CHAPTER I

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1. Atopic dermatitis (AD)

AD is a critical inflammatory skin disease (chronic) with symptoms like rough skin, itching, swelling, debris, and scarring. Furthermore, physical symptoms such as behavioural problems, mood disorders, enhanced risk of depression, sleep disorder, finances, and also, in addition to physical symptoms are also found in AD disease. This disease can cause erythematous plaques, recurring rashes, small blisters, and persistent scratching that may leak extracellular fluid. AD frequently increases in developing countries and can occur at any age, generally in children and adults. The reasons for AD disease are complex and multi-factorial (Lin *et al*., 2021; Torres *et al*., 2019), but some reasons, such as environmental, pharmacological, immunological, psychological, skin dysfunction, and genetic factors, play a fundamental role in pathogenic mechanisms (Yun *et al*., 2014).

A subset of AD patients has null gene mutations in filaggrins, resulting from an autosomal dominant inheritance. Even in people lacking germline filaggrin mutations, barrier breakdown can happen due to exogenous insults, potentially due to direct skin barrier insults or epigenetic changes. These elements include stress, pollution, pruritogens, scents, and weather. Damaged keratinocytes emit thymic stromal lymphopoietin and other cytokines from the compromised skin barrier, which cause skin inflammation and in gene-environment interactions in AD disease (Cork *et al*., 2006; Kantor & Silverberg, 2017). Transcutaneous allergen penetration is more likely due to compromised barrier function, and antigen sensitization and presentation could also happen (Owen *et al*., 2018).

The disease AD is determined by type 2 immunity (T2) and characterized by the CD4+ T helper (Th2) lymphocyte cells, which induce and produce IL-5, IL-13 & IL-4 (type-2 cytokines). These cytokines can stimulate the produce antibodies and innate immune cells that transform into B-cells, and producing IgE. This illness is frequent, recurrent, and extremely itchy. It can also cause skin injuries to become inflamed. The primary goals of treating AD involve reducing inflammation, controlling itching, and resetting the skin barrier through pharmacological and non-pharmacological methods based on emollients and moisturizers that improve the skin's moisture and barrier function (Napolitano *et al.*, 2023; Ramos Campos *et al.*, 2020).

The primary epidermal cells, known as keratinocytes, are thought to be essential in AD. In response to different stimuli, the keratinocyte releases chemokines and inflammatory cytokines. These mediators facilitate the entry of inflammatory cells into the skin's inflammatory regions. According to recent research, inflammatory cytokines can play an important role in the AD disease. Creating substances that can stop inflammatory cytokines from acting is of tremendous interest (S.-J. Kim, 2017; Vestergaard *et al.*, 2000).

Patients with this illness show abnormalities in the generation of lipids in the epidermis are critical for developing and maintaining dermal alterations. In addition, AD patients frequently get viral and bacterial skin infections (El-Salamouni *et al.*, 2020). Recent data indicates a progressive increase, and which can be attributed to environmental changes by the world's the disease AD can rapidly development. In the context of India, the increasing trend is likewise fundamental. Research on the epidemiology, etiopathogenesis, and management of AD worldwide has recently increased, although the literature on these topics in India is not as strong. In India, population-based studies on the epidemiology of AD are uncommon, and most of the

epidemiological data currently accessible comes from hospital studies. Over the past forty years, an increasing trend in AD has also been noted in India (Kanwar & De, 2011).

This chronic disease has a high global burden on healthcare costs and morbidity. Although many areas of uncertainty persist (panel 2), clinical medicine, epidemiology, molecular biology, and discoveries from genetics have spurred new disease concepts. In most patients' cases, AD founds a lifetime with activity manifestations and clinical variables. The pathogenic fundamental mechanisms are type-2-dominated cutaneous inflammation and dysfunctional epidermal barrier, which small-molecule therapies and innovative biological therapies can target. There is an increasing appreciation of that therapy. The multidimensional assessment is essential to identify the appropriate requirements of the patients. There is a genuine expectation that therapies targeted specifically and novel prevention strategies against AD disease developments will decrease morbidity and the global burden of healthcare costs (Thomsen, 2014; Torres *et al*., 2019).

2. Medicinal plant

The practice of alternative and complementary medicines in the healthcare sector has expanded due to community patient empowerment. Since the health care experts were practitioners who worked mainly with Phyto remedies, historically, allopathic drugs were considered the alternate type of treatment. It was only within the last century that allopathic regimens replaced alternative herbal medicine. Only ten to thirty percent of health care parts are currently provided by allopathic practitioners, and the remaining seventy to ninety percent of services are still offered by complementary and alternative medicine practitioners, according to the WHO (World Health Organization). The

alternative modalities of care provided within a standardized healthcare system are based on standard or believed practice (Hussain *et al*., 2017; Templeman & Robinson, 2011).

2.1. *Mesua ferrea* Linn.

