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PHYTOSOMES: NOVEL CARRIERS FOR DELIVERY OF PHYTOCONSTITUENTS

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ABSTRACT

Phytosome technology as a novel delivery system utilizes water soluble phytoconstituents and phospholipids for drug delivery. "Phyto" means plants and "some" resembles a covering around/or a structure over the substance. Phytosomes are prepared by reacting one or two polyphenolic phytoconstituents and phospholipids in 1:1 or 1:2 ratio. Phytosomes acts as a bridge between novel and traditional drug delivery systems. Poor solubility of phytoconstituents and polyphenolic compound is barrier for their absorption and bioavailability of several phytoconstituents. Phytosomal technology can overcome the solubility problems of phytoconstituents and increases the bioavailability and absorption of water soluble phytoconstituents. Phospholipid improves both hydrophilicity and lipophilicity of phytoconstituents and hence acts as emulsifiers. Hydrophilicity (helps in dissolution of phytoconstituents in gastro-intestinal fluids) and hydrophobicity (helps phytoconstituents to cross lipid rich cell membranes) are required for better absorption and bioavailability of natural phytoconstituents. For passage of drugs through acidic and basic environment before being get absorbed into systemic circulation the DDS must have optimum water and lipid solubility. Phytosomal preparations of Olive oil, Ginkgo biloba, Grape seed and Silybum are available in the market. The current review highlights the potential scope and emerging technologies in the field of NDDS for the benefit of herbal and traditional medicines prepared from plant origins. This review covers the preparation techniques employed for phytosome preparation, advantages, applications and characterization techniques employed for phytosomal evaluation.

KEYWORDS: Phytosomes, Phytoconstituent, Liposomes, Novel drug delivery system, DDS, Phospholipids.

1. INTRODUCTION

During ancient period, plants were used for preparation of medicinal products. Plant extracts were used for treating of various diseases because of their pharmacological and therapeutic efficiency.[1] The major barrier for absorption of phytoconstituents is their polar (water soluble) nature which affects both solubility and bioavailability of plant products. Hence there is need for balance between both hydrophilic and lipophilic content for easy passage of drug products through biological membranes.

In recent years, due to advancement in novel drug delivery systems (NDDS) the barriers for absorption of phytoconstituents can be removed by preparation of phytosomes.[2] Various methods of fabrication are available for incorporating phytoconstituents into phospholipids. These products show less side effects and targeted drug delivery to specific site. The therapeutic efficiency of phytosomes is better than the traditional plant extracts due to higher absorption and bioavailability.

The chemical constituents like flavonoids and glycosides present in herbal medicines are less soluble or hydrophobic which affects their absorption through biological membranes hence affects efficiency and potency of preparation. Novel preparations like phytosomes and liposomal preparations shown better solubility, targeted site specific drug delivery and medicinal properties compared to conventional herbal extracts.

Phytosomes are nano sized drug loaded vesicles, which envelope active herbal constituent inside the vesicles. [3] The vesicle which envelopes or cover the active phytoconstituents protect the drug from degradation by gastro intestinal digestive secretions and bacteria. Hence the active drug remains safe while passing through the stomach and intestine to reach systemic circulation. Phytosomes can be utilized in treatment of major ailments without loss of their therapeutic properties.

The lower absorption and bioavailability of polyphenolic constituents is mainly due to two factors. These chief constituents are ringed molecules which are large in size that obstructs absorption by diffusion process. Second factor is that flavonoid molecule or chief constituents of polyphenol which have poor solubility with lipids. These are the limitations that inhibit their absorption through biological membrane. Phytosome technology is mainly based on complexation of polyphenols with phospholipid in 1:1 ratio or 1:2which results in the formation of phytosomal complex with lipid covering around the constituents.

First Phytosome: Milk Thistle

The first commercial phytosome preparation was extracted from the milk thistle fruit containing flavonolignan silybin, the major constituent of silymarin.[4] Flavonolignans complex are the major bioactive components extracted from the milk thistle fruit, *Silybum marianum*, *belonging to* family of Asteraceae/Compositae.

