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Liposomes as Drug Delivery Systems: Properties and Applications

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ABSTRACT

Liposomes are polymeric nanoparticles used for drug delivery due to their unique properties. Lipisomes can encapsulate both hydrophobic and hydrophilic drug. liposomes deliver the drugs into cells by fusion or endocytosis mechanisms. In the last few decades, liposomes have been considered as ideal models for mimic biological membranes and also they are suitable carriers for drugs, diagnostics, vaccines, and other bioactive agents. Drugs can be distributed non-specifically throughout the body, lead to death of normal and malignant cells. Entrapment of drugs into liposomes results in increasing circulation life time, protection from the metabolic degradation, enhancement of deposition in the infected tissues and decreased uptake in the kidney, myocardium and brain. There are now some liposomal formulations of conventional drugs that have received clinical approval.

Keywords: liposomes, drug delivery, nanoparticles

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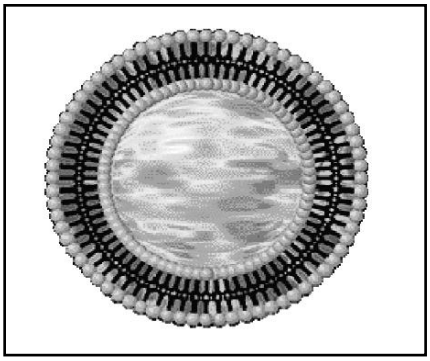
INTRODUCTION

Liposomes are spherical vesicles with concentric phospholipid bilayers [1] that are formed spontaneously in aqueous solution [2]. The word liposome comes from two Greek words, lipos (fat) and soma (body or structure) [3, 4]. Lipid bilayered membrane encloses an aqueous core and hydrophilic drugs may get entrapped in the central aqueous core of the vesicles while lipophilic drugs are entrapped within the bilayered membrane [5]. A schematic structure of a liposome is shown in fig (1) [5]. They were firstly introduced by British hematologist, Bangham and his students in the mid-1960s. Bangham was found phospholipids combined with water immediately formed a sphere because one end of each molecule is water soluble, while the opposite end is water insoluble [6]. Liposomes were introduced as drug-delivery vehicles in the 1970s [7].

Fig (1): Structure of liposome [5]



Liposomes can be prepared from natural phospholipids (egg or soya) or synthetic lipids such as DOPE (dioleoylphosphatidyl ethanolamine) [1, 8]. Size, charge and surface properties of liposomes can be altered by adding new ingredients to the lipid mixture during liposome preparation [9]. Encapsulated drugs in liposomes can be transported without rapid degradation and minimum side effects to the recipients [10]. Liposomes have many advantages such as, biocompatibility and biodegradability, prolonging release of active pharmaceutical agents [11], ability to entrap both lipophilic and hydrophilic drugs [8], protecting encapsulated agents from metabolic processes, increased circulation life times of drug [12, 13], nontoxicity and enhancement of drug penetration [14, 15]. Liposomes are used to administer drugs by several routes such as topical, oral and parenteral [1] and have many applications in the fields of immunology, tumor therapy, vaccine adjuvant, antimicrobial therapy, gene therapy and delivery of radiopharmaceuticals for diagnostic imaging [8, 10, 16]. Also liposomes can be used for cosmetic delivery to skin, delivery of dyes to textiles, enzymes and nutritional supplements in foods, pesticides on plants and can be used as biosensors [6, 17].

History

The history of liposomes can be divided into three periods: genesis, middle age and modern era [5].

Genesis (1968-75): The physiochemical characterizations of liposomes have been investigated. In this period liposomes were used to study the nature of biological membrane and thin lipid film hydration method was developed to prepare liposomes.

Middle age (1975 – 85): In this period, advantages, stability and interaction characteristic, liposomes, physico-chemical properties of liposomes, their interaction with the cells and their behavior within the body were studied. Also, various methods for the preparation of liposomes discovered.

Modern era (1985 onwards): Today, liposomes are used in different scientific fields such as biophysics (properties of cell membranes and channels), mathematics, biochemistry (function of membrane proteins), theoretical physics (topology of two-dimensional surfaces floating in a three dimensional continuum) and biology (excretion, cell function, signaling, gene delivery and function).

Disadvantages

Many researchers have studied and worked on liposomes, but small numbers of liposomal products have been approved to be used in human. This may be due to many reasons such as: High cost of liposome production especially in large scales, toxicity of some liposomal formulations, relative short half life, instability, low solubility, low entrapment of molecules and compounds into vesicles and sometimes phospholipid undergoes oxidation and hydrolysis [18, 19, 20].

Mechanism of formation

When phospholipids are placed in water and sufficient energy is provided from sonication, heating, homogenization and ect, results in the arrangement of the lipids and formation of bilayer vesicles (fig 2) [21]. The phenomenon can be result of the critical micelle concentration (CMC) of phospholipids in water. The CMC may be a defined as the concentration of lipid in water, above which the lipid forms micelles or bilayer structures rather than remaining in solution as monomers [5].