Mesua ferrea Linn. is a member of the Calophyllaceae family and is found in tropical regions such as China, India, Burma, Thailand, and New Guinea. It grows in the Eastern Himalayan Mountains, East Bengal, Assam, Burma, and the Andaman Islands; it also grows in the North and South Konkan evergreen rain forests; and it grows in the Western Ghats woods, which stretch from South Canara to Travancore (K. Chahar, 2013; Kritikar KR, Basu BD., 1981).

This medicinal tree, which may reach 18 to 30 meters, is medium to large. This tree's bark is reddish-brown, thin flakes peeled off, and durable wood. Simple, sharp, lanceolate, and leathery leaves with a waxy bloom underneath exist. Mature leaves are red, grouped in opposition in colour, measure seven and thirteen centimetres in length and two to four centimetres in width and have a diameter of 7.5 cm and many golden-coloured filaments that are shorter than the petals' length, and the white blossoms have a fragrant trace. The style, which can be axillary borne unaccompanied or in pairs, is double as long as stamens. The fruits are ovoid and also have a conical point, with a woody husk that contains one to four seeds (Dassanayake MD., 1980; *India Biodiversity Portal Mesua Ferrea L.*, 2023). The image of *Mesua ferrea* Linn tree, flowering buds, and leaves of are shown in (**Figure 1)**.

Figure 1. In right image of *Mesua ferrea* Linn. tree and in left flowering buds with leaves

This medicinal plant is traditionally used in dyspepsia, dyspepsia, haemorrhoids, fever, neurasthenia, liver problems, depression, diarrhoea, and renal diseases. According to phytochemical analysis, this medicinal plant contains xanthones, coumarins, flavones, flavanone glycosides, derivatives of cyclohexanedione, and essential oil. The methanolic extract of the *Mesua ferrea* Linn. flowering buds yielded recognized 4-alkyl-5,7-dihydroxycoumarins, and 4-alkyl-5,7-dihydroxycoumarins, essential oils, mesuaferol G–K, coumarins, flavones, flavanone glycosides, and derivatives of cyclohexanedione (S. Wang *et al.*, 2019). In addition to its many other uses, the plant has anti-inflammatory properties, antibacterial, analgesic, anticancer, and antioxidant. It is a component of several Unani and Ayurveda remedies. The phytochemical screening verifies that the primary components of the plant include phenyl coumarins, xanthones, triterpenoids, lipids, and flavonoids (K. Chahar, 2013).

Rasayana is founded on similar ideas, and immune response modification to treat illnesses has long been of interest. Included in the literature on traditional Indian medicine, the class Rasayana comprises various plants believed to improve physical and mental well-being, strengthen the body's defines mechanisms, and lengthen life. The immunomodulatory properties which can strongly stimulate oxidative processes, immune cells are particularly vulnerable to oxidative damage. The immune system's cellular and humoral components are especially vulnerable to raised oxygen reactive species levels, and which can affect the initial immunosenescence. Combating this oxidative stress and strengthening the body's defences against infection is crucial. The immunomodulators resulting from the natural sources have been exhibited the antioxidant activity and immunomodulatory function (M. K. Chahar *et al.*, 2012; Ramnath & Rekha, 2009).

Ayurvedic medicine utilizes the flower buds of this plant to treat fever, blood-related issues, scabies, sweating, bad breath, skin eruption, itching, heart difficulties, vomiting, sore throat, cough, thirst, small tumours, headache, and dysentery (Pinkesh *et al., 2012;* Roshy *et al.*, 2010). This medicinal plant is recognized for its antibacterial, anti-inflammatory, anticancer, antioxidant, and analgesic properties. It is a component of several Unani and Ayurveda remedies. The phytochemical screening verifies that the primary components of the plant include phenyl coumarins, xanthones, triterpenoids, lipids, and flavonoids (K. Chahar, 2013).

The wood (deep dark red) is strong, dense, and sturdy, appropriate for heavy construction, show posts, including tool handles, railway sleepers, parquet flooring, and heavy-duty furniture. *Mesua ferrea* is a significant forest tree used to produce lumber. The tree is also frequently implanted in avenues, hedgerows, and landscapes. Because of its potent scent, incense sticks manufactured from this plant's blooms are well-liked worldwide. Bridal beds are furnished with pillows and cushions with fragrant stamens. In

addition to its therapeutic applications, it finds commercial use in the polymer sector, painting, firewood production, nanoparticle synthesis, and fuel substitution. Thus, more research may be done to demonstrate the potential of this plant and its separated components (K. Chahar, 2013).

