



Sphingosome Vesicular System

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ABSTRACT

Vesicular drug delivery system has overtaken the pre existing drugs by improving therapeutic efficacy and by sustaining and controlling action. Liposomes, herosomes, sphingosomes, ethosomes, cubosomes, pharmacosomes, niosomes, transferosomes are the newly developed vesicular drug delivery system. This article mainly deals with the sphingosomal drug delivery system. It provides a brief description about advantages and disadvantages of sphingosomes, the main classification of sphingosomes and highlights about its applications. Sphingosomes are vesicular drug delivery system which is made up of a lipid bilayer membrane enclosing an aqueous volume. Because of its wide variety of application in treatment of cancer, cosmetics, gene delivery, ocular drug delivery; it has become a better area of field of study. These systems have enhanced therapeutic index of all drugs by encapsulating these medications inside aqueous volume.

Keywords: Vesicular drug delivery, sphingolipids, lipid bilayer, cholesterol.

INTRODUCTION

Novel drug delivery system is one of the most suitable and approachable in developing delivery system. This system must deliver the drug to the body at a rate which is needed by the body, for a period of treatment. The entity is directed to the site of action.

Most of the conventional dosage doesn't fulfil these requirements so currently vesicular drug delivery system is the choice drug delivery for enhanced action¹.

Vesicular systems consist of several circular lipid bilayer which are formed when amphiphilic molecules interact with water. In 1965 Bingham reported this and is named as Bingham bodies^{2,3,4}.

These vesicular drug delivery system have emerged with different applications in the field of cosmetics, food industries and pharmaceutics. This delivery system directly delivers the drug to the infection site, which leads to less or no adverse effects with reduced toxicity. The lipid vesicular system as improved absorption as well as bioavailability of poorly soluble drugs.

Various cardiovascular agents, antidiabetic drugs, NSAIDS, proteins, drugs used in treatment of glaucoma incorporated in these vesicles has showed improved bioavailability and action in humans⁵.

Advantages

- Used for hydrophilic and hydrophobic drugs.
- These systems deliver the drug to the site of action.
- Reduce the toxicity of drugs, no adverse reactions.
- Cost of therapy is reduced by improving bioavailability of medications.
- Enhance the stability of medications.

- Elimination of rapidly metabolized drug is avoided.

Types⁶

Vesicular drug delivery is classified based on their composition. Two types are:

1. Lipoidal biocarriers
2. Non lipoidal biocarriers

Table 1 shows few examples of Lipoidal Biocarriers and Non-lipoidal Biocarriers

Table 1: Examples of lipoidal biocarrier and non-lipoidal biocarrier.

Lipoidal Biocarriers	Non-Lipoidal Biocarriers
1. Liposomes	
2. Emulsomes	
3. Ethosomes	1. Niosomes
4. Sphingosomes	2. Bilosomes
5. Transferosomes	3. Aquasomes
6. Pharmacosomes	
7. Virosomes	

Sphingosomes

Sphingosomes are the important lipid vesicular drug delivery system. They are vesicle consisting of membranous lipid bilayer which encloses an aqueous space inside where the drug can be enclosed (Figure 1).

The membranous bilayer is made of either natural or synthetic sphingolipid. Sphingosomes are mainly composed of cholesterol and sphingolipid.

The pH of the polar interior environment is less than exterior environment. Sphingosomes are having more advantages than liposomes & niosomes because of its high stability to acid hydrolysis and have improved drug



retention properties. There are several route through which sphingosomes can be administered like parenteral, inhalation oral, trasdermal route etc.

Sphingosomes are vesicular lipodal drug delivery system which is composed of many lipids like sphingolipid and cholesterol. Sphingolipid is mainly made of amide and ester linkage^{7,8}.

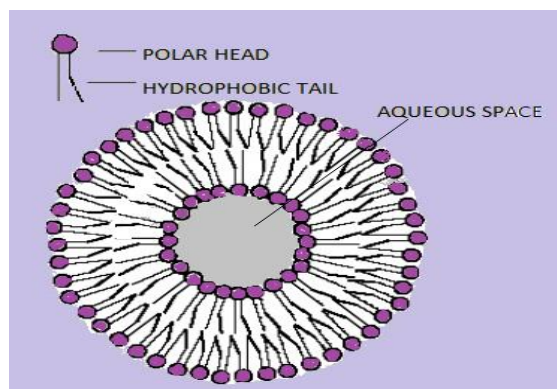


Figure 1: Structure of Sphingosome

Advantages^{9,10}

- Efficacy will be improved.
- Reduced harmfulness.
- Stability will be enhanced.
- Better transmission. (pharmacokinetic effect).
- Resist hydrolysis.
- Less chances of rancidity.
- Tumour tissues get targeted selectively.
- More targeting is achieved by coupling site with specific ligand.

