Original Article

Journal of Pharmaceutical and Medicinal Chemistry
Volume 3 Number 1, January - June 2017
DOI: http://dx.doi.org/10.21088/jpmc.2395.6615.3117.3

Gellified Emulsion of Voriconazole for Transdermal Drug Delivery

Gandla Kumaraswamy*, G. Ramesh, K. Vijay Prakash***

*Care College of Pharmacy, Oglapur (Vill), Damera (Mdl), Warangal, Telanagana-506006. **Sankar Reddy Institute of Pharmaceutical Sciences, salakalaveedu (post), Besthavaripeta (Mdl) prakasham (Dist), Andhra Pradesh-523334.

Abstract

Transdermal drug delivery has gained a tremendous interest in today´s pharmaceutical formulation design and still research is going on in achieving better product. Transdermal drug delivery system is having a plenty of advantages over other drug delivery system. Voriconazole is a triazole antifungal medication that is generally used to treat serious, invasive fungal infections drug with biological half life of Dose Dependent with oral bioavaibality of 58%. The objective of the present study was to develop gellified emulsion of Voriconazole to decrease the systemic side effects and to create a more pronounced effect with lower doses of the drug. Gellified emulsion (Emulsion in gel) have emerged as one of the most interesting topical drug delivery system as it has dual release control system. Also the stability of emulsion is increased when it is incorporated into gel. The gellified emulsion was developed using polymers like carbopol 940 and HPMC K-100 in various ratios (1:1 and 1:1.5) of gel and emulsion.DSC and IR spectral studies were performed to confirm the compatibility of drug and polymers in the formulations. The prepared gellified emulsion was evaluated for their physical appearance, pH evaluation, spreadability, rheological study, drug content and *in- vitro* permeation studies. *In-vitro* drug permeation studies were carried out using keishary chein cell using egg membrane as the permeation membrane.

The optimized formulations VE-4 and VE-4 showed Highest drug permeation as compared to other formulations and followed peppas with non-fickian as a best fit model in the drug release kinetics.The optimized formulations was subjected to stability studies as per ICH guidelines and concluded as stable formulations.

Keywords: Voriconazole; Emulgel; Carbopol 940; HPMC K-100.

Introduction

Transdermal drug delivery system has been in existence for a long time. In the past, the most commonly applied systems were topically applied lotions, creams and ointments for dermatological disorders. The occurrence of systemic side-effects with some of these formulations is indicative of absorption of the drugs throu gh the skin, which lead to the idea of TDDS. In a broad sense, the term transdermal delivery system includes all topically administered drug formulations intended to deliver the active ingredient into the general circulation. Transdermal therapeutic systems have been designed to provide controlled continuous delivery of drugs via the skin to the systemic circulation.

Transdermal drug delivery system (TDDS) established itself as an integral part of novel drug delivery systems [2].

The novel Transdermal drug delivery is defined as self-contained, discrete dosage forms which when applied to the intact skin, deliver the drug through

Reprint Request: Gandla KumaraSwamy, Associate professor & H.O.D Dept.of pharmaceutical Analysis, Care College of pharmacy Warangal, Telangana-506006.

E-mail: kumaraswamy.gandla@gmail.com

(Received on 31.12.2016**, Accepted on** 09.01.2017**)**

the skin at controlled rate to the systemic circulation 1.

Advantages of Transdermal Drug Delivery Systems

- Transdermal medication delivers a steady infusion of a drug over an extended period of time. Adverse effects or therapeutic failures frequently associated with intermittent dosing can also be avoided.
- Transdermal delivery can increase the therapeutic value of many drugs by avoiding specific problems associated with the drug e.g., gastro-intestinal irritation, low absorption, decomposition due to hepatic 'first- pass' effect, formation of metabolites that cause side effects, short half - life necessitating frequent dosing etc.

Disadvantages of Transdermal Drug Delivery Systems

- The drug must have some desirable physicochemical properties for penetration throuVE stratum corneum and if the drug dose required for therapeutic value is more than 10 mg/ day, the transdermal delivery will be very difficult.
- Only relatively potent drugs are suitable candidates for TDDS because of the natural limits of drug entry imposed by the skin impermeability
- Some patients develop contact dermatitis at the site of application for one or more of the system components, necessitating discontinuation.
- Clinical need is another area that has to be examined carefully before a decision is made to develop a transdermal product.
- The barrier function of the skin changes from one site to another on the same person, from person to person and with age.

Historical Perspective

Transdermal delivery of medications was foreshadowed in earlier era as by the use of certain plasters and ointments. The mustard plaster, applied as a home remedy for severe chest congestion, may be considered an example. Powdered mustard seeds were mixed with warm water, and the resulting paste was spread on a strip of flannel, which was applied to the patient s chest with a cloth binding wrapped around the body to hold the plaster in place. The history of plasters has been traced back to antiquity. In addition to mustard plasters, several other plasters were recognized in early 20th century editions of the United States Pharmacopeia (USP) and National Formulary (NF). At one time, Belladonna Plaster, containing 0.25 – 0.30% of belladonna root alkaloids, was believed to act transdermally as an analgesic. Perhaps the most remarkable forerunner of modern transdermal medication was Strong Mercurial Ointment, used as a treatment for syphilis when Salvarsan and other arsenicals were in use, before the discovery of penicillin. For the first time use of transdermal drug delivery system was done by the USFDA in December 1979, which administered scopolamine for motion sickness.

Skin as a Site for Drug Diffusion

The skin of an average adult body covers around 2 m^2 of surface area and receives approximately one third of all blood circulating throuVE the body. It is one of the extensive and redialy accesible organs on the human body. With a thickness of only a fraction of millimeter, the skin separates the underlying blood circulation network from the outside environment and serves as a barrier against physical, chemical and microbial attacks, acts as a thermostast in maintaining body temperature, plays a role in the regulation of blood pressure and protects against the penetration of ultra violet rays.

The skin is a multilayered organ composed of many histological layers. It is generally described in terms of three major tissue layers: the epidermis, the dermis and the hypodermis. Microscopically, the epidermis is further divided into five anatomical layers with stratum corneum forming the outermost layer of the epidermis.

The stratum corneum consists of many layers of compacted, flattened, dehydrated and keratinised cells. They are dead cells converted to protein and are continuously shed, requiring replacement from the underlying viable epidermal tissues. The stratum corneum has a water content of only ~ 20% as compared to the normal 70% in the physiologically active stratum germinativum (regenerative layer of the epidermis).

An average human skin surface is known to contain 40-70 hair follicles and 200- 250 sweat ducts on each square centimeter of the skin area. These skin appendages, however, occupy grossily, only 0.1% of the total human skin surface. Even though the forein agents, especially the water soluble ones may be able to penetrate into the skin via these skin appendages at a rate which is faster than though the impact area of the stratum corneum. This transappendageal route of percutaneous absorption has, at steady state, a very limited contribution to the

overall kinetic profile of transdermal permeation. Therefore the transdermal permeation of most neutral molecules can, thus, be considered as, process of passive diffusion though the intact stratum corneum in the interfolicular region.

For many decades, the skin has been commonly used as the site for the administration of dermatological drugs to achieve a localised pharmacologic action in the skin tissues. Most recently, there is an increasing recognition that the skin can also serves as port of administration for systematically active drugs. In this case, the drug applied topically will be absorbed first into the blood circulation and then be transported to target tissues, which could be rather remote from the site of drug application, to achieve its therapeutic responses. It is exemplified by controlled administration of nitroglycerin for the treatment of angina pectoris and scopolamine for the prevention of motion sickness [3].