3. Novel herbal drug delivery systems

The medicinal qualities of plants have long been acknowledged, and herbal traditional medicines have been used throughout the world to cure an extensive variety of local and regional illnesses. At least eighty percent of individuals trust on conventional herbal treatments for their medical needs. Many people depend on traditional healers, therapies, and herbal medications, and traditional herbal medicines raise standards for their historical and cultural values (Li & Weng., 2017; *World Health Organization.*, 2023). Recent research study shows that many pharmacological therapies for diseases, symptoms and indicators while ignoring its underlying causes. In the meantime, more prosperous and better disease outcomes are shown using natural herbal medication (Nagalingam, 2017).

The majority of conventional herbal medicine delivery methods fall short of what makes for an optimal drug delivery system. Certain limitations are seen when using traditional herbal treatment, such as **(a).** The short half-lives of most herbal medications, which increase the likelihood of skipping a dose; (**b).** The liver may process several components before entering the systemic circulation, at which point they cannot have any therapeutic impact; (**c).** The acidic pH of the stomach increases the likelihood that the herbal ingredients will be destroyed; **(d).** Polyphenols have low absorption and bioavailability while having good water solubility; (**e).** The inability of traditional herbal

delivery systems to overcome the poor lipid solubility, instability, and inappropriate molecular size of herbal pharmaceuticals has controlled the progress of novel herbal drug delivery systems with essential alterations of herbal drugs. The important sources of chemical sustenance and medicinal qualities, plants are used to build traditional drug delivery systems by the modern scientific methods, which reduces the need for human subject error (Li & Weng, 2017; Nagalingam, 2017; Raeiszadeh *et al.*, 2018).

The limits of conventional plant drug delivery systems have recently been overcome by researchers using a modern strategy herbal formulation of NDDS (Novel drug delivery systems). These advanced herbal drug formulations that target specific body sites, release their ingredients continuously over extended periods with minimal dosages, and enhance the effects of herbal medications are characteristics of these formulations. NDDS offers many advantages, including increased stability, solubility bioavailability, and pharmacological activity, protection against toxicity, protection against physical and chemical degradation, and sustained and controlled delivery of herbal formulations. A variety of novel formulations, such as ethosomes, liposomes, nano-emulsion, transferases, transdermal, solid lipid nanoparticles, and mucoadhesive drug delivery systems. Several novel formulations are investigated, some in the research laboratory (Ajazuddin & Saraf, 2010; N. Jain *et al.*, 2010).

4. Phytosome

The conventional method of delivering herbal medications has significant drawbacks. Phytomedicine is responsible for insufficient absorption and bioavailability due to its higher likelihood of disintegration in the acidic pH of the gastrointestinal tract, first-pass metabolism that necessitates frequent dosing, instability, improper molecular

size, and low lipid solubility. The development of innovative herbal drug delivery systems, such as NDDS for phytopharmaceuticals, has garnered increasing attention in research in recent years. The best new carriers would target the intended site of action and administer the medication with controlled release throughout the treatment. Certain intrinsic benefits of NDDS over traditional formulations include increased potency and safety, improved stability, solubility, bioavailability, and less physicochemical degradation (Ajazuddin & Saraf, 2010; Nagalingam, 2017).

The patented phytosome technology was initially created by the Italian business (company) Indena and incorporates Phospholipid and water-soluble phytoconstituents. Phytolipid delivery systems, another name for phytosome technology, are primarily used to improve the oral absorption of hydrophilic phytoconstituents. The Greek words 'phyto' (meaning plant) and 'some' (meaning cell-like) are combined to get the phrase phytosome (Nagalingam, 2017). Chemical bonds form between poorly absorbed compounds, such as flavonoids, and the hydrophilic head of the phospholipid, phosphatidylcholine, in its inner core (Kidd, 2009; Lu *et al*., 2019). Phytosomes structure is shown in (**Figure 2)**.

