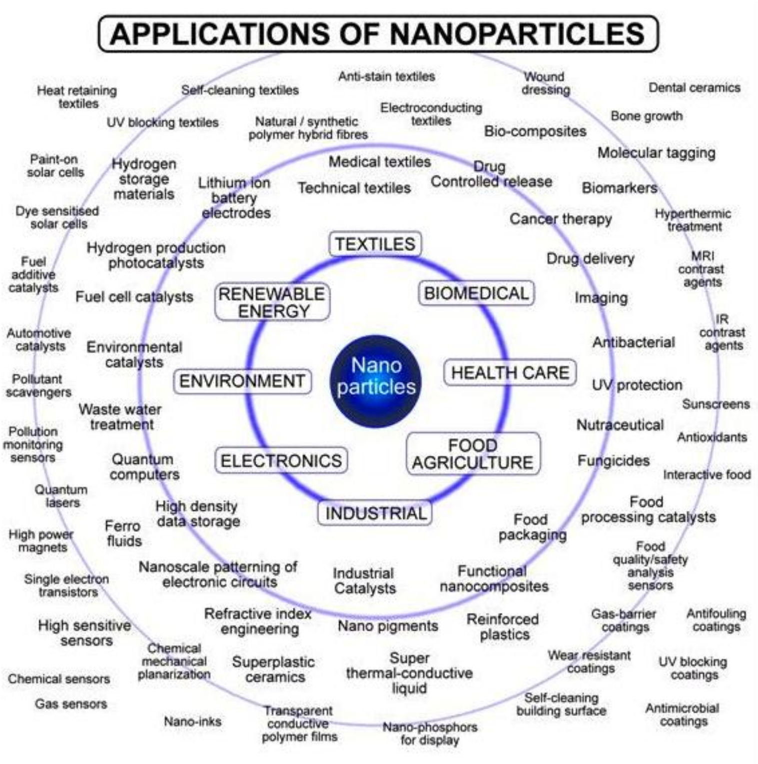

Fig.2

Because of the fact that basic elements of living organisms, components of cells, are of nano size, application of nanotechnology at this field, nano biotechnology, seems to be natural consequence.

Medicine is the field, which can benefit from utilization of nanotechnology most, as it deals with specimens on the smallest levels. On top of this development of tiny nano devices can make it possible to enter the body and explore it from inside, what used to be only science fiction even a decade ago.

There are plenty of areas in medicine, which can be revolutionized by the use of nanotechnology.

Nanobots – nano devices, which can be used for some interventions inside the body; they can be even programmed in the way to build new nanobots. Nano computers must be built to supervise the work of nanobots. They can be small enough to repair cells working on atomic/ molecular level.

They can be used in heart diseases to repair damaged heart cells as well as to remove cholesterol deposits from the inside of coronary arteries.

Nanotechnology can help in cancer treatment and diagnosis. Nanobots can be sent directly to the side of tumor to destroy cancer cells, not affecting surrounding normal tissues. With the use of nano particles drugs can be sent and released directly into cancer cells making treatment more effective, and reducing side effects.

Instead of using implants, such as current practice, it may be possible to send nanobots to build required structures in situ. Nanoparticles of different composition can be used as well for bone repair, helping to restore normal bone structure following fracture.

Nanobots injected in to a vein can be used as valuable diagnostic device, a kind of nano endoscopy, providing a medical team with important data about patients' condition.

With further development it may be possible to use nanobots on cellular level to provide patients with advanced gene therapy, where abnormal genes can be swapped with normal ones.

Nanotechnology can be utilized in drug delivery systems to ensure particular drugs are released at appropriate times to eliminate human errors, for example in elderly patients.

It can make life much easier for diabetics – nanocomposite contact lenses with the property of changing color depending on blood sugar level can be used instead of invasive blood tests.

Robots are already implemented in surgery to help with precise operations. It can however come in to nano level where surgery can be performed with the use of lasers and different nano devices.

Medical imaging can be pushed in to molecular level providing far more accurate diagnosis of various diseases.

Nanotechnology can help stem cells to differentiate in to the line actually needed. Nanodevices can be used for prenatal diagnostics as they can go in to uterus or even in to the fetus not causing any harm. Possibly they can be used to repair eventual problems in the womb.

