AN OVERVIEW: NANOMEDICINE AND FUTURE PROSPECTUS

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Summary

Nanomedicine is the process of diagnosing, treating, and preventing disease and traumatic injury, of relieving pain, and of preserving and improving human health, using molecular tools and molecular knowledge of the human body.Medicine is constantly evolving and new technologies are incorporated into the diagnosis and treatment of patients. Regenerative medicine aims to work with the body's own repair mechanisms to prevent and treat disabling chronic diseases such as diabetes, osteoarthritis, and degenerative disorders of the cardiovascular and central nervous system and to help victims of disabling injuries. nanomedicine applications include activity monitors, chemotherapy, pacemakers, biochips, OTC tests, insulin pumps, nebulizers, needleless injectors, hearing aids, medical flow sensors and blood pressure, glucose monitoring and drug delivery systems. Nanomedicine is the medical application of nanotechnology. Nanomedicine ranges from the medical applications of nanomaterials, to nanoelectronic biosensors, and even possible future applications of molecular nanotechnology. Nanoshells possess highly favorable optical and chemical properties it is often used for biomedical imaging, therapeutic applications, fluorescence enhancement of weak molecular emitters, surface enhanced Raman spectroscopy and surface enhanced infrared absorption spectroscopy.

Keywords:- Nanomedicine, Advancement in Nano medicine, Future Aspects of Nanomedicine.

1.1 Introduction

Medicine is constantly evolving and new technologies are incorporated into the diagnosis and treatment of patients. This process is sometimes slow and there can be a gap of years before new technologies are integrated in medical practice. The reasons for the delay are as follows:

- Establishing the safety and efficacy of innovative treatments is a long process, particularly with clinical trials and regulatory reviews.
- The current generation of medical practitioners are still not well oriented toward biotechnology and conservative elements of the profession may be slow in accepting and learning about nanobiotechnology, which is at the cutting edge of biotechnology.
- The high cost of new technologies is a concern for healthcare providers. Cost benefit studies are needed to convince the skeptics that some of the new technologies may actually reduce the overall cost of healthcare.

Molecular medicine is already a recognized term. It should not be considered a subspecialty of medicine as molecular technologies would have an overall impact on the evolution of medicine. Recognition of the usefulness of biotechnology has enabled progress in the concept of personalized medicine, which again is not a branch of medicine but simply indicates a trend in healthcare and simply means the prescription of specific treatments and therapeutics best suited for an individual . Various nanomachines and other nanoobjects that are currently under investigation in medical research and diagnostics will soon find applications in the practice of medicine. Nanobiotechnologies are being used to create and study models of human disease, particularly immune disorders. Introduction of nanobiotechnologies in medicine will not create a separate branch of medicine but simply implies improvement in diagnosis as well as therapy and can be referred to as nanomedicine [1, 3]

1.2 Basics of Nanobiotechnology in Relation to Nanomedicine

Nanotechnology (Greek word nano means dwarf) is the creation and utilization of materials, devices and systems through the control of matter on the nanometer length scale, i.e., at the level of atoms, molecules, and supramolecular structures. Nanotechnology, as defined by the National Nanotechnology Initiative is the understanding and control of matter at dimensions of roughly 1–100 nm, where unique phenomenon enable novel applications.[2]

1.3 NANOMEDICINE: PRINCIPLE

Nanomedicine is an emerging discipline of nanotechnology with a large subject area and includes nanoparticals that act as biological mimetics (e.g. functionalized carbon nanotubes), "nanomachines" (e.g., those made form interchangeable DNA parts and DNA scaffolds such as octahedron and stick cube), nanofibers and polymeric nanoconstructs as biomaterials (e.g., molecular self assembly and nanofibers of peptides and peptide amphiphiles for tissue engineering, shape memory polymers as molecular switches, nonporous membranes), and nanoscale microfabrication based devices (e.g., silicon microchips for drug release and micro machined hollow needles and two dimensional needle arrays from single crystal silicon), sensors and laboratory diagnostics. Nanomedicines have widespread applications. it is used for diagnosing, treating, preventing diseases and traumatic injury, relieving pain, repair, construction and control of human biological system at the molecular level. nanomedicine applications include activity monitors, chemotherapy, pacemakers, biochips, OTC tests, insulin pumps, nebulizers, needlelessinjectors, hearing aids, medical flow sensors and blood pressure, glucose monitoring and drug delivery systems.[38]

Here are a few examples of how nanomedicine could transform common medical procedures:

- Diagnostic nanomachines could be employed to monitor the internal chemistry of the body. Mobile nano robots could circulate in the blood and lymph systems, and send out warnings when chemical imbalances occur .
- Similar fixed nanomachines could be planted in the nervous system to monitor pulse, brain-wave activity, and other functions. [39]
- Implanted nanotechnology devices could dispense drugs or hormones as needed in people with chronic imbalance or deficiency states.
- In heart defibrillators and pacemakers, nanomachines could affect the behavior of individual cells.

The most advanced nanomedicine involves the use of nanorobots as miniature surgeons. Such machines might repair damaged cells, or get inside cells and replace or assist damaged intracellular structures. At the extreme, nanomachines might replicate themselves, or correct genetic deficiencies by altering or replacing DNA (deoxyribonucleic acid) molecules.

The key elements of nanotechnology applied to nanomedicine are:

- The use of analytical tools and devices to bring a better understanding of the molecular basis of disease, patient predisposition and response to therapy, and to allow imaging at the molecular, cellular and patient levels.
- The design of nano-sized multifunctional therapeutics and drug delivery systems to yield more effective therapies.
- The most important medical areas where nanotechnology will have a great impact as identified by the European Science Foundation and the European Union are:
  - Nanodiagnostics including imaging
  - Targeted drug delivery and controlled release
  - Regenerative medicine

a) Nanodiagnostics

The main goal of nanodiagnostics is to identify diseases at a very early stage at the level of a single cell. Nanotechnology can provide tools for better sensitivity, specificity and reliability. It The use of nanoelectronics will improve the sensitivity of sensors based on already established methods. Nanotechnology will improve the microscopic and spectroscopic techniques to achieve ultra-high spatial resolution, molecular resolution and ultra-high sensitivity which will provide a better understanding of the cell's complex mechanisms in basic research. [40]

b) Targeted Drug Delivery and Controlled Release

The drug delivery systems enabled by nanotechnology aims to target selected cells or receptors in the body. Nanoformulations which make use of enlarged surface/volume ratio for enhanced reactivity and nanoparticles that can be used as drug carriers will improve the present targeted delivery systems reducing the costs and increasing the patient acceptance. When a drug is suitably encapsulated, in nanoparticulate form, it can be delivered to the appropriate site, released in a controlled way and protected from undergoing premature degradation. These kinds of controlled release techniques enabled by nanotechnology will have less side effects and high efficiency which can be successfully used for the treatment of cancer and wide range of other diseases. [(41)]

c) Regenerative Medicine

Regenerative medicine aims to work with the body's own repair mechanisms to prevent and treat disabling chronic diseases such as diabetes, osteoarthritis, and degenerative disorders of the cardiovascular and central nervous system and to help victims of disabling injuries. Nanotechnology has established a cellular and molecular basis for the development of innovative disease-modifying therapies for in-situ tissue

regeneration and repair, requiring only minimally invasive surgery. The basic elements of importance in this new 'nanobiomimetic' strategy are intelligent biomaterials, bioactive signalling molecules, and cells.. The sequential signalling triggers the regenerative events at the cellular level which is necessary for the fabrication and repair of cells. Regenerative medicine also aims to effectively exploit the enormous self-repair potential that has been observed in adult stem cells.[14]