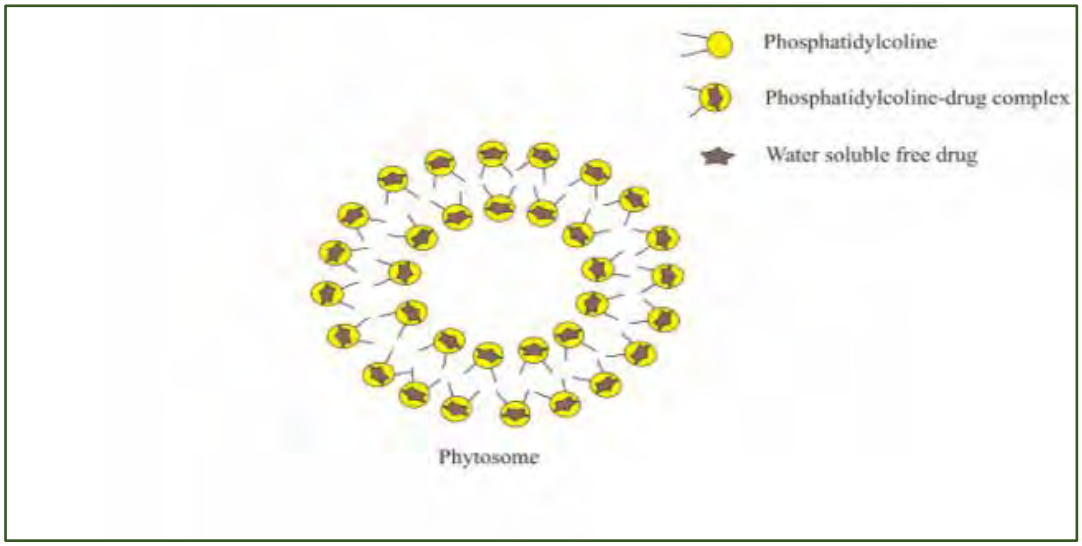

Figure 2. Structure of molecular organization of the phytosomes complex

The phospholipids molecules and phytoconstituents create chemical bonds, and phytosomes generate a superior stability profile. A lower dose is needed because the primary ingredients have the highest absorption rate. Creating photo-phospholipid complexes significantly increases the active components' membrane permeability and oil-water partition coefficients. The absorption of these complexes improves bioavailability (Lu *et al*., 2019; Upase *et al.*, 2019). Due to their high lipid profile and excessive skin penetration, phytosomes are widely employed in cosmetic preparation. Their lipid layer in the phytoconstituent area allows them to permeate through the skin (Karimi *et al*., 2015; Kumar *et al.*, 2017).

4.1 Structure of phospholipids

Phospholipids are simple to find in various foods, including fish, sunflower, rapeseed, soy, and chicken eggs. Sphingomyelins and glycerophospholipids are the two main categories into which phospholipids can be separated. The phosphatidyl-glycerol, phosphatidylcholine, phosphatidylethanolamine, phosphatidylserine, phosphatidic acid, phosphatidylinositol, and phosphatidylserine are further subdivided under the category of glycerophospholipids. Phosphatidylethanolamine, phosphatidylserine, and phosphatidylcholine are the three main phospholipids that are supportive to the phytosomes formation (Ghanbarzadeh *et al.*, 2016). Phospholipid's structures are shown in (**Figure 3)**.

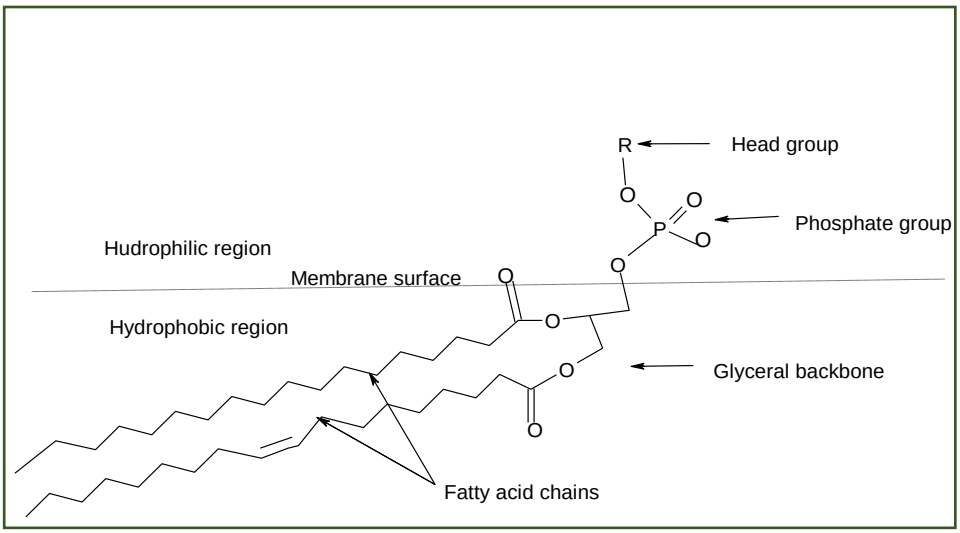

Figure 3. Chemical structure of phosphatidylcholine

A bilayer structure of phospholipids results from the combined action of hydrophilic and hydrophobic interactions. The bilayer structure stabilizes when dipole-dipole contact between the ion pairs at the bilayer's surface. Water-soluble substances can directly bond with phosphatidylcholine or phospholipids, increasing the water-soluble standard extract's bioavailability. The lipid component of lipoprotein phosphatidylcholine is circulating in the bloodstream. Its surface-active characteristic shields the gastrointestinal tract and lungs' epithelial luminal interfaces. The two components of phospholipids—the hydrophilic head, which has polar groups, and the hydrophobic tail, which is made up of a lengthy hydrocarbon chain of fatty acids—play a crucial role in creating phospholipid bilayers (Ebrahimi *et al.*, 2015; Lu *et al.*, 2019; Singer & Nicolson, 1972).

