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Historical perspectives

Skin permeation: The years of enlightenment☆

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Abstract

Considerable advances in our understanding of the mechanisms by which drugs permeate the skin barrier have been made over the past 60 years. The key publications, which have influenced the field of skin permeation research are highlighted in the present review. The methodologies commonly employed for estimation of skin permeability are discussed as are the mechanisms proposed for skin permeation. The principal findings from the commonly employed in vitro and in vivo models are considered as well as the applications of enhancers and surfactants for optimization of skin delivery. As these studies presaged the emergence of transdermal drug delivery research in the 1970s, early approaches to model and predict dermal and transdermal absorption are also outlined. The published work on skin permeability in this period embodies the fundamental literature sources for consultation by scientists new to and currently engaged in transdermal drug delivery.

Keywords: Skin absorption; Topical formulation; Enhancers

Contents

☆ *Editor's note:* This paper is one of an occasional series of articles that describe the historical development of a subject. They are written often from a personal perspective and do not aim to be comprehensive reviews, nor do they give detailed historical accounts. The papers in the series have been commissioned by the Editor-in-Chief (Europe) and have been reviewed for their content and coverage. The aim is to give a perspective of past efforts and the way in which these have influenced current research. The Editor-in-Chief welcomes comments on these and indeed all papers in the journal.

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1. Introduction

Topical delivery has been used as a route of medicinal delivery for many thousands of years and there have been considerable advances in our mechanistic understanding of the process in the past 60 years or so. There is a period roughly from the 1940s to 1980, which largely remains unreferenced because of the difficulties in accessing the information using standard databases. This review article considers some of the publications, which we feel have been useful in the development of research in this field. It also demonstrates the range of techniques used and the strategies adopted to understand percutaneous penetration in its widest context. It is surprising how much 'reinvention of the wheel' there has been and how many fundamental facts were known many years ago. In many ways, the 1940s, 1950s, 1960s and 1970s were the formative years for our modern understanding of topical and transdermal delivery.

The review will not consider the historical development of the topic but will separate the major components that have, in our view, been influential. We have chosen to explore the following general areas and clearly some are interrelated and there is some overlap between them. One of the most important areas is that of formulation design, but elements of this are included under most of the different headings outlined below.

- General reviews
- General concepts
- Methodologies
- Mechanisms
- In vitro experiments
- In vivo experiments
- Enhancers
- Transdermal
- Site variation
- Models

It is interesting to see what have been the major drivers in the field, with a general appreciation of topical delivery being important overall. Considerable interest in chemical defence aspects came to the fore in the 1950s and 1960s. The development of synthetic corticosteroids clearly shaped formulation development in the 1960s and early 1970s. After the early 1970s, transdermal delivery started to dominate the area and research interest refocused. More recently, advances in biophysical techniques have contributed significantly to the mechanistic interpretation of data.

2. General reviews and basic concepts

Rothman (1943) produced an excellent review of the state of the art at the time and appreciated the general importance of the physicochemical properties of the permeant. For example, he described the importance of solubility and showed that the vehicle had a significant impact on absorption rate. He also showed the importance of skin lipids. Other representative reviews of skin permeability in the 1950s include (Davies, 1950; Hadgraft and Somers, 1956; Treherne, 1956; Vallette, 1953).

REVIEW ARTICLE

PERCUTANEOUS ABSORPTION

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The application of preparations to the skin for cosmetic and medical purposes is as old as the history of medicine itself and references to the use of ointments, salves and pomades may be found in the early records of Babylonian and Egyptian medicine. In Roman times, drugs were sold by the "ungentarii" who were the compounders of ointments. Galen's formula for cold cream has persisted through the ages and, with some modification, it is still in use to-day.

