

**The Future of Clinical Nanotechnology in
General and Cardiovascular Medicine**

BY

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PASS WITH MERIT

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Abstract

Nanotechnology is a new discipline of science and engineering that has led to innovative approaches in many areas of medicine. Its applications in the screening, diagnosis, and treatment of disease are collectively referred to as —nanomedicine—an emerging field that has the potential to revolutionize individual and population-based health this century. The three-in-one functions of imaging, targeting, and transporting, play a central role in nano- and microparticle-based imaging of cardiovascular interventions. While nano- and microparticlebased imaging of cardiovascular interventions is still in its developing phase, it has already presented the exciting potential to monitor primary interventional procedures for precise therapeutic delivery, enhance the effectiveness of delivered therapeutics, and monitor therapeutic efficiency after interventions performed to treat cardiovascular diseases.

Introduction

The term nanotechnology refers to the ability to measure, design and manipulate materials at atomic, molecular and supramolecular level in order to understand, create and apply structures and systems with specific functions attributable to their size. Nanotechnology classically refers to matter in the size range of 1–100 nm, but it is often extended to include materials below 1 μm . A key goal is to assemble nanoparticles and integrate them into ordered structures in order to obtain useful materials.

The rapid growth of nanotechnology and nanoscience could greatly expand the clinical opportunities for molecular imaging¹⁸. By precisely engineering atoms and molecules, nanotechnology can give rise to new molecular assemblies on the scale of individual cells, organelles, or even smaller components, generally in the range of 5 to 500 nm. The specific organization of such nanoscale materials is anticipated to confer unique chemical and biological properties on the basis of interactions that occur at their surfaces.

The prefix “nano” derives from the Greek word for “dwarf.” One nanometer (nm) is equal to one-billionth of a meter, or about the width of 6 carbon atoms or 10 water molecules. Atoms are smaller than 1 nm, whereas many molecules including some proteins can far exceed this size¹⁷. The conceptual underpinnings of nanotechnologies were first laid out in 1959 by the physicist Richard Feynman in his lecture, “There’s plenty of room at the bottom.” Feynman explored the possibility of manipulating material at the scale of individual atoms and molecules, imagining the whole of the Encyclopedia Britannica written on the head of a pin and foreseeing the increasing ability to examine and control matter at the nanoscale. The term “nanotechnology” was not used until 1974, when Norio Taniguchi, a researcher at the University of Tokyo, used it to refer to the ability to engineer materials precisely at the nanometer level. The primary driving force for miniaturization at that time came from the electronics industry, which aimed to develop tools to create smaller (and therefore faster and more complex) electronic devices on silicon chips.

In recognition of the enormous scientific and commercial potential for nanotechnology, President Clinton established the National Nanotechnology Initiative (NNI) in 2000. NNI is a multiagency umbrella program to build, characterize, and understand nanoscale devices. The NNI lists medicine, manufacturing, material sciences, information technology, energy, and environmental sciences as target beneficiaries. The program is slated to spend nearly \$1 billion in fiscal year 2005 as compared with \$464 million in 2001¹².

Advances in nanotechnology have led to the development of new nanomaterials whose physiochemical properties differ from those of their larger counterparts due to their higher surface-to-volume ratio. These novel properties make them excellent candidates for biomedical applications, given the range of biological processes that occur at nanometer scale⁹.

Discussion

Cardiovascular disease (CVD) remains the leading cause of death in the USA; one in four Americans is diagnosed with CVD and one person world-wide dies from it every 30 seconds. Although significant advances have been made in the management and treatment of this disease, the effectiveness of early detection and treatment in preventing heart attacks is still questionable.

One of the fundamental and unresolved problems in cardiovascular biology is the in vivo detection of atherosclerotic disease and the evaluation of atherosclerotic disease activity. Currently technology limits clinicians to diagnostic techniques that either image or functionally assess the significance of large obstructive vascular lesions. Techniques have been developed recently that achieve molecular and cellular imaging with most imaging modalities, including nuclear, optical, ultrasound and MRI. However, current imaging modalities are not capable of imaging atherosclerotic disease at its earliest stages, nor do available techniques allow routine assessment for atherosclerotic lesions for the general population at a feasible cost.