4.2. Benefits of phytosome

The phytosome has several benefits, such as **(a).** The phytosomes preparation is simple; **(b).** In addition to being quickly absorbed by the skin, it increases efficacy; **(c).** It nourishes the skin and readily absorbs into the cell membrane; **(d).** The phytoconstituent and PC (phosphatidylcholine) establish a chemical connection, improving stability; **(e).** Water-soluble phytoconstituents in phytosomes are encased in PC, which stops the pre-systemic metabolism; **(f).** Because of the active ingredient's improved absorption, this compound lowers the dosage requirement; **(g).** By using water-soluble phytoconstituents, it improves oral and topical absorption; **(h).** It has a much higher therapeutic value and exhibits greater bioavailability; and **(i).** Enhanced bioavailability and fewer dosages may increase market demand (Kumar *et al.*, 2017; Upase *et al.*, 2019).

4.3. Properties of phytosome

4.3.1. Physicochemical properties

The aprotic solvent reacts with a stoichiometric quantity of phospholipids and herbal phytoconstituents and finally produces the phytosome complex. The size range of the phospholipids is fifty to hundred micrometres. Hydrogen bonds are which formed with the phosphate group due to substrate interactions with the phytomolecule and phospholipid poler head. When handled with water, the Phyto-phospholipid complex comprises a micellar structure and becomes readily soluble in non-polar solvents. It also exhibits a distinct melting point (Bhagyashree, 2015; Kumar *et al.*, 2017).

4.3.2. Biological properties

Phytosomes are an improved herbal product that is more bioavailable, readily absorbed, and effective than traditional herbal extract administration methods. The pharmacodynamic and pharmacokinetic parameters in human volunteers (healthy) and animals (experimental) have demonstrated the biological properties (Agarwal *et al.*, 2012; Kumar *et al.*, 2017).

4.4. Method of preparation

Typically, the processes of, anti-solvent precipitation, solvent evaporation and lyophilization or freeze-drying are used for the preparation of phytosomes. Generally speaking, preparing the phytosome involves combining Phospholipid from a vegetable or other source in a 1:1 or 1:2 ratio with polyphenolic phytoconstituent. Typically, freeze-drying, solvent evaporation, and anti-solvent precipitation techniques are used to

manufacture phytosomes. One mole of Phospholipid (natural or synthetic), such as phosphatidylcholine or phosphatidylserine, and one mole of a constituent mixed in a solvent like ethyl acetate, methylene chloride, dioxane, etc., are characteristically used in its formulation. More recently, protic solvents like ethanol and tetrahydrofuran have been used. Therefore, the complex can be separated using various techniques such as solvent evaporation as well as spray drying to form a thin film. In this complex Equation, the difference between these moieties is approximately from 0.5 to 0.2 moles other than the most required ratio of 0.1:0.1 (J. Khan *et al.*, 2013; Lu *et al.*, 2019; Tawheed Amin &, Suman Vikas Bhat, 2012).

Singh *et al.* prepared gingerol phytosomes by using anti-solvent methods and varying the molar ratios of gingerol plant extract and soya lecithin. The mixture ratios were recorded by stirring with 30 ml of methanol solvent dissolved in 100 ml beaker. Twenty millilitres of n-hexane (anti-solvent) were added with stirring. Then, the mixture was concentrated, and the precipitate was collected and stored in a desiccator. The formulated phytosome was stored at room temperature in a glass bottle with a golden tint (R. P. Singh *et al.*, 2018).

Freag *et al*. formulated the phytosomes by the lyophilization process. The diosmin was mixed in dimethyl sulphoxide, and the diosmin solution was further added to the Phospholipid (soybean) solution, which was dissolved with solvent (t-butyl alcohol), and stirred continuously for three hours until the lyophilization formed the complex. Diosmin was fully dissolved in dimethyl sulphoxide for lyophilization and after adding the obtained diosmin 2.5 % w/v solution to the Phospholipid (soybean) solution that dissolved in t-butyl alcohol. The mixture was stirred for at least three hours until the complex formation occurred. The complex was separated and frozen after lyophilization. It was then gradually cooled to -40^{0}C before being dried at 25^{0}C. After

being removed from the dryer, this sample was preserved in desiccators (Elnaggar *et al.*, 2013).

Kim *et al.* used solvent evaporation methods for the formulation of chrysin phytosomes with soya phosphatidylcholine. They employed phospholipid and chrysin molar ratios of 1:2 and 1:3, and this combination was dissolved in 12.5 ml of protic solvent tetrahydrofuran and agitated constantly at 40^0C for four hours. The solvent was extracted at the same temperature using a rotary evaporator (S.-M. Kim *et al.*, 2019).

4.5. The factors prompting the phytosome preparation

Several parameters affect the stability and efficacy of phytosomes, including **(a).** Drug release percentage; **(b).** Membrane permeability; **(c).** Drug entrapment efficiency; **(d).** Chemical composition; **(e).** Phytosome size and shape; and **(f).** Component purity (Nagalingam, 2017).

