Review Article

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Nanoimaging in cardiovascular diseases: Current state of the art

Suryyani Deb, Kanjaksha Ghosh & Shrimati Dharmapal Shetty

Department of Hemostasis & Thrombosis, National Institute of Immunohaematology (ICMR), Mumbai, India

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Nanotechnology has been integrated into healthcare system in terms of diagnosis as well as therapy. The massive impact of imaging nanotechnology has a deeper intervention in cardiology *i.e.* as contrast agents , to target vulnerable plaques with site specificity and in a theranostic approach to treat these plaques, stem cell delivery in necrotic myocardium, *etc.* Thus cardiovascular nanoimaging is not limited to simple diagnosis but also can help real time tracking during therapy as well as surgery. The present review provides a comprehensive description of the molecular imaging techniques for cardiovascular diseases with the help of nanotechnology and the potential clinical implications of nanotechnology for future applications.

Key words: Cardiovascular disease - nanoimaging - nanotoxicity - theranostic - thrombus imaging

Introduction

Nanotechnology is considered as a cutting edge technology in the 21st century. In ancient period also people prepared and used nanoparticles in different fields of art and medicine, without knowing their in-depth physico-chemical properties, but believing their potential to prevent diseases. Window glasses and ceramic containers in the Roman Empire were found to contain gold, copper and silver nanoparticles to give them eternal bright colour[1,2]. Gold nanoparticles were being used in medicines in China and India. In India, till now gold nanoparticles are used in medicine as '*Swarno Vasmo*'[3,4]. In 1857, Michael Faraday first prepared the pure colloidal gold nanoparticles, and named it as 'activated gold'[5], though the first introduction of the concept of modern nanotechnology was by renowned Noble laureate Physicist, Richard Phillips Feymann in 1959 in his famous talk called "There's Plenty of Room at the Bottom" [6]. Robert Curl, Harold Kroto, and Richard Smalley were awarded Nobel Prize in Chemistry in 1996 for their roles in the discovery of buckyballs or fullerenes (spherical carbon nanoparticles), the first synthetic nanoform with known characteristics[7].

The word 'Nano' is derived from the Greek word 'Nanos' which means dwarf, denoting a factor of 10^{-9} (1meter= 1,000,000,00 nano meter). According to National Nanotechnology Initiative "Nanotechnology is the understanding and control of matter at the nanoscale, at dimensions between approximately 1 and 100 nanometers, where unique phenomena enable novel applications"[8,9]. Nanoscale dimension acquires some special characteristics (*e.g.* optical, magnetic, electrochemical, *etc*.) which are neither present in bulk

material nor in molecular state[8, 10-13]. A good example is gold nanoparticles which remain in colloidal phase and are red in colour, though bulk gold is solid in nature and is yellow in colour. This unique colour phenomenon of gold nanoparticles is due to surface plasmon resonance (SPR), found only in nanoscale dimension. Therefore, these exclusive nano-specific properties make them unique entity in classical chemistry. Interestingly, one can easily modify or customize these properties just by modulating shape, size and/or surface topology of nanoforms[14]. In a nutshell, the maneuverability of designing the materials at the nanoscale and tunability on its surface make this an independent branch of science having wide applicability .

Nanobiotechnology

Nanotechnology is truly an interdisciplinary area of modern science. It involves vast area of chemistry, physics, electronics, material science as well as biology[15,16]. Among all of these areas, biology is the most recently introduced subject. Application of nanotechnology in biology is thought to be one of the most successful and wide spread areas of utility, which includes basic understanding of biological event as well as medical diagnosis, surgery and therapy[17-20]. Biomolecule compatible size distribution of nanoforms along with their tuneable properties (physical, chemical, topological) help nanotechnology to merge with biology to give birth to one of the most advanced fields of science - Nanobiotechnology[21-23]. Depending on the constituting material, mode of synthesis, surface topologies, mode of applications, *etc*., scientists are classifying different forms of nanomaterials. Some of these basic nanoforms synthesized so far and are proposed to be used in biomedical field are given in Table I.

The ultra small dimension and uniform size distribution (compared to liposome), specialized optical, physico-chemical, electrical, magnetic properties; high cellular penetration power, tuneable size, shape, texture, unique surface topology and surface chemistry make nanoparticles to play promising role in the biomedical field (*e.g*. therapy, drug delivery, diagnostics, *etc*.)[40- 42]. The nanoforms that belong to this category (bio-medical) are listed in Table II.