S. marianum is a medicinal plant, which has been using for thousands of years as a remedy for a variety of ailments. Since the 70s of the last century, flavonolignans presented in silymarin have been regarded to the official medicine as substances having an hepatoprotective properties. Structurally, flavonolignans are composed of a taxifolin and a coniferyl alcohol. Flavonolignans have been also studied as a potential anticancer agents, antioxidant, anti-inflammatory, and liver detoxification benefits. First phytosome preparation was initially christened IDB 1016 or Silipide and subsequently recast as Siliphos* Phytosome™.

2. COMPARISON BETWEEN PHYTOSOME AND LIPOSOME

The basic difference between liposomes and phytosomes is that in liposomes, the drug is dissolved in medium contained in layer of membrane, whereas in phytosome drug is an integral part of membrane in which the drug is attached to polar heads of phospholipids. [5]Phosphatidyl choline is mixed with water soluble substance in liposomes. In phytosomes water soluble substances are enclosed in phosphatidyl choline without formation of bonds. Difference between phytosome and liposome are tabulated in table 1. Chemical bonds are formed in liposomes. No chemical bonds are formed in phytosomes. In phytosome, phosphatidylcholine and plant compound form 1:1 or 2:1 complex depending on substance. In liposomes, hundreds or thousands of phosphatidylcholine molecules surround the water soluble molecule. Phytosomes are much better absorbed than liposomes hence they show better bioavailability. Contents of phospholipids are lesser in phytosomes compared to liposomes. The phytosome is a combination of few molecular complexes which bounded together, while the liposome is a combination of number of phospholipids which react with chief constituent but without complete bonding with them. Difference between phytosome and liposome is represented in Fig. 1. Various types of novel vesicular system and their applications are tabulated in Table 2.

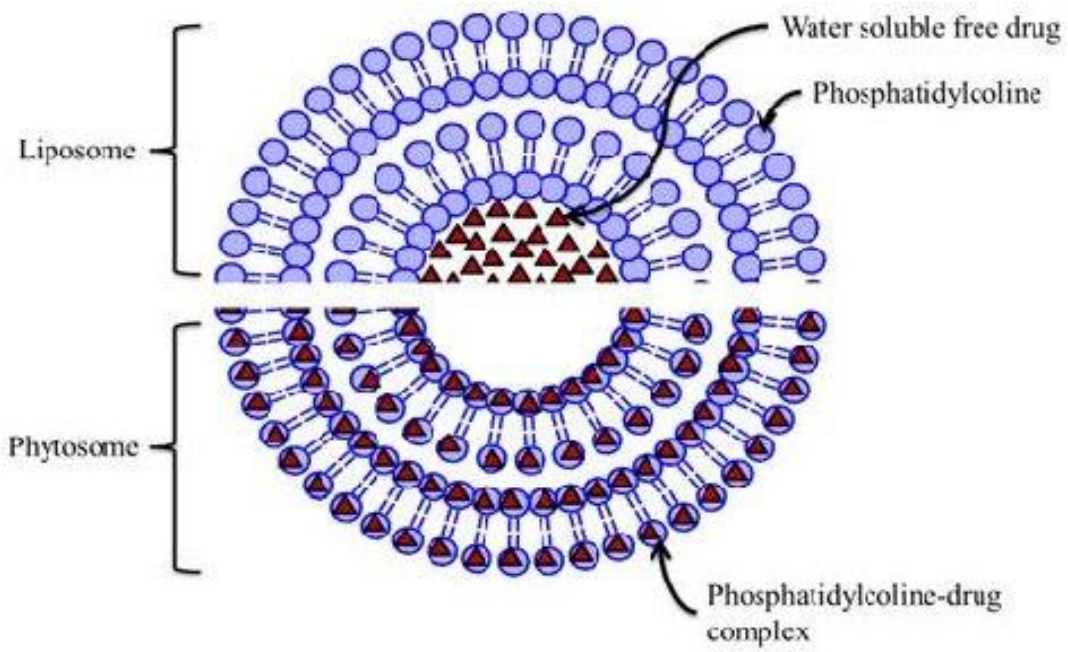


Fig. 1: Difference between phytosome and liposome.

Table 1: Difference between phytosome and liposome.

