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Transfersomes: New Dominants for Transdermal Drug Delivery

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ABSTRACT

With oral and parenteral drug delivery systems, poor patient compliance is a frequent problem in daily clinical practice. So, the transdermal route of drug delivery has gained great interest of pharmaceutical research. But the big hurdle in transdermal delivery of drug is the skin, the stratum corneum, & the outermost envelop of the skin. Recently, various strategies have been used to augment the transdermal delivery of bioactive. Mainly, they include iontophoresis, electrophoresis, sonophoresis, chemical permeation enhancers, micro needles, and vesicular system (liposomes, niosomes, elastic liposomes such as ethosomes and transfersomes). Transfersomes possess an infrastructure consisting of hydrophobic and hydrophilic moieties together and as a result can accommodate drug molecules with wide range of solubility. The high and self-optimizing deformability of typical composite transfersomes membrane, which are adaptable to ambient tress allow the ultra deformable transfersomes to change its membrane composition locally and reversibly, when it is pressed against or attracted into narrow pore. Transfersomes can deform and pass through narrow constriction (from 5 to 10 times less than their own diameter) without measurable loss. This high deformability gives better penetration of intact vesicles. They can act as a carrier for low as well as high molecular weight drugs e.g. analgesic, anesthetic, corticosteroids, sex hormone, anticancer, insulin, gap junction protein, and albumin.

Keywords: Transfersomes, Ultra-Deformable vesicles, Transdermal, osmotic gradient

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INTRODUCTION:

The term Transfersome and the underlying concept were introduced in 1991 by Gregor Cevc. In broadest sense, a Transfersome is a highly adaptable and stress-responsive, complex aggregate. Its preferred form is an ultra-deformable vesicle possessing an aqueous core surrounded by the complex lipid bilayer. Interdependency of local composition and shape of the bilayer makes the vesicle both self-regulating and self-optimizing. This enables the Transfersome to cross various transport barriers efficiently, and then act as a Drug carrier for non-invasive targeted drug delivery and sustained release of therapeutic agents.

Transfersome is a term registered as a trademark by the German company IDEA AG, and used by it to refer to its proprietary drug delivery technology. The name means “carrying body”, and is derived from the Latin word 'transferred', meaning ‘to carry across’, and the Greek word ‘soma’, for a ‘body’. A Transfersome carrier is an artificial vesicle designed to be like a cell vesicle or a cell engaged in exocytosis, and thus suitable for controlled and, potentially targeted, drug delivery.

Transfersome is a highly adaptable and stress-responsive, complex aggregate. Its preferred form is an ultra deformable vesicle possessing an aqueous core surrounded by the complex lipid bilayer. Interdependency of local composition and shape of the bilayer makes the vesicle both self-regulating and self-optimizing. This enables the Transfersome to cross various transport barriers efficiently, and then act as a Drug carrier for non-invasive targeted drug delivery and sustained release of therapeutic agents.[1]

Delivery via the transdermal route is an interesting option in this respect because a transdermal route is convenient and safe. This offers several potential advantages over conventional routes[2] like avoidance of first pass metabolism, predictable and extended duration of activity, minimizing undesirable side effects, utility of short half-life drugs, improving physiological and pharmacological response, avoiding the fluctuation in drug levels, inter-and intra-patient variations, and most importantly, it provides patients convenience.

To date many chemical and physical approaches have been applied to increase the efficacy of the material transfer across the intact skin, by use of the penetration enhancers, iontophoresis, sonophoresis and the use of colloidal carriers such as lipid vesicles (liposomes and proliposomes) and nonionic surfactant vesicles (niosomes and proniosomes). Table 1 shows some advantages and disadvantages of different approaches[3] used to increase the material transport through the skin to systemic circulation.

Table 1: Advantages and disadvantages of different vesicular approaches[3]

Methods	Advantages	Disadvantages
Penetration enhancers	Increase penetration through skin and give both local and systemic effect	Skin irritation Immunogenicity, only for low molecular weight drugs
Physical methods e.g. Iontophoresis	Increase penetration of intermediate size charged molecule	Only for charged drugs, Transfer efficiency is low (less than 10%)
Liposomes	Phospholipid vesicle, biocompatible, biodegradable	Less skin penetration, less stable
Proliposome	Phospholipid vesicle, more stable than liposomes	Less penetration, cause aggregation and fusion of vesicles
Niosomes	Non-ionic surfactants vesicles, greater stability,	Less skin penetration easy handling
Proniosomes	Will convert into niosome in situ, stable	But will not reach upto deeper skin layer
Transfersomes and Protransfersomes	More stable, high penetration due to high deformability, biocompatible and biodegradable, suitable for both low and high molecular weight and also for lipophilic as well as hydrophilic drugs and reach up to deeper skin layers.	None, but for some limitations

Vesicular systems show importance because of their ability to give sustained release action of drugs. These systems have several advantages [4-10]:

- They can encapsulate both hydrophilic and lipophilic moieties,
- Prolong half lives of drugs by increasing duration in systemic circulation due to encapsulation,
- Ability to target organs for drug delivery,
- Biodegradability, and
- Lack of toxicity.