Nanotechnology and cardiovascular diseases

Biomedical application of primitive era nanotechnology was mostly in the field of cancer[43,44,49], though with advanced exploration, it encompasses almost all fields of biomedical research. Cardiovascular nanomedicine is the most recent area. Diagnosis, drug delivery, stem cell therapy, tissue engineering, stent surgery, are the few other areas where nanotechnology imprints its signature. The most promising area however, is the nanomaterial based improved clinical imaging, *e.g*. nanoimaging of cardiovascular diseases[50].

Cardiovascular imaging is one of the most reliable diagnostic tools for cardiovascular diseases[51]. After extensive research, it was hypothesised that nanoparticles could be unique contributors in the field of the medical imaging, due to their special features, which are as follows:

(i) Biocompatible size distribution : The ultra small nano size helps them to accommodate with different biocomponents even inside the subcellular organelle[52].

(ii) High penetration power: This is another aspect fulfilled by nanoparticles for bio-medical imaging[53].

(iii) Image contrasting ability : Paramagnetic nanoparticles are magnetic resonance imaging (MRI) contrast agents. Iodinated nanoparticles can be used as computed tomography (CT) contrast agents, whereas quantum dots can act as fluorescent enhancers[54].

(iv) Surface tuneable property : Nanosurface can be modified with the molecules of choice. Thus, it is possible to conjugate a nanomaterial with multimodal entity, for example, target specific molecules (targeted delivery), imaging probes and/or therapeutic molecules[55,56].

(iv) Stability : Contrast enhancer nanomaterials are much more stable than a chemical image probe[57].

(v) Half life : In case of carrier nanoforms, used as image contrast agents, the half life of the chemical image probes is also increased due to their conjugation with nanoparticles.

Thus, atypical size distribution, target specific delivery, high contrast capability, increase lifetime are the key features that make nanomaterials indispensable in the future medical imaging.

Nano based cardiovascular imaging can monitor the live physiological system in a noninvasive manner, with almost no pain. This live imaging is not only important for proper diagnosis or therapy, but is also beneficial for the basic understanding of the pathological conditions, which in turn helps us to develop future advanced techniques. Though majority of the nano-based cardiovascular imaging modules are in the field of diagnosis, but with advancement of this

Table I. Different types of synthesized nanoforms

Types of nanoforms	Characteristics	Biomedical advantage
Polymeric nanoparticles	Solid or encapsulated nanoparticles composed of natural or synthetic polymers[24, 25].	Biodegradable in nature. Minimum retention within body [24].
Inorganic nanoparticles	Mainly metallic solid nanoparticles[24, 26].	Special optical, magnetic, electro chemical properties. Applied in drug delivery, tissue engineering and diagnostics [24].
Nanoshell	Dielectric core covered by thin layer of metallic shell[24, 27].	Broad wavelength tuneable optical properties, biocopmpatibility. Thermal killing of cancer cells[28].
Nano wires	In nanowire the diameter is of in order of nanometre, but the length can be in micro meter range [29].	Special electronic properties. Useful in stem cell engineering, surgery and diagnosis [30].
Quantum dots (QD)	Quantum dots are smallest in this series that contain a tiny droplet of free electrons. QDs are mostly semiconductor nanocrystals that are less than 10 nanometers in diameter[31].	Special optical properties with large wavelength spectrum and longer life time. Useful in diagnostics and *in vitro* imaging[32].
Carbon nanoparticles	Hollow cage spherical structure of pure carbon, known as fullerene [33].	Antiviral, antibacterial, photodynamic and anti-tumour effect[24,33].
Carbon nanotubes	Carbon nanotubes (CNTs) are cylindrical nanostructure. CNTs are allotropes of carbon and may be of single (SWCNTs) or multi wall (MWCNTSs)[34].	High cell membrane penetration power useful for delivery agent. High tensile strength useful in tissue engineering[24,34].
Dendrimers	These are mono disperse globular molecules with highly branched (3D) architecture[24].	High availability, can be conjugate with large quantity of drugs. Effective in control release of drugs. Also useful in imaging and bio sensing[35].
Nano crystal	Nanoparticle with a crystalline structure is called nanocrystal[36].	Effective for poorly soluble drug[24,36].
Solid lipid nanoparticles	These nanoforms are spherical in structure, with a solid lipid core that is stabilised by an outer lipophillic layer[37]	Control release and high content of drug. Enhanced biocompatibility [38]
Nano silk (Nano biomaterials)	Silk fibroin protein based nanosphere[39] .	Biocompatible, biodegradable, tunable drug loading release capacities[39].

Source: Refs 24-39

technology, it has entered in the domain of therapy and surgery also.