Stratum Corneum: Biological and Biochemical Consideration

In order to fully understand permeation process of the drug though the skin, it is necessary to understand the components and function of the stratum corneum. In this case, components and barrier function of stratum corneum is given below:

Components of the Stratum Corneum

A. Keratin

keratins are a family of α-helical polypeptides ranging from 40000 to 70000 daltons in size. They are relatively poor in cystine, rich in serine and glycine and contain N- acetyl serine at the amino terminus. Keratins accumulate throuVEout epidermal differentiation and represent the major component of stratum corneum as well as of epidermal appendages such as hair, nail and hoof.

The keratin polypeptides seem to be synthesized as pair of relatively acidic and basic polypeptides. Varying degrees of phosphorylation of serine residues may contribute to charge heterogeneity. The individual keratin molecules aggregate to form superhelicles, the detailed structures of which are still under investigation. This aggregation is facilitated by histidine rich protein called filaggrin that is derived from the keratohylin granules.

The filaments found in the stratum corneum are 7-10 nm in diameter and many micron in length. They are stabilized by the formation of disulphide bridges and cannot be solubilised in the absence of reducing agent. The keratin filaments fill interior space of corneocyte. They are probably responsible for maintaining the flat hexagonal shape of the corneocyte, and may contribute to the touVEness and flexibility of the stratum corneum.

B. The Corneocyte Envelope

The cornified cells ofof the stratum corneum are bounded by an envelope produced in the final step of terminal differentiation. In the transmission electron microscopy, this envelope appears as an uniform 12 nm thick electron dense band that has replaced or been added to the electron dense polar region of the inner leaflet of the granular cell plasma membrane. The lucent hydrophobic interior of the plasma membrane and the outer polar region appear to remain intact.

The thickened inner portion of the envelope consists of cross-linked proteins, predominantly involucrin. Involucrin becomes cross-linked through γ-glutamyl-ε-amino lysyl isopeptide bonds introduced by the action of γ-glutamyl transpeptidase.

This enzyme is apparantly activated by an influx of calcium resulting from a change in the permeability of the plasma membrane late in the differentiation prosess. In addition to involucrin, at least six other soluble and membrane associated protein become incorporated into the cross linked protein envelope.

Several of these are specific keratinocyte proteins, where as several others are nonspecifically incorporated into envelope superstructure.

C. Intercellular Lamellae

Intercellular spaces of the stratum cornuem are completely filled with broad, multiple lipid lamellae. These lamellae were first noted by Breathnach and coworkers, who applied freeze-fracture electron microscopy to the skin. In osmium- post fixed thin sections, these lamellae are rarely evident and the intercellular spaces appear empty, but recent use of ruthenium tetroxide as postfixative has permitted routine visualization of the intercellular membranes. Use of this technique has revealed that lamellae are found throughout the stratum corneum and even persist after desquamation. These extracellular membranes in the stratum corneum appear to be produced by edge to edge fusion of the flattened lipid vesicles that are extruded from the lamellar granules. Before extrusion, the stacks of the disks in the lamellar granules appear to have alternating major and minor electron dense bands with electron lucent material in between. Each minor

dense band is thouVEt to represent the apposition of two polar regions on the interior of a flattened bilayer vesicles, where as the major dense bands represent the polar regions between adjacent vesicles [5].

Mechanism of Absorption

Drug Delivery Routes Across Human Skin

- Drug molecules in contact with the skin surface can penetrate by three potential pathways: throuVE the sweat ducts, *via* the hair follicles and sebaceous glands (collectively called the shunt or appendageal route), or directly across the stratum corneum.
- The relative importance of the shunt or appendageal route versus transport across the stratum corneum has been debated by scientists over the years and is further complicated by the lack of a suitable experimental model to permit separation of the three pathways.
- *In vitro* experiments tend to involve the use of hydrated skin or epidermal membranes so that appendages are closed by the swelling associated with hydration. Scheuplein and colleagues proposed that a follicular shunt route was responsible for the presteady-state permeation of polar molecules and flux of large polar molecules or ions that have difficulty in diffusing across the intact stratum corneum.
- However it is generally accepted that as the appendages comprise a fractional area for permeation of approximately 0.1%, their contribution to steady state flux of most drugs is minimal. This assumption has resulted in the majority of skin penetration enhancement techniques being focused on increasing transport across the stratum corneum rather than via the appendages. Exceptions are iontophoretic drug delivery which uses an electrical charge to drive molecules into the skin primarily via the shunt routes as they provide less electrical resistance, and vesicular delivery.
- The stratum corneum consists of 10-15 layers of corneocytes and varies in thickness from approximately 10-15 ìm in the dry state to 40 μm when hydrated. It comprises a multi-layered "brick and mortar" like structure of keratin-rich corneocytes (bricks) in an intercellular matrix (mortar) composed primarily of long chain ceramides, free fatty acids, triglycerides, cholesterol, cholesterol sulfate and sterol/wax esters. However it is important to view this model in the context that the corneocytes are not brick shaped but are polygonal, elongated and flat (0.2-1.5 μm thick, 34-46 μm in diameter).
- The intercellular lipid matrix is generated by keratinocytes in the mid to upper part of the stratum granulosum discharging their lamellar contents into the intercellular space. In the initial layers of the stratum corneum this extruded material rearranges to form broad intercellular lipid lamellae, which then associate into lipid bilayers, with the hydrocarbon chains aligned and polar head groups dissolved in an aqueous layer.
- As a result of the stratum corneum lipid composition, the lipid phase behaviour is different from that of other biological membranes. The hydrocarbon chains are arranged into regions of crystalline, lamellar gel and lamellar liquid crystal phases thereby creating various domains within the lipid bilayers. The presence of intrinsic and extrinsic proteins, such as enzymes, may also affect the lamellar structure of the stratum corneum. Water is an essential component of the stratum corneum, which acts as a plasticizer to prevent cracking of the stratum corneum and is also involved in the generation of natural moisturizing factor (NMF), which helps to maintain suppleness. In order to understand how the physicochemical properties of the diffusing drug and vehicle influence permeation across the stratum corneum and thereby optimise delivery, it is essential to determine the predominant route of drug permeation within the stratum corneum.
- Traditionally it was thought that hydrophilic chemicals diffuse within the aqueous regions near the outer surface of intracellular keratin filaments (intracellular or transcellular route) while lipophilic chemicals diffuse through the lipid matrix between the filaments (intercellular route) [6].

Kinetics of Transdermal Permeation

Knowledge of skin permeation kinetics is vital to the successful development of transdermal therapeutic systems. Transdermal permeation of a drug involves the following steps:

1. Sorption by stratum corneum.
2. Penetration of drug through epidermis.
3. Uptake of the drug by the capillary network in the dermal papillary layer.