Disadvantages

- Use of vesicular system is limited due to its high cost.
- Entrapment efficacy is low.

Classification¹¹⁻¹³

Classification of sphingosomes is based on the number of lipid bilayer formed and the size of vesicle. Sphingosomes can be unilamellar or multilamellar.

The typical mean diameter of sphingosomes was found to be 0.05-0.45micron. Most desirable diameter range was 0.05-0.2micron.

- Small unilamellar vesicles (SUV)
- Large unilamellar vesicles (LUV)
- Multilamellar vesicles (MLV)
- Oligolamellar vesicles (OLV)
- Multivesicular vesicles (MVV)
- Vesicles above 1 micrometre are called as Giant vesicles (GV).

Composition^{14,15}

Sphingosomes are comprised of sphingolipids and cholesterol. Sphingolipids and cholesterol occurs in a ratio of seventy five by twenty five to thirty by fifty. Most desirable ratio is 55:45.

Sphingolipids

Sphingolipids are one of the cell component and has a polar head which is joined to the hydrophobic body (Figure 2). Phospholipids functions in transmission of signals and recognition of cells. It belongs to a complex family of compounds that has a sphingoid base backbone. Sphingolipids are derived from aminoalcohol with eighteen carbon atom and unsaturated C chain, sphingosine. It is synthesized biologically from acyl-coA and serine and then it is converted in to ceramides, other lipids and other species^{16,17}.

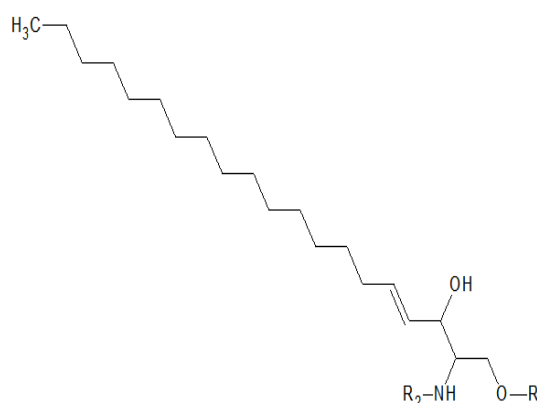


Figure 2: Structure of Sphingolipid

By considering the origin, sphingolipid can be classified as^{18,19}:

Natural Sphingolipids:

Sources:

- 1) Egg, brain, milk
- 2) Soy-bean
- 3) Plant (yeast)

Functions of Sphingolipids in Biological System

- Sphingolipids forms a stable layer of plasma membrane that protects cell membrane.
- Complex glycosphingolipids play an important role in physiology of cell by acting as antigens, growth factors and binding agents.
- Ceramide funtions as second messenger, signals the proliferation, differentiation, apoptosis, regulation and function of immune system.

Sphingosine-1-Phosphate act as 1st and 2nd messenger. It also regulate angiogenesis, cardiac development and cell growth.

Cholesterol

It is used in the production of sphingosomes. Major change has been brought in the preparation of sphingosomal bilayer membrane by incorporation of sterol. Cholesterol: sphingolipid is incorporated in the ratio 1:1 / 2:1. These results in increased separation between choline groups and normal electrostatic and H₂ bond interactions are eliminated. Targeting drug loaded sphingosomes to specific cell types can be done by addition of many other components.

Methods of Preparations of Sphingosomes

Different methods used in the preparation are

- Lipid hydration method.
- French pressure cell method.
- Solvent spherule method.
- Calcium –induced fusion method.
- Sonication method.

Lipid Film Formation (Hand Shaking Method)^{20,21}

Sphingolipids, Surfactant/cholesterol, lipophilic drug mixed together and dissolved in an organic solvent (ether) present in R.B flask. By using a rotary film evaporator the organic solvent is removed under reduced pressure. The surfactant which is dried or lipid casted film so obtained is hydrated with aqueous phase at 50-60°C. On hydration dried lipid layer swells & detaches from the inner side of RB flask and forms multi lamellar sphingosomal vesicles.

Non shaking method is also done to get large unilamellar sphingosomal vesicles, where the film is exposed to a steam of N₂ gas for 15min followed by swelling of lipid layer in aqueous media without shaking. (Figure 3.)

