# DRUG CARRIERS

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## ABSTRACT

In recent years, there has been an exponential interest in the development of novel drug delivery systems using drug carriers. Drug carriers offer significant advantages over the conventional drug delivery systems in terms of high stability, high specificity, high drug loading capacity, controlled release of drug and ability to deliver both hydrophilic and hydrophobic drugs. As a result of their unique behaviors, drug carriers have a wide range of biomedical and industrial applications. Nanospheres are associated with a lot of benefits such as ease of administration to target sites, reduction in toxicity level and ease of passage via the capillary vessels. Hydrogel nanoparticles are useful in the treatment of inflammatory diseases, as bioresponsive hydrogels in drug delivery system and as a carrier in controlled drug delivery system. Carbon nanotubes have a large surface area which has the ability to adsorb or conjugate with a wide variety of therapeutic and diagnostic agents. They are useful in the areas of gene delivery, tissue regeneration and biosensor diagnosis. Liposomes are known to target a drug to a specific site. They entrap drugs which are released for subsequent absorption. They are used to achieve active targeting, increase efficacy and therapeutic index of drugs. Niosomes improve the solubility and oral bioavailability of poorly soluble drugs. They protect drugs from biological environment, increase the stability of entrapped drugs and they can easily reach the site of action. Aquasomes are nanoparticulate carriers that can be characterized for structural analysis. They preserve conformational integrity and biochemical stability of drugs. Ethosomes are noninvasive delivery carriers that enable drugs to reach the deep skin layers and the systemic circulation. They contain phospholipids which could be in form of phosphatidyl choline (PC), hydrogenated PC, phosphatidic acid (PA), Phosphatidyl serine (PS) and phosphatidyl inositol (PI). Ethosomes are known to increase skin permeation of drugs, improve biological activity and pharmacodynamics profile of drugs. This review aims to emphasize the importance of drug carriers in drug delivery system, and applications of drug carriers in various areas of research, technology and treatment.

**Keywords**: Drug carriers, Niosomes, Nanoparticles, Nanocapsules, Nanotubes, Hydrogels.

## INTRODUCTION

A drug carrier is defined as a substrate which is used in the process of drug delivery that serves to improve the effectiveness and safety of drug administration (Sakthivel *et al*., 2003). The controlled drug release can be regulated either by slow release of the drug over a long period of time or by triggered

release by some stimulus such as pH, heat and light (Sakthivel *et al.*, 2003). The main advantage associated with the use of drug carriers is their ability to protect drugs during the administration time, enclosing them in external protective barriers which will reduce losses of active substances and side effects in patients (Park *et al*, 2002). There are three major factors to consider during the design of any drug delivery system. They include: site specificity, longevity and external stimuli sensitivity (Henglein *et al.*, 1999). Site specificity has to do with the capability of the drug carrier to recognize the target tissue where the drug acts (Qiu *et al*., 1999), while longevity is the capability of reaching the desired tissue without being phagocytized by the phagocytic system before reaching the target site of delivery (Wuister *et al.*, 2004). Drug carriers offer advantages over the known conventional drug delivery systems in terms of high stability, high specificity, high drug loading capacity, controlled release of drugs and ability to deliver both hydrophilic and hydrophobic drugs (Wuister *et al*., 2004).

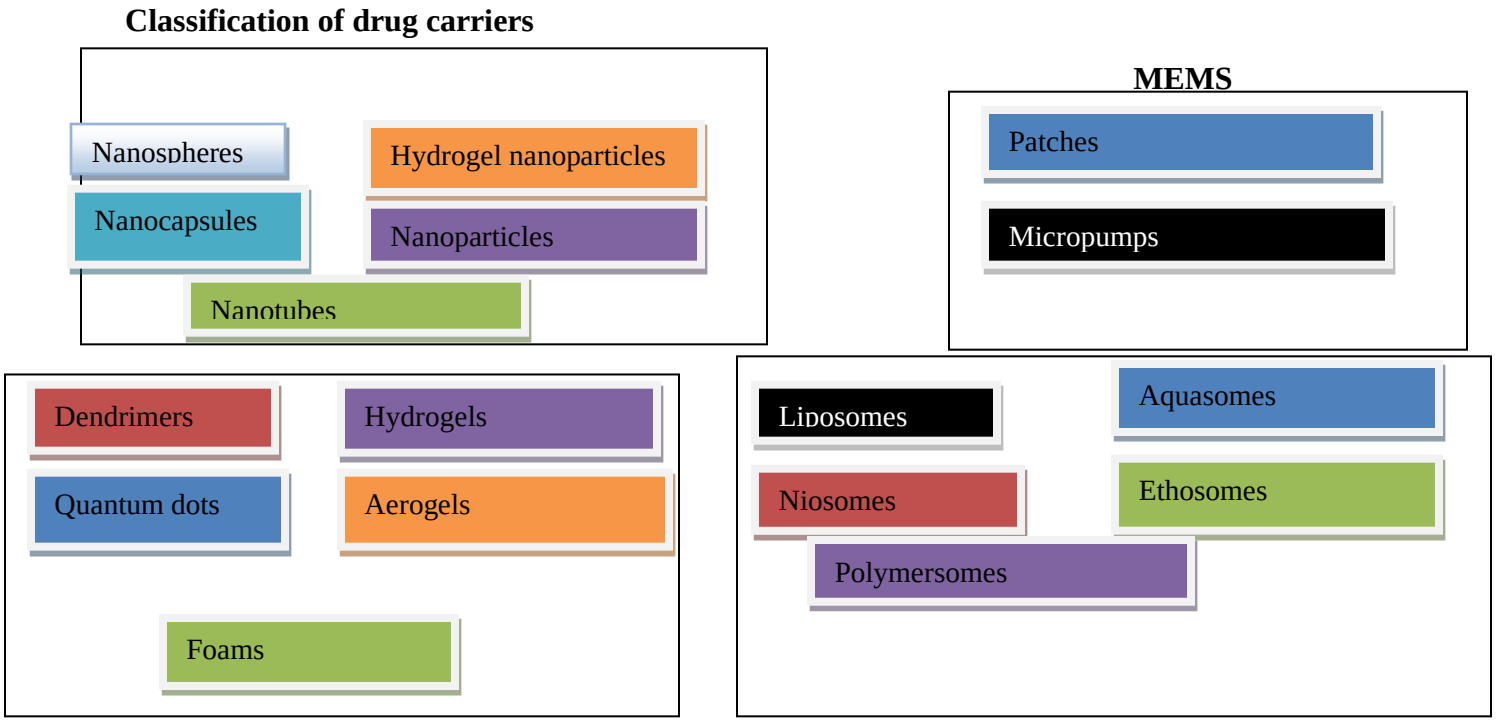


Figure 1: A representation of the set and sub-sets of artificial Drug Delivery Systems (DDS) among nanosystems, micro-electromechanical systems (MEMS), other specific systems and their derivatives.

Nanoparticles are defined as particulate objects that exist within the range of 1-100 nm. Due to the nature of their sizes, they are considered as sub-sets of colloidal particles (Kreyling *et al.*, 2006). A nanoparticle is known to have multiple layers which can be split apart (Lee, 2005). They consist of surface layer, shell and the core material. Most of the major properties associated with nanoparticles are found within the core material (Mohan, 2006). Nanoparticle surfaces may be incorporated with metal

ions, small molecules, polymers and surfactants. For nanoparticles that easily disperse in aqueous media, they are usually prepared with a charged surface. For those materials that are not easily dispersed in aqueous media, they are prepared using a molecule that could covalently bind to the surface of the particle. One of such example is the stabilization of gold and silver sols using citrate (Mu, 2003). Surfactants such as sodium dodecyl sulfate (SDS) are reliable agents that can be used in the formulation of a stable dispersion of nanoparticles (Zhang, 2001). This stability can be achieved by allowing the nanoparticle to form in the core of a micelle were the surfactant tail forms a hydrophobic interaction. The main component of the nanoparticle is the core (Zhang, 2001). Nanoparticles can be divided into two main families: nanospheres and nanocapsules. Examples of nanocapsules include silver, gold, alloy and magnetic nanoparticles (Zonghua *et al.*, 2008)

Nanospheres are defined as particles within the size range of 10-200 nm (Zonghua *et al*., 2008). In nature, they can exist in either crystalline or amorphous state. They have the ability of protecting any drug from the effect of enzymatic and chemical degradation (Aberturas, 2000). Although elimination of drugs may be slowed due to the size of the nanospheres, the major limitation associated with them, is the issue of clearance. This is because the hydrophobic surfaces of the nanospheres are highly susceptible to opsonization and clearance by the reticulo-endothelial system (RES) (Zonghua *et al*., 2008). Another major challenge associated with nanospheres is the ability for them to maintain their own structure in relation to the final aim of drug delivery, polymer biocompatibility and the physico-chemical properties of the drug (Mu, 2003). Nanospheres are differentiated from the vesicles, in that the former consist of the skeletal structure, while the latter consist of capsule of drug store house (Zhang, 2001). One major advantage associated with nanospheres is the ability to administer medication via this system, thus they can be used for site specific drug delivery. They are further sub-divided into biodegradable and non-biodegradable nanospheres (Abra, 1981). Examples of biodegradable nanospheres include: gelatin, albumin, modified starch and polypropylene dextran nanospheres, while that of non-biodegradable nanospheres include the immune and magnetic nanospheres (Gelindo, 2004). Nanospheres are designed in order to ensure that the drug reaches its target site, maintaining its optimal rate and dose regimen (Mu, 2003). Other benefits associated with nanospheres include:

i. Ease of passage via the capillary vessels.
ii. Rapid clearance associated with phagocytes can be avoided thus prolonging its duration in the bloodstream.
iii. Easy penetration via the cells and tissues to enable the drug reach its target site.
iv. Ease of administration to target sites.
v. Reduction in toxicity level.

Major drawbacks associated with nanospheres include (zhang, 2001):

a. They are very difficult to handle either in liquid or dry form.
b. High risk of particle aggregation considering their small size
c. The large surface area affects the drug loading and burst release.

Some of the methods used in the preparation of nanospheres include (Alivisatus, 2004): solvent displacement technique, phase inversion temperature methods,

emulsification polymerization and solvent evaporation (Zhang *et al.*, 2001).