1.4 Landmarks in the Evolution of Nanomedicine

Table 1.2 Historical landmarks in the evolution of nanomedicine [4]

Year	Landmark
1985	Discovery of buckyballs (fullerenes) by Robert Curl, Richard Smalley, and Harold Kroto, which led to the award of the 1996 Nobel Prize in Chemistry (Smalley 1985; Curl et al 1997)
1987	Cancer targeting with nanoparticles coated with monoclonal antibodies (Douglas et al 1987)
1988	Maturation of the field of supramolecular chemistry relevant to nanotechnology: construction of artificial molecules that interact with each other leading to award f the Nobel prize (Lehn 1988). Awarded the Nobel Prize
1990	Atoms visualized by the scanning tunneling microscope discovered in the 1980s at the IBM Z¨urich Laboratory (Z¨urich, Switzerland), which led to the award of a Nobel Prize (Eigler and Schweizer 1990).
1991	Discovery of carbon nanotubes (Iijima et al 1992).
1994	Nanoparticle-based drug delivery (Kreuter 1994).
1995	FDA approved Doxil, a liposomal formulation of doxorubicin, as an intravenous chemotherapy agent for Kaposi's sarcoma. Drug carried by nanosize liposomes is less toxic with targeted delivery
1998	First use of nanocrystals as biological labels, which were shown to be superior to existing fluorophores (Bruchez et al 1998)and Use of DNA-gelatin nanospheres for controlled gene delivery
2000	First FDA approval of a product incorporating the NanoCrystal_R technology (Elan, King of Prussia, PA, USA), a solid-dose formulation of the Immuno suppressant sirolimus — Rapamune_R (Wyeth)
2003	The US Senate passed the Nanotechnology Research and Development Act, making the National Nanotechnology Initiative a legal entity, and authorized.

2005 FDA approved AbraxaneTM, a taxane based on nanotechnology, for the treatment of breast cancer. The nanoparticle form of the drug overcomes insolubility problems encountered with paclitaxel and avoids the use of toxic solvents.

2.1. POTENTIAL TARGETES IN THE DEVELOPMENT OF NANOMEDICINE

2.1.1 Macrophage as a target:

The propensity of macrophages of the reticuloendothelial system for rapid recognition and clearance of particulate matter has provided a rational approach to macrophage-specific targeting with nanocarriers. The macrophage is a specialized host defense cell whose contribution to pathogenesis is well known.[42]

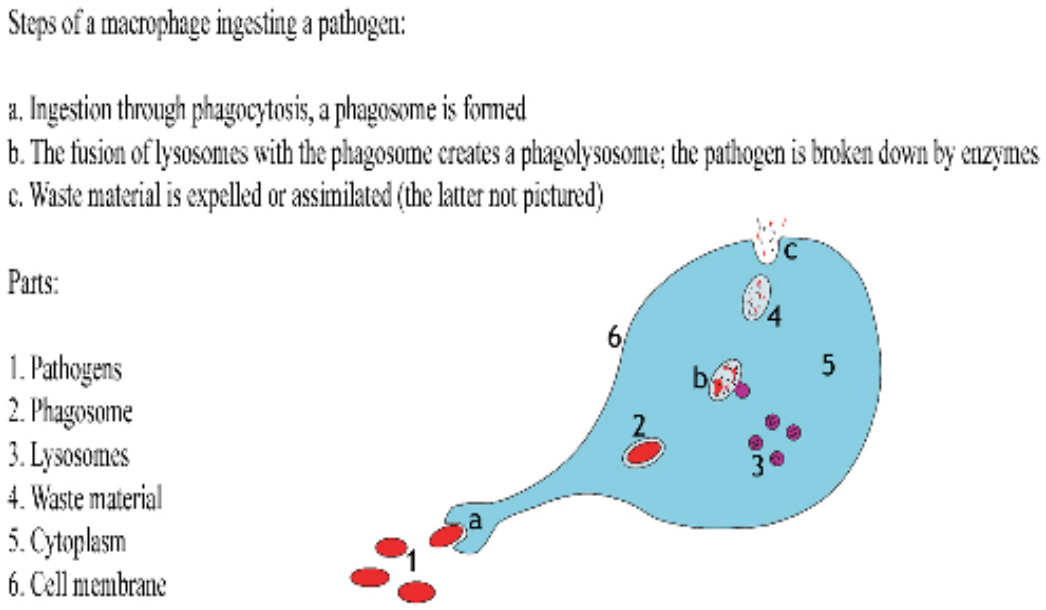


Figure 1: Steps of macrophages in ingesting pathogens[28]

Alterations in macrophage clearance and immune effector functions contribute to common disorders such as atherosclerosis, autoimmunity, and major infections. The macrophage, therefore, is a valid pharmaceutical target and there are numerous opportunities for a focused macrophage-targeted approach [5–8]. For example, although most microorganisms are killed by macrophages, many pathogenic organisms have developed means for resisting macrophage destruction following phagocytosis. In certain cases, the macrophage lysosome and/or cytoplasm is the obligate intracellular home of the microorganism, examples include Toxoplasma gondii, various species of Leishmania, Mycobacterium tuberculosis, and Listeria monocytogenes. Passive

targeting of nanoparticulate vehicles with encapsulated antimicrobial agents to infected macrophages is therefore a logical strategy for effective microbial killing [5, 6, 9–11]. The endocytic pathway will direct the carrier to lysosomes where pathogens are resident. Degradation of the carrier by lysosomal enzymes releases drug into the phagosome-lysosome vesicle itself or into the cytoplasm either by diffusion or by specific transporters, depending on the physicochemical nature of the drug molecule. Approved formulations for human subjects are limited to lipid-based nanosytems (100–200 nm) with entrapped amophotericin B (Amp-B), and are recommended for treatment of visceral eishmaniasis or confirmed infections caused by specific fungal species [5, 9, 11]. This mode of targeting has significantly reduced the required clinically effective quantity of Amp-B for treatment, achieving therapeutic drug concentrations in the infected macrophages. Other beneficial effects include significant reduction in nephrotoxicity, a common side effect associated with Amp-B administration, and pro inflammatory cytokine release [12, 13].

