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Uses of Nanoparticles in CNS Imaging and Therapy

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Annasaheb B Jagnar[*1], Priyanka M.Wavare[2], Trivedi Krishnaprem U[3], Akash Pawar[4]

1.Amrutvahini Institute of Pharmacy, Sangamner, Sangamner,Ahemednagar, Maharashtra,422 605,India.

2.S.P.C.O.P Otur,Tal-Junnar,Dist-Pune. India.

3. Parul Institute of Pharmacy and Research, Parul Institute, Limda, Waghodia, Vadodara, Department of Pharmaceutical Technology. Gujarat-391760, India.

4.School of Pharmacy,S.RT.M University Nanded. India.

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ABSTRACT

Nanoparticles are the devices that are in nanosize intended for administration into the body through parenteral routes. Nanoparticles are of very much importance nowadays due to their nano size level. Nanoparticles are the particles in the size range of nanometer. Due to their nano size level of the particle, they are easily absorbed from the stomach, and due to ease of absorption they can easily be solubilized and enhancement of bioavailability. Due to this significance of nanoparticles, they are widely useful in the treatment of various diseases. Nanoparticles are used as nanosponge, nanosphere, nanocapsule, nanosuspension, etc. Nanoparticles are widely used in the treatment of cancer and some brain-related diseases. This article gives an overview of various uses of nanoparticles in CNS imaging and therapy.

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INTRODUCTION:

Basic Composition [1]

Nanoparticles can be made from many different types of materials. Besides polymers, nanoparticles can be composed of ceramics, carbon, and various metals. They can be designed in several shapes, such as tubes, rods, hollow or solid spheres, and complex strands. Some materials and shapes are optimal for a specific application. Thus, there is no single preferred type of nanoparticle; rather, the adequacy of the material and shape used is dependent on the use for which the nanoparticle is intended. Nanoparticles for biologic imaging and/or delivery of therapeutic agents often consist of a central structure on which (or within which) an imaging agent or drug is held.

TYPES OF NANO PARTICLES-[2]

(1) Iron oxide particles

(2 Nanoshells

(3) Quantum dots

(4) Liposomes and Micelles

(1) Iron oxide particles

Iron oxide particles were one of the first nanoparticles developed and arguably have had the widest range of applications yet, compared with other nanoparticles. These particles have already received approval by the US Food and Drug Administration (FDA) for some clinical uses, specifically in imaging the bowel and abdominal viscera. Iron oxide nanoparticles contain ultrasmall paramagnetic iron oxide particles, which allow them to produce an increase in signal intensity on T1-weighted images, similar to that seen with commercially available gadolinium-containing MR imaging contrast agents. These agents have the advantage of also producing signal-intensity decrease on T2*-weighted images.

(2) Nanoshells

The application of a Dextran coating on the surface of the nanoparticles provides them with a much longer half-life (on the order of 24–30 hours) than conventional MR imaging contrast

agents; as a result, imaging can be performed many hours after infusion. Advantages include allowing a single infusion of contrast material many hours before surgery while still being able to perform intraoperative MR imaging by using the same dose of contrast material. For instance, one type of iron oxide nanoparticle termed ferumoxtran-10 can be detected in surgical and biopsy specimens by using an iron stain. Thus, surgical specimens can be microscopically examined to determine whether the resected specimen was removed from enhancing portions of the tumor. This process allows better identification of sites on imaging studies from which tissue has been resected as well as the closer correlation between imaging findings and histologic features than would be available with routine contrast agents.

(3) Quantum dots

Advantages of quantum dots include light emission that is size-dependent (ie, the wavelength of the emitted light is related to small changes in the semiconductor core of the nanoparticle due to a quantum mechanical phenomenon called quantum confinement), very bright signal intensity, and resistance to photobleaching with time (ie, photostability). This last feature is important because it allows quantum dots to be used for imaging for longer periods than is allowed with other optical imaging agents, such as organic fluorophores. The relationship between the size of the nanoparticle and the wavelength of light emission (tunability) is important because it allows different particles to be used simultaneously in the same biologic preparation or tissue; the presence and location of individual quantum dots can be determined by the color they emit. For example, quantum dots of a few different sizes can be used, with a different antibody used for each size of the quantum dot, allowing imaging of multiple biologic targets (so-called "multiplexing capability"). For instance, the use of 3 different sizes of quantum dots in an experiment, with each size quantum dot having a specific antibody, would allow 3 different molecular targets (eg, 3 different types of the receptor) to be imaged. The 3 different types of quantum dots (bound to their respective 3 targets) will be distinguishable based on the emission of a specific wavelength for each type of quantum do.

