Review Article

PENETRATION ENHANCEMENT TECHNOLOGIES FOR TRANSDERMAL DRUG DELIVERY SYSTEMS

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ABSTRACT

There is considerable interest in the skin as a site of drug application for both local and systemic effect. The transdermal route has numerous advantages over the more traditional drug delivery routes. These include high bioavailability, absence of first pass hepatic metabolism, steady drug plasma concentrations and the fact that therapy is non-invasive. The main obstacle to permit drug molecules is the outermost layer of the skin, the stratum corneum. However, the major limitation of this route is the difficulty of permeation of drug through the skin. In this review, we have discussed the physical, chemical, biological and other penetration enhancement technology for transdermal drug delivery as well as the probable mechanisms of action.

Keywords: Biological enhancement, chemical enhancement, physical enhancement, transdermal delivery.

INTRODUCTION

Transdermal delivery of drugs through the skin to the systemic circulation provides a convenient route of administration for a variety of clinical indications. Transdermal delivery systems are currently available containing scopolamine (hyoscine) for motion sickness, clonidine and nitroglycerin for cardiovascular disease, fentanyl for chronic pain, nicotine to aid smoking cessation, oestradiol (alone or in combination with levonorgestrel or norethisterone) for hormone replacement, testosterone for hypogonadism. Despite the small number of drugs currently delivered via this route, it is estimated that worldwide market revenues for transdermal products are US$3B, shared between the USA at 56%, Europe at 32% and Japan at 7%. In a recent market report it was suggested that the growth rate for transdermal delivery systems will increase 12% annually through to 2007. Transdermal products for cardiovascular disease, Parkinson's disease, Alzheimer's disease, depression, anxiety, and attention deficit hyperactivity disorder (ADHD), skin cancer, female sexual dysfunction, post-menopausal bone loss and urinary incontinence are at various stages of formulation and clinical development. The application of transdermal delivery to a wider range of drugs is limited due to the significant barrier to penetration across the skin which is associated primarily with the outermost stratum corneum layer of the epidermis. Consequently the daily dose of drug that can be delivered from a transdermal patch is 5-10 mg, effectively limiting this route of administration to potent drugs. Moreover, achieving high and constant drug flux through the skin is a daunting task, with a low probability of success, unless one compromises the protective skin barrier function. Rather sophisticated techniques must therefore be used to overcome the skin barrier by means other than a hypodermic needle. It was not until recently that several minimally invasive techniques became available. Significant effort has been devoted to developing strategies to overcome the impermeability of intact human skin. These strategies include passive and active penetration enhancement and technologies to bypass the stratum corneum. This review describes the routes of penetration, how drug properties influence penetration and the techniques that have been used to enhance penetration across human skin.[1-4]

A Brief Review of Skin Structure:

The skin can be considered to have four distinct layers of tissue. Figure 1 illustrates view of skin structure.[5]

1. Non-viable epidermis (stratum corneum)
2. Viable epidermis
3. Viable dermis
4. Subcutaneous connective tissue

   (Hypodermis)

Non-viable Epidermis (stratum corneum)

Stratum corneum (SC) is the outer most layer of skin, which is the actual physical barrier to most substance that comes in contact with the skin. The stratum corneum is 10 to 20 cell layer thick over most of the body. Each cell is a flat, plate-like structure - 34-44 µm long, 25-36 µm wide, 0.5 to 0.20 µm thick - with a surface area of 750 to 1200 μm^2 stocked up to each other in brick like fashion. Stratum corneum consists of lipid (5-15%) including phospholipids, glycosphingolipid, cholesterol sulfate and neutral lipid, protein (75-85%) which is mainly keratin.[6]

Viable epidermis

This layer of the skin resides between the stratum corneum and the dermis and has a thickness ranging from 50- 100 µm. The density of this region is not much different than water. The water content is about 90%.[6]

Dermis

Just beneath the viable epidermis is the dermis. It is a structural fibrin and it can be found histologically in normal tissue. Dermis thickness range from 2000 to 3000 µm and consists of a matrix of loose connective tissue composed of fibers.[6]

Subcutaneous connective tissue

The subcutaneous tissue or hypodermis is not actually considered a true part of the structured connective tissue is composed of loose textured, white, fibrous connective tissue containing blood and lymph vessels, secretary pores of the sweat gland and cutaneous nerves. Most investigators consider drug permeating through the skin enter the circulatory system before reaching the hypodermis, although the fatty tissue could serve as a depot of the drug.[6]

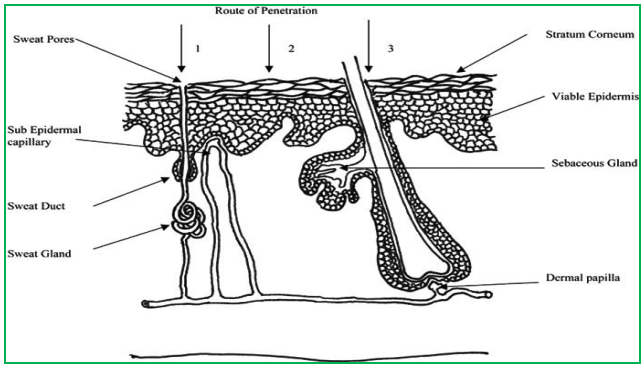

Figure 1: View of skin structure

Pathway of Transdermal Permeation:

Permeation can occur by diffusion via:

1. Transdermal permeation, through the stratum corneum.

2. Intercellular permeation, through the stratum corneum.

3. Transappendaged permeation, via the hair follicle, sebaceous and sweat Glands.[6]

Most molecules penetrate through skin via intercellular micro route and therefore many enhancing techniques aim to disrupt or bypass its elegant molecular architecture. Ideally, penetration enhancers reversibly reduce the barrier resistance of the stratum corneum without damaging viable cells. They should be non-toxic, non-irritating and non-allergenic. They would ideally work rapidly; the activity and duration of effect should be both predictable and reproducible. They should have no pharmacological activity within the body. The penetration enhancers should work unidirectional, i.e., they should allow therapeutic agents into the body whilst preventing the loss of endogenous materials from the body. When removed from the skin, barrier properties should return both rapidly and fully to normal. They should be cosmetically acceptable with an appropriate skin feel. Not surprisingly, no such material that possesses the above ideal properties has yet been discovered although some chemicals demonstrate several of the above attributes.[6]