2.1.2 Endothelium as a target

The concept of targeting to the blood vessels is an attractive one, particularly with the view that the endothelium plays an important role in a number of pathological processes including cancer (dysregulated angiogenesis), inflammation, oxidative stress and thrombosis. Indeed, a number of studies have demonstrated a level of control of arrest and distribution of passively targeted nanoparticles by specific endothelial cells, and these were linked to the surface properties of the carrier. Recent studies have shown that cationic liposomes within 1 h of entering the circulation, are internalized into endosomes and lysosomes of endothelial cells in a characteristic organ- and vessel-specific manner [14.] These patterns seem to bear no relationship to the morphological characteristics of the endothelium associated with a particular site, but probably reflect vessel-specific expression of receptors for which such particles, or their surface-associated blood proteins, are ligands. But recent dramatic progress in the development of a human vascular map, in particular through the application of cell and molecular biological tools such as serial analysis of gene expression (SAGE), subtractive proteomic mapping, and in vivo phage display, is generating yet another level of possibilities for specific targeting of drugs and biological agents [15–18]. For example, SAGE examines the spectrum of mRNA species within a cell or tissue and allows rapid comparison with other cell types or tissues. Phage, however, behaves as a nanomachine; it can be engineered to display numerous peptides on its surface and

after injection one can select peptides that make the phage home to a given target . The authors surveyed 47,160 sequences that localized to different organs within the patient, and determined that in many cases these sequences were similar to those of known ligands for endothelial cell-surface proteins, thus validating his technique for identifying novel endothelial targets in humans. This approach is now being used in a number of ways to target therapeutic agents, particularly to the vasculature of solid tumors. Examples include integrins alpha v beta 3, alpha v beta 5 and alpha 5 beta 1, which are up-regulated in angiogenic endothelial cells and play a role in the process of angiogenesis [15, 18, 19, 20]. They bind with high affinity to sequences containing a characteristic RGD (Arg-Gly-Asp) motif, which seems to be central to anti-integrin approaches. Indeed, in vivo phage display studies by Assa-Munt et al. have led to the development of a cyclic nonapeptide RGD-4C, which avidly binds to the integrins alpha v beta 3 and alpha v beta 5. Coupling of RGD-4C to doxorubicin yielded a compound significantly more effective than doxorubicin alone, and with less side effects to the heart and liver, the main sites of doxorubicin toxicity.[43]

2.1.3 Extravasation: targeting of solid cancers

The development of "stealth" technologies has provided opportunities for passive accumulation of intravenously injected nanoparticles (20–150 nm) in pathological sites expressing "leaky" vasculature by extravasation [6]. In spite of these limitations, there are regulatory approved formulations of long circulating liposomes with entrapped doxorubicin for management/treatment of AIDS-related Kaposi's sarcoma, refractory ovarian cancer, and metastatic breast cancer [21]. These formulations exhibit favorable pharmacokinetics when compared with the free drug, for example the area under the curve after a dose of 50 mg/m2 doxorubicin encapsulated in stealth liposomes is 300-fold greater than that of free doxorubicin. As a result of these promising pharmacokinetic profiles, stealth doxorubicin containing liposomes are currently undergoing additional early- to late-phase clinical trials.(56) A number of engineering issues must be considered for cancer drug delivery. First, the carrier must have a high drug loading capacity and remain stable within the vasculature with minimum drug loss.

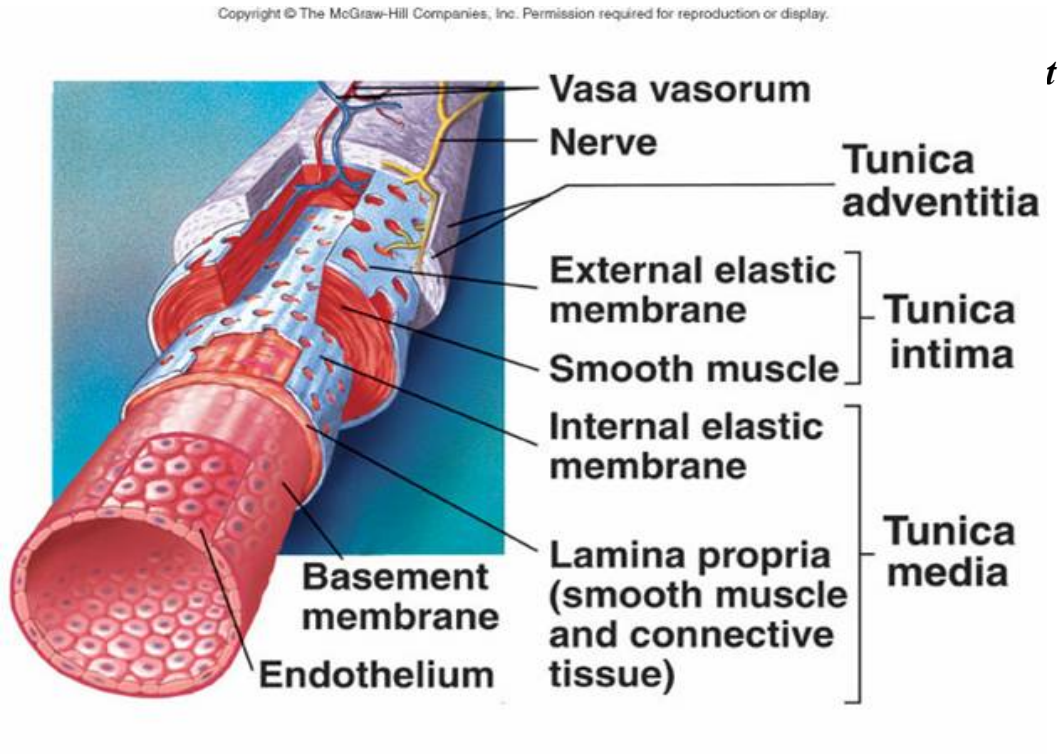


Figure 2: ***Location of Endothelium***[29]

Here, doxorubicin is loaded actively by an ammonium sulfate gradient (as doxorubicin sulfate) yielding highly stable liposomes with high contents of doxorubicin aggregates [22]. Second, it has been widely established that the majority of extravasated particulate systems, such as liposomes, do not interact with target cancer cells [23]. They are often distributed heterogeneously in perivascular clusters that do not move significantly. The process of particle extravasation must be followed by the efflux of drug from the carrier, resulting in target exposure (being tumor cells, tumor-associated macrophages, components of tumor vasculature, or extracellular mediators such as proangiogenic proteases) to drug molecules. Here, the drug must be released at a rate that maintains free drug levels in the therapeutic range.[44]