(4) Liposomes and Micelles

Liposomes" are nanoscale vesicles having a phospholipid bilayer membrane and an aqueous core. The aqueous core provides an environment in which therapeutic drugs can be sequestered for transport to target sites, thereby protecting drugs from actions at unintended targets and degradation. Like many other types of nanoparticles, ligands that bind biologic

targets can be attached to the surface of liposomes for targeted delivery. These particles can be made to provide sustained time-release of their contents. Liposomal formulations of doxorubicin have been approved by the FDA for clinical use in tumors such as ovarian cancer and multiple myeloma; others are present in the stage of testing of feasibility for human use. Liposomes have recently been developed for the treatment of brain tumors but are still in the investigational stage in that setting.

Micelles" are spheric aggregates of molecules, in which the hydrophilic regions of the molecules face outward and the hydrophobic portions of the molecules face inward. Micelles offer a means of allowing a compound that is normally insoluble in a particular solvent to become soluble by being sequestered in the hydrophobic core of the micelle. The external hydrophilic shell allows the micelle to serve as a nanocarrier, which permits the delivery of greater amounts of a drug to a target tissue compared with the intravenous administration of the free drug. Thus, micelles may offer one mechanism for increased distribution of drugs across the BBB, an issue that is discussed further below.

Nanoparticle Delivery[3]

Issue of Bio- Distribution

One of the major challenges investigators face is the effective delivery of nanoparticles to the organ of interest rather than to unintended targets. One major impediment in the delivery of nanoparticles administered by intravenous infusion is sequestration by the reticuloendothelial system. Nanoparticles will typically be captured in the liver and spleen unless, during the manufacturing process, a deliberate attempt is made to provide a means to escape capture by the reticuloendothelial system. The most commonly used technique is to coat the nanoparticles with a covalent attachment of polyethylene glycol (PEG), which renders the nanoparticle essentially invisible to the reticuloendothelial system; such particles are sometimes referred to as stealth particles. This process (referred to as PEGylation) substantially prolongs circulation time and allows the nanoparticle to be delivered to the organ of interest.

One agent of interest is a poly(butyl cyanoacrylate) nanoparticle coated with polysorbate 80, which adsorbs apolipoproteins B and E and allows receptor-mediated endocytosis by brain capillary endothelial cells. Polysorbate 80 nanoparticles have also been used for CNS drug therapies for nonneoplastic disorders and have similarly been shown to cross the intact BBB

in reasonable amounts. In several studies, doxorubicin bound to nanoparticles has been shown to cross the intact BBB and reach therapeutic levels in the brain as well as to prolong survival times significantly in rats with glioblastomas. A significant challenge facing the use of these nanotechnologies for delivering drugs and other small molecules across the BBB is that in addition to their primary function of having to deliver enough drug to elicit a therapeutic effect, at the same time, the nanoparticle-drug conjugates must curb unintended systemic side effects by limiting undesired molecular interactions with cell types other than those they are designed to target. Thus, simply increasing the concentration of a nanoparticle that crosses the BBB to deliver a greater quantity of drugs may not be possible if the higher concentration results in increased nonspecific molecular interaction events. This is a formidable but unavoidable challenge that faces the development and use of nanotechnologies aimed at CNS drug delivery.

Many other techniques for enhanced delivery across the BBB are being evaluated. However, because the topic is a broad one that cannot be easily summarized here, the reader is referred to a review article specifically addressing the issue of BBB transport.

Nano Particles directed against target[4]

Nanoparticles can be targeted (ie, modified with specific binding properties directed against molecular targets) or nontargeted. Nontargeted nanoparticles passively accumulate at the site of interest (eg, within a tumor). Thus, nanoparticles administered intravenously are passively delivered under the guidance of normal blood circulation. Two phenomena allow non-targeted nanoparticles to congregate within tumors. First, vessels produced by the angiogenesis that accompanies tumor growth are known to exhibit marked leakiness, allowing extravasation of nanoparticles into the tumor microenvironment. Also, because tumors typically lack an effective lymphatic drainage system, the egress of nanoparticles away from the tumor is impaired. This dual phenomenon has been termed the “enhanced permeability and retention effect”.