Nanochips implanted in your body can be responsible for continuous monitoring of your body systems and can send information to your personal computer. In these settings prevention of diseases can be easier and some problems can be repaired before causing serious health problems.

Nanotechnology is based upon the use of specific nanoparticles. Information about properties of currently used nanoparticles are summarized in the table 1 below.

Name	**Size**	**Composition Details**
Quantum Dots	2-10 nm	Colloidal fluorescent semiconductor nanocrystals. Central core consists of elements from groups II - VI of the periodic table.
Dendrimers	<15 nm	Highly branched synthetic polymers with a layered architecture - consisting of a central core, an internal region, and several terminal groups
Magnetic nanoparticles	10-20 nm	Spherical nanocrystals with Fe^{2+} and Fe3+ core surrounded by dextran or PEG (polyethelene glycol) molecules
Gold nanoparticles	<50 nm	Can be prepared into different geometries - nanospheres, nanoshells, nanorods, or nanocages
Carbon Nanotubes (CNT)	<100 nm	Coaxial graphite sheets
Liposomes	50-100 nm	Phospholipid vesicles. Classified by size and the number of layers - multi-, oligo-, or uni-lamellar.

Table 1

Discussion

Cancer

Cancer (malignant neoplasm) is the kind of disease where abnormal cells are dividing in uncontrolled manner and eventually invade surrounding tissues, destroying them. In some cases cancer can spread in to distal location in the body forming metastases. Cancer cells can be carried within lymph or blood.

There are more then 100 types of cancer, which can be grouped in five categories, depending on their origin:

Carcinoma – originating from skin or tissues lining or covering internal organs

Sarcoma – begins in bones, cartilages, fat, muscle, blood, vessels and connective tissues

Leukemia – starts from blood forming tissue, like bone marrow

Lymphoma and Myeloma – coming from immune system

Central nervous system cancers – originated from brain and spinal cord tissues. (1)

Cancer should be distinguished from tumors, not invading adjacent tissues neither giving metastases (so called benign tumor).

Some of them can be included into a group of environmental diseases, while others seem to have genetic causes,

All cancers begin in cells. Occasionally the genetic material of cells can become damaged or changed, sometimes as the consequence of environmental influences. In effect mutations are produced. A mutated cell can become the source of cancer when a regulatory mechanism of apoptosis is affected.

Action of oncogenes stimulating cell growth and reproduction are normally balanced by tumour suppressor genes. When this equilibrium is affected for some reason, cell proliferation occurs. These extra cells can form a mass called tumor.

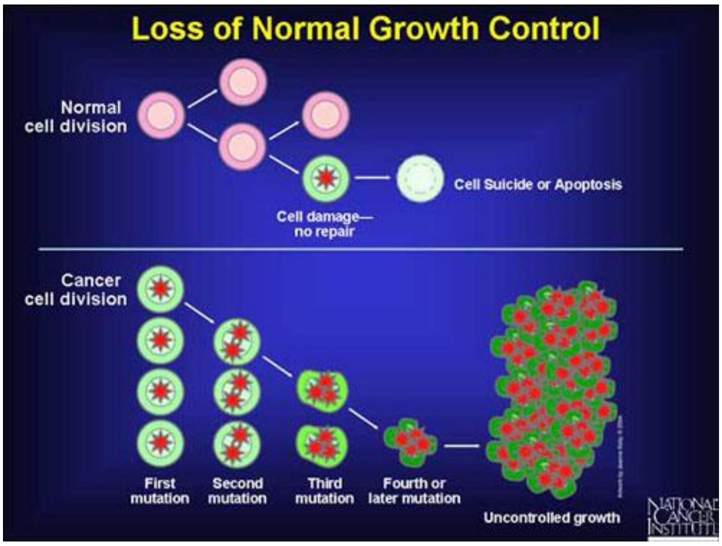

Fig. 3

Cancer diagnosis.

In most cases initial diagnosis is made on the basis of symptoms or results of cancer screening. Early symptoms however are not diagnostic in vast majority of cases, while screening tests are not specific for particular types of cancer.

Pathology tests (invasive): tissue sampling for histopathological examination – only biopsy can provide with the definite diagnosis.