Vesicles have a unique structure which is capable of entrapping hydrophilic, lipophilic, amphiphilic and charged hydrophilic drugs. Vesicles are colloidal particles having a water filled core surrounded by a wall of lipids and surfactants (amphiphiles) arranges in bilayer. If the proportion of water is increased, these amphiphiles can form one or more concentric bilayers. Hydrophilic drugs find a place in the internal aqueous environment while amphiphilic, lipophilic drugs get entrapped in the bilayered wall with electrostatic and/or hydrophobic forces. The flexible or deformable vesicles are called elastic vesicles or Transfersomes.

Why only transfersomes for skin?

Transfersomes are advantageous as phospholipids vesicles for transdermal drug delivery. Because of their self-optimized and ultra flexible membrane properties, they are able to deliver

the drug reproducibly either into or through the skin, depending on the choice of administration or application, with high efficiency. The vesicular transfersomes are more elastic than the standard liposomes and thus well suited for the skin penetration. Transfersomes overcome the skin penetration difficulty by squeezing themselves along the intracellular sealing lipid of the stratum corneum. This mechanism can be well understood from the figure 1. These are characteristic with transfersomes, because of the high vesicle deformability, which permits the entry due to the mechanical stress of surrounding, in a self-adapting manner.

Flexibility of transfersomes membrane is governed by mixing suitable surface-active components in the proper ratios with phospholipids [11]. The resulting flexibility of transfersome membrane minimizes the risk of complete vesicle rupture in the skin and allows transfersomes to follow the natural water gradient across the epidermis, when applied under non-occlusive condition. Transfersomes can penetrate the intact stratum corneum spontaneously along two routes in the intracellular lipid that differ in their bilayers properties[12]. The figure 2 shows possible micro routes for drug penetration across human skin intracellular and transcellular[6].

Bangham discovered liposomes in 1963 and since then vesicular systems have attracted increasing attention[13]. But recently it has become evident that classic liposomes are of minor values in terms of penetration. Confocal microscopic studies have shown that intact liposomes are not able to penetrate into granular layer of epidermis but, they rather remain on the upper layer of stratum corneum. The modification of the vesicular compositions or surface properties can adjust the drug release rate and the deposition to the target site [14].

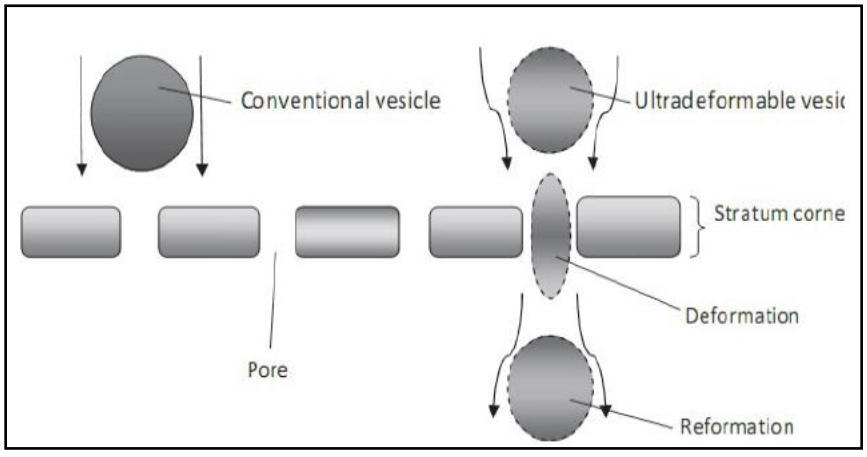

Figure 1: Deformability of transfersomes in to skin pores.[15]