In most of the cases, the nano based imaging are not discrete, but are inter-connected between the fields of diagnosis, therapy or surgery. Thus for the better understanding, nano-cardiovascular-imaging can be broadly divided into four categories depending on the site of detection and/or mode of action: *(i)* Thrombus imaging; *(ii)* Theranostic approach; *(iii)* Stem cell imaging; and *(iv)* Graft imaging

(i) Thrombus imaging : Acute coronary syndrome (ACS) is one of the leading causes of death in the world[58-62]. Atherosclerotic plaques in humans consist of different bio-components which are heterogeneous in nature, *i.e*. macrophages, smooth muscle, endothelial cells, other undefined mesenchymal cells, *etc* [63]. Proper detection of the plaques, in a non-invasive way is crucial and is the most demanding diagnostic procedure for the accurate treatment of the disease. In modern physics different non-invasive imaging techniques have been developed for the detection of plaques which generally require contrast agents[64,65]. The choice of the contrast agent depends on the type of technique used. Most of the clinically applied contrast agents pose two significant difficulties. First, these show sometimes toxic effects

Table II. Use of nanoparticles in different field of biomedicine.

Biomedical field	Nanoparticle	Application
Therapy		
	Gold	Tumour treatment[23,43,44]
	Silica gold nanoshell	Tumour treatment[24]
	Carbon	Tissue engineering[24,34]
	Iron oxide	Stem cell therapy[47]
Delivery agent		
	Gold	Drug, gene delivery[26]
	Polymer	Drug, gene delivery[25]
	Iron oxide	Stem cell delivery agent[46,47]
Diagnosis		
	Iron oxide	MRI contrast agent[41]
	Quantum dot	Microscopic contrast agent[32]
	Perfluoro carbon	MRI contrast agent[45]
	Silver	ELISA agent[42]
	Gold	ELISA agent[17,42]
Surgery		
	Hydroxy apatite	Cardiac stent[48]
	Titania	

Source: Refs 17, 23-26, 32, 34, 41-48

and second, these get non-specifically distributed to the whole body by circulation due to absence of any target specificity. This is where nano-based imaging system can come as a rescuer over the conventional clinical imaging techniques. For example, one of the most reliable imaging methods for the detection of plaques is by cardiac magnetic resonance imaging (CMRI), which requires a contrast agent gadolinium (GD), that often exerts toxic effects[66]. This toxic effect can be minimized by nanotechnology based approaches. It has been found recently that intracellular self-assembled gadolinium nanoparticles show enhanced MRI contrast ability with reduced toxicity[67]. The toxicity of GD is due to free ions. One way to reduce this toxicity is by using chelating agents, for example, diethylene triamine pentaacetic (DTPA). It has been shown that GD-DTPA exhibits less toxicity than GD alone[68,69], though there are some reports about the possibility of leakage of free ions from GD- chelate complex[70]. It has also been shown that nano-GD complex exhibits large loading capacity as well as large ionic relaxivity which in turn increases the contrast ability[71,72]. Anti-fibrin antibody conjugated oleate modified GD-DTPA nanoparticles (microemulsion) can effectively detect fibrin in vulnerable plaques[73]. Again, GD-chelate conjugation, incorporating nanoparticles reduces the toxicity to a significant extent[74]. Though nanotechnology based approaches manage some initial success to reduce GD toxicity, search is still on for new nontoxic agents for MRI. In that direction, ultra-small super-paramagnetic iron oxide nanoparticles (SPION) are thought to be an ideal substitution, as in the laboratory conditions these have shown high contrast ability with no toxicity[75]. One report shows that SPIONs have high efficacy for CMRI without manifesting any toxic effect in humans[76]. Same as SPION, perfluorocarbon nanoparticles contain fluorine, generate good contrast without any background signal, and can also be used in CMRI[45]. Another advantage of SPION is that these can be easily conjugated by any surface ligand, therefore, are efficient enough for targeting and therapy, compared to GD-chelate complex. The last and most effective point is that SPION are degraded by lysosomes and free iron from particles is released into the intracellular iron pool; hence, there is no chance of deposition inside the body[77] .

The atherosclerotic plaques are mainly of two types; stable plaques (fibrous plaque) and unstable plaques (lipid plaque). Unstable plaques are the main culprits for thrombosis, also known as vulnerable plaques[78]. Vulnerable plaques are made up of large amount of lipids, covered by a thin fibrous cap. Destruction of this fibrous cap makes the plaques unstable and these become detached from the endothelial layer[79]. This

phenomenon activates circulating resting platelets and the consequence is the formation of platelet rich thrombus. Thrombus blocks the artery, inhibits local circulation, resulting in muscle necrosis. Detection of the vulnerable plaques is a crucial step to initiate therapy for this disease.

