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A REVIEW ON CLOTRIMAZOLE LOADED TRANSETHOSOMAL GEL FOR CANDIDIASIS

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ABSRACT

Candidiasis is one of the most common fungal infections. For the treatment, antifungal therapy is used. Clotrimazole is a topical broad spectrum antifungal agent used for the treatment of a wide variety of dermatophyte infections and candidiasis. Various disadvantages occur when the drug is administered as conventional form. In this dealing with the latest form of vesicular system like transethosomes. Transethosomes are one of the potential innovative nano-carriers for improving the solubility and permeation of poorly soluble and permeable drugs. When the drug is incorporated in the form of transethosomes which nullify the all the disadvantages when it is formulated in any other conventional systems and there by provide stable and more effective formulation, and also having high patient compliance. Transethosome containing high concentration of ethanol along with edge activator which provide high penetration to the skin and leads to better therapeutic action. Clotrimazole loaded transethosomes to overcome the skin barrier function. Clotrimazole loaded transethosomes fabricated by the cold method and followed by their incorporation into Carbopol 940 as a gel. The prepared clotrimazole loaded transethosomes were evaluated for percentage yield, % Entrapment efficiency (EE), surface morphology, particle size, zeta potential. The development of transethosomal formulation can serve as an efficient drug delivery system through a topical route with enhanced efficacy and better patient compliance.

KEYWORDS: Fungal infection, Clotrimazole, Transethosomes, Candidiasis, Edge activator.

INTRODUCTION

The fungal infection caused by Candida species is known as Candidiasis. The prevalence of superficial fungal infections of the skin, hair, and nails has risen globally. In undeveloped and underdeveloped countries, it is believed that roughly 40 million people have been infected with fungi. Due to compromised immune function, the course of fungal infections can be fast and serious. One of the most common causes of tinea and onchomycosis is dermatophytes. Candida infections are also among the most common fungal infections of the skin. When the immune system is weakened, candida can further infect deeper tissues and blood, resulting in life threatening systemic candidiasis.

Topical treatment of fungal infections has various advantages, including the ability to target the infection site, reduced risk of systemic adverse effects, improved treatment efficacy, and high patient compliance. A range of topical antifungal agents have been employed in treatment of various dermatological infections. These antifungal medicines are currently accessible in creams, gels, lotions, and sprays in traditional dose forms. The effectiveness of topical antifungal treatment is determined on drug penetration through the target tissue. As a result, the effective medication concentration levels in the skin should be obtained. When antifungals are applied topically, the drug components must penetrate through the stratum corneum; the skin's outermost layer, to reach the lower layers, notably the viable epidermis. New carrier systems for licensed and investigational medications are being developed as alternate techniques for topical treatment of fungal infections of the skin. Antifungal chemicals can be delivered to the skin more effectively using carriers such as colloidal systems and vesicular carriers. The vesicles are the colloidal systems in which the hydrophilic core is surrounded by amphiphilic molecules in a double layered fasion. Vesicular systems have capability to encapsulate wide variety of drug hydrophilic, lipophilic and charged hydrophilic, and amphiphilic. The effectiveness of a vesicular systems as a carrier depends on various physicochemical characteristics like surface charge, size, thermodynamic phase, and lamellarity. The conventional liposomes show the drawback of less permeation into the deeper region of skin and they accumulate at the outer layer of stratum corneum. Transfersomes and the liposomes having the addition of edge activator like span 60, Span80, span25, tween80, and Sodium deoxycholate and sodium cholate. Transfersomes improve the skin deposition of many drugs. But they can’t reach the stratum corneum deep enough. Ethosomes are the composition of the phospholipid, ethanol and water and fluidization caused by ethanol may increases intercellular space between corneocytes and enhance the