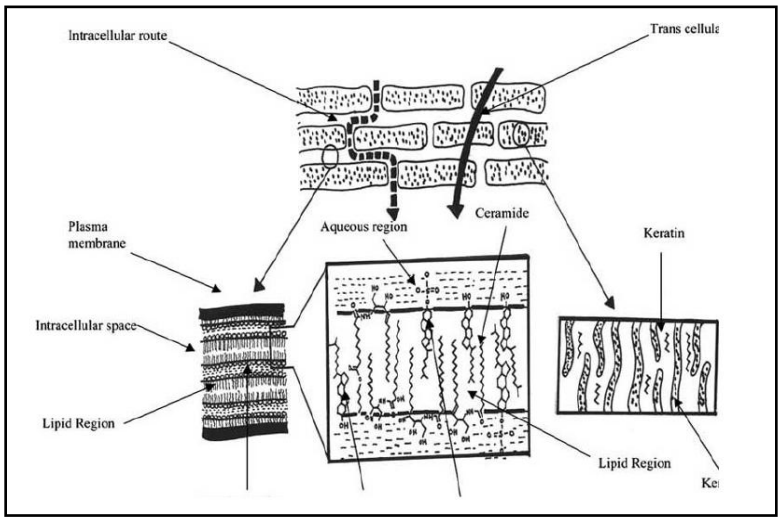

Figure 2: Micro routes for drug penetration across human skin

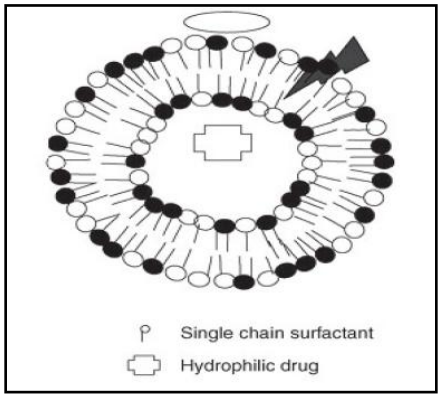

Figure 3: Deformable transfersomes vesicle

Novel characteristics of transfersomes

- Transfersomes possess an infrastructure consisting of hydrophobic and hydrophilic moieties together and as a result can accommodate drug molecules with wide range of solubility as shown in fig 3.
- Transfersomes can deform and pass through narrow constriction (from 5 to 10 times less than their own diameter) without measurable loss. This high deformability gives better penetration of intact vesicles.
- They can act as a carrier for low as well as high molecular weight drugs e.g. analgesic, anesthetic, corticosteroids, sex hormone, anticancer, insulin, gap junction protein, and albumin. They are biocompatible and biodegradable as they are made from natural phospholipids similar to liposomes.
- They have high entrapment efficiency, in case of lipophilic drug near to 90%.
- They protect the encapsulated drug from metabolic degradation.

- They act as depot, releasing their contents slowly and gradually.
- They can be used for both systemic as well as topical delivery of drug.
- Easy to scale up, as procedure is simple, do not involve lengthy procedure and unnecessary use or pharmaceutically unacceptable additives [16].

Limitations of transfersomes [16]

- They are chemically unstable due to their predisposition to oxidative degradation.
- Purity of natural phospholipids is difficult to achieve so, world is against adoption of transfersomes as drug delivery vehicles.
- These formulations are expensive.

Composition of transfersomes [17]

The transfersome is composed of two main aggregates namely,

- First one Amphipathic (such as phosphatidylcholine) ingredient, which in aqueous solvents self-assembles into lipid bilayer that closes into a simple lipid vesicle.
- Second one bilayer softening component (such as a biocompatible surfactant or an amphiphile drug) so, lipid bilayer flexibility and permeability are greatly increased.

The resulting, flexibility and permeability optimized, Transfersome vesicle can therefore adapt its shape to ambient easily and rapidly, by adjusting local concentration of each bilayer component to the local stress experienced by the bilayer as shown in fig 2. Therefore, the Transfersome thus differs from such more conventional vesicle primarily by its "softer", more deformable, and better adjustable artificial membrane.

Mechanism of action

Briefly, the mechanism for penetration is the generation of “osmotic gradient” due to evaporation of water while applying the lipid suspension (Transfersomes) on the skin surface. Transfersomes have strong bilayer deformability and therefore they have increased affinity to bind and retain water. An ultra deformable and highly hydrophilic vesicle always seeks to avoid dehydration; this may involve a transport process related to but not identical with forward osmosis. When they are applied on an open biological surface, such as non-occluded skin, tends to penetrate its barrier and migrate into the water-rich deeper strata to secure its adequate hydration. Natural trans-epidermal water activity gradient enables them to deliver actives to the deeper epidermal layers through dehydration of the lipid vesicles within the stratum corneum. Therefore, transfersome uptake is driven by the hydration gradient that exists across the epidermis, stratum corneum, and ambient atmosphere[18, 19]. Barrier penetration involves reversible bilayer

deformation, but must not compromise unacceptably either the vesicle integrity or the barrier properties for the underlying hydration affinity and gradient to remain in place.