In emulsification polymerization, polymerization medium could be used in dissolving the drug or by adsorbing the drug onto the nanospheres which is subsequently purified (zonghua *et al.*, 2008) In solvent evaporation, the macromolecules are dissolved in the organic solvent. The organic solvent is subsequently removed and precipitated (Al-jamal *et al.*, 2011). In solvent displacement technique, the nanospheres are formed by low-energy method (Chiannil *et al.*, 1990). The polymer is dissolved in an organic water miscible solvent, subsequently added into the aqueous phase in the presence of surfactant. In phase inversion temperature method, nano-emulsion droplets are used to de-solubilize the polymer. Nanocapsules are defined as a nanoscale shell that is usually made from a non-toxic polymer (Echilarasi *et al.*, 2012). As a vesicular system, they are known to encapsulate inner liquid core. Due to the fact that they are used in biological systems, biodegradable polyesters are normally used in its formulation. The polymers that are commonly used include: poly-e-caprolactone (PCL), poly lactide (PLA) (Vartholomeos *et al.*, 2011). Also natural polymers such as chitosan, gelatin, sodium alginate and albumin are used in nanocapsule formulation (Long *et al.*, 2012) Oil surfactant is one of the major core components of the nanocapsule (Nagoverma *et al.*, 2012). The oil that is used in the formulation is one in which the drug is highly soluble and show non-toxic features. The oil-drug emulsion must show high solubility in order to ensure that the drug is carried properly throughout the system. The drug must also be uniformly dispersed throughout the polymeric membrane (Amidon *et al.*, 1995).

**Classification of nanoparticles**

1. **One dimension nanoparticles**: They are mainly useful in the areas of biological and chemical sensors, fiber-optic systems, magneto-optic and optical devices, electronics, chemistry and engineering. The thin films are within the range of 1-100 µm (Vergas *et al.*, 2004).

2. **Two dimension nanoparticles**. They consist of:

a. Carbon nanotubes (CNTs): They are hexagonal network of carbon atoms. There are two types of CNTs: multi-walled carbon nanotubes (MWCNTs) and single-walled carbon nano-tubes (SWCNTs). The carbon nanotubes are unique materials because, they possess remarkable physical, mechanical and electrical properties (Vargas *et al.*, 2014). They are chemically stable and have a great capacity for molecular absorption.

**3** **Three dimension nanoparticles**.

a. **Fullerenes (Carbon 60)**: They are spherical in shape and contain more than 100 carbon atoms. When subjected to extreme pressure, they regain their original shape when the pressure is released. Since they are empty structures that possess dimensions similar to that of biological active molecules, they are useful in the area of medical applications (Tomala *et al.*, 2004)

b. **Dendrimers**: They are controlled polymers that possess nanometric dimensions. They are ideal carriers for target drug delivery due to the multiple functional groups on their surfaces (Li *et al.*, 2007). They are useful in large scale synthesis of organic and in-organic nanostructures with dimensions of 1- 100 µm. Dendrimers can also be fabricated to

metallic nanostructure and nanotubes (Fu *et al.*, 2007). Over the years, dendrimers have shown pharmaceutical applications in the areas of anti-inflammatory formulations, antimicrobial, antiviral drugs and anticancer agents (Cheng *et al*., 2005). One major disadvantage associated with dendrimers is toxicity. This is due to their ability to disrupt cell membranes as a result of a positive charge on their surfaces (Cheng *et al.*, 2005).

c. **Quantum dots (QDs):** They are small devices that contain a tiny droplet of free electrons (Cham *et al.*, 2002). QDs are semiconductor nanocrystals that have diameters in the range of 2-10 nm. Examples include: cadmium selenide (CdSe), cadmium telluride (CdTe), indium phosphide (InP) and indium arsenide (InAs). QDs have the ability to attach therapeutic agents for simultaneous drug deliver, *in vivo* imaging and tissue engineering (Goldberg *et al.*, 2007).

**Preparation of nanoparticles**

1. **Spontaneous Emulsification/Solvent Diffusion Method**

This is a modification of the solvent evaporation technique (Song *et al*., 1997). In this method, water-miscible solvents such as acetone or methanol are used as the organic phase I and water immiscible solvents such as dichloromethane or chloroform are used as the organic phase II. The interfacial turbulence that is created when these two phases are mixed leads to the formation of nanoparticles (Vargas *et al*., 2004). Even though this method produces significantly smaller sized particles, there are some disadvantages such as the presence of residual organic solvent.

**2. Double emulsion evaporation method**: This is used to encapsulate hydrophilic drugs due to the limitation of poor entrapment of these drugs exhibited in emulsion and evaporation method (Jaiswal *et al*., 2004). It involves the addition of aqueous drug solutions to organic polymer solution under vigorous stirring to form w/o emulsion. The w/o emulsion formed is then added into the second aqueous phase with continuous stirring to form w/o/w emulsion. The double emulsion formed is then subjected to solvent removal by evaporation and after centrifugation, the nanoparticles are obtained. They are washed and lyophilized (Lemarchard *et al*., 2006). Uchida *et al.*, *(*1995), prepared ovalbumin loaded poly (lactide-co-glycolide) (PLGA) microparticles by a w/o/w emulsion solvent evaporation technique using NaCl for protection of the aqueous droplets. The addition of NaCl into the external aqueous phase improved ovalbumin loading efficiency (Uchida *et al*., 1995). Graves et a*l*., (2004), investigated the effect of blending of low molecular weight (Mw) and high molecular weight (Mw) PLGA on the characteristics of microparticles prepared by double emulsion solvent evaporation technique (Jaiswal *et al*., 2004). The drug encapsulation efficiency increased significantly when high Mw PLGA (RG 506) was mixed with low Mw PLGA (RG 502) at a ratio of 1:7 (Graves *et al*., 2004). Blanco-Prieto *et al*., (2002), used water soluble peptide (pBC 264) to encapsulate in PLGA microparticles using double emulsion solvent evaporation method. There was significant improvement (90 %) in the encapsulation efficiency (Blanco *et al*., 2002).

**3 Salting-out method**: In this method, water-soluble polymers are dissolved in a highly concentrated solution of electrolytes or nonelectrolytes to obtain a viscous gel (aqueous phase) (Dezhi, 2018). This aqueous

gel is added to an organic phase such as acetone to obtain an oil-in-water emulsion under vigorous stirring. The formation of nanoparticles is facilitated by adding an excess amount of water, which diffuses the acetone out. The residual organic solvent is removed by continuous stirring or high-speed homogenization Dezhi *et al*., (2018) studied the effect of salting out agents on extraction. He discovered that salting-out agents had a lot of functions which include: the hydration function of the salting out agents attracted one part of water molecule to reduce free water molecule in the system (Dezhi, *et al.,* 2018), salting out agents reduced the polymerization of the metal ions. Some of the solvents used were ethanol, acetone and methanol, while some of the salting-out agents used were calcium chloride, magnesium chloride and magnesium acetate. Keemi *et al*., (2010), discovered that salting-out agents play prominent role in manufacturing polymer encapsulated drugs nanoparticles (Keemi *et al.,* 2010). The salts initially impede the miscibility of organic phase into aqueous solution, forming emulsion. Subsequently, reverse salting-out effect leads to precipitation which aided in entrapment of drug into the polymer matrix forming the nanoparticles. Salting out method has a lot of advantages such as high yield, high encapsulation efficiency and small particle size.

**4 Emulsion-diffusion method**: The encapsulating polymer is dissolved in a partially water-miscible solvent which is saturated with water to ensure thermodynamic equilibrium of both liquids. The polymer-water saturated solvent phase is then emulsified in an aqueous solution, which leads to formation of nanospheres or nanocapsules (Elizabeth *et al*., 2015). The solvent is eliminated by evaporation. This method offers a lot of advantages such as high encapsulation efficiency (> 70 %), high reproducibility, simple, easy to scale up and narrow size distribution. It has its own disadvantage such as: elimination of high volume of water, leakage of water-soluble drugs into the saturated aqueous external phase during emulsification. Elizabeth Pinon *et al*; used the emulsion-diffusion method to obtain polymeric nanoparticles. They recorded high reproducibility and versatility with its simple implementation (Elizabeth *et al.*, 2015).

**5 Solvent displacement/precipitation method**: It involves the precipitation of a preformed polymer from an organic solution. The polymer and drug are dissolved in a semi polar water miscible solvent (ethanol or acetone). The solution formed, is subsequently poured into an aqueous solution containing stabilizer under magnetic stirring. This leads to the formation of nanoparticles by the rapid solvent diffusion. The solvent is then removed, by low or reduced pressure. Chandramani *et al.,* (2019), used solvent displacement method to produce nanoparticles. The polymer was precipitated from the organic solution and the organic solvent diffused in the aqueous medium. Under magnetic stirring conditions, the prepared solution was added into the aqueous solution containing stabilizers. By rapid solvent diffusion, nanoparticles were formed instantly. Some of the stabilizers commonly used include: sodium alginate, sodium carboxymethyl cellulose, guar gum, locust bean gum, gelatin and pectin (Chandramani *et al.,* 2019). Generally stabilizers are added at the rate of 0.2 to 0.3 % of the mix. They have high water-holding capacity, give uniformity of product, desired resistance to melting and improve handling properties.

## Characterization of nanoparticles

Nanoparticles are characterized based on size, morphology and surface charges using microscopic techniques such as scanning

electron microscopy (SEM), transmission electron microscopy (TEM) and atomic force microscopy (AFM) (Gabindo *et al*., 2004)

a) **Particle size:** Electron microscopy is used to measure the morphology and size of nanoparticles. Indirect relationship exists between the particle size and surface area. Smaller particles offer large surface area, while larger particles offer smaller surface area (Betancor *et al*., 2005). Particle size is also affected by polymer degradation.

**Methods used to determine nanoparticle size**

1. **Dynamic light scattering (DLS):** This is also known as photon-correlation spectroscopy (PCS). It is used to determine the size of nanoparticles in colloidal suspension. This is achieved by shining a monochromatic light onto a solution of spherical particles. This causes a Doppler shift when the light hits the particles in motion, thus changing the wavelength of the incoming light (Deassi *et al*., 2008).

2 **Scanning electron microscopy (SEM)**: It is used to determine the morphological examination of nanoparticles. One of its limitations is that it doesn't give enough information about the size distribution (Molpeceres *et al.*, 2008). For characterization of a sample material using SEM, the nanoparticles solution should be converted to powder, which is then mounted on a sample holder followed by coating with a conductive metal. With a focused fine beam of electrons, the sample is scanned. The secondary electrons emitted from the sample surface gives an information on the surface characteristics of the sample (Jores *et al*., 2004).

3 **Atomic force microscopy (AFM):** It is used to scan samples at sub-micron level using a probe tip of atomic scale (Molpeceres *et al.*, 2000). Depending on the properties of the sample, they could be scanned in contact or non-contact mode. In contact mode, the topographical map is generated by tapping the probe onto the surface across the sample and probe hovers over the conducting surface in non-contact mode. One of the advantages associated with AFM, is the ability to allow imaging of delicate biological and polymeric nano and micro structures.

