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NANOEMULSION AS A NOVEL TRANSDERMAL DRUG DELIVERY SYSTEM

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ABSTRACT

The nanoemulsion is one of the most efficient dispersed nanosystems of droplet size ranging to submicron size. Nanoemulsion are generally transparent or semi transparent system characterized by high stability. They are characterized by high stability. Submicron droplet size and high surfactant concentration makes it an efficient transdermal delivery vehicle. Research works proves that nanoemulsion is far more efficient drug delivery system than other transdermal drug delivery system. The efficacy of the nanoemulsion is enhanced by the nature and type of surfactant and co-surfactant used. Thus in this article ability of the nanoemulsion as a transdermal and topical delivery system is reviewed.

INTRODUCTION: "There is plenty of room at the bottom" was once told by Richard Feynman, a Nobel laureate who in 1959 first proposed the concept of nano scale structures for use. This was the phrase which gave birth to the Nanotechnology. Nanotechnology scales up to one billionth of a meter. Generally, they are considered to be in the range of 10 nm to 100 nm. Various effects such as surface area and area to volume ratio and many other physical properties get magnified when reduced to nanoscale [1].

Most of the current research works in almost all technical and biomedical fields is based on nanosize. Nanoemulsions are thermodynamically stable transparent (translucent) dispersions of oil and water stabilized by an interfacial film of surfactant and cosurfactant molecules having a droplet size of less than 100 nm [2]. Nanoemulsion, which is categorized as multiphase colloidal dispersion, is generally characterized by its stability and clarity. There is an application of high shear generally obtained by micro fluid or ultrasonic approach generally used to reduce the droplet size to nanoscale. Various works are done to correlate the different transdermal drug release from various delivery systems. Studies suggested that the drug release from various formulations such as solid lipid nanoparticles, nanoemulsion and polymeric nanosuspension, nanoemulsion proved to be efficient transdermal delivery vehicle.

The skin: The skin (cutis) is the largest human organ and an excellent biological barrier. **Fig. 1** gives a pictorial representation of skin. They are normally less than ≤2mm thin, and contribute around 4% to a body weight. The outer skin part (epidermis) is in humans generally 0.02-0.2 mm, and 50-150µm, thin. The horny skin layer, of stratum corneum, is the main skin barrier to diffusion. The excellence of this lipid enhanced [4] obstacle to transport is best illustrated by the associated, steep water concentration reduction from around 75% in the viable epidermis to a 10- 30% at an air-exposed skin surface [5-7]. Gender and body also affect skin surface pH, which is mainly below neutral [8] and slightly above pH=5 in humans.