Propensity of penetration

Since transfersomes is too large to diffuse through the skin, the Transfersome needs to find and enforce its own route through the organ. The magnitude of the transport driving force, can be calculated by: Flow = Area x (Barrier) Permeability x (Trans-barrier) force. Therefore, the chemically driven lipid flow across the skin always decreases dramatically when lipid solution is replaced by the some amount of lipids in a suspension [16].

MATERIALS AND METHOD

Materials which are widely used in the formulation of transferosomes are various phospholipids, Surfactants, alcohol, dye; buffering agent etc different additives used in the formulation of transferosomes are summarized in Table 2.[18, 20-23]

Table 2: Different additives used in formulation of transfersomes

Class	Example	Use
Phospholipids	Soya phosphatidyl choline, egg phosphatidyl choline, dipalmitoyl phosphatidyl choline	Vesicles forming component
Surfactants	Sod.cholate,Sod.deoxycholate,Tween-80,Span-80, Tween 20	Vesicles forming Component
solvents	Ethanol, methanol, isopropyl alcohol, chloroform	As a solvent
Buffering agent	Saline phosphate buffer (pH 6.4), phosphate buffer pH 7.4	As a hydrating medium
Dye	Rhodamine-123 Rhodamine-DHPE Fluorescein-DHPE Nile-red	For CSLM study

Preparation of Transfersomes

A. Thin film hydration technique is employed for the preparation of transfersomes which comprised of three steps:[3 ,17, 25]

1. A thin film is prepared from the mixture of vesicles forming ingredients that is phospholipids and surfactant by dissolving in volatile organic solvent (chloroform-methanol). Organic solvent is then evaporated above the lipid transition temperature (room temp. for pure PC vesicles, or 50^0C for dipalmitoyl phosphatidyl choline) using rotary evaporator. Final traces of solvent were removed under vacuum for overnight.
2. A prepared thin film is hydrated with buffer (pH 6.5) by rotation at 60 rpm for 1 hr at the corresponding temperature. The resulting vesicles were swollen for 2 hr at room temperature.

3. To prepare small vesicles, resulting vesicles were sonicated at room temperature or 50^{0}C for 30 min. using a bath sonicator or probe sonicated at 4^{0}C for 30 min. The sonicated vesicles were homogenized by manual extrusion 10 times through a sandwich of 200 and 100 nm polycarbonate membranes.

B. Modified hand shaking, lipid film hydration technique is also founded for the preparation of transfersomes which comprised following steps[26, 27, 28]

1. Drug, lecithin (PC) and edge activator were dissolved in ethanol: chloroform (1:1) mixture. Organic solvent was removed by evaporation while hand shaking above lipid transition temperature (43°C). A thin lipid film was formed inside the flask wall with rotation. The thin film was kept overnight for complete evaporation of solvent
2. The film was then hydrated with phosphate buffer (pH 7.4) with gentle shaking for 15 minute at corresponding temperature. The transfersome suspension further hydrated up to 1 hour at 2-8^{0}C.

OPTIMIZATION OF FORMULATION CONTAINING TRANSFERSOMES[28-30]

There are various process variables which could affect the preparation and properties of the transfersomes. The preparation procedure was accordingly optimized and validated. The process variables are depending upon the procedure involved for manufacturing of formulation. The preparation of transfersomes involves various process variables such as,

- Lecithin : surfactant ratio
- Effect of various solvents
- Effect of various surfactants
- Hydration medium

Optimization was done by selecting entrapment efficiency of drug. During the preparation of a particular system, the other variables were kept constant.

Characterization of Transfersomes

The characterization of transfersomes is generally similar to liposomes, niosomes and micelles [31, 32]. Following characterization parameters have to be checked for transfersomes.

Entrapment efficiency[25]

The entrapment efficiency is expressed as the percentage entrapment of the drug added. Entrapment efficiency was determined by first separation of the un-entrapped drug by use of mini-column centrifugation method. After centrifugation, the vesicles were disrupted using 0.1% Triton X-100 or 50% n-propanol. The entrapment efficiency is expressed as:

$$\frac{\text{Amount entrapped}}{\text{Total amount added}} \times 100$$

Drug content[29]

The drug content can be determined using one of the instrumental analytical methods such as modified high performance liquid chromatography method (HPLC) method using a UV detector, column oven, auto sample, pump, and computerized analysis program depending upon the analytical method of the pharmacopoeial drug.