This permeation can be possible only if the drug possesses certain physiochemical properties. The rate of permeation across the skin is given by-

$dQ/dt = Ps (Cd - Cr)$

Where Cd and Cr are the concentration of the skin penetrant in the donor compartment i.e. on the surface of stratum corneum and in the receptor compartment i.e. body respectively. Ps is the overall permeability coefficient of the skin tissue to the penetrant. This permeability coefficient is given by the relationship:

$Ps = Dss\ Ks / hs$

Where Ks is the partition coefficient for the interfacial partitioning of the penetrant molecule from a solution medium or a transdermal therapeutic system on to the stratum corneum, Dss is the apparent diffusivity for the steady state diffusion of the penetrant molecule throuVE a thickness of skin tissues and hs is the overall thickness of skin tissues. As Ks, Dss and hs are constant under given conditions the permeability coefficient Ps for a skin penetrant can be considered to be constant. From equation (1) it is clear that a constant rate of drug permeation can be obtained only when Cd >> Cr i.e. the drug concentration at the surface of the stratum corneum Cd is consistently and substantially greater than the drug concentration in the body Cr. The equation becomes:

$dQ/dt = Ps\ Cd$

The rate of skin permeation is constant provided the magnitude of Cd remains fairly constant throughout the course of skin permeation. For keeping Cd constant the drug should be released from the device at a rate Rr i.e. either constant or greater than the rate of skin uptake Ra i.e. Rr>> Ra. Since Rr>> Ra, the drug concentration on the skin surface Cd is maintained at a level equal to or greater than the equilibrium solubility of the drug in the stratum corneum Cs .i.e. Cd>>Cs. Therefore a maximum rate of skin permeation is obtained and is given by the equation:

$(dQ/dt)\ m = PsCs$

From the above equation it can be seen that the maximum rate of skin permeation depends upon the skin permeability coefficient Ps and is equilibrium solubility in the stratum corneum Cs. Thus skin permeation appears to be stratum corneum limited.

Basic Components of Transdermal Drug Delivery

1. Drug
2. Polymer matrix or matrices
3. Permeation enhancers
4. Other excipients

Drug

For successfully developing a transdermal drug delivery system, the drug should be chosen with great care. The following are some of the desirable properties of a drug for transdermal delivery.

Physicochemical Properties

- The drug should have a molecular weiVEt less than approximately 1000 Daltons.
- The drug should have affinity for both lipophilic and hydrophilic phases. Extreme partitioning characteristics are not conducive to successful drug delivery via the skin.
- The drug should have low melting point.
- Along with these properties the drug should be potent, having short half life and be non-irritating.

Polymer Matrix

The polymer controls the release of the drug from the device. Possible useful polymers for transdermal devices are:

Natural Polymers: Cellulose derivatives, Zein, Gelatin, Shellac, Waxes, Proteins, Gums and their derivatives, Natural rubber, Starch etc.

Synthetic Elastomers: Polybutadieine, Hydrin rubber, Polysiloxane, Silicone rubber, Nitrile, Acrylonitrile, Butyl rubber, Styrenebutadieine rubber, Neoprene etc.

Synthetic Polymers: Polyvinyl alcohol, Polyvinyl chloride, Polyethylene, Polypropylene, Polyacrylate, Polyamide, Polyurea, Polyvinyl pyrrolidone, Polymethylmethacrylate, Epoxy etc [7].

Penetration Enhancers

There is great interest among pharmaceutical scientists to develop chemical penetration enhancers and physical methods that can increase percutaneous absorption of therapeutic agents.

Chemical Enhancers: A chemical skin penetration enhancers increases skin permeability by reversibly damaging or altering the physico-chemical nature of the stratum corneum to reduced its diffusional resistance. Among the alterations are increased hydration of stratum corneum, a change in the structure of the lipids and lipoproteins in the intercellular channels through solvent action or denaturation or both. Some drugs have an inherent capacity to permeate the skin without chemical

enhancers. However, when this is not the case, chemical penetration enhancers may render an otherwise impenetrable substance useful in transdermal drug delivery. More than 275 chemical compounds have been cited in the literature as skin penetration enhancers; they include acetone, azone, diethyl acetamide, dimethyl formamide, DMSO, ethanol, oleic acid, polyethylene glycol, propylene glycol and sodium lauryl sulphate. The selection of a penetration enhancer should be based not only on its efficacy in enhancing skin permeation but also on its dermal toxicity and its physicochemical and biologic compatibility with the systems other components. Some of the examples are:

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

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

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

D. Alcohol, Glycol, and Glycerides: Ethanol is the most commonly used alcohol as a transdermal penetration enhancer. It increases the permeation of ketoprofen from a gel- spray formulation and triethanolamine salicylate from a hydrophilic emulsion base. It also acts as a vehicle for menthol in increasing the penetration of methyl paraben.

E. Sulfoxides, Ulfoxides and Similar Compounds: Dimethyl sulfoxide (DMSO), the most important compound belonging to the category of sulfoxides and similar compounds, enhances the transdermal permeation of a variety of drugs, like β-blockers, ephedrine hydrochloride, and papaverine hydrochloride. It also enhances the release of azapropazone from its ointments. Fourier transform Raman spectroscopic studies revealed that DMSO changes the stratum corneum keratin from alpha-helical to β-sheet conformation. At concentrations greater than 60% v/v, at which DMSO enhances the flux, there was evidence of its interaction with stratum corneum lipids. It also produces alteration in protein structure, but may also be related to alterations in stratum corneum organization besides any increased drug-partitioning effect. Decylmethylsulfoxide (DCMS) in combination with ethanol increased the flux of oxymorphone hydrochloride. A 4% aqueous solution of DCMS increased the permeation of 5-FU 35times across human skin, but it was rapidly washed out of the tissues. Dimethylacetamide (DMA) enhances thepermeation of indomethacin from creams andointments in rats. N, N- Dimethyloctanamide and N,Ndimethyldecanamide were found to be effective enhancers of the NSAIDs like ibuprofen and naproxen from 50% aqueous PG vehicles across rat skin.

F. Terpenes, Terpenoids, Essential Oils: Terpenes have been utilized for a number of therapeutic purposes, such as in antispasmodics, carminatives, perfumery, and others, but a few reports also suggest their potential as percutaneous absorption enhancers. The effect of three essential oils (eucalyptus, peppermint, turpentine oil) on the permeation of 5-fluorouracil (5-FU) were studied using excised rat skin. Eucalyptus oil was found to be the most active, causing a 60-fold increase, while peppermint and turpentine oil showed 48- and 28-fold increases, respectively. Mode of action of these enhancers may be due to a combined process of partition and diffusion, the latter being dominant.

G. Fatty Acids and Esters: A large number of fatty acids and their esters have been used as permeation enhancers. A general trend has been seen that unsaturated fatty acids are more effective in enhancing percutaneous absorption of drugs than their saturated counterparts. Addition of oleic acid to an Ethanol: water (50:50) cosolvent system markedly improved the skin permeation of zalcitabine, didanosine, and zidovudine, whereas addition of the same to ethanol: TCP (50:50) produced no enhancement across hairless rat skin. The fatty acid extract of cod liver oil was found to be as good a permeation enhancer as oleic acid [8].

Iontophorosis and Sonophoresis

In addition to chemical means, some physical methods are being used to enhance transdermal drug delivery and penetration namely, iontophoresis and sonophoresis. Iontophoresis is delivery of a charged

chemical compound across the skin membrane using an electric field. A number of drugs have been the subject of iontophoretic studies; they include lidocaine, dexamethasone, amino acids, peptides and insulin. There is particular interest to develop alternative routes for delivery of biologically active peptides. At present these agents are delivered by injection because of their rapid metabolism and poor absorption after oral delivery. They are also poorly absorbed by the transdermal route because of their large molecular size and ionic character. However, iontophoresis has shown some promise as a means of peptides and protein administration. Sonophoresis is also being studied as a means to enhance transdermal drug delivery. Among the agents examined are hydrocortisone, lidocaine and salicylic acid in such formulations as gels, creams and lotions. It is thought that high frequency ultra sound can influence the integrity of the stratum corneum and thus affect its penetrability [9].