Medical: blood tests

Non – invasive, imaging: X-rays, CT, MRI, PET

The problems are low sensitivity and poor specificity.
Current diagnostic techniques are often ineffective for making early diagnosis as they are not designed to detect singular cancer cells. Cancer can be only detected if visible change to the tissue is made – at that stage there are thousands of cells and cancer is usually already metastatic.

Management of cancer.

The best treatment available for cancer is surgery, but it is not always possible to determine surgical margins or whether to localize microscopic metastases, which makes surgery not a definite solution.

Chemotherapy: not very precisely delivered, affecting normal cells and tissues. Cancer medications are not free from side effects as doses of drugs are higher, then they could be if methods of delivery are more precise and action of drugs is restricted only to cancer tissue.

Radiation therapy: radiation used to kill cancer cells and reduce the size of tumor. Not very precise: affecting surrounding healthy cells by damaging their genetic material.

Statistics of cancer in the UK.

The graph below is reflecting cancer morbidity and mortality per 100.000 population, which is not changing very much over last years.

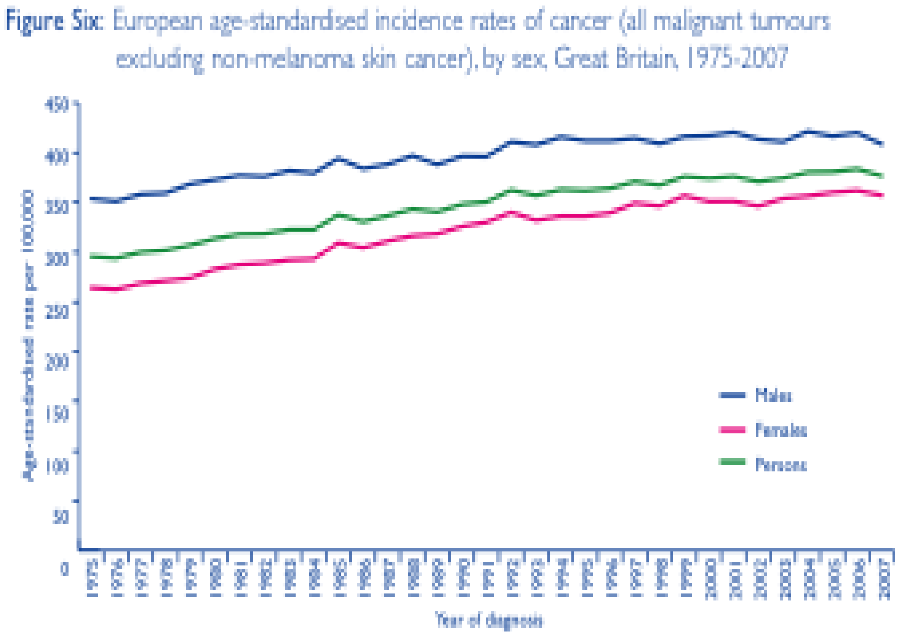

Fig. 4

From the next graph we can see, that while mortality from cardiovascular, respiratory and infectious diseases has come significantly down over last century, the cancer mortality is not changing very much. It is expected that over next 10-20 years the largest number of deaths in the UK will be caused by cancer.
It is why new diagnostic and treatment modalities in this field are very welcome.