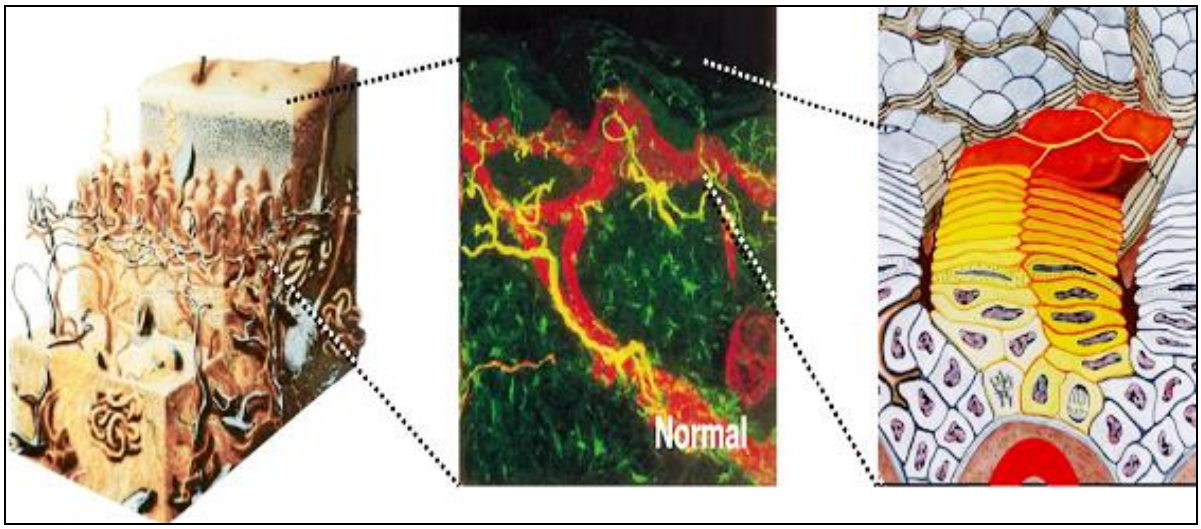

FIG. 1: SKIN DRAWINGS (left: low magnification of the whole skin modified from Casella-Riedel de Haan textbook; right: a higher magnification of the epidermis with the stratum corneum, including a highlighted corneocytes cluster). The confocal-laser-scanning-microscopy visualization of the outer skin region in a cross-section is a blow-up from [22] and shows blood vessels and basal membrane (in red), cutaneous nerves, including the epidermal nerve fibers (in yellow), and epidermis, E, the papillary and the upper reticular dermis, D (in green)

The superficial pH is also ≥1 pH unit higher on excised skin in relation to a living organ [9]. The layer below epidermis is known as dermis. It consists of the outer papillary and the inner reticular dermis, together are usually 5-20 times thicker than epidermis and thus normally measure up to 2 mm in thickness. Cutaneous microvasculature provides the secondary skin barrier, by acting as a sink for the molecules that had diffused across the primary skin barrier. The papillary dermis moreover contains relatively thin elastic elaunin fibers that are broadly perpendicular to skin surface. These fibers merge with the microfibrillar cascade (oxytalan fibers) and then intercalate into the dermal-epidermal junction.

The collagen fibers in the reticular dermis generally are thicker and largely parallel to skin surface. Dermal collagen forms continuous, elastic network and imparts mechanical elasticity to the skin from the reticular and papillary dermis and up to the epidermis [10]. The dermis transitions seamlessly into the hypodermis which together with the dermal fatty deposit helps to absorb mechanical shocks that might otherwise endanger skin vasculature and nerves. The same structures and deposits also contribute to skin elasticity. Owing to its unique biochemical and anatomical properties, each individual skin layer is mechanically different.

The stratum corneum is rather stiff. The underlying epidermis and papillar dermis are less resistant to mechanical stress (low-strain, static stiffness: 0.11kPa), reticular dermis having an intermediate stiffness. Owing to the voluminous nature of the reticular dermis, this layer normally dominates “skin mechanics”, also due to its coupling to the viscoelastic hypodermis that laterally dissipates locally imposed mechanical stress on skin [11]. Being a tactile organ, the skin is also well innervated, yet contains only few myelinated nerve fibers. When the vertically oriented nerve bundles coming from the reticular epidermis enter the papillary dermis they form a horizontal sub-epidermal neural plexus. This plexus is the origin of very fine, non-myelinated epidermal nerve fiber branches that extend between keratinocytes to convey slow cutaneous nociception.

Natural pathways through the Skin Permeability Barrier: The relationship between the skin permeability and epidermal anatomy is not clear. It has not been known for long that hydrophilic transepidermal ‘aqueous pathways’—other than intracutaneous glands and follicles plays a vital role in polar and amphiphilic molecules transport through skin barrier. Enhancement to the transdermal drug transport enhanced by exposing the stratum corneum to a strong electrical current (electroporation/

iontophoresis), mechanical stimulus (e.g. sonoporation /sonophoresis), or thermal stimulus. Alternatively, but less irritatingly, suitable skin penetrants meet the same goal [12]. The best known are the largest "transcutaneous" hydrophilic pores, such as hair follicles [13], or artificial skin micro-perforations, such as localized transport regions (LTR) created by electro- , sono- or optoporation [14]. Separation of transdermal pathways into quantitatively different hydrophilic "pore classes" is therefore difficult until now. It is safe to posit, that hydrophilic conduits through skin barrier have effective openings between ≥5µm (skin appendages) and ≤10 nm (narrow intercorneocyte pores, Cevc, in preparation).

It is furthermore doubtless that sweat ducts (N50µm), pilosebaceous units (5-70µm), and sebaceous glands (5-15µm) represent the largest width/lowest resistance end of the range. Junctions of corneocytes-clusters and cluster boundaries fall within the range. Intracluster pathways, leading along lipidic lamellae for hydrophilic entities, or diffusion pathways, typically running within lipidic layer(s) for lipophilic molecules, form the small width/high resistance end of the range. Diversity of calculated widths and pore expansions suggests that various entities or skin treatments may sense, or help generate, hydrophilic microconduits with different widths or width distributions within the skin permeability barrier.

