CHAPTER 1

INTRODUCTION

As the present investigation is concerned mainly with topical preparations, it appears appropriate to go through the historical developments of ointments and creams.

1.1. History :

Ancient and universal as cosmetic arts and practices are known to be, the scientific development of cosmetic products and treatments for the hygienic care and embellishment of the human body was not initiated until late in the nineteenth century; and the growth of the great industry that comprises them all has been entirely in the twentieth century. The formulations and its application to the skin for cosmetic and medical purposes is as old as the history of medicine itself. Various fats from animal kingdom were used as vehicles for the dermatological preparations in the early Babylonian-Assyrian[1] Era about 3000-5000 B.C. The examination of remains of such preparations found in the excavated tombs of Egyptian Kings support the view that the fats derived from almost every possible source and mixtures of these with therapeutically active and inert materials were used as ointments in olden days.[2] In second century A.D. 150 Claudius Galenes[3], a Greek physician gave a formula for an ointment base containing animal and vegetable fats and oils namely olive oil, rose oil and bees wax. Galen conceived the idea of incorporating water into a molten

mixture of bees wax and olive oil. In the resulting product, the emollient effect of the oil was accelerated, and a pleasant cooling effect was obtained from the evaporation of the water. The process of manufacture was slow and laborious. The product was unstable and subject to rancidity. This original formula with certain modifications, has been incorporated in most of the text books all over the world as cold cream. Honey, wax, gums and resins were also employed. A greaseless ointment was included among the formulae of Papyrus Ebers,[1] this consisted of Hartshorn blended with incense and floor and mixed with sweet ale.

Until the end of 18th century there was little change in the preparation or evaluation of ointments but then the scientific study began to reveal new substances, and this stimulated the development of new ideas. In the beginning, the ingredients offered by the nature were purified and used or partly replaced by synthesis. It was in 1858 that a radical change took place in ointment technology when apothecary Schacht found that glycerin and starch if heated in certain proportions at a particular temperature formed a translucent jelly which he named a Plasma. This plasma was recommended as an ointment and was included in continental pharmacopoeias as Unguentum Glycerini and in English Pharmacopoeia and U.S.P. as Glycerinum Amyli and Glyceritum Amyli respectively. Lard was introduced as the chief

constituent in the first official ointment of U.S.P. The addition of wax or spermaceti was permitted to give it better consistancy. Later, Lard because of its instability has largely been replaced by benzoinated lard, a product possessing a pleasant odour. Paraffins were used by Miller in 1873 and Lanolin in 1885. Since then stearic acid, wool fat and wool alcohols were in use either singly or in combinations as vehicles for topical formulations.

In 1907, Unna, a great Pharmacist of his days introduced a base consisting of 20% water. Numerous combinations of hydrogenated, sulphated, sulphonated oils as well as stearic acid, sodium stearate and glycerin had appeared after 1920. Before 1948, with the exception of hydrous ointments, official ointments were made with the fatty materials such as soft paraffins, anhydrous wool fat or bees wax or combinations of these substances. It was hardly appreciated that the therapeutic usefulness of an ointment depends as much on the kind of the base used as on the active medicament. A brief history of modern developments of ointment bases was reviewed by Zopf[4].

In the past few decades considerable attention has been given to the development and application of hydrophilic ointments usually called as water-washable bases. Bases of this type are often desired as they are nongreasy, pleasant in appearance, non staining and easily washable with water.

For many years ointments were limited by definition and through use to admixtures of fatty substances. But the present concept of this type of preparation is much broader. Today ointments and creams, in addition to such oleaginous admixtures, include preparations of greater efficiency possessing more or less the same general consistancy but different formula and appearance. They may be entirely free from oleaginous substances. In many instances, they are emulsions of fatty or wax like materials with comparatively high proportion of water. These emulsions may be of the either type. Water soluble substances are also widely used for the preparation of ointment bases.

There are many contradictory definitions among the ointments.

According to I.P. ointments are semisolid preparations consisting of medicament or a mixture of medicaments dissolved or dispersed in a suitable base.

According to USP XX the term ointment is understood to mean only a non-aqueous spreadable preparation and according to USP XXI creams refer to a viscous liquid or semisolid emulsions of either the oil-in-water or water-in-oil type. Whereas in German Pharmacopoeia the term ointment is used for all spreadable preparations including creams and aqueous gels.

Much confusion over the usage of a cream and an ointment should be overcome, if a cream is defined as on o/w emulsion and an ointment the opposite[5].

1.2. Relationships between Vehicle and Drug :

The importance of the vehicle for the absorption process has been neglected till recently. One of the few requirements demanded of the vehicle has been that it acts as an inert medium and incorporates the drug in the most homogeneous distribution possible. An ointment base forms a major proportion of an ointment and hence influences the action of medicament which constitutes only a very minor proportion. Recent studies attach more importance to the chemical and physical relationship between the base and medicament incorporated in it than to the penetration properties of the base itself. This idea was initiated by Sauerland[6] in 1912, when he suggested that the influence of the ointment base varies according to the substances which constitute the bases. It was also suggested that the oily bases delay the absorption of fat-soluble substances such as phenols, mustard oil and others. Since bases constitute more than 90 to 99% in the final make up of an ointment, it is natural that their physico-chemical properties have tremendous influence on the rate of diffusion, penetration and absorption of the drug and hence they should be given

full consideration in their selection for various drugs.

There is no single base which possesses all the required characteristics of an ideal base even today, it is often remarked that the pursuit of an ideal ointment base is a pharmaceutical dream and is likely to remain so without becoming a reality. In this connection the observation made by Wales and Reddish[7] in 1929 that no single base is wholly satisfactory as a vehicle for all antiseptics holds true even after four decades.

These days the term ointment has been used widely and covers up various types of applications used externally. Its meaning is also with respect to the active therapeutic agents incorporated as well as the types of bases used in their preparation. However for centuries the term ointment implied emperical application by innuction of medicaments contained in fatty bases and was concerned either with primary treatment of superficial skin disorders or with the attempt to obtain systemic absorption by penetration as high tissue concentration of the drug at the site of infection for a shorter period of time with less amount of drug is definitely advantageous compared with systemic mode of administration of some drugs. This has given a challenge to research pharmacists who have to evolve, modify, alter and suggest such bases which meet the requirement of maximum absorption, minimum or no

irritation, non greasiness, water-washability, compatibility, low or nil microbial contamination and good stability.

Channing[8] has stressed that the ultimate aim of efficient ointment therapy will only be gained by intelligent choice of the therapeutic end: There has been considerable efforts made by pharmaceutical and medical professions to set up specifications for bases for topical application. These listings cover requirements ranging from chemical inertness to the cosmetic niceties. However, one must not loose sight, on the main objective, that the primary and indispensable quality of a vehicle or carrier is that, it allows adequate release, desired penetration and ultimately required absorption of the drug. The lack of these qualities nullifies whatever other values the base may possess.

The commonly used terms release, penetration and absorption need a clear-cut definition in the first instance because very often they have been used without making any distinction. 'Release' refers to the availability of a medicament from a base at the surface. 'Penetration' means the entry of the drug inside the skin, whereas 'absorption' means drug concentration in the blood and is ready for the systemic distribution. The term penetration and absorption apply to both base and medicament or individually for each. If the release of drug is more the penetration and ultimately

absorption may be more.

Effective topical medication requires an uninterrupted release of the medicament and for this purpose a knowledge of the factors responsible for such release is required. Each drug has peculiar properties of its own and they become more pronounced when more than one drug is present in a formulation. Also the components of the base may offer similar considerations to be taken into account. Thus the problem becomes more complex than it appears at the first thought. Even today, many formulations are based on empirical experience. They are composed of a broad range of compounds with such different chemical structures as antioxidants, buffer systems, emollients, emulsifiers, matrix-builders, moisturisers, preservatives, solvents, thickners and other additives whose precise function is difficult to determine. The result has been a scarcely comprehensible. Profusion of interactions within the vehicle and at the same time, a physico-chemical environment for the drug with an almost immeasurable distribution in the various phases on the vehicle. However, the drugs distribution in the vehicle is of supreme importance for its release. This inevitably leads to a requirement for combination products on the least possible complexity, since the desired release of active ingredient can only be attained via the determination of the inner structure

on the vehicle. The selection of the base to use in the formulation of the creams depends upon the careful assessment of a number of factors.

(a) The desired release rate of particular drug substance from the base.

(b) The desirability for enhancement by the base on the percutaneous absorption of the drug.

(c) The advisability of occlusion of moisture from the skin by the base.

(d) Minimum or no irritation due to base on the skin.

(e) Absence of sensitization.

(f) The short term and long term stability and compatibility of the drug in the base.

(g) Cheap and made from freely available ingredients.

(h) Low or no microbial contamination.

(i) Extrusion from the tube and spreadability.

(j) Shine and cosmetic appeal.

