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Review Article

PHYTOSOME: RECENT ADVANCE RESEARCH FOR NOVEL DRUG DELIVERY SYSTEM

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ABSTRACT

Development of novel drug delivery systems (NDDS) is a new approach for plant extracts and active components. The available approaches for novel herbal formulations like polymeric nanoparticles, nanocapsules, liposomes, phytosomes, nanoemulsions, microsphere, niosomes, planterosomes, cubosomes, transferosomes, and ethosomes has been reported using bioactive and plant extracts. Phytosome is a patented technology that was introduced and developed by a leading herbal drug manufacturer and nutraceuticals. In phytosomes, incorporated the standardized extracts of plant or water soluble phytoconstituents was improved into phospholipids to form a lipid compatible molecular complex. These phytosomes improve the absorption and bioavailability of drug. This novel formulation has remarkable advantages over conventional formulations of plant actives and extracts which includes enhancement of solubility, bioavailability, ability to cross the cell membranes, protection from toxicity, enhancement of stability, sustained delivery, and protection from physical and chemical degradation. The present review summarizes the water soluble phytoconstituents (like flavonoids, tannins, terpenoids, etc.) that are poorly absorbed either due to their large molecular size or due to their poor lipid solubility, resulting of poor bioavailability, methods of preparation, particle size and shape, entrapment efficiency, route of administration, biological activity and applications of novel formulations.

Keywords: *Novel drug delivery systems (NDDS); Phytosomes; Herbal drugs; Phospholipids.*

INTRODUCTION

Over the last century, there are valuable criteria has been proceed on the development of novel drug delivery system (NDDS) for herbal drugs. Development of novel drug delivery systems (NDDS) is a new approach for plant extracts and active components [1]. Novel drug delivery system aims to deliver the drug at a rate directed by the needs of the body during the period of treatment, and channel the active entity to the site of action. A number of novel drug delivery systems have emerged encompassing various routes of administration, to achieve controlled and targeted drug delivery.

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Encapsulation of the drug in vesicular structures is one such system, which can be predicted to prolong the existence of the drug in systemic circulation, and reduce the toxicity, if selective uptake can be achieved [2].

The term “Phyto” means plant while “some” means cell-like structure [3, 4]. It is a patented technology that was introduced and developed by a leading herbal drug manufacturer and nutraceuticals®. The bioavailability of active principles of plants has become an issue of concern for researchers and scholars because of poor oral bioavailability of many plants specifically those containing polyphenolic rings in their structures such as flavonoids and other water soluble constituents like terpenoids and tannins. Some of the basic reasons for the poor bioavailability of these substances are low water or lipid solubility, high molecular

weight/size and poor plasma membrane permeability [5 - 8]. To overcome these problems and to make herbal therapy more effective, these drugs have been incorporated into several novel delivery systems in the recent time. Some of the approaches for bioavailability enhancement are formulating at nano scale as nanoparticles, binding with lipids as liposomes or herbosomes / phytosomes, delivery in the form of micro emulsions, modification in chemical structures, delivery as prodrug and complexation with cyclodextrins [5, 9, and 10].

In recent years, the technique of complexing plant drugs or extracts with phospholipids has emerged as a challenging and one of the most successful methods for improving bioavailability and therapeutic efficacy of a number of poorly absorbed plant constituents. This technique incorporates the phospholipids molecules containing phosphatidylcholine in their structure to form complexes with standardized herbal extracts and/or the specific bioactive ingredient of plant which improves the membrane permeability, water–oil partition coefficient, enhance the systemic bioavailability, enhancement of solubility, ability to cross the cell membranes, protection from toxicity, enhancement of stability, sustained delivery, and protection from physical and chemical degradation of the drugs, called as phytosomes [11-12]. The incorporation of water soluble drugs into their phospholipids complexes has considerably enhanced their bioavailability by increasing penetration through the lipoid plasma membrane while the phospholipids complexation of poorly water soluble drugs had increased their bioavailability by improving their solubility in gastric fluids [13-14]. The phyto-phospholipid complexation technique in recent years has made it possible to administer high efficacy plant actives with improved biological profile.

Phospholipids molecule has arisen as a potential and unique carrier system for improving the bioavailability of poorly absorbed plant extracts/actives because of their unique structural components, which are similar to the lipid content of the mammalian cell membrane that makes them highly compatible with the human physiological system [15]. It is present in egg yolk, brain tissue and a wide variety of animal fat and plant oils. It is routinely present in the bile fluid, to help emulsify food ingredient for absorption.

Phospholipids molecules are amphipathic having considerable solubility in aqueous and oily mediums. They have a polar and a non-polar portion in their structures [16]. It possesses a cylindrical shape with highest entropy and is involved in formation of bilayer. It contains one saturated and one unsaturated chain in its structure. Phosphatidylethanolamine is cone shaped and doesn't form bilayer itself [17].