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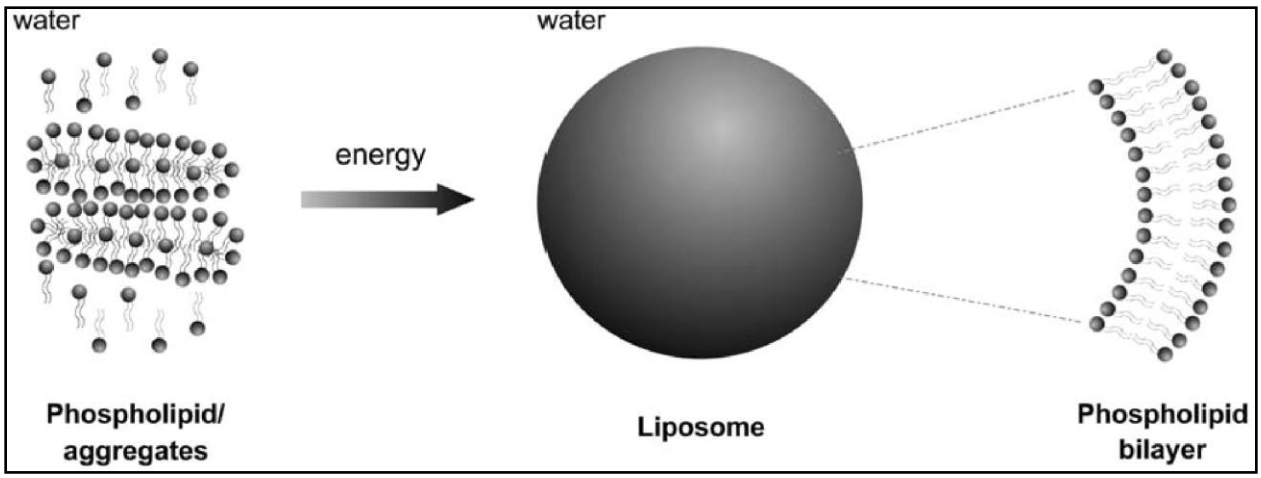

Fig (2): The mechanism of formation of liposomes [21]

Classification

Liposomes may be classified on the basis of their structure, method of preparation and their composition and applications.

Structural classification

Multilamellar liposomes (MLV) consist of several (up to 14) lipid layers separated from one another by a layer of aqueous solution and usually are in size range of 500 nm to 10 µm. MLV liposomes are prepare by using the aqueous hydration of dried lipid films and are more suitable for the encapsulation of a variety of lipophilic materials. Small unilamellar vesicles (SUV) are surrounded by a single lipid layer and have only 25–50 nm. Large unilamellar vesicles (LUV) are also surrounded by a single lipid layer and are usually larger than 50 nm. LUV liposomes are suitable delivery systems for nucleic acid drugs. Giant vesicles (GVs) are very large, from 10 µm to 1000 µm and are prepare by the process of electroformation. GV liposomes are ideal models of cell membranes and can be used as microscale bioreactors [3, 21, 22, 23, 24].

Classification based on method of preparation

Table (1): Classification of liposome based on the method of preparation

Vesicle Type	Preparation Method
VET	Vesicle prepared by extrusion technique
REV	Single or oligo lamellar vesicle made by reverse phase evaporation method
SPLV	Stable pluri lamellar vesicle
DR V	Dehydration- Rehydration method
FATMLV	Frozen and thawed multi lamellar vesicle
MLV-REV	Multi lamellar vesicle made by reverse phase evaporation method

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Classification based on composition and application

Table (2): Classification of liposome based on their composition and application.

Type of Liposome	Abbreviation	Composition
Fusogenic liposome	RSVE	Reconstituted sendai virus envelops
Long circulatory liposome	LCL	Neutral high temp, cholesterol, and 5- 10% PEG, DSP
pH sensitive liposomes	-	Phospholipids such as PER or DOPE with either CHEMS or OA
Conventional liposome	CL	Neutral or negatively charge phospholipids and cholesterol
Cationic liposome	-	Cationic lipid with DOPE
Immuno liposome	IL	With attached monoclonal antibody or recognition sequences

Methods of preparation

Thin film method: In this method, liposomes are prepared by hydrating the thin lipid film in an organic solvent and then organic solvent is removed under vacuum. After completely removing the solvent, the solid lipid mixture is hydrated by aqueous buffer. The lipids spontaneously swell and hydrate to form liposome. This method has low encapsulation efficiency [25].

Reverse Phase Evaporation*:* In this method**,** the lipid mixture is added to a round bottom flask and the organic solvent (diethyl ether and isopropyl ether) is removed under reduced pressure by a rotary evaporator. The liposomes are formed when residual solvent is removed by continued rotary evaporation under reduced pressure. With this method, high encapsulation efficiency (up to 65%) can be obtained in a medium of low ionic strength for example 0.01M NaCl. This method can encapsulate large macromolecules with high efficiency. Disadvantage of the method is the exposure of the encapsulated materials to organic solvents [13, 22, 25].

Freeze-thaw method: In FT method, liposomes formed by the film method are vortexed with the solute to be entrapped until the entire film is suspended.Then liposomes are frozen in dry ice-ethanol (-80 ˚C) or in liquid nitrogen and are thawed and then vortexed again. The freeze and thawing cycles are repeated. This method is used widely for encapsulation of protein [25, 26, 27].

Ultrasonic Method: The method is used for the preparation of SUV liposomes. Ultrasonication of an aqueous dispersion of phospholipids is done by two types of sonicators; probe sonicators or bath sonicators for the small volumes and large volumes, respectively [25].

Calcium induced fusion method: The method is used for preparation of LUV liposomes from acidic phospholipids. Calcium is added to SUV liposomes that induce fusion and cause formation of multilamellar vesicle. The addition of EDTA to the preparations results in the formation of LUV liposomes. The advantage of calcium induced fusion method is that

macromolecules can be encapsulated , while their disadvantages is that LUV liposomes can only be obtained from acidic phospholipids [22].

