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PHYSICOCHEMICAL PROPERTIES AND TARGETING STRATEGIES FOR LIPOSOMES: A VERSATILE DRUG DELIVERY SYSTEM

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ABSTRACT

Liposomes are phospholipid-based bilayer vesicular systems. It can encapsulate both hydrophilic and hydrophobic drugs. Its properties like biocompatibility and site-specific action are the two most attractive features of the systems. Their architecture has been satisfactorily accepting a great technical advance such as remote drug loading, combining passive, active drug targeting concepts, etc. It can be used in diverse areas like carrier for anticancer, bio-active molecules, diagnostics, and therapeutic agents. In present review, we discuss in detailed about structure of liposomes and liposomes as effective targeting delivery system.

Keywords *– Liposomes; Vesicle; Physicochemistry; Targeting site.*

1. INTRODUCTION

Development of suitable drug delivery system has become a challenging task to deliver particular type of drug to the targeted site. There are number of drug delivery systems so far developed for various drugs. These systems include ethosomes, liposomes, tablets, multilayered tablets, emulsion, suspension, etc. New drg delivery Liposomes are phospholipid-based bilayer vesicular systems. They are discovered by Alec Bangham in 1964. It was found that phospholipids in aqueous systems can form closed bilayered spherical vesicular structures **(Fig.1**) [1-27]. They have emerged as one of the most promising tools for drug targeting in medical fields. Mostly it is composed of natural substances like cholesterol, phosphatidylglycerol, dicetylphosphate, distearoylphosphatidylcholine, dipalmitoylphosphatidylcholine, etc. It is nontoxic, biodegradable, non-immunogenic and biocompatible. The size varies from a few nanometers to several micrometers [28-29].

In medical use, the range of liposomes applied in between 50-450 nm. The properties of liposomes are mostly dependent on lipid composition, surface charge, size, and the method of preparation. The rigidity of bilayer and the charge are depending on the selection of the bilayer components [30-32]. Conventional liposomes tend to form aggregate with each other resulting in immature release of liposomal contents over time [33-34]. To overcome such problems associated with liposomes, the surface-

modification strategies were adopted by coating the surface of liposomes with inert, biocompatible hydrophilic polymers, such as polyethylene glycol. These polymers confer steric stabilization to the liposomal surface via formation of a protective layer over the liposome surface and slows down liposome recognition by opsonins, and therefore, subsequent clearance of liposomes [35-45].

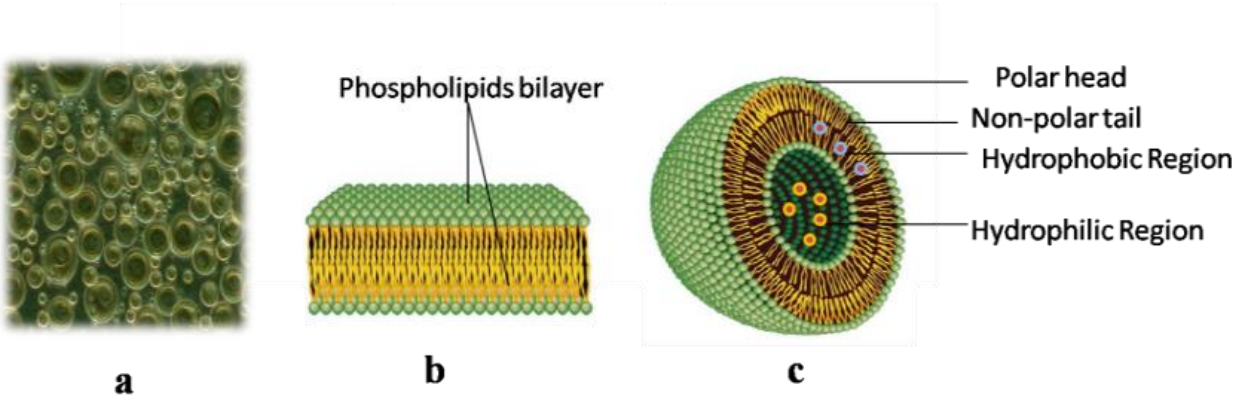

Fig. 1: Liposomes (a) Microscopic, (b) Phospholipid bilayer, and (c) Spherical Vesicle.

