Nanomaterials: A realistic business case for Pharma

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ABSTRACT

In this presentation, an attempt will be made to identify the realistic potential of nanotechnology based medicines, including e.g. improved tissue targeting of biopharmaceuticals; raising the therapeutic index of existing marketed drugs; reducing off-target interactions and improving the side effect profile of candidate molecules; modifying pharmacokinetic properties and hence allowing more convenient dosing regimes and improving patient compliance. The barriers to the successful integration of nanotechnology based therapeutics will also be explored, including, their compatibility with the existing drug discovery and development paradigm. The potential challenges associated with the manufacturability and product quality control of nanotechnology based drugs will be discussed. Finally, examples of in-house projects employing nanomaterials will be presented.

Keywords: nanomaterial, patients, drug delivery systems, development partnerships, barriers

1 NANOMATERIAL OVERVIEW

Nanoscale science studies the phenomena, properties and responses of materials at atomic, molecular and macromolecular scales, and in general at sizes between 1-100 nm. Nanotechnology is then the design, the manipulation, the building, the production and application, by controlling the shape and size, the properties responses and functionality of structures, devices and systems of the order or less than 100 nm.

The focus of nanomaterials is a bottom up approach to structures and functional effects whereby the building blocks of materials are designed and assembled in controlled ways. Nanomaterials exhibit bulk mesoscale (i.e. mid-range between micro and macro) properties that are unique to the material, and are not possessed by the constituent molecules that make them up. Nanomaterials have the potential to perform multiple specific functions at once or in a predefined sequence, important properties for the clinically successful targeting and delivery of drugs [1].

Two principal factors cause the properties of nanomaterials to differ significantly from other materials which are (1) increased relative surface area, and (2) quantum effects. These factors can change or enhance properties such as reactivity, strength and electrical characteristics.

Understanding how the human body clears particles is essential to developing effective nanomaterial-based delivery and targeting systems. Intravenously injected particles are scavenged and cleared from the circulation by Kupffer cells and macrophages in a process that is facilitated by surface deposition of blood opsonic factors and complement proteins on the injected drug particle. Both clearance and opsonization can, however, be influenced by the size, shape & surface characteristics of particulates [2].

Although the definition of what constitutes "nano" is that a material has features between 1 to 100 nm, GSK takes a more pragmatic approach and we would consider anything below 500 nm (the practical limit of most nanomilling systems) to be nanomaterial.

2 PATENT LANDSCAPE

Patents [3] are a good measure of what organisations are planning and the level of commitment shown in specific area. It is expensive to set up a patent.

Close to 3000 patents were issued in the USA from 1996-2006 with the term 'nano' in the patents, with a considerable number having application in nanomedicine. The majority of therapeutic patents are focused on drug delivery systems employing nanomaterials to achieve superior drug absorption, controlled drug release or reduced side-effects.

Nanopharmaceutical patents are centred mainly on non-communicable diseases, with cancer receiving the greatest focus, followed by hepatitis. Other non-communicable diseases with significant nanomaterial based patent activity include osteoporosis, beri-beri, stroke and diabetes mellitus. Malaria and tuberculosis, are noticeably absent from any significant level of nanopharmaceutical patenting.

3..DRUG DELIVERY SYSTEMS

GSK is constantly reviewing a selection of platform technologies. The assessment criteria's are against a set of aspirations which address the needs of the patient and the imperatives of the industry e.g. regulatory acceptance, applicability to different delivery routs, cost, development stage, manufacturability & scalability, EHS, time to market. As a result, a broad assessment of their key features has been done. Furthermore, published examples will be given of collaborations between GSK and an external party e.g. Alkermes, SIRNA, and Nektar as well as in-house activities presented during the presentation.

3.1 Polymeric particulates

The rapid advances in nanomaterials have led to the intense research of nanoparticulate drugs as therapeutic carriers. The use of nanoparticulate drugs has the potential to significantly improving the drug systemic bioavailability due to their higher dissolution rate [4]. Therapeutic active can either be physically dispersed within the matrix of polymer-based carriers [5], encapsulated within or loaded on the surface of the inert carriers [6], such that the drug release rate and pattern can be controlled. Particles may be produced by polymerization of synthetic monomers, or dispersion of synthetic polymers or natural macromolecules [7]. A wide range of nanoparticulate drugs for oral and parenteral delivery has been investigated which is evident in the number of commercialized products.