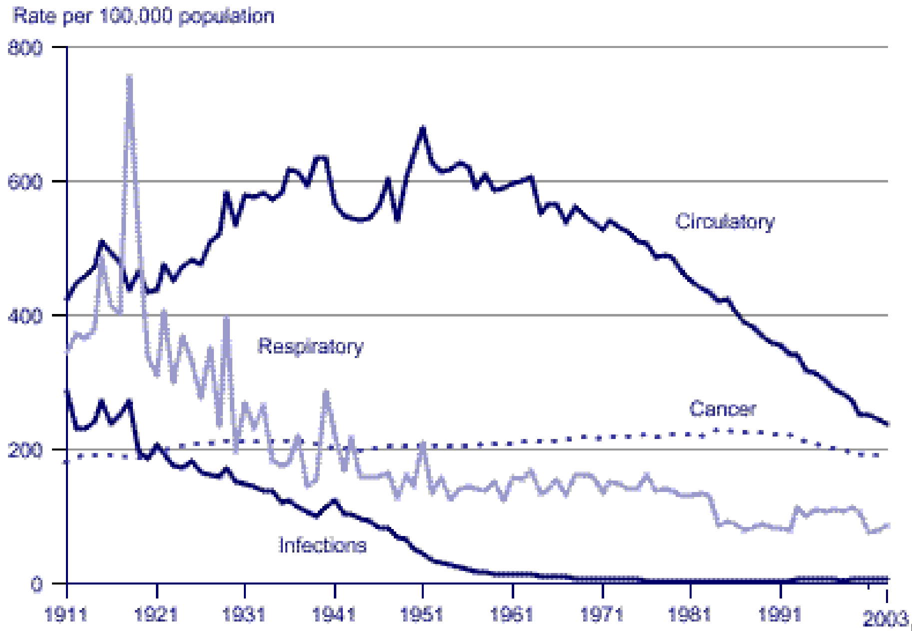

Fig. 5

Early symptoms of cancer are usually not diagnostic, but at the same time early diagnosis of cancer is crucial as some types of cancer early diagnosed can be curable.

Nanotechnology can be the one to change the very disappointing survival rate in amongst cancer patients

Nanotechnology in cancer.

The use of nanotechnology in the management of cancer is currently the most important fragment of nanomedicine.

The diagram below is summarizing the applications of nanotechnology in cancer: detection, treatment and monitoring, which will be discussed in the next part of the paper.

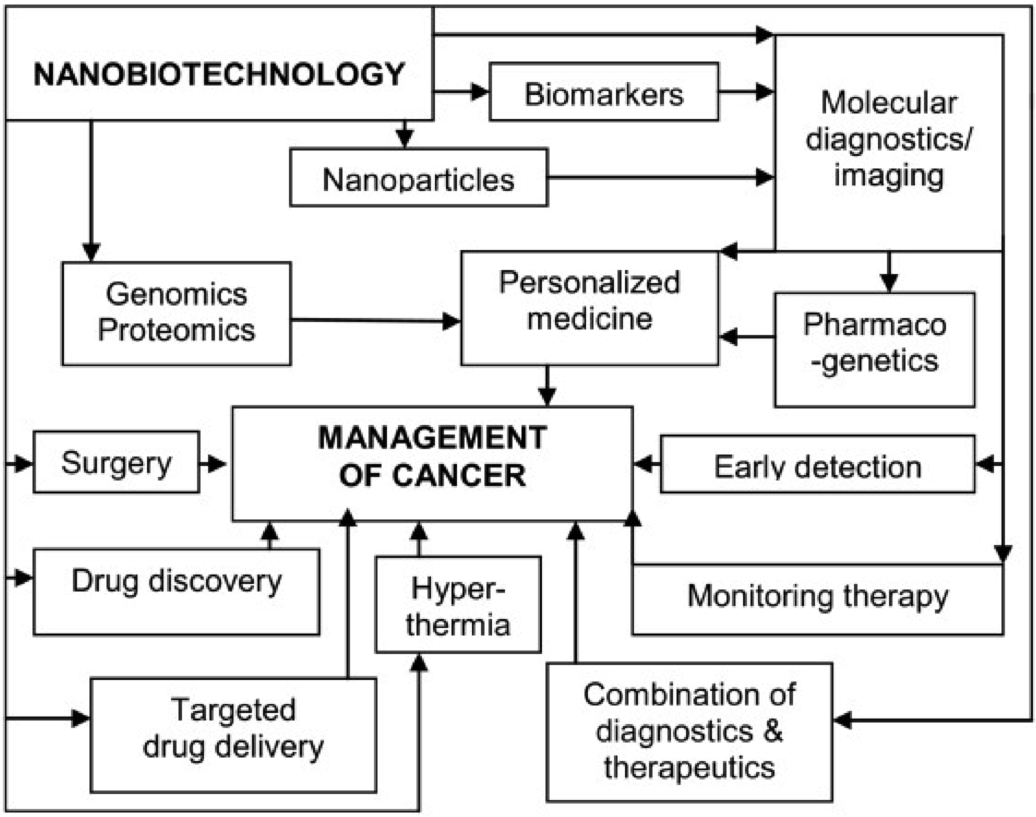

Fig. 6

Role of nanotechnology in cancer diagnosis.

It is complementary to already existing technologies, seems to be very useful in biomarkers research, brings better sensitivity to tests and can be used for tumour imaging.