S. No.	Phytosome	Liposome
1.	Phytosome are associated with few molecule (mainly with Phospholipid and polyphenol extract)	Liposomes are associated with number of molecules
2.	Best for oral delivery with good bioavailability	Poor oral bioavailability
3.	Preferably 1:1, 1:2 Phospholipid ratio is referred for its preparation	Lipid ratio is increased up to 10 times than the chief active constituents
4.	The drug is an integral part of membrane in which the drug substance is attached to polar heads of phospholipids	The drug is dissolved in medium contained in layer of membrane
5.	No chemical bonds are formed	Chemical bonds are formed in liposomes
6.	Phosphatidylcholine and plant compound form 1:1 or 2:1 complex depending on substance.	Hundreds or thousands of phosphatidylcholine molecules surround the water soluble molecule

Table 2: Various types of novel vesicular system and their applications.

S. No.	Vesicular system	Application
1.	Pharmacosomes	Pharmacosomes are colloidal dispersion mainly consist of phospholipid designed for improvement of entrapment efficiency of polar drugs. These are also called phytosomes
2.	Ethosomes	Ethosomes designed to penetrate the skin of mammals
3.	Ufasomes	Ufasomes develop vesicles having size range nano to submicron used in topical delivery system. Ufasomes shows better stability, great entrapment efficiency both in hydrophobic/hydrphillic drugs.
4.	Sphingosomes	Sphingosomes are vesicular system which composed of either natural or synthetic sphingolipids. These are better resistant to hydrolysis and having better drug retention.
5.	Quantasomes	Quantasomes are unilamellar vesicles formed by surfactant ammonium and sterol. These are also having long stability, and their morphology do not change with time
6.	Virosomes	Vesicles having size in nano and submicron range.

3. COMPOSITION OF PHYTOSOMES

Phospholipids

In today's world, industrially produced phospholipid drug delivery play a major role and are becoming highly widespread. Soya lecithin, chicken egg, and other additives are used to prepare phytosomes.[6] The most important component in all of this is phospholipid, which is mainly composed of a glycerol unit linked to two fatty acids and a phosphate group connecting the remaining linkages. The main ingredient used in phytosomal preparation is phospholipid. The molecular structure of Phosphatidylcholine is represented in Fig. 2. Various phospholipids obtained from plants, their structure and flavonoid /chief chemical constituents are tabulated in Table No. 3

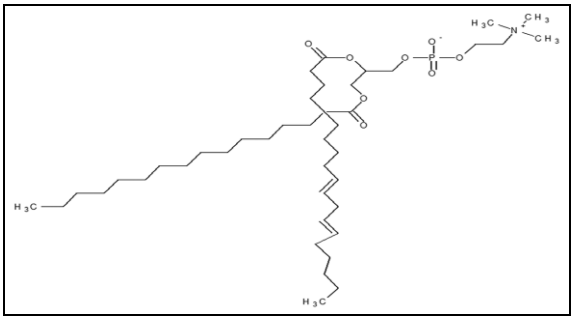


Fig. 2: Molecular structure of phosphatidylcholine.

4. COMMERCIALLY AVAILABLE PHYTOSOMAL PREPARATIONS

There are several commercially available products based on phytosomes having good therapeutic action as compared to traditional dosage form. Flavonoids are used in the preparation of phytosomes. Each active component has its own properties and therapeutic action. A number of important flavonoids are represented in table no.3 together with their source and molecular structure. Commercially available phytosomal preparations are obtained from Silybium marianum, Ginkgo biloba, Panax ginseng, Thea sinensis and Olea europaea oil. Commercially available phytosomal preparations with their source, dose and application are tabulated in table no. 4

Table 3: Plant name, structure of chief chemical constituents and name flavonoid /chief chemical constituents.

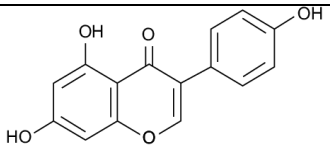
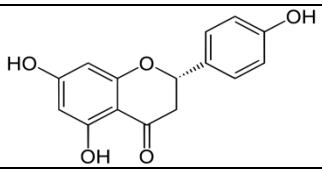
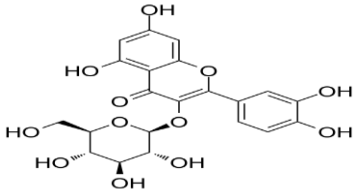
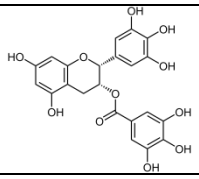
S. No.	Plant name	Structure of chief chemical constituents	Flavonoid /chief chemical constituents
1	Soy tea		Genistein
2	Orange		Naringenin
3	Onion buckwheat *Hyptis fasciculata*		Isoquercetin
4	Green tea		EGCG

Table 4: Commercially available phytosomes with their source, dose and application.