A. CHEMICAL PENETRATION ENHANCERS

1. Oxazolidinones

They have ability to localize co-administered drug in skin layers, resulting in low systemic permeation. Oxazolidinones such as 4-decyloxazolidin-2-one has been reported to localize the delivery of many active ingredients such as retinoic acid and diclofenac sodium in skin layers.[7]

2. Urea

Cyclic urea permeation enhancers are biodegradable and non-toxic molecules consisting of a polar parent moiety and a long chain alkyl ester group. As a result, enhancement mechanism may be a consequence of both hydrophilic activity and lipid disruption mechanism.[8]

3. Pyrrolidones

N-methyl-2-pyrolidone was employed with limited success as a penetration enhancer for captopril when formulated in a matrix-type transdermal patch. The pyrrolidones partition well into human stratum corneum within the tissue and they may act by altering the solvent nature of the membrane. Pyrrolidones have been used to generate reservoirs within the skin membrane. Such a reservoir effect offers a potential for sustained release of a permeant from the stratum corneum over extended time periods.[9]

4. Alcohol, Glycol and Glycerides

Ethanol is the most commonly used alcohol as a transdermal penetration enhancer. It increases the permeation of ketoprofen from a gel-spray formulation and triethanolamine salicylate from a hydrophilic emulsion base. It also acts as a vehicle for menthol in increasing the penetration of methyl paraben. Ethanols in combination with trichloropropanol and with water were used as two cosolvent systems for zalcitabine, didanosine, zidovudine, tegafur, aceclofenac, and ibuprofen. The permeation rate of zalcitabine, didanosine, and zidovudine increased as the volume fraction of ethanol in the two cosolvent systems was increased, and it reached a maximum at 50–60% v/v of ethanol. Flux of tegafur, aceclofenac and ibuprofen was higher from the ethanol-water cosolvent system than from the ethanol-TCP system. Propylene glycol (PG) promoted the flux of heparin sodium, verapamil hydrochloride, and ketoprofen, but at higher concentrations, it inhibited the flux of ketoprofen. A saturated solution of terpenes in a PG-water cosolvent system enhanced the flux of 5-FU, terpene activity being dependent on PG content and with the maximum flux obtained from formulations containing 80% PG. Also, PG increases drug partitioning and drug permeation. PG, in combination with azone, increases the flux of methotrexate, piroxicam, cyclosporin A and 5-Fluorouracil (5-FU). Flux of estradiol was 10 times higher when PG was used in conjunction with 5% oleic acid. Urea

analogues were effective in enhancing the permeation of 5-FU only when PG was used as a vehicle. Short-chain glycerides are also effective as permeation enhancers (e.g., TCP). For instance, glycerin tricaprylate (caprylic acid triglyceride) in combination with ethanol is used as a solvent system. TCP is an excellent hydrophobic vehicle and promoted the permeability of tegafur combined with ethanol. Glyceryl monocaprylate enhanced the partitioning of papaverine across hairless rat skins. Sefsol 318, a medium-chain glyceride, increased the permeation of papaverine hydrochloride by almost 820 times by increasing the fluidity of the lipoidal membrane of the stratum corneum.[9]

5. Sulphoxides and similar chemicals

Dimethyl sulphoxides (DMSO) is one of the earliest and most widely studied penetration enhancers. It is a powerful aportic solvent which hydrogen bonds with itself rather than with water. It is colourless, odourless and is hydroscopic and is often used in many areas of pharmaceutical sciences as a "universal solvent". DMSO alone has been applied topically to treat systemic inflammation. DMSO works rapidly as a penetration enhancer - spillage of the material onto the skin can be tasted in the mouth within a second. Although DMSO is an excellent accelerant, it does create problems. The effect of the enhancer is concentration dependent and generally cosolvents containing > 60% DMSO are needed for optimum enhancement efficacy. However, at these relative high concentrations, DMSO can cause erythema and wheal of the stratum corneum. Denaturing of some skin proteins results in erythema, scaling, contact urticaria, stinging and burning sensation. Since DMSO is problematic for use as a penetration enhancer, researchers have investigated a similar chemically-related material as an accelerant. DMSO may also extract lipids, making the horny layer more permeable by forming aqueous channels. The mechanism of the sulphoxide penetration enhancers is widely used to denature protein and, on application to human skin, has been shown to change the intercellular keratin conformation, from helical to ß sheet.[10, 11]

6. Azone

Azone (1-dodecylazacycloheptan-2-one or laurocapran) was the first molecule specifically designed as a skin penetration enhancer. Azone is a colourless, odourless liquid with a melting point of -7 ºC and it possesses a smooth, oily but yet non-greasy feel. Azone is a highly lipophilic material with a log p (octanol / water) of around 6.2 and it is soluble in and compatible with most organic solvents including alcohol and propylene glycol. Azone enhances the skin transport of a wide variety of drugs including steroids, antibiotics and antiviral agents. Azone is most effective at low concentrations, being employed typically between 0.1- 5% but more often between 1- 3%. Azone partitions into a bilayer lipid to disrupt their packing arrangement but integration into the lipid is unlikely to be homogeneous. Azone molecules may exist dispersed within the barrier lipoid or separate domains within the bilayer.[12]

7. Fatty Acids and Esters

Percutaneous drug absorption has been increased by a wide variety of long-chain fatty acids, the most popular of which is oleic acid. It is of interest to note that many penetration enhancers such as azone contain saturated or unsaturated hydrocarbon chains and some structure-activity relationships have been drawn from the extensive studies of Aungst who employed a range of fatty acids, acids, alcohols, sulphoxides, surfactants and amides as enhancers for naloxone. Shin and Lee studied various penetration enhancers like glycols (diethylene glycol and tetraethylene glycol), fatty acids (lauric acid, myristic acid and capric acid) and anionic surfactant (polyoxyethylene-2-oleyl ether, polyoxy ethylene-2-stearly ether) on the release of triprolidone.[13]