All of them are in nano-dimensions. Lipophilic cutaneous barrier is more narrowly defined and, unlike the hydrophilic barrier, is governed by molecular weight and distribution coefficient rather than molecular size [15]. The relative height of cutaneous lipophilic barrier consequently decreases with permeant's lipophily, but molecules heavier than 400-500 Da are in any case nearly completely confined to skin surface. The reason is inability of so large permeants to find sufficiently wide defects in the intercellular lipidic matrix to start diffusing through the lipidic parts of cutaneous barrier [16].

Thus, while targeting delivery systems the above mentioned points are taken to consideration. The delivery system is designed in such a way that it shows more permeation and less skin entrapment. The brief process for drug passage across the skin is shown in (**fig. 2)**. The various carrier used to deliver the drug is also mentioned. The best possible carrier is the Nanoemulsion which has proved to be the better carrier than other drug delivery systems.

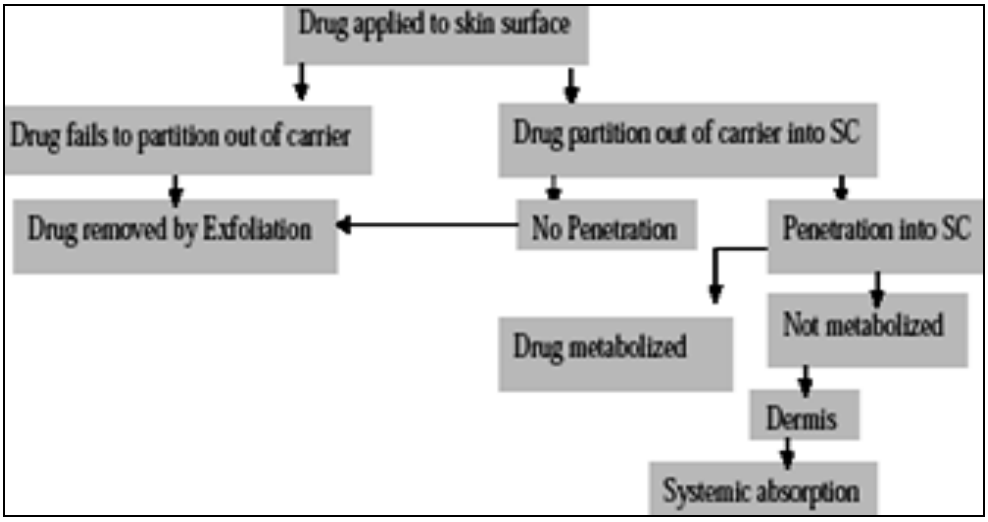

FIG. 2: PASSAGE OF DRUG THROUGH THE SKIN.

Carriers for Transdermal Drug Delivery System:

a. **Micro or nanocapsules:** These are composed of multiple concentric bilayers of surfactant; separated by a polar liquid medium, generally water in which the hydrophilic additives can be incorporated. Their lipid core allows encapsulation of lipid additives and their multi-lamellar (lipid/water) structure creates good skin

affinity leading to cutaneous penetration and good hydration.

b. **Nanoemulsions/ Sub-micron emulsions (SMEs)/Mini-emulsions:** These are oil-in-water emulsions with an average droplet size ranging from 100 to 500 nm. They have very good stability and they do not undergo phase separation during storage. They have a liquid lipophilic core and are appropriate for lipophilic compound transportation. Many studies showed reduced transepidermal water loss, which means support to the barrier function of the skin. Nanoemulsion viscosity is very low, which is interesting because they can be produced as sprays.

c. **Solid Lipid Nanoparticles (SLNs**): These droplets are made by solid lipids. Their sizes range from 50 to 1000 nm. They can also be stabilized by surfactants or polymers. There are mainly three structures: Homogeneous matrix, drug-enriched shell and drug enriched core. They can protect active components against chemical degradation and modulate compound release. SLNs also present occlusive properties because of the formation of a film on the skin. This film formed by lipid fusion is supposed to be a pore-less film with improved skin hydration and protection properties.

d. **Multiple Emulsions:** These W/O/W emulsions consist in the dispersion of a W/O emulsion in an aqueous phase under several conditions. One can incorporate different water-soluble ingredients (even if they are incompatible) and also oil soluble additives. Like SLNs, these substances will be protected and release sustained by controlling droplet breakdown. These systems can have high oily phase contents (65%, Trixera, Bain emollient, Avene) and thus present good hydration. Their efficacy has been demonstrated in dermatology to treat stretch marks (Triffadiane, CS Dermatologie).