Although the topical vehicles themselves may not penetrate the skin to any extent nor actually carry the medicament through the epidermal barrier, it is known that the therapeutic effectiveness of the drug may vary when it is incorporated in different vehicles. The choice of a right vehicle for a particular medicament depends on physical and chemical properties of the medicament alone

in the vehicle as well as on the nature and condition of the skin being treated.

A good deal of information has been collected during the last few decades while much more is still needed in order to treat effectively the various skin disorders encountered. The potential exists for developing vehicles that will enable the drug to reach the site of action rapidly and maintain a sufficient concentration at the site for the required length of time. The proper formulation of dermatological preparations depends on a thorough understanding of percutaneous absorption.

1.3. Routes of Percutaneous Absorption :

When the therapeutic target lies beneath the stratum corneum, topical therapy becomes quite complicated. Regardless of whether the involved tissue is to be the viable epidermis and upper dermis or whether a systemic response is sought, delivery of the drug to the site of action in sufficient amounts is often a constraining problem. There are many potential topically useful drugs which do not find their place in topical therapy due to an inability to adequately penetrate the skin. The phenomenon of diffusive penetration of the skin by drugs and chemicals is known as percutaneous absorption. There

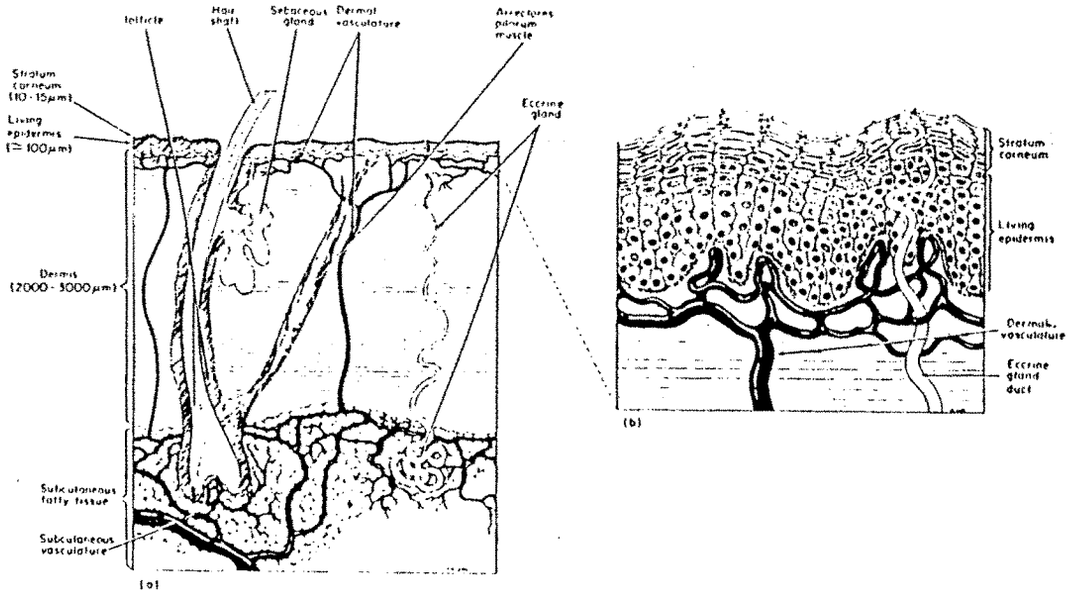
are a number of pathophysiological states which can be treated by concentrating drugs in the surface tissues by percutaneous absorption. Most dermatoses result in inflammed surface tissues, and topical steroidal and nonsteroidal anti-inflammatory drugs are effective in controlling this symptom.

The process of percutaneous absorption can be described as follows. When a drug system is applied topically, the drug diffuses passively out of its carrier or vehicle and into the surface tissues of the skin, specifically and most importantly the stratum corneum and the sebum-filled pilosebaceous gland ducts. A net mass movement continues through the full thickness of the stratum corneum and ducts and into the viable epidermal and dermal strata. A concentration gradient is thus established across the skin that essentially terminates at the outer reaches of the skin's microcirculation in the dermal layer. The systemic circulation acts as a reservoir or "sink" for the drug, and a near - zero concentration of the drug is maintained at the plane where the drug reaches the capillaries and is diluted into the general system.

Once the drug comes in the general circulation it is diluted and distributed very rapidly and, given reasonable rates of systemic metabolism and elimination, there is

generally no appreciable systemic buildup. Thus relatively high local epidermal concentrations of some drugs may be obtained by reason of the fact that the epidermis is without a direct blood supply and the concentration gradient from the outer surface to the micro-circulation cuts directly through the epidermal stratum. However, if massive areas of the body are covered with the topical therapeutic system (> 10% of the total body surface may be used as a crude rule of thumb), then the amounts accumulating systemically can be significant. For instance, corticosteroids have produced serious systemic toxicities on occasion when they have been applied over large fractional areas of the body. Also, if the skin is damaged so that the stratum corneum is not functionally intact, many chemicals can gain systemic entrance at a considerably faster rate and the potential again exists for systemic involvements.

FIG 1 : Schematic Cross Sections of the Skin.



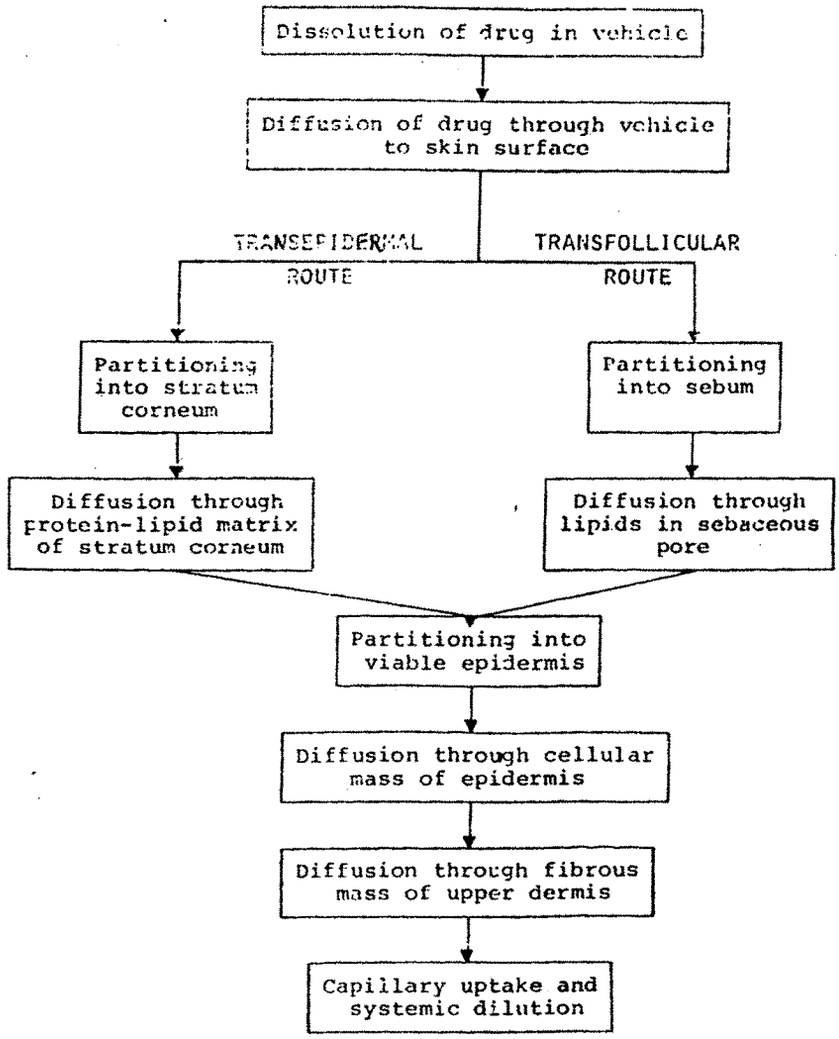


FIG 2 : Schema of Events for Percutaneous Absorption.

A flow diagram of the steps or events requisite to percutaneous absorption following application of a drug in a thin vehicle film is shown in Fig. 2.

The important kinetic processes of dissolution and diffusion within the vehicle layer have been added to processes already described to complete the drug delivery picture. Each step in the diagram is potentially rate limiting, depending on the drug and how it interacts with the vehicle and the skin[9].

Some concept of the complexity of percutaneous absorption may be gained by considering Fig. 2. This diagram represents a simple idealisation of the drug flux which may arise clinically following the common treatment in which we apply a drug to the skin as a solid suspension in a topical vehicle only permeation into the body is considered, and not back-diffusion.