Naturally occurring phospholipids incorporate an unsaturated fatty acid (such as oleic acid, linoleic acid or arachidonic acid) in position 2 and a saturated one (such as stearic acid or palmitic acid) in position 1 [18]. The most commonly used phospholipids (Fig.1) are those derived from soya bean containing higher proportions that is about 76% of phosphatidylcholine with a high content of polyunsaturated fatty acids like linoleic acid about 70%, linolenic acid and oleic acid [19]. The soy phospholipids are absorbed at a rate greater than 90% in humans and reach peak plasma concentration in about 6 h after oral administration. The maximum plasma concentration reached was found to be 20% of the dose administered [20]. The phospholipids especially those containing phosphatidylcholine have shown to be incorporated in the cell membrane to replace cellular phospholipids and thus affect the fluidity of the membrane [21]. The essential or soya phospholipids have shown to be hepatoprotective in nature and prevent liver damage by alcohol, drugs and other toxins [22]. They have also been reported to aid in clearance of serum cholesterol and increase circulating HDL levels in plasma [23]. The presence of proportionally larger amounts of poly-unsaturated fatty acids in soy phospholipids makes it potentially useful in reducing the risk of coronary heart disease. Essential phospholipids have also shown to possess antilipemic and antiatherogenic effects by impeding the upsurge of total lipids in

dietetic hypercholesterolemia in therapeutic as well as prophylactic doses [24].

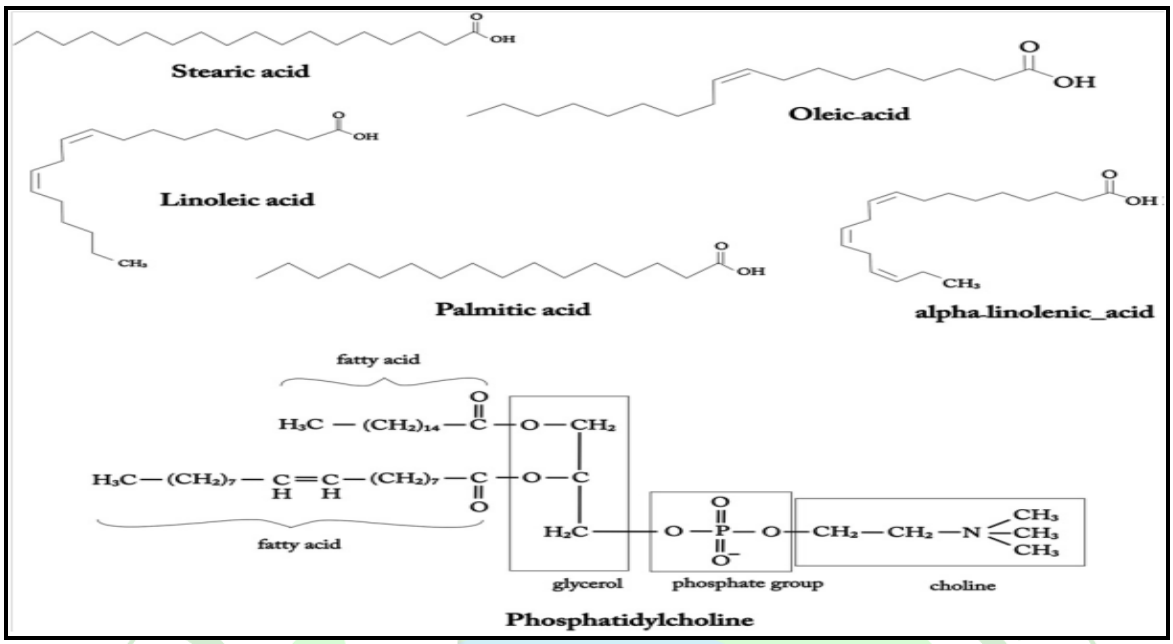

Figure 1: Different fatty acids constituting phosphatidylcholine.

MECHANISM OF PHYTOSOME FORMATION

The polyphenolic constituents of plant extracts lend themselves quite well for direct binding to phosphatidylcholine. Phytosomes are formed from the reaction of the phospholipids like phosphatidylcholine with the standardized extract or polyphenolic constituents like simple flavonoids in aprotic solvent [25-26]. Phosphatidylcholine is a bifunctional compound, the phosphatidyl moiety being lipophilic and the choline moiety being hydrophilic in nature. Specifically the choline head of the phosphatidylcholine binds to these compounds while lipid soluble phosphatidyl portion comprising the body and tail which then envelopes the choline bound material. Hence, the Phytomolecules produce a lipid soluble molecular complex with phospholipids called as phyto-phopholipid complex. Phytomolecules are anchored through chemical bonds to the polar choline head of phospholipids, as can be demonstrated by specific spectroscopic techniques [27]. Often Precise chemical analysis indicates the unit phytosome is usually a flavonoid molecule linked with at least one phosphatidylcholine molecule. The result is a little microsphere or cell is produced [28].