The term “targeted nanoparticles” refers to those manufactured with a surface ligand or other surface modification designed to allow the nanoparticle to bind to a target such as a cell membrane receptor or other protein. Thus, targeted nanoparticles can selectively bind to sites of interest (eg, tumor cell membranes). A wide array of modifications of nanoparticles is

available; binding of ≥1 antibody against the intended target is one of the more common strategies used. Some of these strategies are described in subsequent parts of this review.

Drug delivery using Nano particles[5]

The term "targeted nanoparticles" refers to those manufactured with a surface ligand or other surface modification designed to allow the nanoparticle to bind to a target such as a cell membrane receptor or other protein. Thus, targeted nanoparticles can selectively bind to sites of interest (eg, tumor cell membranes). A wide array of modifications of nanoparticles is available; binding of ≥1 antibody against the intended target is one of the more common strategies used. Some of these strategies are described in subsequent parts of this review.

Nanoparticle-mediated selective drug delivery may allow a means to minimize delivery to unintended sites, potentially allowing larger doses of the drug to be administered and with a greater percentage of drug reaching the target, thereby possibly lowering toxicity. Many methods of selective delivery exist. In addition to targeting strategies using ligands on nanoparticle surfaces mentioned earlier, strategies have been devised for the release of nanoparticle contents by using stimuli external to the body utilizing a focused trigger, such as light (so-called "photodynamic therapy") or heat. Antineoplastic therapy by using heat-labile liposomes is one example. After intravenous administration of the liposomes and passive delivery to the tumor, heating of the tumor (by using various means such as focused sonography) can cause the targeted release of liposomal contents directly at the tumor, thereby minimizing systemic drug effects.

Adaptation of Nanoparticles for Specific Functions[6]

pH-Sensitive Nanoparticles

For effective nanoparticle-borne antineoplastic therapy, nanoparticles must exit the bloodstream into the tumor interstitium, enter cells, and, in many cases, be taken up within endosomes. However, each compartment has its pH. For instance, the pH of the endosomal system (ie, range of 5.0–6.0) is substantially lower than physiologic pH (ie, pH of 7.4) and the extracellular pH of solid tumors (ie, pH of 6.8). The differing pH of various compartments can potentially affect transport and stability of nanoparticles. With this in mind, nanoparticles whose structures are altered by the pH of the local environment have been engineered. Specifically, a PEG coating on a nanoparticle is reversibly removed at a pH

of 6.8 and reattached at a pH of 7.4. Note that a nanoparticle of this type could potentially be altered to provide information about local pH, which might be important in many scenarios, such as understanding drug resistance to chemotherapeutic agents that are pH-sensitive.

Dual Capability Imaging Using Nanoparticles[7]

Another innovative technique is the development of nanoparticles that combine optical nanoprobe systems with the capacity for more conventional imaging techniques, such as MR imaging. As an example, investigators have developed methods to fuse fluorescent dyes and magnetic nanoparticles into a single nanoprobe. In one such probe, a dye-doped silica core is surrounded by water-soluble iron oxide particles, which can be detected by using T2*-weighted imaging. When coated with an appropriate antibody (in this case, an antibody against polysialic acids on neuroblastoma cells), the dual-purpose nanoparticles can be used to image the cells by using both fluorescence imaging and MR imaging. Furthermore, synergistic magnetism between the multiple iron oxide molecules markedly increases the T2 relaxivity of the agents and provides greater conspicuity on MR imaging.

Uses of Nanoparticles in specific function[8]

pH-Sensitive Nanoparticles

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Uses of Nanoparticles in CNS Process[9]

Tracking Functional Responses in Neurons

One advantage of nanoparticles is the enhanced ability to study and track molecular events within neurons and glia over many seconds or even minutes. Types of cells can be identified,

and their number, development, and physiologic responses can be monitored using fluorescence microscopy with quantum dots that are coated with an antibody directed against specific cellular features.

For instance, nanoparticles bound to effector compounds (ie, molecules that initiate a response in cells) can be used to interrogate the functional capabilities of cells in the CNS. Such nanoparticles can serve the dual function of both an imaging probe and a functional probe that induces a cellular response, such as a change in cell metabolism. In one study, investigators bound quantum dots to nerve growth factor, a peptide hormone that targets motor, sensory, and autonomic neurons and is important for neuronal development and survival. Exposure of cells from a highly tumorigenic teratoma-derived cell line to such quantum dots over a few days induced changes in cell-signaling mechanisms and cell behavior as well as the development of neurites extending from cell bodies.