Objectives

The main aim of the present research work is:

- Formulation of Glucosamine sulphate emulgel using various emulsifying and gelling agents in different combinations and ratios by suitable methods.
- Evaluation of the transdermal formulations for its physico-chemical properties like visual appearance, viscosity, pH, spredability, uniformity, drug content etc.
- *In vitro* drug release permeation studies throuVE the suitable membrane models using Franz-Diffusion cell.
- Comparative drug release profile and steady-state flux between the optimized emulgel and hydrogel formulation.
- To predict the shelf life of the formulation by conducting stability studies as per the ICH guidelines.

Voriconazole

Voriconazole has become the new standard of care in the treatment of invasiveaspergillosis, which may occur in immunocom promised patients, including allogeneic BMT, hematologic cancers, and solid organ transplants. This is based on the results of a large, randomized study in which voriconazole proved superior to amphotericin B with 53% complete or partial response, compared with 32% for amphotericin B. Importantly, voriconazole also offered a 22% greater survival benefit over amphotericin B, with 71% of voriconazole patients still alive at week 12. Only 13% of patients who received initial therapy with voriconazole died from invasive aspergillosis, compared with 29% of patients who initially received amphotericin B. Voriconazole was also better tolerated than amphotericin B, with significantly fewer serious adverse effects and a longer duration of therapy. Note that the design of these studies has been called into question, and some still consider (liposomal) amphotericin B as the drug of choice [2]. For multiple site or CNS aspergillosis a combination therapy of voriconazole and caspofungin should be considered. It is also the recommended treatment for the CNS fungal infections transmitted by epidural injection of contaminated steroids. With fewer patients having to switch from initial voriconazole than amphotericin B or its lipid formulations because of intolerance or insufficient response, and limited efficacy of salvage therapy with other licensed antifungals, the importance of effective initial therapy has been demonstrated.

IUPAC name: (2R,3S)-2-(2,4-difluorophenyl)-3-(5-fluoropyrimidin-4-yl)-1-(1H-1,2,4-triazol-1-yl) butan-2-o.

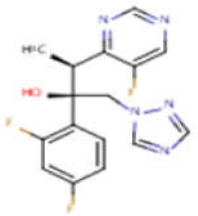

Fig. 1:

Pharmacokinetics

Onset and Duration

1. Initial Response

Antifungal medication used to treat serious fungal infections.

Methodology

Analytical Method Development

Identification and Authenticity of Voriconazole Pure Drug

A. Determination of Melting Point

Melting point of the drug was determined by

taking a small quantity of drug in a capillary tube closed at one end which was then placed in Tehsil´s melting point apparatus. The temperature at which the drug melts was noted using liquid paraffin as a solvent. Average of triplicate readings was recorded.

B. Infrared Spectral Studies

Infrared spectroscopy deals with the infrared region of the electromagnetic spectrum. FTIR is most useful tool for identifying chemicals that are either organic or inorganic by identifying the types of chemical bonds (functional groups).

In FT-IR spectroscopy, IR radiation is passed through a sample. Some of the infrared radiation is absorbed by the sample (the wavelength of light absorbed is characteristic of the chemical bond) and some of it is passed through (transmitted). The resulting spectrum represents the molecular absorption and transmission, creating a molecular fingerprint of the sample. Like a fingerprint no two unique molecular structures produce the same infrared spectrum. By interpreting the infrared absorption spectrum, the chemical bonds in a molecule can be determined. The organic compounds produced spectra are rich and detailed whereas inorganic compounds spectra are much simpler.

Applications

i. It can be utilized to identify components of an unknown mixture. It can be applied to the analysis of solids, liquids, and gasses.
ii. Identification of compounds by matching spectrum of unknown compound with reference spectrum (fingerprinting)
iii. Identification of functional groups in unknown substances
iv. Identification of polymers, plastics, and resins.

Method

In this technique, approximately 2-3 mg of the drug was allowed to mix with about 0.5- 1 g of KBr (which is transparent to IR) and then thoroughly grind the mixture in a mortar, press the mixture in a pellet die manually and placed it in a Fourier transform infrared (FTIR) spectrophotometer (Shimadzu corporation 8400S, Japan).

C. Differential Scanning Calorimetry Studies

- Differential scanning calorimetry or DSC is a thermoanalytical technique in which the difference in the amount of heat required to increase the temperature of a sample and reference are measured as a function of temperature. Both the sample and reference are maintained at very nearly the same temperature throuVEout the experiment.
- The basic principle underlying this technique is that, when the sample undergoes a physical transformation such as phase transitions, more (or less) heat will need to flow to it than the reference to maintain both at the same temperature.
- Whether more or less heat must flow to the sample depends on whether the process is exothermic or endothermic. For example, as a solid sample melts to a liquid it will require more heat flowing to the sample to increase its temperature at the same rate as the reference. This is due to the absorption of heat by the sample as it undergoes the endothermic phase transition from solid to liquid.
- Likewise, as the sample undergoes exothermic processes (such as crystallization) less heat is required to raise the sample temperature. By observing the difference in heat flow between the sample and reference, differential scanning calorimeters are able to measure the amount of heat absorbed or released during such transitions.

Applications

i. DSC can measure important *thermoplastic* properties including: Melting temperature, heat of melting, percent crystallinity, Tg or softening, crystallization, presence of recyclates/regrinds, plasticizers, polymer blends (presence, composition and compatibility) etc.
ii. DSC can also be used to study oxidation as well as other chemical reactions.
iii. DSC is widely used in the pharmaceutical and polymer industries. In the pharmaceutical industry it is necessary to have well-characterized drug compounds in order to define processing parameters. For instance, if it is necessary to deliver a drug in the amorphous form, it is desirable to process the drug at temperatures below those at which crystallization can occur.

Method

Drug sample of about 1-3 mg were weiVEed and placed in aluminium pans and the lids were crimped using a shimadzu crimper. An empty pan sealed in the same way as for the sample was used as a reference. Thermal behavior of the samples was investigated under nitrogen gas at scanning rate of 20°C/min, covering a temperature range of 30- 300°C. The instrument was calibrated with an indium standard.

Determination of ë Max for Voriconazole by UV-Visible Spectroscopy

A. Preparation of 2000 µg/ml Voriconazole Stock Solution

Voriconazole stock solution was prepared by dissolving accurately weighed 200 mg of Voriconazole in 100 ml of volumetric flask using distilled water to get the stock solution of 2000 µg/ml.

B. Preparation of 1200 µg/ml Voriconazole Standard Solution

From the stock solution (2000 µg/ml), 6ml was pipetted out and diluted with distilled water in 10 ml volumetric flask up to the mark to get concentration of 1200 µg/ml.

C. Preparation of 0.2M Phosphate Buffer pH 6.0

D. Determination of Absorption Maxima (λ max) using UV-Visible Spectroscopy

4 ml of VORICONAZOLE standard solution (1200 µg/ml) was taken in a test tube. 0.5 ml 0.8% ninhydrin solution followed by 0.5 ml 0.2M phosphate buffer pH 6.0 was added. Heated for 5 mins at 100°C for reaction to occur. Violet colour solution was obtained. Diluted that solution to 10 folds. The resulting solution was scanned between 400-800nm. The λmax was found to be 558nm and was used as analytical wavelength throuVEout the study.

Calibration curve of Voriconazole

From the stock solution (2000 µg/ml), aliquots of 1, 2, 4, 6 and 8 ml were withdrawn and further diluted to 10 ml with distilled water to obtain concentration range of 0.2-1.6 mg/ml. From these solutions, 4 ml was taken in test tubes separately and reacted with 0.5 ml of 0.8% ninhydrin solution followed by 0.5 ml of 0.2M phosphate buffer pH 6.0. Heated for 5 min to get violet color and diluted to 10 folds. The absorbance was measured immediately within 15 min at 558nm using double beam UV- Visible Spectrophotometer (1700, Shimadzu, Japan). This procedure was performed in triplicate to validate the calibration curve.