Vesicle morphology[3, 25]

Vesicle diameter can be determined using photon correlation spectroscopy or dynamic light scattering (DLS) method. Samples were prepared in distilled water, filtered through a 0.2 mm membrane filter and diluted with filtered saline and then size measurement done by using photon correlation spectroscopy or dynamic light scattering (DLS) measurements. Transfersomes vesicles can be visualized by TEM, phase contrast microscopy, etc. The stability of vesicle can be determined by assessing the size and structure of vesicles over time. Mean size is measured by DLS and structural changes are observed by TEM.

Vesicle size distribution and zeta potential[25]

Vesicle size, size distribution and zeta potential were determined by Dynamic Light Scattering Method (DLS) using a computerized inspection system by Malvern Zetasizer.

No. of vesicles per cubic mm[17]

This is an important parameter for optimizing the composition and other process variables. Non-sonicated transfersome formulations are diluted five times with 0.9% sodium chloride solution. Haemocytometer and optical microscope can then be used for further study. The Transfersomes in 80 small squares are counted and calculated using the following formula:

Total number of Transfersomes per cubic mm = Total number of Transfersomes counted × dilution factor × 4000

Confocal scanning laser microscopy study[12]

Conventional light microscopy and electron microscopy both face problem of fixation, sectioning and staining of the skin samples. Often the structures to be examined are actually incompatible with the corresponding processing techniques; these give rise to misinterpretation, but can be minimized by Confocal Scanning Laser Microscopy (CSLM). In this technique lipophilic fluorescence markers are incorporated into the transfersomes and the light emitted by these markers used for following purpose:

- For investigating the mechanism of penetration of transfersomes across the skin,

- For determining histological organization of the skin (epidermal columns, interdigitation), shapes and architecture of the skin penetration pathways.
- For comparison and differentiation of the mechanism of penetration of transfersomes with liposomes, niosomes and micelles.

Different fluorescence markers used in CSLM study are as –

1. Fluorescein- DHPE (1, 2- dihexadecanoyl- sn- glycero- 3- phosphoethanolamine- N- (5- fluoresdenthiocarbamoyl), triethyl- ammonium salt)
2. Rhodamine- DHPE (1, 2- dihexadecanoyl- sn- glycero- 3ogisogietgabikanube- LissamineTmrhodamine-B- sulfonyl), triethanol- amine salt)
3. NBD- PE (1, 2- dihexadecanoyl- sn-glycero- 3- phosphoethanolamine- N- (7-nitro- Benz- 2- xa- 1,3- diazol- 4- yl) triethanolamine salt)
4. Nile red.

Degree of deformability or permeability measurement[3, 17, 25]

In the case of transfersomes, the permeability study is one of the important and unique parameter for characterization. The deformability study is done against the pure water as standard. Transfersomes preparation is passed through a large number of pores of known size (through a sandwich of different microporous filters, with pore diameter between 50 nm and 400 nm, depending on the starting transfersomes suspension). Particle size and size distributions are noted after each pass by dynamic light scattering (DLS) measurements.

The degree of deformability can be determined using the following formula,

$$D = J * \frac{rv}{rp}$$

Where,

J= the amount of the suspension extruded during 5min;

rv = the size of the vesicle;

rp = pore size of the barrier.

Turbidity measurement[25]

Turbidity of drug in aqueous solution can be measured using nephelometer.

Surface charge and charge density[17]

Surface charge and charge density of Transfersomes can be determined using zetasizer.

Penetration ability[3, 17]

Penetration ability of Transfersomes can be evaluated using fluorescence microscopy.

Occlusion effect[25]

Occlusion of skin is considered to be helpful for permeation of drug in case of traditional topical preparations. But the same proves to be detrimental for elastic vesicles. Hydrotaxis (movement in the direction) of water is the major driving force for permeation of vesicles through the skin, from its relatively dry surface to water rich deeper regions. Occlusion affects hydration forces as it prevents evaporation of water from skin.

Physical stability[29, 30]

The initial percentage of the drug entrapped in the formulation was determined and were stored in sealed glass ampoules. The ampoules were placed at 4 ± 2^0C (refrigeration), 25 ± 2^0C (room temp), and 37 ± 2^0C (body temp) for at least 3 months. Samples from each ampoule were analyzed after 30 days to determine drug leakage. Percent drug lose was calculated by keeping the initial entrapment of drug as 100%.

In-vitro drug release[25]

In vitro drug release study is performed for determining the permeation rate. Time needed to attain steady state permeation and the permeation flux at steady state and the information from in-vitro studies are used to optimize the formulation before more expensive in vivo studies are performed. For determining drug release, transfersomes suspension is incubated at 32^{0}C and samples are taken at different times and the free drug is separated by mini column centrifugation. The amount of drug released is then calculated indirectly from the amount of drug entrapped at zero times as the initial amount (100% entrapped and 0% released).