The rapid growth of nanotechnology and nanoscience could expand the clinical opportunities for molecular imaging greatly. Advanced imaging methods and new, targeted, nanoparticle contrast agents for early characterisation of atherosclerosis and cardiovascular pathology at the cellular and molecular levels might represent the next frontier for combining imaging and rational drug delivery to facilitate personalized medicine.

Nanoparticles carrying therapeutics offer the benefits of decreased systemic toxicity, targeted delivery to specific tissues, and higher doses delivered to the target. The most commonly used nanoparticles include liposomes, emulsions, polymeric microspheres and microbubbles. Liposomes are of similar structure to phospholipid micelles; Emulsions, being chemically distinct from liposomes, are oil-in water– type mixtures and are stabilized with surfactants to maintain their size and shape¹⁹; Polymeric spheres, made from different polymers, are readily produced, are inexpensive, and show no signs of toxicity in vivo⁸; Microbubbles, made of albumin or charged lipid, function by resonating in an ultrasound beam and rapidly contracting and expanding in response to the pressure changes of the sound wave³. In addition to these commonly used particles, new types of nanoscale particles have been recently introduced, such as (a) quantum dots (b) aquasomes (carbohydrate- ceramic nanoparticles), (c) dendrimers, (d) gold nanoparticles, (e) metal nanoshells.

a) “Three in One Functionality”

The three-in-one functions—targeting, imaging and transporting —are the major characteristics of nanoparticles, enabling, for example, the potential use of different imaging techniques to monitor, enhance, and track cardiovascular interventions. By combining each of the three functions, one can generate (a) target-specific molecular imaging for early diagnosis of diseases, (b) target-specific therapy for efficient treatment of diseases, and (c) imaging guided target-specific therapy for further enhancement of target particle based treatment of diseases (Figure 1 and 2).

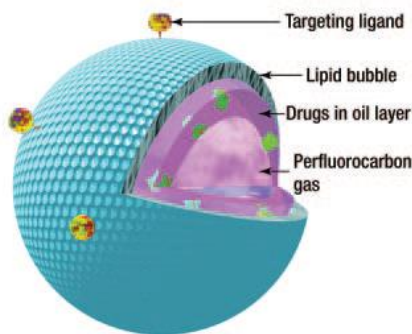


Figure 1: Diagram of microbubble constructed for drug delivery. Gas-filled microspheres can be designed so that their interior is loaded with drugs and gas. A stabilizing material, in this case a lipid, surrounds the perfluorocarbon bubble. Drugs can be incorporated alone, or if insoluble, in water, or in an oil layer. The microsphere can be targeted to specific tissue by incorporating protein ligands on the surface. The microbubble is capable of three functions: visualization at US, site-specific targeting, and drug transportation⁹

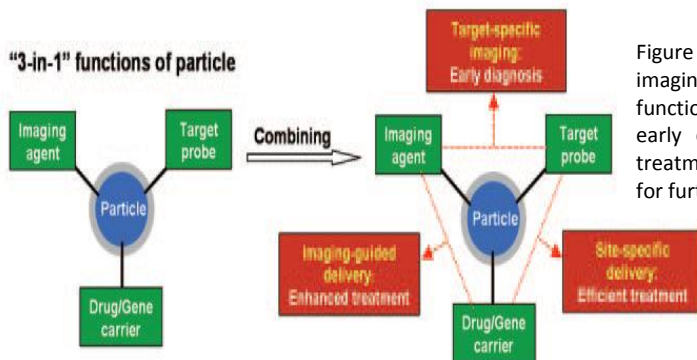


Figure 2: Diagram represents particle with three-in-one functions for imaging, targeting, and transport (carrier). Combination of the three functions enables generation of target-specific molecular imaging for early diagnosis, site-specific delivery of therapeutics for efficient treatment, and imaging-guided site-specific delivery of therapeutics for further enhanced treatment.

i) Target-specific Probe or Tracers

To implement this function, nanoparticles are commonly bound to target specific ligands e.g. monoclonal antibodies or their fragments, peptides, vitamins, or aptamers¹⁸.