The medicament may undergo any or all of the following events. The drug particles must first dissolve so that molecules may diffuse within the vehicle to reach the vehicle-stratum corneum interface. Interfacial effects are not usually considered important, but for the drug to move through the skin it must partition into the stratum corneum and diffuse within this very impermeable barrier. Some drug may bind at a so-called depot site;

the remainder diffuses in the horny layer, meets a second interfacial barrier, and partitions into the viable epidermis. Whereas the initial partition process may have favoured and increased flux (for example, when a lipophilic drug is released to the skin from an aqueous vehicle), the second partitioning will be unfavourable as the viable epidermis provides a more hydrophilic Milieu compared with the stratum corneum. Any substance with a high affinity for the horny layer and a very low water solubility may not be absorbed percutaneously even though it may have penetrated the barrier layer, particularly when it is applied in low concentrations. The thermodynamic activity of the diffusant in the viable epidermis immediately below the barrier may approach that in the vehicle and in the top layer of the stratum corneum. The rate determining step will not now be the penetration of the barrier but rather the clearance rate from the barrier. Metabolism may alter diffusion in the epidermis. The epidermis-to-dermis partition coefficient may usually be assumed to be close to 1 and may be neglected, as both tissues contain much water. Within the dermis, additional depot regions and metabolic sites may intervene in the progress of the drug to a blood capillary, its partitioning into the capillary wall, thence out into the blood, and its subsequent removal by the systemic circulation.

Very little is known about equilibration in the subepidermal environment and the pharmacokinetic factors which operate there. A fraction of the diffusant may partition into the subcutaneous fat to form a further depot.

Although the aforementioned sequence is already too complex for a full theoretical analysis together with a practical investigation, the situation is further complicated. Such factors may be important as the nonhomogeneity of the various tissues; the presence of hair folicles, sweat glands, interstitial fluid, and lymphatics; and the division of cells in the basal layer, their transport through the stratum corneum, and their surface loss. In addition, drugs permeate the skin under dynamic conditions. Thus, the drug, the components of the vehicle, and the disease may progressively modify the skin barrier, as may the healing process. As components of the vehicle may diffuse into the skin, so physiological materials, including sweat, sebum, and cellular debris, may pass into the formulated product and change its physicochemical characteristics. Emulsions may invert or crack when rubbed into the skin, and volatile solvents may evaporate into the atmosphere[10]. Rothman[11] defines that percutaneous absorption is penetration of substances from outside the skin to the inside and them to the blood

stream. Blank[12] suggests that it is of great importance to know how molecules move into the skin from a unit area of a cutaneous surface in unit time and where these molecules go after leaving the skin surface.

Despite the voluminous literature available on this topic the basic mechanism which is largely responsible for absorption is not fully understood. One group of investigators is of the opinion that major route of entry for drugs is directly through the intact epidermis (transepidermal route) while the other group believes that the absorption is mainly through appendages either the pilosebaceous apparatus or spiral sweat glands. There is evidence that all these avenues take part in transfer of substances through the skin.

1.3.a. Transepidermal absorption :

Mailli[13] states that the pathway through the epidermis is much more likely to be the main avenue of penetration for substances than are the sebaceous glands or sweat glands, simply because the epidermis presents a surface area 100 to 1000 times greater than the other two. Various anatomical zones of the skin behave differently with regards to transepidermal absorption[14]. The film on the surface of skin is composed of sebum, sweat and horny layer and has a complex chemical

composition. This surface film is discontinuous and offers relatively little resistance to a penetrating molecule. The horny layer which is 20 to 40 μ in thickness is composed largely of keratin and sulphydril containing proteins which absorb a large amount of water and other polar compounds. It also contains surface lipids which may spread along the channel walls and absorb lipid soluble material. In other words the horny layer may act as a sponge becoming a reservoir for the penetrating agent and maintaining a maximum concentration gradient just above the barrier thus possibly hindering penetration. Blank and Gould[15] using an autoradio-technique have shown that ionic surfactants are bound by horny layer and often do not penetrate beyond the orifice of the hair follicle. The barrier zone was demonstrated by Rein[16] who reported that it was located between horny layer and granular layer underlying it and that it was an electronegatively charged barrier which repelled anions and attracted cations and held them for further penetration. Rothman[17] reported that the barrier layer checked the transfer across the skin. Blank[18] showed graphically that a barrier of water transfer in skin, is a layer, a few microns thick at the base of the stratum corneum. He used the plastic tape stripping method of Wolf[19] and

Pincus[20] to remove the horny layer gradually. He found that the diffusion of water through the skin remained low until the base of the horny layer had been stripped away, thereafter the permeability to water raise sharply.

In 1951, Szakall[21] reported that after the horny layer had been stripped off with tape the entire barrier could be removed on the tape in a single stripping subsequently be removed the barrier layer by dipping the tape in petroleum ether. The barrier layer is 10 μ thick and prevents the penetration of molecules having molecular weight greater than 200-300[22], yet the diameter of the pores in intracellular spaces in the barrier is greater than the largest penetrating molecules. Thus the restraining force must be molecular interaction between penetrant and pore contents. If the substance has high electrostatic charge eg. ions, the interaction is so great that no penetration occurs. If the substance has water/lipid partition coefficient of about 1 : 10, it has the highest permeability, the barrier must be then having polar and non polar groups in the pore contents. Blank et al [23] have confirmed that penetration occurad through transepidermal route. A similar observation was made by Scott and Kalz[24] for hydrocortisone.

It appears that the ratio of the solubilities in water and in lipids as originally advocated by Meyer and Overtone is actually important for the absorption of substances through the skin. Rothman[17] has shown that lipid soluble substances penetrate the skin more readily than polar substances; those soluble in both lipid and water penetrate most rapidly of all. Treherne[22] exclusively has reported that the permeability of the skin is directly proportional to ether/water partition coefficient of these drugs.

1.3.b Absorption through appendages :

Hair follicles and sweat gland ducts open on the surface of the skin in the form of visible pores which are believed to be avenues for the passage of medicaments through the skin. It has been stated that the hair follicles are the major avenues for percutaneous absorption.

1.3.b (i) Pilosebaceous apparatus :

There are many workers whose studies have shown the prominant role of the pilosebaceous apparatus in percutaneous absorption. This concept takes into consideration the solubility of the drug in sebum. In the upper portion of the follicular canal the hair shaft

does not adhere to the follicular walls and therefore a space is formed which is with horny scale and hair. The interspace is continuous with the duct of the sebaceous glands. The sebum from this duct eventually empties into the interspace[25]. Therefore, any medicament possessing solubility in sebum may penetrate this space and reach inside of the sebaceous glands whose membrane is more permeable than epidermal barrier. Similarly the wall of follicular sheath is less resistant to penetration than surface epidermis. From the sebaceous glands and hair follicles, medicament may penetrate downward into the corneum and from there into the blood thus bypassing the barrier. Medicaments may also move upwards from the sebaceous glands into the epidermis but without penetrating the barrier layer. This pathway of absorption was demonstrated by Mackee[26] and others.

1.3.b (2) Sweat ducts :

There is much controversy as to whether the sweat ducts serve as an avenue for percutaneous absorption. Rein[16] reported that he observed the staining of the pores of sweat duct like those of follicles, when he introduced dyes into the human skin by electrophoresis. Ichihashi[27] postulated that this pore pattern indicated absorption of the dye through the sweat duct. Abramson[28]

confirmed the above result and found the development of the sweat pore pattern dependent on electrical charges of the skin. However, many authors are septical of this[29].

1.4 Factors Influencing Percutaneous Absorption :

There are many factors which influence the rate of percutaneous absorption.

1.4.a Skin condition :

It has been realised for many years that the horny layer (stratum corneum) is the main barrier against penetration and permeation of substances of various origin, including drugs such as those in dermatological and cosmetic use.

The definition of intact and damaged skin in regard to the problems of absorption is therefore inextricably linked with the structure and condition of the horny layer and these are closely dependent not only on exogenous factors such as sweat and sebum production and the microcirculation of the skin itself. If the barrier is destroyed by trauma as in cuts, chapping, reptured blisters of eczema, all substances pass freely into the dermis[30].

Baker et al.[31] had demonstrated that the disintegration of the horny layer caused by Dimethyl Sulfoxide or Salicylic Acid is due to reduced adhesion among the horny cells, whereas that seen after high concentrations of urea is due to the splitting of keratin caused by the incorporation of urea of hydrogen receptors.

A somewhat surprising result was obtained by Sinha et al.[32]. They applied diflorasone diacetate cream to rats at depilated sites and at depilated sites which had been abrated to produce hyperemia. No marked differences in the excretion of the topical steroid were observed between rats with unabraded and with abraded skin. In the monkey, a significant result was the prolonged retention of the steroid or its metabolites in the superficial cell layers of the epidermis of animals with abraded skin. The investigators concluded that such retention is related to the skin damage.

Blank et al.[33] had demonstrated that there was a striking increase in penetration of sarrin following a superficial scratch extending just barely through the barrier. Similar data were obtained from trauma by adhesive tape stripping and puncturing wound.

Some workers[34,35], observed upto 90% penetration of applied dose of radioactive hydrocortisone through stripped skin, while the intact epidermis absorbs only 2% or less.

