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PHOSPHATIDYLCHOLINE: A REVOLUTION IN DRUG DELIVERY TECHNOLOGY

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ABSTRACT

In recent years Phosphatidylcholine has greatly impacted the drug delivery technology. The very first and most important advantage of phospholipid based vesicular system is the compatibility of phospholipids with membrane of human either internal membrane as well as skin (external membrane). For a drug to be absorbed and distributed into organs and tissues and eliminated from the body, it must pass through one or more biological membranes/ barriers at various locations. Such a movement of drug across the membrane is called as drug transport. For the drugs to be delivered to the body should cross the membranous barrier. Either it would be from oral route or topical/transdermal route. Therefore the phospholipid based carrier systems are of considerable interest in this era. A number of drug delivery systems are based entirely on Phosphatidylcholine such as Liposome, Ethosome, Phytosome, Transferosomes, and Nanocochelates.

INTRODUCTION:

Since last decades the popularity of vesicular systems has been increased because of a lot of advantages associated with it. Vesicular systems are mainly composed of phospholipids. Phospholipids are amphipathic (having affinity for both aqueous and polar moieties) molecules as they have a hydrophobic tail and a hydrophilic or polar head.

The hydrophobic tail is composed of two fatty acid chains containing 10-24 carbon atoms and 0-6 double bonds in each chain. The polar end of the molecule is mainly phosphoric acid bound to a water- soluble molecule. The hydrophilic and hydrophobic domains/ segments within the molecular geometry of amphiphilic lipids orient and self organize in ordered supramolecular structure when confronted with solvents [1]. Some commonly used synthetic phospholipids are dioleoyl- phosphatidyl- choline (DOPC), Dioleoyl-phosphatidyl- ethanolamine (DOPE), Distearoyl- phosphatidyl- choline (DSPC), Distearoyl- phosphatidyl- ethanolamine (DSPE)[2].

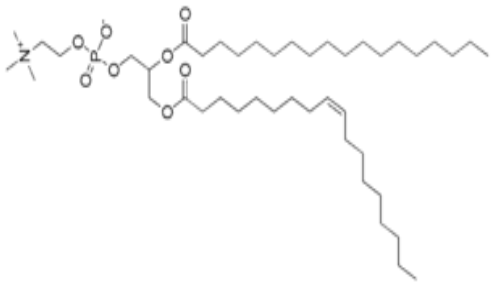

Fig 1: Phophatidylcholine containing oleyl and stearoyl side chains.[3]

Among these all phospholipids, phosphatidylcholine classes of phospholipids are very important in the drug delivery technology.

The very first and most important advantage of phospholipid based vesicular systems is the compatibility of phospholipids with membrane of human either internal membrane as well as skin. For a drug to be absorbed and distributed into organs and tissues and eliminated from the body, it must pass through one or more biological membranes/ barriers at various locations. Such a movement of drug across the membrane is called as drug transport. The cellular membrane consists of a double layer of amphiphilic phospholipids molecules arranged in such a fashion that their hydrocarbon chains are oriented inwards to form the hydrophobic or lipophilic phase and their polar heads oriented to form the outer and inner hydrophilic boundaries of the cellular membrane that face the surrounding aqueous environment.

Globular protein molecules are associated on either side of these hydrophilic boundaries and also interspersed within the membrane structure. In short, the membrane is a mayonnaise sandwich where a bimolecular layer of lipids is contained between two parallel monomolecular layers of proteins. The hydrophobic core of the membrane is responsible for the relative impermeability of polar molecules. Aqueous filled pores or perforations of 4 to 10 A^0 in diameter are also present in the membrane structure through which organic ions and small organic water- soluble molecules like urea can pass. In general, the biomembrane

acts like a semi permeable barrier permitting rapid and limited passage of some compounds while restricting that of others[4]. The GI linings constituting the absorption barrier allows most nutrients like glucose, amino acids, fatty acids, vitamins, etc., to pass rapidly through it into the systemic circulation but prevent the entry of certain toxins and medicaments. Thus, for a drug to get absorbed after oral administration, it must first pass through this biological barrier[4].