Particles that function as target-specific imaging probes or tracers are based on the mechanism of ligand-molecule binding i.e. the interaction of ligands with their complementarily shaped targets (such as receptors expressed on cell surfaces). The specific ligand-target binding ensures that particles are highly localized at the site of interest. Thus, when a sufficient number of nanoparticles are bound to the target, the particle-delineated target can be visualized with imaging. Configuration of multivalent ligands (or mixture ligands) is desirable to maintain the particle-target interaction and reduce “off rates,” which thus permits the success of target-specific imaging within a sufficient time window after administration of target-specific imaging particles¹¹.

ii) **Diagnostic Imaging Agents**

The imaging capability of nanoparticles involves the construction of particles with use of imaging contrast agents, such as paramagnetic or superparamagnetic metals for magnetic resonance (MR) imaging, optically active compounds for optical imaging, or gases for ultrasonographic (US) imaging.

iii) **Therapeutic Carriers**

The ability of target-specific nanoparticles to carry therapeutic agents (such as drugs, genes, or proteins) is essential for the application of nanotechnology in modern medicine. Therapeutic agents can be attached to the surface of a target-specific imaging particle, incorporated into its structure, or encapsulated within the core of a hollow agent⁵. Combining therapeutics with a target-specific nanoparticle allows efficient drug delivery while minimizing systemic toxicity¹⁶.

b) Application of Nanoparticles in Cardiovascular Medicine

Imaging of cardiovascular interventions should help address several issues pertinent to the success of cardiovascular interventional therapies, such as (a) monitoring the primary interventional procedure to assess desirable distribution and localization of delivered therapeutics at the targets, (b) enhancing the effectiveness of the delivered therapeutics to achieve a sufficient level of therapeutic effect at the targets, and (c) monitoring the function and functional period of the delivered therapeutics at the targets²¹. The application of the three-in-one functions of nanoparticles, including imaging, targeting, and transport, has opened new avenues for using imaging to monitor, enhance, and track cardiovascular interventions.

Several studies¹ have demonstrated the usefulness of combining drug-carrying nanoparticles with arterial stent placement to reduce in-stent restenosis. In addition to the application of nanotechnology to catheter-based gene and drug delivery, non-catheter-based nanoparticle imaging techniques such as tissue factor imaging may offer a non-invasive molecular imaging tool to monitor primary therapeutic delivery.

Several molecules, such as endothelial interleukin-2, fibrin, fibrinogen, IIb-IIa factors, $\alpha_v\beta_3$ -integrin and myeloperoxidase are expressed from vulnerable plaques and activated platelets. Thus, these molecules become the targets for tissue factor imaging^{18,5,4}. For example, the success of nanoparticle-targeted fibrin imaging, which is based on physiopathologic evidence of fibrin deposition, one of the earliest signs of plaque rupture or erosion and intraplaque haemorrhage, has been reported¹⁴. Through systemic administration, the fibrin-specific nanoparticles accumulate at the site of arterial thrombi, which can then be detected with either US or MR imaging¹⁰.

c) General Benefits of Nanotechnology

i) **Biosensors and Diagnostics**

Currently, diagnostics are largely confined to in vitro use, with diagnostic tests performed individually in centralized clinical laboratories. Nanotechnology has enormous potential both for multiplexing in vitro diagnostic tests and for allowing miniaturization of sensors for use in vivo. The former applications are likely to predominate in the short term, but in vivo devices that report health problems in real time could be powerful tools for disease management.

Nanotubes or nanowires, which are starting to find use as components of very small computer circuits, show promise for diagnostic testing. They can be used to measure pH or decorated with specific capture molecules to detect minute quantities of biological and chemical species. Nanocantilevers can measure the content of specific DNA moieties or can be used for simultaneous rapid monitoring of multiple serum protein markers⁶.