4.6. Evaluation of phytosomes

4.6.1. Percentage of entrapment efficiency

The centrifugation method is used to ascertain the percentage of entrapment efficiency (% EE) of a developed phytosome mixture (Ghanbarzadeh *et al.*, 2016; Lu *et al.*, 2019). For determine the drug entrapment percentage, apply the initial amount of sample minus final amount of sample divided by initial amount of sample multiply by hundred.

4.6.2. Transition temperature

The DSC (Differential scanning calorimetry) has been estimated the transition temperature for the vesicular lipid systems such as phytosomes. The complex's produced picks can be examined and contrasted with the Phospholipid and active components using the DSC (Kumar *et al.*, 2017).

4.6.3. Vesicle size and zeta potential

The PCS (Photon correlation spectroscopy) and DLS (Dynamic light scattering) are used to measure the particle size distribution, zeta potential, and particle size of the phytosome. The physical stability of the phytosome depends on the effects of Brownian motion are crucial characteristics for the evaluation of the phytosome. The surface morphology and entrapment mechanism of phytosomes are determined by AFM (Atomic force microscopy), TEM (Transmission electron microscopy), and SEM (Scanning electron microscopy) (Babazadeh *et al.*, 2018; Ghanbarzadeh *et al.*, 2016).

4.6.4. Spectroscopic evaluation

The production of phytosomes, the complex analysis between the Phospholipid and the phytoconstituent moiety, and the identification of the interactions between the involved components are all studied using spectroscopic evaluations. Using 1H NMR techniques, phytoconstituents and phospholipid complex were estimates and the results indicate a widened signal originating from the proton of the constituent phytoconstituents. The C-13 NMR signal shift has signified the complex formulation's chemical interactions between PC and phytoconstituents. The spectroscopy method is used to identify the formulation complex structure, phospholipids, or phytoconstituents

(active ingredient), and formulated phytosome interaction (Bhagyashree, 2015; Gabetta *et al.*, 1989; Kumar *et al.*, 2017). By comparing the unique spectra of the constituent materials and the prepared phytosome complexes, the FTIR investigations can validate the complex formation (Ghanbarzadeh *et al.*, 2016).

4.6.5. Chromatographic evaluations

The chromatographic techniques are used to determine the phytosome's retention duration, characterize phytosomes, phytoconstituents, and phospholipid retention factor, as well as separate complexes by the HPLC (High-performance chromatography) and TLC (Thin-layer chromatography) (Babazadeh *et al.*, 2018).

5. Topical application

The body's most significant, most intricate, multipurpose, and cellular organ that contributes to its construction is the skin. The skin, an exterior covering of the body, can regulate body temperature, prevent bodily fluid loss, and support or shield the individual from external environmental influences. The three primary layers are the epidermis, dermis, hypodermis, or subcutaneous tissue. Numerous specialized cells and structures are found in the skin. The epidermis, dermis, and hypodermis are its three primary layers. Every layer contributes differently to the skin's overall functionality. The thickness of the epidermis, the skin's outermost layer, varies depending on the body part. The subcutaneous connective tissue, or hypodermis, is connected to the dermis. Larger blood arteries and nerves are housed in the subcutaneous tissue, which is connective to the tissue layer and fat. This layer is crucial for controlling the body's and the skin's internal temperature. This layer is different in size in other body parts and different people. The

primary skin appendages are hair follicles, sweat glands, and sebaceous glands (Elias, 1991; Proksch *et al.*, 2008; Reuter *et al.*, 2010). The human skin structure is shown in (**Figure 4)**. An estimated 34 % of all occupational diseases are related to skin conditions. The skin serves various functions, including protection, thermoregulation, percutaneous absorption, and secretory and sensory functions.