Type of phospholipids

Table (3): Phospholipids which are used for the preparation of liposome [5, 28, 29].

Type of phospholipids	Phospholipids
Neutral phospholipids	egg lecithin, soya lecithin, cholesterol, sphingomyelin, phosphatidylethanolamine and phosphatidylcholine
Positively charged phospholipids	1, 2-dihexadecyl-N,N-dimethyl–N-trimethyl amine methyl ethanol amine and stearyl amine (SA)
Negatively charged phospholipids	dipalmitoyl phosphatidylcholine, dipalmitoyl phosphatidyl acid (DDPA), distearoyl phosphatidyl choline (DSPC), dioleoyl phosphatidyl choline (DOPC) and dicetyl phosphate (DCP)

Several sources of lipids may be used for preparation of lipoomes. Przeworska *et al* in 2001 prepared liposomes using resorcinolic lipids isolated from *Anacardium occidentale* nut oil extract and entraped of patent blue violet (PBV) as a marker in the liposome. Results showed resorcinolic lipid liposomes (AR-osomes) highly entrapped the marker and induced their size stability [30]. Gupta *et al* in 2008 prepared liposomes from natural lipids derived from *E.coli* that are economically compared to the synthetic lipids. The results showed that these bacterial liposomes had potential applications in delivery of aqueous molecules to cancer cells [31].

Characterization

After preparation, liposomes should be characterized using physical, chemical and biological methods. Table (4) shows the methods of evaluation of liposomes [25].

Table (4): Different methods of characterization of liposomes.

Characterization parameters	Instrument for analysis
Drug concentration	Assay method
Phospholipids hydrolysis	HPLC/ TLC
Drug release	Diffuse cell/ dialysis
Vesicle shape, and surface morphology	TEM and SEM
Phase behavior	DSC, freeze fracture electron microscopy
Vesicle size and size distribution	Dynamic light scattering ,TEM
Phospholipids per oxidation	UV observance
Lamellarity	P31NMR
Pyrogenicity	Rabbit fever response
pH	PH meter
Sterility	Aerobic/anaerobic culture
Animal toxicity	Monitoring survival rats
Electrical surface potential and surface pH	Zeta potential measurement and pH sensitive probes

Triton X-100, sodium chlorate, sodium dodecyl sulphate, chloroform, octyl-β-glucoside, methanol, bile salts and acidified isopropanol can be used to extract the encapsulated drugs from liposome [26, 32, 33, 34, 35].

Factors affecting the properties of liposomes

Temperature of solvent evaporation is an important factor in preparation of liposome. In a study, cyproterone acetare (CA) was loaded in liposomes using film method. The data showed that the best temperature for solvent evaporation was 40˚C and also optimum loading was at phophatidylcholine (PC): cholesterol/drug ratio of 1:2:0.5 with 74 ± 6.11% loading efficiency [8]. Begum *et al* reported there was a significant decrease in encapsulation of flurbiprofen in liposome that stored at room temperature and 37±2 ºC, while there was no significant change in flurbiprofen encapsulation for the formulations stored at -20 ºC and 4 ºC [36].

In a study, incorporation efficiency of essential oil of *Anethi fructus* was evaluated in different liposomal formulation and observed decrease oil content with the increasing of cholesterol content [37]. Membrane permeability is regulated by the lipid composition. The EPC/chol formulations lost half of the entrapped doxorubicin within 1 h while doxorubicin does not significantly leak from DSPC/chol liposomes over a period of 24 h [38].

Jonathan *et al* in 1999 studied factors affecting the size distribution of liposomes produced by freeze-thawing extrusion. The results of the study indicated that the average size of 2µm MLV liposomes composed of egg PC was increased when cholesterol was added in the bilayers. Moreover dispersion of liposomes in NaCl solution increased the diameter following freez- thawing in comparison to aqueous dispersion. NaCl may cause aggregation and fusion. But poloxamer P338 and P404 inhibited the size increase in during freeze-thawing for liposomes dispersed in 1.0 NaCl and it has suggested that poloxamer may prevent the aggregation and fusion [39].

Ramana *et al* in 2010 reported that the best encapsulation of nevirapine in liposome was observed when using egg phosholipid to cholesterol ratio of 9:1 at physiological pH. It is believed that the faster drug release in the acidic and basic media may cause the hydrolysis of the carrier [40].

In a study, it was reported that egg leicthin, cholesterol and charged lipid with ratio of 7:2:1 used in the preparation of liposomes can be promote antibody production [28]. Researchers demonstrated that cholesterol may decrease the permeability of negative, neutral and positively charged membranes to Na+, K+, Cl- and glucose. Also cholesterol stabilized the membranes against temperature changes [25].

Egg lecithin or phosphatidylcholine is an important lipid in preparation of liposome that may promote adjuvant activity in contrast to phosphatidyl inositol, phosphatidyl glycerol and phosphatidic acid. phosphatidyl choline is a very poor antigenicity. Another study, has

suggested that sphingomyelin, used in preparation of liposme, is elicited a more sever immune response to incorporated antigen than liposomes from phsophatidylcholine. Also liposomes composed of dipalmitoyl phosphatidyl-choline (DPPC) and distearyl phosphatidylcholine (DSPC) are more effective immunogens in comparison with liposome composed of egg lecithin [28].
It has been shown that cryoprotective agents such as trehalose, sucrose, mannitol, dimethylsulphoxide and glycerol protect phospholipid bilayers from damage during freeze-drying and freeze-thawing. The protection mechanism by the sugars is to form an amorphous matrix during freezing and exhibiting a low molecular mobility after drying [39, 41].