8. Essential oil, Terpenes and Terpenoids

Terpenes are found in essential oils, and are compounds comprising of only carbon, hydrogen and oxygen atoms, but which are not aromatic. Numerous terpenes have long been used as medicines as well as flavoring and fragrance agents. The essential oils of eucalyptus, chenopodium and ylang-ylang have been found to be effective penetration enhancers for 5-flouorouracil transversing human skin in vivo. Cornwell et al investigated the effect of 12 sesquiterpenes on the permeation of 5-flurouracil in human skin. Pretreatment of epidermal membranes with sesquiterpene oil or using solid sesquiterpenes saturated in dimethyl isosorbide increased the absorption of 5- flurouracil. L-menthol has been used to facilitate in vitro permeation of morphine hydrochloride through hairless rat skin as well as diffusion of imipramine hydrochloride across rat skin and hydrocortisone through hairless mouse skin. One mechanism by which this agent operates is to modify the solvent nature of the stratum corneum, thus improving drug partitioning into the tissue. Many terpenes permeate human skin well and large amounts of terpene have been found in the epidermis after application from a matrix-type patch. Terpenes may also modify drug diffusivity through the membrane. During steady state permeation experiment, terpenes as penetration enhancers, the lag time for permeation was usually reduced, indicating some increase in drug diffusivity through the membrane following terpene treatment.[14, 15]

9. Cyclodextrin Complexes

Cyclodextrin complexes of a number of drugs have been formed, and such a combination usually enhances the permeation of drugs. For instance, an inclusion complex of piroxicam with B-cyclodextrin increased the drug flux three times across hairless mouse skin, and a similar complex of clonazepam with methyl-B-cyclodextrin improved its release profile from Carbopol hydrogel through cellulose nitrate membrane. In solution, cyclodextrin forms a complex with enhancers like

quaternary ammonium salts and shifts their critical micellar concentration to higher values, thereby decreasing the toxic effect of such enhancers. Transdermal absorption of alprostadil (AP) from its B-cyclodextrin complex and O-carboxymethyl-O-ethyl-B-cyclodextrin (CME-B-CD) complexes were compared across hairless mouse skin. HPE-101 (1-[2-(decylthio) ethyl] azacyclopentan-2 one) was included as a permeation enhancer in both cases. Flux from the latter complex was 10 times higher than from the former one. It was concluded that a combination of CME-B-CD and HPE-101 enhances the topical bioavailability of the drug.[16]

10. Phospholipids

Phosphatidyl glycerol derivative increased the accumulation of bifonazole in skin and the percutaneous penetration of tenoxicam; phosphatidyl choline derivatives promoted the percutaneous penetration of erythromycin. Results suggest that phospholipids containing unsaturated fatty acids in the hydrophobic group are strong permeation enhancers for percutaneous delivery of some topically applied drugs.[17]

11. Lipid Synthesis Inhibitors

The barrier layer (i.e., stratum corneum) consists of a mixture of cholesterol, free fatty acids, and ceramides, and these three classes of lipids are required for normal barrier function. Addition of inhibitors of lipid synthesis enhances the delivery of some drugs like lidocaine and caffeine. Fatty acid synthesis inhibitors like 5-(tetradecyloxy)- 2-furancarboxylic acid (TOFA) and the cholesterol synthesis inhibitors fluvastatin (FLU) or cholesterol sulfate (CS) delay the recovery of barrier damage produced by prior application of penetration enhancers like DMSO, acetone, and the like. It was concluded that modulation of lipid biosynthesis following the application of conventional chemical penetration enhancers causes a further boost in the transdermal permeation.[18]

12. Dodecyl-N, N-Dimethylamino Acetate (DDAA)

DDAA increased the transdermal permeation of a number of drugs, like propranolol hydrochloride and timolol maleate. It was found to be as effective an enhancer as azone, but it possesses an advantage over azone: Skin irritation with DDAA is reversed in a short time compared to azone. DDAA also increased the transdermal flux of 5-FU through snake skin. Moreover, substitution of one of the hydrogen atoms of the acetate moiety with a methyl group greatly increased its penetration power. The increase in the flux of tetrapeptidehisetal by DDAA was 1.5-fold more than azone across hairless mouse skin. The permeability-enhancing effect was due to changes in the lipid structure of the stratum corneum, like azone and oleic acid. The improvement in transdermal permeation of sotalol by DDAA was the same as that produced by iontophoresis. DDAA causes the disruption of the lipoidal bilayer of the stratum corneum. Its duration of action is shorter than that of azone and dodecyl alcohol because of the presence of hydrophilic groups. So, there is faster recovery of the skin structure and hence less irritation potential. It also exerts a hydrating effect on the skin.[19]

13. Amino Acid Derivatives

Various amino acid derivatives have been investigated for their potential in improving percutaneous permeation of drugs. N-Dodecyl-l-amino acid methyl ester and N-pentyl-N-acetyl prolinate were studied. Application of these two enhancers on excised hairless mouse skin 1 hour prior to drug treatment produced greater penetration of hydrocortisone from its suspension. N-Pentyl-N-acetyl prolinate also enhances the flux of benzoic acid across human cadaver skin; it is nontoxic at low doses, but at higher doses produces dose-dependent central nervous system toxicity. Esters of omega amino acids like octyl-6-aminohexanoate and decyl-6-aminohexanoate enhanced the transdermal permeation of theophylline in aqueous and oily vehicles, respectively. The effectiveness of the biodegradable penetration enhancer dodecyl N, N-dimethylamino isopropionate (DDAIP; dodecyl-N, N-dimethyl-l-alanine) was compared to dodecyl-N,N-dimethylamino acetate (DDAA), azone, and other known permeation enhancers. DDAIP showed a dose-dependent increase in the flux of 5-FU. Also, DDAIP produced better enhancement than DDAA and azone. It increased the transdermal flux of indomethacin. Hydrogen bonding and dipole-dipole interactions were reported between the drug and DDAIP.[20]