3.1.2 Absorption of polymeric particulates via the gastrointestinal tract [1]

Oral absorption of particulates has been described as early as 1844. Particles are described as crossing either at the level of Peyer's patches or through the enterocyte layer.

The number and location of Peyer's patches vary widely between species and individuals and are also age dependent. Physicochemical properties of particles influence greatly their rate of uptake by the intestinal tract. The two main factors are the size and the nature of the polymer used to make the particles. The absorption takes place primarily, but not exclusively, in the Peyer's patches at the level of the M cells. Uptake is very fast and seems to be the result of a transcellular mechanism but a paracellular pathway, although very unlikely, cannot be excluded. The particulate/cell membranes interface appears to play an important role and all the parameters involved in the binding of particles at the cell surface have not been yet identified. The modification of particle surface properties due to the nature of the polymer or due to the binding of specific proteins or lectins can modulate the absorption rate.

3.2.2 Nanoparticle approaches for crossing the blood brain barrier (BBB)

The approaches for crossing the BBB [1] have focused on 1) the delivery of antineoplastic drugs to CNS tumors and 2) treatment of a variety of CNS disorders: Radiolabeled PEG-coated hexadecylcyanoacrylate nanospheres (rat model of gliosarcoma), showed a several fold increase in accumulation in tumors. Encapsulated antineoplastic drug paclitaxel in PLGA nanoparticles showed targeted cytotoxicity some 13 times greater than with the drug alone in 29 different cancer cell lines.

Neuropeptides such as enkephalins, the NMDA receptor antagonist MRZ 2/576, and the chemotherapeutic drug doxorubicin have been attached onto the surface of poly(butylcyanoacrylate) nanoparticles coated with polysorbate 80 [8]. [^{3}H] Dalargin conjugated to poly(butylcyanoacrylate) nanoparticles and injected systemically into mice was shown by radiolabeling to cross the BBB and accumulate in the brain. Studies have also shown the delivery of dalargin by using polysorbate 80–coated nanoparticles.

PEG-treated polyalkylcyanoacrylate nanoparticles were shown to cross the BBB and accumulate at high densities in the brain in EAE, a model for multiple sclerosis. The polysorbate on the surface of the nanoparticles is thought to adsorb apolipoproteins B and E and is taken up by brain capillary endothelial cells via receptor-mediated endocytosis.

Molecules other than drugs need to cross the BBB for therapeutic or diagnostic reasons, including oligonucleotides, genes, and contrast agents. Nanoparticles loaded with iron oxide, injected systemically into rats have been shown to cross the BBB and accumulate in the brain. Iron oxides are superparamagnetic MRI contrast agents that directly affect both T1 and T2 water molecule relaxation times. Nanoparticles complexed with iron oxides may provide a novel approach to imaging the CNS by MRI.

Abraxane is a polymeric nanoparticle based product from American Bioscience, Inc. Approved in 2005, the product consists of albumin-bound paclitaxel nanoparticles. This product is free of toxic solvents such as cremophor-EL, which had been employed to solubilise paclitaxel for IV administration.

3.2 Liposomes

Liposomes or lipid vesicles are colloidal particles composed of (phosphor) lipid molecules as the major constituent in formation of enclosed lipid bilayers or lipid-drug sheet-disk complexes. Most of the liposome formulations approved for human use contain phosphatidylcholine (neutral charge), with fatty acyl chains of varying lengths and degrees of saturation, as a major membrane building block. A fraction of cholesterol (~30 mol%) is often included in the lipid formulation to modulate rigidity and to reduce serum-induced instability caused by the binding of serum protein to the liposome membrane.