S. No.	Trade name	Chief constituents	Source	Dose	Use
1	Ginselect phytosomes	Ginsenosides	Gingko biloba	120 mg	Adaptogenic
2	Leucoselect	Polyphenols	Vitis vinifera	300 mg	Antioxidant
3	Meriva	Curcuminoids	Curcuma longa	200mg	Anti-inflammatory
4	Oleaselect TM phytosome	Polyphenols of olive oil	Olea europaea	-	Anti-inflammatory
5	Crataegus phytosomes	Vitexin-2'-O-rhamonoside	Crataegus Mexicana	-	Antioxidant

5. PROPERTIES OF PHYTOSOMES

a. Phytosomes are complex complexes made up of phytoconstituents and herbal phospholipids and the complex is generated by the reaction of phospholipids, drug and solvent.[7]
b. The association between phospholipids and substrates is due to the improvement of hydrogen bonding between the polar head of phospholipids and the polar functions of the main components.
c. When treated with hydrophilic components, phytosomes form cell-like form similar to liposomes, but in liposomes, the main constituent interacts within the inner pocket, whereas in phytosomes the drugs are wrapped in the polar head of phospholipids.
d. The phytosome is an aggregate of few molecular complexes which bounded collectively, whilst the liposome aggregate of a number of phospholipids which react with drug.

6. ADVANTAGE OF PHYTOSOMES

a) Phytosomes can be utilized to target the liver by expanding the solvency in bile salt and guarantees appropriate delivery of the drug to the targeted tissues.[8]
b) These are modern herbal drugs more successful than herbal extracts.
c) Phosphatidylcholine is utilized in the formulation of phytosomes which acts as a carrier (vehicle) moreover acts as a hepatoprotective as a result it confers a synergistic impact.
d) The retention of hydrophilic constituents is increased which increases the efficacy of phytosomes.
e) As the efficacy increases dosage requirement is decreased which leads to enhancement in the bioavailability of drug.
f) Phytosomes have better stability and duration of action.
g) Phytsosome has the capacity to pass through the skin due to its lipid layer around the phytoconstituent and in this way improve the effectiveness of the drug.[9]

h) Low dissolvability of phytosomes in fluid media allows the formation of stable emulsions or creams.
i) Phytosomes embody important components of herbal extracts, subsequently, they are ensured from destruction by digestive enzymes.

The increased bioavailability of the phytosome over the non-complexed natural derivatives has been demonstrated by pharmacokinetic studies or by pharmacodynamics tests in experimental animals and in human subjects.

7. BIOAVAILABILITY OF PHYTOSOMES[9]

It is evident from many research studies that phytosomes have an improved absorption and bioavailability when compared to the conventional herbal products. Most of the research studies are focused on *Silybum marianum* (milk thistle), the fruit of which contains a water-soluble phytoconstituent (flavonoids) which is known to have a hepatoprotective effect. But these flavonoids are poorly absorbed.[10]

8. FORMULATION OF PHYTOSOME[11]

Phytosome® complexes can be manufactured as oral and topical preparations. In order to achieve the highest efficiency of both in terms of formulation and bioavailability, phytosomes are prepared as capsules, tablets and topical preparations.

Soft gelatin capsules
Soft gelatin capsules are the perfect alternative for the formulation of Phytosomes® .The Phytosome® complex may be spread in oily vehicles to procure suspensions to be equipped in soft gelatin capsules. Vegetable or semi-synthetic oils can be used for this purpose. Not all phytosome® complexes act in the same manner when spread in oily vehicles; hence, preliminary feasibility studies should be conducted to choose the most appropriate vehicle.

Hard gelatin capsules
Hard gelatin capsules can also be utilized for filling of the phytosome® complex. 300 mg of phytosome® complex can be filled in size 0 capsules, if the powder has high density. Precompression is not required for direct volumetric filling process for high density phytosome® complex. Piston tamp capsule filling technique is utilized for low density powder filling in a capsule. Disintegration time might be affected because of precompression process. For best control of parameter preformulation studies and dry granulation might be preferable. Careful monitoring of all physical parameters is recommended for formulation of a best phytosomal complex.