Although the use of ointments goes back to antiquity, their use has, until recently, been largely empirical. It is only with the advances in dermatology that there has been an appreciation of the requirements of different ointment bases for different skin conditions. Before 1948, with the exception of hydrous ointment, official ointments were made with fatty materials such as soft paraffin, anhydrous wool fat, or beeswax or combinations of these substances. It was hardly appreciated that the therapeutic usefulness of an ointment depends as much on the kind of base used as on the active medicament. To-day, there are available many different synthetic substances allowing the formulation of a wide variety of preparations for application to the skin. The clinician has, therefore, a considerable choice of bases in the prescribing of dermatological preparations and the pharmacist needs to have an expert knowledge of the properties of the different preparations. Both should understand the main principles concerned in percutaneous absorption.

The percutaneous route has been used as a method of drug administration and, although of strictly limited value, may be useful in particular circumstances. With the introduction of toxic synthetic chemicals such as plasticisers in industry, and the use of highly potent insecticides in agriculture, hazards from the toxic effects after percutaneous absorption have become very real ones. The study of percutaneous absorption is of importance also in the elucidation of the normal functioning of the skin.

This review describes the main factors affecting percutaneous absorption, their assessment and application in preparations used in dermatology and drug administration.

Structure and Physical Properties of the Skin

Structure

The skin consists of an outer layer, the epidermis, and an inner layer, the dermis. The epidermis is a horny layer of keratinised epithelial cells, rich in lipoids and cholesterol. The thickness of this layer depends much on the position on the body and is largely determined by the amount

Extract from Hadgraft and Somers (1956).

Reprinted with permission from The Journal of Pharmacy and Pharmacology, 1956, 8, pp. 625-634.

It is interesting to note, at an early time, that it was well documented that molecules with balanced partition behaviour (molecules that possess a log [octanol–water partition coefficient] between 1 and 3) and good solubility in oils and water permeate the skin well. Despite this knowledge, very few molecules have been developed for topical and transdermal use that possesses these preferred physicochemical characteristics. With the more recent commercialisation of transdermal delivery systems, the molecules that are delivered with most ease are nicotine and nitroglycerin which have the physicochemical properties identified in the 1950s as being important. It is surprising that chemical entities that have been specifically designed for topical delivery in recent years are often very lipophilic and insoluble. This clearly creates problems in their delivery, which cannot necessarily be overcome by formulation science.

In the 1960s, there were notable reviews by Blank and Tregear (Blank, 1965, 1969; Blank and Scheuplein, 1964, 1969; Tregear, 1964, 1966). Advances in the general area of skin permeation throughout the 1950s and 1960s occurred as a result of general research in chemical defence and there were efforts to identify the reasons for the barrier properties of the skin and how these properties could be modified in a systematic and predictable way. Mathematical modelling and physicochemical evaluation of the process of percutaneous penetration came to the fore in the 1960s and influential papers by Higuchi (1960,1961) and Higuchi (1962) were published. The seminal paper by T. Higuchi using very basic and well established, physicochemical principles showed the importance of thermodynamic activity on permeation (Higuchi, 1960). Coldman et al. (1969) followed these concepts and produced topical formulations with volatile solvents. As the solvent evaporated the permeant became supersaturated and had a transient higher thermodynamic activity (or chemical potential). This was the fore-runner of much of the work conducted in the 1990s and more recent years on supersaturation.

Release from semi-solid formulations was shown to follow simple solutions to Fick's second law of diffusion and the well known 'square root of time' equation was applied to show a linear dependence between release and square root of time. Towards the late 1960s, there was interest in the corticosteroids for topical delivery; this followed the synthesis of more potent fluorinated compounds requiring appropriate formulations for optimum delivery. Key researchers in the field included Katz and Poulsen (1971, 1972) and two of the most comprehensive reviews of skin permeation during this time frame were co-authored by these researchers.