Imaging.

Nanoparticles of superparamagnetic iron oxide (SPIONs) – iron oxide core with a hydrophilic coat can be applied as a contrast agent for MRI; they can alter magnetic field gradients in target tissue. They are lymphotropic and when administered intravenously they are trapped in lymph nodes. They are very valuable for detection of metastatic lymph nodes, not detected in standard MRI.

Breast cancer cells have on their surface over-expressed HER-2 receptor. If HER-2 antibody (Trastuzumab) is conjugated to nanoparticles of iron oxide or iron oxide/gold breast cancer cells will be labelled with nanoparticles and even small breast tumours can be properly identified.

Similarly colon cancer can be identified in MRI if SPIONs are conjugated with an antibody for specific colon cancer receptor Ganyl Cyclase C (GCC)

Another strategy would be the use of Quantum Dots (QD) with appropriate ligand, administered systemically. Visible fluorescence should be assessed after stimulation with near-infrared light. Quantum Dots are however not in use in vivo because of toxicity (contain heavy metals).

SPIONs can be applied for detection of sentinel lymph nodes (SLN) in breast cancer. Injected subcutaneously they alter node color to black and make them easily detectable using hand-held magnetometer or MRI.

Gold is attractive element because it demonstrates low toxicity and easy protein binding on the surface. When coated with polyethylene glycol it is resistant against immunological attack. It can be used as contrast agent for CT – detection of hepatic cancer cells.

LDLs (low density lipoproteins) are nanostructures existing naturally. Several tumors are over-expressing LDL receptor. With incorporated gadolinium chelate they can be used as MRI contrast for in-vivo detection of tumors, which would be labeled with gadolinium.

The conjugation of SPIONs with optical dye can make possible dual MRI/ optical imaging. You can get high concentration of SPION in tumor even without targeting ligand (EPR effect),

Similarly Quantum Dots, highly specific for optical imaging, while conjugated with the particle of gadolinium can be used for MRI imaging as well.

Brain tumors, especially surrounded by oedema are very difficult to assess. Nanoparticles coated with specific proteins can cross the blood-brain barrier to improve cerebral images. They can have therapeutic properties as well when conjugated with appropriate drug.

Drug delivery.

Nanoparticles used for drug delivery can be made from different materials.

A. Polymeric nanoparticles can be made from natural or synthetic polymers. In most cases they are built from hydrophobic core used as drug container and hydrophilic shell to stabilize particle in aqueous surrounding. A drug can be put into nanoparticle by physical entrapment or chemical conjugation. For chemical conjugation special pH-sensitive linkers have been developed, which are stable in blood stream (pH 7), while in the tumor, where pH is below 5,5 drug is easily released. Dendrimers - synthetic molecules with branched 3D structure are important materials for drug and gene delivery as well as for nanoparticle encapsulation in imaging.

B. Liposomal nanoparticles.

They are self-assembling particles with a membrane made of phospholipid bilayers. They are 25nm to 10um in size. Unmodified liposomes were considered for drug delivery for years, but their blood circulation time was very short (several minutes). In second generation of liposomes: polymer-coated, that time is extended up to 3 days.

C. Gold and iron oxide nanoparticles.

Both can be used for anticancer drugs delivery.

Gold nanorod formulation can be used for photothermal therapy – it can generate heat following radiation with infrared laser (not making any damage to healthy tissues).

Iron oxide nanoparticles can retain their properties of imaging agents for MRI whilst carrying drugs.

Schematic pictures of particles described above are presented on Fig 7.

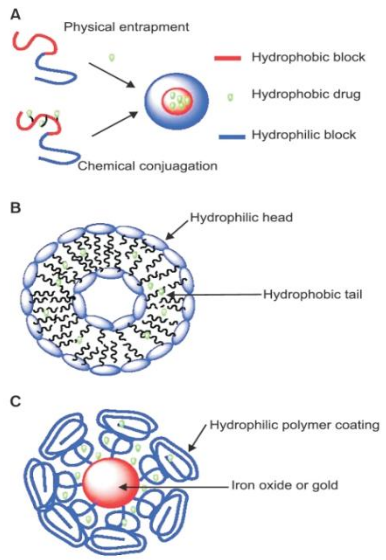

Fig 7

Properties of nanoparticles depend very much on their size. Optimal size for drug carriers is 10 – 100nm. Particles smaller than 10nm can be eliminated in kidneys, while larger then 100 nm can be captured by reticulo-endothelial system. For stability in circulation surface coating with appropriate materials is very important.