Tablets
For best release of phytosomal preparation from solid dosage forms, dry granulation technique can be opted. Low quantity unit doses of phytosome® complex are prepared by direct compression process because of the limited flow ability, stickiness and low apparent density of the phytosome® complex. Phytosome® complex tablets with required characteristics can be prepared using 60-70% of diluents .The physical stability of the phytosomes is affected by wet granulation techniques due to presence of water and application of thermal procedures for drying of granules. Hence the wet granulation is not recommended for preparation of phytosomes.

Topical dosage forms
The Phytosome® complex can also be formulated as topical preparation. The best process for incorporating the phytosome® complex into the emulsion is to spread the phospholipids in a small volume of the lipid phase and mix it to the emulsion already produced at low temperatures $\leq$ 40°C.Phytosome® complexes are dispersible in the lipid solvents used in topical formulations. In the case of formulations with a small number of lipids, the phytosome® complex may also be distributed into the aqueous phase and introduced into the final formulation at temperatures $\leq$ 40°C.

9. PREPARATION OF PHYTOSOME

Accurate amount of phospholipid, i.e., Soya lecithin with herbal extracts in an aprotic solvent are required for preparation of phytosomes. [12] Soya lecithin contains main constituent, i.e., phosphatidylcholine which is having a dual function. Phosphatidyl part is hydrophobic in nature and choline part is lipophobic in nature. Choline part is attached with hydrophilic constituents, and phosphatidyl compound attached with choline bound complex. It results in the formation of lipid complex with better stability and bioavailability. Phytosomes are obtained by reacting phospholipid with selected phytoconstituents with an suitable solvent, and due to their physical and chemical efficiency, these phytocomplex can be considered as a novel entity.

2 or 3 moles (ideally with one mole) of a natural or synthetic phospholipid, such as phosphatidylcholine, phosphotidyl- ethanolamine or phosphatidyl serine, are allowed to react with one mole of phytoconstituents in an aprotic solvent, such as dioxane or acetone, in equal proportion. The ideal proportion of phospholipid to phytoconstituent is 1:1. The complex in this way shaped can be separated by precipitation with an aliphatic hydrocarbon or lyophilization or spray drying. Some liposomal drug complexes operate within the presence of water or buffer solution where the phytosomes connected with a solvent with a reduced dielectric constant.

Various techniques for phytosome preparation
a. Phytosome vesicles were made by thin layer **rotary evaporator vacuum method.**[13] The phytosomal constituents are mixed in ethanol in 250 ml RBF. The bottle was connected to the rotary evaporator. The solvent evaporates at a 60°C temperature, forming a thin coating of film around the flask. The

film is hydrated by 7.4 phosphate buffer and the lipid coating is stripped off in a phosphate buffer producing a vesicle suspension. The phytosomal suspension was subjected to sonication at 60 percent amplitude using probe sonicator. Phytosomal suspension was preserved in the refrigerator for 24 hours before evaluation.

b. Phospholipid, i.e. soya lecithin was reacted with polyphenol extract at the same ratio. 5 mL of dichloromethane (DCM) was added with stirring till it gets evaporated. After evaporation of dichloromethane, 5mL of n-hexane was added to the thin film with stirring and left in the fuming hood for removal of the solvent. After the full removal of n-hexane, the thin film was hydrated and sonicated to obtain desired phytosomal compound.
c. Weigh the exact amount of phospholipids and polyphenol extract. Place in 100 ml round bottom flask and reflux with 30 mL of DCM at 60°C for 3 hours, evaporate it, till it reaches to 5-10 mL and add 30 ml of n-hexane with continuous stirring to get a precipitate. Filter the precipitate and place it overnight in a vacuum desiccator.
d. Phytosomes can also be made using a reflux technique. Polyphenolic extract and phospholipid were being suspended in a 100 mL round bottom flask and refluxed in DCM for 1 hour at a temperature not beyond 40°C.The transparent solution was evaporated and 15 mL of n-hexane was added till precipitate was produced. The precipitate was taken and kept in the desiccator.
e. Precisely weigh required amount of phospholipids and cholesterol in a round bottom flask and dissolved in 10 mL of chloroform followed by 10 minutes of sonication using a bath sonicator. Organic solvent separation can be achieved by subjecting it to decreased pressure in a rotary evaporator at 40°C temperature. After removal of the solvent, a thin layer is formed and is hydrated by the polyphenol extract of the substance in a rotary evaporator. The phospholipids were sonicated in an ice bath for heat dissipation.