2.1.4 Nanoparticles for cytoplasmic drug delivery

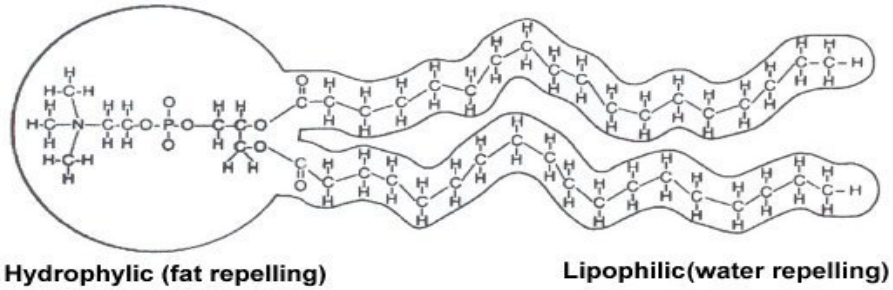
The endosomal membrane is particularly important for priming MHC class I-restricted cytotoxic T lymphocyte responses, for survival of genetic materials against nuclease degradation in the lysosomal compartment, or for those drugs that must reach cytoplasm in sufficient quantities (as for treatment of cytoplasmic infections or reaching nuclear receptors) after endocytic delivery with nanoparticulate carriers. Here, there are advances in particle engineering too. For instance, nanoparticles made from poly(DL-lactide-co-glycolide) can escape the endo-lysosomal compartment within minutes of internalization in intact form and reach the cytoplasm [23]. The mechanism of rapid escape is by selective reversal of the surface charge of

nanoparticles from the anionic to the cationic state in endo-lysosomes, thus resulting in a local particle-membrane interaction with subsequent cytoplasmic release. Another impressive approach for cytoplasmic delivery of nanoparticles is their surface manipulation with short peptides known as protein transduction domains such as HIV-1 TAT protein transduction domain (TAT PTD), which is a short basic region comprising residues ,or heterologous recombinant TAT-fusion peptides.The electrostatic interaction between the cationic TAT PTD and negatively charged cellsurface constituents, such as heparan sulfate proteoglycans and glycoproteins containing sialic acids, is a necessary event before internalization . [57]After this ionic interaction, cellular uptake occurs by lipid raftdependent macropinocytosis in a receptor-independent manner; this is followed by a pH drop and destabilization of integrity of the macropinosome vesicle lipid bilayer, which ultimately results in the release of TAT-cargo into the cytosol . This mode of entry may further suggest the avidity of TAT PTD for glycophosphoinositol- anchored glycoproteins, which are present in lipid rafts, or binding to cholesterol membrane constituents that trigger macropinocytosis. Indeed, an influenza vaccine based on the latter principle is currently available for human use . This vaccine is administered parenterally and is well tolerated in children, young adults and the elderly. The influenza strains chosen are dependent on the yearly recommendations of the World Health Organization. The delivery system is comprised of unilamellar vesicles of 150 nm in diameter, but intercalated into their lipid bilayer are viral components, which include neuraminidase and hemagglutinnin (HA) glycoprotein. The mode of action of these virus-like liposomes (virosomes) is dependent on HA glycoprotein, the major antigens of the influenza virus. The HA is composed of two subunits, HA1 and HA2. The first subunit has high affinity for sialic acid present on the surface of antigen presenting cells thus facilitating virosome binding. The HA2 subunit is a fusion peptide and is activated at low pH (5.0). Hence, in late endosomes, where pH is acidic, the virosome becomes fusion-competent; this process releases entrapped antigens into the cytosol for subsequent processing and presentation . [45]

2.2 TYPES OF NANOMATERIALS USED IN NANOMEDICINE

2.2.1 LIPOSOME

A liposome is a tiny bubble (vesicle), made out of the same material as a cell membrane. Liposomes can be filled with drugs, and used to deliver drugs for cancer and other diseases. The membranes are usually made of phospholipids, which are



Shape of phospholipid molecule

Figure 3: Shape of phospholipid molecule[29]

molecules that have a head group and a tail group. The head is attracted to water, and the tail, which is made of a long hydrocarbon chain, is repelled by water. Phospholipids are found in stable membranes composed of two layers (a bilayer). In the presence of water, the heads are attracted to water and line up to form a surface facing the water. The tails are repelled by water, and line up to form a surface away from the water. In a cell, one layer of heads faces outside of the cell, attracted to the water in the environment. Another layer of heads faces inside the cell, attracted by the water inside the cell. The hydrocarbon tails of one layer face the hydrocarbon tails of the other layer, and the combined structure forms a bilayer.[24]

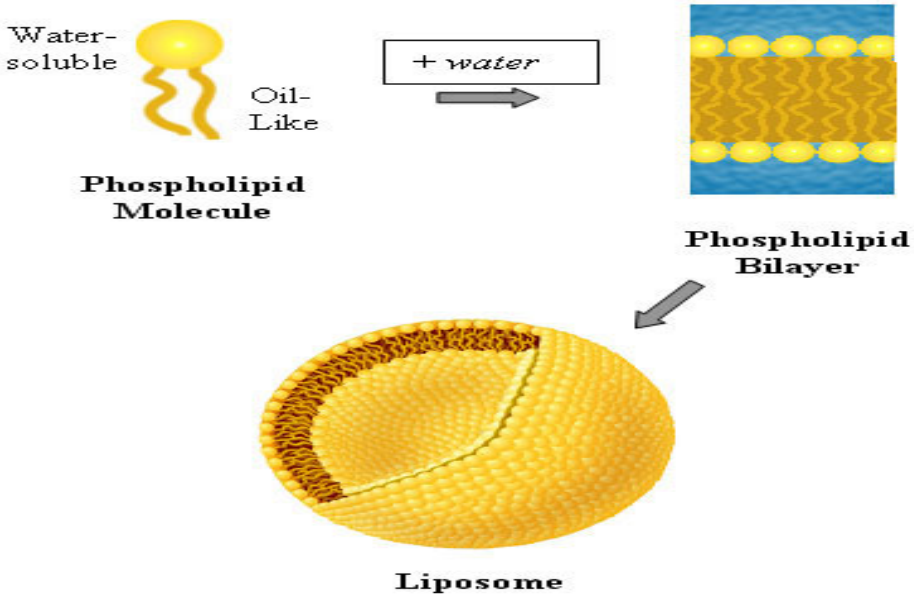


Figure 4: Liposome[30]

When membrane phospholipids are disrupted, they can reassemble themselves into tiny spheres, smaller than a normal cell, either as bilayers or monolayers. The bilayer structures are liposomes. The monolayer structures are called micelles. The lipids in the plasma membrane are chiefly phospholipids like phosphatidylethanolamine and phosphatidylcholine. Phospholipids are amphiphilic with the hydrocarbon tail of the molecule being hydrophobic; its polar head hydrophilic. As the plasma membrane faces watery olutions on both sides, its phospholipids accommodate this by forming a phospholipid bilayer with the hydrophobic tails facing each other. Liposomes can be composed of naturally-derived phospholipids with mixed lipid chains (like egg phosphatidylethanolamine),or of pure surfactant components like DOPE (Di-oleoyl Phosphatidyl Ethanolamine).