These drug-phospholipids complexes can be formulated in the form of solution, suspension, emulsion, syrup, lotion, gel, cream, aqueous micro dispersion, pill, capsule, powder, granules and chewable tablet phosphatidylcholine resulting in a product that is better absorbed and produces better result than the conventional herbal extracts.

ADVANTAGES OF PHYTOSOMES OVER CONVENTIONAL DOSAGE FORMS

Improved absorption

There is a dramatic enhancement of the bioavailability of plant extracts or bioactive components due to their complexation with phospholipids and improved absorption in the intestinal tract [29-30].

Cosmetic use

The formulation of phytosomes is safe and the components have all been approved for pharmaceutical aid and cosmetic use [27, 31]. They can be also used for enhanced permeation of drug through skin for transdermal and dermal delivery [32]. They can be widely used in cosmetics due to their improved skin penetration and have a high

lipid profile. Phytosomal formulations can be used as functional cosmetics [27].

Protective in nature

They have been used to deliver liver-protecting flavonoids because they can be made easily bioavailable by phytosomes [33]. In addition to this, phosphatidylcholine is also hepatoprotective and so provides a synergistic effect for liver protection.

Cost-effective

This technology offers cost-effective delivery of phytoconstituents and synergistic benefits when used as functional cosmetics to protect the skin against exogenous or endogenous hazards in normal as well as stressful environmental conditions [27].

As a carrier

Phosphatidylcholine, an essential part of the cell membrane used in phytosome technology that acts as a carrier and also nourishes the skin.

Enhance the entrapment efficiency

There is no problem with drug entrapment during formulation preparation. Also, the entrapment efficiency is high and more over-predetermined, because the drug itself forms vesicles after conjugation with lipid.

Improve the stability

They offer a better stability profile because chemical bonds are formed between the phosphatidylcholine molecules and phytoconstituents. The phytosomal system is passive, non-invasive and is suitable for immediate commercialization.

Dose reduction

The dose requirement is reduced due to improved absorption of the main constituent. They can also be given in smaller quantities to achieve the desired results.

Low risk profile

This technology has no large-scale drug development risk since the toxicological profiles of the phytosomal components are well documented in the scientific literature.

PROPERTIES OF PHYTOSOMES

Physico - Chemical properties

Phytosome is a complex between a natural product and natural phospholipids, like soy phospholipids. Such a complex is obtained by reaction of stoichiometric amounts of phospholipids and the substrate in an appropriate solvent. On the basis of spectroscopic data it has been shown that the main phospholipids-substrate interaction is due to the formation of hydrogen bonds between the polar head of phospholipids (i.e. phosphate and ammonium groups) and the polar functionalities of the substrate [25, 29].

Biological properties

Pharmacokinetic and pharmacodynamic studies in experimental animals and in human subjects have been used to demonstrate the biological behavior of phytosomes [29].

METHODS OF PREPARATION OF PHYTOSOME

The phytosome is a cell like structure, which is a combination of soy lecithin with standardized extracts containing polyphenolic compounds, which improves their absorption and utilization [34].

Solvent evaporation technique

The complex of plant extracts or specific active principles with dietary phospholipids is generally prepared by solvent evaporation techniques using alcoholic or organic solvents as reaction medium. In the more frequently used solvent evaporation technique the drug and the phospholipids are placed in the same flask containing a suitable solvent system such as tetrahydrofuran or ethanol. The reaction is allowed to be carried out at suitable fixed temperature for a fixed duration of time to get maximum possible yield and drug entrapment.

Research work based on formulated marsupsin–phospholipid complex using mechanical dispersion oriented liquid antisolvent precipitation process. They dissolved soy lecithin in diethyl ether by sonication and marsupsin in double distilled water. The drug solution was then added drop wise to the phospholipids solution with

sonication. The resultant formulation was then refrigerated and on analyzing the complex showed 44% entrapment of marsupsin with 20% cumulative drug release [35].

Super critical fluids (SCF)

The super critical fluids (SCF) have emerged as an effective tool for preparing particles of size ranging from 5 to 2000 nm. Different methods of supercritical fluid have been utilized for improving solubility profiles of poorly soluble drug candidates some of which are compressed antisolvent process (PCA), supercritical antisolvent method (SAS), rapid expansion of supercritical solutions (RESS), gas anti-solvent technique (GAS) and solution Enhanced dispersion by supercritical fluids (SEDS).

Research work incorporated the supercritical fluid technique for preparing puerarin-phospholipids complex. They have formulated the complex by three different conventional methods viz. solvent evaporation, lyophilization and micronized puerarin and compared them qualitatively with the complex prepared by the supercritical antisolvent precipitation technique. Two SCF techniques viz. GAS and SEDS were used for preparation of complexes [36, 37].

Gas anti-solvent technique (GAS)

In the GAS technique, a supercritical antisolvent was added to the drug and phospholipids solutions separately until the final pressure was reached. The reaction vessel was then kept for 3 h without any agitation at a fixed temperature of 38 °C with 10 mPa of pressure.