And for topical/ transdermal delivery of drugs, it has to cross the skin. During the past decades there has been wide interest in exploring new techniques to increase drug absorption through skin[5, 6,7]. Topical delivery of drugs by lipid vesicles has evoked a considerable interest. The skin, the heaviest single organ of the body, combines with the mucosal linings of the respiratory, digestive, and urogenital tracts to form a capsule, which separates the internal body structures from the external environment. The skin has various functions such as protection from external environment, maintenance of body posture, regulation of temperature, etc. Including these various primary functions, it also acts as a site for drug delivery.

The skin itself has two main layers: the epidermis, which is the outermost layer of the skin, covering the dermis that is the active part of the skin, holding the hair muscles, blood supply, sebaceous glands, and nerve receptors. There is a fat layer underneath the dermis. The skin is a very heterogeneous membrane and has a variety of cell types, but the layer that controls the penetration of drugs is called the stratum corneum and, despite its thickness of only 15–20 µm, it provides a very effective barrier to penetration. The permeation of the drug through the skin has several routes: transcellular, intercellular, and appendageal (through eccrine (sweat) glands or hair follicles). Since the appendages occupy a very low surface area, this means of permeation is less significant under normal conditions[8].

Nevertheless, in iontophoretics delivery this route is more significant[9]. So for the drugs to be delivered to the body should cross the membranous barrier. Either it would be from oral route or topical/transdermal route. Therefore the phospholipid based carrier systems are of considerable interest in this era.

History: Phosphatidylcholine was first isolated in Odessa, Ukraine some 50 years ago. This was followed by further research in Germany and Russia. It has been marketed by Sanofi- Aventis for over 30 years and, at present; the substance phosphatidylcholine is registered in 53 countries. Its main application nowadays lies in the intravenous treatment and prevention of fat embolisms in polytraumatized patients in the treatment of metabolic disorders and as a liver-protecting substance[10].

Mechanism of vesicle formation: In aqueous medium the molecules in self-assembled structures are oriented in such a way that the polar portion of the molecule remains in contact with the polar environment and at the same time shields the non-polar part. Among the amphiphilic used in the drug delivery, viz soaps, detergents, polar lipids, the latter (polar lipids) are often employed to form

concentric bilayered structures. However, in aqueous mixtures these molecules are able to form various phases, some of them are stable and others remain in the metastable state[11]. At high concentrations of these polar lipids, liquid- crystalline phases are formed that upon dilution with an excess of water can be dispersed into relatively stable colloidal particles. The macroscopic structures most often formed include lamellar, hexagonal or cubic phases dispersed as colloidal nanoconstructs (artificial membranes) referred to as liposomes, hexasomes or cubosomes, respectively[12]. The most common natural polar phospholipids are Phophatidylcholine (PC).

These are amphipathic molecules in which a glycerol bridges links to a pair of hydrophilic polar head group, phosphatidylcholine. Fatty chains are embedded in the hydrophilic inner region of the membrane surface, the hydrophilic head group, including the phosphate portion, points out towards the hydrophilic aqueous environment. Molecules of PC are not soluble (rather dispersible) in aqueous milieu in the physical chemistry sense, as in aqueous media they align themselves closely in planer bilayer sheets to minimize the unfavorable interactions between the bulk aqueous phase and long hydrocarbon fatty acyl chain. Such interactions are completely eliminated when the sheets fold over themselves to form closed sealed and concentric vesicles.

The large free energy change between an aqueous and hydrophobic environment explains the most favored orientation of lipids to assemble as concentric bilayer structures that exclude confrontation between aqueous and hydrophobic domains. This distinctive behavior derives in the lowest free energy state and hence ensures the maximum stability to self- assembled structures[11]. The phosphatidylcholine and its synthetic analogues differ markedly from amphiphilic molecules of differ markedly from amphiphilic molecules of other origin (soaps, detergents, lysolecithin) in that they preferably orient to form bilayer sheets rather than micellar structures.

This presumably attributed to the double fatty acid chain that imparts the molecule an overall tubular shape are more suitable for assemblage in planer sheets. In contrast, the detergent molecule with a polar head and single acyl chain has a conical shape and facilitate the formation of spherical micellar structures. Depending on the hydrophobic environment and aqueous phase, homogenous smectic phases of parallel lipid bilayers (lyotropic phases) or heterogeneous dispersion of multilamellar or single- walled vesicles can be observed.