2. PHYSICOCHEMISTRY OF SYSTEM

Liposomes are the drug carrier for delivering both the hydrophilic and the lipophilic drugs. The strength of liposomes as a carrier system for drugs depend on the number of physicochemical properties of the liposomal membranes, on the nature of their components, their size, surface charge, and the lipid organization [46]. As liposomes composed of phospholipids, dispersed in aqueous phase, due to their amphiphatic nature they have a strong tendency to form membranes by polar head and nonpolar tail [47]. Polar heads prefer to interact with the aqueous environment and long non-polar aliphatic chains promote interaction with one another **(Fig.1)** [48].

Hydrophobic interactions are behind the formation of these lipid bilayers and the forces like Van der Waals keep the long hydrocarbon tails together, thus it helps for strengthening of this architecture. Stabilization of this vesicle is due to hydrogen bonds and polar interactions between the water molecules of the aqueous environment and the polar heads of an organization. Whole structural organization of lipids depends on their nature, concentration, temperature, and geometric form. If ions or other drug molecules are present during the formulation process of liposomes, they can be encapsulated inside these membranes [49-50].

The addition of cholesterol in the liposomal formulation reduces their permeability and increases its stability by interacting with the core of the membrane, because the presence of cholesterol induces a dense packing of phospholipids and inhibits their transfer to high-density lipoprotein and low-density lipoprotein [51]. Also, the use of phosphatidyl choline with saturated fatty acyl chains and materials that stretch the transition temperature beyond 37°C important for better stabilization [52-54].

3. CLASSIFICATION

Liposomes can be classified on the basis of the method of preparation like reverse-phase evaporation vesicles or vesicle extruded technique, size small, intermediate, or large, and lamellarity uni-, oligo-, and multilamellar vesicles **(Fig. 2)**. The formation of

unilamellar vesicles (ULVs) composed of one lipid bilayer with size 50–250 nm. Ideally it is important for the encapsulation of hydrophilic drugs [55].

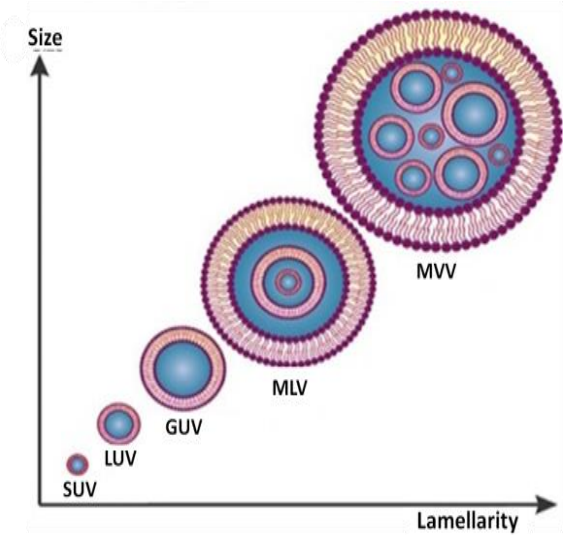

Fig. 2: Liposomes according to size and lamellarity.

The multilamellar vesicles (MLVs) depend on the synthesis methods and post formation processing used for their preparation. It is composed of two or more concentric lipid bilayers organized by packing of an onion, having size1-5µm preferentially entrap lipid-soluble drugs [56]. The multi vesicular vesicles (MVV) composed of multiple vesicles entrapped under the lipid bilayer. According to the lamellar arrangement MLVs are formed more easily at larger hydrodynamic diameters. It has greater entrapped volume than ULVs. [57] **(Table 1)**.

Table 1: Types of liposomes with their size and lipid layers [58].