Macrophages being one of the key components of atherosclerotic plaque play a decisive role in plaque destabilization[80]. These get attached to the thin fibrous cap of the plaques and secrete proteolytic enzymes which dissolve the fibrous cap[81]. Therefore, conceptually macrophages can be used as a good identifier of vulnerable plaques[82]. Phagocytic activity, which is the key feature of macrophages, has been exploited for the identification of the vulnerable plaque. It has been shown that macrophages can effectively take up a wide range of nanomaterials, including contrast enhancing nanoforms[83]. Therefore, nanoform loaded inflammatory macrophages on the plaque can be easily identified by non-invasive imaging techniques[84-88]. Now, the choice of imaging techniques will depend on the constituent of nanomaterials, or vice versa. For example, if macrophages are loaded with SPION, then CMRI will be the ideal technique, whereas CT scan can be done if the particles are iodinated. Gold nanoparticles can be used as good CT contrast agents. It has been found that gold nanoparticles have three times greater photon absorption capacity compared to iodinated contrast agent and, therefore, can enhance image contrast ability[89,90]. In addition, high contrast ability, inert character and surface modification are the other additional advantages of gold nanoparticles (AuNP). It has been found that CNA35 (small peptide which has excellent affinity for collagen) conjugated AuNP effectively identify myocardial scar signature by CT scan[91]. Au-HDL nanoparticles (gold nanoparticles coated with apolipoprotein A1, phospholipid and rhodamine lipid) specifically target macrophages of plaques and are identified in multicolour CT[92]. Specific targets of the plaque, choice of nanoforms and corresponding imaging techniques are listed in Table III.

Amino acid sequence specific targeting of plaque destabilize proteases (secreted by the inflammatory macrophages) can also be used as an identifier of atheromata. It was found that polymeric nanoparticles with fluorochrome labelled oligo-L-Lysine cleavage sequence (target for plaque specific proteases) can efficiently detect inflammatory plaque[93]. Factor XIII is another important constituent of acute thrombus; it converts linear fibrin to crosslink fibrin (fibrin α- and γ-chains), ultimately increases fibrinolytic resistance, and increases the plaque lifetime. Therefore, nano-mediated factor XIII specific targeting and imaging is another approach to detect thrombus. This detection process can also be applicable for other fibrin specific molecules. Magnetic nanoparticles coated with factor XIII, as well as fibrin specific peptide act as effective contrast agents for the detection acute thrombus, especially when thrombus is in its growing phase[94].

Reactive oxygen species (ROS) and reactive nitrogen species (RNS) are generated by atheroscerosis induced inflammation. Under certain circumstances these oxidizing species can neutralize local antioxidant defences, thus leading to oxidative stress and tissue injury. These oxidation reactions are mainly catalyzed by myeloperoxidase (MPO), a heme protein secreted from activated phagocytes in human atherosclerotic lesions[99]. Though *in vivo* imaging of ROS/RNS has significant clinical impact, yet there is no conventional method for their detection. An oxazine nano based imaging method has been developed to monitor hypochlorous acid (HOCl/OCl−) formation by peroxynitrite, a reactive nitrogen species and myeloperoxidase (MPO), thereby identify the oxidative damage by atherosclerosis[95,100].

(ii) Theranostic approach: 'Theranostics' is a newly established term in clinical medicine which deals with a treatment strategy in combination with therapeutics and diagnostics[101]. It can be defined as 'a modified diagnostic procedure equipped with therapeutic molecules/ device'. Theranosis has created a huge expectation in medical sciences because of its multimodal applications. It can reduce the steps and costs of both diagnosis and the therapy. Nanoparticles can themselves act as diagnostic probes (image contrast agent) and get conjugated with therapeutic or diagnostic molecules or vice versa .

Nanoimaging mediated cardiovascular theranosis is a recently introduced area. Simultaneous detection and volume reduction (thrombolysis/fibrinolysis) of thrombus is one such approach. The sole component of the thrombolytic / fibrinolytic pathway is plasminogen, which gets converted into plasmin (serine proteinase) by plasminogen activators, *i.e.* tissue-type PA (tPA) and urokinasetype PA (uPA). This plasmin then degrades fibrin and different extracellular matrix proteins (fibronectin, laminin, proteoglycan, and type IV collagen), thus reducing the plaque volume[102]. Recombinant tissue plasminogen activator (rTPA)

Table III. Different nano-conjugates and their mode of action.