At lower water content and higher temperature, other lyotropic liquid crystalline phases exist, such as the hexagonal, the cubic and the ribbon phases. Table presents different vesicle forming lipids with their molecular weights and phase transition temperature (Tc^0). The vesicles have been well known for their importance in cellular communication and particle transportation for many years. Researchers have understood the properties of vesicles structure for use in better drug delivery within their cavities,

which would to tag the vesicle for cell specificity.

Advantages of phospholipid based carrier systems[2] in comparison to other delivery systems:-

1. These systems show enhanced permeation of drug through skin for transdermal and dermal delivery.
2. These are platform for the delivery of large and diverse group of drugs (peptides, protein molecules).
3. Their composition is safe and the components are approved for pharmaceutical and cosmetic use.
4. Low risk profile- The toxicological profiles of the phospholipids are well documented in the scientific literature.
5. High market attractiveness for products with proprietary technology. Relatively simple to manufacture with no complicated technical investments required for production of Ethosomes.
6. The vesicular system is passive, non-invasive and is available for immediate commercialization.

Brief introduction to the phospholipid based carrier systems:

Liposome: Liposomes are colloidal, vesicular structures composed of one or more lipid bilayers surrounding an equal numbers of aqueous compartments. Since, 1960's pharmaceutical researchers used liposomes as therapeutic tools in medicinal field. A number of liposomal formulations of such drugs have available in the market such as Doxil® (Doxorubicin), Fungizone® (Amphotericin-B), Novasome® (Smallpox vaccine) and Nyotran™ (Nystatin). Liposomes used as potential carriers in field like tumor targeting, gene and antisense therapy, genetic vaccination, immunomodulation and skin care and topical cosmetics products. The present review highlights the composition, method of preparation, characterization, therapeutic applications of liposomes and its marketed products[13].

Ethosome: Ethosomes are soft, malleable vesicles composed mainly of phospholipids, ethanol (relatively high concentration) and water. These "soft vesicles" represents novel vesicular carrier for enhanced delivery to/through skin. The size of Ethosomes vesicles can be modulated from tens of nanometers to microns.

Ethosomes are provides a number of important benefits including improving the drug's efficacy, enhancing patient compliance and comfort and reducing the total cost of treatment. The Ethosomes were found to be suitable for various applications within the pharmaceutical,biotechnology, veterinary, cosmetic, and nutraceutical markets[14].

Phytosome: Phytosome are created when the standardized extract and active ingredients of an herb are bound to the phospholipids on a molecular level. Phytosome structures contain the active ingredients of the herb surrounded by the phospholipids. The phospholipid molecular structure includes a water-soluble head and two fat-soluble tails, because of this dual solubility, the phospholipid acts as an effective emulsifier, which is also one of the chief components of the membranes in our cells. Phytosomes are advanced forms of

herbal products that are better absorbed, utilized, and as a result produce better results than conventional herbal extracts[15].

Transferosomes: In functional terms, may be described as lipid droplets of such deformability that permits its easy penetration through the pores much smaller than the droplets size. Transferosomes have been developed in order to take advantage of phospholipids vesicles as transdermal drug carrier.

These self optimized aggregates, with ultraflexible membrane, are able to deliver the drug reproducibly either into or through the skin, depending on the choice of administration or application, with high efficiency. Transferosomes overcome the skin penetration difficulty by squeezing themselves along the intracellular sealing lipids of stratum corneum. There is provision for this, because of the high vesicle deformability, which permits the entry due to mechanical stress of surrounding, in a self adapting manner.

Flexibility of transferosomes membrane is achieved by mixing suitable surface active agents in the proper ratios. The resulting flexibility of transferosome membrane minimize the risk of complete vesicle rupture in the skin and allow transferosomes to follow the natural water gradient across the epidermis ,when applied under non occlusive condition. Transferosomes can penetrate the intact stratum corneum spontaneously either through intracellular route or transcellular route[16].

Long circulating liposomes: The major limitation of liposomes is their fast elimination from the blood and localization in reticuloendothelial system primarily kupfer cells of liver. Different methods have been reported to achieve long circulation of liposomes *in vivo*, including modification with certain lipids such as monosialoganglioside, palmityl-D-Glucoronic acid and PEG-PE. These liposomes show significantly longer circulation in blood than the liposomes without these lipids[17, 18, 19, 20, 21].