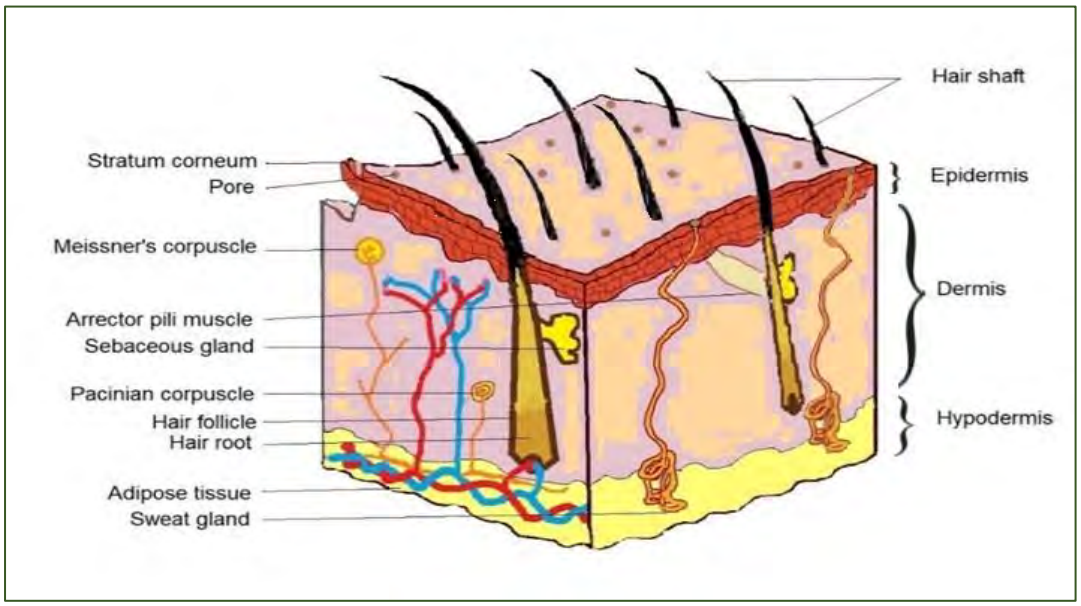

Figure 4. Structure of human skin

The skin provides the first defence against broad injury by microbial and chemical agents. Infectious diseases, mucosal infections, and skin infections, are common among most tribal inhabitants due to potable water, an absence of sanitation, and awareness of hygienic food habits. The main interface to the body and the external environment and the skin, interacts with the environment and is crucial in shielding the body from

infections and excessive water loss. Other roles include insulation, controlling body temperature, feeling, storing and synthesizing vitamin D through UV light, protecting vitamin B folates, absorbing oxygen and medications, and being water resistant. Scar tissue is the body's way of mending severely injured skin. This needs to be depigmented and discoloured more frequently (Grice *et al.*, 2009; Madison, 2003).

Medications and cosmetics are used to protect the skin from external and internal hazardous substances while maintaining the skin's natural structure, integrity, beauty, and attractiveness. The topical method is superior to conventional routes, such as preventing hepatic first-pass effects, constantly delivering medications and phytoconstituents, enhancing patient compliance, and minimizing side effects. The primary skin barrier, the stratum corneum, keeps the skin from drying out and limits the absorption of certain medications and cosmetics. Although a variety of synthetic pharmaceuticals are used as photo-protective agents, their application is restricted due to their possible toxicity to people and their capacity to obstruct specific routes of carcinogenesis (Sahu *et al.*, 2013; Tabassum & Hamdani, 2014). Several fundamental factors have been found for skin diseases which is related to the skin infections. The harmful result of skin disturbance is the entry of infectious microorganisms, and several microorganisms thrive on the skin, making it a sanctuary for them and a potential source of skin illnesses (Guirat *et al.*, 2018).

5.1. Nanocarriers skin penetration

Because of the protective layers of the skin, traditional formulations exhibit inadequate penetration of the active components. The more recent formulations of nanocarriers have an exceptional method for enhancement of drug delivery and penetration into the skin's deeper layers. The drug delivery through nanocarriers are

shows better skin penetration and be beneficial for treating skin conditions. Additionally, longer-lasting drug release, improved drug stability, and fewer side effects are all possible with nanosized pharmaceuticals (Kahraman *et al.*, 2017).

Many factors, including skin age, skin site, various species, application area, skin state, degree of hydration, normal and abraded state of the skin, contact time, and skin temperature, can affect the penetration of the skin (Alexander *et al.*, 2012). Passive diffusion allows the applied nanocarriers or active compounds to penetrate the skin along three different pathways like shunt (transappendageal), paracellular (intercellular), and intracellular (transcellular). The three penetration pathways are shown in (**Figure 5).** The drugs are penetration through a high-concentration region to a lower-concentration region, performing a passive kinetic process also called diffusion. Fick's first law can be used to express this passive diffusion (Kováčik *et al.*, 2020; Lane, 2013; Ramos Campos *et al.*, 2020).

The primary drug molecule penetration route of active components through the stratum corneum of intercellular lipid bilayers is intercellular pathway. The majority of small-molecular medications nevertheless have a route length significantly longer than the thickness of the stratum corneum. Another potential route for penetration through the stratum corneum is the transcellular pathway. This channel facilitates the direct transfer of active chemicals to the deeper layers of skin by passing *via* keratinocytes and intercellular lipid bilayers. The majority of molecules are not able to use this channel to penetrate through the stratum corneum. Lipophilic log (P) $^{(1\text{-}3)}$ and small molecular weights (<500 Da) are known to permeate through this channel (Kahraman *et al.*, 2017; Kováčik *et al.*, 2020).