Application of liposome

- Liposomes are used for drug delivery due to their unique properties. A liposome encapsulates a region on aqueous solution inside a hydrophobic membrane; dissolved hydrophilic solutes cannot readily pass through the lipids.
- Hydrophobic chemicals can be dissolved into the membrane, and in his way liposome can carry both hydrophobic molecules and hydrophilic molecules to deliver the molecules to sites of action, the lipid bilayer can fuse with other bilayers such as the cell membrane, thus delivering the liposome contents. By making liposomes in a solution of DNA or drugs (which would normally be unable to diffuse through the membrane) they can be (indiscriminately) delivered past the lipid bilayer.[55]
- Liposomes are used as models for artificial cells. Liposomes can also be designed to deliver drugs in other ways. Liposomes that contain low (or high) pH can be constructed such that dissolved aqueous drugs will be charged in solution. As the pH naturally neutralizes within the liposome (protons can pass through some membranes), the drug will also be neutralized, allowing it to freely pass through a membrane. These liposomes work to deliver drug by diffusion rather than by direct cell fusion. .
- The use of liposomes for transformation or transfection of DNA into a host cell is known as lipofection.

2.2.2 Dendrimers

Dendrimers (dendri means tree, mer means branch) are a novel class of 3-D nano scale, core–shell structures that can be precisely synthesized for a wide range of applications. Specialized chemistry techniques allow for precise control over the physical and hemical properties of the dendrimers. They are constructed generation by generation in a series of controlled steps that increase the number of small branching molecules around a central core molecule. Up to 10 generations can be incorporated into a single dendrimer molecule. The core, branching, and surface molecules are chosen to give desired properties and functions.[54]

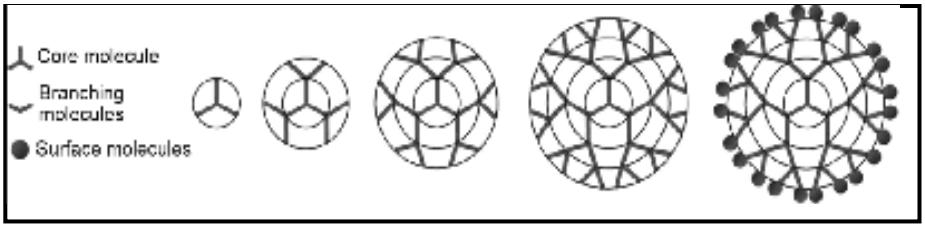


Figure 5: The core, branching, and surface molecules of Dendrimers [31]

As a result of their unique architecture and construction, dendrimers possess inherently valuable physical, chemical, and biological properties. These are as follows:

- Precise architecture, size, and shape control—Dendrimers branch out in a highly predictable fashion to form amplified 3D structures with highly ordered architectures.
- High uniformity and purity—The proprietary stepwise synthetic process used produces dendrimers with highly uniform sizes (monodispersity) possessing precisely defined surface functionality and very low impurity levels.
- High loading capacity—Internal cavities intrinsic to dendrimer structures can be used to carry and store a wide range of metals, organic, or inorganic molecules
- Low toxicity—Most dendrimer systems display very low cytotoxicity levels.
- Low immunogenicity when injected or used topically.

a)Properties of Dendrimers

The surface properties of dendrimers may be manipulated by the use of appropriate "capping" reagents on the outermost generation. In this way dendrimers can be readily decorated to yield a novel range of functional properties. These are as follows:

- Polyvalency—The outer shell of each dendrimer can be manipulated to contain a large number of reactive groups. Each of these reactive sites has the potential to interact with a target entity, often resulting in polyvalent interactions.
- Flexible charge and solubility properties—Through use of appropriate capping groups on the dendrimer exterior, the charge and solubility of dendrimers can be readily manipulated.[53]
- Flexible binding properties—By using appropriate capping groups on the dendrimer exterior, dendrimers can be designed to exhibit strong affinity for specific targets.

b) Applications

Potential applications of dendrimers in nanomedicine are as follows:

i. Diagnostics
    1. Sensors
    2. Imaging contrast agents

ii. Drug delivery
    1. Improved delivery of existing drugs
    2. Improved solubility of existing drugs

iii. Drug development
    1. Polyvalent dendrimers interacting simultaneously with multiple drug targets
    2. Development of new pharmaceuticals with novel activities
    3. Improving pharmacological activity of existing drugs
    4. Improving bioavailability of existing drugs

iv. Medicine and surgery
    1. Prevention of scar tissue formation after surgery

2.2.3 VIROSOMES

A Virosome is a unilamellar phospholipid bilayer vesicle with a mean diameter of 150 nm. Essentially, virosomes represent reconstituted empty influenza virus envelopes, devoid of the nucleocapsid including the genetic material of the source virus.[26] Virosomes are not able to replicate but are pure fusion-active vesicles. In contrast to liposomes, virosomes contain functional viral envelope glycoproteins: influenza

virus hemagglutinin(HA) and neuraminidase (NA) intercalated in the phospholipid bilayer membrane.[52]

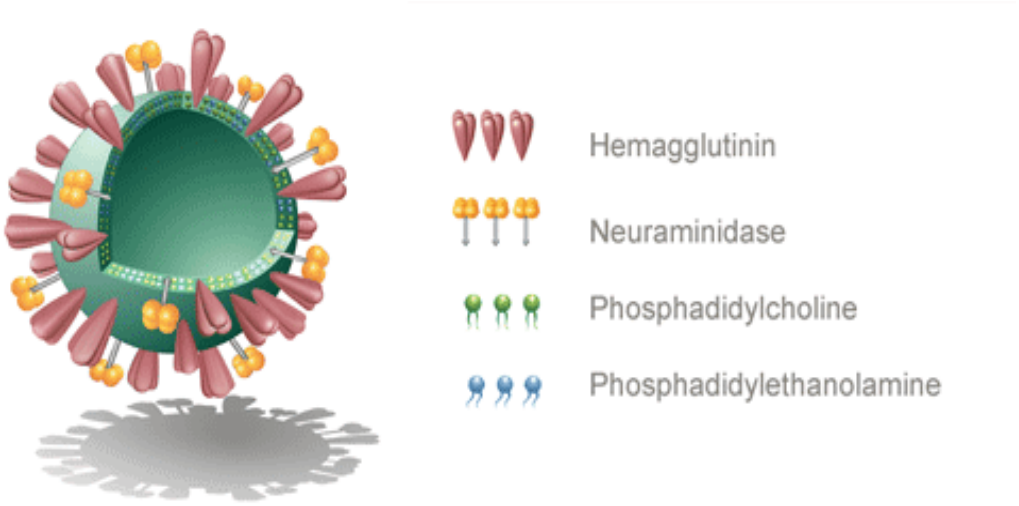


Figure 6: Structure of Virosomes[32]

The unique properties of virosomes partially relate to the presence of biologically active influenza HA in their membrane. It has been shown that a physical association between the virosome and the antigen of interest is necessary for the full adjuvant effect of virosomes . Such physical association can be achieved by a variety of methods, depending on the properties of the antigen. Antigens can be incorporated into virosomes, adsorbed to the virosome surface, or integrated into the lipid membrane, either via hydrophobic domains or lipid moieties cross-linked to the antigen. Virosomes therefore represent an innovative, broadly applicable adjuvant and carrier system with prospective applications in areas beyond conventional vaccines.