Nanotechnology offers several very interesting methods to monitor the fundamental features of cellular organization in real-time. An important issue in the understanding of synaptic development and neurotransmitter function is the mobility of neurotransmitter receptors along cell surfaces. Investigators have used quantum dots to track the lateral motion of glycine receptors (the major inhibitory neurotransmitter in the adult spinal cord) within the membranes of living cells for many minutes Here, the photostability (i.e., sustained ability to provide signal intensity with time) and improved signal intensity–to-noise ratio (ie, fluorescence signal intensity from the quantum dot relative to nonspecific background fluorescence) of quantum dots provides a real advantage relative to standard fluorophores, which are more photolabile (i.e., their fluorescent properties degrade with time). Also, the in-plane resolution offered by quantum dots is markedly superior to that offered by standard fluorophores.

Iron Oxide Nanoparticles for MR Imaging of Brain Tumors[10]

Because ultrasmall paramagnetic iron oxide−labeled nanoparticles were among the first nanoparticles to be used in humans, investigators have gained a fair amount of experience with the use of these particles. MR imaging agents based on such technology have been used for preoperative and intraoperative imaging of human brain tumors. MR imaging contrast agents that use these nanoparticles have the unique capability of contrast enhancement of brain tumors for days after administration. Most interesting, the nanoparticles appear to

accumulate in macrophages and reactive astrocytes immediately adjacent to tumor cells rather than within tumor cells.

Superparamagnetic nanoparticles have been developed for targeted therapy of brain tumors. Conjugation of the surface of the nanoparticle with chlorotoxin allows targeting of tumors of neuroectodermal origin (including gliomas) that contain membrane-bound matrix metalloproteinase-2. Experiments have shown selective targeting of tumors by the chlorotoxin-conjugated nanoparticles compared with nanoparticles that are not chlorotoxin-bound. Although a very small amount of nontargeted nanoparticles were found in the tumor, the number of targeted nanoparticles in the tumor was markedly greater. Also, nanoparticles were not reported in organs other than the brain.

Nanoparticles as Neuroprotection Devices[11]

In the past few decades, increased emphasis has been placed on the development of agents that could limit the effect of injurious CNS events such as cerebral infarction and brain trauma. Following such injuries, several chemical species are released that contribute to ongoing tissue damage as part of secondary injury mechanisms; examples include oxygen free radicals and superoxide and peroxide molecules. The accumulation of these products leads to some processes such as impaired mitochondrial energy production, inactivation of transporter proteins, increases in intracellular calcium concentrations promoted by elevated glutamate levels, and promotion of apoptosis. Various approaches, mostly pharmacologic, have been attempted to diminish the local concentrations of such substances. Nanoparticles represent another possible method for limiting brain injury. For example, fullerenes (nanoparticles comprising arrays of regularly spaced carbon atoms) have been investigated for this purpose and have shown promise as antioxidants with the capacity to scavenge free radicals. In theory, nanoparticles can be developed that are capable of releasing therapeutic agents directly at the site of CNS injury and are a potential novel approach for achieving neuroprotection.

Nanoparticles for limiting the degree of tissue injury following events such as cerebral infarction or trauma. Nanoparticles could potentially be used as neuroprotective agents by limiting the effect of substances produced by injury, such as free radicals. In principle, nanoparticles could be loaded with materials that negate the injurious effects of free radicals or could serve as scavengers of free radicals. *B*, The use of nanoparticles to produce self-

assembled scaffold materials that can provide the structural environment for neural regeneration, such as a medium for regrowth of neurons. *C*, Enhanced delivery of therapeutic agents by nanoparticles specifically designed to cross the Blood-Brain Barrier.

Monitoring of Stem Cell Migration within the CNS[12]

Application of stem cells for the treatment of CNS diseases is a topic that is gaining intense interest among neuroscientists and clinicians alike because it may provide a means to regenerate damaged brain tissue (eg, after trauma, degeneration, or cerebral infarction) or replace missing enzymes in enzymatic disorders. As an example, stem cells are already in clinical use as a form of enzyme replacement for the treatment of pediatric leukodystrophies such as Krabbe disease. At present, stem cells are typically provided intravenously, but they could potentially be stereotactically placed within CNS tissue.

Iron oxide nanoparticles are a means by which the migration of nanoparticle-tagged cells can be monitored by MR imaging. In one study, investigators produced cerebral infarction in mice and then implanted stem cells containing iron oxide nanoparticles. MR imaging was successfully used to detect the location of transplanted cells. In another study of mice in which brain or spinal cord injury had been produced, researchers followed the migration of iron oxide−labeled stem cells implanted in the CNS to sites of injury. In combination, these studies show a potential role for MR imaging surveillance of labeled stem cell migration for CNS therapies in humans.