Pre-Formulation Studies

Pre-formulation testing is the first step in rational development of dosage forms of a drug substance. Pre-formulation study is the process of optimizing the delivery of the drug throuVE determination of physico-chemical properties of the new compound that could affect the drug performance and development of an efficacious, stable and safe dosage form. It gives the information needed to define the nature of the drug substances and provide a framework for the drug combination with pharmaceutical excipients in the dosage form.

Drug and Polymer Interaction Studies

A. FTIR Studies

FTIR spectroscopy was carried out to check the compatibility between drug and polymer. The IR spectra of Voriconazole, carbopol 940, HPMC K-100 and physical mixtures of drug and polymer was carried out by KBr disc method using FTIR 8400S, Shimadzu, Japan. The wave numbers of characteristic peaks of physical mixtures were compared with the pure samples and interpreted.

B. DSC Studies

Thermal analysis can be used to investigate and predict any physico- chemical interaction between components in a formulation and therefore can be applied to the selection of suitable chemically compatible components.

An interaction on DSC will show changes in melting point and or appearance of the transition. However there is found to be some changes in transition temperature, peak shape and area by virtue of the mixing of two components. This is not due to any detrimental interaction.

The advantages of DSC over more traditional, routine compatibility screening methods like TLC, is that no long term storage of mixture is required for prior evaluation nor any inappropriate thermal stress required to accelerate the interactions.

DSC studies were performed using METTLER 7 system equipped with thermal analysis data system. The drug and polymers were analysed separately

and in combination mixture of drug: polymer (1:1) from 0-300°C at 5°C/min in a nitrogen environment.

Formulation of Gellified Emulsion of Voriconazole

Gel: The composition of voriconazole emulgel- 10% w/w has been shown in Table 1 and 2. The carbopol gel was prepared by dispersing 1.5g carbopol 940 in purified water with constant stirring at a moderate speed and soaked overniVEt. The gel was obtained by neutralizing the dispersion with triethanolamine and adjusted the pH to 6.5 and purified water was added to adjust the weight to 50g. In case of HPMC K-100, gel was prepared by dispersing HPMC K-100 in hot purified water (80°C), and the dispersion was cooled. Weight adjusted to 50g with purified water.

Emulsion: The oil phase of emulsion was prepared by dissolving span 60 in liquid paraffin while the aqueous phase was prepared by dissolving tween 20 and drug in 5 ml purified water. Methyl paraben and propyl paraben were dissolved in propylene glycol and this solution was mixed with aqueous phase. Both oil and aqueous phase were heated separately to 70°C and then oily phase was mixed with aqueous phase slowly by triturating in mortar and pestle to get an emulsion and finally volume made upto 20g with purified water.

Emulgel: The obtained emulsion was mixed with the gel in 1:1 and 1:1.5 ratios according to the formula given in Table 1 and 2 to get an emulgel, weight made upto 50g with distilled water and homogenized to get an emulgel of voriconazole 10% w/w.

Formulation of Hydrogel of Voriconazole

The gel formulation of GAS with polymer concentration equivalent to the selected emulgel formulation of both HPMC K-100 and carbopol 940 was made with drug concentration of 10% w/w as discussed above.

Evaluation Parameters

Physical Appearance

The prepared emulgel formulations were inspected visually for their color, homogenecity, consistency, and phase separation.

pH Evaluation

pH evaluation is the important criteria especially for the topical formulation. The pH of the emulgel should be between 5-7 to mimic the skin condition. If the pH of the prepared emulgel is acidic or basic, it may cause irritation to the patient.

pH of the prepared emulgel was measured using

Table 1: Formulation of emulgel using carbopol 940

Ingredients	Formulation code							
	VE-1	VE-2	VE-3	VE-4	VE-5	VE-6	VE-7	VE-8
				Emulsion				
VORICONAZOLE (g)	5	5	5	5	5	5	5	5
Liquid paraffin (ml)	7.5	7.5	7.5	7.5	7.5	7.5	7.5	7.5
Span 60 (g)	0.63	1.25	0.63	1.25	0.63	1.25	0.63	1.25
Tween 20 (ml)	0.63	0.63	1.25	1.25	0.63	0.63	1.25	1.25
Propylene glycol (ml)	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Methyl paraben (g)	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03
Propyl paraben (g)	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
Purified water (q.s)	20 ml							
	Gel							
Carbopol 940- 3%w/w (g)	20	20	20	20	30	30	30	30
Purified water (q.s)	50 g							
Emulsion: Gel	**1:1**	**1:1**	**1:1**	**1:1**	**1:1.5**	**1:1.5**	**1:1.5**	**1:1.5**

Table 2: Formulation of emulgel using HPMC K-100

Ingredients	Formulation code							
	VE-1	VE-2	VE-3	VE-4	VE-5	VE-6	VE-7	VE-8
				Emulsion				
VORICONAZOLE (g)	5	5	5	5	5	5	5	5
Liquid paraffin (ml)	7.5	7.5	7.5	7.5	7.5	7.5	7.5	7.5
Span 60 (g)	0.63	1.25	0.63	1.25	0.63	1.25	0.63	1.25

Tween 20 (ml)	0.63	0.63	1.25	1.25	0.63	0.63	1.25	1.25
Propylene glycol (ml)	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Methyl paraben (g)	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03
Propyl paraben (g)	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
Purified water (q.s)	**20 g**							
	Gel							
HPMC K-100- 5%w/w (g)	20	20	20	20	30	30	30	30
Purified water (q.s)	50 g							
Emulsion: Gel	**1:1**	**1:1**	**1:1**	**1:1**	**1:1.5**	**1:1.5**	**1:1.5**	**1:1.5**

digital pH meter (ELICO LI 613) by dipping the glass electrode into an emulgel. The measurement of pH of each formulation was done in triplicate and average values were calculated.

Rheological Study

The viscosity of the gel during handling, transport and storage is an important criteria. The viscosity of different emulgel formulation was determined at 25°C using a Brook field viscometer (Brookfield DV II+ viscometer). The emulgels were rotated using spindle 1 at 10, 50 and 100 rpm and the viscosities were measured.

Spreadability Test

One of the criteria for a dermatological preparation is to meet the ideal qualities is that it should possess good spreadability. Spreadability is a term expressed to denote the extent of area to which the gel readily spreads on application to skin or the affected area. The therapeutic efficiency of the formulation also depends on its spreadability values. Hence determination of spreadability is important in evaluating gel characteristics. Spreadability is measured as:

The spreadability of each sample was evaluated in triplicate by using fabricated spreadability apparatus which consists of two glass plates. 0.5 g of sample was placed on the lower plate and the upper plate was placed on the top of the sample. Force was generated by adding increasing weight slowly at 1 min interval into the pan connected to the upper plate. Each sample was tested at least three times at constant temperature and exerted weight and the mean values of spread surface area on the lower plate were calculated.

Extrudability

It is a usual empirical test to measure the force required to extrude the material from tube. The method applied for determination of applied shear in the region of the rheogram corresponding to a shear rate exceeding the yield value and exhibiting consequent plug flow. The emulgels were filled into collapsible tubes, crimped and the extrudability of the formulation from the packed material was tested.