10. APPLICATIONS

Various therapeutic uses of phytosomes have been investigated in order to investigate their ability to improve the bioavailability of polar phytoconstituents.[14] Phytosomes increase capillary tone, decrease abnormal blood vessel permeability, and show potent antioxidant property. They have considerable effect for the treatment of complications with retinal blood vessels and venous insufficiency. Phytosomes delivers fatty acids, alcohols and sterols that support the wellbeing of the prostate gland. Phytosomes are used for treatment of enlargement of the prostate. Silybum marianum which contains liver protectant flavonoids are mostly used for preparing phytosomes. Milk thistle fruit plant contains flavonoids known for acion. Silymarin used to treat fatty infiltration of the liver (chemical and alcohol induced fatty liver), hepatitis, cirrhosis, and inflammation of the bile duct.

The antioxidant capacity of silymarin substantially boosts the liver's resistance to toxic substances. Green tea leaves *(Theasinensis)* is characterized by presence of a polyphenolic compound epigallo catechin 3-O-gallate as the key component. These compounds are potent modulators of several biochemical process linked to the breakdown of homeostasis in major chronic-degenerative diseases such as cancer and atherosclerosis. Green tea also furnishes us with a number of beneficial activities such as antioxidant, anti carcinogenic, anti mutagenic, hypo cholesterolemic, and cardio protective effects.

11. ADVANCES IN PHYTOSOME TECHNOLOGY[15]

There are number of research articles reveal the importance of phytosomal delivery system over conventional herbal extracts. Advances in phytosomal delivery system are as follows:

a) Bacopaside well-known chief constituents present in *Bacopa monnieri* plant having anti amnesic activity. There is remarkably great change in the therapeutic efficacy of the compound prepared by phospholipid as compare to simple *B. monnnieri* extract.
b) Another study also reveals that there is the preparation of berberine phospholipid complex solid dispersion, which not only increase the solubility of the compound but also increase its flow ability and dissolution rate for industrial production.
c) Another research states that there is the preparation of sinigrin phytosome. The study was carried out for *in vitro* wound healing capacity and the result is also appreciable as compare to sinigrin alone.
d) One research reported silymarin phytosomes with better anti hepato toxic activity as compare to silymarin alone and it also have great role for the protection against B1 aflatoxin on broiler chicks.
e) The phytosomes from standardized extract of seeds of *S. marianum* have administered orally which is having great effect on fetus from maternally ingested alcohol.
f) Grape seed phytosome also having great role in ischemia induced damage in the heart, also having protective against atherosclerosis. The main chief constituents responsible for this effect are proanthocyanidins /procyanidins.
g) Further clinical trial suggested that phytosomes of green tea free from caffeine also having a significant effect on anti obesity and antioxidant activity. It is also having effect on low-density lipoprotein.
h) Quercitin phytosomal complex reveals the better therapeutic property in rat liver injury induced by carbon tetra chloride.

12. PATENTED TECHNOLOGY OF PHYTOSOME[16]

There is numerous work has been done for commercialization of Phytosome, out of them few patents technology are representing in table 5 along with

their patent title, description of innovation and patent number.

Table 5: Patented technology of phytosome.

S. No.	Patent title	Description of innovation	Patent no.
1	Phospholipid complexes of olive fruits or leaves extracts having improved bioavailability	Having improved bioavailability	EP/1844785
2	Compositions comprising *Gingko biloba* derivatives for the treatment of asthmatic and allergic conditions	Useful for asthma and allergic condition	EP1813280
3	Treatment of skin and wound repair with thymosin beta-4	Composition of thymosin for treatment of skin	US/2007/0015698
4	Soluble isoflavone compositions	Exhibit improved solubility	WO/2004/045541
5	Phospholipid curcumin complex and piperine as chemosensitizing agent	Treatment of drug resistant	EP2228062 A1
6	Fatty acid monoesters of sorbityl furfural and composition for cosmetic and dermatological use	Fatty acid monosester of sorbityl furfural selected from two different series of compounds in which side chain is linear alkyl radical optionally containing at least one ethylenic unsaturation	EP1690862
7	Cosmetic and dermatological compositions for the treatment of aging and photo damaged skin	Cosmetic or dermatological composition for topical treatment	EP1640041
8	Complex of saponin with phospholipid and pharmaceutical and cosmetic compositions containing them	High lipophilic and improved bioavilability and suitable for use in pharmaceutical cosmetic compositions	EP0283713
9	An antioxidant preparation based on plant extract for the treatment of aging or photodamaged skin	Used in circulation problems, arteriosclerosis and high blood pressure	EP1214084