14. Clofibric Acid

Esters and amides of clofibric acid were studied for their permeation-enhancing property using nude mice skin. The best enhancement of hydrocortisone acetate and betamethasone-17-valerate was observed with clofibric acid octyl amide when applied 1 hour prior to each steroid. Amide analogues are generally more effective than ester derivatives of the same carbon chain length.[21]

15. Enzymes

Due to the importance of the phosphatidyl choline metabolism during maturation of the barrier lipids, the topical application of the phosphatidyl choline–dependent enzyme phospholipase C produced an increase in the transdermal flux of benzoic acid, mannitol, and testosterone. Three epidermal enzymes (triacylglycerol hydrolase [TGH], acid phosphatase, phospholipase A2) were also studied for their effect. Acid phosphatase was ineffective; TGH increased the permeation of mannitol, while phospholipase phospholipase A2 increased the flux of both benzoic acid and mannitol. Pretreatment of skin with papain produced reversible alterations in the protein structure of the stratum corneum. These alterations resulted in increased permeation of proteins of various molecular weights, with the effect decreasing with increasing molecular weight.[22]

16. Water

Water has been well-characterized as an agent that can increase drug flux across the skin. The water content of the SC in humans is typically 15 to 20% under normal physiological conditions. However, when fully hydrated, such as may occur when the skin is occluded, the highest water content can increase the weight of the SC by up to 400%. The hydration of the occluded area increases because little or no water is lost through evaporation. Topical formulations that employ hydrophobic bases as vehicles can also achieve this effect as oily bases limit transepidermal water loss (TEWL). It has been suggested that hydration causes swelling of the corneocytes, which in turn affects the arrangement of the SC lipid bilayers. These disruptions cause a merging of the interrupted polar and continuous non-polar intercellular routes to form a continuous combined polar and non-polar route across the SC. However, it has been argued that there is actually no major modification in the SC lipid packing except for water pools in the SC lipid bilayer. It seems certain that SC hydration can increase drug flux, but more research is needed to clearly address the exact mechanism of TDD enhancement.[23, 24]

17. Surfactants

Surface active agents or surfactants are molecules that possess both polar and non-polar regions within one molecule. They are widely used in the detergents and in cosmetic and pharmaceutical industries to solubilize poorly soluble molecules in the desired vehicle. The surfactants can be divided into four major groups: cationic surfactants, anionic surfactants, zwitter ionic surfactants, and non-ionic surfactants. The cationic surfactants such as cetyl-trimethyl-ammonium bromide (CTAB), cetylpyridinium chloride and benzalkonium chloride have been demonstrated to significantly increase the transdermal flux of some drugs. However, the irritation and damage that they cause to the skin means that this class has only limited use as a chemical enhancer. Similarly, the zwitterionic surfactants also have limited use as transdermal enhancers, as they also can cause skin irritation.[26] Consequently, most research in this area is carried out using the anionic and non-ionic surfactants that tend to cause less skin irritation. The anionic surfactant class which includes sodium laurate and sodium lauryl sulfate, disrupts the packing of SC lipids leading to improved drug penetration across the skin. The non-ionic surfactants such as Polysorbates, Spans and Tweens tend to cause less irritation and less damage to the skin, but the enhancement factor tends to be lower than the anionic class.[25, 26]

18. Liposomes, niosomes, transfersomes and ethosomes

Lipid-based encapsulation techniques have been used in drug delivery via several routes for many years. These particulates can be characterized as lipid or lipid and surfactant bilayer spheres. Hydrophilic drugs can be incorporated within the aqueous core of these complex structures, whereas, hydrophobic drugs may be incorporated within the bilayer. This strategy has been successfully developed and applied to many parenteral and oral formulations, especially for tissue-specific drug delivery. Liposome and niosome formulations have also been utilized in topical drug delivery where improved drug retention within the SC has been accompanied by minimal transdermal flux. Liposomes for topical delivery are usually constructed using phospholipids or ceramides, whereas, the niosome (a liposome variant) has an additional non-ionic surfactant included in its formulation. Although showing promise in the area of topical drug delivery, the use of conventional liposomes (including niosomes) is unlikely to lead to any advances in transdermal drug delivery. This topical drug delivery by conventional liposomes is thought to be linked to the large size of the liposomes and the deformation that takes place within the SC. Interestingly, a liposome derivative, the transfersome, has been shown to permeate the skin into the systemic circulation. Transfersomes consist of phospholipids, surfactants and a small amount of ethanol, which is added in the final preparative stage. Transfersomes are ultra-flexible, thus allowing them to squeeze through narrow intercellular spaces within the skin. The driving force of transfersome permeation is claimed to be generated by xerophobia, a condition where an entity moves from a low water potential area to a high water potential area, to remain hydrated and avoid dryness. This force is manifested in vivo (where the water gradient increases as one moves away from the skin surface) to facilitate transfer of some movements to the deeper skin layers, until it is taken up by the systemic circulation. Further research into these highly deformable vehicles is needed to assess whether they are likely to be capable of delivering drugs at clinically efficacious level. A further variant of the liposome is the ethosome, which is a soft vesicle consisting of phospholipids and a high amount of ethanol. This variant is also highly deformable and has also been shown to be capable of delivering a variety of drugs, including testosterone, trihexyphenidyl hydrochloride, and insulin across the skin. The mode of action of these flexible liposomes may be associated with the presence of ethanol, which can enhance drug percutaneous penetration, by unknown mechanism.[27-29]

B. PHYSICAL PENETRATION ENHANCERS

1. Electroporation

The use of electropermeabilization as a method of enhancing diffusion across biological barriers dates back as far as 100 years. Electroporation involves the application of high-voltage pulses to induce skin perturbation. High voltages (≥100 V) and short treatment durations (milliseconds) are most frequently employed. Other electrical parameters that affect delivery include pulse properties such as waveform, rate and number. The increase in skin permeability is suggested to be caused by the generation of transient pores during electroporation. The technology has been successfully used to enhance the skin permeability of molecules with differing

lipophilicity and size (i.e., small molecules, proteins, peptides, and oligonucleotides), including biopharmaceuticals with a molecular weight greater that 7 kDa, the current limit for iontophoresis. Figure 2 describes schematic representation of electroporetion.[30]