In-vitro Skin permeation Studies[28]

Modified Franz diffusion cell with a receiver compartment volume of 50ml and effective diffusion area of 2.50cm^2 was used for this study. In vitro drug study was performed by using goat skin in phosphate buffer solution (pH 7.4). Fresh Abdominal skin of goat were collected from slaughterhouse and used in the permeation experiments. Abdominal skin hairs were removed and the skin was hydrated in normal saline solution. The adipose tissue layer of the skin was removed by rubbing with a cotton swab. Skin was kept in isopropyl alcohol solution and stored at 0-4^{0}C[33].

To perform skin permeation study, treated skin was mounted horizontally on the receptor compartment with the stratum corneum side facing upwards towards the donor compartment of Franz diffusion cell. The effective permeation area of donor compartment exposed to receptor compartment was 2.50cm^2 and capacity of receptor compartment was 50ml. The receptor compartment was filled with 50ml of phosphate buffer (pH 7.4) saline maintained at 37± 0.5^{0}C

and stirred by a magnetic bar at 100RPM. Formulation (equivalent to 10mg drug) was placed on the skin and the top of the diffusion cell was covered. At appropriate time intervals 1 ml aliquots of the receptor medium were withdrawn and immediately replaced by an equal volume of fresh phosphate buffers (pH 7.4) to maintain sink conditions. Correction factors for each aliquot were considered in calculation of release profile. The samples were analyzed by any instrumental analytical technique.

Skin deposition studies of optimized formulation[25]

At the end of the permeation experiments (after 24hr), the skin surface was washed five times with ethanol: PBS pH 7.4 (1:1), then with water to remove excess drug from surface. The skin was then cut into small pieces. The tissue was further homogenized with ethanol: buffer solution pH 7.4 (1:1) and left for 6hr at room temperature. After shaking for 5 minutes and centrifuging for 5 minutes at 5000rpm, the drug content was analyzed after appropriate dilutions with Phosphate buffer solution (pH 7.4). The result was compared with the control group using student's t-test.

In Vivo Fate of Transfersomes and Kinetics of Transfersomes Penetration[3, 25]

Once the transfersomes passes the outermost skin layers, they will go into blood circulation via the lymph and distributed throughout the body, if applied under suitable conditions. Transdermally Transfersomes can supply the drug to all such body tissues that are accessible to the subcutaneously injected liposomes. The kinetics of action of an epicutaneously applied agent depends on the velocity of carrier penetration as well as on the speed of drug distribution and the action after this passage. The most important single factors in this process are:

I. Carrier in-flow
II. Carrier accumulation at the targets site
III. Carrier elimination

The onset of penetration-driving force depends on the volume of the suspension medium that must evaporate from the skin surface before the sufficiently strong trans-cutaneous chemical potential or water activity gradient is established. Using less solvent is favorable in this respect. The rate of carrier passage across the skin is chiefly determined by the activation energy for the carrier deformation. If penetration of transfersomes involves the occlusion of an application site or the use of too strongly diluted suspension then it will hamper the penetration process. Carrier elimination from the sub cutis is primarily affected by the lymphatic flow, general anesthesia or any other factor that affects this flow, consequently, is prone to modify the rate of transcutaneous carrier transport.

Further, drug distribution is also sensitive to the number of carrier used, as this may affect the rate of vehicle degradation and / or filtration in the lymph nodes. The lag between the time of application and the time of drug appearance in the body, therefore, is always quite long, complex and strongly sensitive to the type of drug and formulation administration. In the best case, the skin penetration lag amounts to approximately 15 min. if rapidly exchanging agents such as local analgesics are detected right under the skin permeability barrier. Less rapidly exchanging molecules or molecules measured in the blood compartment are typically detected with a lag time between 2 and 6 hr. depending on the details of drug formulation. Molecules that do not diffuse readily from the carriers or agents delivered with the suboptimal carriers normally fall in this category. The kinetics of vesicle penetration into and across the skin can be controlled to a large extent by fixing the physicochemical characteristics of the drug carrier suspension. Kinetics of the transfersomes penetration through the intact skin is best studied in the direct biological assays in which vesicle associated drugs exert their action directly under the skin surface. Local analgesics are useful for this purpose, for determining the kinetics of penetration, various lidocaine loaded vesicles were left to dry out on the intact skin. Corresponding subcutaneous injection is used as control. The animal's sensitivity to pain at the treated site after each application was then measured as a function of time. Dermally applied standard drug carrying liposomes or simple lidocaine solution have never caused any analgesic effect. It was necessary to inject such agent preparations to achieve significant pain suppression. In contrast to this, the lidocaine-loaded transfersomes were analgesic ally active even when applied dermally. Maximum analgesic effect with the latter type of drug application was typically observed 15 minutes after the drug application. A marked analgesic effect was still noticeable after very long time. The precise reach as well as kinetics of transfersomes penetration through the skin are affected by: drug carrier interaction, application condition or form, skin characteristics, applied dose[34].