Passive targeting.

Passive targeting is connected with small sizes of nanoparticles and the specific properties of neoplasmatic vasculature. The growing tumor is recruiting a blood supply in the process known as angiogenesis, but these new vessels, unlike normal ones, have large gaps between endothelial cells (600 – 800 nm). Together with poor lymphatic drainage, defective vessels induce so called enhanced permeability and retention effect (ERP). See Fig 8. Nanoparticles can accumulate in tumor tissue.

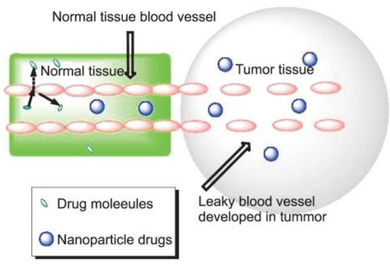

Fig 8

Active targeting.

Accumulation of drug in tumor environment in the consequence of ERP is not always connected with therapeutic efficiency as for most of anticancer medication internalization into the cancer cell is essential. Targeted nanoparticles can bind in to tumor associated receptor or antigen, facilitating delivery of nanoparticle together with the drug into tumour cells. As a result in the case of targeted nanoparticles we can observe a substantive increase in antitumor efficiency.

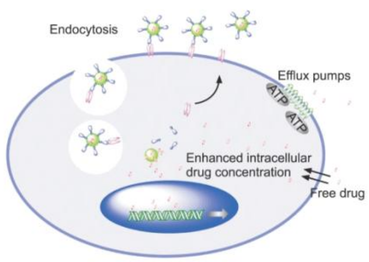

Fig 9

Most of targeted nanoparticles have not yet reached the stage of clinical trials. One of the few is immunoliposome- encapsulated doxorubicin.

Very important is selection of the target receptor or antigen on cancer cell; ideally that should be uniquely expressed on cancer cells and not on normal ones, and they should have a high density on tumor cell. A targeting ligand should promote internalization of nanoparticle into cancer cell to get a good response.

Other therapeutic application.

Nonobubbles use for targeted chemotherapy and ultrasonic tumor imaging.

A drug is delivered intravenously in the form of drug loaded micelles and nanobubbles, which are concentrated selectively in tumor interstitium. Nanobubbles produce microbubbles. Exposed to ultrasound they generate echoes – it is possible to get image of tumor. The sound energy from the ultrasound pops the bubbles and drug is released, what is causing enhanced drug uptake into the cells.

Nanoparticle-based cancer thermal ablation.

Used for tumors, which cannot be reached by conventional surgery. Thermal ablation with the use of invasive needle placement is limited by incomplete tumor destruction and damage to surrounding tissues. With the application of nanoparticles it is much more efficient. Gold nanomaterials, iron and magnetic nanoparticles as well as carbon nanotubes were used. Heat is induced by magnets, lasers, ultrasound or low-power X-ray. Hyperthermia offers additional benefit as hypoxic cells, resistant to radiation, are destroyed by heat as efficiently as normal cells.

Gold nanomaterials for thermal ablation.

Photothermal therapy can be used to heat gold nanoparticles to destroy the tumour. Optionally capsules made from gold particles can be filled with cancer medication, which is eventually released locally following heating.

Magnetic nanoparticles for thermal ablation.

These particles were effectively used in an animal model to destroy small breast tumors using magnetically induced heating. An antibody used for

HER-2 receptor over-expressed on tumour cells, can provide nanoparticles with tumour targeting properties.

<u>Laser induced thermal destruction of cancer using nanoparticles</u>

Selected cancer cells can be destroyed by targeting single wall carbon nanotubes to tumor cells. These nanotubes can emit heat following absorption of energy from near infrared light. No harm to normal cells.

Improving surgical outcomes.

Surgery is still the most effective procedure in treating human cancers, and most important single predictor of patients' survival is complete surgical resection. Nanotechnology can be used to localise a tumour, clearly detect tumour margins, identification of important structures surrounding tumour, mapping of sentinel lymph nodes and detecting residual tumor cells or small metastases.