Contemporaneously with this work, Scheuplein and Blank published a series of papers considering the mechanisms of percutaneous absorption and the role of physical chemistry in the process (Blank et al., 1967; Scheuplein, 1965, 1967; Scheuplein and Blank, 1973; Scheuplein and Ross, 1974). The corticosteroids were known to act as vasoconstrictors and, as will be shown later, provided considerable insight into formulation effects. Researchers such as McKenzie and Stoughton (McKenzie, 1962, 1966; McKenzie and Stoughton, 1962) were instrumental in showing the importance of the blanching effect which remains, today, a powerful tool in determining the pharmacodynamics of topical steroid therapy (Stoughton, 1969).

3. Methodologies

Most of the recent advances in our understanding of topical and transdermal delivery have resulted from application of biophysical techniques that are becoming ever sensitive and sophisticated. However, it is evident that some of the techniques had been considered over 30 years ago. For example, Puttnam (1972) used attenuated total reflectance infra red (ATR) spectroscopy to examine skin in vivo. Infra Red was also used to trace actives and vehicle components on the skin surface (Fischmeister et al., 1975). Skin conductivity, the fore-runner of impedance measurements, was described in the early 1970s (Woolley-Hart, 1972a,b). The application of nuclear magnetic resonance (NMR) to probe diffusion in stratum corneum and to identify various forms of water was reported in the late 1970s (Foreman, 1976; Foreman et al., 1979; Packer and Sellwood, 1978a,b). At the same time, transmission electron microscopy (TEM) was used to investigate the ultra-structural details of the skin (Brody, 1977). Substantial research has been conducted on tape stripping to evaluate bioavailability of topical products but in 1971, Marks and Dawber (1971) described a skin surface biopsy technique using cyanoacrylate adhesives which can be used to estimate agents that have permeated into or accumulated in the stratum corneum. Other

methods that were used routinely will be mentioned in the sections on in vitro and in vivo evaluations.

4. Mechanisms

Routes of permeation have been debated for many years and the precise mechanism of transfer across the stratum corneum has not been unequivocally determined. There is a weight of evidence that suggests that the intercellular route is important and in 1975 Elias recognized that 'intercellular regions of the stratum corneum comprise an expanded, structurally complex, presumably lipid-rich region which may play an important role in percutaneous transport' (Elias and Friend, 1975). The structure of the stratum corneum was examined by Michaels et al. (1975) and likened to a brick and mortar wall. Follicular transport has also had its share of proponents as discussed by Tregear (1961) and Wahlberg (1968). Scheuplein postulated that there was a shunt mechanism at short times during the diffusion process, which was overtaken at longer times by general partition and diffusion through the stratum corneum (Scheuplein, 1967). If a route through the intercellular channels is significant then stacking of the corneocytes is important as this will dictate the path length for diffusion (Christophers et al., 1974).

The outer layer of the skin has been recognized for some time as the rate controlling membrane that acts predominantly as a lipophilic barrier. Therefore, partitioning is a dominant physicochemical determinant in controlling absorption. The importance of partition coefficients and the whole area of structure activity relationships (SAR) have been described for weak electrolytes (Clendenning and Stoughton, 1962) and the steroids (Katz and Shaikh, 1965). One of the first reviews of the available data on SAR and skin permeation was published by Lien and Tong (1973).

In any mechanistic study, it is instructive to determine how rates are influenced by temperature. This can lead to a thermodynamic evaluation of the process and energies of activation may be determined. This is an area that has perhaps not received as much attention as anticipated, but there are some early studies that have probed temperature effects (Allenby et al., 1969; Blank et al., 1967). Some of the problems associated with this type of study are a change in permeation mechanism with temperature as well as possible phase transition changes, which will compromise analyses based on simple Arrhenius approaches.

With the interest in the clinical applications of steroids applied topically a number of studies considered the reservoir function of the skin (Vickers, 1963, 1972) and it was found that the applied steroid could persist in the skin for a significant period of time (days). This has implications for dosage regimens of steroids and the reasons underlying this reservoir function of the skin remain to be elucidated.