TRANSFERSOMES VS OTHER CARRIER SYSTEMS

Liposomes Vs Transfersomes

Structurally, Transfersomes are very similar to lipid bilayers vesicle, liposomes. However in functional terms, transfersomes differ vastly from commonly used liposomes in that they are much more flexible and adaptable because of edge activator. The extremely high flexibility of their membrane permits transfersomes to squeeze themselves even through pores much smaller than their own diameter. This is due to high flexibility of the transfersomes membrane and is

achieved by judiciously combining at least two lipophilic/amphiphilic components (phospholipids plus bio surfactant) with sufficiently different packing characteristics into a single bilayer. The high resulting aggregate deformability permits transfersomes to penetrate the skin spontaneously. This tendency is supported by the high transfersomes surface hydrophilicity that enforces the search for surrounding of high water activity.[35]

Mixed micelles Vs Transfersomes

It is almost certain that the high penetration potential of the transfersomes is not primarily a consequence of stratum corneum fluidization by the surfactant because micellar suspension contains much more surfactant than transfersomes (PC/Sodium cholate 65/35 w/w %, respectively). Thus, if the penetration enhancement via the solubilization of the skin lipids was the reason for the superior penetration capability of transfersomes, one would expect an even better penetration performance of the micelles. In contrast to this postulate, the higher surfactant concentration in the mixed micelles does not improve the efficacy of material transport into the skin. On the contrary, mixed micelles stay confined to the topmost part of the stratum corneum even they are applied none occlusively. The reason for this is that mixed micelles are much less sensitive to the trans-epidermal water activity gradient than transfersomes.[35]

Transfersomes differ in at least two basic features from the mixed micelles,

- A transfersomes is normally by one to two orders of magnitude (in size) greater than standard lipid micelles.
- Each vesicular transfersomes contains a water filled core whereas a micelle is just a simple fatty droplet. Transfersomes thus carry water as well as fat-soluble agent in comparison to micelles that can only incorporate lipoidal substances [36, 37].

Penetration ability

To differentiate the penetration ability of all these carrier systems proposed the distribution profiles of fluorescently labeled mixed lipid micelles, liposomes and transfersomes as measured by the Confocal Scanning Laser Microscopy (CSLM) in the intact murine skin. In all these vesicles the highly deformable transfersomes transverse the stratum corneum and enter into the viable epidermis in significant quantity[21]. Chapman & Walsh [22] also showed that the former two types of aggregates are confined to the outer half of the horny layer, where the cellular packing and intercellular seals are already compromised by the desquamation process. Pure lipid vesicles or micelles seem to have access to the low-resistance pathway only and thus very seldom reach the lower stratum cornea or even get into the viable part of the skin in significant quantities.

APPLICATION OF TRANSFERSOMES

Different drugs are successfully loaded on to transfersomes which provides targeted as well as controlled drug delivery to the various body tissues (Table 3).

Table 3: Application of transfersomes

Sr.no.	Name of drug	Inference
1	Curcumin[28]	Better permeation for anti-inflammatory activity
2	Indinavir sulfate[30]	Improved influx for activity against acquired immune deficiency syndrome (AIDS)
3	Ketoprofen[45]	Improved penetration for anti-inflammatory activity
4	Insulin[40]	induce therapeutically significant hypoglycemia with good efficacy and reproducibility
5	Capsaicin[43, 44]	Increase skin penetration
6	Colchicine[46]	Increase skin penetration
7	Vincristine[46]	Increase entrapment efficiency and skin permeation
1.	Interferon-α[41]	Efficient delivery means (because delivery other route is difficult). Controlled release. Overcome stability problem.
2.	Norgesterol[47]	Improved transdermal flux
3.	Tamoxifen[17]	Improved transdermal flux
4.	Methotrexate[38]	Improved transdermal flux
5.	Oestradiol[39]	Improved transdermal flux
6.	Tetracaine,[39] Lignocain[34]	Suitable means for the noninvasive treatment of local pain on direct topical drug application.
7.	Corticosteroids[22]	Improved site specificity and overall drug safety.
8.	Hydrocortisone[22]	Biologically active at dose several times lower than currently used formulation.
9.	Triamcinolone acetonide[22]	Used for both local and systemic delivery.
10.	Human serum albumin[17]	Antibody titer is similar or even slightly higher than subcutaneous injection.
11.	Stavudine[29]	Improved the in vitro skin delivery of Stavudine for antiretroviral activity
12.	Tetanus toxoid[48]	For transdermal immunization