Bioengineered nanopores allow sequence-specific detection of individual DNA strands with single-base resolution⁶, and similar pores may be useful as components of quantitative sensors for cell signalling molecules. Quantum dots are highly fluorescent semiconductor nanocrystals that are excited by a broad range of wavelengths but have narrow, tunable emission spectra. Linked to antibodies or DNA probes, they can detect specific protein or DNA targets. Their narrow emission spectra allow numerous probes to be used simultaneously for multiplexed high throughput screening⁶.

In vivo nanosensors show promise for real-time monitoring of biological signals, such as the release of proteins or antibodies in response to cardiac or inflammatory events.

ii) **Drug Delivery and Therapeutics**

The current generation of drugs is largely based on small molecules with a mass of 1000 Da or less that circulate systemically. Common deleterious consequences of systemic biodistribution include toxicity to non-target tissues, difficulty in maintaining drug concentrations within therapeutic windows, and metabolism and excretion of drugs, all of which can reduce efficacy. Drug solubility and cell permeability issues are also common with small molecules and biologics.

Nanotechnology-based delivery systems could mitigate these problems by combining tissue- or organ-specific targeting with therapeutic action. Multifunctional nanodelivery systems could also combine targeting, diagnostic, and therapeutic actions. Research has already shown that drugs can be encapsulated in nanospheres or erodible self-assembled structures or conjugated to well-defined multivalent macromolecules such as dendrimers (highly branched polymers). These mechanisms can improve bioavailability and enable continued release, thereby controlling the initial dose, improving effectiveness, and widening the therapeutic window.

Potential targets include proliferating smooth muscle cells, neoplastic cells, inflammatory mediators, proteins expressed in viral infections, or even distinct subcellular localizations such as mitochondria¹⁵ and other cytoplasmic organelles¹³. Specific targeting of drugs should mitigate systemic toxicity, and encapsulating or conjugating drugs to nanoscale carriers can protect them from systemic metabolism or excretion. In addition, nanoparticulate or macromolecular targeting systems can be used to give triggered release in response to internal triggers such as pH or to externally administered signals such as

ultrasound, near-infrared light, magnetic fields, or radiofrequency pulses. For example, magnetic microparticles or nanoparticles that bind to specific cells could be heated with an alternating magnetic field to kill neighbouring cells thermally⁶.

iii) **Imaging**

Established imaging modalities such as CT, MRI, and ultrasound focus primarily on anatomy and physiology. The emerging field of molecular imaging uses novel reagents and methods to image specific molecular pathways non-invasively in vivo, particularly pathways involved in disease processes. As these technologies mature, non-invasive diagnostics with high-affinity homing molecules should become faster, cheaper, and more accurate, allowing physicians to detect early disease and improve patient outcomes.

iv) **Tissue Engineering and Biomaterials**

An additional approach is the use of microtechnologies and nanotechnologies to direct tissue remodelling in vivo. Microspheres that provide sustained perivascular release of elastase can create a chemotactant gradient across the arterial wall to direct smooth muscle cell migration away from the lumen, reducing pathological neointima formation in a balloon injury model²⁰. Directed migration could also attract cells to injury sites for repair purposes.

d) **Clinical Cardiovascular Benefits of Nanotechnology**

i) **Unstable or Vulnerable Plaque**

The diagnosis and treatment of unstable plaque is an area in which nanotechnology could have an immediate impact. Research is under way using probes targeted to plaque components for non-invasive detection of patients at risk⁷. In an extension of this approach, targeted nanoparticles, multifunctional macromolecules, or nanotechnology-based devices could deliver therapy to a specific site, localized drug release being achieved either passively (by proximity alone) or actively (through supply of energy as ultrasound, near infrared, or magnetic field). Targeted nanoparticles or devices could also stabilize vulnerable plaque by removing material, for example, oxidized LDL. Devices able to attach to unstable plaques and warn patients and emergency medical services of plaque rupture would facilitate timely medical intervention.

ii) **Tissue Repair, Engineering, and Remodelling**

Nanotechnology may facilitate repair and replacement of blood vessels, myocardium and myocardial valves, and lung tissue. It also may be used to stimulate regenerative processes such as angiogenesis. Cellular function is integrally related to morphology, so the ability to control cell shape in tissue engineering is essential to ensure proper cellular function in final products. Precisely constructed nanoscaffoldings and microscaffoldings are needed to guide tissue repair and replacement in vessels and organs.