Drug Content Determination

Drug concentration in emulsified gel was measured by UV-Visible spectrophotometer. Voriconazole content in emulsified gel was measured by dissolving accurately weighed (1g) of emulsified gel in solvent (purified water) by Sonication. This solution was treated with 0.5ml of 0.8 % ninhydrin solution followed by 0.5ml of 0.2M phosphate buffer pH 6.0. Heated for 5 min and diluted to 10 folds prior to absorbance. Absorbance was measured at 588nm using UV-Visible spectrophotometer 1700 (Shimadzu, Japan). The test was conducted in triplicate and the average % drug content was determined.

Fig. 2: Photograph of spreadability apparatus

Determination of Globule Size

The globule size analysis of the optimized formulation VE-4 and VE-4 were determined by treating the emulgel sample with scarlet red dye and spreaded over as a thin film on the glass slide and observed under the 10X of the Olympus microscope.

Isolation of egg Membrane

Egg was taken and made a small hole on the tip portion of the egg. The contents of the egg were removed via that hole. Then egg shell was washed internally with water and dipped into 0.1N Hcl solution for four hours. The outer shell of the egg would dissolve and egg membrane was isolated from it.

In-Vitro Drug Permeation Study

In-vitro permeation study was carried out using keisshary chein cell having capacity of 16ml volume. Egg membrane was isolated and used for the study. PreweiVEed (1.5g) emulgel was spread evenly on to the egg membrane. The egg membrane was clamped between donor and receptor compartment. The receptor compartment was filled with 16 ml of purified water maintained at 37ºC and stirred by using magnetic stirrer. The sample (2ml) was collected at suitable time intervals and analyzed for drug content by UV-Visible Spectrophotometer 1700 (Shimadzu, Japan) at 558 nm after appropriate dilutions as discussed earlier.

The same procedure was opted for VORICONAZOLE hydrogel 10% w/w prepared by using carbopol 940 of 3% w/w and HPMC K-100 of 5% w/w.

In-Vitro Drug Release Kinetics43,44,45

In-vitro drug release mechanism was determined by using PCP DISSO V2 software. Depending upon R and k values obtained from different models, the best-fit model was selected.

Zero Order Release

- Drug dissolution from dosage forms that do not disaggregate and release the drug slowly can be represented as:

  Q = Q0 + K0t

Fig. 3: Photograph of Keishary chein cell

Where Q is the amount of drug released or dissolved (assuming that release occurs rapidly after the drug dissolves), Q0 is the initial amount of drug in solution (it is usually zero), and K0 is the zero order release constant. The plot made: cumulative % drug release vs. time (zero order kinetic model).

- Zero order drug release mechanism is mainly applicable to dosage forms like transdermal systems, coated forms, osmotic systems as well as matrix tablets with low soluble drugs.

First Order Release

- To study the first order release rate kinetics the release rate data were fitted into the following equation,

  LoVE =LoVEo- kt / 2.303

  where Cis the amount of drug released at time t, C o is the initial amount of drug in the solution and K1 is the first order release constant.
- This model is applicable to study of hydrolysis kinetics and to study the release profiles of pharmaceutical dosage forms such as those containing water-soluble drugs in porous matrices.

Higuchi Model

- This model is based on the hypotheses that (i) initial drug concentration in the matrix is much hiVEer than drug solubility; (ii) drug diffusion takes place only in one dimension (edge effect must be negligible); (iii) drug particles are much smaller than system thickness; (iv) matrix swelling and dissolution are negligible; (v) drug diffusivity is constant and (vi) perfect sink conditions are always attained in the release environment.
- Higuchi described the release of drugs from insoluble matrix as a square root of time dependent process based on Fickian diffusion

  Q = Kt1/ 2

  Where, K is the constant reflecting the design variables of the system.
- This model is applicable to systems with drug dispersed in uniform swellable polymer matrix as in case of matrix tablets with water soluble drug.

Hixson-Crowell Model

- The Hixson-Crowell cube root law describes the release from systems where there is a change in surface area and diameter of particles or tablets.

  $Q01/3 - Qt1/3 = KHC\ t$

  Where, Qt is the amount of drug released in time t, Q0 is the initial amount of the drug in tablet and KHC is the rate constant for Hixson-Crowell rate equation.

- This expression applies to pharmaceutical dosage form such as tablets, where the dissolution occurs in planes that are parallel to the drug surface if the tablet dimensions diminish proportionally, in such a manner that the initial geometrical form keeps constant all the time. When this model is used, it is assumed that the release is limited by the drug particles dissolution rate and not by the diffusion that miVEt occur throuVE the polymeric matrix.

- Korsmeyer-Peppas Model

  Korsmeyer et al (1983) derived a simple relationship which described drug release from a polymeric system. To find out the mechanism of drug release, first 60% drug release data was fitted in Korsmeyer–Peppas model:

$Mt/M'' = Ktn$

Where Mt / M"is fraction of drug released at time t, k is the rate constant and n is the release exponent. The n value is used to characterize different release mechanisms as given in table 1 for cylindrical shaped matrices,

Stability Studies

Stability of a drug has been defined as the ability of particular formulation in specific container to remain within in its physical, chemical, therapeutic and toxicological specification.

Factor Affecting Stability

Extrinsic: Temperature, liVEt, gases, moisture

Intrinsic: pH, complexation, microbial growth

Boundary: Container composition, porosity, dosage form interaction.

Table 3: Diffusion exponent and release mechanism

Diffusion exponent (n)	Diffusion mechanism
0.5	Fickian diffusion (Higuchi matrix)
0.5 < n < 1	Anomalous (non- Fickian) diffusion
1	Case-II transport (zero order release)
n > 1	Super Case-II transport

Stability testing is an integral part of formulation development. It generates information on which to base proposals for shelf lives of drug substance and products and their recommended storage conditions. Stability data also are a part of the dossier submission to regulatory agencies for licensing approvals.

The complexity and diversity of pharmaceuticals have increased so much in recent years that designing the stability testing protocol for a particular product can be difficult and finding the riVEt approach for estimating retest, shelf-life and expiry periods also can be challenging. Fortunately, useful guidance's are available to address most of those issues.

The International Conference of Harmonization (ICH) tripartite guideline "stability testing of new drug substance and products" describes the stability test requirements for drug registration applications in the European Union, Japan and the United States of America. But it does not seek to cover the testing that may be required for registration in other areas of the world with different climatic conditions.

Further to harmonize and simplify worldwide stability testing, the extension of the ICH tripartite guideline described the stability requirements. The world has been divided into four climate zones. India comes under the climatic zones of III and IV and hence storage conditions protocol mentioned below should be followed:

Stability test	Conditions	Period of test
Long-term testing	30±20C/70±5%RH	12months
Accelerated testing	40±20C/75±5%RH	6months

Salient Features of ICH Guidelines

- The stability test should be conducted using the containers and closures proposed for storage and distribution.

Table 4: Definition and storage conditions for the four climatic zones

Climatic zone	Definition	Storage conditions
I	Temperate climate	21^0C/45%RH
II	Subtropical and Mediterranean climate	25^0C/60%RH
III	Hot, dry climate	30^0C/35%RH
IV	Hot, humid climate	30^0C/70%RH

- The stability plan must include different types of containers and closures such as those used for marketing, physician and promotional samples and bulk storage. However, for bulk containers testing in prototype container that simulates the actual packaging is allowed in ICH and FDA guidelines.
- A sampling frequency of every 3months during the first year, every 6 months during the second year and then annually for drug substances and products stored for real time testing.
- At least two containers are required to be sampled during the stability study.
- To predict the shelf life of the dosage form for clinical zone III and IV, the predictive factor is 3.3 at 300C (6 months at 400C corresponds to 20months at 300C).