Nallamothu *et al* in 2006 investigated the effect of cholesterol content of formulation on *in vitro* release of combretastatin A4 from the liposomes and the results showed that at 30% cholesterol level, there was no drug release within 48 h [42].

Charge of liposome is another parameter that affects the release of agents encapsulated. Holovati *et al* in 2008 investigated delivery of trehalose into red blood cell (RBC). The results showed that negatively charged liposomes containing phospholipids in their membrane delivered about 100 times more trehalose than neutral liposomes into RBC [43].

The presence of polyethylene glycol (PEG) on the surface of liposomes improved stability, increased long circulating and intracellular uptake , protected drug from metabolic degradation, reduce protein binding and plasma elimination of liposomes [44, 45]. Chiu *et al* in 2001 showed that compared to PEG 750, DSPE-PEG 2000(1, 2-distearoyl-*sn*-glycero-3-phosphoethanolamine- N-[poly (ethylene glycol) 2000]) was significantly increased the protecting liposome from clearances [44].

D-alpha-tocopheryl polyethylene glycol succinate (TPGS) has been used in preparation of nanoparticles as an emulsifier, absorption enhancer and solubilizer. Zhai *et al* in 2008 used TPGS as a component in liposomal formulations for loading of gossypol as a potential anticancer agent. Results showed TPGS can be used as a component for the preparation of long-circulating liposomal gossypol *in vivo* [46].

Begum *et al* in 2012 prepared flurbiprofen loaded stealth liposomes. They indicated that by increasing alkyl chain length of lipids, drug encapsulation of the liposomes increased. It suggested that longer alkyl chain lipids increase the hydrophobic area in the lipid bilayer membrane [36].

Clinical applications

Some drugs that are successfully loaded in liposome are commercially available. Table (5) shows the liposomal drug formulations.

The outer layer of epidermis blocks drug permeation across the skin and topical treatment of cutaneous diseases represent an important challenge [51]. A way to overcome this problem is encapsulation of drugs in liposomal vesicles. Miconazol nitrate is an antifungal

agent that is widely used, but poor skin penetration of the drug is a problem in the treatment of cutaneous disease by topical application. Agarwal *et al* in 2002 evaluated miconasol nitrate loaded topical liposome *in vitro*. The results showed the entrapment of miconasol nitrate in liposome may facilitate localized delivery of the drug [14]. Incorporation of rifabutin in liposomes resulted in a significant enhancement of activity against *Mycobacterium avium* infection compared to free rifabutin [10].

Table (5): Liposomal drug formulations

Properiatry name	Generic name	Application	Ref
Abelcet®	Amphotericin B	Systemic fungal infections	10 , 38, 48
Doxil™	Doxorubicin	Metastatic ovarian cancer and advanced Kaposi's sarcoma	10, 47, 38
Amphocil®	Amphotericin B		10
L-AMP-LRC-1	Amphotericin B		10
MiKasome	Amikacin	Bacterial infections	3
AmBisome®	Amphotericin B	Antifungal activity	10, 48
DaunoXome	Daunorubicin	Cancers	3
VincaXome	Vincristin	Cancers	3
Annamycin	Annamycin	Kaposi's sarcoma, Breast cancer, Leukemia	3
Myocet™	Doxorubicin	Metastatic breast cancer	38
NX211	Lurtotecan	Ovarian cancer	9
Epaxal	Inactivated hepatitis A virus (strain RGSB)	Hepatitis A	49
Inflexal V	Inactivated hemaglutinine of Influenza virus strains A and B	Influenza	49
Depocyt	Cytarabine	Neoplastic meningitis and lymphomatous meningitis	49
Repithel®	Povidone-iodine	Anti-inflammatory	50

In a study, cyproterone acetare (CA) was loaded into liposome. According to the results, liposomal formulation of the drug may increase percutaneous permeability if compared to conventional formulations [8].

Topical application of liposome vesicles has many advantages over the conventional dosage forms. It has been previously shown that the liposomal gel of Lidocaine HCL may perform therapeutically better effects than the conventional formulation. It has been suggested that the formulated liposomes may be applied onto the skin as gel [52].

Using different enhancers, enhancement permeability of gentamicin sulfate through shed snake skin and liposomal membranes has been studied. The results indicated that direct effect of surfactants on shed snake skin and liposomal membranes is responsible for their enhancing effects [53].

Anticancer drugs encapsulated in liposomes reduced tissue toxicity and the rate of drug metabolism, controled drug release and extended circulation time of drugs [54].

Myocet™ is doxorubicin encapsulated in EPC/chol liposomes. This formulation is cleared from circulation more slowly than the free drugs. Doxil® is a stabilized liposome formulation by which doxurubicine distributes over the body in a lower rate if compared to free drug [38]. Doxil was the first liposomal drug approved by the FDA and has been on the market since 1995 [7].

Celecoxib is a non-steroiddal anti-inflammatory drug (NSAID) that acts on cyclooxygenase-2 (COX-2). Preparation and evaluation of celecoxib loaded liposomes and the study of their in vitro release behavior have been previously investigated. The results of characterization of the vesicles indicated the potential application of celecoxib loaded liposome as carrier system [55]. Fluconazole is a synthetic antifungal agent that is used in the treatment of esophageal, vulvovaginal disorder and is effective against dermatophytoses. Fluconazole has adverse effects such as bloating, nausea, abdominal problems and vomiting. Mitkari *et al* in 2010 have formulated and evaluated a topical liposomal gel of fluconazole. Their investigations showed that the entrapment efficiency was 57.78 – 66.64% and liposomal gel was increased the skin permeation and deposition of fluconazole compared to control. They suggested that by designing a fluconazole liposomal topical preparation, the risk of gastrointestinal side effect may decrease [56].