4 **Surface charge**: This determines the interaction of nanoparticles with the biological environment and the electrostatic interaction with bioactive compounds. Zeta potential is used to analyze the colloidal stability of nanoparticles. The zeta potential measurement helps to predict about the storage stability of colloidal dispersions (Pangi *et al*., 2003).

5 **Surface hydrophobicity**: There are different techniques used to determine the surface hydrophobicity of a nanoparticle. They include: hydrophobic interaction chromatography, biphasic partitioning adsorption of probes and contact angle measurements (Scholes *et al*., 1999).

**Table 1: Application of nanoparticles (Pangi *et al*., 2003)**

| | Applied field | Application |
|---|---|---|
| 1 | Nanomedicines | Nanodrugs, medical devices, tissue engineering. |
| 2 | Chemicals and cosmetics | Nanoscale chemicals, paints, coatings |
| 3 | Materials | Nanoparticles, carbon nanotubes, biopolymer |

| | | |
|---|---|---|
| 4 | Food science | Processing, nutraceuticals, nanocapsules |
| 5 | Military and energy | Biosensors, weapons |
| 6 | Electronics | Semiconductor chips, memory storage. |
| 7 | Scientific tools | Atomic force, microscopic techniques |
| 8 | Agriculture | Atomic force microscopy and scanning tunneling |

**Hydrogel nanoparticles**

Hydrogels are hydrophilic polymers that have the ability to imbibe large volume of water (Hugaard *et al*., 1995). Due to their network structure, they are insoluble in water. When hydrogels are held together by molecular entanglements, they are known as reversible or physical gel (Hofman, 2002). There are two types of hydrogels: Physical and chemical hydrogels.

Physical hydrogels are formed when a polyelectrolyte is combined with a multivalent ion of the opposite charge. Chemical hydrogels are formed by cross-linking water soluble polymers to form a network (Lim *et al*., 1980) Both the physical and chemical hydrogels are not homogenous, due to the fact that they contain regions of low water swelling and high cross-link density which are dispersed within the swelling region (Drumbellar, 1995).

**Applications of hydrogel nanoparticles**

1 **Treatment of inflammatory conditions:** Hydroxyl radicals are normally produced from inflammation cells during inflammation. Yui and co-workers (Yui *et al*., 1992), developed drug delivery system that is related to the hydroxyl radicals using hyaluronic acid (HA). The HA is degraded either by hyaluronidase or hydroxyl radical in the human body. When HA is injected at inflammatory site, the degradation via hydroxyl radicals becomes rapid.

2 **Bioresponsive hydrogels for drug delivery system**: This is applicable in the area of insulin release in response to raised blood sugar levels (Kin *et al*., 1990). A research was carried out by Ishihara et al., (1984), where glucose oxidase molecules were immobilized onto a basic polymer carriers. Due to the enzyme reaction that converts glucose to gluconic acid, there was lowering of the pH, which caused the basic groups on the polymer to be protonated, thus inducing swelling and enhancing insulin release. In summary, the system works as a feedback loop, where the release of insulin causes the sugar level to drop, resulting in an increased pH (Ishihara *et al.*, 1984).

3 **Glucose sensitive hydrogels**: Hydrogels that have lectins are carbohydrate binding proteins which interact with glycoproteins and glycolipids on the cell surface, resulting in cell agglutination, cell adhesion and hormone like actions (Brownlee *et al.*, 1979).

4 **Bioresponsive hydrogels for sensing**: Kirsch et al, (2000), developed a hydrogel-based photonic crystal that acts as a glucose sensor for patients with diabetes mellitus (Kirsch *et al*., 2000). They achieved this by attaching a glucose oxidase to arrays of polystyrene nanospheres. This caused the material to swell in the presence of glucose.

The swelling increases the mean separation between the immobilized nanospheres, causing a shift in the Bragg peak of diffracted light to lower wavelengths and producing a red-shift in the optical properties.

**Table 2: Recent development in the hydrogel based biosensor (Akihiki et al., 2002)**

| Stimulus | hydrogel | Application | Output signal |
|---|---|---|---|
| Glucose | PA-PEG | Glucose biosensor | Optical colour |
| Protein | PNIPAmcoAAc | Avidin, antibiotin | Optical focusing |
| Peptide | PEG | Liver cell biosensor | Biochemical fluoresence |

1. **The use of hydrogels as carriers in controlled drug delivery system**: Hydrogels are used as carriers for pharmaceutical applications, for delivery of drugs, peptides or proteins, to regulate drug released in reservoir-based controlled release system, and as carriers in swellable and swelling controlled release devices (Ende *et al.*, 1997). In swelling controlled systems, the drug which is dispersed in the polymer, diffuses out as water uptake occurs and the polymer swells. The drug release rate is dependent both on water diffusion and polymer chain relaxation.

**Carbon Nanotubes (CNTs)**

They have wide application in pharmacy due to their high surface area that is able to adsorb or conjugate with a wide variety of therapeutic and diagnostic agents (Lijima, 1991). They are excellent vehicle for drug delivery by penetrating into the cells directly and keeping the drug intact without metabolism during transport in the body (Digge *et al.*, 2012). CNTs belong to the family of fullerenes. There are two types of carbon nanoparticles: (a) single-walled carbon nanotubes (SWCNTs), (b) multi-walled carbon nanotubes (MWCNTs). The SWCNTs consist of two to several coaxial cylinders with a graphine sheet surrounding a hollow core. There are three major techniques used in the production of SWCNTs and MWCNTs:

a. Arc-discharge method (uses arc-vaporization of two carbon rods)

b. Laser-ablation method (uses graphite)

c. Chemical vapour deposition (uses hydrocarbon sources).

There are two main approaches for the functionalization of CNTs:

a. Covalent attachment (chemical bond formation)

b. Non-covalent attachment (physioadsorption).

The covalent form is obtained by oxidation with strong acid (Madami, 2011). This leads to the formation of carboxyl groups at the open side, while the non-covalent functionalization is obtained by coating CNTs with amphiphilic surfactant molecules.

After functionalization, CNTs can then be linked with drugs or biomolecules for the delivery into the target cells (Liu, 2007).

**Applications of carbon nanotubes**

They are useful in the areas of gene delivery, tissue regeneration, biosensor diagnostics and analysis. The functionalized CNTs can carry molecules across the cytoplasmic membrane without producing a toxic effect. CNTs can easily cross cell membrane due to certain features they possess which include: Simple-hydrophobic interaction, electrostatic adsorption and covalent bonds. They do not only promote cellular uptake of therapeutic molecules, but also keep them intact during transportation and cellular penetration.

**1. Carbon nanotubes for cancer therapy**

a. **By drug delivery**: CNTs are often used as drug carriers to treat tumors. The conventional anticancer drugs have some limitations which include: systemic toxicity, drug resistance and limited penetration. The use of CNTs as drug carriers offers a better advantage because they penetrate into the cells and also promote the cellular uptake of therapeutic molecules. The anticancer drugs transported via CNTs will be liberated in situ with intact concentration. Some of the anticancer drugs that have been conjugated with functionalized CNTs include: epirubicin, doxorubicin, cisplatin, methotrexate, quercetin and paclitaxel.

b. **By antitumor immunotherapy**: CNTs have been used as carriers in antitumor immunotherapy. This was achieved by stimulating the patients' immune system to attack the malignant tumor cells (Ende *et al*, 1997).

c. **By local antitumor hyperthermia therapy**: CNTs have been used in hyperthermia therapy for cancer treatment due to the fact that they generate significant amount of heat upon excitation (Jiang *et al*., 2012).

2. **Carbon nanotubes for infection therapy**: CNTs have been used to resolve the problem associated with resistance of infectious agents against antiviral and antibacterial drugs. Functionalized CNTs are a good carrier system for the delivery of candidate vaccine antigens (Li *et al.*, 2010)

3. **Carbon nanotubes for gene therapy by DNA delivery**: Gene therapy which involves the introduction of a DNA molecule into the cell nucleus is used to correct a defective gene caused by hereditary diseases. CNTs have the ability to transport genes inside mammalian cells and keep them intact (Lay *et al*., 2011).

4. **Carbon nanotubes for solid phase extraction of drugs and biochemical**: CNTs are known to possess excellent adsorption ability. Many drugs such as benzodiazepines, sulfonamides, antidepressants have been extracted using solid phase extraction adsorbents. CNTs can also be used to extract inorganic ions and organometallic compounds to prepare stationary phases of gas and liquid chromatography (Bekyarova, 2005).

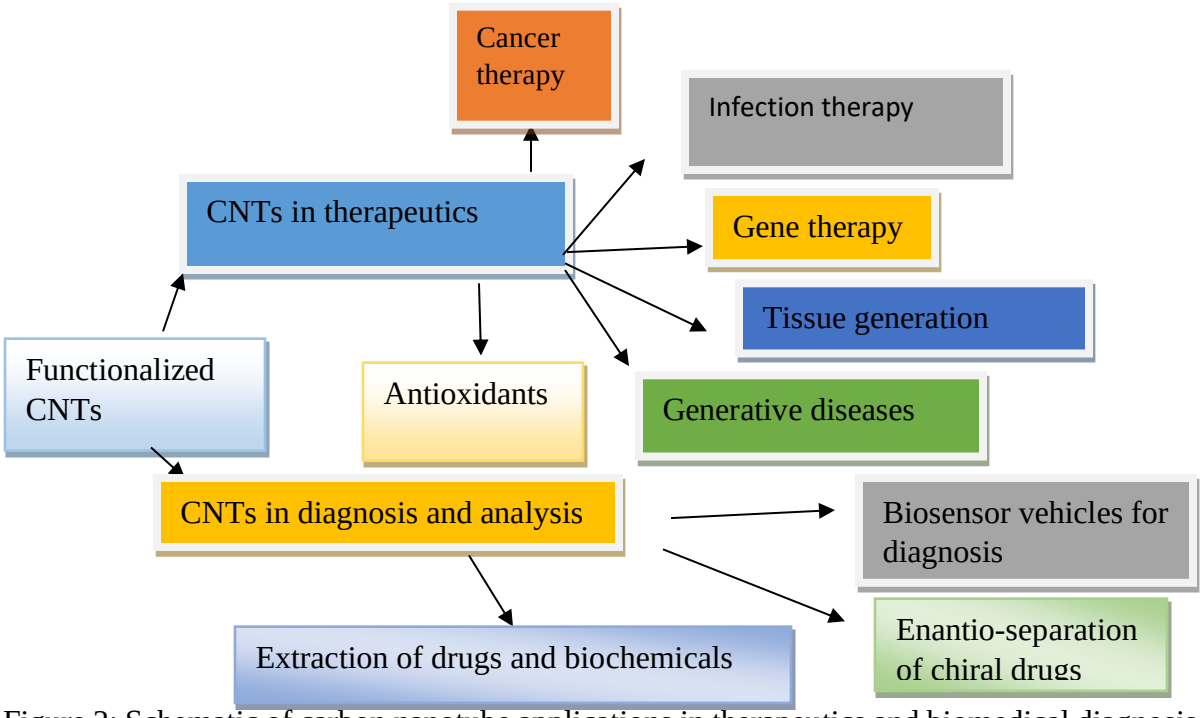


Figure 2: Schematic of carbon nanotube applications in therapeutics and biomedical diagnosis and analysis**.**

**Dendrimers**

A dendrimer is a macromolecule that is characterized by highly branched three-dimensional structure which provides a high degree of surface functionality and versatility (Frechet, 2002). They possess three architectural components which include: an initiator core, interior layers and exterior (Barbara, 2001).