The *in vitro* percutaneous absorption of topically applied hydrocortisone increases in the experimental epidermal hyperproliferation of mice deficient is essential fatty acids[36]. A similar increase occurs in abnormal hairless mouse epidermis produced by UV light irradiation, topical Vitamin A acid, or topical 10% acetic acid in acetone[37]. They concluded that a combination of abnormal cell membrane phospholipids and abnormal stratum corneum increase skin permeability.

The evidence for permeability increases which may arise from UV, infra-red (IR), and ionising radiation, as well as mild thermal burns, was reviewed by Malkinson[38], although some of the claims are unconfirmed.

Many solvents markedly alter the permeability of the skin barrier, often opening up the complex, dense structure of stratum corneum.[39,42]

Shelmire[43] pointed out that other vehicles of topical products assume more importance when the stratum corneum was intact and differences in drug penetration attributable to the vehicle were more pronounced on

abraded or deseased skin, there might be a large increase in both the rate and the extent of absorption of drug from vehicle.

Blank and Scheuplein[44] pointed out that a mixture of a non-polar and a polar solvent such as chloroform and methanol removes the lipid fraction of the stratum corneum, forming artificial shunts in the membrane through which molecules pass more easily.

A useful physical technique for assessing water permeability determines the electrical impedance of the skin. Some aliphatic acids, bases, the neutral compounds, including dimethylsulfoxide, dimethylformamide, markedly change the impedance of excised human skin. The corresponding increases in water permeability arise from a combination of mechanisms, including the relaxation of binding forces between skin elements, the dissolution of components, and the hydration and subsequent swelling of the skin to form additional channels for permeation[45].

Malkinson[46] observed that the penetration of testosterone and pyribenzamine was more in psoriatic plaques than at normal skin sites and drug concentrations in diseased skins can be higher than in normal skin sites adjacent to the lesion.

It had been shown experimentally that the percutaneous absorption of a drug coming in contact with the skin was significantly reduced in old age[47]. This might be due in part to the atropic changes on the pilosebaceous apparatus through which such substances have been absorbed.

It is a clinical fact that sensitisation reaction to contactants are less commonly encountered in patients. Cramer[48] described that the presence of cholesterol in skin fats had the effect of permitting the surface of the skin to be readily moistened with water and taking water more easily.

1.4.b. Skin age :

The relation between a patient's age and the permeability of his or her skin to drugs has rarely been investigated. Fetal and infant skin appears more permeable than adult skin[49]. Percutaneous absorption of topical steroids occurs more rapidly in normal or inflammed site on infants than comparable areas in adults, as judged by reports of cushingoid side effects consequent to topical steroid treatment.[46-50] The significant dermal atropic and gross epidermal changes in the elderly delay absorption influence.[52-53] It is usually assumed that the skins of the fetus, the young, and the elderly

are more permeable than adult tissue.[50,53]

Rasmussen[44], in a balanced treatment, reviewed percutaneous absorption in children, particularly for allergens and irritants, boric acid and borates, phenol, salicylic acid, mercury, hexachlorophene, lindane (gamma benzene hexachloride), and topical steroids.

1.4.c Regional Skin Sites :

There are relatively few studies done on the variations in absorption from one side to another. In different normal individuals, there are wider variations in the absorption rate of a given substance through the same skin site[55]. One may predict that variations in cutaneous permeability will depend on the thickness of the stratum corneum, its nature, and to a degree over-emphasized in some publications - the density of skin appendages. Many reports conflict, with variable epidermal cell counts and histochemistry[56,58]. Estimation of horny layer thickness provides values of 8.9, 9.4, 10.9 and 12.9 μm for abdoman, back, thigh and flexor forearm, respectively. On an average, 19 cells each 0.55 μm thick give stratum corneum an overall thickness of 10.4 μm.[60]

The inferior barrier nature of palmar and plantar callus is also indicated in the way in which weakly basic solutions, and even water, will eventually dissolve

it[60] and parathion will penetrate it[58]. These observations bring into question much of the earlier work on skin permeability which attempted to make general observations from experiments performed on horny pads. Regional variations in water permeability are not nearly as large as they would be if the stratum corneum were equal in thickness over all the body. As the thickness of the horny layer increases, so diffusivity increases so as to provide the skin with a relatively uniform steady state permeability.

Turning to materials other than water, Smith et al.[61] found that compunds such as salicylic acid, hydrogen sulfide gas, and Lidocaine base penetrate the scrotum more readily than the abdomen scrotal and postauricular skin are the most permeable to tributyl phosphate[62]. Feldmann and Maibach[63] using radioactive hydrocortisone and analysing its urine excretion, showed that the scrotum absorbs the greatest total amount, with absorption decreasing in the following order : forehead, scalp, back, forearms, palms and plantar surface of the foot arch. Normal valvar skin (labia majora) absorbs hydrocortisone to a greater extent than does forearm skin[64].

Among the most interesting biopharmaceutical applications of such studies is the selection of a

suitable regional site to use as a window for systemic therapy. Because of its relatively high permeability and its ease of access, the Transiderm, Transderm, or Transdermal Therapeutic Systems (TTS) employ the postauricular skin as the site of application to insert drugs percutaneously into the blood stream[65].

Horhota and Fung[66] investigated the percutaneous absorption of nitroglycerin through the shaved abdomen and back of the rat by measuring plasma concentrations after topical administration. Abdominal absorption was significant, whereas adhesive tape had to be used to strip the dorsal site before plasma concentration of nitroglycerin could be detected. Site dependance for absorption of nitroglycerin has been reported in human,[67] but not in the rhesus monkey[68].

1.4.d Species differences :

Human and animals display wide differences in physical characteristics. These physical and structural differences obviously affect the penetration pathways and penetration resistance of skin[69].

Frequently, laboratory animals such as rat, mice and rabbits are used to assess percutaneous absorption, but their skins have more hair follicles than human skin and they lack sweat glands. To apply experimental

samples to such animals, often the hair must first be clipped and the skin shaved. It is often assumed that such shaving damages the stratum corneum and artificially increases penetration. Thus, when vasoconstrictors are applied to the shaved human forearm, the sites immediately blanche; a time delay intervenes for the unshaved limb. However, testosterone penetration through the shaved and unshaved forearm of the rhesus monkey shows no significant difference[70].

Some workers correlated a number of publications in an attempt to obtain a perspective as to how percutaneous absorption in several animal models compares with penetration in humans[68-71].

Wester and Maibach[70] compared percutaneous absorption in the rhesus monkey and in man. In both the species, the order of increasing absorption is hydrocortisone, testosterone, and benzoic acid. Among different species like rat, rabbit, miniature swine, man and rhesus monkey, absorption of testosterone in the rhesus monkey is clossest to that in man. For the guinea pig, absorption of hydrocortisone and benzoic acid is similar to man, but testosterone is absorbed to a greater extent[72].

The percutaneous absorption and disposition of a new topical corticosteroid, diflorasone diacetate, were

studied in the rat, cynomogus monkey, and man after a single cutaneous application of a 0.05% steroid cream and showed wide variation of percutaneous absorption in above species[32].

Benzoic acid, progesterone, and testosterone penetrate less readily into the skin of the maxican hairless dog than into human skin[73].

In some investigations, mouse skin proved to be the most permeable, much more so than human skin. However, Stoughton[75] reported that, in vitro, human and hairless mouse skin behaved similarly towards some steroids.

Stolar et al.[75] used rabbits to study the absorption of sodium salicylate from hydrophilic ointment.

Chowhan and Pritchard[76] concluded that rabbit and rat skin are not comparable with human skin. They also emphasized that the rat is not a good in vivo model for man[77].

Desgroseilliers et al.[78] studied the topical steroid absorption in pigs.

Sweeney et al.[79] studied the effect of dimethylsulfoxide on water passage in hairless mice.

The black skin is harder and more resistant to toxic chemicals, partly because it has greater density and more cell layers than caucasian skin[80]. Fair-skinned people of celtic ancestry (Irish, Welsh, or Scottish) sunburn easily, and their skins are hyperirritable to a variety of toxic chemicals such as Croton oil, kerosene, alkalies and sodium lauryl sulphate, compared with dark complexioned Caucasoids from the Mediterranean region[81].

1.4.e. Circulatory effect :

Theoretically, changes in the peripheral circulation, or blood flow through the dermis, could effect percutaneous absorption. Thus, an increased blood flow could reduce the time for which a penetrant remains in the dermis and also raise the concentration gradient across the skin. For example, the penetration of tributyl phosphate in perfused dog skin preparations depends to some extent on the perfusate flow rate[82].

In clinically hyperemic skin, any consequent increase in absorption almost always arises from a disease process damaging the skin barrier[83].

Topical application of 6-methyl prednisolone to stripped skin sites slows the absorption of (^{14}C)

testosterone applied subsequently[34].