e. **Microemulsions:** These formulations have been shown to be superior for cutaneous delivery compared to other conventional vehicles. These systems are identified as transparent mixtures of water, oil and surfactants. They are thermodynamically stable and optically isotropic. Microemulsions are spontaneously produced in a narrow range of oil-water-surfactant composition, represented on pseudo-ternary diagram phases. They are dynamic systems with continuously fluctuating interfaces. Their good dermal and transdermal delivery properties could be attributed to their excellent solubilizing properties.

f. Their high solubilizing properties improve biodispensibility and thus reduce the efficient dose thereby increasing tolerability. Furthermore, their restructuring effect on skin and hair (due to their high lipid content) make microemulsion formulations adapt to altered skin and hair conditions.

g. Vesicular carriers

- **Liposomes:** These are colloidal particles formed as concentric biomolecular layers that are capable of encapsulating drugs. Their delivery mechanism is reported to be associated with accumulation of the liposomes and associated drug in the stratum corneum and upper skin layers, with minimal drug penetrating to the deeper tissues and systemic circulation. It is interesting that the most effective liposomes are reported to be those composed of lipids similar to stratum corneum lipids, 17 which are most likely to enter stratum corneum lipid lamellae and fuse with endogenous lipids.

- **Niosomes:** These are vesicles composed of nonionic surfactants that have been evaluated as carriers for a number of drug and cosmetic applications. This carrier has more permeability than liposomes for transdermal drug delivery.

- **Transfersomes:** These are vesicles composed of phospholipids as their main ingredient with 10-25% surfactant (such as sodium cholate) and 3-10% ethanol. The surfactant molecules act as “edge activators”, conferring ultra deformability on the transfersomes, which

reportedly allows them to squeeze through channels in the stratum corneum that are less than one-tenth the diameter of the transfersome.

- **Ethosomes:** These are leptosomes with high alcohol content capable of enhancing penetration to deep tissues and the systemic circulation [18-21]. It is proposed that alcohol fluidizes.

Various works has been done to correlate the different transdermal drug release from various delivery systems. Shakeel *et al*. suggested that the drug release of aceclofenac was done by various formulations such as solid lipid nanoparticles, nanoemulsion and polymeric nanosuspension. The work finally concluded that the *in- vitro* drug release was found to be optimum

Nanoemulsions: A novel drug delivery for transdermal treatment: Nanoemulsions are thermodynamically stable transparent (translucent) dispersions of oil and water stabilized by an interfacial film of surfactant and cosurfactant molecules having a droplet size of less than 100 nm[16-17] as shown in **(Fig 3)**.

FIG 3: PICTURE OF NANOEMULSION (LEFT) OF SIZE 35 NM AND MICRO EMULSION (RIGHT) OF SIZE 1µm.[23]

Many studies have shown that Nanoemulsion formulations possess improved transdermal and dermal delivery properties *in- vitro*, [20-21] as well as *in-vivo* [18-19]. Nanoemulsions have improved transdermal permeation of many drugs over the conventional topical formulations such as emulsions and gels[16, 17, 22]. This present work describes the potential of Nanoemulsion systems in transdermal delivery of Flurbiprofen using nonirritating, pharmaceutically acceptable ingredients without using additional permeation enhancers, because excipients of Nanoemulsion themselves act as permeation enhancers. The formation and stabilization of nanoemulsion is the topic to be discussed, nanoemulsion are highly stabilized system thus the composition of the nanoemulsion plays a vital role in the in its formation and stabilization. The selection of proper oil, surfactant and cosurfactants is crucial. In this topic an overview of the theory and principle behind the formation of the nanoemulsion is discussed.

Method of preparation [16-18]:

a) **High pressure homogenization:** The technique has use of high pressure homogenizer. The proess generally involves a piston system. This produces high shear which generally causes the reduction of particle size to sub-micron level causes the formation of Nanoemulsion.

b) **Microfludization:** It's a patented mixing technology that involves a high pressure positive displacement pump with a pressure of 500- 20000 psi as depicted in **(fig 4)**. This pressure forces the phases to pass through micro channels. The two phases mixed and proceed as coarse emulsion when made to pass through interaction chamber results in the formation of Nanoemulsion. This is made to pass through micro channels.