**Synthesis of dendrimers**

1 **Divergent dendrimer growth:** It is one of the synthetic methods used due to the nature of the dendrimer to grow outwards from the core, diverging into space. This approach is used for the production of large quantities of dendrimers (Lay *et al*., 2011)

2 **Convergent dendrimer growth:** This approach was developed due to the weakness of divergent syntheses. It works by linking surface units together. The major disadvantages associated with this approach is the low yield in the synthesis of large structures (Li *et al*., 2007).

**Properties of dendrimers (Bossman *et al.*, 1999)**

1 It is useful in nanoscale sizes that have similar dimensions

2 It is useful in terminal surface groups that are suitable for bio-conjugation of drugs

3 It is suitable for surfaces that may be designed with functional groups to

augment or resist transcellular permeability

4 An interior void space may be used to encapsulate small molecular drugs.

5 It is suitable for surface groups that can be modified to optimize bio-distribution, receptor mediated targeting or controlled release of drug.

**Applications of dendrimers**

1. **Oral drug delivery**: The use of polyoxyethylene glycol chains or ionic groups can reduce the problem of flocculation and aggregation of the system

2. **Ocular drug delivery**: Dendrimers help to solve most of the problems associated with ocular delivery such as bioavailability, irritation, sterility and biocompatibility (Colton, 2004).

3. **Transdermal drug delivery**: Highly water soluble and biocompatible dendrimers have the capacity to improve drug properties such as solubility and plasma circulation time to enhance drug delivery system for nonsteroidal anti-inflammatory drugs, antiviral and anticancer drugs (Gajbhiye *et al.*, 2008).

4. **Targeted gene delivery**: They act as carriers in gene therapy. They transfer genes via the cell membrane into the nucleus. Cationic dendrimers are used to form compact complexes with DNA.

5. **CNS delivery**: They are used as delivery vehicles for drug therapy or molecular imaging (Vandamme, 2005)

6. **Anticancer drug delivery**: Dendrimers are used for the encapsulation and conjugation of multiple entities either on the surface or in the core making them ideal carriers for various anticancer drugs. In 2005, Vandamme *et al* (2005), worked on the possibility of a 2,2-bis propanoic acid dendritic scaffold as a delivery carrier for doxorubicin *in vitro*. PEGylated dendrimers possess low level of toxicity and lower accumulation in different organs, and also they exhibit prolong blood circulation.

**Quantum dots (QDs)**

They are nanomaterials that are commonly used as drug targeting and in-vivo biomedical imaging (Gao *et al*., 2004). They are also semi-conductor nanocrystals that exist within the range of 2-100 nm. They are used in biomedial imaging due to the fluorophores (Colton *et al.*, 2004). Fluorescent QDs can be conjugated with antibodies and receptor ligands to target specific biological events and cellular structures. Newly synthesized QDs are hydrophobic in nature and are thus not biologically useful. To render them biologically useful, they are given secondary core durability and desired bioactivity. The functionalization can be activated via: electrostatic interactions, adsorption, multivalent chelation or covalent bonding. QDs can be designed in such a way as to give them specific bioactivities such as therapeutic and diagnostic purposes (Deassin, 2008).

**Aerogels**

An aerogel is a solid that has an extremely low density and low thermal conductivity (Jaypee et al., 2016). It is a nanoporous insulation material that has an open cell structure. They are nontoxic, lower flammability limit, light weight and are permeable. There are two types of aerogels: monolithic and granular aerogels. The former has higher solar transmittance, than the latter. Due to their unique structures, they are used as insulators (Donaruma *et al.,* 1985).

**Properties of aerogel**

1. It has low density

2. It has 90-99.9 % porosity

3. Low thermal conductivity

4. High melting point (950 $^{o}$C)

**Applications of aerogels**

1. They are used as heat insulating materials

2. They are used as energy applications

3. They are used to improve polyester thermal insulators properties

**Advantages of aerogels**

1. They are the best type of insulators

2. They have high thermal resistance

3. They have reduced sensitivity to mechanical damage

4. They are water repellant.

**Limitations of aerogels**

1. They have high cost of production

2. They are soft and light weight

3. They are prepared from chemicals that may affect health

4. They can be destroyed by contact with liquids

**Liposomes**

They are colloidal and vesicular structures that compose of one or more lipid bilayers surrounding an aqueous compartment (Sunil *et al*, 2005). Liposomes entrap drugs which are released for subsequent absorption at the intestinal membrane surface. Various carriers like liposomes, nanoparticles, microparticles, polysaccharides, and lecithin can be used to target the drug to a specific site. They have a size range of 0.01 – 5.0 µm in diameter. Liposomes can encapsulate both hydrophilic and hydrophobic drugs.

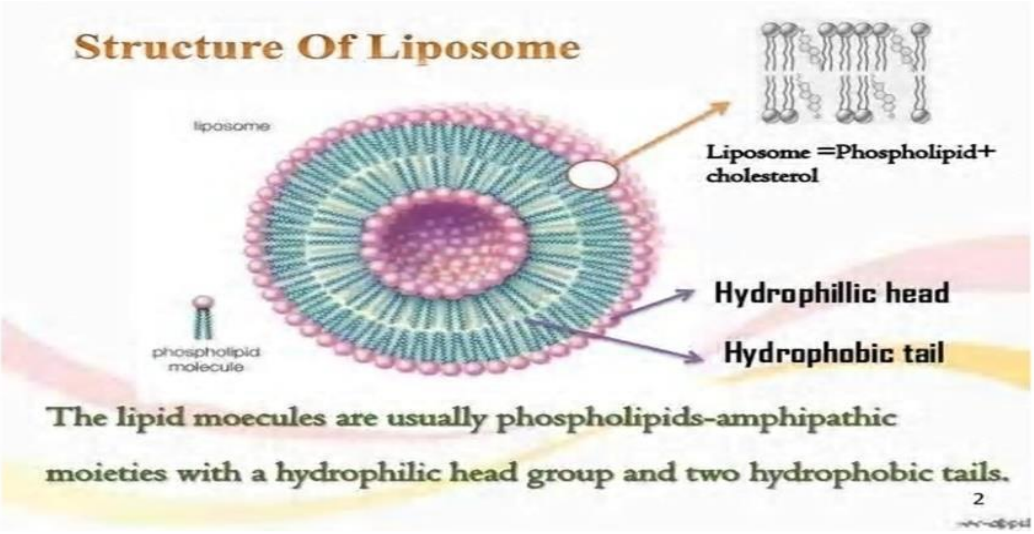


**Figure 3: Structure of liposome (Sunil *et al*., 2005).**

**Advantages of liposomes (Sharma *et al.*, 1997).**

1. They are biocompatible, biodegradable and non-toxic

2. They are suitable for delivery of hydrophobic, amphipathic and hydrophilic drugs

3. They protect encapsulated drugs from the external environment

4. Reduced toxicity of encapsulated agent

5. Increased stability

6. Reduced exposure of sensitive tissues to toxic drugs

7. They increase efficacy and therapeutic index of drug

8. They are used to achieve active targeting.

**Disadvantages of liposomes**

1. High cost of production

2. They have short-half life

3. Leakage and fusion of encapsulated drug

4. They have low solubility

5. Phospholipids can undergo oxidation and hydrolysis.

**Types of liposomes**

1. **Based on structural parameters**

a) Unilamellar vesicles (ULV)

| Types | Size ranges (nm) |
|---|---|
| Small unilamellar vesicles (SUV) | 20-40 |
| Medium unilamellar vesicles (MUV) | 40-80 |
| Large unilamellar vesicles (LUV) | 100-1000 |

b Oligolamellar vesicles (OLV): They consist of 2-10 bilayers of lipids.

b) Multilamellar vesicles (MLV): They consist of several bilayers.

2. **Based on method of preparation**

i. REV: Reverse-phase evaporation method is used for single or oligolamellar vesicles

ii. MLV-REV: Multilamellar vesicle made by Reverse Phase Evaporation Method

iii. SPLV: Stable plurilamellar vesicles

iv. FATLV: Frozen and thawed MLV

**3.** VET: Vesicles prepared by extrusion technique

vi. DRV: Dehydration-rehydration method

**3 Based on composition and application**

i. Conventional liposomes (CL): They consist of neutral or negatively charged phospholipids and cholesterol

ii. pH sensitive liposomes: consist of phospholipids such as phosphatidyl ethanolamine

iii. cationic liposomes: consist of cationic lipids

iv. Long-circulatory liposomes (LCL): They consist of polyethylene glycol derivatives (PEG). They are attached to the surface of liposomes to decrease their detection by phagocyte system

v. Immuno-liposomes: consist of conventional or long circulatory liposomes with monoclonal antibody.

**Structural components of liposomes**

1. **Phospholipids**: The backbone of the molecule is glycerol moiety. Examples include:

a. Phosphatidyl choline (lecithin)

b. Phosphatidyl ethanolamine (PE)

c. Phosphatidyl serine (PS)

d. Phosphatidyl inositol (PI)

e. Phosphatidyl glycerol (PG)

**2 Sphingolipids**: The backbone is sphingosine. They consist of three building blocks: fatty acid, sphingosine and a head group of simple alcohol and complex carbohydrate.

**3. Sterol**: They consist of cholesterol and its derivatives. They are very important in decreasing the fluidity or micro viscosity of the bilayer. They reduce the permeability of the membrane, and they stabilize the membrane in the presence of biological fluids.

**4. Synthetic phospholipids**: They are classified as saturated and unsaturated phospholipids (Tirrel, 1976). The saturated types consist of:

a. Dipalmitoyl phosphatidyl choline (DPPC)

b. Distearoyl phosphatidyl choline (DSPC)

c. Dipalmitoyl phosphatidyl ethanolamine (DPPE)

d. Dipalmitoyl phosphatidyl serine (DPPS)

e. Dipalmitoyl phosphatidic acid (DPPA)

f. Dipalmitoyl phosphatidyl glycerol (DPPG).