Solution Enhanced dispersion by supercritical fluids (SEDS)

The SEDS technique, the liquid solution and the supercritical antisolvent were continuously added into the precipitation unit. Carbon dioxide gas was allowed to pass through a nozzle of 0.1 mm diameter into the mixture of phospholipids and puerarin in the solvent. The experimental conditions were optimized with temperature of 35 °C, pressure of 10 mPa, 1% mass ratio of drug to phospholipids and a 100 mg/ml concentration of puerarin. The resultant method produced a complex of 93% yield. The morphology of the product obtained from the SEDS technique was found to be in the form of aggregated particles with ordered appearance ranging in size of about 1 µm, while those formed by conventional methods were in the form of nubby granules with fused or viscous plates. Surface area of particles formed by the SEDS technique increased from 0.50 to 1.08. The phospholipids complex prepared by the supercritical techniques exhibited rapid dissolution with an increase of 1.91 folds from 2.87 mg/ml of puerarin to 5.49 mg/ml of its phospholipids complex.

The product of the supercritical GAS technique has shown to be having more precisely controlled morphological characteristics while the particles from the SEDS technique represented complete loss of crystalline.

Anti-solvent precipitation technique

The traditional anti-solvent precipitation technique has also been utilized by many researchers by incorporating n-hexane as the antisolvent to precipitate out the drug phospholipids complex (Fig. 2) from the organic solvent [29].

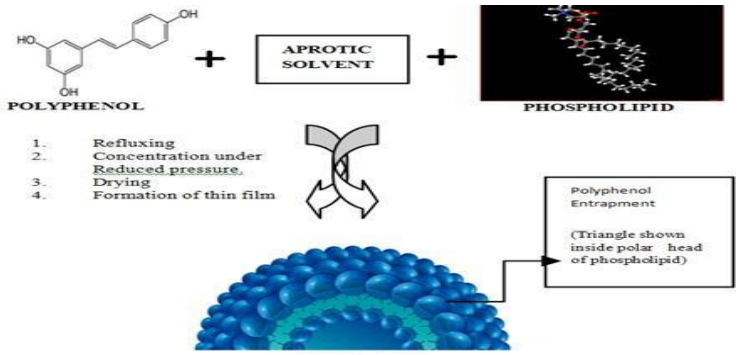

Figure 2: Common stages for preparation of phytosome.

Research work based on patented a similar method for preparing a phyto-phospholipid complex of andrographolide using dichloromethane as the reaction medium and n-hexane as the anti -solvent for final precipitation of the product. The solution is then evaporated off and residue dried usually under vacuum [38]. In a more recent work based on prepared a rutin–phospholipid complex by an anhydrous co-solvent lyophilization method in which the drug and the phospholipids were dissolved in methanol but in separate vessels. Both the solutions were mixed with mechanical stirring till all the solvents evaporated out. The photomicrography showed the rutin - phospholipid complex in amorphous state in contrast to rutin which is crystalline. A drug to Phospholipids ratio of 1:3 yielded comparatively superior experimental results [39].

Some specifications

Complexation of a specific plant active constituent or a group of structurally similar plant actives with phospholipids has been performed at different molar ratios ranging from 0.5:1 to 3:1. In most of the research works a stoichiometric ratio of 1:1 has been considered most suitable for formulating a complex. Research work based on optimized the formulation of the oxymatrine–phospholipid complex by using a composite design technique and had used drug to phospholipids ratios of 1:1, 1.4:1, 2:1, 2.6:1 and 3:1, respectively. The ratio of 3:1 at 60 °C for 3 h produced the complex with highest yield [40]. Research work based on optimized the formulation of Bergenin with phospholipids using a statistical model incorporating polynomial and interactive terms. The optimal formulation was formed with a drug to phospholipids ratio of 0.9, drug concentration of 80 g/l and a temperature of 60 °C. The combination percent of the resultant formulation was found to be 100% and drug content in the complex was 45.98% [41].Research work based on prepared an embelin–soya phosphatidylcholine complex using molar ratios of 1:0.5 to 1:3. The formulation at a ratio of 1:3 exhibited the highest entrapment efficiency of 83.4%. They concluded that the entrapment efficiency of embelin which is lipophilic in nature increases with an increase in its aqueous solubility [42]. Different solvents have been utilized by different researchers as the reaction medium for formulating herbosomes. Aprotic solvents like methylene chloride, ethyl acetate etc. has been used for preparing complexes but they have been largely replaced by protic solvents like ethanol. Others have used tetrahydrofuran as the reaction medium [43]. Others have used dichloromethane as the solvent and n-hexane as the medium for precipitation of the complex [38]. Most of the recent work has been done using absolute ethanol as the reaction medium. Apart from the solvent system, different researchers have used phospholipids from different sources. The common criterion for selection was the ratio of phosphatidyl group present in them. Soy lecithin, phosphatidylserine, and 1, 2-distearoyl-sn-glycero-3-phosphocholine is some of the phospholipids used. However, the phospholipids of soya bean have been the phospholipids of choice because of the higher content of phosphatidylcholine in them, which offers compatibility and similarity with the mammalian plasma membrane [44].