Nanofiber meshes may enable vascular grafts with superior mechanical properties to avoid patency problems common in synthetic grafts, particularly small-diameter grafts. Cytokines, growth factors, and angiogenic factors can be encapsulated in biodegradable microparticles or nanoparticles and embedded in tissue scaffolds and substrates to enhance tissue

regeneration. Scaffoldings capable of mimicking cellular matrices should be able to stimulate the growth of new heart and lung tissue and direct revascularization.

iii) Sleep Apnea and Related Cardiovascular Consequences

Because sleep apnea is a cause of irregular heartbeat, hypertension, heart attack, and stroke, it is important that patients be diagnosed and treated before these highly deleterious sequelae occur. For patients suspected of experiencing sleep apnea, in vivo sensors could constantly monitor O₂ blood concentrations and cardiac function to detect problems during sleep.

In addition, heart-specific antibodies tagged with nanoparticles may allow doctors to visualize heart movement while a patient experiences sleep apnea to determine both short- and long-term effects of apnea on cardiac function.

Conclusion and Ethics

The rapid progress of nanoscience and the application of nanotechnology are changing the foundations of diagnosis, treatment, and prevention of cardiovascular diseases. Nanoparticles, with the three-in-one functions of imaging, targeting, and transporting, play a central role in particle-based imaging of cardiovascular interventions, presenting the exciting potential to monitor primary interventional procedures for precise therapeutic delivery, enhance the effectiveness of delivered therapeutics, and monitor therapeutic efficiency after performed interventions. As refinements of nanotechniques continue with additional effort on their subsequent translation to clinical practice, the rapid growth of nanoscience and the application of nanotechnology in modern medicine are changing the field of imaging and imaging-guided interventions for cardiovascular diseases.

Examined extensively by Dr. Raj Bawa, nanomedical ethics throw up some well known ethical arguments²:

i) What's the limit on how clinically & socially relevant nanotechnological data can be?

One must ask the question whether the capacity to analyse every disease risk factor via nanotechnological means is both clinically and socially relevant; profiling the likelihood of developing diseases may allow lifestyle adjustments to be made accordingly, improving long term quality of life. Alternatively, it may throw up statistically worrying results that cause anxiety and mental maleficence to a patient, while never manifesting clinically and thus being consequentially unjustifiable. Certainly, no benefit to the patients is guaranteed and autonomy, for example, with health insurance may be severely curtailed.

ii) What are the implications with regards to potential Transhumanism?

This, in itself, is a vast philosophical idea. The concept entails the possibility to enhance the human body – physically, psychologically or intellectually – thus transforming the human race into ‘posthumans’. This could be perceived to suit the orthodox perception of better quality of life. However, possible ethical implications include practical criticisms i.e. it is very

unlikely to happen successfully, or moral objections in that it may pose a threat to human values and increase social divides.

*iii) **Nanotechnological Risks***

As with any drug, it is difficult to see how clinically effective – or alternatively toxic – it is until it has been trialled on humans. The ethical implications of therapeutics are already well documented. However, other questions must be fairly considered, such as; a) what are the long term side-effects? b) How will nanowaste be disposed of? & c) What environmental implications are possible? One argument is that if patients are fully informed of the risks then clinical use of nanotechnology should be allowed, respecting the patient's autonomy. On the other hand, one must then ask why a fully consenting, fully informed and competent adult is not allowed to be put at 'more than minimal risk' in clinical research. Here we can see where double standards within medical ethics may appear.

*iv) **Privacy & Confidentiality***

Live data harvesting sounds brilliant; the ability to be monitored and diagnosed the instance a medical problem manifests. Or does it? With such a vast population, we do not have the capacity to store and analyse everyone. Therefore allocation ethics come into play. Also, with such intimate and potentially threatening information, rigorous and expensive safeguards must be put in place.

This ethical overview is generalised to nanotechnology in medicine. However, with cardiovascular incidences on the rise and such a large interest in cardiovascular nanomedicine, all are proportionately applicable.

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