Vesicle Type	Number of lipid bilayers	Size of vesicles
Small Unilamellar Vesicles (SUV)	1	50-100 nm
Large Unilamillar Vesicles (LUV)	1	All size ranges
Giant Unilamellar Vesicles (GUV)	1	More than 1 micrometer
Multilamellar Vesicles (MLV)	5-25	More than 0.5 micrometer
Multi Vesicular Vesicles (MLV)	Multi compartmental str.	More than 0.5micrometer

4. FORMATION OF TARGETED LONG-CIRCULATING LIPOSOMES

Targeted long circulating liposome is a critical step to achieve development of liposomes. It must survive in the systemic circulation for longer time to reach and bind to the targeted site. Strategies to increase the circulation time of liposomes, that is reduction in liposomal size or the inclusion of cholesterol and high phase transition lipids provided a modest decrease in the

clearance rate of liposomes. More success came from the strategy of including a hydrophilic molecule at the liposomal surface, e.g., phosphatidylinositol or lipid derivatives of polymers like PEG, poly(acrylamide), poly(vinylpyrrolidone), etc. [59].

Coupling strategies of molecule is via cross-linking molecules of a ligand is coupled, often through a spacer molecule, to a hydrophobic molecule. The stable insertion of the conjugate into the lipid bilayer of a liposomes is important. Generally, the carrier of choice has been phosphatidylethanolamine (PE) because of the reactive amine in its head group and the availability of various acyl chain lengths of different degrees of unsaturation. PEG- at concentrations of approximately 5% or more in the bilayer, probably sterically interfere with the accessibility of the antibody to the liposomal surface. PEG could be incorporated into liposomes after ligand coupling occurs, to overcome the interference of PEG in the conjugation of antibodies onto the liposomal surface [60-62].

5. LIPOSOME TARGETING STRATEGIES

A. Passive targeting

Drugs associated with a carrier, exhibit two pharmacokinetic processes related to the liposomal system and the drug itself. When the drug is released slowly, the kinetics will be determined essentially by that of the liposomes. Accordingly, drugs with short biological half-lives, such as retinoic acid and Curcumin owing to rapid clearance can be encapsulated into a liposome which protects the drug providing minimal to no leakage [63]. The macrophagial uptake requires binding of specific serum proteins to the drug-particulate system for identification and consequent elimination, a phenomenon commonly known as opsonization [64].

The most widely used polymers to formulate stealth liposomes are PEG-based conjugates, it has unique characteristics of this polymer including non-ionic nature, high solubility in both aqueous and organic media, excellent biocompatibility, lack of toxicity, low immunogenicity and antigenicity, good excretion kinetics, as well as ease of synthesis with a broad range of molecular weight and a low polydispersity index is < 1.1 and a variety end-group functionalities [65-66]. Although other surface modifying polymers are much less common, a recent patent claimed that liposomes stabilized by poly 2-methacryloyloxyethyl phosphorylcholinecan enhance retention in vivo to a greater extent than do PEGylated liposomes [67-69].