5. In vitro experiments

There has been a plethora of different cells to determine percutaneous absorption in vitro and it is important to select a design where the transport is limited by the skin and not in any stagnant diffusion (non-stirred) layers adjacent to the skin surface. The effect of perfusion rate in the receptor medium has been shown to influence the results particularly if the permeant is very lipophilic in nature (Crutcher and Maibach, 1969). Most static cell designs are based on the publications by Franz in the mid 1970s (Franz, 1975, 1978). These were also used in the early tests on in vitro–in vivo comparisons.

It is also interesting to note that information can be obtained using membranes other than the skin but the results have to be treated with caution. Various model membranes have been used including cellulose acetate (Barry and El Eini, 1976) and zeolites (Dyer et al., 1979).

Clearly most of the information regarding the mechanisms of penetration has been gained from in vitro permeation studies and from the 1940s to the 1980s there is a vast array of data. Over the decades, attention has been paid to the penetration of water and it is interesting that this small polar molecule has a profound influence on the barrier properties of the skin. It is an enhancer, as hydrated skin is, in general, more permeable than non-hydrated skin but even today the precise mechanism of action is unclear. Several publications examine the penetration of water, examples being Bettley and Grice (1967), Blank (1952, 1953), Burch and Winsor (1946), DeLong et al. (1954), Idson (1973) and Scheuplein and Morgan (1967).

Probably the simplest homologous series studied is the alcohols and again Blank's research group

investigated these in the mid 1960s (Scheuplein and Blank, 1973). Part of the problem in evaluating materials like these is that they can themselves influence the barrier properties of the skin. For example, the neat alcohols can extract the skin lipids and their presence in the skin will influence the solubility properties of the intercellular channels. Long chain alcohols can intercalate into the structured stratum corneum lipids and disrupt their packing. This will be a function of their degree of uptake. The phenols have also been investigated (Roberts and Anderson, 1975; Roberts et al., 1977) and these also are not without their problems as high concentrations of phenol can damage the integrity of the stratum corneum. Probably the largest class of compounds examined have been the steroids which is because of their clinical utility. The majority of the early work was conducted with radiolabelled compounds. Not much of the steroids permeated the skin and analytical methodology as late as the 1960s relied on radiolabelled compounds to achieve adequate sensitivity. There are obvious problems associated with this type of study, for example, it is important to check the integrity of the label during the time course of the experiment. Examples of in vitro studies on steroids include Busse et al. (1969), Chowhan and Pritchard (1975), Foreman et al. (1978), Foreman and Kelly (1976), Scheuplein et al. (1969) and Zesch and Schaefer (1973).

Many compounds that are placed on the skin are ionized but to date, little research has been conducted on the ionization effects. Salicylic acid has been examined in the 1970s (Marcus et al., 1970) together with methotrexate (Wallace et al., 1978). The first drug to be administered transdermally, scopolamine, is also subject to ionization over the pH range encountered on, and through the skin and has been investigated by Chandrasekaran et al. (1976).

6. In vivo experiments

There is some commonality in the compounds examined between the in vitro and in vivo experiments. In vivo work has been conducted on both animals and human subjects and animal comparisons made by Bartek et al., 1972, 1971). In general, rodent skin is more permeable than human skin and probably the best model for human tissue is pig skin. Some work has been conducted on rhesus monkeys and comparisons made with human skin permeation (Wester and Maibach, 1975, 1976).

Early work on methyl salicylate examined the effect of temperature and ethanol on its dermal penetration (Brown, 1934). Further in vivo studies on the salicylates have been published (Cotty et al., 1960; Fritsch and Stoughton, 1963). Histamine was examined in the late 1940s (Shelley and Melton, 1949), eserine (Hadgraft and Somers, 1954), local anaesthetics (Brockemeyer and Guth, 1955) and nicotine (Johnston, 1957).

30 *The Disease of Tobacco Smoking*

for the second time—about 6 or 7 minutes after the application, I estimate—it had reached my elbow.