The unsaturated types consist of:

a. Dioleoyl phosphatidyl choline (DOPC)

b. Dioleoyl phosphatidyl glycerol (DOPG).

**5. Polymeric materials**: Consist of synthetic phospholipids with diactylenic group. They polymerize when exposed to UV leading to formation of polymerized liposomes.

6. Polymer bearing lipids.

**Preparation of liposomes**

1. General method of preparation

2. Specific method of preparation.

The general method of preparation involves four stages:

a. Drying down lipids from organic solvent

b. Dispersing the lipids in aqueous media

c. Purifying the resultant liposome

d. Analyzing the final product.

The specific methods are classified into three types:

a. Physical dispersion methods

b. Solvent displacement methods

c. Detergent solubilization methods

**Physical dispersion methods**: The aqueous volumes enclosed within lipid membrane is about 5-10 %. This method is favourable to liquid soluble drugs which can be encapsulated to high percentage, but does not favour water soluble drugs which are easily lost during preparation. Physical dispersion involves three methods:

i. **Hand shaking method**: The liquid mixture and charged components are dissolved in chloroform and methanol mixture in the ratio of 2:1. The mixture is introduced into a 250 ml round bottomed flask. The flask is attached to rotary evaporator connected with vacuum pump, and rotated at 60 rpm for 30 min. The organic solvents are evaporated, which helps to form a dry residue. The evaporator is detached from vacuum pump and nitrogen is introduced into it. The flask is removed from evaporator and fixed onto lyophilizer to remove the residual solvent. The flask is subsequently flushed with nitrogen and 5 ml of phosphate buffer is added. The flask is again attached to evaporator and rotated at 60 rpm for 30 min. A milky suspension is formed which is then allowed to stand for 2 h, so as to complete the swelling process to give MLVs.

ii. **Non-shaking method**: The solutions of lipid in chloroform and methanol are spread over the flat bottom of the conical flask. The solution is evaporated at room temperature by flow of nitrogen via the flask. Saturated nitrogen is then passed via the flask after drying to remove the opacity of the dried films. The flask is inclined to enable the introduction of 10-20 ml of 0.2 M sucrose in distilled water. The flask is slowly returned to upright position. The flask is flushed with nitrogen and allowed to stand for 2 h, to facilitate swelling. The vesicles are mixed to yield a milky suspension which is then centrifuged at 1200 rpm for 10 min to form LUVs (Uchida *et al*., 1995).

2. **Freeze drying**: This method involves the removal of solvents from a material via the process of sublimation. Controlled freeze drying keeps the temperature of the product low enough during the process to avoid changes in the dried product appearance and characteristics. This method is used to preserve pharmaceuticals, proteins and microbes. Certain requirements must be met in order to successfully freeze dry a sample. They include: the collector coil of the freeze dryer must be 15-20 $^{o}$C colder than the freezing point of your preferred sample, presence of a vacuum pump, a drying accessory such as manifold chamber. The lyophilization process involves three stages such as pre-freezing, primary drying and secondary drying. In the pre-freezing stage, the sample material will need to be cooled to at least the temperature of the melting point of the sample. Primary drying occurs when you start your freeze dryer and vacuum pump. With the low pressure environment, evaporative cooling of the sample begins, thus allowing for energy in the form of heat to speed the freeze drying process. Secondary drying involves the release of the water molecules that are bound to the sample. Secondary drying is applied mainly in samples that are being prepared for a long term preservation and storage.

3. **Solvent dispersion methods**: It involves dissolving the lipids in an organic solution and making it come in contact with an aqueous phase containing materials to be entrapped within liposome. It involves two methods:

a. **Ethanol injection method**: It involves injecting an ethanol solution to an

aqueous medium via a fine needle. The ethanol is diluted in water and the phospholipid molecules are evenly dispersed via the medium to yield high proportion of SUVs.

b. **Ether injection**: The organic solution is injected slowly into an aqueous phase via the narrow needle at temperature where the organic solvent is evaporated.

4. **Detergent solubilization technique**: A detergent is used to make the phospholipids come in contact with the aqueous phase. This leads to the formation of micelle [116].

**Applications of liposomes**

1. Cancer chemotherapy
2. Gene therapy
3. Liposome as carriers for vaccine
4. Liposomes as carriers of drug in oral treatment
5. Liposomes for topical applications
6. Liposomes for pulmonary delivery
7. Cell biological application
8. Ophthalmic delivery of drugs.

**Niosomes**

**Niosomes**: They are drug delivery system which are amphiphilic in nature and are made of non-ionic surfactants that can entrap both hydrophilic and hydrophobic drugs in the core cavity and non-polar region respectively (Gregoriadis, 1979). The major difference between niosomes and liposomes is that the former consist of non-ionic surfactants, while the latter consist of phospholipids. The niosomal system is chemically stable, biodegradable, biocompatible, low production cost, easy storage and handling and low toxicity (Ezhilarasi *et al*., 2012).

**Advantages of niosomes.**

1. They can entrap solutes
2. They are osmotically stable and active
3. They discharge the medication in a controlled manner
4. They improve solubility and oral bioavailability of poorly soluble drugs
5. They protect the drug from biological environment
6. They increase the stability of entrapped drugs
7. The surfactants are easy to handle
8. They can reach the site of action of various routes of drug administration.

**Types of niosomes**

1. Proniosomes: They are formed from the carrier and surfactant mixture
2. Aspasomes
3. Niosomes in carbopol gel
4. Vesicles in water and oil system (v/w/o)
5. Niosomes of hydroxyl propyl methyl cellulose
6. Deformable niosomes: consist of non-ionic surfactants, ethanol and water.

**Classification of niosomes based on number and size of bilayer (Tomalia, 2004)**

1. Multilamellar vesicles (MLV): They have a diameter of 0.5–10 µm. They are the most widely used because they are simple to make and they have high mechanical stability upon storage.
2. Large unilamellar vesicles (LUV)
3. Small unilamellar vesicles (SUV).

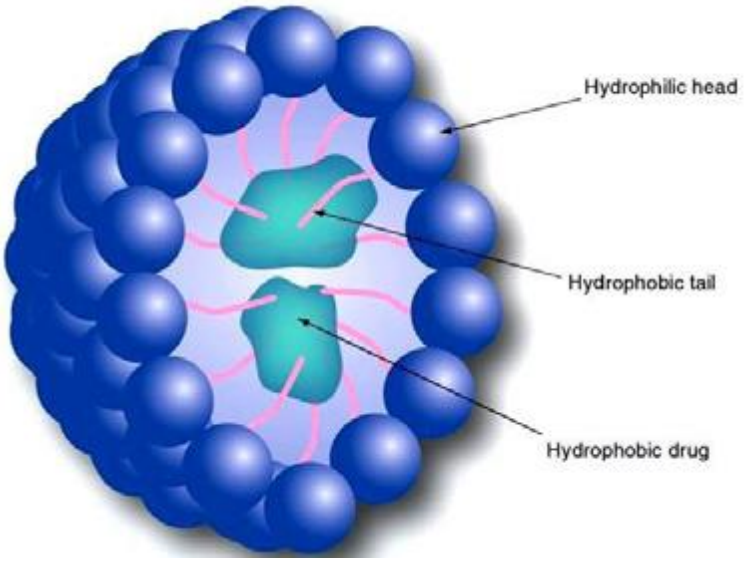


**Figure 4: structure of noisome (Jaypee *et al.*, 2016).**

**Difference between noisome and liposomes**

| Liposomes | Niosomes |
|---|---|
| They are more expensive | Less expensive |
| Phospholipids are prone to oxidative degradation | Surfactants are stable |
| Requires special method of storage, handling and purification | Does not require any special method |
| Phospholipids may be neutral charged | Surfactants are uncharged. |

**Components of niosomes (Jaypee *et al.*, 2016).**

1. Cholesterol
2. Non-ionic surfactants

**Methods of preparation of niosomes**

1. **Ether injection**: It involves the slow injection of surfactants and cholesterol in preheated aqueous phase at 60 $^{o}$C. The ether solution is evaporated using the rotary evaporator which forms the single layered vesicles.

2. **Sonication**: the surfactant: cholesterol mixture is dispersed in 2 ml aqueous phase. The dispersion is subjected to probe sonication for 3 min at 60 $^{o}$C. Probe sonicator is used for small sample volume, while bath sonicator is used for large sample volume.

3. **Hand shaking method**: This method is used in the synthesis of MLVs. It involves the dissolution of the surfactants and some additives such as cholesterol in an organic solvent in a round bottomed flask. The organic solvent is then removed using a

rotary evaporator to form a thin film on the outside wall of the flask. The dried film is subsequently hydrated with aqueous solution containing the drug for about 1 h with mechanical shaking to form niosomal dispersions that have a milky appearance. Other drugs that have been entrapped include: diclofenac sodium, luteinizing hormone releasing hormone and Adriamycin.

4. **Thin film hydration method (TFH):** This method is widely used because of its simplicity. It involves dissolving the surfactants and some additives in an organic solvent in a round-bottomed flask. The thin film is formed on the inside wall of the flask when the organic solvent is removed using a rotary vacuum evaporator. In addition, aqueous solution such as water is added to the dry film which is hydrated above the transition temperature of the surfactant. This method has been used to form MLVs. Also, TFH method has been used to form niosomes entrapped in minoxidil nimesulide insulin and antioxidants.

5. **Reverse phase evaporation technique (REV)**: This method is used to prepare LUVs. It involves the dissolution of the niosomal ingredients, surfactants and additives in an organic solvent. Subsequently, the aqueous phase which contains the drug is added to the organic phase, and the mixture is sonicated in order to form an emulsion. The organic solvent is then removed slowly using a rotary vacuum evaporator at about 40-60 $^{o}$C. At this point, LUVs are formed after complete hydration. REV method has been used to prepare niosomes entrapped in diclofenac sodium, acetazolamide and naltrexone. The main advantage associated with this method is that it gives a very high entrapment efficiency.

6. **Bubble method**: This is a novel technique that does not require any organic solvent. It consists of round-bottomed flasks that has three necks placed in water bath to control the temperature. The first and second neck consist of water-cooled reflux and thermometer respectively, while the third neck consist of nitrogen supply. Cholesterol and surfactants are dispersed together in pH 7.4 at 70 $^{o}$C. A continuous stream of nitrogen gas bubbles is generated and introduced via the dispersion to form niosomes

**Factors that affect the physico-chemical properties of niosomes**

1. Amount and type of surfactant: Increased HLB of surfactants, increases the mean size of niosomes, because the surface free energy decreases with an increase in hydrophobicity of surfactant.