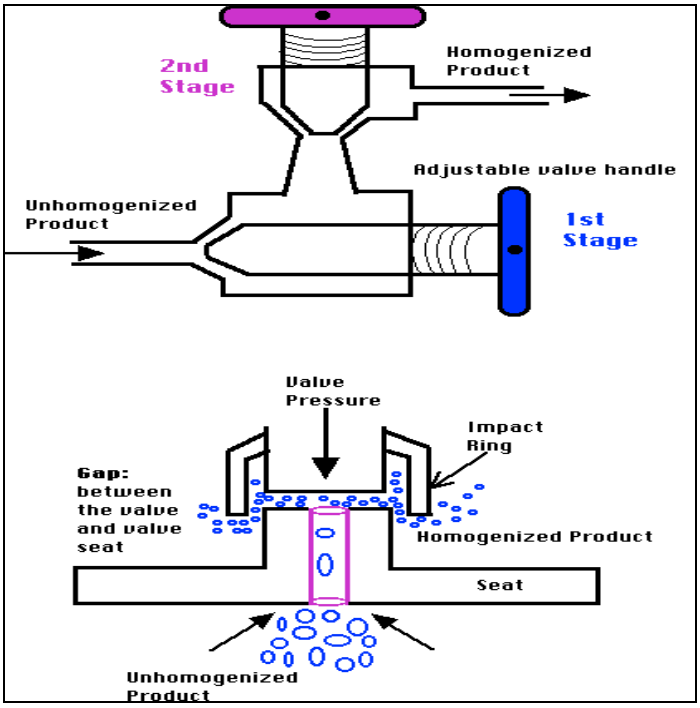

FIG. 4: MICRO FLUIDIZER WITH THE MECHANISM OF WORKING

c) **Ultrasonication:** The preparation of Nanoemulsion is reported in various research papers which aim to use the ultrasonic sound frequency for the reduction of the droplet size. Another approach is the use of a constant amplitude sonotrode at system pressures in excess of the ambient value shown in (**fig 5)**.

FIG. 5: PROBE SONICATOR USED IN LABORATORY SCALE FOR PREPARATION OF NANOEMULSION

It is well known that increasing the external pressure increases the cavitations threshold within an ultrasonic field and thus fewer bubbles form. However, increasing the external pressure also increases the collapse pressure of cavitations bubbles. This means that the collapse of the bubbles when cavitation occurs becomes stronger and more violent than when the pressure is at atmospheric conditions. As cavitation is the most important mechanism of power dissipation in a low frequency ultrasonic system, these changes in navigational intensity can be related directly to changes in the power density. The system also uses a water jacket to control the temperature to optimum level [26].

d) **Phase inversion method:** In this method, fine dispersion is obtained by chemical energy resulting of phase transitions produced by emulsification pathway. The phase transition is produced by varying the composition of the emulsion and keeping temperature constant or vice versa. The phase inversion temperature was first done by Shinoda *et al*. it was concluded that increase in temperature results in the chemical changes of polyoxyethelene surfactants by degradation of the polymer chain with the temperature [27].

Theory of the formation of Nanoemulsion [28]: In Nanoemulsion which is categorized as multiphase colloidal dispersion which is generally characterized by its stability and clarity. There is an application of high shear generally obtained by micro fluid or ultrasonic approach generally used to reduce the droplet size to nanoscale. There is a marginal difference between the terms Nanoemulsion and microemulsion also known as micellar phase or mesophase.

The microemulsion generally forms through thermodynamic self assembly whereas nanoemulsion requires external shear for rupturing the droplets. In retrospect, the historical choice of the word "microemulsion" to describe the nanoscale is unfortunate since they are structurally between 1 to 100 nm as for Nanoemulsion. Micro emulsions are not the emulsions of micro scale droplets. They are formed by self assembled equation phase in which the surface tension dose not play a significant role.[28] The Nanoemulsions underlines the basic principle in its formulation. They generally comprises of two immiscible phase with an interfacial tension between them reduced by addition of surfactant.

Stability factors of Nanoemulsion: As a general consideration the stability of nanoemulsion largely depends upon the following factors;

a) Coalescence of the two droplets of dispersed phase due to the surface tension and intermolecular attractions. This is mainly reduced by addition of suitable surfactants.

b) If the dispersed phase has high solubility in the dispersed medium. This results in diffusive migration of smaller droplets with low Laplace pressure to larger droplets of high Laplace pressure also known as Ostwald's ripening. The dispersed phase should be selected such that it should have minimum or no solubility in the continuous phase.