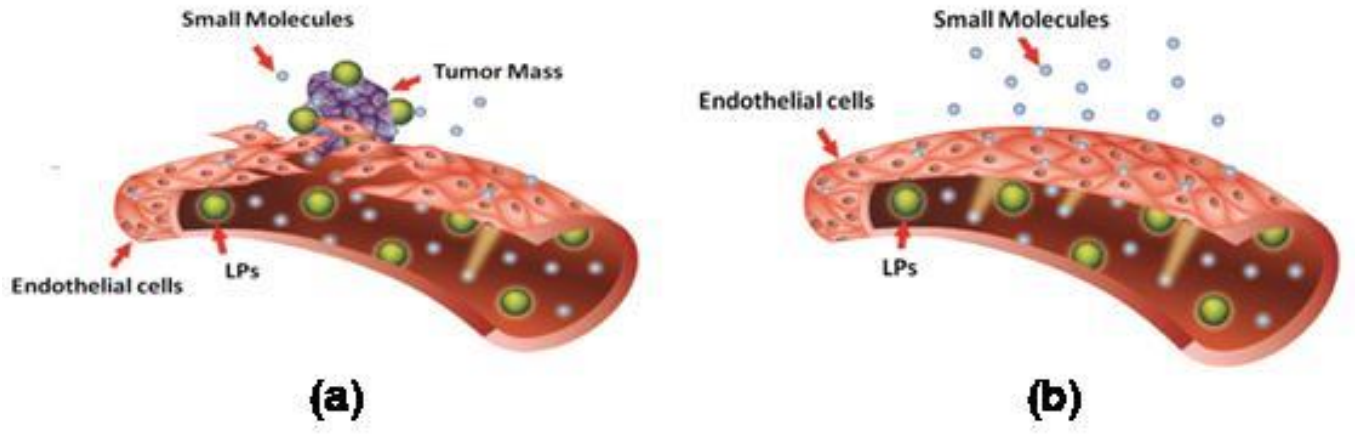

Fig. 3: Targeting of LPs on tumor (a) and normal (b) vessels by enhanced permeability and retention effect.

B. Ligand-mediated targeting

Ligand or receptor-mediated targeting is based on the surface charges of the particle with ligands that bind specifically to receptors. Ligand-receptor binding initiate the uptake of the liposomes by the targeted cell. Targeting ligands such as proteins, peptides, antibodies, antibody fragments, vitamins, carbohydrates and glycoproteins have shown the ability to bind selectively target cells such as tumor cells. For e.g., ginsenosides which are substrates of glucose-related transporters overexpressed in certain tumors and have the ability to cross the blood-brain barrier (BBB) via glucose transporter 1-mediated transport can therefore function as targeting ligands [70-72].

When the objective is imaging a specific target, it is crucial for the imaging agent to accumulate at the desired site in sufficient concentration. A possible strategy to enhance imaging outcomes is the use of ligand-mediated targeting Liposomes, carrying a suitable imaging agent, which exhibit the ability to bind specifically to the receptor of interest [73-74]. The BBB, which is formed by endothelial capillary endothelium and separates the vascular system from the brain, is a strict permeability barrier that efficiently restricts penetration of therapeutic compounds to the brain. A fusion protein containing a six amino acid peptide with the ability to target the BBB has been incorporated in a liposomal formulation, comprising CHOL: phosphatidyl phosphoric acid:PC (30-50:5-20:40-60), in which the targeting protein is anchored and conjugated to 1,2-dioleoyl-sn-glycero-3-succinate within the LP bilayer [75-76].

C. Combined targeting

Ligand-mediated targeting would require extended residence time in blood of the liposomal system for an effective access and accumulation at the site of action. Therefore, it is very common to engineer therapeutic Liposomes to exhibit combined passive and ligand-mediated targeting capabilities (passive + ligand-mediated). Combined targeting is attained by functionalizing the surface of Liposomes with both a targeting ligand and a hydrophilic polymer, mostly PEG chains, in which the ligand is usually coupled to the distal end of the polymeric chain. The PEG linkers may enhance ligand's binding affinity for the receptors and the flexible PEG spacers are known to enhance ligand-receptor binding. However, it was shown that the distal conjugation of the trigalactosyl on the PEG chain reduced the uptake of the particle by hepatocytes by comprising with the non-PEGylated ligand [77-79].

D. Stimuli-responsive liposomes

Stimuli-responsive LPs are intelligent systems that have the ability to deliver and release their cargo in a site-specific manner. The composition of these liposomal systems can be specifically designed to facilitate triggered or controlled release in response to an endogenous stimulus like pH, redox potential, or specific enzymatic activity or an applied external stimulus these are heat, light and ultrasound (US) waves. Triggered drug release is based on the concept of membrane destabilization induced by local defects within bilayer membranes to promote payloads release [80].

6. METHODS OF ASSAY OF LIPOSOMES

There are many analytical tools that are used for the analysis of various pharmaceutical drugs, formulations, including liposomes. These methods include UV-Vis spectrophotometry, gas chromatography, HPLC, HPTLC, etc. [81- 115].

7. CONCLUSION

Liposomal drug delivery systems can be used for delivery of both hydrophilic and lipophilic drugs. Thus, it concludes that it could be effectively used in delivery of drugs for severe diseases.

8. DISCLOSURE OF CONFLICT OF INTEREST

The author declares no conflict of interest.

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