Some workers conclude that if the bioavailability of a topical steroid is low, the blood flow may remain unaltered or may decrease; but if the steroid is applied at high concentrations or under conditions which promote percutaneous absorption, blood flow will increase[84].

1.4.f Skin metabolism :

When the pharmaceutical industry develops a new systemically active drug, a full biopharmaceutical investigation considers in detail the pharmacokinetics of the compound, including its absorption, distribution, metabolism, and excretion. However, in the past, for a topical drug, what happens to it after it penetrates the stratum corneum barrier has received much less attention than the fundamentals of percutaneous process itself. In particular, this has been the situation with respect to the metabolism of drugs by the skin.

Several investigators have studied androgen metabolism in whole human skin and in plucked hair follicles[85-86].

Little is known about the biotransformation of highly potent fluorinated corticosteroids in the skin. However, esterases in the skin of rats and guinea pigs

rapidly hydrolyse diflucortolone valerate, whereas *in vitro* tests with human skin reveal a very slow degradation rate for this steroid[87].

Schaefer[88] stated that there is little skin metabolism of topically applied anti-inflammatory steroids at therapeutic concentrations. Longcope[89] concluded that normal skin contributes to the overall metabolism of estrogens, but the tissue is not the major site of metabolism despite its mass and blood flow. Ando *et al.*[90] proposed an *in vitro* model for determining the simultaneous transport and metabolism of the antiviral agent Vidarabine.

1.4.g Thermodynamic Properties of drug :

It is axiomatic and irrefutable that the rate of percutaneous delivery of any drug substance whose absorption is rate limited by its passage across the stratum corneum is directly proportional to the thermodynamic activity (the escaping tendency) of the diffusible species in the formulation.

Higuchi[40] had reported that the important factor in permeation is distribution coefficient. Rothman[91] had also emphasised the importance of the distribution coefficient of the drug between its vehicle and membrane and between membrane and its recepter solution on permeability.

It depends chiefly on molecular interaction between the membrane and penetrating molecule. A substance may enter the membrane when the partition coefficient (membrane/vehicle) is high. It cannot easily leave the membrane when partition coefficient (recepter solution membrane) is low in complex structure such as skin, where membrane may be nonpolar and receptor tissue fluid polar, substance whose partition coefficient between polar and non polar is 1.00 will have the highest penetration rate. Thus permeability is more if there is some, if not too much, affinity between molecule and membrane. Under these conditions the membrane will attract penetrating molecule but not so strongly that it fails to release it on the other side. It is recognised that vehicles which lower affinity (poor solvent power) will normally produce faster penetration. Wagner[92] has also supported this theory.

1.4.h Effect Of moisture :

There are, infact, two opposite views on the influence of moisture on percutaneous absorption. Based on the concept that skin is relatively impermeable to water, one school assumes that moisture on the outside has little to do with promotion of absorption.[7] On the other hand since it has been long standing

clinical experience that water-tight covering of the skin surface does influence percutaneous absorption, many investigators believe that moisture is the most important factor in promoting the passage of medicament through the skin.

Renshaw[93] and Cullumbine[94] reported that more severe lesions develop on human skin if mustard and nitrogen gases were applied to wetted skin. It was interpreted that, this might be because of moisture which played an important role in promoting skin penetration, and this might be responsible for Sevear Lesion. Laug et al.[95] demonstrated that increased moisture promoted transfollicular absorption.

Leslie Roberts[96] reported that moisture caused maceration of horny layer and provided a condition promoting the retention of substances in contact with the skin. According to Higuchi, the transfer properties of the several layers of the skin were probably strongly influenced by the presence of water. Wurster and Cramer[97] studied the absorption of the three salicylate esters and reported that their absorption was produced by increased moisture conditioning. They concluded that percutaneous absorption involved a diffusional process ie. a spontaneous movement of a substance from high concentration to an area of low concentration in

the tissue fluids.

Behl *et al.*[98,99] studied the influence of hydration on n-alkanol permeation through rat skin and compared the results with hairless and swiss mice. They also studied the influence of stripping and scalding on hydration and alteration of the permeability of hairless mouse skin to water and n-alkanols.

Hydration of the epidermis has been shown to increase the percutaneous absorption of nicotinic acid[100] and salicylic acid[60].

Fritsch and Stoughton[101,102] showed the dual importance of temperature and humidity on the penetration of excised skin.

Further evidence of the importance of hydration can be found in investigations employing occlusive plastic films in steroid therapy. Here the prevention of water loss from the stratum corneum and the subsequent increased water concentration in the skin layer apparently enhance the penetration of the steroids[103-107].

Shelmire[108] had also emphasized the importance of hydration of stratum corneum while discussing the penetration of the skin by a medicament. Hydration might physically alter the skin tissues and also result

in changes both in the diffusion coefficient and activity co-efficient of the penetrating medicaments, thereby increasing passage through the skin.

It follows that ointment containing water available for hydration of the keratin layer, such as o/w emulsion bases are likely to increase percutaneous penetration of certain drugs conversely the bases which do not maintain hydration of skin surface would decrease percutaneous absorption. He also suggested that the mechanism of hydration was to increase the size of the pores.

Higuchi[39] suggested that there is no physical alteration of the tissue due to hydration, but at high water activities there are also changes in both the diffusion coefficient and activity coefficients of the penetrating agent.

One would expect the rate of penetration of water soluble drugs to be faster through hydrated than through normal stratum corneum[109].

The hydrated stratum corneum is one of the almost water impermeable biological membranes found in nature, although it is slightly more efficient before extensive hydration and presumably <u>in vivo</u>[110].

A two phase series model for the permeability behavior of the fully hydrated stratum corneum shared resonable correlation between experimental permeability coefficients and partition coefficients[111].

It has been suggested that steroids such as pregnenolone and estrogens benefit aging skin by hydrating it. Thus, pregnenolone acetate reduces the degree of wrinkling in older skin[112]. Barry and Woodford[113] assessed the vasoconstrictor activity and bioavailability of hydrocortisone in six commercial cream formulations. They used a single application technique in which the creams were applied for only 6h under occlusion. Results for the two preparations which contained Urea-Calmurid HC and Alphaderm - suggested that under such conditions the Urea did not preferentially promote the skin penetration of hydrocortisone. Two possible reasons for this result were the short application time and the possible swamping effect of occlusion which would rapidly saturate all stratum corneum sites with water.

Laden and Spitzer[114] identified the main humectant in the water soluble extract from skin as sodium pyrrolidone carboxilate. Middleton and Roberts[115] found that a cream containing 5% sodium pyrrolidone. Carboxilate increased by some 13% the water holding

capacity of the isolated stratum corneum of the footpads of Guinea pigs. In a consumer trial which assessed skin dryness and flakiness, this cream was more effective than the control product.

Rieger and Deem[116] claimed that humectants in the skin increase the transepidermal water loss *in vitro*, which may be undesirable *in vivo*.

Several reports suggested that a commercial cream containing 10% urea (Calmurid) is valuable in the management of ichthyosis. This disease is characterized by very rough skin which presents a dry, cracked appearance resembling fish scales[117,118].

1.4.1 *Effect of temperature* :

Relatively little attention has been paid to the effects of temperature on percutaneous absorption.

Blank and Scheuplein[109] observed little alteration of the permeability of the barrier from exposure for several hours to temperatures as high as 60°C. However, Allenby *et al*.[119] showed that the stratum corneum undergoes irreversible structural changes when heated above 65°C.

The dual effects of both temperature and humidity were shown clearly by Stoughton and Fritsch[102], who demonstrated considerable increase in the percutaneous

absorption of aspirin and corticosteroids as the temperature rose and especially, as humidity increased.

In vivo work with occlusion[103,104] and clinical studies[62] have correlated the roles of temperature and humidity.

Mckensie and Stoughton[103] showed that the minimum effective concentration of certain steroids was reduced by a factor of 100 when the site of application was occluded.

1.4.j Effect of Vehicle :

It has been thought by majority of workers in this field that the functions of the vehicle in percutaneous absorption is to facilitate contact between the medicament and absorbing cells. Absorption is best from vehicles which spread easily over the skin surface, readily mix with the sebum and ultimately bring the medicament into contact with the absorption cells. It was believed in the past that the primary factor influencing penetration through the skin was vehicle itself. Thus the subdivision of the ointment bases into epidermatic, endodermatic and didermatic type became popular[120].

However, it generally became apparent that the thermodynamic activity and the diffusion coefficient

and many other factors decide the absorption of the medicament than the base itself. It is believed that the vehicle itself is not capable of promoting the absorption of non-absorbable drugs dispersed in it but rather modify the degree of penetration of absorbable drug[121].

Organic solvents such as ether, chloroform, benzene and acetone penetrate the skin with ease and enhance the absorption of drug to such an extent that toxic effects can occur[122]. The results obtained from Patch test[123] and analysis of blood and urine[124] after topical application of ointment vehicles containing salicylic acid indicated difference in absorption from various emulsion type of bases. Strakasch's[125] observations are not in accordance with this.