CHARACTERIZATION AND EVALUATION OF PHYTOSOMES

The behavior of phytosomes in both physical and biological systems is governed by factors such as the physical size, membrane permeability, percentage of entrapped solutes, and chemical composition as well as the quantity and purity of the starting materials. Therefore, phytosomes can be characterized in terms of their physical attributes i.e. shape, size, distribution, percentage drug captured, entrapped volume, percentage drug released and chemical composition [29, 45].

Different characterization techniques used for phytosomes

Entrapment efficiency

The entrapment efficiency of a drug by phytosomes can be measured by the ultracentrifugation technique [46].

Vesicle size and Zeta potential

The particle size and zeta potential can be determined by dynamic light scattering (DLS) using a computerized inspection system and photon correlation spectroscopy (PCS) [47].

Visualization

Visualization of phytosomes can be achieved using transmission electron microscopy (TEM) and by scanning electron microscopy (SEM) [48-49].

Drug content

The amount of drug can be quantified by a modified high performance liquid chromatographic method or by a suitable spectroscopic method [50].

Transition temperature

The transition temperature of the vesicular lipid systems can be determined by differential scanning calorimetry [49].

Surface tension activity measurement

The surface tension activity of the drug in aqueous solution can be measured by the ring method in a Du Nouy ring tensiometer [51].

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Vesicle stability

The stability of vesicles can be determined by assessing the size and structure of the vesicles over time. The mean size is measured by DLS and structural changes are monitored by TEM [49].

Spectroscopic evaluations

To confirm the formation of a complex or to study the reciprocal interaction between the phytoconstituent and the phospholipids, the following spectroscopic methods are used [27, 52].

Fourier Transform Infrared (FTIR) spectroscopy studies

The formation of the complex can be also be confirmed by IR spectroscopy by comparing the spectrum of the complex with the spectrum of the individual components and their mechanical mixtures. FTIR spectroscopy is also a useful tool for the control of the stability of phytosomes when micro-dispersed in water or when incorporated in very simple cosmetic gels. From a practical point of view, the stability can be confirmed by comparing the spectrum of the complex in solid form (phytosomes) with the spectrum of its micro-dispersion in water after lyophilization, at different times. In the case of simple formulations, it is necessary to subtract the spectrum of the excipients (blank) from the spectrum of the cosmetic form at different times, comparing the remaining spectrum of the complex itself.

^{1}H-NMR

NMR spectra of (+) - catechin and its stoichiometric complex with distearoylphosphatidylcholine have been studied [27]. In nonpolar solvents, there is a marked change of the ^{1}H-NMR signal originating from the atoms involved in the formation of the complex, without any summation of the signal peculiar to the individual molecules. The signals from the protons belonging to the flavonoids are to be broadened that the proton cannot be relieved. In phospholipids, there is broadening of all the signals while the singlet corresponding to the N-$(CH3)_3$ of choline undergo an uplift shift. Heating the sample to 60˚ results in the appearance of some new broad bands, which corresponding mainly to the resonance of the flavonoid moiety.

^{13}C-NMR

In the ^{13}C-NMR spectrum of (+)-catechin and its stoichiometric complex with distearoylphosphatidylcholine, particularly when recorded in C_6D_6 at room temperature, all the flavonoid carbons are clearly invisible. The signals corresponding to the glycerol and choline portion of the lipid (between 60–80 ppm) are broadened and some are shifted, while most of the resonances of the fatty acid chains retain their original sharp line shape. After heating to 60˚, all the signals belonging to the flavonoid moieties reappear, although they are still very broad and partially overlapping.

In vitro and in vivo evaluations

Models of in-vitro and in-vivo evaluations are selected on the basis of the expected therapeutic activity of the biologically active phytoconstituents present in the phytosomes [53]. For example, in-vitro antihepatotoxic activity can be assessed by the antioxidant and free radical scavenging activity of the phytosomes. For assessing antihepatotoxic activity in-vivo, the effect of prepared phytosomes on animals against thioacetamide-, paracetamol or alcohol- induced hepatoxicity can be examined [54]. Skin sensitization and tolerability studies of glycyrrhetinic acid-Phytosome® ointment, a commercial product, describe the in vivo safety evaluation methodology [55].

DIFFERENCE BETWEEN PHYTOSOMES AND LIPOSOMES

Layer of membrane

The basic difference between liposomes and phytosomes is that in liposomes the active principle is dissolved in the medium contained in the cavity or in the layers of the membrane, whereas in the phytosomes it is an integral part of the membrane, being the molecules anchored through chemical bonds to the polar head of the phospholipids shown in (Fig. 3).