Mode of action	Nanoparticles	Function	Target site	Imaging types
Thrombus detection	Albumin nanoparticles (HSA-NPs) loaded with gadolinium-diethylenetriamine pentaacetic acid (Gd-DTPA) and coupled with transferring. And	Provide a contrast enhancement	Brain, heart, liver, and skeletal muscle	MRI[72,74]
	Gold nanoparticle functionalized Gadolinium -DTPA anti-fibrin antibody conjugated oleate modified GD-DTPA nanoparticles (microemulsion)	Detection of vulnerable plaque	Fibrin clot targeting	MRI[73]
	Super paramagnetic nanoparticle	Detection of thrombus	Macrophage	Ultrasound[83]
	Magnetic nanoparticle coated with DTPA, fluorescent & PET tracer (64Cu)	Detection of thrombus	Macrophage	PET, MRI, Fluorescence imaging[84]
	Monocrystalline iron oxide nanoparticles(MION)	Detection of thrombus	Atherosclerotic plaque	PAT (Photoacoustic tomography)[85]
	Gold nanoparticles (Au-HDL) tagged with iodine based contrast agent.	Detection of thrombus	Macrophage	Multi colour CT scan[86]
	Iodinated nanoparticles (N1177)	Detection of thrombus	Macrophages	CT scan[87,88]
	Polymeric nanoparticle with oligo-L-Lysine cleavage sequence (PS)	Detection of thrombus	Proteases of inflammatory atherosclerotic plaque	FMT, CT[93]
	CNA35 (small peptide which has excellent affinity for collagen) conjugated with AuNP	Detection of myocardial scar	Collagen	CT[91]
	Au-HDL nanoparticles	Detection of atherosclerotic plaque	Macrophage	CT[92]
	Fluorescent and peptide coated magnetic nanoparticles.	Detection of thrombus	Fibrin and factor XIII	Fluorescence imaging, MRI[94]
	Oxazine conjugated nanoparticles	Reactive \ oxygen sp, Reactive nitrogen sp	Detection of myeloperoxidase, and hypochlorous acid.	Flow cytometry, FRI, MFI[95]
Stem cell delivery	SPION	Stem cell delivery	MCS	MRI[47]
graft rejection	Fluorescent conjugated magnetic nanoparticles	Phagocyte activity	Macrophages	MRI, fluorescence-mediated tomography (FMT)[50]
	ProSense-680 nano-construct (fluorogenic particle)	Protease activity	Cathepsin activity	FMT, CT[50]
Theragnosis	Magnetic nanoparticles with NIRF and light activated therapeutic moieties tagged	Macrophages in atherosclerotic plaque	Detection and destroy macrophages in inflammatory atherosclerotic plaque	Fluorescence microscopy[98]
	Iron oxide nanoparticles coated with factor XIII and tPA	Atherosclerotic plaque	Detection and treatment of thrombo embolism	Fluorescence reflectance imaging[96]
	Intergin $\alpha_v\beta_3$ targeted perfluorocarbon.	Neo angiogenesis	Intergin $\alpha_v\beta_3$	MRI[97]

Source: Refs 47, 50, 72, 73, 83-88, 91-98

MRI, magnetic resonance imaging; PET, positron emission tomography; PAT, photoacoustic tomography; CT, circular dichroism; FMT, fluorescence mediated tomography; FRI, fluorescence reflecting imaging; NIRF, near infrared fluorescence; SIPION, super paramagnetic iron oxide nanoparticle; tPA, tissue plasminogen activator

is now being recognized as an effective clinically used therapeutic molecule to dissolve plaque[103]. In this context, a nano-based theranostic approach can be conceptualised to monitor as well as to reduce plaque volume. It has been already found that iron oxide nanoparticles tagged with rTPA can efficiently dissolve clot[96]. A real time monitoring on thrombolytic effect has been done using nanoparticles coated with fluorophores. Therefore, diagnosis of plaque and reduction in its volume can be carried out simultaneously with the help of nanotechnology.

Another theranostic approach is detection and inhibition of angiogenesis. Angiogenesis is an important phenomenon during development of atherosclerotic plaque[104]. Neovascularisation is directly associated with plaque progression, risk of plaque rupture; therefore the subsequent consequence is myocardial infarction[105]. Integrin $\alpha_v\beta_3$ is only expressed in angiogenic vasculature, not in mature vasculature; hence can act as a marker of active angiogenesis[106]. To get molecular image (MRI) of angiogenesis, ultra small super paramagnetic iron oxide nanoparticle has been developed to target integrin $\alpha_v\beta_3$ receptor[107]. The research in this direction has further led to the detection and quantification of early angiogenesis (through MRI) by integrin $\alpha_v\beta_3$ targeted perfluorocarbon[108]. The ultimate nanotechnology based theranostic approach shows that fumagillin (potent angiogenic inhibitor) incorporated with paramagnetic nanoparticle not only detects early angiogenesis, but also effectively inhibits it[98].