Bourget[126] and Kimura[127], while studying fatty bases reported that salicylic acid was rapidly absorbed, and the amount absorbed was dependent on the type of vehicle. The greatest absorption occured from lard and lanolin while petroleum was observed to be the least effective. The salicylic acid was absorbed more from o/w type than w/o type of bases[124]. There is no doubt that a continued oil phase would tend to reduce absorption, while a continued aqueous phase would be miscible with the

secretion of the denuded and oozing surfaces of the abraded skin and thereby promote release of medicament[128]. Peck, et al.[129] reported that water miscible emulsion bases were found to be best for pyribenzamine hydrochloride.

Poulsen et al.[130] studied the effect of topical vehicle composition on the in vitro release of fluocinolone acetonide and its acetate ester prepared in Carbopol 934 resin gel, where he used isopropyl myristate as a barrier for release.

Cheng-dev Yu et al.[131] studied the effect of propylene-glycol on subcutaneous absorption of a benzimidazole hydrochloride.

Cole et al.[132] studied the vehicle effects in percutaneous absorption. They studied in vitro the influence of solvent power and microscopic viscosity of vehicle on benzocain release from suspension hydrogels.

Wurster[133] and Higuchi[134] reported that if the drug is only partially soluble in the vehicle, release from the vehicle may be less than maximal, possibly compromising bio-availability.

Burdick et al.[135] studied and reported that commercial products if diluted or modified, adversely affect bioavailability from carefully designed system.

Shahi and Zatz[136] studied the effect of formulation factors on penetration of hydrocortisone through mouse skin.

Asker et al.[137] studied the effect of formulation and processing techniques on release of salicylic acid from ointments. He reported a wide variation in release of salicylic acid in different formulation.

Ostrenga et al.[138] studied the relationship between topical vehicle composition, skin permeability, and clinical efficacy.

Conflicting reports concerning the importance of the vehicle in percutaneous absorption may be related to the fact that many absorption studies are carried out in animals whose skin permeability differs considerably from that of man[139].

1.4.k Effect of pH :

The rate of absorption of acidic or basic drugs is pH dependent. Higuchi[41] demonstrated that the rate of absorption of histamine would be 10 times greater from a base buffered at pH 7.5 than from a base at pH 5.5. Harry[140] had suggested that ointments should be compounded with a pH range of approximately 5.3 to 5.6 to coincide the acid pH of the skin. Bhatia and Barter[141]

have reported that maximum local anaesthetic activity of benzocaine could be obtained when the pH of the ointment base was between 6 and 7. The activity decreased with varied pH, this view was also substantiated by the work of Reques[142].

1.4.1 Effect of concentration of medicament :

The amount of drug percutaneously absorbed per unit surface area per time interval increases as the concentration of the drug in the vehicle is increased. Also, more drug is absorbed per time interval at a constant drug concentration if the drug is applied to a larger surface area. However, with a few compounds, increasing concentrations produce significant decrease in absorption rates as in the case of phenol[143] and hydrogen sulfide gas[144].

Higuchi developed a relationship to indicate that this had a definite effect on release pattern. According to him the drug concentration in the base, the diffusion coefficient of drug molecule and solubility of the drug in the base are the important factors. Skog and Wahlberg[145] showed a definite increase in the absorption of various compounds with increasing concentration in guinea pig.

Bettari *et al*.[146] studied the influence of drug concentration on *in vitro* release of salicylic acid from ointment bases.

Tregear[147] showed that the permeability constant of thioglycollic acid is dependent on concentration and probably on time of contact.

The positive penetrative effects of increased concentrations of the steroids like betamethasone, hydrocortisone and cortisone have been demonstrated.[148-150]

One of the anamolies in the action of dimethyl sulfoxide on skin penetration is low concentrations are virtually without effect, as the concentration is increased, there is rapid enhancement of percutaneous penetration[160]. A direct relationship was obtained between the concentration of dimethylsulfoxide and the rate of penetration of potassium methyl sulfate[151].

1.4.m Effect of surfactants :

Surfactants offer possibilities of improving topical vehicles and promoting a more through diffusion of a medicament from the vehicle, thus influencing therapeutic performance[152,153]. Such substances are superior vehicles because of their ability to lower interfacial tension and promote more diffusion of drugs. They provide

ointments washable with water alone allowing easy removal from the injured tissues. They act as medicated cleansing agents. Dodd, Hartmann and Ward[153] have studied the surface-active agents for their suitability as major components in compounding hydrophilic ointment bases. Sweet[154] has suggested that the mode of action of surfactants is due to their capability to emulsify the sebum and thus increase absorption. Duemling[155] studied wetting agents and found that fats alone penetrated chiefly around the hair follicle to a depth just below the skin; while fats in combinations with wetting agents penetrated to the base of hair follicles rapidly and soon penetrated to a depth just below the skin surface but spread through fatty tissues. Howse et al.[156] also reported that oleagineous bases exhibited less release of drug while emulsion bases indicated a better release. Duemling[155] observed more release of ammoniated mercury from paraffin bases by the addition of wetting agents. This was confirmed by Laug et al.[95] who reported that ointments containing surfactants exhibited more absorption of mercury by skin. The mercury absorption was more from base containing sodium lauryl sulphate. Sharma[157] had also observed that surface-active agents enhanced the capacity of the petroleum type base to release antibiotics. In case of tetracyclines, tweens, polyglycol laurate and glyceryl monosterate had

enchanced release of the antibiotic; the release of hamycin was comparatively better from bases containing anionic surfactants.

Stark et al.[158] while studying the influence of nature of surfactants present in ointment bases, on the release of various substances, reported that ionic and nonionic surfactants, substantially influenced it and the nonionic surfactants increased it to a greater extent. Similarly Vinson and Choman[159] found that the nonionic surfactants had promoted a greater release of iodine than did the ionics and especially cationics but same was not true for mercury. However many investigators are of the opinion that nonionic surfactants have little effect in promoting skin penetration. It has been claimed that nonionic surfactants have less initiating effect.

On the other hand, several anionic surfactants have received a widespread use in pharmaceutical products. A few anionic and cationic surfactants have been studied in formulation of hydrophilic ointment bases. It was reported that sodium lauryl sulphate and dodecyl benzene sulphonate released maximum amount of nickel sulphate. Barker and Dekay[160] had also shown that hydrophilic ointments containing anionic surfactants like sodium lauryl sulphate showed a greater release of medicament than those containing nonionic surfactants. Amongst

anionics, the laurate ions are reported to have the greatest penetration and greater effect on the penetration of other solute. Soaps of different fatty acids have this property of varying degrees. The concentration of surfactants have also been found to have influence on the release of the drug[160,161].

One of the factors that has made the surfactants to influence the release of drug is its affinity for water and when a base containing surfactants comes in contact with the aqueous medium, the surfactants diffuse out of the base into the medium followed by the active drug. Another factor may be that the surfactants bring out emulsification of base and aqueous flow in the area, resulting in a watery ointment which is more miscible with the surroundings and thereby exposing greater surface area of the ointment and allowing effective diffusion[61].

Peteanu[162] studied the release of tetracycline from carbohydrate gels associated with non-ionic surfactants. The influence of Tweens and Spans on the rheological properties of vaseline and on the release of tetracycline hydrochloride was studied. The release of tetracycline was found to be influenced by the presence of surfactants which in turn was HLB dependent.

Dermatitic effect of non-ionic surfactants was studied by Mezei[163] Non-ionic surfactants were applied to the rabbit skin to determine the physiological properties and irritative potential of some selected surfactants. The metabolic measurements indicated a two, three and four fold increase in the oxygen consumption of the inflammed, treated skin sample, depending on the length of the treatment and the type of the agent used.

Mezei[164] also studied the dermatitic effect of non-ionic surfactants which caused changes in phospholipid and deoxyribonucleic acid content of the rabbit epidermis. He concluded that after application of non-ionic surfactants in white petrolatum, there was an increase in the phospherus and deoxyribonucleic acid content of the blood.

Peterson[165] studied the emulsifying effects of some of the non-ionic surfactants on a non-aqueous immiscible system. No relation between HLB values and emulsifying capacity, method of mixing or emulsion type was apparent. The chemical nature of the surfactant appeared to have an effect on the method of mixing and emulsion type. Only stearate ester category of the surfactants induced emulsification when the surfactant was added to the

olive oil. In addition only stearate ester surfactants induced Glycerin-in-oil emulsification. Several non ionic surfactants were found to be important in producing emulsions of glycerin and olive oil. Emulsification was apparent at lower concentration of oil but not at the 50% level.

Gibaldi et al.[166] studied the mechanism of the effect of surfactants on the drug absorption.