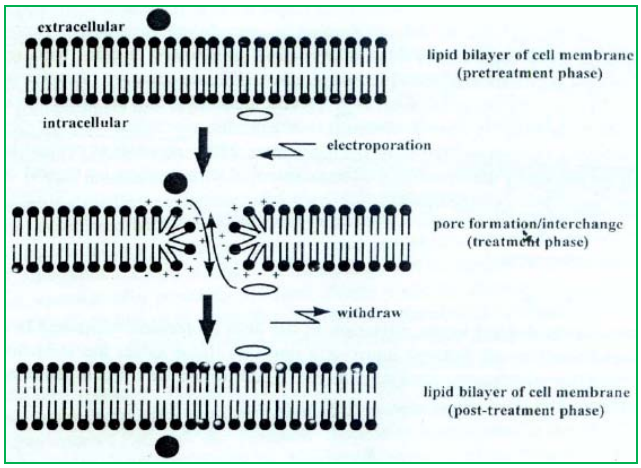

Figure 2: Schematic representation showing electroporation

2. Iontophoresis

This method involves enhancing the permeation of a topically applied therapeutic agent by the application of a low-level electric current, either directly to the skin or indirectly via the dosage form. Increase in drug permeation as a result of this methodology can be attributed to one or a combination of electrorepulsion (for charged solutes), electro-osmosis (for uncharged solutes), and electroperturbation (for both charged and uncharged) mechanisms. Parameters that affect design of an iontophoretic skin delivery system include electrode type, current intensity, pH of the system, competitive ion effect, and permeant type. The launch of commercialised systems of this technology either has occurred or is currently under investigation by various companies. Extensive literature exists on the various types of drugs investigated using iontophoretic delivery. The Phoresor™ device (Iomed Inc., Salt Lake City, UT) was the first iontophoretic system to be approved by the Food and Drug Administration in the late 1970s as a physical medicine therapeutic device. In order to enhance patient compliance, the use of patient-friendly, portable and efficient iontophoretic systems have been under intense development over the years. Such improved systems include the Vyteris (Figure 3) and E-Trans iontophoretic devices. Previous work has also reported that the combined use of iontophoresis and electroporation is much more effective than either technique used alone in the delivery of molecules across the skin. The limitations of iontophoretic systems include the regulatory limits on the amount of current that can be used in humans (currently set at 0.5 mA/cm^2) and the irreversible damage such currents could do to the barrier properties of the skin. In addition, iontophoresis has failed to significantly improve the transdermal delivery of macromolecules of greater than 7,000 Da.[30]

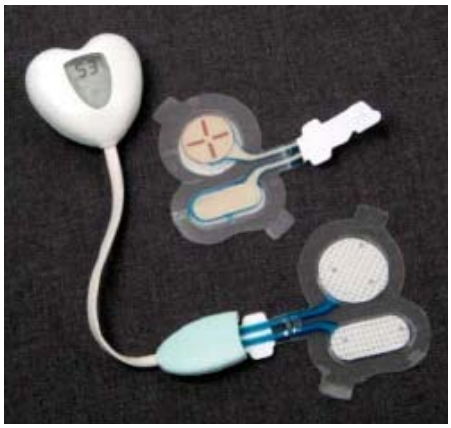

Figure 3: Vyteris iontophoretic device

3. Ultrasound or sonophoresis

Ultrasound involves the use of ultrasonic energy to enhance the transdermal delivery of solutes either simultaneously or through pretreatment, and is frequently referred to as sonophoresis. The proposed mechanism behind the increase in skin permeability is attributed to the formation of gaseous cavities within the intercellular lipids on exposure to ultrasound, resulting in disruption of the stratum corneum. Ultrasound parameters such as treatment duration, intensity and frequency are all known to affect percutaneous absorption, with the latter being the most important. Although frequencies between 20 kHz to 16 MHz have been reported to enhance skin permeation, frequencies at the lower end of this range (<100 kHz) are believed to have a more significant effect on transdermal drug delivery, with the delivery of macromolecules of molecular weight up to 48 kDa being reported. Figure 4 showing sonophoretic device.[31]

Figure 4: Sonophoretic device

4. Laser radiation and photomechanical waves

Lasers have been used in clinical therapies for decades, and therefore their effects on biological membranes are well documented. Lasers are frequently used for the treatment of dermatological conditions such as acne and to confer facial rejuvenation, where the laser radiation destroys the target cells over a short frame of time (~300 ns). Such direct and controlled exposure of the skin to laser radiation results in ablation of the stratum corneum

without significant damage to the underlying epidermis. Removal of the stratum corneum by this method has been shown to enhance the delivery of lipophilic and hydrophilic drugs. The extent of barrier disruption by laser radiation is known to be controlled by parameters such wavelength, pulse length, pulse energy, pulse number and pulse repetition rate. Pressure waves which can be generated by intense laser radiation, without incurring direct ablative effects on the skin, have also been recently found to increase the permeability of the skin. It is thought that pressure waves form a continuous or hydrophilic pathway across the skin due to expansion of the lacunae domains in the stratum corneum. Important parameters affecting delivery such as peak pressure, rise time and duration have been demonstrated. The use of pressure waves may also serve as a means of avoiding problems associated with direct laser radiation. Permeants that have been successfully delivered in vivo include insulin, 40 kDa dextran, and 20 nm latex particles. A design concept for a transdermal drug delivery patch based on the use of pressure waves has been proposed by Doukas and Kollias.[32, 33]