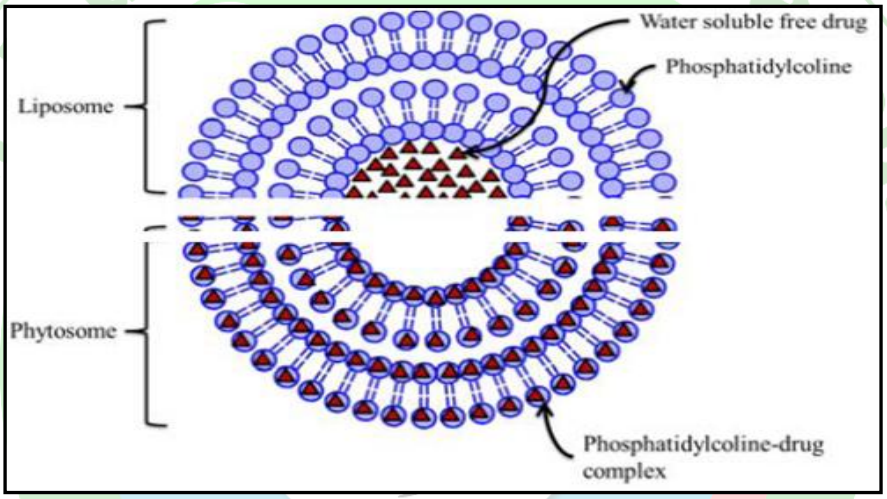

Figure 3: Difference between phytosome and liposome. The molecular organization of phytosomes (lower segment) liposome (upper segment).

Layer of phospholipids

Liposomes are used primarily in cosmetics to deliver water-soluble substance to the skin. A liposome is formed by mixing a water-soluble substance. There may be hundreds or even thousands of phosphatidylcholine and the individual plant components actually from a 1:1 or a 2:1 complex depending on the substance. On the contrary, in a phytosomes, the active principle can be compared to an integral part of the lipid membrane [25].

Content of phospholipids

Furthermore, in liposomes the content of phospholipids is much higher; about five times one in phytosomes, making this delivery from not suitable for oral clinical realistic dosages for natural compounds.

Absorbance profile

This difference result in phytosomes being much better absorbed than liposomes.

THERAPEUTIC APPLICATIONS OF PHYTOSOMES

To examine the various advantages of phytosomes, especially their ability to enhance the bioavailability of polar phytoconstituents, various therapeutic applications of phytosomes have been explored. The details of the type of phytosomes, active constituents, the daily dose And specific indications are given in Table 1.

TABLE I: Therapeutic Applications Of Different Phytosomes With Their Dose

Sr. No	Trade Name	Phytoconstituents complex	Daily Dose (mg)	Indications
1	Silybin phytosome	Silybin from *Silibium marianum*	120	Hepatoprotective, Antioxidant
2	Silyphos milk thistle	Silybin from *Silibium marianum*	150	Antioxidant, Hepatoprotective
3	Grape seed phytosome	Procyanidins from *vitis vinifera*	50-300	Antioxidant, Anticancer
4	Ginseng phytosome	Ginsenosides from *panax ginseng*	150	Immunomodulator
5	Hawthorn phytosome	Flavonoids from *crataegus species*	100	Antihypertensive, Cardioprotective
6	Sericoside phytosome	Sericoside from *Terminalia sericea*	–	Skin improver, Anti-Wrinkles
7	Ginko phytosome	Flavonoids from *Ginkobiloba*	120	Anti aging, Protects Brain & Vascular lining
8	Olea phytosome	Polyphenols from *Oleaeuropea*	120	Anti–hyperlipidemic, Anti-inflammatory
9	Green phytosome	Epigallocatechin from *Thea sinensis*	50-300	Anti-Cancer, Antioxidant
10	Echinacea phytosome	Echinacosides from *Echinacea angustifolia*	–	Immunomodulatory, Nutraceuticals
11	Bilberry (Mertoselect)	Anthocyanosides from *Vaccinium myritillus*	–	Antioxidant, Improvement of
12	Palmetto (sabalselect)	Fattyacids,alcohols&sterols from *Serenoarepens*	–	Anti-oxidant, Benign Prostatic hyperplasia
13	Visnadine (visnadax)	Visnadine from *Ammi visnaga*	–	Circulation Improver, Vasokinetic
14	Centella phytosome	Terpens from *Centella asitica*	–	Brain tonic, Vein and Skin Disorder
15	Glycyrrhiza phytosome	18-β glycyrrhetinic acid from *Glycyrrhiza glabra*	–	Anti-inflammatory ,Soothing
16	Melilotus phytosome	Triterpens from *Melilotus officinalis*	–	Hypotensive, Indicated in Insomnia
17	Curcumin phytosomes	Polyphenol from *Curcuma longa*	200-300	Cancer Chemo preventive Agent
18	Merto phytosome	Polyphenols, Antcinoside from *Vaccinium myrtilus*	–	Antioxidant
19	PA_2 phytosome	Proanthocyanidin A_2 from horse *Chestnut bark*	–	Anti-Wrinkles, UV protectant
20	Escin β sitosterol phytosome	Escin β-sitosterol from horse *Chestnut fruit*	–	Anti-Odema

SOME PATENTED TECHNOLOGIES RELATED TO PHYTOSOMES

There are a number of innovative processes and formulation research studies in the field of phytosomes carried out by a number of academic scientists as well as by industrial laboratories. Some patents for phytosomes and other related technologies along with their applications and innovations are listed in Table 2.