Procedure

The selected formulations were filled in Aluminium collapsible tube and crimped. They were placed horizontally in the Stability Chamber and subjected to stability studies at accelerated testing conditions (40±20C/75±5%RH) for 6 months. At specified intervals of time, the samples were withdrawn and evaluated for parameters such as physical appearance, pH, spreadability, drug content and *in-vitro* permeation study.

Result

Identification and Authenticity of Voriconazole Pure Drug

Determination of Melting Point

Table 7: Data of melting point of drug

Compound name	Melting point	
	Observed	Standard
Voriconazole	205°C	205-210°C

IR Spectroscopy

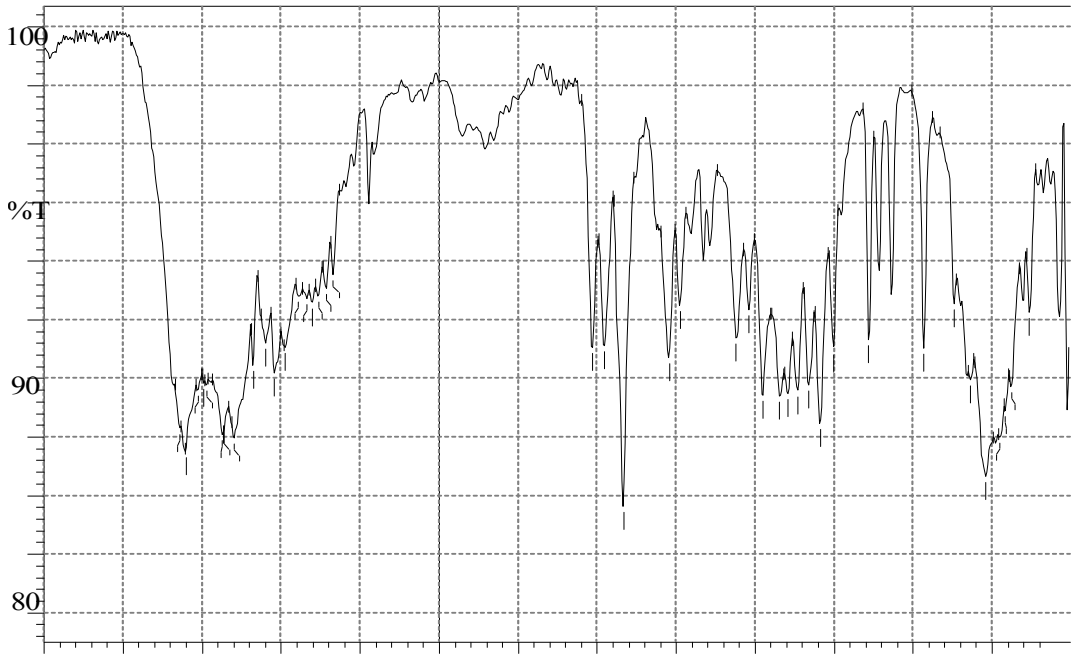

Fig. 4: IR Spectroscopy

Differential Scanning Calorimetry Studies

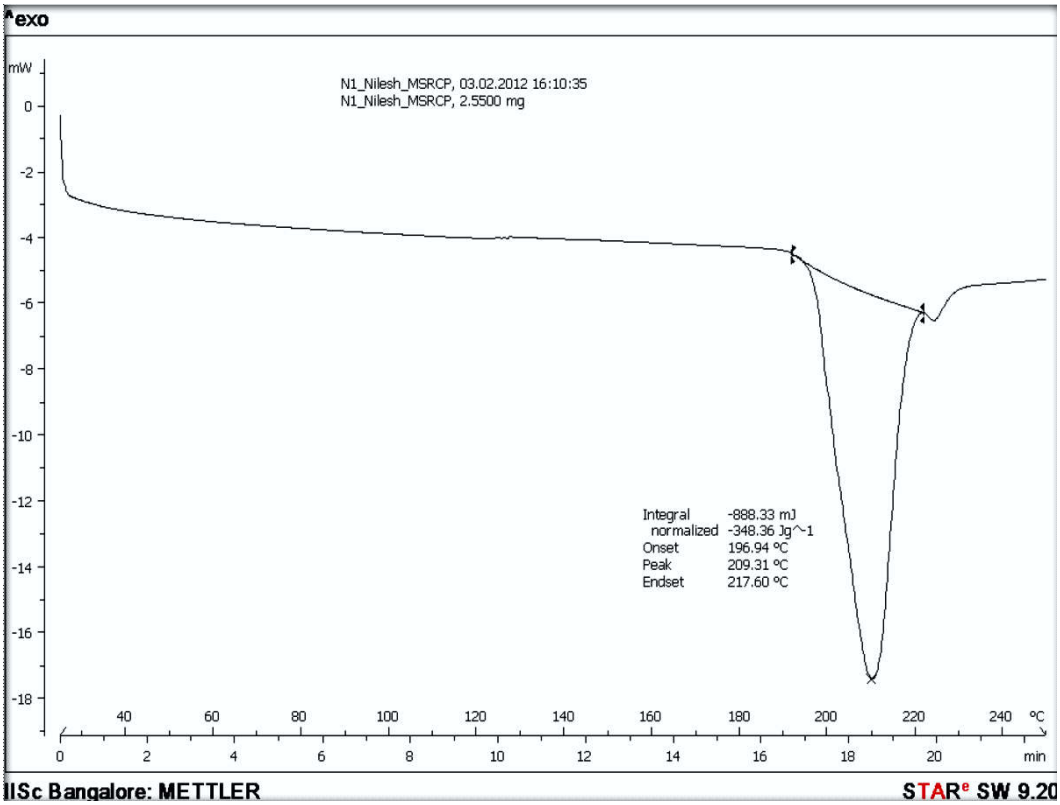

Fig. 5: DSC thermogram of VORICONAZOLE

Determination of λmax for VORICONAZOLE by UV-Visible Spectrophotometer

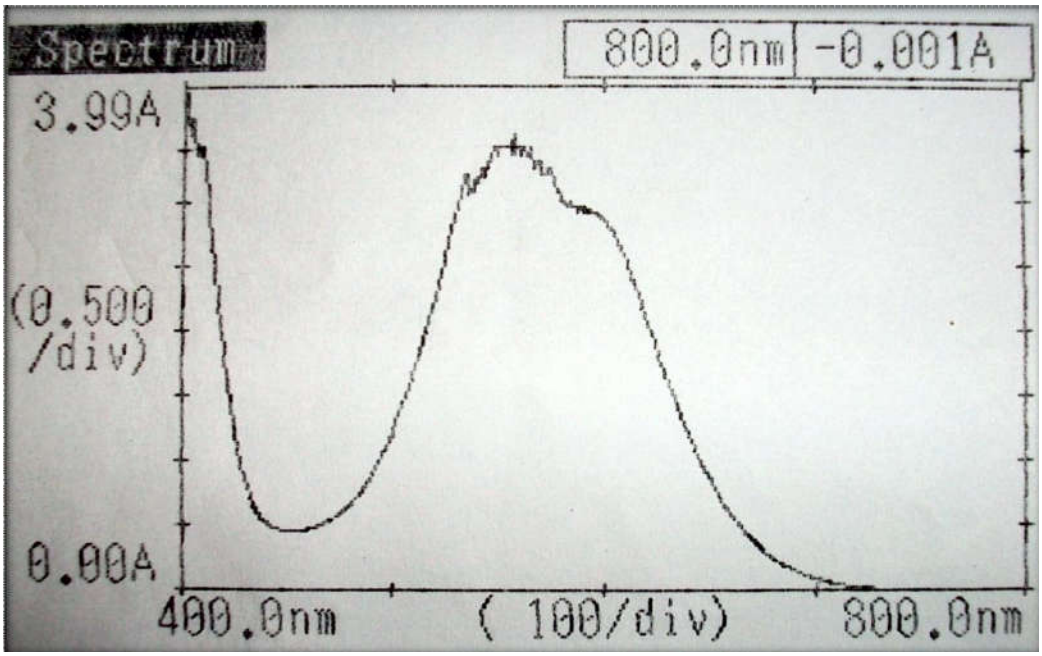

Fig. 6: UV Spectrum of VORICONAZOLE from 400-800 nm

Calibration Curve of VORICONAZOLE

Table 6: Calibration curve of VORICONAZOLE

Concentration (µg/ml)	Absorbance			Mean± SD
	Trial 1	Trial II	Trial III	
200	0.070	0.072	0.074	0.072±0.02
400	0.180	0.181	0.181	0.181±0.01
800	0.390	0.391	0.389	0.390±0.01
1200	0.602	0.602	0.601	0.602±0.01
1600	0.821	0.820	0.821	0.821±0.01

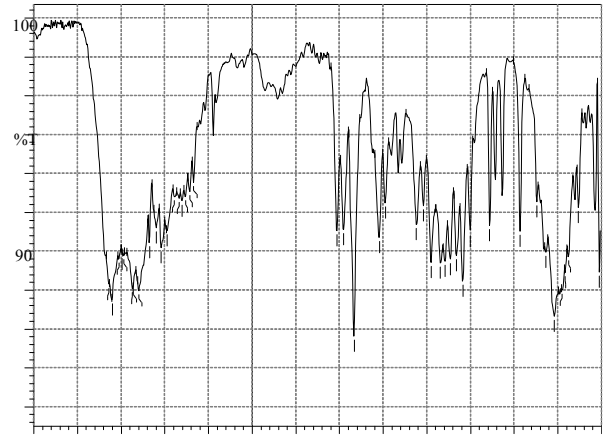

Fig. 7:

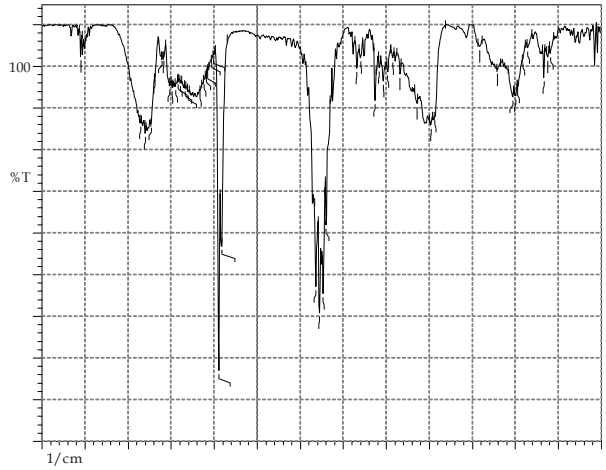

Fig. 8:

Table 7: IR spectral data of HPMC K-100

Compound name	Functional group	Standard range	Observed peak
HPMC K-100	C-C aliphatic	1100-800	1035.70
	C-O-C	1300-1000	1116.66
	CH (methylene) stretch	3000-2840	2831.31
	C-O-R stretch	1200-1100	1163.00

Transdermal drug delivery system (TDDS) facilitates the passage of therapeutic quantities of drug substances through the skin and into general circulation for their systematic effects. Evidence of percutaneous drug absorption may be found through measurable blood levels of the drug, detectable excretion of the drugs and its metabolites in urine and clinical response of the patients to the therapy.

The purpose of the topical dosage form is to conveniently deliver drugs across a localized area of the skin. To develop an ideal dosage form, one must take into account flux of the drug across the skin, nature of the drugs, patient´s acceptability of formulation etc.

Although having plenty of advantages over other routes of drug administration, transdermal drug delivery system is having certain limitations including hydrophilic drugs cannot easily penetrate across the skin. To overcome this problem, the drugs have to made sufficiently lipophillic or use of penetration enhancers will help to achieve the desired results.

VORICONAZOLE is a hydrophilic, anti-arthritic drug with biological half life of 1.5-2 hrs with oral bioavalability of 26%. On this context, gellified emulsion was formulated using hydrogel polymers like carbopol 940 and HPMC K-100, liquid paraffin as oil phase, emulsifying agents like span 60, tween 20 and propylene glycol as permeation enhancers.