Nanoparticles as a Means for CNS Tissue Regeneration[13]

Nanoparticles offer a potential means to enhance the body's reparative mechanisms by providing building-block molecular materials or products needed for CNS repair. Alternatively, self-assembling nanodevices can be administered that can serve as platforms for regenerative processes. One example is a constellation of nanofiber scaffolds that can self-assemble within the CNS to promote the growth of neurites. For instance, peptide amphiphile molecules have been designed that self-assemble in a physiologic environment in response to appropriate cation concentrations in a configuration that allows physiologically active peptide sequences to act as ligands for cell surface receptors. The peptides can then engage in cell signaling, which promotes cell growth and differentiation. In one experiment, scaffolds of the type outlined above trap water molecules the following self-assembly into nanofibers and form a gel-like material in which neural progenitor cells are encapsulated.

Rapid (ie, within a day) and robust differentiation of the neural progenitor cells into mature neuronal cells was induced by a combination of the 3D nanostructure of the gel and the bioactive signaling peptide on the surface of the nanofibers, thereby potentially providing a method to regenerate CNS tissues. Furthermore, very little astrocyte development was noted, which suggests that reactive gliosis and glial scarring have been minimized.

Nanoparticles as Sensors within the CNS [14]

One intriguing possible use of nanoparticles involves indwelling sensors within the CNS, which could provide intermittent or continuous measurement of physiologic processes within the local environment. As an example, a nanosensor that is based on the principle of a magnetic nanoswitch has been proposed. A nanoswitch is a functionalized iron oxide particle that can be used to monitor in vivo dynamic events such as changes in concentrations of molecules in the local nano environment. These nanoparticles undergo reversible assembly and disassembly in the presence of specific molecules (eg, glucose, enzymes, and other proteins). The assembly state and the disassembly state are each associated with a different transverse magnetic relaxivity. The different relaxivities can be detected by using MR imaging based on T2 relaxation times, thus providing a noninvasive means for detecting physiologic changes that are reflected by varying concentrations of specific molecules.

Nanoparticles for Imaging of Angiogenesis [15]

One of the major applications of molecular imaging for the assessment of tumors is the development of nanoparticles for imaging of angiogenesis. One interesting advance is the development of a lipid-encapsulated perfluorocarbon nanoparticle that can be modified for imaging by using sonography, nuclear medicine techniques, or MR imaging. The nanoparticle is optimized for MR imaging through conjugation with very large numbers of gadolinium particles, thereby compensating for the poor signal-intensity contrast, which is one of the inherent limitations of MR molecular imaging agents. By conjugating ≥1 ligand to the surface of the nanoparticle, targeted imaging can be accomplished. Specifically, for imaging of angiogenesis, the nanoparticle is targeted against a protein that is expressed on the luminal surface of angioblasts within neovasculature (ie, $\alpha_v\beta_3$-integrin). With this technique, imaging of small animals has been successfully conducted on a 1.5T scanner, raising the promise that if such imaging could be performed in humans, MR imaging scanners that are

presently clinically relevant in humans could be used (as opposed to the very high-field-strength scanners often used for small-animal imaging).

In addition to serving as an imaging agent, angiogenesis-targeted nanoparticles can serve as a therapy-delivery vehicle. In one study, investigators developed a polyacrylamide nanoparticle-containing iron oxide particles (thereby allowing use as an MR imaging contrast agent) and also having a tumor vasculature−targeting peptide on its surface. The nanoparticle also contained a photosensitizing molecule that, when activated by light, produces microvascular injury and tumor cell death. The study showed good deposition of the nanoparticles within 9L glioma tumors in rats and substantial improvement in the survival rate of treated animals after administration of phototherapy.

CONCLUSION:

This review has provided an introduction to a number of the most important applications of nanotechnology to imaging of the CNS. The topics here are necessarily presented in limited detail; the reader is encouraged to learn more about this important and exciting field of investigation by reading articles that have been cited. The fact that many of the topics covered here are still in discovery mode should not discourage neuroradiologists from taking a serious interest in topics in nanotechnology-related to imaging. Although the nanotechnology applications are not ready for clinical translation at this time, little doubt can be entertained that, in one form or another, some of the general principles described here will prove clinically relevant. After all, many techniques that are presently the mainstay of neuroradiologic practice were once also regarded (as nanotechnology applications might now be considered) as solely exciting theoretic principles without practical application.

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