At this point the nicotine must have reached my general circulation because three things happened simultaneously : (1) I heaved and salivated—3 or 4 times, and would undoubtedly have been sick had not my stomach been empty : I had not eaten for over 3 hours ; (2) All went black before my eyes except for myriad minute flashes of light. I retained my central field of vision, however, and was able to focus. My mind was clear ; (3) and most striking of all, my heart pounded furiously—quickly and so forcibly that I felt it must burst in my chest and there was sharp pain along both sides of my neck from the distension of my carotid arteries. Realizing that this must be acute nicotine poisoning, I hastily applied the cuff of my sphygmomanometer as a tourniquet and raised the pressure to 200 mm. I could think of nothing else and to do so I lay down on the floor (and wondered if this was the end !). At this point my wife came in and, seeing me on the floor, looking like death (she afterwards told me), 'phoned in a panic to my partner to come at once. However, a few moments later, and before he came, I noticed, with infinite relief, that my pulses were slowing and my vision clearing. In a few minutes I sat up feeling little the worse. My arm was uncomfortable, however, so I released the pressure in my sphygmomanometer. Almost immediately I felt faint—as though I were going to die—but this lasted only a few seconds, after which I felt well again.

Next day, as I moved the bottle of nicotine, I received a drip on my middle finger. Precisely the same symptoms occurred, but much less severely. Later on the same thing happened again—the side of my hand

Extract from Johnston (1957).

The account by Lennox Johnston on self-inflicted nicotine absorption is very poignant.

'On one occasion I painted an area 3" by 2" on the flexor aspect of my forearm with nicotine 5%. In about

7 minutes I felt wretched—nauseated, headachy and faint.'

Other compounds include methyl ethyl ketone (Munies and Wurster, 1965; Wurster and Munies, 1965), ephedrine (Beckett et al., 1972) and nitroglycerin (Attia, 1972).

In the 1960s and 1970s, the nicotinates were examined, the esters possessed a range of physicochemical properties and they permeated the skin to create vasodilatation when they reached a triggering level near the dermal vasculature. It was therefore, relatively easy to use them as markers for skin permeation and they were therefore investigated to probe both physicochemical and formulation effects (Albery and Hadgraft, 1979a,b,c; Baker et al., 1969; Cronin and Stoughton, 1962; Hadgraft et al., 1972, 1973; Stoughton et al., 1960).

It was also possible to use a physiological response, that of vasoconstriction, to look at similar effects for the corticosteroids. Release effects from different vehicles were examined together with structure–activity relationships and estimates of bio-equivalence (Busse et al., 1969; Coldman et al., 1971; Feldmann and Maibach, 1965, 1966, 1969; Hellman et al., 1954; Katz and Poulsen, 1972; Malone et al., 1974; Ostrenga et al., 1971; Sarkany and Hadgraft, 1969; Sarkany et al., 1965; Stoughton, 1969). The clinical significance of the vasoconstriction effect was reviewed in 1968 (Hadgraft and Sarkany, 1968). The particle size of suspended steroids can influence vasoconstriction, and hence, their clinical effect, showing that, in some circumstances, dissolution of the particle and diffusion to the skin surface is comparable to diffusion through the skin (Barrett et al., 1965).

7. Enhancers

It has been recognized for many years that the skin is an excellent barrier to the ingress of many compounds and to increase the therapeutic effectiveness of topical medicines permeation enhancers are needed. Materials have been examined in the past and, as mentioned above, the effects of simple structures such as ethanol examined. Real progress on enhancers and their mechanism of action did not get underway until dimethyl sulfoxide (DMSO) was recognized as a good solvent for many compounds and one that accelerated the dermal absorption of a spectrum of permeants (Horita and Weber, 1964; Jacob et al., 1964; Stoughton, 1964, 1965; Coldman et al., 1971; Chandrasekaran et al., 1976).