13. CHARACTERIZATION TECHNIQUES OF PHYTOSOMES[17]

1. Entrapment efficiency

By subjecting the formulation to the ultracentrifugation technology and transition temperature, the entanglement quality of a phytosomal formulation can be determined. The transition temperature of the vesicular lipid system can be calculated by differential calorimetric scanning. The drug phytosomal complex was centrifuged at 10000 rpm at 4°C for 90 minutes to separate the phytosome from the untrapped drug. [18] The concentration of drug can be determined by UV-spectroscopy. The percentage of drug concentration can be determined as the following formula:

$$\text{Entrapment efficiency} = \frac{\text{Weight of total drug} - \text{weight of free drug}}{\text{Weight of total drug}} * 100$$

2. Vesicle size and Zeta potential

The size of the particle and its appearance were measured using scanning electron microscopy. On an electron microscope, brass stab coated with gold in an ion sputter, a dry sample was mounted and screened at 100^0c. Phytosomes can be calculated by dynamic light scattering, which uses a computerized inspection test and spectroscopy of the photon correlation to determine their particle size and zeta potential. TEM has been used to characterize the phytosomes at 1000 magnification. Malvern Zetasizer is also used for the monitoring of phytosomal complex particle size and zeta size. This particle size and zeta potential can be characterized by Argon laser.

3. Surface tension activity measurement

The surface tension of phytosomal solutions can be measured by Du Nouy ring tensiometer method.

4. Spectroscopic evaluation

1H NMR: The complex formation of a phytoconstituent active with the phosphatidylcholine molecule can be predicted with 1H NMR method. Without summing up the signals which specific to specific molecules, the 1H NMR signal is considerably affected by some atoms engaged in complex formation.

13C NMR: In C6D6 at room temperature all the phytoconstituent carbons are invisible in the 13C NMR. The glycerol and choline segment signaling are broadened and others are shifted, while the majority of the fatty acid chains resonance maintains their sharp original shape.

FTIR: The structure and chemical stability of medication and phospholipid, spectroscopic examination of the complex can be validated by FTIR clearly by comparing the spectrum of the complex and the individual components and that of the mechanical mixtures. FTIR can also be seen as a valuable instrument to confirm the phytosomal complex stability. The stability can be demonstrated by comparing the complex spectrum in solid form with the micro-dispersion of water spectrum at different times following lyophilization. The phytosomes are crushed with KBr to obtain pellets at 600 kg/cm^2 pressure. FTIR scanning will be carried out between the ranges of 4000-400 cm−1.

***5.* In vitro and in vivo evaluation**

Phytosomes are subjected to a variety of spectroscopic, in vitro, and in vivo tests. To compare pharmacokinetics parameters between pure extracts and its phospholipid complex, in-vivo experiments are carried out on Beagle dogs, rodents, and wistar rats.[19] It will be determined by the drug's properties, as well as the main phytoconstituents bound to the phospholipid layer, and on this basis specific animal model will be chosen for testing.[20]

14. CONCLUSION

Denaturation and bioavailability are major constraints for development and use of natural drug products. Several approaches are available in the form NDDS to increase bioavailability and denaturation of phyto products. Despite these efforts, liposomes and phytosomes are the most promising new ways for herbal medicines to address these issues. The pharmacotherapeutic and pharmacokinetics of herbal medicines have changed because of these novel delivery mechanisms. This kind of delivery mechanism is also used in the nutraceutical and cosmoceutical industries to improve therapeutic activity and skin absorption. The preparation of phytosomes is easy and consistent, and the phospholipids used in their preparation provide their own health benefits in the body.