Zhai *et al* in 2008 have investigated a novel formulation using TPGS (D-alpha-tocopheryl polyethylene glycol succinate) as a lipid component of liposome preparation and incorporated gossypol (GP) in liposomes. The results of their study showed that GP was stably encapsulated in liposomes and exhibited cytotoxicity on cancer cells [46].

Essential oils are natural compounds, volatile and aromatic oily liquids that are commonly extracted from plants. Essential oils have great importance in several fields, from food chemistry to pharmaceutics, although the most of them are instable and poorly soluble in water. It has been reported that using the liposomal encapsulation may overcome these problems [37, 57]. Liolios *et al* in 2009 have studied encapsulation of carvacrol and thymol isolated from the essential oil of *Origanum dictamuns* and their *in vitro* antimicrobial activity. The results of the study showed the antimicrobial activity was enhanced after encapsulation in liposome [57]. Ortan *et al* in 2009 have prepared *Anethum graveolens* loaded liposome and determined the influence of liposome composition on the entrapment of the essential oil. Their results showed that phosphatidyl cholin (PC) and cholesterol influence the encapsulation of the essential oil and incorporation the essential oil in liposome was 98% [37]. The effectiveness and encapsulation of essential oil of *E.camaldulensis* have been previously investigated. They formulated and characterized a liposomal gel loaded with the essential oil. The results of the study concluded that the presence of *E.camadulensis* in liposomes may effectively enhance its stability and entrapped oil may be stable during an extended period of time [58].

Mugabe *et al* in 2006 have investigated the antibacterial activity of liposome-entrapped aminoglycosides against *P.aeruginosa* and their mechanism of action. The results showed that the incorporation of antibiotics into liposomes significantly increased their antibacterial activity against the resistant strains used in the study (MICs of ≥32 versus ≤8 µg/ml). TEM observations showed that liposomes interact with the outer membrane of *P. aeruginosa* that leads membrane disruption [59]. Shafaa *et al* in 2008 reported that negative liposomes encapsulating cephalexin had antibacterial activity against *Staphylococcus aureus* compared to neutral and positive liposomes [29]. Vitas *et al* in 1996 reported that gentamicin incorporated into cationic liposomes containing phosphatidylcholine, 30% cholesterol, and 10% stearylamine eliminated all of the intracellular *Brucella abortus* (4.6 log), while free gentamicin reduced the number of intracellular bacteria (0.3 log) [60].

DNA vaccines can be encapsulated into cationic liposomes. DNA-containing liposomes may suitably deliver their contents directly to antigen-presenting cells (APC) and also protect DNA from nuclease degradation [61].

Targeted liposomes

A number of developments have been used to target of liposome for enhancement of therapeutic of drugs that loaded in liposomes. Among of them, passive targeting and active targeting are used.

Active targeting: For enhancing liposomal drug accumulation in the tissues, the use of targeted liposomes with surface-attached ligands capable of recognizing and binding to cells has been used. Antibodies such as IgG and their fragments are the most widely used targeting ligands for liposomes. Folate receptors (FR) are frequently over expressed in a range of tumor cells, so targeting tumors with folate-modified liposomes is another strategy. Folate receptor (FR) has two glycosylphosphatidylinositol (GPI)-anchored isoforms, α and β. Normal tissues do not express FR. But, FR-α is frequently overexpressed in epithelial cancers including over 90% of ovarian carcinomas. FR-β, is expressed in a non-functional form in placenta and mature neutrophils and in a functional form in activated macrophages, chronic myelogenous leukemias and about 70% of acute myelogenous leukemias. It has been suggested that FR-targeted liposomes are useful in the selective delivery of therapeutic agents to tumor cells both *in vitro* and *in vivo* [62]. Liposomal daunorubicin and doxorubicin have been delivered into tumor cells using FR .This method is used for treatment of acute myelogenous leukaemia [9]. Turk *et al* investigated the distribution of folate-targeted liposomes in a mouse model of ovarian cancer. The results showed that the folate-conjugation of liposomes significantly enhanced their uptake into ovarian cancer cells [63]. Transferrin (Tf) receptors (TfR) are over expressed on the surface of many tumor cells too, so antibodies against TfR can be suitable ligands for liposome targeting to tumor cells [9]. Tf-coupled doxorubicin- loaded liposomes increased binding and toxicity against C6 glioma cells. Also vitamin, growth factor receptors, vasoactive intestinal peptide (VIP) and RGD peptides have been used as targeted liposome to the tumor cells. Using of galactosylated liposomes is another way to target drugs to liver for the treatment of liver tumors or metastases. Cisplatin-loaded liposomes that specifically bind to chondroitin sulphate,

which is over expressed in many tumor cells, have been used for reduction of tumor growth and metastases [9]. Also galactose, mannose and asialofetuin have been used in liposomal systems to target specific cells [64].

Passive targeting: Biocompatible polymers, such as PEG form a protective layer over the liposome surface and decreases liposome recognition by opsonins and caused long circulation of liposomes. Studies have done on the conjugation of Tf to PEG on PEGylated liposomes to combine longevity and targetability for drug delivery into solid tumors [9, 64].