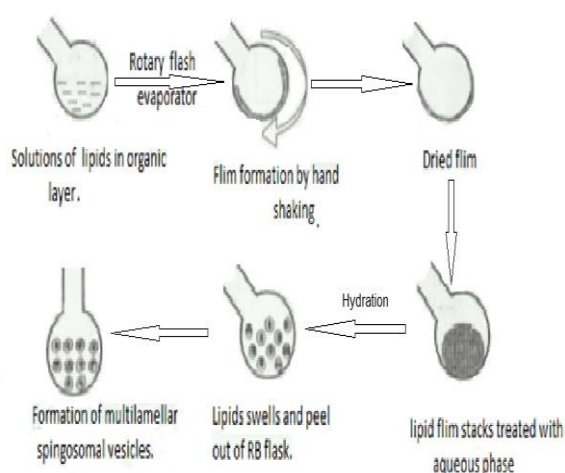


Figure 3: Preparation of Sphingosomes

Sonication Method²²

Sonication method is most commonly used method for producing small vesicles. The size of sphingosomes is reduced further on exposure to high energy levels. The

multilamellar sphingosomal vesicles are exposed to ultra sonic irradiation, which results in formation of small vesicles. 2 types of sonication methods based on both bath and probe. The widely used one is ultrasonic disintegration bath sonicator.

Solvent Spherule Method^{23,24}

In solvent spherules, the sphingolipids are dissolved in a volatile hydrophilic solvent which is then dispersed as small spheres in to an aqueous solutions. When the volatile hydrophilic organic solvent is evaporated in water bath under controlled conditions multi lamellar vesicles are formed.

Calcium Induced Fusion Method²⁵

Formation of multilamellar vesicles results, when calcium added get fused with SUV sphingosomes. Then on addition of EDTA large unilamellar vesicles sphingosomes can be prepared from multilamellar sphingosome vesicles. This method is used for encapsulation macromolecules.

French Pressure Cell Method²⁶

French pressure cell method is more useful method for producing more stable uni or oligolamellar sphingosomes when compared to sonicated vesicles.

This technique is carried out under very high pressure by using a French press. Here extrusion of performed sphingosomes is done.

Transport Mechanism of Sphingosomes²⁷

Transport mechanism at cellular level. There are various mechanisms by which small unilamellar sphingosomal vesicles (SUSV's) interact with cell such as stable adsorption, endocytosis, fusion, lipid transfer. (Figure 4.)

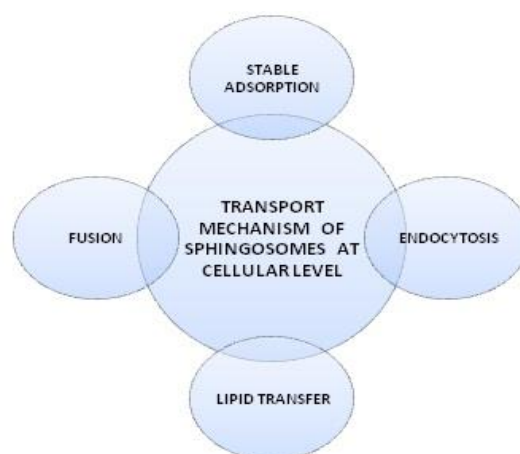


Figure 4: Transport Mechanism of Sphingosomes

Stable Adsorption

The association of intact vesicles with the cell surface is one of the method known as Adsorption.

Such process is mediated by non-specific electrostatic, hydrophobic or other forces or component present at the vesicles or cell surface. (Figure 5)



Figure 5: Adsorption Phenomena

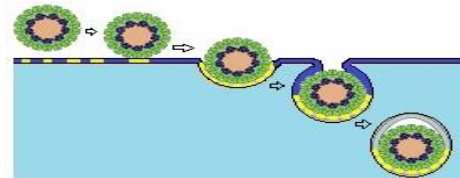


Figure 6: Endocytosis

Endocytosis

The endocytotic vesicles uptake the intact vesicles and help in their delivery to the lysosomal apparatus, this phenomenon is known as Endocytosis. (Figure 6)

Fusion

The vesicular bilayer simply merge with the plasma membrane and releases the vesicular content in to the cytoplasmic space. (Figure 7)

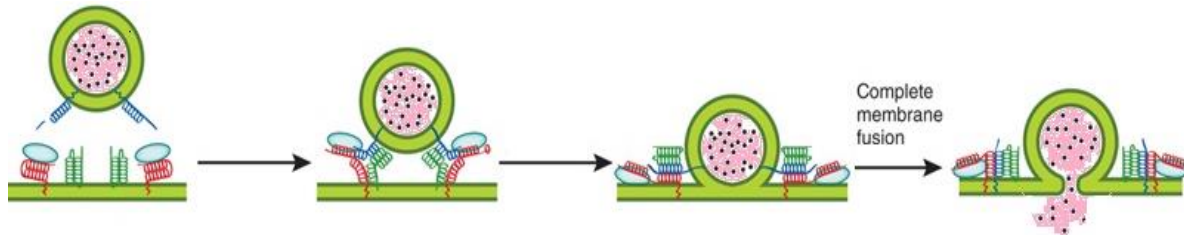


Figure 7: Fusion

Table 2: Applications of Sphingosomes in Drug Delivery System.