Laplace pressure: At a low volume of dispersed phase (ϕ) an isolated droplet with a curved radius (a). The curved interface formed due to surfactant exerts a pressure on the molecule inside the droplet. This pressure is called the Laplace pressure (Π_L). For non-spherical droplets, Π_L is proportional to the sum of the inverse of the two principal radii of curvature of the interface. Due to the inverse dependence of Π_L on a, the molecules in smaller droplets experience a higher pressure than those in larger droplets.

$$\Pi_L = 2\sigma/a \quad \text{(1)}$$

Π_L= Laplace pressure; σ= surface tension

The formulation of Nanoemulsion involves stress applied (τ) which should exceed the Laplace pressure inorder to stretch and break the droplet to smaller size.

$$\tau = \eta_c \dot{\gamma} \quad \text{(2)}$$

η_c= viscosity of continuous phase; $\dot{\gamma}$= rate of share

As per above consideration,

$$\tau >> \Pi_L$$

In general, due to the presence of high quantity of surfactants in continuous phase as soon as the interfacial area is created by shear surfactant coats the interface.

Factors affecting the formulation of Nanoemulsion:

- Appropriate composition is required to avoid Oswald ripening the dispersed phase should be highly insoluble in the dispersed medium.
- The surfactant is an essential part of the Nanoemulsion. They should not form lyotropic liquid crystalline "microemulsion" phases. Systems containing short chain alkanes, alcohols, water, and surfactants form the phases which are generally used with the co surfactant.
- The presence of excess surfactants enables new surface area of nano scale to be rapidly coated during emulsification there by inhibiting induced coalescence.
- Extreme share must be applied to rupture microscale droplets to nanoscale by providing the stress level to reach above the Laplace pressure of the droplets with a pressure of 10-

100 atm. Out of various methods ultrasonication is widely used in laboratory.

Effect of surfactant in the formation of Nanoemulsion: Out of various surfactants required to prepare nanoemulsion, the most important components are primary surfactant and secondary surfactant (also known as cosurfactant). As the following system prepared are isotropic in nature, its difficult to produce as it involves the interaction of the other constituents. Thus it is necessary to study the phase diagram to establish the effects of the constituents which mainly involves the study of oil, surfactant and cosurfactant at various ratios and proportion.

Formulation of nanoemulsion by spontaneous emulsification method can be done when the interfacial tension is low enough such that increase in the free energy due to creation of free surface can be compensated by increasing the entropy. This is the reason behind the consumption of large amount of the surfactant.

It is generally proposed to select a surfactant that can reduce interfacial tension, provide flexibility to the film to form nanodroplets. It is generally proposed that the surfactant hydrocarbon volume **(V)**, effective chain length **(l_c)** and head group area **(a)**.

$V=a_o.l_c$ ..... (3)

According to the equation 3 surfactant with balanced solubility in oil and water phase can meet with the criteria. Cosurfactant along with surfactant too playas a important role in the formation of nanoemulsion. A cosurfactant with a hydrocarbon moiety of different size is used with respect to the surfactants. The cosurfactants are generally used to enhance the interfacial fluidity by penetration into the surfactant film. They ensure that surfactant film is flexible enough to deform readily deform to form fine nanodroplets. There are a lot of examples of the surfactants and cosurfactants used till now in various research works, some of them are,

Surfactants = Lecithin, Sodium Dodecyl Sulphate, Labrasol, Tween (20, 40, 60, 80), Span, Triton-X, Igepal, Polyoxyethelene surfactants, etc.

Cosurfactant= Ethanol, Propanol, Isopropanol, Butanol, Cremophor, Propylene Glycol, etc.

Mechanism of Transmembrane Permeation: Nanoemulsion permeation involves various factors they are;

- Solubility of the drug.
- Hair follicles in percutaneous absorption
- The presence of stratum corneum and lipid layer.
- Presence of surfactant and cosurfactants too plays a role in the formation of nanoemulsion.

Nanoemulsion as transdermal drug delivery system varies a lot from the other sub micron drug delivery systems. Many formulations were prepared and investigation was done using permeation enhancers. The various studies regarding the investigation of transdermal permeation of Nanoemulsion was done by Fiyaz *et al*.

The result obtained from the study of the skin done by FTIR and DSC showed a marked difference in the skin composition of the treated and untreated skin with nanoemulsion. It was found that the breaking of the tight hydrogen bonding between proteins and lipids in the subcutaneous layer as the main changes shown in the study. These alterations results in the disruption of the lipid layer, thus producing distinct void spaces in the epidermis. Further histopathological study clearly showed the morphological changes in the epidermal layer.

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