Nanocochelates: Nanocochelates consists of a purified soy based phospholipid that contains at least about 75% by weight of lipid which can be phosphatidyl- serine (PS), dioleoyl-phosphatidyl- serine (DOPS), phosphatidic acid (PA), phosphatidyl-inositol (PI), phosphatidyl glycerol (PG) and /or a mixture of one or more of these lipids with other lipids. Additionally or alternatively, the lipid can include phosphatidylcholine (PC), phosphatidyl-ethanolamine (PE), diphosphotidyl-glycerol (DPG), dioleoyl phosphatidic acid (DOPA), distearoyl phosphatidylserine (DSPS), and dimyristoyl phosphatidylserine (DMPS), dipalmitoyl phosphatidylgycerol (DPPG) [7].

A multivalent cation, which can be Zn^{+2} or Ca^{+2} or Mg^{+2} or Ba^{+2} and a drug, which can be protein, peptide, polynucleotide, antiviral agent, anesthetic, anticancer agent, immunosuppressant, steroidal anti inflammatory agent, non steroidal anti inflammatory agents, tranquilizer, nutritional supplement, herbal product, vitamin and/or vasodilatory agent[22].

Application of phospholipid based carrier systems:

1. Liposomes

Table 1: Therapeutic applications of liposomes:

Drug	Route of administration	Application	Targeted Diseases
Amphotericin-B[23]	Oral delivery	Ergosterol membrane	Mycotic infection
Insulin [24]	Oral, Ocular, Pulmonary and Transdermal delivery	Decrease glucose level	Diabetic mellitus
Ketoprofen [26]	Ocular delivery	Cyclo-oxygenase enzyme inhibitor	Pain muscle condition
Pentoxyfylline[26]	Pulmonary delivery	Phosphodiesterase	Asthma
Tobramycin[27]	Pulmonary delivery	Protein synthesis inhibitor	Pseudomonas infection, aeruginosa
Salbutamol[28]	Pulmonary delivery	β_2- adrenoceptor antagonist	Asthma
Cytarabin[29]	Pulmonary delivery	DNA-polymerase inhibition	Acute-leukemias
Benzocain[30]	Transdermal	Inhibition of nerve impulse from sensory nerves	ulcer on mucous surface with pain
Ketoconazole[30]	Transdermal	Inhibit ergosterol membrane	Candida- albican's
Levonogesterol[30]	Transdermal	Rhamnose receptor	Skin disorder
Hydroxyzine[30]	Transdermal	H_1- receptor antagonist	Urtecaria, allergic skin disorder
Ibuprofen[30]	Oral delivery	Chemoreceptor, free nerve ending	Rheumatoid arthritis
Pentamidine isethionate[30]	Oral delivery	Killed protozoa	Protozoal infection
Idoxiuridine[30]	Ocular delivery	DNA-synthesis, Protein synthesis	Herpex- simplex, Keratitis
Adrenaline[30]	Ocular delivery	Decreases intra-ocular pressure	Glucoma, Conjectivitis
Triamcinolone[30]	Ocular delivery Transdermal	Inhibition of prostaglandin	Anti-inflammatory
Penicillin G[30]	Pulmonary delivery	Inhibit synthesis of bacterial cell wall	Meningococal, staphylococcal infection
Terbutaline sulphate[30]	Pulmonary delivery	β_2- adrenoceptor	Asthma
Methotrexate[30]	Transdermal	Dihydrofolate reductase	Cancer
Doxorubicin[31, 32]	Oral delivery	Inhibit DNA/ Protein synthesis	Cancer