Antigens can be incorporated into virosomes, adsorbed to the virosome surface, or integrated into the lipid membrane, either via hydrophobic domains or lipid moieties cross-linked to the antigen

Application of Virosomes

1. Virosome technology provides a broadly applicable delivery system for antigens or DNA/RNA encoding specific immune stimulatory proteins.
2. Virosome technology enables target-specific delivery of antigens and amplification of the immune response.
3. Virosomes stimulate both arms of the immune system – eliciting antibody and cellular immune responses - against inserted immune stimulatory proteins derived from human pathogens.

4. Virosomes are completely biodegradable and can exert an immune response via different routes of administration.[51]

2.2.4 AQUASOMES

Aquasomes are the nanobiopharmaceutical carrier system contains the particle core composed of nanocrystalline calcium phosphate or ceramic diamond, and is covered by a polyhydroxyl oligomeric film. Aquasomes are spherical 60–300 nm particles used for drug and antigen delivery Three types of core materials are mainly used for producing aquasomes: tin oxide, nanocrystalline carbon ceramics (diamonds) and brushite (calcium phosphate dihydrate). The brushite is unstable and converts to hydroxyapatite upon prolong storage. Hydroxyapatite, a better core for the preparation of aquasomes. [50]

APPLICATIONS OF AQUASOMES

1. Aquasomes as red blood cell substitutes, haemoglobin immobilized on oligomer surface because release of oxygen by haemoglobin is conformationally sensitive. By this toxicity is reduced, haemoglobin concentration of 80% achieved and reported to deliver blood in non linear manner like natural blood cells.
2. Aquasomes used as vaccines for delivery of viral antigen i.e. Epstein-Barr and Immune deficiency virus to evoke correct antibody.
3. Aquasomes have been used for successful targeted intracellular gene therapy, a five layered composition comprised of ceramic core, polyoxyoligomeric film, therapeutic gene segment, additional carbohydrate film and a targeting layer of conformationally conserved viral membrane protein.
4. Aquasomes for pharmaceuticals delivery i.e. insulin, developed because drug activity is conformationally specific.Bioactivity preserved and activity increased to 60% as compared to i.v. administration and toxicity not reported .
5. Aquasomes also used for delivery of enzymes like DNAase and pigments/dyes because enzymes activity fluctuates with molecular conformation and cosmetic properties of pigments are sensitive to molecular conformation.[49]

2.2.5 Quantum Dots :

QDs are nanoscale crystals of semiconductor material that glow, or fluoresce when excited by a light source such as a laser. QD nanocrystals of cadmium selenide 200–10,000 atoms wide, coated with zinc sulfide. Quantum dots are tiny crystals that glow when stimulated by UV light. The wavelength or colour of light depends on size of

crystals. Latex beads filled with these nanoscale semiconductor dots can be designed to level specific DNA sequence by combining different sized quantum dots within a single bead, we can create probes that release distincts colours and intensity of the light. When the crystals are stimulated by UV light, each beam emits lights that serves as spectral barcode identifying a particular region of DNA. The size of the QD determines the frequency of light emitted when irradiated with low energy light Eviflours are quantum dots which bind to antibodies and proteins and are used in research assays.[44]

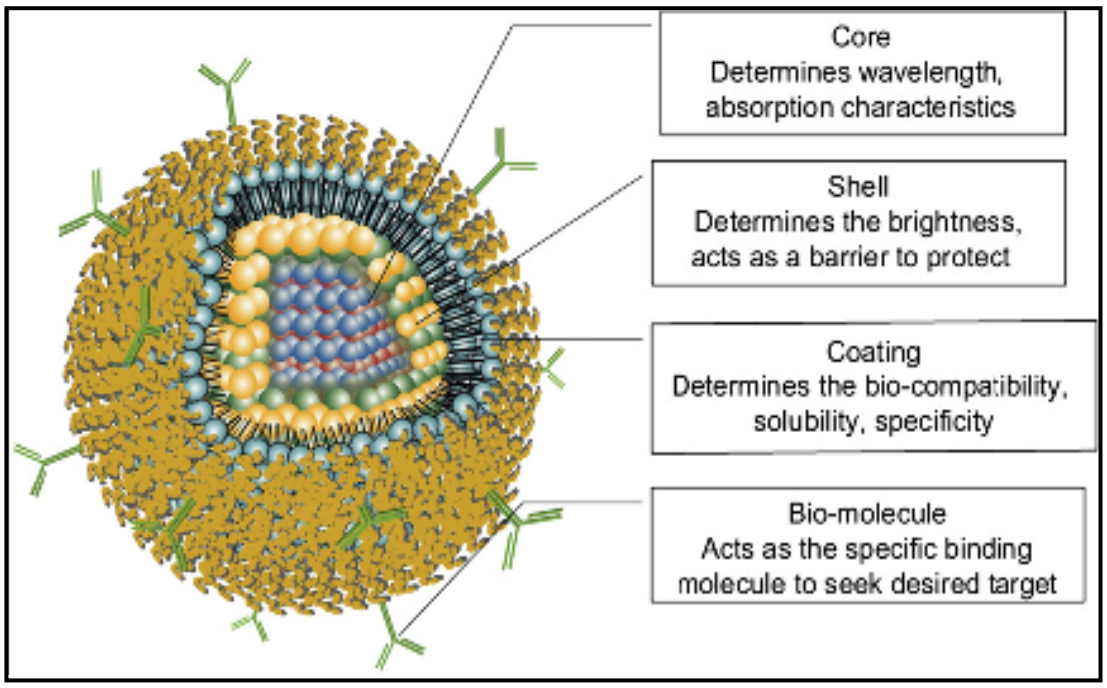


Figure 7:*Structure and applications of Q-dots.*[33]