Magnetic nanoparticles tagged with near infrared fluorophores and light activated therapeutic moieties can be used to detect and destroy inflammatory macrophages in atherosclerotic plaques. Intravenous administration of these nanoparticles in murine system showed that these were readily taken up by the macrophages and killed (phototoxic effect due to activation of therapeutic moiety THPC) them after exposure at 650 nm light. It is thought to be highly effective theranosis for atherosclerosis[97]. All the imaging based theranostic approaches are listed in Table III.

(iii) Stem cell imaging: The most recent approach though is under in-depth investigation, shows a hope of using stem cell technology in the treatment of cardiovascular diseases[109,110]. Infarcted myocardium cannot be replaced spontaneously; the reason behind it is that human cardiomyocytes are post-mitotic cells; therefore cannot proliferate after birth[111]. Recent findings show that mesenchymal stem cells (MSCs) are the bone marrow stromal cells which can differentiate into cardiomyocytes in an appropriate condition[112,113]. Most excitingly, transplantation of MSCs can improve cardiac activity in patients with myocardial infarction (MI)[114,115]. But the proper implementation of the MSCs that are going to be transplanted in terms of fraction (%) of cells reached to the infracted myocytes is of great importance in respect to prognosis of the disease. So far, SPION are found effective markers in this regard. Super paramagnetic iron oxide nanoparticle labelled stem cell tracking and targeting has been piloted effectively in animal models with chronic MI[46,116]. Cellular magnetic resonance imaging is found to be convenient method for the study of SPION guided delivery of MSCs to the infarcted muscle[47].

(iv) Graft imaging: Heart transplantation is the only treatment for patients with end-stage heart failure or severe coronary artery disease[117]. Even after heart transplantation, patients have to undergo repeated endomyocardial biopsies to see transplant graft rejection[118]. This procedure has significant risk, prone to sampling error and can induce fibrotic tissue build up at the site of biopsies[119]. A recent nanotechnology based approach has shown that fluorophore tagged iron oxide nanoparticle can efficiently diagnose this pathological condition[96,97]. Macrophages and cathepsin (protease) play a key role during graft rejection; therefore, these are attractive molecular imaging targets. These fluorescent conjugated magnetic nanoparticles have been used as a marker for macrophages with phagocytic activity and ProSense-680 nano-construct (fluorogenic particle) for determination of cathepsin activity[50] (Table III).

Concern

With the increasing demand of nanotechnology in day to day life, one should be concerned about its negative effects also. It is already established that one of the main targets that can be affected by nanotoxicity is cardiovascular system. The general toxicity effect is mainly due to nanoparticles present in atmosphere, in fuel exhausts from car, though in some cases it has also been found that designer nanoparticles (chemically synthesised nanoparticles in laboratory) can also exert toxic effect, if not properly modified.

Peters and colleagues[120] have shown that there is a consistent and clear dependence of duration of exposure to traffic with onset of myocardial infarction. The most common and ambient nanoparticles in traffic are carbon nanoparticles generated from diesel exhaust

which show toxic effect on vascular cells[121]. Air borne particulate matter enters into our body through alveolar wall during inhalation. After penetrating the alveolar wall it comes in contact with blood and thus gets access into the cardiovascular system[122] and induces cytotoxic injury, inflammation in endothelium, inhibition of cell growth, and cardiovascular toxicity[123].

Apart from the carbon black nanoparticles most of the metallic nanoparticles are found to induce platelet activation and aggregation thus increase the cardiac risk[124-126]. Cosmetics with nanoparticles [titanium oxide (TiO_2), silicon oxide (SiO_2)] also can increase the risk of cardiac arrest by inducing plaque progression, vasodialatory dysfunction, myocardial ischaemic damage, atrio-ventricular blockage, *etc.*[127,128]. Copper oxide (CuO) nanoparticle increases the oxidative stress, and ROS generation which ultimately activates plasminogen activator inhibitor-1 expression, and increases the risk of myocardial infarction[129]. Nickel nanoparticles have been shown to induce atherosclerosis during long term exposure in mice model[130]. The known toxicity of industrial nanoforms is listed in Table IV.