On the basis of quality of emulgel produced, sixteen formulations namely VE-1 to VE-8 and VE-1 - VE-8 were selected. They were evaluated for physical appearance, pH evaluation, rheological study,

spreadability study, drug content and *in-vitro* permeation study.

Prior to formulation, drug polymer interaction studies were carried by IR and DSC studies and found to be compatible. Due to lack of chromophore in the structure of the drug, it cannot be detected directly by UV-Visible spectrophotometer. To overcome this problem, it was treated with ninhydrin solutions and phosphate buffer pH 6.0 by heating method and converted to detectable moieties.

The effect of the formulation variables on the drug permeation kinetics and the increased permeation of emulgel incontrast to hydrogel with the optimized formulation were also studied

The accelerated stability study was conducted as per ICH guidelines and concluded with the prediction of its shelf life.

Thus, the formulated gellified emulsion had a distinct advantage over existing conventional dosage form in that the drug permeation was rapid across the skin and hence increased therapeutic response bypassing the first pass metabolism and also patient compliance.

References

1. Arunachalam A et al. Transdermal drug delivery systems: A review. Current Pharma Res 2010; 1(1): 70-81.
2. Meera C SinVE, Ajinkya S Naik, Sawant SD. Transdermal drug delivery system with major emphasis on transdermal patches. J Pharm Res 2010; 3(10):2537-2543.
3. Robinson JR, Lee VH. Controlled drug delivery fundamentals and applications. 2nd ed. New Delhi: CBS Publishers & Distributors; 2005; 29: 523-552.
4. Kotta Kranthi Kumar, Sasikanth K, Sabareesh M, Dorababu N. Formulation and evaluation of diacerein cream. Asian J Pharm Clinical Res 2011; 4(2):93-98.
5. Jonathan Hadgraft, Richard H Guy. Transdermal drug delivery: development issues and research initiative. Marcel Dekker. Vol 35:1-16.
6. Divyesh Patel, Nirav Patel, MeVEal Parmar, Navpreet Kaur. Transdermal drug delivery system: review. Int J BiopharmToxicol Res 2011; 1(1): 61-80.
7. Virendra Yadav. Transdermal drug delivery system: review. Int J Pharm Sci Res 2012; 3(2):376-382.
8. Ankush I Shembale, Dipak K Borole, Rajesh T Lohiya. Useful permeation enhancers for transdermal drug delivery: A review. Int J Pharma Res Dev 2010; 2(5):1-6.
9. Loyd VA, Nicholas GP, Ansel HC. Pharmaceutical dosage forms and drug delivery systems. 8th ed. Philadelphia: Lippincott Williams and Wilkins 2005; 298-315.
10. Bhavsar JD, Brahambhatt VG, Patel MR, Patel KR, Patel NM. Review article: Novel approaches in semisolids. Int J Pharm World Res 2011; 2(1):1-22.
11. Patil Bharat, Mandore Paresh, Sharma RK, Tekade BW, Thakre VM, Patil VR. A review: novel advances in semisolid dosage form and patented technology in semisolid dosage forms. Int J Pharm Tech Res 2011; 3(1):4.