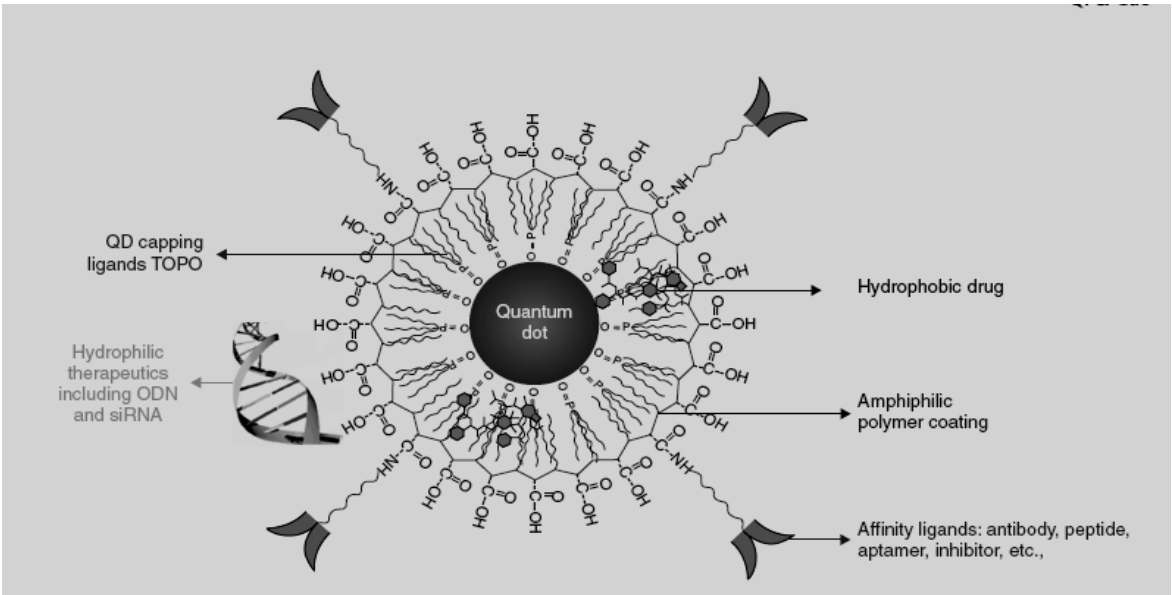


Figure 8: *Multifunctional Quantum Dot Coated With Amphiphilic Polymer*[34]

Applications of QDs

- Fluorescence detection—microscopy, biosensors, multicolor flow cytometry
- Molecular diagnostics, Ex vivo live cell imaging
- In vivo targeting of cells, tissues, and tumors with monitoring by PET and MRI
- High-throughput screening and also used as a multifunctional quantum dot coated

2.2.6 Carbon Nanotubes

Carbon nanotubes are rolled-up sheets of carbon . These nanotubes can go down to 1 nm in diameter, are stronger than any material in the universe, and can be of any length. These can be used as probes for AFMs(atomic force microscopy) that can image individual molecules in both wet and dry environments. The bonding in carbon nanotubes is sp^2, with each atom joined to three neighbours, as in graphite. The tubes can therefore be considered as rolled-up graphene sheets (graphene is an individual graphite layer). [43]

Types of Carbon Nanotubes

Carbon nanotubes are cylindrical sheets of carbon atoms with diameters of about 1 nanometer. Carbon nanotubes can be thought of as a rolled up sheet of graphite. Depending on how the sheet is rolled into a tube, different nanotube structures are produced. The image to the right shows several types of nanotubes, each with a different atomic structure. The structures can be clearly distinguished by looking at the cross section or along the axis of the nanotube the cathode. It is the deposit on the cathode which contains the carbon nanotubes. Single-walled nanotubes are produced when Co and Ni or some other metal is added to the anode. carbon nanotubes can also be made by passing a carbon-containing gas, such as a hydrocarbon, over a catalyst. The catalyst consists of nano-sized particles of metal, usually Fe, Co or Ni.

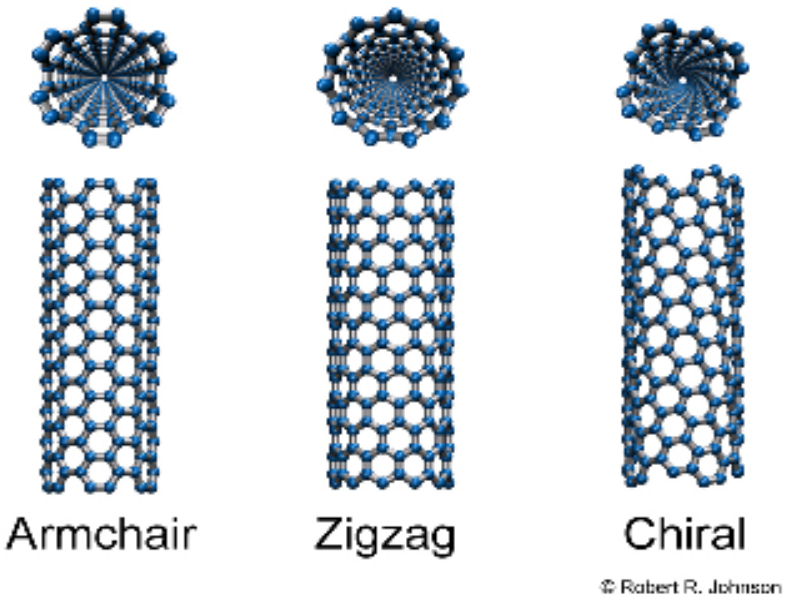


Figure 9: ***Types of Carbon Nanotubes***[35]

These particles catalyse the breakdown of the gaseous molecules into carbon, and a tube then begins to grow with a metal particle at the tip . The third important method for making carbon nanotubes involves using a powerful laser to vaporise a metal-graphite target. This can be used to produce single-walled tubes with high yield.

Medical Applications of Nanotubes:

- Cyclic peptide nanotubes can act as a new type of antibiotic against bacterial pathogens.
- Nanoscale electromechanical systems (nanotweezers) based on carbon nanotubes have been developed for manipulation and interrogation of nanostructures within a cell.
- Carbon nanotubes can be used as tips for AFM.
- Nanotubes can be used in biosensors.
- Blood-compatible carbon nanotubes, with heparin immobilized on the surface, are building blocks for in vivo nanodevices. Activated partial thromboplastin time and thromboelastography studies prove that heparinization can significantly enhance the blood compatibility of nanomaterials

2.2.7 Fullerenes

A molecule of 60 carbon atoms that form a hollow sphere 1 nm in diameter. The molecule was named buckyball or fullerene. fullerenes represent a family of related

structures containing 20, 40, 60, 70, or 84 carbons. C60, however, is the most abundant member of this family.

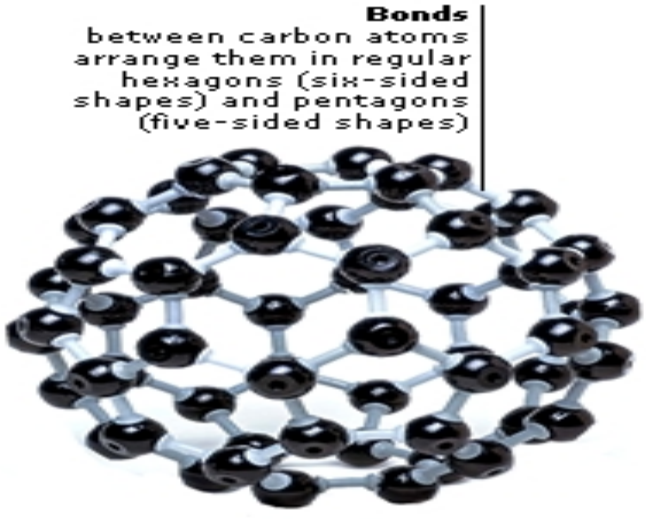


Figure 10:*Fullerenes*[35]

Fullerenes are entirely insoluble in water, but suitable functionalization makes the molecules soluble . Upon contact with water, under a variety of conditions, C60 spontaneously forms a stable aggregate with nanoscale dimensions (25–500 nm), termed nano-C60 that are both soluble and toxic to bacteria .