The exact mechanisms of biodistribution and disposition *in vivo* vary depending on the lipid composition, size, charge, and degree of surface hydration/steric hindrance. In addition, the route of administration may also influence the *in vivo* disposition of liposomes. Immediately after intravenous administration, liposomes are usually coated with serum proteins and taken up by cells of RES and eventually eliminated. Plasma proteins that may interact with liposomes include albumin, lipoproteins [i.e., high-density lipoprotein (HDL), low-density lipoprotein (LDL),

etc.] and other cell-associated proteins. Some of these proteins (e.g., HDL) may remove phospholipids in the liposome bilayer, thereby destabilizing the liposomes.

The use of high quantities of the carrier can lead to problems of carrier toxicity, metabolism and elimination, or biodegradability [9]. Because each liposome can entrap up to tens of thousands of drug molecules, drug potency is less of an issue for this type of carrier. However, even the relatively high carrying capacity of liposomes becomes problematic for very large therapeutic molecules such as proteins, particularly if small liposome diameters are desirable for reasons of biodistribution. Hydrophilic drugs can be readily entrapped with a high degree of latency within the liposome aqueous interior, but neutral hydrophobic drugs or those with intermediate solubility's tend to be rapidly released in the presence of plasma proteins or cell membranes. Excellent retention of drugs that are hydrophobic weak bases (such as doxorubicin and vincristine) has been achieved through "remote loading" techniques that rely on pH or chemical gradients across the liposome bilayer to accumulate and retain the drug [9].

Doxil, a pegylated liposomal formulation of doxorubicin was one of the initial nanopharmaceuticals and liposomal formulations introduced to the market. Their long circulating 'STEALTH' liposomes with hydrophilic PEG surface and diameters <200nm were found to target tumour tissue by the mechanism of enhanced permeation and retention. The liposomal formulation of doxorubicin thus considerably reduced the cardio-toxicity of the drug.

However, despite having already regulatory accepted drug carriers in form of liposomes on the market, at this point no site-specific drug carrier system is on the market [10]. Liposomal products on the market such as Doxil® (Doxorubicin) or DaunoXome® (Daunorubicin) are not targeting systems, they are controlled release systems to minimise side effects (heart toxicity) by reducing the concentration of free drug in the blood.

There are again several reasons for this lack of targeted systems, examples are:

- Using molecules as targeting moiety (e.g. mannose) has the problem that receptors for these molecules are on many different cells, not only on the target cell.
- Binding of targeting moieties in form of molecules or monoclonal antibody leads very often again to recognition by the MPS.
- Most of the technologies are complex,

3.3 Dendrimers

Dendrimers [11] are highly branched, globular macromolecules with many arms emanating from a central core. The stepwise synthesis of dendrimers affords molecules with a highly regular branching pattern, a unique molecular weight or a low polydispersity index, and a well-defined number of peripheral groups. A comparison with linear polymers shows that the dendritic architecture can provide several advantages for drug delivery applications. For example:

- Controlled multivalency of dendrimers can be used to attach several drug molecules
- The low polydispersity of dendrimers should provide reproducible pharmacokinetic behaviour
- The globular shape of dendrimers, as opposed to the random coil structure of most linear polymers, could affect their biological properties

The research in dendrimer mediated drug delivery has mainly been focused on the delivery of DNA drugs (genes or gene inhibitors) into the cell nucleus [12]. Current dendrimers based systems under development include those which are, biocompatible, water soluble, biodegradable, and formed with monomer units that are chemical intermediates or products in metabolic pathways.

Anticancer drug carriers exploit multivalency in the covalent attachment of drug molecules to the dendrimer periphery. The drug loading can be tuned by varying the generation number of the dendrimer, and release controlled by incorporating degradable linkages.

One of the areas that remain to be addressed in more detail is the biodistribution behaviour of dendrimers.

Products based on dendrimers are progressively entering the market and Vivagel, developed by Starpharma, is the first product based on dendrimers. VivaGel is a Topical Vaginal Gel instituted in the prevention of HIV and STIs.

3.4 Nanocrystals

Nanocrystals [10] are aggregates of around hundreds or thousands of molecules that combine in a crystalline form, composed of pure drug with only a thin coating comprised of surfactant or combination of surfactants. To produce nanosuspensions, the drug powder is dispersed in an aqueous surfactant solution by high speed stirring. The obtained macrosuspension is then homogenized to nanosize by wet milling, high-pressure homogenization, and nanocrystallisation from supersaturated solution and spray drying. Problems typical of poorly soluble drugs like reduced bioavailability, improper absorption pattern and problems of preparing the parenteral dosage form may be resolved by formulation as nanocrystals.