2. Nature of surfactants: The surfactant used in noisome formation must have a hydrophilic head and hydrophobic tail. The surfactants with alkyl chain length from $C_{12}$ to $C_{18}$ are suitable for preparation of niosomes.

3. Nature of encapsulated drug

**Applications of niosomes**

1. As a carrier for hemoglobin: They can be used as a carrier for hemoglobin because they show a visible spectrum that is super imposable to that of hemoglobin.

2. As a drug carrier

3. Ophthalmic drug delivery

4. Delivery for peptide drugs

5. Transdermal delivery

6. Neoplasia

7. In studying immune response

8. Anti-inflammatory agent

9. Immunological application: They are used to study the nature of the immune response provoked by antigens.

## Aquasomes

They are nanoparticulate carrier system that are made up of three layered self-assembled structures which contain calcium phosphate or ceramic diamond covered with a polyhydroxyoligomeric film.

## Properties of aquasomes

1. They maintain molecular confirmation and optimum pharmacological activity
2. They possess large size and active surface
3. They preserve conformational integrity and biochemical stability
4. They avoid clearance by reticuloendothelial system
5. They can be characterized for structural analyses.

## Formulation of aquasomes

1. **By principle of self-assembly**: This is governed by three physicochemical processes. The interactions of charged groups, dehydration effects and structural stability.

**Interactions between charged groups**: The interactions of charged groups facilitates long range approach of self-assembly. Such groups such as amino-, carboxyl-; sulfate- and phosphate groups facilitate the approach of self-assembling sub-units.

**Hydrogen bonding and dehydration effects**: Secondary proteins are stabilized by hydrogen bonding. They also have a major role in base pair matching. Molecules forming hydrogen bonds are hydrophilic in nature and this increases the organization of surrounding water molecules.

**Structural stability**: Both hydrogen bonding and Van dar Waals forces play a major role in maintaining the internal and external molecular surfaces. They also maintain molecular conformation during self-assembly.

## Methods of preparations of aquasomes

### 1. Formation of an inorganic core

It includes the construction of a ceramic core and the process hinge on upon the materials used. Examples of ceramic cores commonly used are: calcium phosphate and diamond.

### a Preparation of nanocrystalline tin oxide core ceramic

It involves the process were 3 inches' diameter of highly purified tin is sputtered by a blend of argon and oxygen under high pressure. Ultrafine particles formed in the gaseous phase are collected on Cu tubes cooled by nitrogen under 77°C.

### b Self-assembled nanocrystalline brushite (calcium phosphate-dihydrate)

These can be manufactured by colloidal precipitation and sonication by reacting solution of $Na_2HSO_4$ and $CaCl_2$.

### c Nanocrystalline carbon ceramic, diamond particles

After ultracleaning and sonication, nanocrystalline carbon ceramic, diamond particles can be employed for core synthesis.

### 2 Covering of core with polyhydroxy oligomer

In this process, the ceramic cores are layered with polyhydroxyl oligomer. Carbohydrate is added into an aqueous dispersion of the cores under sonication to coat the ceramic cores. Then, it is lyophilized to elevate an irretrievable adsorption of carbohydrate on the ceramic surface. Unadsorbed carbohydrate is removed by centrifugation. Some of the coating materials commonly

used includes: citrate, cellobiose, sucrose, trehalose, and pyridoxal-5-phosphate.

**3 Charging of the drug of choice to the core**

The drug is loaded to the coated particles by adsorption which involves the dispersing of the coated particles into a solution of drug prepared in pH buffer. This dispersion is either lyophilized or reserved overnight at minimum temperature to form drug-laden aquasomes.

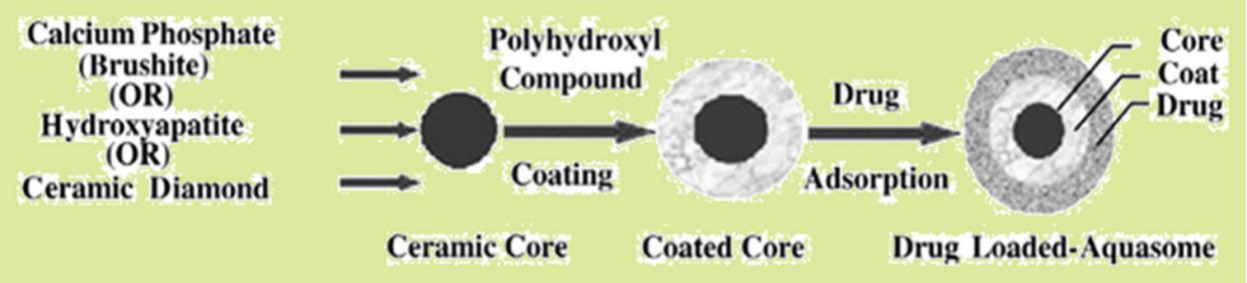


**Figure 5: Method of preparation of aquasomes (Sharme *et al*., 1997).**

**Application of aquasomes.**

1. In insulin delivery: Aquasomes were prepared using calcium phosphate ceramic core for the parenteral delivery of insulin. Kanitakis *et al* (2005), coated the cores with cellobiose, trehalose and pyridoxal-5-phosphate, and the drug was loaded to these particles by adsorption method. He discovered that the drug coated with pyridoxal-5-phosphate was more effective in reducing blood glucose levels in albino rats. This could be attributed to the high degree of molecular preservation and slow release of drug.

2. Oral delivery of enzymes

3. As oxygen carrier

4. Antigen delivery

5. Delivery of gene: Aquasomes maintain the structural integrity of gene segment.

**Advantages of aquasomes**

1. They conserve structural properties of drug particles

2. They evade the reticuloendothelial system

3. They display colloidal characteristics

4. They contain biodegradable nanoparticles

5. The drug release can be controlled by altering the surface structure.

**Ethosomes**

They are noninvasive delivery carriers that enable drugs to reach the deep skin layers and the systemic circulation. They are vesicular carriers consisting of hydro-alcoholic phospholipids in high concentration. The phospholipids comes in various forms such as phosphatidyl choline (PC), hydrogenated PC, Phosphatidic acid (PA), phosphatidyl serine (PS), phosphatidyl inositol (PI). In ethosome preparation, cholesterol can be used in the range of 0.5 – 10 % w/w in order to increase stability (Long *et al*., 2012).

**Table 3: Additives employed in ethosome formulation.**

| Class | Example | Uses |
|---|---|---|
| Phospholipid | Soya phosphatidyl choline | Vesicle forming component |
| Polyglycerol | Propylene glycol | As a skin penetration enhancer |
| Alcohol | Ethanol | Provides softness for vesicle membrane |
| Cholesterol | Cholesterol | Provides stability for vesicle membrane |
| Dye | Rhodamine-123 | For characterization study |
| Vehicle | Carbopol D934 | As a gel former |

**Methods of preparation of ethosomes**

1. **Cold method**: Phospholipids, drug and other lipid materials are dissolved in ethanol by vigorous stirring with a mixer and covered in a vessel at room temperature. During stirring, propylene glycol is added. The mixture is heated to 30 ºC in a water bath. The water heated to 30 ºC in a separate vessel, is added to the mixture which is stirred for 5 min in a covered vessel.

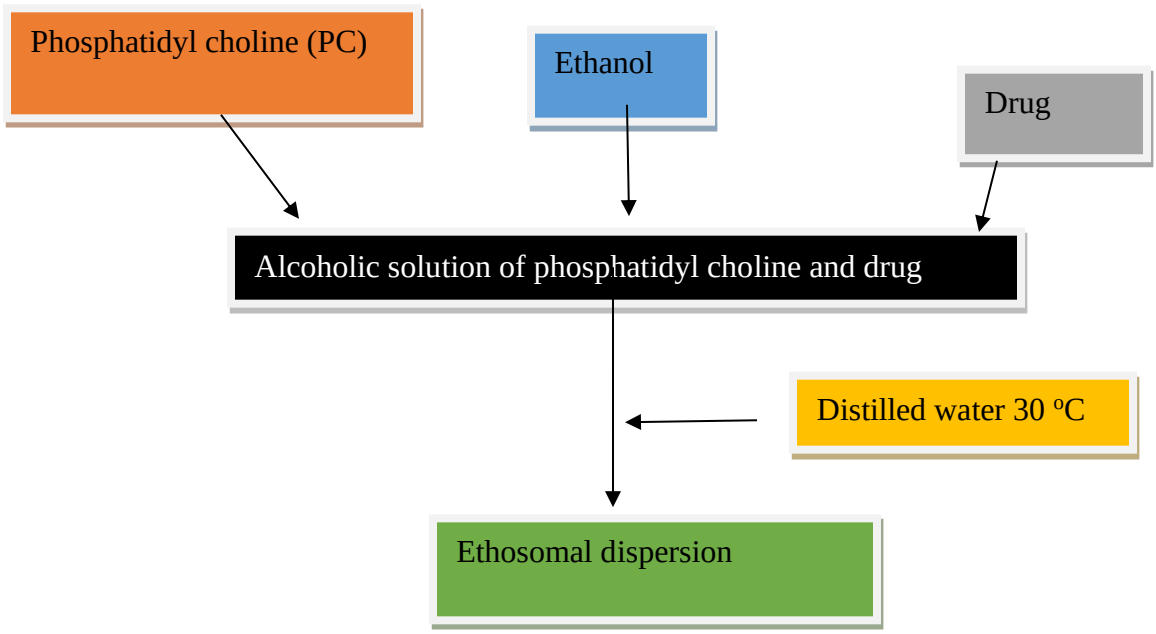


**Figure 6: Preparation of ethosomes by cold method**

**Hot method**: Phospholipid is dispersed in water by heating in a water bath at 40 °C to form a colloidal solution. Ethanol and propylene glycol are mixed and heated to 40 °C in a separate vessel. The organic phase is added to the aqueous phase, and the drug is dissolved in the appropriate solvent (Sunil *et al.*, 2005).