15. REFERENCES

1. Petrovska BB. Historical review of medicinal plants' usage. Pharmacogn Rev, 2012; 6(11): 1-5.
2. Rahman HS, Othman HH, Hammadi NI, et al. Novel Drug Delivery Systems for Loading of Natural Plant Extracts and Their Biomedical Applications. Int J Nanomedicine, 2020; 15: 2439-2483.
3. Lu M, Qiu Q, Luo X, et al. Phyto-phospholipid complexes (phytosomes): A novel strategy to improve the bioavailability of active constituents. Asian J Pharm Sci, 2019; 14(3): 265-274.
4. Di Costanzo A, Angelico R. Formulation Strategies for Enhancing the Bioavailability of Silymarin: The State of the Art. Molecules, 2019; 24(11): 2155. Published 2019 Jun 7.
5. Lu M, Qiu Q, Luo X, et al. Phyto-phospholipid complexes (phytosomes): A novel strategy to improve the bioavailability of active constituents. Asian J Pharm Sci, 2019; 14(3): 265-274.
6. Budai L, Kaszás N, Gróf P, et al. Liposomes for topical use: a physico-chemical comparison of vesicles prepared from egg or soy lecithin. Sci Pharm, 2013; 81(4): 1151-1166.
7. Kumar S, Baldi A, Sharma DK.Phytosomes: A Modernistic Approach for Novel Herbal Drug Delivery - Enhancing Bioavailability and Revealing Endless Frontier of Phytopharmaceuticals. J Develop Drugs, 2020; 9: 22-27.
8. Pavlović N, Goločorbin-Kon S, Đanić M, et al. Bile Acids and Their Derivatives as Potential Modifiers of Drug Release and Pharmacokinetic Profiles. Front Pharmacol, 2018; 9: 1283.
9. Kidd PM. Bioavailability and activity of phytosome complexes from botanical polyphenols: the silymarin, curcumin, green tea, and grape seed extracts. Altern Med Rev, 2009; 14(3): 226-246.
10. Bijak M. Silybin, a Major Bioactive Component of Milk Thistle (Silybum marianum L. Gaernt.)-Chemistry, Bioavailability, and Metabolism. Molecules, 2017; 22(11): 1942.
11. Sharma S, Sahu AN. Development, Characterization, and Evaluation of Hepatoprotective Effect of Abutilon indicum and Piper longum Phytosomes. Pharmacognosy Res, 2016; 8(1): 29-36.
12. Gnananath K, Sri Nataraj K, Ganga Rao B. Phospholipid Complex Technique for Superior Bioavailability of Phytoconstituents. Adv Pharm Bull, 2017; 7(1): 35-42.
13. Xu L, Xu D, Li Z, Gao Y, Chen H. Synthesis and potent cytotoxic activity of a novel diosgenin derivative and its phytosomes against lung cancer cells. Beilstein J Nanotechnol, 2019; 10: 1933-1942.
14. Gauttam VK, Kalia AN. Development of polyherbal antidiabetic formulation encapsulated in the phospholipids vesicle system. J Adv Pharm Technol Res, 2013; 4(2): 108-117.
15. Babazadeh A, Zeinali M, Hamishehkar H. Nano-Phytosome: A Developing Platform for Herbal Anti-Cancer Agents in Cancer Therapy. Curr Drug Targets, 2018; 19(2): 170-180.
16. Matias D, Rijo P, Reis CP. Phytosomes as Biocompatible Carriers of Natural Drugs. Curr Med Chem, 2017; 24(6): 568-589.
17. Gnananath K, Sri Nataraj K, Ganga Rao B. Phospholipid Complex Technique for Superior Bioavailability of Phytoconstituents. Adv Pharm Bull, 2017; 7(1): 35-42.

18. Javed S, Kohli K, Ali M. Reassessing bioavailability of silymarin. Altern Med Rev, 2011; 16(3): 239-249.
19. Adouani I, Qureshi AS, Hang TJ. Preparation, evaluation and pharmacokinetics of diosmin herbosome in beagle dogs. Pak J Pharm Sci, 2020; 33(1): 33-40.
20. Patra JK, Das G, Fraceto LF, et al. Nano based drug delivery systems: recent developments and future prospects. J Nanobiotechnology, 2018; 16(1): 71.