Table 2: list of marketed products of Liposome

Marketed product	Drug used	Target diseases	Company
Doxil™ or Caelyx™ [33]	Doxorubicin	Kaposi's sarcoma	SEQUUS, USA
DaunoXome™ [34]	Daunorubicin	Kaposi's sarcoma, breast & lung cancer	NeXstar, USA
Amphotec™ [35]	Amphotericin-B	fungal infections, Leishmaniasis	SEQUUS, USA
Fungizone® 36	Amphotericin-B	fungal infections, Leishmaniasis	Bristol-squibb, Netherland
VENTUS™ [12]	Prostaglandin-E_1	Systemic inflammatory diseases	The liposome company, USA
ALEC™ [37]	Dry protein free powder of DPPC-PG	Expanding lung diseases in babies	Britannia Pharm, UK
Topex-Br[34]	Terbutaline sulphate	Asthma	Ozone, USA
Depocyt[38]	Cytarabine	Cancer therapy	Skye Pharm, USA
Novasome®[38]	Smallpox vaccine	Smallpox	Novavax, USA
Avian retrovirus vaccine[39]	Killed avian retrovirus	Chicken pox	Vineland lab, USA
Epaxal –Berna Vaccine[39]	Inactivated hepatitis-A Virions	Hepatitis A	Swiss serum & vaccine institute, Switzerland
Doxil®40	Doxorubicin HCl	Refractory ovarian cancer	ALZA, USA
Evacet™ [41]	Doxorubicin	Metastatic breast cancer	The liposome company, USA
VincaXome[41]	Vincristine	Solid Tumours	NeXstar, USA
Mikasome® [41]	Amikacin	Bacterial infection	NeXstar, USA
Autragen™ [41]	Tretinoin	Kaposi's sarcoma	Aronex Pharm, USA
Shigella Flexneri 2A [41]Vaccine	Shigella flexneri 2A	Shigella Flexneri 2A infections	Novavax, USA
Nyotran™ [41]	Nystatin	Systemic fungal infections	Aronex Pharm, USA

2. Ethosome:-

Table 3: Therapeutic applications of Ethosomes

Drug	Applications	Comments
Acyclovir[42]	Treatment of Herpetic infection	Improved drug delivery
Zidovudine[43]	Treatment of AIDS	Improved transdermal flux
Trihexypenidyl HCl[44, 45]	Treatment of Parkinsonian syndrome	Increased drug entrapment efficiency, reduced side effect & constant systemic levels
Erythromycin[46]	Efficient healing of *S. aureus* - induced deep dermal infections	Improved drug penetration and systemic effect.
Insulin[47]	Treatment of Diabetes	Improved therapeutic efficacy of drug
Testosterone[48]	Treatment of male hypogonodism	Enhance skin permeation
Cannabidol[49]	Prevents inflammation and edema	Significant accumulation of the drug in the skin
Minodixil[50]	Hair growth promotion effect	Higher skin retention
Bacitracin[51]	Treatment of dermal infections	Reduced drug toxicity

3. Phytosome:-

Table 4: Therapeutic applications of Phytosomes

Molecule	Application	Targeted Diseases
Botanical extracts[52]	Enhance bioavailability due to their complex with phospholipids and delivers faster and improved absorption in intestinal tract.	Diabetic mellitus
Non-lipophilic botanical extract[53]	To be better absorbed in intestinal lumen	For treatment of Infections
Cosmetics, gap junction protein[53]	Transdermal immunization	More skin penetration and have a high lipid profile

4. Transferosomes:-

Table 5: Therapeutic applications of Transferosomes

Drug	Application	Targeted Diseases
Insulin[54]	Decreases glucose level	Diabetic mellitus
Interferons, for example leukocytic derived interferone-α (INF-α)[54]	Antiviral, antiproliferive and some immunomodulatory effects	For treatment of Infections
Soluble protein like integral membrane protein, human serum albumin, gap junction protein[54]	Transdermal immunization	Immunization
Corticosteroids[54]	Treatment of skin diseases	Skin diseases
Anesthetics[54]	Topical anesthesia	For the operation purposes

5. Long circulating liposomes:-

Table 6: Therapeutic applications of long circulating liposomes

Drug/ molecule	Application	Ligand
Serum proteins[17]	Increase the circulating time	Sialic acid and glyco- conjugates

6. Nanocochelates

Table 7: Therapeutic applications of Nanocochelates

System	Application	Purpose
Biogeode™ Nanocochleates[55]	Stabilize and protect an extended range of micronutrients and the potential to increase the nutritional value of processed foods	Nutrition
Nanocochleates[55]	To deliver proteins, peptides and DNA	for vaccine and gene therapy applications
Nanocochleates[55]	In the delivery of antibacterial agents: (clofazimine)	For reducing the toxicity and improving the bactericidal activity

CONCLUSION:

A number of drug delivery systems are emerging today. But the delivery systems based on Phosphatidylcholine are of much importance because of intense advantages associated with them. The delivery systems described above have proved their ability and efficacy to deliver the active moiety to the desired location of the body. But this is not the limit of these Phosphatidylcholine based drug delivery systems. In future the Phosphatidylcholine based drug delivery systems can be utilized to the maximum to proof these systems as a revolution in drug delivery technology. This review is only an attempt to attract the attention of researchers to these types of systems.

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