Mechanism of interaction liposome with cell membrane

Drug-loaded liposomes can specifically or nonspecifically adsorb onto the cell surface or fuse with the cell membrane, release their contents into the cell cytoplasm, or can be destabilized by certain cell membrane components when adsorbed on the surface, so drug can enter cell via micropinocytosis. Also liposome can undergo the direct or transfer-protein-mediated exchange of lipid components with the cell membrane or by specific or nonspecific endocytosis that caused the drug release into the cell cytoplasm [9]. Fig (3) shows interaction of liposome with cell membrane.

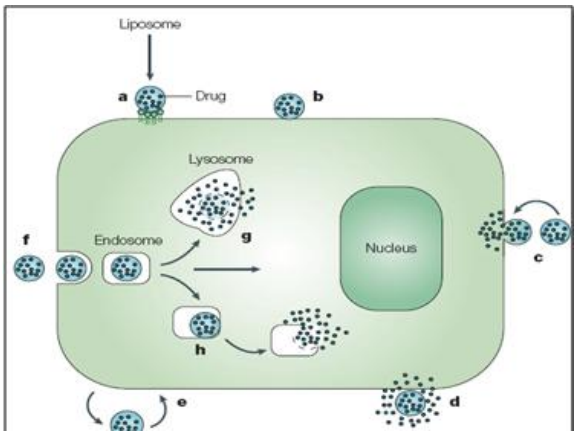

Fig (3): Interaction liposome with cell membrane: Drug-loaded liposomes can specifically (a) or nonspecifically (b) adsorb onto the cell Surface, or fuse with the cell membrane (c), and release their contents into the cell cytoplasm, or can be destabilized by certain cell membrane components when adsorbed on the surface (d) so that the released drug can enter cell via micropinocytosis. Liposome can undergo the direct or transfer-protein-mediated exchange of lipid components with the cell membrane (e) or be subjected to a specific or nonspecific endocytosis (f). In the case of endocytosis, a liposome can be delivered by the endosome into the lysosome (g) or, en route to the lysosome, the liposome can provoke endosome destabilization (h) [9].

New generation of liposomes

Magnetic liposomes: liposomes loaded with a drug and a ferromagnetic are known as magnetic liposomes and are used for targeted drug delivery. Magnetic liposomes loaded with adriamycin were better accumuled in tumor vasculature and resulted in enhanced tumor-growth inhibition [9].

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ATP liposomes: Adenosine triphosphate (ATP) is a source of energy in cells and lack of ATP causes many problems. ATP liposomes protect human endothelial cells from energy failure in a cell culture model. It was shown that ATP-loaded liposomes effectively preserve mechanical properties of the heart under ischaemic conditions in an isolated rat heart model [9].

L**iposomal haemoglobin (haemosomes)**: Liposomal haemoglobin (haemosomes) is used as a blood substitute. PEGylated liposomal haemoglobin is shown to be stable at storage for 1 year even at room temperature and to circulate longer in rabbits. Haemoglobin vesicles suspended in recombinant human serum albumin used to treat haemorrhagic shock in rats, but some side effects were reported for PEG haemosomes such as sensitivity to phagocytosis by human peripheral blood monocytes and macrophages [9].

Virosomes: Virosome is a type of liposome in which the surface of vesicle is modified with fusogenic viral envelope proteins. Virosomes are useful for enhancing tissue targeting. Virosomes were used primarily for the intracellular delivery of drugs and then they became a suitable candidate for development of new vaccines for human and animal immunization. The delivery of protein antigens to the immune system by fusion-acting virosomes was found to be very effective (REF) [9]. Virosomes containing the spike proteins of influenza virus elicit high titers of influenza-specific antibodies. They are also suitable means of the efficient delivery of various antigens and many drugs such as nucleic acids, toxoids and cytotoxic drugs, although there are certain problems associated with their stability and immunogenicity [9]. Fig (4) shows virosome –cell interaction.

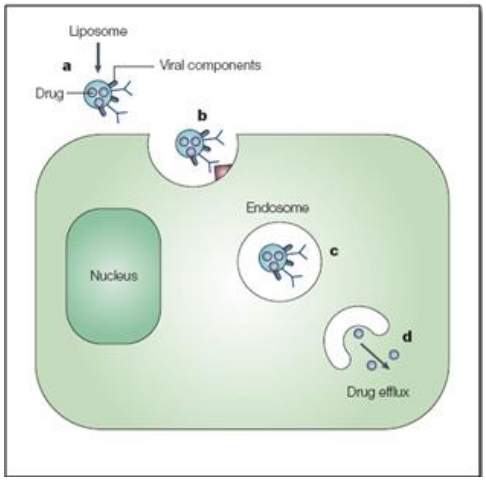

Fig (4): Interaction of virosome with cell membrane [9].

Phytosome: Phytosomes are novel compounds that are composed of phospholipids and natural products from various plants such as *Silybum Marianum, Ginkgo Biloba* and *ginseng*. They are more bioavailable than pure extracts, and improve absorption in intestinal tract. Phytosomes are used in treatment of various acute diseases [65, 66].

Photosome: Replication errors and a variety of mutagens can alter the nucleotide sequence in DNA. Thymine dimers are formed by ultraviolet radiation. Photoreactivation is the process of repairing thymine dimmers, by splitting them apart into separate thymines, with the help of visible light in a photochemical reaction catalyzed by the enzyme photolyase [68]. Photolyase encapsulated liposomes release their contents by photo triggered charges in membrane permeability characteristics. Photosomes found their application in photodynamic therapy [68].