S. No	Drugs	Targeted Disease	Application
1)	-Vincristine -Alocrest	-Non-Hodgkins lymphomas --Non-small cell lung cancer, breast cancer	In cancer therapy
2)	-Prostaglandins -Amphotericin B -Methotrexate -Cisplatin -Vincristine -Vinblastine -Doxorubicin -Ciprofloracine -Progesterone -Testosterone -Estradiol -Doxamethasone -Other steroids	-proliferative diseases -Immune system disorders	As a drug delivery vehicle
3)	-Beclomethasone -Sphingosine Moist	-Skin or dermal therapy -skin cleansing and make-up removal efficiency	In cosmetic industry
4)	-Idoxuridine	-Accute & chronic herpatitics keatitis	In ocular delivery, enhances the penetration
5)	Streptokinase, Urokinase enzymes	-Treatment of malnutrition	Enzyme delivery
6)	-5-Fluoro uracil incombination with sphingomyelin Swasinoline in combination with interferon.	-Colonic tumour -Colon cancer and melanoma	Tumour therapy
7)	Sphingosine-1-phosphate	-Radiation induced lung injury	Gene therapy
8)	Ceramides	Rdiation induced ling injury (RILI)	Regulation of sphingosine 1-phosphate
9)	Sphingosine and sphinganine, free sphingolipids of the stratum corneum	Treating infectious disease	Anti fungal therapy
10)	Ceramides, sphingosine 1-phosphate	Immunology	Regulation of immune response

Marketed Formulations

Table 3 shows various marketed formulation of sphingosomes.

Table 3: Marked Formulations of Sphingosomes in Cancer Drug Delivery

Drugs	Marketed Formulation	Uses
Vincristine	Margibo(T.M) Oncovin®	Treatment of Lymphoblastic leukemia.
Vinorelbine	Hycumtin® Navelbine®	Treatment of lung cancer.
Topotecan	Hycumtin®	Treatment of lung cancer ovarian cancer.

Lipid Transfer

Transfer of individual lipid molecular between vesicles and the cell surface without the cell association of aqueous vesicle content.

Characterisation of Sphingosomes²⁸⁻³²

- 1) Vesicular characterization-used for measuring important vesicular characters like particle size, shape and zeta potential.
- 2) Transition temperature- For evaluating transition temperature of bilayer vesicles differential scanning calorimetry is used.
- 3) Penetration study- The mainly used method for penetration study is Confocal laser scanning microscopy (CLSM).
- 4) Vesicle stability- The vesicular stability depends on the parameters like structure and shape. Changes in shape and structure can be viewed through transmission electron microscopy (TEM).
- 5) Entrapment efficiency- Ultracentrifugation technique can be used for measuring entrapment efficiency.
- 6) Sphingolipid cholesterol interaction– Differential scanning calorimetry and P31 NM can be used.
- 7) Permeation study –By incorporating sphingosomes with gel permeation study can be done. Franz diffusion cell can be used for diffusion studies.
- 8) Surface tension activity measurement- The ring method is for used determination of surface tension in a Du Nouy ring tensiometer can be used.
- 9) Drug content- Ultra violet spectrophotometry and High performance liquid chromatography can be used for measuring the drug content.

Applications³³⁻³⁸

Table 2 summaries brief description about the drug loaded sphingosomes, targeted diseases and its applications.

CONCLUSION

Sphingosomes because of its improved drug loading capacity, stability, targeting to specific organs and tissues, its release and because of its most compatible nature and safe to the host cells is considered as most efficient novel vesicular drug delivery system.

Natural or synthetic sphingolipid is the main composition of lipid bilayer membrane which encloses an aqueous volume. There has been an increased use of sphingosomes in the field of cosmetics, pharmaceuticals, biotechnology, and medical technology.

The sphingosomes has been used for diagnostic purpose, clinically used for delivering chemotherapeutic compounds, treatment of fungal infections and is used in ocular delivery.

Because of its safe nature and compatibility US food and drug administration has approved for its use. Extensive research has been going on with sphingosomes because of its novelty and great potential and many of its applications will be investigated in the future.

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