The toxic effects that seem to be induced by nanoforms might not be due to the nanoscale. The adverse effects are possibly due to the corresponding ions that get adsorbed on the outer surface of bare nanoforms, during the leaching of particles, when they are in solution phase[131,134] (Fig. 1). It is well known that metallic ions can induce oxidative stress in biological system[135]. Compared to the bare nanoparticles, ion coated nanoforms are easily taken up by cells as nanoparticles by virtue of their good penetration power[136]. Therefore, the effects shown are actually by the ions carried by nanoparticles. This concept is well correlated with some experimental facts, where it has been shown that surface modified nanoparticles do not show any toxicity (as leaching of ions are much less due to surface stability) compared to the bare nanoforms[137-140]. The carbon nanoparticle mediated toxic effect is probably due to a different mechanism. In this case, their (carbon nanoparticles) amorphous (nanotubes, or carbon black) and super hydrophobic nature is the major cause of their toxic effect[141].

Conclusion and future prospects

Nanoparticles along with their own unique properties (image contrast capacity, electromagnetic properties, bio-size compatibility, *etc.*) can be customised for individual needs. Nano-based imaging is applicable for both diagnosis and therapy. The nano-based diagnosis covers detection of disease condition, appropriate therapy, as well as detection of post-surgery conditions (Fig. 2). Though several techniques have already been developed to make nanoparticles as a potent candidate in the cardiovascular imaging field, yet there is much more to be done (Fig. 2). One important aspect is related to the stent technology. With the advancement of technology drug eluting stents have been developed which are more potent than the bare stent. Nano-mediated drug eluting stents are more efficient than the only drug eluting stent as nanoforms can increase the half life of the drug, by sustained release. Therefore, a new nano-based technique can be conceptualised with SPION, or nanoparticle with imaging probe, in drug eluting stent that will not only slow down the drug release, but also can be monitored in real time, by imaging devices. Another important

Table IV. Nanotoxicity of industrial nanoforms

Nanoparticles(NP)	Source and/or biomedical implications	Effect on cardiovascular system
Titanium oxide NP	Cosmetic industry	Plaque progression[127]
Silicon oxide NP	Cosmetic industry, drugs, printer toners *etc*	myocardial ischaemic damage[128]
Copper oxide NP	Aviation industry	Endothelial fibrinolytic activities[129]
Nickel NP	Alloys, Battery, *etc.*	Oxidative stress, atherosclerosis[130]
Silver NP	Antibacterial agent	Platelet pro-aggregatory effect [126,131]
Gold NP	Drug or drug carrier	Platelet pro-aggregatory effect[124,125]
Quantum Dots	Fluorescent probe	Pulmonary vascular thrombosis[132]
Iron NP	MRI contrast agent	Platelet pro-aggregatory effect[126]
Carbon Black	Petroleum exhaust	Vascular effect[121], Platelet aggregation[133]
Ultra fine particles	Air pollution	Atherosclerosis[123]