Mezei and Ryan[167] studied the effect of surfactants on the epidermal permeability in rabbits. The rate of water desorption of untreated and surfactant treated rabbit skin was investigated by these authors. The compounds applied were Petrolatum USP alone, (Control) and petrolatum containing 10% polysorbate 80 or 10% sorbitan trioleate.

Bradshow[168] worked on the effect of non ionic surfactant on the bactericidal activity of cetylpyridinium chloride. The interaction of cetylpyridinium chloride with a non-ionic surfactant was examined, ie. the effect of surfactant on the biological activity of bactericide was predicted. Determination of the degree of reduction of biological activity showed good agreement with the predicted results over a concentration range of 10-100 ppm of the bactericide.

Withington and Collett[169] studied the transfer of salicylic acid across a cellophane membrane from micellar solution of polysorbate 20 and polysorbate 80.

Kundu et al.[170] studied the effect of surfactants on the release of nitrofurasone from polyethylene glycol base (hydrous).

Shen et al.[171] studied the effect of non-ionic surfactants on the percutaneous absorption of salicylic acid and sodium salicylate in the presence of dimethyl sulfoxide.

Mariaux et al.[172] studied the drug release from a lipophilic ointment base as influenced by chain lenth of added surfactant.

Effect of dimethyl sulfoxide (DMSO) :

Stelzer and Collaizzi[173] studied the influence of DMSO on the percutaneous absorption of salycylic acid and salicylate ointments. The object of research was to determine whether DMSO, would alter the precutaneous absorption pattern of salicylic acid and sodium salicylate when incorporated into hydrophilic petrolatum USP and a polyoxyethylene (20) stearyl ether (PEG) gel system.

Whilworth and Stephenson[174] studied the effect of three liquid additives on the diffusion of atropine from

various ointment bases. It was found that the diffusion was significantly enhanced by the presence of liquids but the effect of these liquids depended on the type of base used.

Scott and Vincent[175] worked on the effect of DMSO on the release of antiseptics from the ointment bases. The effect of different concentrations of DMSO on the release of phenol, hexachlorophene and merbromine from five varieties of Hydrophilic Petrolatum USP and Hydrophilic Ointment USP was investigated.

Ansel et al.[176] studied the effect of DMSO as an antimicrobial agent.

Sneader et al.[177] proposed a possible mechanism for the action of DMSO on percutaneous absorption.

Cramer and Cates[178] studied the effect of DMSO and trimethyl phosphite oxide on percutaneous absorption of corticosteroids in rat.

Mezei and Ryan[167] studied the effect of surfactant on epidermal permeability in rabbits. They demonstrated that non-ionic surfactants which are used in pharmaceutical formulation modify the dissolution or emulsification of active ingredients and change the viscosity of ointments and hence absorption process.

1.4.n. Miscellaneous factors :

Several other factors which may also influence the percutaneous absorption of medicaments are the site of application, the length of time such application remains in contact with the skin, the amount of rubbing and temperature of the skin. Blank et al.[179] and Marzulli[180] showed that excised scrotal skin was more permeable than abdominal skin to salicylic acid, hydrogen sulphite and water vapours. Maikinson[181] showed that the rate of penetration of a drug decreased with time as the tissues became saturated with the drug.

Mackee[182] and Peck[183] showed that, in general, the longer period of induction, the greater is the amount of drug absorbed.

1.5. Classification of Bases :

Ointment bases are classified in many ways. However the following classification has been accepted universally by all pharmacists and dermatologists. According to this classification ointment bases are classified into hydrocarbons, fats and fixed oils, silicones, absorption bases, emulsifying bases, and water-soluble bases.

1.5.a. Hydrocarbon bases :

These usually consists of soft paraffin (vaseline,

petroleum jelly or petrolatum) or a mixture of soft paraffin with hard paraffin to produce a suitable consistancy. Paraffins are used extensively and have been found to be very good so far as protective action is concerned[199]. These bases are neither prone to decompositon nor do they get oxidised. The major advantage of these bases is that they do not get rancid and further their inertness have made them more popular. They are used as occlusive, emollients, protectives or protective coverings for the skin on account of these useful properties, paraffins have still remained official in pharmacopoeias.

On the other hand, the hydrophobic properties of paraffins have rendered them to be somewhat unpopular. An antagonism exists between the oily phase constituted by the paraffins and the water phase which is represented by the aqueous exudation of the skin. It is known that the release of medicament from these bases is very limited. Petrolatum is known to inhibit antiseptic action of drugs incorporated in it.

Woodford and Barry[184] showed that white soft paraffin produced a response in the skin blanching test used to assess topical steroid activity.

Hard paraffin and microcrystalline waxes are chemically similar to white soft paraffin but contain no fluid components.

Schindler[185] reviewed the history of soft paraffin since the first patent relating to this material appeared in 1872.

The properties of the various paraffins vary considerably between different batches and grades of petrolatum, and this complexity can lead to difficulties with quality control.

The effect of diluting petrolatum with liquid paraffin was considered by Barry and Grace[186].

Some workers[187] investigated the change in rheological parameters on dilution of white soft paraffin with oil or wax, lanolin, complex emulsifiers, simple surfactants, emulsified water, and other ingredients.

Hydrocarbon bases may contain ingredients additional to petrolatum; for instance, Paraffin Ointment BP is a blend of white beeswax, hard paraffin, cetostearyl alcohol, and soft paraffin. Ozokerite is a mineral wax consisting mainly of C_{35} to C_{55} saturated hydrocarbons Ceresin is a mixture of ozekerite and paraffin wax (hard paraffin).

The Plastibases provide a series of hydrocarbon vehicles in which the manufacturing process incorporate polyethylene into mineral oil at high temperature,

followed by rapid cooling. Foster et al.[188] suggested that more drug released from above vehicle compared to petrolatum system.

1.5.b Fats and fixed oil bases :

These bases which include lard, vegetable and mineral fats have been in use since Greece-Roman times.

Lard, the purified internal fat of abdomen of the hog, and lard containing vehicles are now only of historical interest since we rarely use them in morderndermatological therapy. Castor oil[189] hydrogenated castor oil, palm Kernel oil[190] hydrogenated palm kernel oil, which can take up some water and phosphated or hydrogenated oils have been used as ointment[191] bases. But all these oils turn rancid and hence cannot be used over long periods of time. Other oils include peanut, sesame, olive, cotton seed, almond, archis, maize and persic oils.

In conclusion it can be said that though the fatty bases played an important role in ointment therapy and enjoyed a reputation in past, they need to be replaced by other bases because of number of drawbacks encountered in their use.

1.5.c Silicones :

Silicones[192] are synthetic polymers and are widely used in similar preparations. They are chemically and

physiologically inert[195]. However they are considered objectionable in the treatment of wounds. Because the aqueous discharge provides a barrier which prevents the medication from reaching the diseased tissues, and at the same time, the greasy bases do not allow the aqueous fluid to come out.

Creams, lotions and ointments containing 10 to 30% of a dimethicone prevents rash and bedsores and protect the skin against the trauma associated with colostomy discharge or incontinence[194].

1.5.d Absorption bases :

The term absorption used in this context does not refer to the absorption of the bases by the skin but to the hydrophilic or water absorbing property possessed by the bases. In general they are anhydrous vehicles composed of hydrocarbon base and a substance that is miscible with the hydrocarbon but also carries polar groups and therefore functions as a w/o emulsifier. Higher fatty alcohols, lanolin and glycol stearate are found to be of considerable interest as constituents of these bases[195]. It is claimed that commercial absorption bases usually contain 5 to 10% of cholesterol in hydrocarbon bases. Due to the presence of cholesterol, lard and petrolatum can be used as absorption bases and

have been found to release more amount of drugs[196,197]. It is reported that the hydrophilic properties of paraffins can be increased by the addition of 1% Cholesterol[198].

Absorption bases such as wool alcohols ointment and simple ointment deposit a greasy film on the skin in a similar manner to that of a hydrocarbon base, but they suppress less the transepidermal water loss[199]. Cetyl alcohol is found to increase the water holding property of petrolatum. Liftschuets[200] discussed these bases and claimed that the absorbing properties of lanolin are due to its content of alcohol. Hydrophilic Petrolatum USP, Wool Alcohol Ointment I.P, are the examples of this category. According to Newcomb[201] some individuals are sensitive to lanolin.

Absorption bases generally have a high index of compatibility towards the majority of medicaments used topically. As a class they are relatively heat-stable and can be used in hydrous or anhydrous conditions. These bases, however, possess the undesirable property of greasiness, but are more readily removable from the skin than the oleagenous bases.

1.5.e Emulsion bases :

These essentially anhydrous bases contain o/w

emulsifying agents which make them miscible with water and so washable or "self-emulsifying".