Delivery of proteins and peptides:

Transfersomes have been widely used as a carrier for the transport of proteins and peptides. Proteins and peptide are large biogenic molecules which are very difficult to transport into the body, when given orally they are completely degraded in the GI tract. These are the reasons why these peptides and proteins still have to be introduced into the body through injections. Various approaches have been developed to improve these situations. The bioavaibility obtained from transferosomes is somewhat similar to that resulting from subcutaneous injection of the same protein suspension. The transferosomal preparations of this protein also induced strong immune response after the repeated epicutaneous application, for example the adjuvant immunogenic

bovine serum albumin in transferosomes, after several dermal challenges is as active immunologically as is the corresponding injected proteo-transferosomes preparations [38, 39].

Delivery of insulin

By transferosomes is the successful means of non invasive therapeutic use of such large molecular weight drugs on the skin. Insulin is generally administered by subcutaneous route that is inconvenient. Encapsulation of insulin into transferosomes (transfersulin) overcomes these entire problems. After transfersulin application on the intact skin, the first sign of systemic hypoglycemia are observed after 90 to 180 min, depending on the specific carrier composition.[40]

Delivery of interferons:

Transferosomes have also been used as a carrier for interferons, for example leukocytic derived interferone-α (INF-α) is a naturally occurring protein having antiviral, antiproliferive and some immunomodulatory effects. Transferosomes as drug delivery systems have the potential for providing controlled release of the administered drug and increasing the stability of labile drugs. Hafer et al studied the formulation of interleukin-2 and interferone-α containing transferosmes for potential transdermal application .they reported delivery of IL-2 and INF- α trapped by transferosomes in sufficient concentration for immunotherapy.[41]

Delivery of corticosteroids:

Transferosomes have also used for the delivery of corticosteroids. Transferosomes improves the site specificity and overall drug safety of corticosteroid delivery into skin by optimizing the epicutaneously administered drug dose. Transferosomes based corticosteroids are biologically active at dose several times lower than the currently used formulation for the treatment of skin diseases[22].

Transdermal immunization:

Another most important application of transferosomes is transdermal immunization using transferosomes loaded with soluble protein like integral membrane protein, human serum albumin, gap junction protein. These approach offers at least two advantages, first they are applicable without injection and second, they give rise to rather high titer and possibly, to relatively high IgA levels.

Delivery of anesthetics:

Application of anesthetics in the suspension of highly deformable vesicles, transferosomes, induces a topical anesthesia, under appropriate conditions, with less than 10 min. Maximum resulting pain insensitivity is nearly as strong (80%) as that of a comparable subcutaneous bolus injection, but the effect of transferosomal anesthetics last longer.

Delivery of NSAIDS:

NSAIDS are associated with number of GI side effects. These can be overcome by transdermal delivery using ultra-deformable vesicles. Studies have been carried out on Diclofenac and Ketoprofen. Ketoprofen in a Transfersome formulation gained marketing approval by the Swiss regulatory agency (SwissMedic) in 2007; the product is expected to be marketed under the trademark Diractin. Further therapeutic products based on the Transfersome technology, according to IDEA AG, are in clinical development.[42]

Delivery of Anticancer Drugs:

Anti cancer drugs like methotrexate were tried for transdermal delivery using transfersome technology. The results were favorable. This provided a new approach for treatment especially of skin cancer.

Delivery of Herbal Drugs:

Transfersomes can penetrate stratum corneum and supply the nutrients locally to maintain its functions resulting maintenance of skin in this connection the Transfersomes of Capsaicin has been prepared by Xiao-Ying et al. which shows the better topical absorption in comparison to pure capsaicin[43, 44].

CONCLUSION

Ultra-deformable vesicles can provide the novel solution for the transport related problems. They are free from the rigid nature of conventional vesicles and can transport even the large molecules. They work on number of mechanisms working together to provide an excellent carrier system for the drug transport. When tested in artificial systems, Transfersomes can pass through even tiny pores (100 mm) nearly as efficiently as water, which is 1500 times smaller. Drug laden transfersomes can carry unprecedented amount of drug per unit time across the skin (up to 100mg cm2h-1). Ultra-deformable vesicles hold great prospective in delivery of huge range of drug substances which includes large molecules like peptides, hormones and antibiotics, drugs with poor penetration due to unfavorable physicochemical characters, drugs for quicker and targeted action, etc. All above discussed properties of this technology strongly advocate its good future in transdermal drug delivery.

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