Drug administration *via* the appendageal route is crucial for the penetration of slowly diffusing molecules like nanoparticles and high molecular weight compounds. The

appendageal pathway may help drugs diffuse quickly into the early stages of diffusion. More than twenty distinct cell populations comprise the hair follicle's intricate, potentially three-dimensional structure, which is responsible for a range of metabolic, immunological, and biochemical processes. Various factors, such as morphology, follicular density, and regional variations in hair cycle activity influence as well as control the penetration of follicles (Knorr *et al.*, 2009; Lane, 2013). When an exogenous or endogenous trigger signal is released, the nanocarrier medicines are transported through hair follicles and exhibit deeper penetration. Current carrier technologies are appropriate for unique physicochemical properties such as pH sensitivity, stability in the skin environment, and temperature-sensitive enzyme-mediated cleavage site deprivation (Patzelt & Lademann, 2020; Vogt *et al.*, 2016).

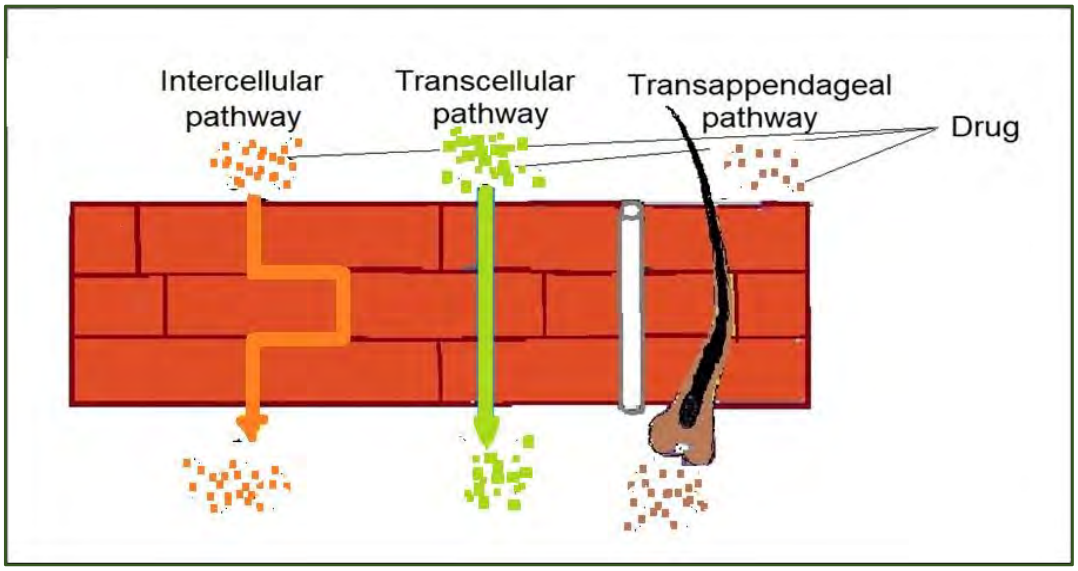

Figure 5. Drug penetration pathway through skin

The shape of the particle may play a significant role in skin penetration. The investigations using mouse and human models have been conducted infrequently. Compared to spheres, the study on tiny gold nanorods demonstrates increased epidermal penetration. According to some researchers, when topically applied to the hairless mice's skin, tiny silver nanorods exhibited the highest blood concentration compared to triangle and shaped particles (Vogt *et al.*, 2016).

5.2. Involvement of nanocarriers for skin penetration

With the encapsulation of active pharmaceutical substances to form specific characteristics and permeate through hair follicles related to the skin's lipids to transport and generate prolonged release, the nanocarriers may help with medication delivery. The nanocarriers' enhanced surface area and volume ratio effects increased the penetration to each pathway. Drug nanocarriers have been used for the treatment of dermatological disorders, and skin penetration of active components. Along with lowering adverse effects and increasing chemical stability, these have also allowed drugs to be released continuously for more extended periods (Kahraman *et al.*, 2017; Makhmalzade & Chavoshy, 2018).

When using nanocarriers directly to deliver drugs *via* the skin with different nanocarrier formulations, such as liposomes, ethosomes, transfersomes, invasomes, and transethosomes, might improve penetration capability. Lipid and polymer-based nanoparticles, dendrimers, micelles, various nanoparticles, and microemulsions have been extensively active in skin delivery to enhance drug penetration.

Peptides containing three to one hundred cyclic amino acids bind to skin proteins, most likely keratin, to improve skin penetration. Lipid-based nanocarriers

(nanostructured and solid lipid nanoparticles) allow the skin to penetrate more easily because they form a thin layer on the skin's surface, hydrate the skin, and lower trans-epidermal water (Dragicevic & Maibach, 2018; Koushki *et al.*, 2020; Patel & Prabhu, 2020).

Skin penetration augmentation procedures can make use of chemical techniques, and physical including sonophoresis, electroporation, high-voltage, laser-light pulse sources, iontophoresis, and cyclodextrins, fatty acids, lectin, glycols, urea, terpenes, and surfactants. Recently, the use of release systems based on nanocarriers for cosmetic and topical medicinal has been evidence of possible techniques across the skin (Medeiros-Neves *et al.*, 2019).