Fantus[202] pointed out that except the cream ointments and emulsions, other ointments are contraindicated in acute inflammatory conditions due to their 'heating quality'. Mumford[203] while discussing the role of emulsified base in dermatology observed that the application of petrolatum or lanolin or a mixture of these offered an objectionable barrier for the aqueous discharge of the skin. In order to overcome this difficulty, emulsion bases can be used as they are oil-soluble as well as water-soluble. It is observed that antiseptic substances exhibited better results when incorporated in emulsion bases rather then when combined with greasy bases. Another appealing advantage of the emulsified ointment base is that they do not stain th) clothings. Besides being less conspicuous when applied on the skin they can be easily removed by washing with water. These emulsified ointments do not interfere with the physiological processes of the skin and are found to be cosmetically more acceptable to the consumers.

Kuever[204] prepared w/o type of emulsion ointment base for incorporating mercuric nitrate. It was found that this preparation was three times more effective in

emulsion base than the base given in N.F. VI, with same percentage of drug similar observations are reported by other workers[135]. Apparently, the most logical approach to formulating a stable ointment is to use an o/w type of emulsion base which is easy to formulate. Often, the preparation incorporates the emulsifying agents in the form of a wax, a granular material which a formulator can more easily handle and weigh, and which one can also use separately to produce a semisolid emulsion. Depending on the ionic nature of the surface-active portion of the water-soluble emulsifying agent, one can classify emulsifying bases into three types - anionic, cationic, or nonionic.

A recurrent feature of these bases is that they contain a mixture of emulsifiers of the opposite type. The w/o emulsifier is cetostearyl alcohol; it combines with the o/w emulsifier which may be ionic or nonionic. Among the two types of emulsion bases it is reported that o/w type of bases promote percutaneous absorption of drug due to either presence of surface-active agents, which may help to bring the medicament into more intimate contact with the skin or the miscibility of bases with aqueous skin secretions[205].

Such bases produce ointments of cosmetic appearance, easy to apply, simple to remove and hence bring about a

desirable psychological effect on the patient ensuring better compliance resulting in more efficacious therapy.

1.5.f Water-Washable bases :

Water-washable bases consitute a group of so called greaseless ointment bases which contain ingredients either water-soluble or water-washable. There are relatively a few materials that possess the physical property of water solubility and still maintain the characteristics that make them desirable as semisolid vehicles for dermal application of medicinal substances.

Carbowaxes are polyethylene glycol derivatives having a molecular weight from 200 to above thousand. They are water-soluble, non volatile, unctuous compounds and this feature combined with their chemical inertness and ability to form an emollient base made them ideally suited for cutaneous therapy. These compounds neither deterierate on standing nor do they support mould growth. It has been shown that carbowaxes are not more irritating than lanolin and petrolatum when applied to the skin[206]. Smyth et al.[207] have reported that acute oral and dermal toxicity and the irritating power of carbowax are very low. Benzoic acid, salicylic acid and phenol are found to exert a solubilising effect and further to interact with ointment bases containing the high molecular weight

carbowaxes. Although *in vitro* studies have shown that medicaments diffuse readily from carbowax bases[208]. It is also observed that little percutaneous absorption occurs from them. Clark[209] has indicated that sulphathiazole is found to be more diffused from carbowax which is an excellent base for the sulpha drugs on account of its high penetrating power.

Pectin which is a purified carbohydrate obtained from the inner rinds of citrus fruits, has been used as an ingredient of ointment bases. Evans[210] suggested the use of pectin as a vehicle for tannic acid and sulphadiazine preparations.

Carbopol 934 has been reported for use as a vehicle for dermatological preparations. The thickening efficiency of Carbopol 934 can be employed in the preparation of such pharmaceuticals as creams, ointments, lotions, suspensions and emulsions[211]. The toxicity of Carbopol is reported to be negligible.

Glyceryl monostearate has been widely used in cosmetics and ointment bases. The glyceryl monostearates serves as a good emulsifying agent[212]. Sodium carboxymethylcellulose (CMC) a granular substance can be used alone or in combination as a base for ointments. It is commercially available with different degree of substitution and in various viscosity grades[212].

Tragacanth is also used alone or in combination as a base for topical vehicles[213].

Bentonite has been used as an ingredient for topical preparations and has been found to be effective for wide range of drugs[214]. Darlington and Guth[215] reported that pH of bentonite may be adjusted by using buffers and *in vitro* activity of ammoniated mercury was enhanced in these bases. Bentonite bases containing antibacterial drugs demonstrate greater antimicrobial activity than certain other bases.

2. RESEARCH ENVISAGED :

Ointments including creams, are the second largest selling formulation of the pharmaceutical industry. Ointment should be compatible with the skin, stable, permanent, smooth and pliable, non irritating, non sensitising, inert and able readily to release its incorporated medication. Since there is no single ointment base which possess all these characteristics, a huge amount of work has been and is being done on this dosage form.

The consistancy of a topically applied semisolid product plays a very significant role in its performance. There are so many semisolid preparations available in the world as well as in India. Now, India is a big

country where so much fluctuations in the climatic conditions like temperature differences, and humidity changes occur during the year. Even during a day there is lot of fluctuation in the temperature and humidity at different regions of India. Ultimate aim of the formulator should be to give a product to the patient in the same condition at the time of manufacturing. It is common man's experience in India that during the time of use of topical preparations there are so many physical problems associated with the preparations, like pourable liquid instead of semisolid mass coming out from tube, separation of phase observed, sometimes hard mass coming out from tube which is very difficult to spread on the skin, sometimes air along with the preparations coming out, sometimes pungent odour or change in colour observed etc. To overcome these problems systematic rheological and stability studies at different conditions on creams and ointment are required which are helpful for a number of things like:

1) as an aid to the fundamental understanding of the nature of the system.

2) quality control of raw materials, final products and manufacturing processes, such as mixing, pumping, filling etc.

3) to study the effect of changes in the formulation, storage time, temperature etc on a product.

4) to assess a product with regard to its usage; and

5) to assess the capacity to suspend solids or immiscible liquids.

Corticosteroids preparations are widely used in dermatology and during the last quarter of century they have revolutionised the treatment of many inflammatory skin diseases like localized neurodermatitis, pruritus ani, allergic, irritant contact dermatitis, inflammatory phase of xerosis and psoriasis etc. Research departments of pharmacentical companies continue to develop more potent steroids in different bases for topical application and it has been demonstrated that different formulations containing the same drug in the same concentration or different drugs in the same vehicle give wide variations in the cutaneous availability or wide variations in the complete blanching profile. Four drugs selected for the present investigation are Triamcinolone Acetonide, Betamethasone Valerate , Fluocinolone Acetonide and Halcinonide.

In above corticosteroids first three are well established and the last one is relatively newest Incorporation of the above steroids in different vehicles

is required and systematic studies of above formulations like stability of the active ingredients, stability of the adjuvants, visual appearance, colour, odour, viscosity, loss of water, phase distribution, phase seperation, particle size distribution of dispersed phases, pH, texture, feel upon application, particulate contamination etc. should be obviously carried out.

Sometimes ointment causes irritation or allergic reactions. Irritant reactions are more frequent and more important, hence a number of test procedures have been devised to test for irritancy levels, both in animal and in man. Thus before introducing a formulation in the market primary irritation test on bases is to be carried out in rabits.

Now according to new concept of USP for non-sterile topical preparations it being necessary to maintain the potency and integrity of product forms and to protect the health and safety of the consumer, the significance of micro-organisms in non sterile products should be evaluated in terms of the use of the product, the nature of the product, and the potential hazard to the user. According to the USP, preparations should be free from P.aeruginosa and S.aureus.

Micro-organisms could cause disease or product

deterioration. So it is very dangerous to have a pathogenic micro-organism or any micro-organism which is dangerous to the health. Hence a systematic study of total viable count in the preparations and the identification of pathogenic organisms if present in the formulations, is to be carried out and also optimum concentration of suitable preservative can be found out.

3. PLAN OF WORK :

The present study was planned to proceed on the following lines.

1) Formulation of the cream bases suitable for incorporating an adequate concentration of the Glucocorticosteroids.

   Here it was planned to prepare Hydrophilic bases o/w emulsion type bases, w/o type emulsion bases and absorption bases with commonly used ingredients.

2) First, second and third screening evaluation of all the prepared cream bases.

   The aim was to study the properties of the cream bases, such as Grainy, Separation, Satisfactory, Unsatisfactory, pH, Spreadability, Washability, Consistancy, congealing point and water content.

3) Rheological evaluation of promising cream bases.

   Promising cream bases depending on the results of above three screening evaluation shall be subjected to Rheological evaluation.

4) To study the primary skin irritation test of selected cream bases.

5) Stability studies of selected creams and the effect on the physical parameters on storage conditions and time.

6) Total viable count of micro-organisms in selected cream bases.

7) In vitro release studies and studies on liberation of drugs through Sartorius ointment Chamber.

8) In vivo medicament release studies (Topical availability studies).

9) In vitro - In vivo correlation.

10) Based on the results most promising bases for the preparation of medicated creams of the four corticosteroid drugs shall be suggested.

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