TABLE II: Some Patented Technologies Related To Phytosome.

Sr. No	Title of Patent	Innovation	Patent No	References
1	Phospholipid complexes of olive fruits or leaves extracts having improved bioavailability	Phospholipids complexes of olive fruits or leaves extracts or compositions containing it having improved bioavailability	EP/1844785	Franceschi et al. 2007
2	Compositions comprising Ginkgo biloba derivatives for the treatment of asthmatic and allergic conditions	Compositions containing fractions deriving from Ginkgo biloba, useful for the treatment of asthmatic and allergic conditions	EP1813280	Pierro, 2007
3	Fatty acid monoesters of sorbityl furfural and compositions for cosmetic and dermatological use	Fatty acid monoesters of sorbityl furfural selected from two diff series of compounds in which side chain is a linear or branched C3 -C19 alkyl radical optionally containing at least one ethylenic unsaturation least one ethylenic unsaturation	EP1690862	Bertelli et al. 2006
4	Cosmetic and dermatological composition for the treatment of aging or photo damaged skin	Composition for topical treatment of the skin comprises a substance that stimulates collagen synthesis and a substance that enhances the interaction between extracellular matrix and fibroblasts Cosmetic or dermatological composition for topical treatment	EP1640041	Doering et al. 2006
5	Treatment of skin, and wound repair, with thymosin beta 4	Compositions and methods for treatment of skin utilizing thymosin β4	US/2007/ 0015698	Kleinman et al. 2007
6	Soluble isoflavone composition	Isoflavone compositions exhibiting improved solubility (e.g., light transmittance), taste, colour, and texture characteristics, and methods for making the same	WO/2004/ 045541	Khare, 2004
7	An anti-oxidant preparation based on plant extracts for the treatment of circulation and adiposity problems	Preparation based on plant extracts which has an anti-oxidant effect and is particularly useful in treatment of circulation problems such as phlebitis, varicose vein, arteriosclerosis, hemorrhoid and high blood pressure.	EP1214084	Merizzi, 2002
8	Complexes of saponins with phospholipids and pharmaceutical and cosmetic compositions containing them	Complexes of saponins with natural or synthetic phospholipids have high lipophilic and improved bioavailability and are suitable for use as active principle in pharmaceutical, dermatologic and cosmetic compositions	EP0283713	Bombardelli et al. 1987

RECENT ADVANCED RESEARCH IN PHYTO - PHOSPHOLIPIDS COMPLEXATION

Numerous researches is being conducted by the researchers and the recent researches reveals that the phytosome technology is a novel method for improving the absorption and bioavailability of plant extracts significantly reducing the dose level. Some plant extracts are getting more focus now-a-days due to their potential pharmacological effects, such as, silymarin, grape seed extract, quercetin, curcumin, hesperetin, ginkgo biloba extract, andrographolide etc. The suitability of this technique and increased demand of herbal medicines for various disease management in current scenario, has paved the way of newer researches. Some of the crucial works of various researchers are briefly given below.

Researcher observed that phospholipids exhibit a marked affinity for some classes of flavonoids. They developed a new series of compounds called Phytosomes, which he obtained by complexation of phospholipid with very polar botanical derivatives, i.e. complexes between a pure phospholipids and pure active principle. He first established some chemicophysical properties of phytosomes of catechin, quercetin, escin and glycyrrhetinic acid and gathered some pharmacological data. He, from the beginning described phytosomes to be a good carrier for phytoconstituents across the skin [27].

Researchers used nine human volunteer patients and tested the hepatoprotective activity of silymarin and reported that phytosomal form of silybin possess four times greater passage through the liver [56, 57].

Scientist prepared Gingko biloba terpenes phytosomes and reported that the phytosome was effective on soothing individual contact reaction to other substances contained in topical formulation [58].

Researcher conducted series of experiments on silymarin phytosomes and reported the better fetoprotectant activity of the phytosomal form [59].

Researcher also reported better fetoprotectant activity of silymarin phytosome than uncomplexed silymarin against ethanol-induced behavioural deficit [60].

Scientist reported that the silymarin phytosomes showed much higher specific activity and a longer lasting action than the single constituent, with respect to percent reduction of edema, inhibition of myeloperoxidase activity, antioxidant and free radical scavenging activity [61].

Researchers also developed naringenin and curcumin phytosomes in two different studies and reported that antioxidant activity of the phytosomal complex has better therapeutic efficacy. They developed simple methods of preparation of phytosomes [62, 63].