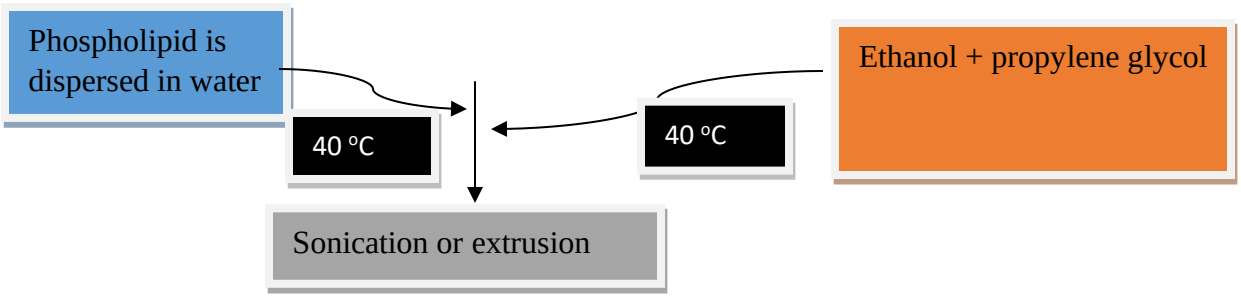


**Figure 7: Preparation of ethosomes by hot method**

Skin delivery from the ethosomal system occurs in 2 phases:

a) Ethanol effect

b) Ethosomes effect

**Ethanol effect**: Ethanol acts as a penetration enhancer via the skin. It penetrates into intracellular lipids and increases the cell membrane fluidity.

**Ethosome effect**: Ethosomes permeate very easily inside the deep skin layers due to the increased lipid fluidity caused by ethanol.

**Characterization of ethosomes**

1. **Vesicle shape**: This is achieved by using the transmission electron microscopy (TEM) and scanning electronic microscopy (SEM).

2. **Vesicle size and zeta potential**: This is achieved using the dynamic light scattering (DLS) and photon correlation spectroscopy.

3. **Entrapment efficiency**: This is achieved using the ultra-centrifugation at 20,000 rpm for 90 min at 40 °C. The sediments and supernatant liquids are separated using methanol to lyse them.

4. **Penetration and permeation studies**: This is achieved using confocal laser scanning microscopy (CLSM).

5. **Surface tension**: This is achieved using the Du Nouy ring tensiometer.

6. **Drug content**: This is achieved using the high performance liquid chromatography.

**Table 4: Applications of ethosomes (Gao *et al.*, 2004)**

| Drug | Results |
|---|---|
| NSAIDs (Diclofenac) | Selective delivery of drug to desired site for an extended period of time |

| | |
|---|---|
| Acyclovir | Increase skin permeation, improved biological activity and pharmacodynamics profile |
| Insulin | Significant decrease in blood glucose level |
| DNA | Better expression of genes, selective targeting to dermal cells. |
| Antibiotic | Improved skin deposition |
| Anti-HIV agents | Improved in biological activity, improved in pharmacodynamics profile. |

**Steps for drug carriers' production**

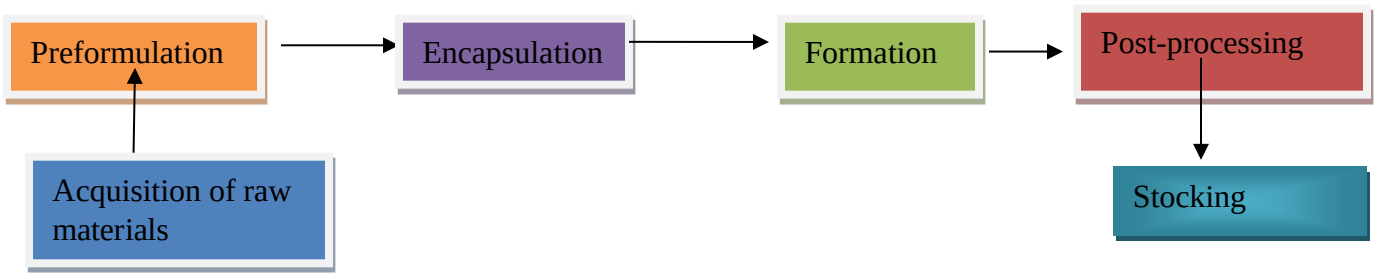


**Figure 8: Summary of the description of the main steps for drug carriers' production (Gao *et al*., 2004).**

1. **Acquisition of raw materials**: These include drugs, surfactants, gases, water and organic solvents. Preformulation of the carrier takes place, followed by incorporation of a drug or active molecule, the formation of a protective barrier and finally the closure or hardening of the structure.

2. **Post-processing steps**: This is characterized by the addition of elements on the surface of the carriers such as antibodies, antigens and peptides. The post-processing steps could be sonication, extrusion and filtration. The essence of the post-processing steps is to decrease mean size, homogenize drug carrier population and obtain a narrow distribution.

3. **Stocking**: The produced samples are subsequently stocked in proper conditions such as freezing, refrigerating or kept at room temperature (Chrais *et al*., 2001)

**Conclusion.**

With the emergence of novel drug carriers, some of the challenges associated with conventional drug delivery system such as poor solubility, low encapsulation efficiency and toxicity will be drastically reduced. A drug carrier has the ability to target a drug candidate which is highly potent, with low therapeutic indication to its required diseased state. The efficacy of any drug carrier depends on its ability to deliver the drug molecule to the targeted site over a prolonged period of time, and show reduction in drug toxicity. In any drug carrier formulation, the drug concentration, drug to lipid ratio and encapsulation efficiency must be put into consideration.

**Declaration of interest**

The authors declare no conflict of interest.

## REFERENCES


Aberturas MR., Guzman M. (2000). Biodegradable nanoparticles as a delivery system for cyclosporine: preparation and characterization. J Microencapsul, 17, 599-614.

Abra, R.M., Hunt, C.A. (1981). Liposome disposition in vivo. III. Dose and vesicle size effects, Biochim Biophys Acta, 666, 493-503.

Allen TM (1997). Liposomes. Opportunities in drug delivery. Drugs, 54 (4), 8–14.

Akihiko KT (2002). Pulsatile drug release control using hydrogels, Advanced Drug Delivery Reviews, 54, 53–77.

Alivisatos P. (2004). The use of nanocrystals in biological detection. Nat Biotechnol, 22, 47–52.

Al-Jamal K.T, Nerl H, M¨uller K. (2011). "Cellular uptake mechanisms of functionalised multi-walled carbon nanotubes by 3D electron tomography imaging," *Nanoscale*, 3 (6), 2627–2635.

Amidon, G.L., Lennernas, H., Shah, V.P., Crison, J.R. (1995). A theoretical basis for a biopharmaceutic drug classification: the correlation of in vitro drug product dissolution and *in vivo* bioavailability. Pharm Res, 12, 413-420.

Barbara K, Maria B. (2001). Dendrimers: properties and applications. Acta Biochimica Polonica, 48, 199–208

Bekyarova E, Ni Malarkey Y.E. (2005). "Applications of carbon nanotubes in biotechnology and biomedicine," Journal of Biomedical Nanotechnology, 1 (1), 3–17.

Betancor L. and Luckarift HR. (2008). Trends Biotechnol, 4, 265-66

Blanco-Prieto M, Decaroz C, Renedo M, Kunkova J, Gamazo C. (2002). *In-vitro* evaluation of gentamicin released from microparticles. Int. J. Pharm, 242 (2), 203-206.

Bosman AW, Janssen HM, Meijer EW (1999). About dendrimers: Structure, physical properties, and applications. Chemical Reviews, 99, 1665-1688.

Brownlee M., Cerami A. (1979). A glucose-controlled insulin delivery system: semisynthetic insulin bound to lectin. Science, 206, 1190–1191.

Catarina PR., Ronald JN., Antonio JR. (2006). Nano capsulation. Method of preparation of drug – loaded polymeric nanoparticles: Nanotechnology, Biology and medicine, 2, 8-21.

Chandramani P, Foram U, Shashibhal M (2019). Applications of targeted nano-drugs and delivery systems. Micro and nanotechnologies, 35-67.

Chan WCW, Maxwell DJ, Gao X, Bailey RE, Han M, Nie S. (2002). Luminescent

quantum dots for multiplexed biological detection and imaging. Curr Opin Biotechnol, 13(1), 40–46

Cheng Y., Wang J., Rao T., He X., Xu T (2008). Pharmaceutical applications of dendrimers: promising nanocarriers for drug delivery. Front Biosci, 13, 1447-71.

Chiannil kulchai N; Ammoury N; Caillou B, Devissaguet JP, Couvreur P. (1990). Hepatic tissue distribution of doxorubicin-loaded nanoparticles after i.v. administration in reticules arcoma M 5076 metastasis-bearing mice, Cancer Chemotherapy and Pharmacology, 26, 122-6.

Chrai SS, Murari R, Imran A (2001). Liposomes: a review. Bio Pharm, 14 (11), 10–14.

Colton HM, Falls JG, Ni H, Kwanyuen P, Creech D, McNeil E.(2004). Visualization and quantitation of peroxisomes using fluorescent nanocrystals: treatment of rats and monkeys with fibrates and detection in the liver. Toxicol Sci, 80(1), 183–192.

Dezhi Qi. (2018). Extractants used in solvent extraction-separation of rare earths: extraction, mechanism, properties and features. Separation and extraction, 7 (2), 187-387.

DeAssis DN., Mosqueira VC., Vilela JM., Andrade M.S., Cardoso VN. (2008). Release profiles and morphological characterization by atomic force microscopy and photon correlation spectroscopy of 99mTechnetium – fluconazole nanocapsules. Int J Pharm, 349, 152 –160.

Demanuele A, Jevprasesphant R, Penny J, Attwood D (2004). The use of a dendrimer-propranolol prodrug to bypass efflux transporters and enhance oral bioavailability. J Control Release, 95:447-453.

Digge M.S, Moon R, and Gattani,S. (2012). “Applications of carbon nanotubes in drug delivery: a review,” International Journal of Pharm Tech Research, 4 (2), 839–847.

Donaruma, L.G., Turek, A.B. (1985). Macromolecules as drugs and drug as carriers for biologically active material. Ann NY Acad Sci, 446, 105-115.

Drumheller P., Hubbell Densely J.A. (1995). Crosslinked polymer networks of PEG in trimethylolpropane triacrylate for cell adhesion-resistant surfaces, J. Biomed. Mater Res, 29, 201–215.

Elhissi A, Ahmed W, Hassan I, Dhanak, V and D’Emanuele A. (2012) “Carbon nanotubes in cancer therapy and drug delivery,” Journal of Drug Delivery, 5, 12-20

Elizabeth P, Viridiana G, Gerardo L, Zarda U, Nestor M, David Q.(2015). Nanoscale, fabrication, optimization, scale up and biological aspects of pharmaceuticals. Nanotechnology, 9, 51-83.

El-Sheikh A.H and Sweileh J.A. (2011). “Recent applications of carbon nanotubes in solid phase extraction and preconcentration: a review,” Jordan Journal of Chemistry, 6 (1), 1-16.

Ende M., Mikos A.G. (1997). Diffusion controlled delivery of proteins from hydrogels and other hydrophilic systems, in: L.M. Sanders, R.W. Hendren (Eds.), Protein Delivery: Physical Systems, Plenum, New York, 3, 139–165.