Cryptosome: Cryptosomes are lipid vesicles coated by of phosphotidylcholine and polyoxyethylene derivative of phosphatidyl ethanolamin. Cryptosomes is used as ligand mediated drug delivery [68].

Archaeosomes: Archaeosomes are made from lipids found in archaeal bacteria or synthetic archaeal lipids and more stable against high temperature, alkaline or acidic pH, serum media, phospholipases, oxidative conditions, high pressure and bile salts than conventional liposome. Archaeosomes have applications in drug and gene delivery [19, 69].

Stealth liposome: Stealth liposomes are new generation of liposomal formulations that alter the biodistribution of liposomes, retain the encapsulated drug in circulation for longer period of time and reduce susceptibility to protein induced leakage. Stealth liposomes have greater stability and less opsonized and removed by phagocytes. Polyethylene glycol (PEG) increases circulation time of liposome. Stealth liposomes can be used in tumor therapy [36, 70].

CONCLUSION

Liposomes as drug delivery systems have played a significant role in formulation of drugs to improve their therapeutics, reduce their toxicity, and enhance the efficacy of drugs for the treatment of diseases. Liposomes are currently more applicated in the dermatology, vaccine adjuvant, infective disease, immunology, eye disorders, and in tumor therapy. A number of developments have been used to target liposomes such as attachment of antibodies, proteins, peptides, etc., which lead to drug accumulation at disease sites and reduced distribution to sensitive tissues. Stealth liposomes as a new generation are especially used as carriers for hydrophilic anticancer drugs like doxorubicin.

References

[1] Chetanachan P, Akarachalanon P, Worawirunwong D, Dararutana P, Bangtrakulonoth A, Bunjop M, Kongmuang S. Advanced Materials Research 2008; 55(57): 709-71.
[2] Trommer H, Neubert RHH. J Pharm Pharmaceut Sci 2005; 8(3):494-506.
[3] Kozubek A, Gubernator J, Przeworska E, Stasiuk M. Acta Biochimica Polonica 2000; 43(7): 639-649.
[4] Honarpisheh H, Mallesware Rao VSN, Mozafari MR. Micron 2007; 38: 804-818.
[5] Sharma VK, Mishra DN, Sharma AK, Srivastava B. IJCPR 2010; 1(2): 6- 16.
[6] Kumar KPS, Bhowmik D, Deb L. The Pharma Innovation 2012; 1(1): 29-38.
[7] Lasic DD. Tibtech 1998; 16: 307-321.

[8] Mohammadi- Samani S, Montaseri H, Jamshidnejad M. Iranian J Pharmaceutical Sciences 2009; 5(4): 199-204.
[9] Torchilin VP. Drug Discovery 2005; 4: 145-160.
[10] Goyal P, Goyal K, Kumar SGV, Singh A, Katare OP, Mishra DN. Acta Pharm 2005; 55: 1–25.
[11] Shazly G, Nawroth T, Langguth P. Dissolution Technologyies 2008; 7-10.
[12] Paolo DD, Pastorino F, Brignole C, Marimpietri D, Loi M, Ponzoni M, Pagnan G. Tumori 2008; 94: 245-252.
[13] Dua JS, Rana AC, Bhandari AK. IJPSR 2012; 3(2): 14-20.
[14] Agarwal a, Katare OP. Pharmaceutical Technology 2002; November: 48-60.
[15] Dodov MG, Kumbaradzi EF, Goracinova K, Calis S, Simonoska M, Hincal AA. Acta Pharm 2003; 53: 241–250.
[16] Huang YZ, Qing Gao J, Liang WQ, Nakagawa S. Biol Pharm Bull 2005; 28(2): 387-390.
[17] Liang X, Mao G, Simon Ng KY. J Colloid Interface Sci 2004; 278: 53–62.
[18] Mozafari MR. Cellular & Molecular Biology Letters 2005; 10: 711 – 719.
[19] Barbeau J, Cammas-Marion S, Auvray P, Benvegnu T. J Drug Delivery 2011; 2011: 1-11.
[20] Anwekar H, Patel S, Singhai AK. Int. J. of Pharm. & Life Sci. (IJPLS) 2011; 2(7): 945- 951.
[21] Mozafari MR, Johnson C, Hatziantoniou S, Demetzos C. J Liposome Research 2008; 18: 309–327.
[22] Riaz M. Pakistan J Pharm Sci 1996; 19(1): 65-77.
[23] MacLachlan I. Antisense Drug Technology. Taylor and Franac Group, 2007, pp.237-270.
[24] Sriwongsitanont S, Ueno M. The Open Colloid Science J 2011; 4:1-6.
[25] Samad A, Sultana Y, Aqil M. Current Drug Delivery 2007; 4: 297-305.
[26] Onaderra M, Mancheno JM, Gasset M, Lacadena J, Schiavo G, Pozo AMD, Gavilanes G. Biochem J 1993; 295: 221-225.
[27] Tuan LQ, Umakoshi H, Shimanouchi T, Kuboi R. Enzyme and Microbial Technology 2009; 44: 101-106.
[28] N Latif, Bachhawat BK. J Biosci 1984; 6: 491–502.
[29] Shafaa MW, Dayem SA, Elshemey WM, Osman HM. Romanian J Biophys 2008. 18(4): 293–300.
[30] Przeworska E, Gubernator J, Kozubek A. Biochimica et Biophysica Acta 2001; 1513 :75-81.
[31] Gupta V, Gupta R, Grover R, Khanna R, Jangra V, Mittal A. J Nanosci Nanotech 2008; 8: 2328-2333.
[32] Lutsiak MEC, Kwon GS, Samuel J. J Pharm Pharmaceut ScI 2002; 5(3): 279-284.
[33] Haginoya K, Kato T, Higuchi M, Shitomi Y, Asakura T, Hayakawa T, Mitsui T, Hori H. Bull. Facul. Niigata Univ 2005; 57(2): 115-120.
[34] Mourtas S, Fotopoulou S, Stela D, Sfika V, Tsakiroglou C, Antimisiaris SG. Colloide and Surfaces B: Biointerfaces 2007; 55: 212-221.
[35] Purwaningsih EH, Schubert R. Makara, Kesehatan 2004; 8(2): 49-52.
[36] Begum MY, Abbulu K, Sudhakar M. Chem Sci Trans 2012; 1(1): 201-209.
[37] Ortan A, Campeanu GH, Pirvu CD, Popescu L. Roumanian Biotechnological letters 2009; 14(3): 4411-4417.
[38] Maurer X, Fenske DB, Cullis PR. Expert Opin Biol Ther 2001; 1(6): 1-25.