5. Magnetophoresis

This method involves the application of a magnetic field which acts as an external driving force to enhance the diffusion of a diamagnetic solute across the skin. Skin exposure to a magnetic field might also induce structural alterations that could contribute to an increase in permeability. In vitro studies showed a magnetically induced enhancement in benzoic acid flux, which was observed to increase with the strength of the applied magnetic field. Other in vitro studies using a magnet attached to transdermal patches containing terbutaline sulphate demonstrated an enhancement in permeant flux which was comparable with that attained when 4% isopropyl myristate was used as a chemical enhancer.[34]

6. Thermophoresis

The skin surface temperature is usually maintained at 32°C in humans by a range of homeostatic controls. The effect of elevated temperature (nonphysiological) on percutaneous absorption was initially reported. Recently, there has been a surge in the interest of using thermoregulation as a means of improving the delivery profile of topical medicaments. Previous in vitro studies have demonstrated a 2- to 3-fold increase in flux for every 7 to 8°C rise in skin surface temperature. The increased permeation following heat treatment has been attributed to an increase in drug diffusivity in the vehicle and in the skin because of increased lipid fluidity. Vasodilation of the subcutaneous blood vessels as a homeostatic response to a rise in skin temperature also plays an important role in enhancing the transdermal delivery of topically applied compounds. The in vivo delivery of nitroglycerin, testosterone, lidocaine, tetracaine, and fentanyl from transdermal patches with attached heating devices was shown to increase as a result of the elevated temperature at the site of delivery. However, the effect of temperature on the delivery of penetrants greater than 500 Da has not been reported.[35-37]

7. Microneedle-based devices

One of the first patents ever filed for a drug delivery device for the percutaneous administration of drugs was based on this method. The device as described in the patent consists of a drug reservoir and a plurality of projections extending from the reservoir. These microneedles of length 50 to 110 mm will penetrate the stratum corneum and epidermis to deliver the drug from the reservoir. The reservoir may contain drug, solution of drug, gel, or solid particulates, and the various embodiments of the invention include the use of a membrane to separate the drug from the skin and control release of the drug from its reservoir. As a result of the current advancement in microfabrication technology in the past 10 years, cost-effective means of developing devices in this area are now becoming increasingly common.[38, 39]

8. Needleless injection

Needleless injection is reported to involve a pain-free method of administering drugs to the skin. This method therefore avoids the issues of pain and fear associated with the use of hypodermic needles. Transdermal delivery is achieved by firing the liquid or solid particles at supersonic speeds through the outer layers of the skin by using a suitable energy source. Over the years there have been numerous examples of both liquid (Ped-O-Jet®, Ject®, Biojector2000®, Medi-jector® and Interject®) and powder (PMED™ device, formerly known as PowderJect® injector) systems. The latter has been reported to deliver successfully testosterone, lidocaine HCL and macromolecules such as calcitonin and insulin.[40, 41]

9. Radio frequency

Radio frequency involves the exposure of skin to high-frequency alternating current (~100 kHz), resulting in the formation of heat-induced microchannels in the membrane in the same way as when laser radiation is employed. The rate of drug delivery is controlled by the number and depth of the microchannels formed by the device, which is dependent on the properties of the microelectrodes used in the device. The Viaderm device (Transpharma Ltd., Lod, Israel) is a hand-held electronic device consisting of a micro-projection array (100 microelectrodes/cm^2) and a drug patch. The microneedle array is attached to the electronic device and placed in contact with the skin to facilitate the formation of the microchannels. Treatment duration takes less than a second, with a feedback mechanism incorporated within the electronic control providing a signal when the microchannels have been created, so as to ensure reproducibility of action. The drug patch is then placed on the treated area. Experiments in rats have shown that the device enhances the delivery of granisetron HCl, with blood plasma levels recorded after 12 hours raising 30

times the levels recorded for untreated skin after 24 hours.[42]

10. Suction ablation

Formation of a suction blister involves the application of vacuum or negative pressure to remove the epidermis whilst leaving the basal membrane intact. The cellpatch® (Epiport Pain Relief, Sweden) is a commercially available product based on this mechanism. It comprises of a suction cup, epidermatome (to form a blister), and device (which contains morphine solution) to be attached to the skin. This method which avoids dermal invasivity, thereby avoiding pain and bleeding, is also referred to as skin erosion. Such devices have also been shown to induce hyperaemia in the underlying dermis in in vivo studies, [44] which was detected by laser Doppler flowmetry and confirmed by microscopy, and is thought to further contribute to the enhancement of dextran and morphine seen with this method.[43]

11. Skin abrasion

These techniques, many of which are based on techniques employed by dermatologists in the treatment of acne and skin blemishes, involve the direct removal or disruption of the upper layers of the skin to enhance the permeation of topically applied compounds. The delivery potential of skin abrasion techniques is not restricted by the physicochemical properties of the drug, and previous work has illustrated that such methods enhance and control the delivery of a hydrophilic permeant, vitamin C, vaccines and biopharmaceuticals. One current method is performed using a stream of aluminium oxide crystals and motor-driven fraises. Sage and Bock also described a method of pretreating the skin before transdermal drug delivery, which consists of a plurality of microabraders of length 50 to 200 mm. The device is rubbed against the area of interest to abrade the site, in order to enhance delivery or extraction.[44, 45]

12. Carriers and vehicles

12.1 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 multilamellar (lipid/water) structure creates good skin affinity leading to cutaneous penetration and good hydration.