Surface-enhanced Raman scattering (SERS) nanoparticles are applied for intra-operative imaging to determine tumor edges, as they are much brighter than near-infrared quantum dots.

Researches in progress.

There is a lot of research in progress connected with application of nanotechnology into cancer treatment. Investigators are trying to find new solutions. Most research is carried on animal models and there is still long time before human clinical trials will be started. Few reports from recent trials are presented to highlight that despite of great hopes connected with the use of nanoparticles we are still some distance away from final answers.

1. "Researchers from Massachusetts institute of Technology have shown in animal study, that they can deliver cancer drug Cisplatine more effectively and safely in the form that has been encapsulated in a nanoparticle targeted to prostate tumor cells "(Feb 2011)
2. "Investigators from Northwestern University have demonstrated that using nanoparticles to deliver multiple drugs simultaneously can produce a synergistic effect that boosts the cell-killing ability of both drugs. They can combine two powerful but extremely toxic anticancer agents - cisplatin and doxorubicin - in one polymer nanoparticle, producing a substantial boost in their ability of the

combination to destroy tumors. In addition, the two-in-one nanoparticle reduces the amount of both drugs needed to kill cancer cells, which presumably would reduce the toxic side effects associated with these drugs" (Dec 2010)

3. "Nanoparticle that is small enough to escape the leaky blood vessels that surround tumors but large enough to avoid rapid clearance from the blood stream via the kidneys. Balancing these two requirements usually results in using nanoparticles that are indeed small enough to accumulate in the vicinity of tumors, but that are really too large to penetrate deeply enough into tumors to have the maximum therapeutic effect.team of researchers from the Massachusetts Institute of Technology, Massachusetts General Hospital, and Harvard Medical School have developed a solution to this problem (animal study): multilayered, or multistage, nanoparticles that partially dissolve once they accumulate around tumors, leaving behind a payload of nanoparticles a mere one-tenth the size of the original delivery vehicle. The remaining 10-nanometer-diameter nanoparticles, loaded with anticancer drugs, can then diffuse deeply into a tumor's dense interior." (Jan 2011)
4. "Rice University bioengineers and physician-scientists at Baylor College of Medicine and Texas Children's Hospital have successfully destroyed tumors of human brain cancer cells in the first animal tests of a minimally invasive treatment that zaps glioma tumors with heat. The tests involved nanoshells, light-activated nanoparticles that are designed to destroy tumors with heat and avoid the unwanted side effects of drug and radiation therapies.

   The researchers injected the mice with gold nanoshells and waited 24 hours for the nanoparticles to accumulate in the tumors. Laser-generated near-infrared light, which passes safely through biological tissues was shined on the tumor for three minutes. The nanoshells converted the laser light into tumor-killing heat. All seven animals that received the nanoshell treatment responded, but cancer returned in three. The other four remained cancer-free 90 days after treatment."

   (Feb 2011)

Conclusions

There are still many questions to be answered before use of nanobiotechnology will be used on a daily basis in oncology and other fields of medicine.

Whatever medicine is going to be used in nanoparticles must go through all the tests, as its properties can become very different.

Issue of safety is still not resolved. Some nanoparticles, especially containing metals are considered to be toxic, while others, like biodegradable polymer nanoparticles are suitable for drug delivery as there is no significant toxicity.

Most research is still performed on animal models, where nanoparticles can potentially behave differently than in humans. We must still wait for clinical trials results before new techniques are widely introduced.

There are however strong rationales for using nanobiotechnology in oncology. Structural, optical and magnetic properties of nanoparticles are not available from larger molecules. Connected with targeting ligands they can become very specific. Direct delivery of cancer medication into their place of action should reduce doses needed making treatment safer and more effective.

Diagnosis of cancer can be made earlier in the course of disease giving patients better chances for recovery. Knowledge about molecular profile of particular patients will enable doctors to set more personalized cancer therapies. Nanooncology has a promising future, and further advances are anticipated in the next 5 – 10 years. We can expect introduction of nanobots to clinical practice and may be personal computers as well which will monitor our health and will use preventive measures rather than treatment.

We must remember about all risks and limitations but there are good reasons to be optimistic about the future of nanotechnology in medicine.

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