2.2.8 NANOSHELLS

Nanoshells are ball-shaped, about the size of a virus or 1/20th of an RBC, and consist of a core of nonconducting glass that is covered by a metallic shell, typically either gold or silver. These nanoshells involve a quasiparticle called plasmon which is a collective excitation or quantum plasma oscillation where the electrons simultaneously oscillate with respect to all the ions. The simultaneous oscillation can be called Plasmon hybridization where the tenability of the oscillation is associated with mixture of the inner and outer shell where they hybridize to give a lower energy or higher energy. This lower energy couples strongly to incident light whereas, the higher energy is an anti-bonding and weakly combines to incident light. The hybridization interaction is stronger for thinner shell layers, hence, the thickness of the shell and overall particle radius determines which wavelength of light it couples with.[27]

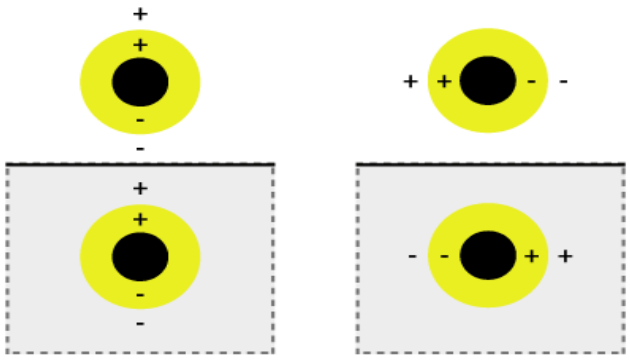


Figure 11: *Nanoshells*[36]

The interaction of light and nanoparticles affects the placements of charges which affects the coupling strength. Incident light polarized parallel to the substrate gives a s-polarization , hence the charges are further from the substrate surface which gives a stronger interaction between the shell and core. Otherwise, a p-polarization is formed which gives a more strongly shifted Plasmon energy causing a weaker interaction and coupling.

Nanoshells possess highly favorable optical and chemical properties for biomedical imaging and therapeutic applications. These particles are also effective substrates for surface-enhanced Raman scattering (SERS) and are easily conjugated to antibodies and other biomolecules . the same conjugation protocols used to bind biomolecules to gold colloid are easily modified for nanoshells. The core/shell ratio and overall size of a gold nanoshell influence its scattering and absorption properties. Gold Nanoshells (Spectra Biosciences, Columbia, MD, USA) possess physical properties similar to gold colloid, in particular a strong optical absorption due to the collective electronic response of the metal to light. The optical absorption of gold colloid yields a brilliant red color . It is a type of spherical nanoparticle consisting of a dielectric core which is covered by a thin metallic shell (usually gold).

Applications of Nanoshells :

Nanoshells possess highly favorable optical and chemical properties it is often used for biomedical imaging, therapeutic applications, fluorescence enhancement of weak molecular emitters, surface enhanced Raman spectroscopy and surface enhanced infrared absorption spectroscopy.

2.2.9 Polymeric Micelles

Polymeric micelles are essentially based on amphiphilic block coplymers such as the pluronics polyoxyethylene polyoxypropylene block co-polymers self assemble into

polymeric micelles. Hydrophobic drugs may be solubilized within the core of micelles or, alternatively, conjugated to the micelles forming polymers. Although micelles are rather dynamic systems, which continuously exchanged units between the micell structure and free units in solution, polyoxyethylene polyaspartic acid micelles are sufficiently stable in the blood and effectively alter the pharmacokinetics of the solubilized drugs. They thus circulate for prolonged periods and are capable of selectively delivering more drugs to tumour tissue as compared to administration of the drugs in solution.

2.2.10 Gold Nanoparticles

DNA molecules are attached to gold nanoparticles, which tangle with other specially designed pieces of DNA into clumps that appear blue. The presence of lead causes the connecting DNA to fall apart. That cuts loose the individual gold nanoparticles and changes the color from blue to red. Gold nanoparticles are also used as a connecting point to build biosensors for detection of disease. A common technique for a diagnostic test consists of an antibody attached to a Fluorescent molecule. When the antibody attaches to a protein associated with the disease, the fluorescent molecule lights up under UV light.

2.2.11 Paramagnetic and Super paramagnetic Nanoparticles

Paramagnetic particles are important tools for cell sorting, protein separation, and single-molecule measurements. The particles used in these applications must meet the following requirements:

- uniform in size,
- highly paramagnetic,
- stable in physiological
- salt buffer,
- functionizable, and 100–1,000nm in size.

They have been used for the detection of model pathogens. Paramagnetic Nano-particles, which are linked to antibodies, enable highly specific biological cell separations. Super paramagnetic iron oxide nanoparticles : (SPIONs) with appropriate surface chemistry have been widely used experimentally in vivo applications such as MRI contrast enhancement, tissue repair, immunoassay, detoxification of biological fluids, hyperthermia, drug delivery, and in cell separation. These applications require that these nanoparticles have high magnetization values

and size smaller than 100 nm with overall narrow particle size distribution, so that the particles have uniform physical and chemical properties. In addition, these applications need special surface coating of the magnetic particles, which not only has to be nontoxic and biocompatible but also has to allow a targetable delivery with particle localization in a specific area. Nature of surface coatings of the nanoparticles not only determines the overall size of the colloid but also plays a significant role in biokinetics and biodistribution of nanoparticles in the body .Magnetic nanoparticles can bind to drugs, proteins, enzymes, antibodies, or nucleotides and can be directed to an organ, tissue, or tumor using an external magnetic field

3.1 NANOMEDICINE TAXONOMY: APPLICATIONS

Nanomedicine research is being funded by government sources, such as the National Institutes of Health (NIH), and by companies in various sectors, including pharmaceutical, biotechnology and medical devices. The nanomedicine taxonomy classifies some of the leading areas that nanotechnology tools, materials, devices, and intelligent materials and machines are currently applied in medical research.

APPLICATIONS

- Biopharmaceutics

Drug Delivery
Drug Encapsulation
Functional Drug Carriers

- Implantable Materials

Tissue Repair and Replacement
Implant Coatings
Tissue Regeneration Scaffolds
Structural Implant Materials
Bone Repair
Bioresorbable Materials

- Surgical Aids

Operating Tools
Smart Instruments
Surgical Robots

- Implantable Devices

Assessment and Treatment Devices,
Implantable Medical Devices
Sensory Aids
Retina Implants
Cochlear Implants

- Diagnostic Tools

Genetic Testing
Ultra-sensitive Labeling and
Detection Technologies
High Throughput Arrays and
Multiple Analyses Imaging

- Role of Nanomedicines in Cance

Nanomedicine is unique among healthcare practices for a number of reasons. Its molecular tools will be able to be manufactured in pollution-free desktop "nanofactories" making nanomedicine highly affordable.

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