Ezhilarasi, P. N.; Karthik, P.; Chhanwal, N.;and Haramakrishnan, C. (2012). "Nanoencapsulation Techniques for Food Bioactive Components: A Review". Food and Bioprocess Technology, 6 (3), 628–647

Frechet JMJ, Donald A. (2002). Dendrimers and other dendritic polymers. 1st ed. New York: Wiley Interscience, 4, 145-158.

Fu HL., Cheng SX., Zhang XZ., Zhuo RX (2007). Dendrimers/DNA complexes encapsulated in a water soluble polymer and supported on fast degrading star poly (DL-lactide) for localized gene delivery. J Control Release. 124:181-8.

Gabizon A, Goren D, Cohen R, Barenholz Y (1998). Development of liposomal anthracyclines: from basics to clinical applications. J Control Release, 53, 275–279.

Galindo-Rodriguez S, Allemann E, Fessi H, and Doelker E. (2004). Physico-chemical Parameters Associated with Nanoparticle Formation in the Salting-Out, Emulsification- Diffusion, and Nano precipitation Methods, Pharmaceutical Research, 2, 1428–439.

Gajbhiye V, Vijayaraj P, Sharma A, Agarwal A, Asthana A, Jain NK (2008). Dendrimeric nanoarchitectures mediated transdermal and oral delivery of bioactives. Indian J Pharm Sci, 70, 431-439.

Gaosheng Wei, Yusong Liu, Xinxin Zhang, Fan Yu, Xiaoze Du (2011). Thermal conductivities study on silica aerogel and its composite insulation Materials, International Journal of Heat and Mass Transfer, 54, 2355–2366.

Gao X, Cui Y, Levenson RM, Chung LW, Nie S. (2004). In vivo cancer targeting and imaging with semiconductor quantum dots. Nat Biotechnol, 22, 969–976.

Graves RA, Pamujul S, Moiseyer R, Freeman T, Bostanian L, Mandal T (2004). Effect of different ratios of high and low molecular weight PLGA blend on the characteristics of pentamidine microcapsules. Int. J. Pharm. 270, 251-262.

Gregoriadis, G. Liposomes, In Gregoriadis, G. (1979). Drug Carriers in Biology and Medicine. Academic Press, New York. P. 287- 341

Goldberg M., Langer R., Jia X. (2007). Nanostructured materials for applications in drug delivery and tissue engineering. *J* Biomater Sci Polym, 18, 241-68.

Henglein A, Giersig M. (1999). Formation of colloidal silver nanoparticles: capping action of citrate. J Phys Chem, 103, 9533–9539.

Hirlekar R, Yamagar M, Garse, H Vij, M and Kadam V. (2009). “Carbon nanotubes and its applications: a review,” Asian Journal of Pharmaceutical and Clinical Research, 2 (4), 17-27.

Hoffman A.S. (2002). Hydrogels for biomedical applications. Advanced Drug Delivery Reviews. 43: 3–12.

Hovgaard L, Brøndsted H. (1995) Dextran hydrogels for colon specific drug delivery. J. Controlled Release, 36, 159–166.

Ishihara K., M. Kobayashi, N. Ishimaru I. (1984). Shinohara Glucose induced permeation control of insulin through a complex membrane consisting of immobilized glucose oxidase and a poly (amine). Polym. J, 16, 625–631.

Jaiswal J., Gupta SK., Kreuter J. (2004). Preparation of biodegradable cyclosporine nanoparticles by high-pressure emulsification solvent evaporation process. J Control Release, 96, 78-92.

Jiang L, Liu T, He H. (2012) "Adsorption behavior of pazufloxacin mesilate on amino-functionalized carbon nanotubes,"*Journal of* Nanoscience and Nanotechnology, 12, 1–9.

Jores K., Mehnert W., Drecusler M., Bunyes H., Johan C., MAder K. (2004). Investigation on the stricter of solid lipid nanopartuicles and oil-loaded solid nanoparticles by photon correlation spectroscopy, fieldflow fractionasition and transmission electron microscopy. J Control Release, 17, 217- 227.

Keemi L, Itamid Z, Zuratul A (2018). Polymer nanoparticles carriers in drug delivery systems: Research trend. Woodhead publishing series in Biomedicals, 217-237.

Kim S.W., Pai, K C.M.. Makino, L.A. Seminoff, D.L. Holmberg, J.M. Gleeson, D.E. Wilson, E.J. (1990). Self- regulated glycosylated insulin delivery. J. Controlled Release, 11, 193–201.

Kreyling WG, Semmler-Behnke M, Moller W. (2006). Health implications of nanoparticles. J Nanopart Res, 8, 543–562.

Lay C.L, Liu J, and Liu Y (2011). "Functionalized carbon nanotubes for anticancer drug delivery," Expert Review of Medical Devices, 8 (5), 561–566.

Lee M, Kim SW. (2005). Polyethylene glycol-conjugated copolymers for plasmid DNA delivery, Pharmaceutical Research, 22, 1-10.

Lemarchand C., Gref R., Passirani C., Garcion E., Petri B., Muller R. (2006). Influence of polysaccharide coating on the interactions of nanoparticles with biological systems. Biomaterials, 27:108-18.

Lim F., Sun A.M. (1980). Microencapsulated islets as bioartificial pancreas. Science, 210, 908–910.

Li R, Wu R, Zhao L, Wu M, Yang L. and H. Zou (2010). "Pglycoprotein antibody functionalized carbon nanotube overcomes the multidrug resistance of human leukemia cells*," ACS Nano*, 4, 1399–1408.

Li Y., Cheng Y., Xu T. (2007). Design, synthesis and potent pharmaceutical applications of glycodendrimers: a mini review. Curr Drug Discov Technol, 4, 246-254.

Iijima S. (1991). "Helical microtubules of graphitic carbon," Nature, 354, 56–58.

Liu Z, Sun X, Nakayama-Ratchford N and Dai H. (2007)."Supramolecular chemistry on water-soluble carbon nanotubes for drug loading and delivery," ACS Nano, 1 (1), 50–56.

Long, Li-xia; Yuan, Xu-bo; Chang, Jiang; Zhang, Zhi-hua; Gu, Ming-qi; Song, Tian-Tian; Xing, Ying; Yuan, Xiao-yan. (2012). "Self-assembly of polylactic acid and cholesterol-modified dextran into hollow nanocapsules". Carbohydrate Polymers, 87 (4), 2630–7.

Madani Y, Naderi N, Dissanayake O, Tan A, and Seifalian A (2011). "A new era of cancer treatment: carbon nanotubes as drug delivery tools," International Journal of Nanomedicine, 6, 2963–2979.

Mohan raj VJ, Chen Y. (2006). Nanoparticles- a review, Tropical Journal of Pharmaceutical Research, 5, 561-573.

Molpeceres J., Aberturas MR., Guzman M. (2008). Biodegradable nanoparticles as a delivery system for cyclosporine: preparation and characterization. *J* Microencapsul, 17, 599-614.

Mu L, Feng SS. (2003). A novel controlled release formulation for the anticancer drug paclitaxel (Taxol(R): PLGA nanoparticles containing vitamin ETPGS, Journal of Controlled Release, 86, 33-48.

Nagavarma, B V N; Yadav, Hemant K S; Ayaz, A; Vasudha, L S; Shivakumar, H G. (2012). "Different Techniques for Preparation of Polymeric Nanoparticles – A Review" Asian Journal of Pharmaceutical and Clinical Research, 5 (3*)*, 16–23.

Pangi Z., Beletsi A., Evangelatos K. (2003). PEG-ylated nanoparticles for biological and pharmaceutical application. Adv Drug Del Rev, 24, 403- 419.

Park SK, Kim KD, Kim HT. (2002). Preparation of silica nanoparticles: determination of the optimal synthesis conditions for small and uniform particles. Coll Surf, 197, 7–17.

Qiu S, Dong J, Chen G. (1999). Preparation of Cu nanoparticles from water-in-oil micro emulsions. J Coll Interf Sci,, 216, 230–234.

Wuister SF, Donega CM, Meijerink A. (2004). Influence of thiol capping on the exciton luminescence and decay kinetics of CdTe and CdSe quantum dots. J Phys Chem B, 108, 17393–17397.

Sakthivel T, Florence AT (2003). Adsorption of amphipathic dendrons on polystyrene nanoparticles. Int J Pharm, 254, 23-26.

Scholes PD., Coombes AG., Illum L., Davis SS., Wats JF., Ustariz C., Vert M., Davies MC. (1999). Detection and determination of surface levels of poloxamer and PVA surfactant on biodegradable nanospheres using SSIMS and XPS. J control Release, 59, 261 - 278.

Sharma, A., Sharma, U.S (1997). Liposomes in drug delivery: progress and limitations. Int J Pharm, 154, 123-140.

Tomalia DA (2004). Birth of a new macromolecular architecture: Dendrimers as quantized building blocks for nanoscale synthetic organic chemistry. Aldrichimica Acta, 37 (2), 39-57.

Tirrell, D.A., Heath, T.D., Colley, C.M., Ryman, B.E. New aspects of liposomes, *Biochim Biophys Acta*, 1976, 457: 259.

Uchida T, Yoshida K, Ninomiya A, Goto S. (1995). Optimization of preparative conditions for polylactide (PLA) microspheres containing ovalbumin. *Chem. Pharm. Bull*, 43 (9), 1569-1573.

Vandamme TF, Brobeck L (2005). Poly (amidoamine) dendrimers as ophthalmic vehicles for ocular delivery of pilocarpine nitrate and tropicamide. *J Control Release*, 102, 23-38.

Vartholomeos, P; Fruchard, M.; Ferreira, A.; Mavroidis, C. (2011). *"MRI-Guided N*anorobotic Systems for Therapeutic and Diagnostic Applications". Annu Rev Biomed Eng., 13, 157–84

Vargas A., Pegaz B., Devefve E., Konan-Kouakou Y., Lange N., Ballini JP. (2004). Improved photodynamic activity of porphyrin loaded into nanoparticles: an *in vivo* evaluation using chick embryos. Int J Pharm, 286, 131- 45.

Zhang Q, Shen Z, Nagai T. (2001). Prolonged hypoglycemic effect of insulin-loaded poly butyl cyano acrylate nanoparticles after pulmonary administration to normal rats, International Journal of Pharmaceutics, 218, 75-80.

Zonghua L, Yanpeng J, Yifei W, Changren Z, Ziyong Z. (2008). Polysaccharides-based nanoparticles as drug delivery systems, Advanced Drug Delivery Reviews, 60, 1650–1662.