The NanoCrystal technology from Elan was one of the earliest scalable technologies developed for nanosizing drugs. Elan's NanoCrystal particles are typically less than 1000 nm in diameter (dominantly crystalline as opposed to amorphous) and produced by milling the drug substance using a proprietary, wet-milling technique. The NanoCrystal particles of the drug are stabilized against agglomeration. The result is an aqueous dispersion of the drug substance that behaves as a colloidal dispersion. The NanoCrystal technology can be incorporated into both parenteral and oral, including solid, liquid, fast-melt, pulsed release and controlled release drug delivery systems.

3.5 Nanotubes and Nanowires

Nanotubes and nanowires [13] are the self-assembling sheet of atoms arranged in the form of tubes and thread-like structures of nanoscale range. Nanostructures that have gained much attention are hollow, carbon-based cage like structures—nanotubes and fullerenes.

Biophan Technologies, Inc., a developer of next-generation medical technology, filed a 2006 US patent application for the use of halloysite nanotubes for controlled drug release. The technology utilizes halloysite nanotubes which are hollow inorganic nanostructures derived from clay, and allow the linear, controlled release.

3.6 Nanogels

Nanogels [3] are crosslinked particles of sub-micron size prepared from hydrophilic polymers. They are soluble in water, but have properties different from linear macromolecules of similar molecular mass. Such structures, along with their larger analogues - microgels - have a number of practical applications, particularly in medicine (e.g. stomatology) and pharmaceutics .

Micro- and nanogels can be synthesized by combined polymerization and crosslinking, usually as an emulsion. A drawback of this procedure, especially when the products are designed for biomedical use, is the presence of monomers and crosslinking agents that are usually toxic and have to be removed from the system after the synthesis.

3.7 DNA cage

The basic idea is that a protein-sized DNA cage [14] could be used to deliver a drug (e.g. a protein) - the cage could be engineered to facilitate delivery and targeting without the need to modify the cargo, and the cage could be programmed to release the cargo in response to a specific molecular stimulus.

4 EXECUTIVE SUMMARY

Nanotechnology based approaches for enhancing the clinical profile of medicines, still require considerable further development before they can deliver, across a broad range of disease areas, the much heralded promises expected by patients. Notwithstanding recent advances in controlled release of therapeutics in oncology settings, significant scientific and technical barriers remain in the drive to improve targeted delivery of medicines using the tools offered by nanotechnology e.g. metrology standard are lacking, pre-clinical development process is highly regulated, nanomaterial approaches need to be integrated into the early drug discovery work stream, regulatory framework is still developing.

- The focus of nanomaterials is a bottom up approach to structures and functional effects whereby the building blocks of materials are designed and assembled in controlled ways.
- The majority of therapeutic nano-based IP is directed towards delivery systems employing nanomaterials to achieve superior drug absorption, controlled drug release or reduced side-effects.
- Despite already having regulatory accepted nano-based drug carriers in the clinic, at this point no site-specific targeted drug carrier system is on the market.
- A wide range of nanomaterial based delivery platforms are in development at multiple biotech and mid-sized Pharma organizations. However, most activity is based on creating line extension opportunities, or improved formulations for off-patent medicines.
- Platform based on: Nanoparticles, Liposomes, Dendrimers, Nanocrystals, Nanotubes, Nanogels and DNA cages have been reviewed in detail. At this point in time, no one platform appears to have an advantage in terms of key operational characteristics, i.e. PK/PD modulation, Therapeutic Index enhancement, Biocompatibility, Manufacturing Tractability etc.
- Significant government support for nanomaterial technologies is in place. The EU Nanotechnology program http://cordis.europa.eu/nanotechnology/ and US National Nanotechnology Initiative (http://www.nano.gov) provide high level visions for nanotechnology-based investment by identifying potential R&D targets.

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