Source: Refs 121, 123-125, 127-133

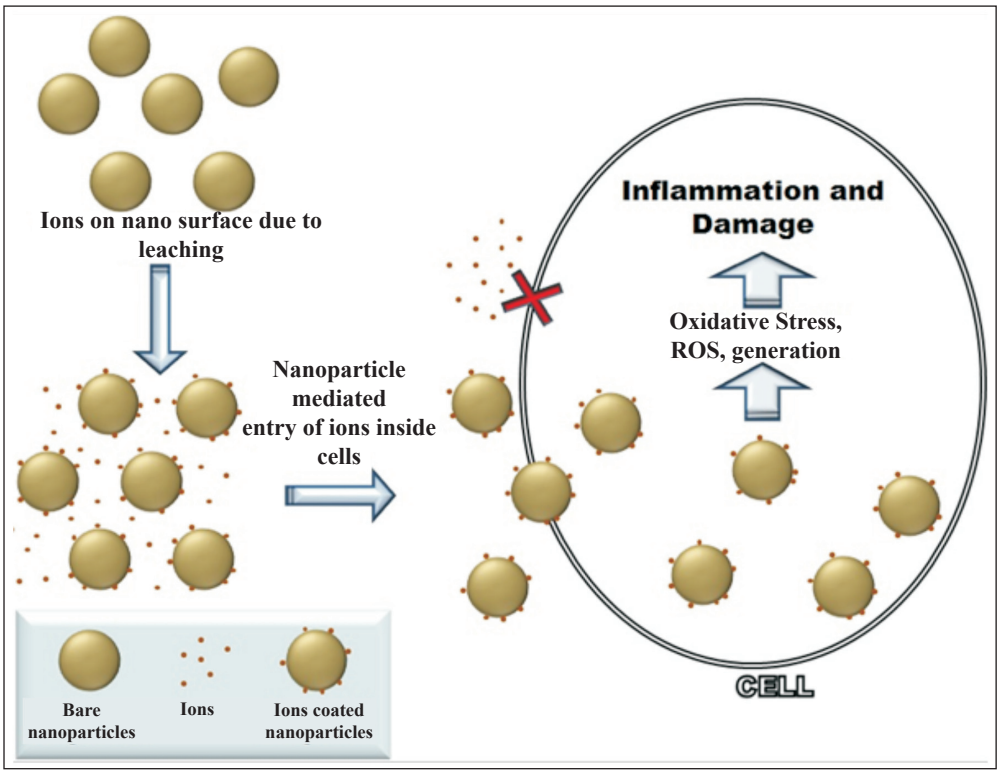

Fig. 1. Schematic representation of metallic nanotoxicity mechanism. Zero valent metallic nanoparticles rarely show toxicity but the ions leaching from it,often adsorbed onto outer surface and can induce oxidative stress while permeating healthy normal cell (ions themselves cannot penetrate cell membrane). ROS, reactive oxygen species.

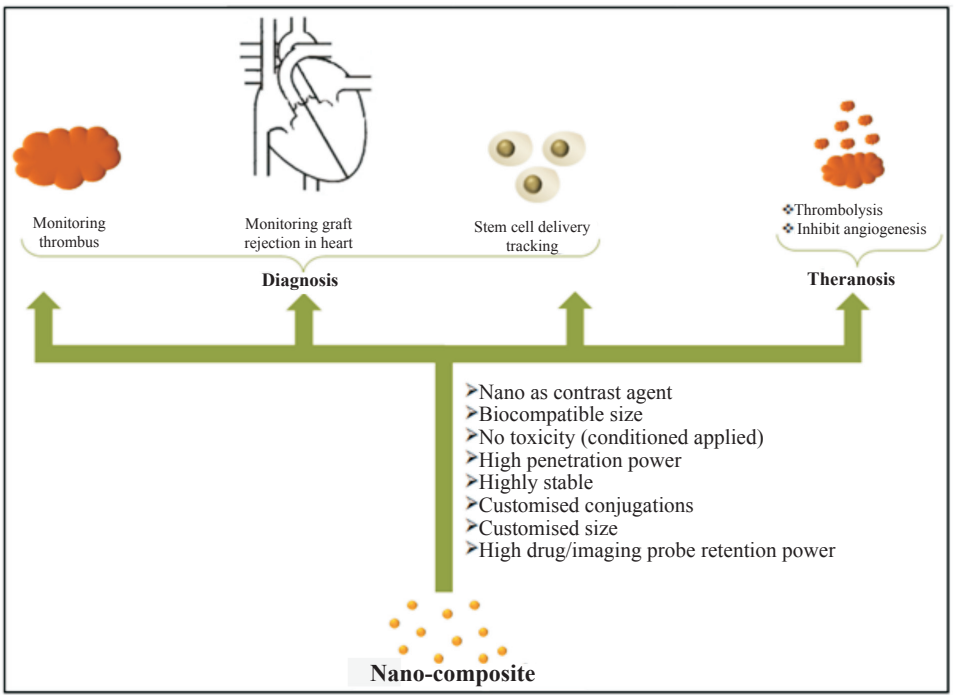

Fig. 2. Schematic representation of potential Nano-mediated cardiovascular imaging in diagnosis and therapy (*e.g.* identification of thrombus, monitoring graft rejection, stem cell tracking, and identification of angiogenesis, *etc*.)

application can be in the area of stem cell delivery. Gold nanowire is a good scaffold for the delivery of stem cell in the infarcted myocardium as it has the ability to synchronize the electrical signal in the cardiac stem cells. Therefore, one can hypothesize a SPION and gold nanowire based model scaffold system that will not only synchronize the rhythm but also be monitored on a real time basis. The same holds good for the theranosis of urokinase mediated thrombus reduction. In conclusion, nanotechnology imposes a huge scope in future clinical imaging field of cardiovascular diseases.

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Reprint requests: Dr Suryyani Deb, Department of Hemostasis & Thrombosis, National Institute of Immunohaematology (ICMR), KEM Hospital Campus, Parel, Mumbai 400 012, Maharashtra, India
e-mail : suryyani@gmail.com