DMSO is a dipolar aprotic solvent and related solvents were also examined as potential permeation enhancers with dimethyl formamide and dimethyl acetamide being examples (Munro and Stoughton, 1965; Baker et al., 1969). Surfactants can also compromise the barrier properties of the skin and early publications concerning these date from the late 1950s (Blank, 1969; Scheuplein and Ross, 1970; Chowhan and Pritchard, 1978). Water has been considered the 'universal' enhancer as hydrated skin is generally more permeable. Therefore, anything that alters the hydration state of the skin, e.g., urea can be considered as an enhancer (Feldmann and Maibach, 1974). The mechanism of action of a number of accelerants including DMSO and urea have been examined (Allenby et al., 1969). Isopropyl myristate, one of the excipients used in modern transdermal patches, has been discussed as a permeation enhancer in past publications (Hadgraft, 1960).

8. Transdermal drug delivery

Towards the middle and end of the 1970s, there was considerable optimism about the delivery of drugs through the skin for systemic effects and this has resulted in a number of commercial patch delivery systems. The pioneering publications in this field described the delivery of scopolamine for travel sickness and considered physicochemical properties such as partition and ionization (Chandrasekaran et al., 1976; Shaw et al., 1975; Shaw and Urquhart, 1979). One of the perceived advantages of transdermal delivery is the lack of first pass metabolism, but the metabolic activity of the skin itself should not be overlooked, a feature that has been recognized for several decades and was reviewed in the mid 1940s (Calvery et al., 1946). A decade later when steroids were under consideration the dermal metabolism of testosterone was reported (Wotiz et al., 1956). Ando et al. (1977) have also investigated the effects of skin metabolism on the bioavailability of topically applied drugs.

9. Site variation

The permeability of skin varies from site to site. It is interesting to note that there are reports in the distant literature about 'flying ointment', which was used by witches to make them fly. The preparations contained hallucinogens from natural extracts and were formulated into lipophilic bases. The ointments were applied on a broomstick, which was held between the legs, showing a clear but early recognition of formulation effects, site variations in permeability and prescience of transdermal delivery for systemic effects (Burton, 1972). Smith et al. (1961) demonstrated the superior permeability of scrotal skin compared with abdominal skin for a range of permeants. The high permeability of this region has subsequently been used in the transdermal delivery of testosterone.

One of the most comprehensive studies of the regional variation in skin permeability was conducted by Feldmann and Maibach (1967). Using cortisol as the permeant, these researchers demonstrated the high permeability of scrotal skin (Feldmann and Maibach, 1967). The permeation of water is also markedly affected across different areas of the skin (Scheuplein, 1978).

10. Models

Numerous models have been developed to interpret skin permeation and have subsequently been modified and improved to predict dermal and transdermal absorption. The fundamental concepts outlining the role of physical chemistry in the diffusion of drugs across the skin were published by Higuchi (1960). The advent of topical corticosteroids led to further work on the release and uptake of steroids from topical vehicles (Poulsen, 1970). Yotsuyanagi recognized the complexity of skin and described a two-phase model of permeation (Yotsuyanagi and Higuchi, 1972). With the realization that pharmacokinetics could be applied to the different stages in skin delivery, several models were reported to describe permeation and uptake into the systemic circulation (Wallace and Barnett, 1978). The evolution of the field of transdermal delivery necessitated the development of predictive methods for selection of candidate drugs for this route of administration. The work of Michaels et al. (1975) on the possible routes of delivery amenable to therapeutic agents culminated in the analogy of the skin as a "brick wall" composite. This paper also demonstrated the significance of partition behaviour in skin penetration.

11. Conclusions

While this review does not purport to be an exhaustive list of publications over the time span addressed, it represents papers that exemplify different points and ones which form the basis of our current knowledge base. There are topics which come in and out of favour and this is reflected in the areas described above. There is clear need to consider some of the historical work, some of which is very informative. Knowledge of it should result in recognition rather than 'reinvention of the wheel' for future prospects and developments in (trans)dermal drug delivery.

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