[39] Castile JD, Taylor KMG. International J Pharmaceutical 1999; 188: 87-95.
[40] Ramana LN, Sethuraman S, Ranga U, Krishnan UM. J Biomedical Science 2010; 17 (57): 1-9.
[41] Lee S, Lee J, Choi YW. Bull. Korean Chem. Soc 2007; 28(1): 99- 102.
[42] Nallamothu R, Wood GC, Kiani MF, Moore BM, Horton FP, Thoma LA. PDA Jof Pharmaceutical Science and Technology 2006; 60(3): 144-155.
[43] Holovati JL, Gyongyossy-Issa MIC, Acker JP. Cell Preservation Technology 2008; 6:207–218.
[44] Chiu GNC, Bally MB, Mayer LD. Biochimica et Biophysica Acta 2001; 1510: 56-69.
[45] Calvagno MG, Celia C, Paolino D, Cosco D, Iannone M, Castelli F, Doldo P, Fresta M. Current Drug Delivery 2007; 4: 89-101.
[46] G Zhai, Wu J, Zhao X, YuB, Li H, Lu Y, Ye W, Lin YC, Lee RJ. Anticancer Research 2008; 28: 2801-2806.
[47] Chen C, Han D, Cai C, Tang X. J of Controlled Release 2010; 142: 299-311.
[48] Mehta J, Blake J, Craddock C. The Open Transplantation Journal 2011; 5: 23-29.
[49] Chang HI, Cheng MY, Yeh MK. Open Access Scientific Reports 2012; 1(3): 1-8.
[50] Beukelman CJ, Van Den Berg AJJ, Hoekstra MJ, Uhl R, Reimer K, Mueller S. Burns 2008; 3 4: 845–855.
[51] Montanari J, Perez AP, Salvo FD, Diz V, Barnadas R, Dicelio L, Doctorovich F, Morilla MJ, Romero EL. International J Pharmaceutics 2007; 330 :183–194.
[52] Dodov MG, Simonoska M, Goracinova K. Bulletin of the Chemists and Technologists of Macedonia 2005; 24(1): 59-65.
[53] Moghimipour E, Sajadi SA, Ramazani M, Lobenberg R. IJBMS 2003; 6(1): 9-19.
[54] Huang X, Caddell R, Yu B, XU S, Theobald B, Lee LJ, Lee RJ. Anticancer Research 2010; 30: 463-466.
[55] Moghimipour E, Handali S. Advanced Pharmaceutical Bulletin 2012; 2(1): 93-98.
[56] Mitkari BV, Korde SA, Mahadik KR, Kokare CR. Indian J.Pharm. Educ. Res 2010; 44(4): 324-333.
[57] Liolios C.C, Gortzi O, Lalus S, Tsakins J, Chinou I. Food Chemistry 2009; 112: 77-83.
[58] Moghimipour E, Aghel N, Mahmoudabadi AZ, Ramezani Z, Handali S. Jundishapur J Nat Pharm Prod 2012; 7(3): 117-122.
[59] Mugabe C, Halwani M, Azghani AO, Lafrenie RM, Omri A. Antimicrobial Agents and Chemotherapy 2006; 50(6): 2016-2022.
[60] Vitas AI, Diaz R, Gamazo C. Antimicrobial Agents and Chemotherapy 1996; 40(1): 146-151.
[61] Perrie Y, Frederik PM, Gregoriadis G. Vaccine 2001; 19: 3301–3310.
[62] PAN XQ, LEE RJ. Anticancer Research 2005; 25: 343-346.
[63] Turk MJ,Waters DJ, Low PS. Cancer Letters 2004; 213:165–172.
[64] Chonn A, Cullis PR. Current Opinion in Biotechnology 1995; 6:698-708.
[65] Changediya V, Khadke M, Devdhe S. Res J Pharm Biol Chem Sci 2011; 2(3): 57-68.
[66] Patil MS, Patil SB, Chittam KP, Wagh RD. AJPSR 2012; 2 (3): 1-9.
[67] Prescott LM. Microbiolog. The McGrow-Hill companies, 2002, pp.249-254.
[68] Rathore P, Swami G. IJPSR 2012; 3(3): 737-755.

ISSN: 0975-8585

[69] Benvegnu T, Lemiegre L, Cammas-Marion S. Recent Patens on Drug Delivery & Formulation 2009; 3: 206-220.
[70] Krishnamachari Y, Geary SM, Lemke CD, Salem AK. Pharm Res 2011; 28(2): 215-23.