12.2 Nanoemulsions/ Submicron emulsions/ Miniemulsions

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.[46]

12.3 Solid lipid nanoparticles

Solid lipid nanoparticles (SLNs) have recently been investigated as carriers for enhanced skin delivery of sunscreens, vitamins A and E, triptolide and glucocorticoids. It is thought that their enhanced skin penetrating ability is primarily due to an increase in skin hydration caused by the occlusive film formed on the skin surface by the SLN. A 31% increase in skin hydration has been reported following 4 weeks application of SLN-enriched cream.[47]

12.4 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) and thus present good hydration. Their efficacy has been demonstrated in dermatology to treat stretch marks (Triffadiane, CS Dermatologie).[48]

12.5 Microemulsions

These formulations have been shown to be superior for cutaneous delivery compared with 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 pseudoternary diagram phases. They are dynamic systems with continuously fluctuating interfaces. Their good dermal and transdermal delivery properties could be attributed to their excellent solubilising properties. Their high solubilising properties improve biodispensibility, and thus reduce the efficient dose thereby increasing tolerability. Furthermore, their restructuring effect on skin and hair (because of their high lipid content) make microemulsion formulations adapt to altered skin and hair conditions.[49]

13. Drug particulate bombardment

This technique typically uses a high speed jet of helium gas to drive solid drug particles across the SC. Other approaches have used a mechanical hammer-like plunger to exert a driving force, which delivers a liquid drug formulation across the skin. Drug particulate bombardment is suitable for delivering a set amount of drug, that is, finite transdermal dosing rather than the zero-order infinite delivery associated with passive diffusion. The main advantage of this technique is that it overcomes the SC by mechanical force and directly deposits the drug into the viable epidermis. In many ways this technique is similar to a subcutaneous injection, but it has the added advantage of avoiding the use of a

needle, which may have a positive impact upon patient compliance. Disadvantages include lower dosage accuracy and the requirement that the device is operated by a healthcare professional. The lower confidence in dosage accuracy is thought to be because there is variation in inter-individual SC thickness and the fact that the SC can swell or shrink depending on the surrounding relative humidity. Incorrect handling of the device can lead to drug particles being delivered to an unintended location or the SC being damaged and its barrier function being temporarily compromised. These devices are mostly still in the development and optimization stage, however, this technique has shown great potential in delivering DNA-based vaccines to the skin in order to have in situ and a lasting immunological effect.[50]

C. BIOLOGICAL ENHANCEMENT TECHNIQUES

Biological enhancement in transdermal drug delivery can be achieved by disrupting the final cell and tissue differentiation process that occurs in the epidermis. This process is critical to the body's ability to develop an efficient barrier against exogenous substances. One of the key processes is the formation of intercellular lipid within the stratum lucidum (SL) and SC layers. The ordered nature of these intercellular lipids is the primary factor that restricts drug permeation across the skin. These lipids are synthesized in situ in the stratum basale (SB), stratum spinosum (SP), and stratum granulosum (SG), where they can be viewed as lamellar bodies (LB). The lipids present in this LB include sphingomyelins, glucosylceramides, phospholipids, and cholesterol-sulfate. However, the lipids found in the intercellular regions of the SC comprise largely of cholesterol, free fatty acids, and notably, ceramides. Glucosylceramides are converted to ceramides with the aid of B-glucocerebrosidase; the failure to form ceramides from glucosylceramides results in severe barrier abnormality and delayed barrier recovery after acute pertubations. Likewise, the sphingomyelins are also converted by acid sphingomylinase to ceramides (type 2 and 5). The phospholipids are degraded by the phopholipase A enzyme to give free fatty acids, which are also the required key to barrier integrity. Recent studies have demonstrated that the fluid intercellular lipid in the SC consists of only cholesterol and ceramides, where the presence of free fatty acid in the fluid cholesterol and ceramide structure yields the crystalline lipid structure. An interesting observation of the epidermal tissue differentiation process is the conversion of cholesterol to cholesterol sulfate from the SB to the SC. Cholesterol sulfate is also thought to play an important role in SC barrier formation. Many inhibitors have been used to alter these biosynthetic processes with the aim of enhancing TDD. The transdermal flux of lidocaine increased by almost an order of magnitude when hairless mouse skin was given the fatty acid synthesis inhibitor 5-(tetradecyloxy)-2-furancarboxylic acid and the cholesterol synthesis inhibitor fluvastatin.[51, 52]

D. DRUG FORMULATION-BASED ENHANCEMENT APPROACHES

Formulation-based enhancement techniques are often based upon our understanding of Equation 1, which is derived from Fick's first law of diffusion. Adolf Fick modified the equation so that it could be used to model steady state diffusion across a membrane of thickness, h.

$$J = PDC_v / h \quad (1)$$

Where J = flux, C_v = permeant concentration in the formulation, D = permeant diffusion coefficient, and P = permeant partition coefficient between the formulation and the membrane.

It is clear that transdermal drug flux can be improved by increasing the permeant partition co-efficient or the concentration of the drug in the vehicle. There are a variety of means by which these factors can be altered, with the result being an increase in permeation flux. However, the practical application of Equation 1 is limited, as where skin is considered, the length of the diffusion pathway varies depending upon the route across the stratum corneum (SC) that the permeating molecule takes. This is overcome by defining the permeability coefficient (kp), which is given by Equation 2.

$$K_p = DP/h \quad (2)$$

1. pH Regulation

According to the pH partition theory, a permeant, which is weakly acidic or basic, may have a low apparent partition coefficient, which can reduce the likelihood of the drug partitioning into the skin. To circumvent this problem a buffered vehicle can be employed where the drug will be largely unionized and thus have a more favorable apparent partition coefficient. The general rule is: pH < pKa for weakly acidic drugs to be unionized and pKa < pH for weakly basic drugs to be unionized. However, the extremes of pH that may be required to prevent drug ionization could result in skin irritation. For example, lidocaine, which has a pKa value of 8, would require a buffered vehicle of pH 9 to ensure that the drug is 90% unionized. Moreover, there is the possibility that pH extremes may also denature some endogenous enzymes localized in the skin. An alternative approach to alter the pH of the formulation is to introduce an ion pair for a charged drug such as the hydrochloride salt of lidocaine or the sulfate of terbutaline. It has been demonstrated that ion paring mechanisms can be employed to enhance the permeation of ionized drugs, as the ionized drugs interact with the counter ion agents that will then permeate the biological membrane more readily. In one study, the permeation of the drug on dansteron was paired with oleic acid, which is also known to act as a permeation enhancer. In another experiment the researcher investigated the effect of ion pairing on meloxicam, with six organic bases and four normal permeation enhancers, which include oleic acid, menthol, azone, and N-methyl-2-pyrrolidone. The cumulative permeation is markedly increased in the presence of

either a counter ion or chemical enhancers, and the results clearly suggest that the degree of enhancement depends only on the structure and hydrophilicity of the counter ions. However, overall only the limited permeation enhancement ratio is achieved with ion pairing techniques. Further detailed review of ion pair techniques for drug delivery across the biological membrane is available in Neubert R.[53, 54]