Scientist investigated that the well known soothing activity of silymarin is increased by more than six times in sylimarin phytosome in experimental models. The improvement in the activity of the phytosome form compared to the free active principle is due to a higher affinity of the complex for skin phospholipids [64].

Scientist reported the hydration of the superficial corneous layer is related to the liposomal like properties of the phospholipids of the complex. Ginselect phytosomes posses a transdermic action which helps the ginseng saponin present in the phospholipid complex to penetrate into the skin [65].

Researchers reported in there review that the topical delivery of plant derived products can be effectively done in cosmetic preparation by phospholipids complexation [66, 67].

Scientist formulated Oxymatrine-phospholipid complex (OMT-PLC) to improve the lipid solubility and effectiveness of OMT. The purpose of their study was to explore the utility of the combination of a micro emulsion and an OMT-PLC as topical delivery vehicle for enhancing the absorption and efficacy of OMT. The solubility of OMT-PLC was determined and phase diagram of micro emulsion were constructed. They developed various physicochemical properties and In Vitro and In Vitro permeability through skin. They concluded that the combination of a micro emulsion and phospholipids complex represents an effective vehicle for topical delivery of OMT [68].

Researchers reported that incorporation of high amount of curcumin in topical

formulation cannot provide a better bioavailability. They prepared complex of curcumin with phosphatidylcholine and characterized them on the basis of TLC, DSC, Melting point and FT-IR. They compared the activity of vesicular systems like liposome, niosome, phytovesicle. In result they got that the phytovesicles are having excellent antioxidant and antiaging properties than the other vesicular systems, which may be due to the amphiphilic nature of the complex, which greatly enhances the water and lipid miscibility of the curcumin [69].

Scientist investigated the relative absorption of a standardized curcuminoids mixture and its corresponding lecithin formulation (Meriva) in a randomized, double blind cross over design human study. They reported the improved absorption and a better plasma curcuminoid profile of the Meriva at a dose significantly lower than unformulated curcuminoid mixture [70].

Scientist has prepared the curcumin-phospholipid complex in a molar ratio of (1:2) of curcumin and phospholipids. They confirmed the formation of complex by FT-IR Spectroscopy and DSC analysis. They compared the skin permeation of curcumin with the complexed curcumin and found that the complexed curcumin showed 60% greater permeation of curcumin through rat skin. They reported that the phospholipids complex has more transdermal penetration than pure curcumin [71].

Scientist studied that Gallic acid and its derivatives are a group of naturally occurring polyphenols antioxidants which have recently been shown to have potential health effects but when administered orally it shows poor absorption because of less lipophilicity. To overcome this limitation, they developed Gallic acid- phospholipids complex in different ratio to improve the lipophilic properties of Gallic acid. The physicochemical properties of the complex were analyzed by ultraviolet-visible spectrometry (UV), infrared spectrometry (IR) and differential scanning calorimetric (DSC), solubility, dissolution, etc. the result showed that Gallic and phospholipids in Gallic-phospholipids complex were joined by non-covalent bond and did not form a new compound and observed that complex was an effective scavenger of DPPH radicals and showed the strong antioxidant activity [72].

Performe Preclinical studies of a novel polyherbal phyto-complex hair growth promoting cream which was incorporated with the aq. extracts of Trichosanthes cucumerica (T.curcumerica) Linn and Abrusprecatorius (A.precatorius) Linn. In the experimental study, extraction of both plants, chemical testing of both extract, then extract were made into phyto-phosphatidylcholine complex, finally preparation of formulation and then evaluation of cream containing polyherbal phyto-complex. Preclinical studies showed that formulated 2% polyherbalphytocomplex hair growth promoting cream was an effective hair growth promoter as the results were analogous to that of minoxidil 2%.it was observed that percentage of hair follicles in the anagen phase increased considerably which predicts that the formulation can be used in alopecia [73].

CONCLUSION

The area of novel drug delivery system is an extensive researches that targeting for plant actives and extracts, so the research in this area is still at the exploratory stage. Regarding from the usefulness of plant products, especially that containing flavonoids and other poly phenolic compounds. The phyto-phospholipid complexation technique has offered a great opportunity and hope in improving the in vivo bioavailability of herbal drugs which in spite of positive in vitro results have failed to deliver a similar response in vivo. The polyphenolic constituents of plants like flavones and others have immense therapeutic potential but because of their inability to cross lipoidal barrier their utilization in treatment of severe illnesses like cancer, hepatic diseases and rheumatic conditions has remained an unresolved apprehension for quite a substantial period of time. Phytosomes are novel formulations which offer improved bioavailability of hydrophilic flavonoids and other similar compounds through the skin or gastrointestinal tract. The formulation methodology for phytosome is simple that can be easily upgraded to a commercial scale. The

characterization methodologies and analytical techniques are well established for this type of novel formulation. Many patents and marketed formulations are already approved for innovative formulations, processes and applications of phytosomes. As far as the potential of phytosome technology is concerned, it has a great future for use in formulation technology and applications of hydrophilic plant compounds.

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