2. Supersaturation

Equations 1 and 2 illustrate how the flux of a drug across a membrane is directly proportional to its concentration in the vehicle. This is because drug concentration in the delivery vehicle is directly related to the thermodynamic activity of the drug in the vehicle. Thermodynamic activity is the driving force behind the diffusion of the drug down a concentration gradient. For this reason, researchers often conduct diffusion experiments with the drug in a vehicle concentration at the saturation solubility level, thereby maximizing the thermodynamic activity to 1. This will theoretically elicit the maximum flux and it will also allow direct comparisons between different formulations or chemical enhancement techniques. Consequently, a given increase in the maximum solubility of the drug can result in a higher flux, and this concept is supported by many studies employing supersaturated systems. Supersaturated systems can be obtained by a variety of techniques such as solvent evaporation, back cooling and the addition of a co-solvent. Supersaturation by solvent evaporation can occur when a volatile solvent such as ethanol is used to dissolve a drug in a non-occluded or open system. The solvent evaporates leaving an increasingly concentrated drug solution with a thermodynamic activity in excess of 1. However, because the supersaturated state is inherently unstable, the drug will often crystallize rapidly, thus reducing the thermodynamic activity. This problem can sometimes be prevented or delayed by the use of anti-nucleating agents such as hydroxypropyl-methylcellulose (HPMC) and poly-vinylpyrrolidone (PVP). The addition of PVP to oestradiol solutions is seen to produce an 18-fold increase in drug saturated solubility, due to inhibition of drug crystallization by PVP. The co-solvent approach involves the dissolution of the drug in two solvents, one in which the drug has a high solubility and the second in which the drug has a much lower solubility. The drug is first dissolved in the better solvent to its maximum solubility and this solution is subsequently diluted with the second solvent. The resultant mixture will initially have a higher drug solubility compared to if the drug was directly dissolved in the binary mixture of this composition. Again, the addition of an anti-nucleating agent will prevent or delay the crystallization of the drug. Supersaturated formulations prepared by the co-solvent method with an anti-nucleating agent have been shown to be stable for an excess of two months, whereas, without an anti-nucleating agent the same formulation lasts for less than an hour in the supersaturated state. A potential commercial weakness of this enhancement technique is that increased quantities of drug are required, which may not be cost-effective.[55, 56]

E. PERMEANT MODIFICATION

1. Prodrug Approach

A prodrug can be defined as a compound that usually has little or no pharmacological activity itself, but becomes active after bioconversion in the body. The expression was first termed in 1958, by Albert, and in the intervening half-century, prodrugs have been administered via all possible routes, including transdermally. The main objective of synthesizing a prodrug for transdermal delivery is to create a compound of optimal Log P (1 - 3), which may result in improved transdermal flux. However, the active moiety must be subsequently liberated for the parent moiety to exert its therapeutic effect. A considerable amount of research has been carried out in the area of transdermal prodrugs, and a number of researchers have found that prodrugs can exhibit improved percutaneous flux compared to the parent compound. In a permeation study using cadaver skin, the transdermal flux of piperazinyl derivatives of naproxen was shown to be up to nine-fold that of the parent drug. In another study two alkyl esters of morphine, morphine propionate and morphine enanthate, were synthesized as potential prodrugs for transdermal delivery, and it was found that both these prodrugs showed two-to-five-fold enhancement, respectively, compared to the parent drug morphine. Conversely, the flux of haloperidol across guinea pig skin was found to exceed that of any of its *O*-acyl derivatives. It was postulated that the lower flux exhibited by the prodrugs was due to a combination of poorer aqueous solubility and increased molecular weight. Prodrugs have also been synthesized by coupling drugs to known chemical penetration enhancers. In one study, using rodent skin, it was found that the flux of the prodrug, 4, 5-dihydroisoxazol-3-yl, synthesized from cycloserine and fatty acids, was 20-fold that of the parent drug.[57-59]

2. Melting Point Depression

The effect of the melting point on transdermal flux can be clearly observed when permeation of enantiomers from a racemic mixture is assessed. In one such experiment, the enantiomers with the lowest melting point, from a selection of three chiral beta-blocker molecules, namely, atenolol, alprenolol and propranolol have been shown to exhibit a heightened flux compared to the other two racemic mixtures. This clearly demonstrates that a lower melting point can increase drug flux. However, the choice of the enantiomer is sometimes limited by therapeutic activity. Similarly, a eutectic system can be prepared to induce lowering in the melting point of a drug. The interaction between a drug and a second compound in a eutectic mixture can lower the melting point of the binary mixture to levels that are lower than the melting points of the individual compounds. Eutectic systems have also been developed by selecting a drug and a second compound that is a potent chemical enhancer. One

system consisting of ibuprofen: thymol (40: 60) gave a flux that was 12 times greater than the flux of ibuprofen, from a saturated aqueous solution. Kang also demonstrated similar findings, by developing a eutectic system consisting of lidocaine and 1-menthol. The permeation of lidocaine from this system was significantly higher than its permeation from an aqueous solution in the presence and absence of propylene glycol across a snake skin membrane. The use of eutectic systems to effect melting point depression appear to be a valuable tool in TDD and can be employed in combination with other techniques to enhance transdermal flux.[60, 61]

CONCLUSION

Transdermal drug delivery has enormous potential as a means of delivering drugs that cannot be administered via the oral route. Skin permeation enhancement technology is a rapidly developing field which would significantly increase the number of drugs suitable for transdermal drug delivery, with the result that skin will become one of the major routes of drug administration in the next decade. Research in this area has proved the usefulness of penetration enhancers in the enhancement of drug permeation through skin. The penetration enhancement methods discussed in this review are promising. Focus should be on skin irritation with a view to selecting penetration enhancers which possess optimum enhancement effects with minimal skin irritation.

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