skin permeation. so the transethosomes is represent the novel lipidic formulation that encompasses the advantage of both transfersomes and ethosomes. The Transethosome show the presence of high amount of ethanol with edge activator or the permeation enhancers. The novel lipid vesicles is also known as the deformable or elastic liposomes- liposomes. They have the great ability to intact the skin and deliver the drug into ultradeformable vesicles (UDV) were developed in the beginning of the 1990's. The UDV are more deformable than the conventional the epidermis and dermis layers or even to the systemic circulation. Currently, there are many types of UDV that have been successfully developed for both pharmaceuticals and cosmeceuticals particularly transferosomes, ethosomes, and recently transethosomes. Transethosomes are lipid vesicles which are combinly based or made up of transfersomes and ethosomes. It was first introduced by Song et al in 2012 where he characterized the high content of ethanol (up to 30%). Transethosomes contain the both advantages of transfersomes and ethosomes. Transethosomes have the irregular spherical shape and higher values in both vesicles elasticity and skin permeation/penetration studies. This cause due to the rearrangement of lipid bilayer in the combination of ethanol and edge activator.

Mechanism of transethosomes

The drug absorption through transethosome probably occurs in following two phases.

- Ethanol effect.
- Transethosome effect.

1. Ethanol effect

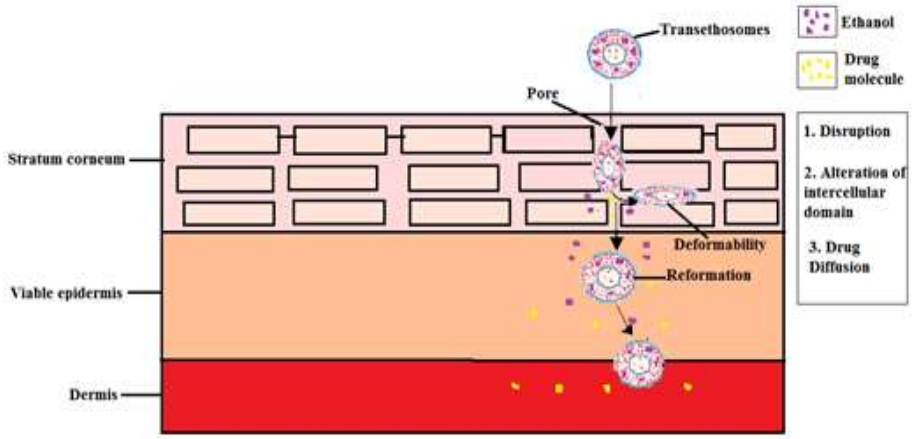
The high concentration of ethanol (30-40%) present in the transethosomes, this ethanol penetrates into the lipids present in the intracellular space of the skin and decreases its density and make it flexible. When the concentration of the ethanol is reduced from 20% may lead to increase in the size of vesicle.

2. Transethosome effect

Permeation enhancer (like tween 20, tween 60, tween 80, span 60, span 65, span 80, sodium cholate, sodium deoxy cholate etc.) present in the transethosome which disrupt the intracellular lipids from stratum corneum. When the intracellular lipids disrupt pores of the skin becomes wider and facilitates the permeation of system across the skin.

The penetration of the transethosomes mechanisms is described in 3 way

1. The interaction between hydrophilic lipid and water makes the polar lipid to attract water molecules induce hydration, lipid vesicles moved to the site of higher water concentration the difference in water contents across skin stratum and epidermis develops transdermal osmotic gradients that leads to penetration of transethosomes across skin.
2. Transethosomes induce hydration that widen pores due to it there is gradual release of drug occurs that binds to targeted organ.
3. Transethosomes act as penetration enhancer which disrupt the intercellular lipids, which results in widen of pores and increase the penetration of system through skin.



Salient features of transethosomes

1. They have high entrapment efficacy, as they are biocompatible and biodegradable in nature.
2. Encapsulation drug is protected from the degradation as due to which they their content slowly and gradually.
3. Easy to prepare, does not involve tedious process and also avoids the unnecessary use of pharmaceutical additives, can be used for both systemic as well as topical delivery.
4. They are highly flexible so have higher flux rate across skin and higher rate of skin penetration as comparisons to other vesicular system.

Advantages

1. The transethosomal system is passive, non-invasive and is available for immediate commercialization.
2. It contains non-toxic raw material in the formulation.
3. The Transethosomes drug is administered in a semisolid form.

4. Transethosomes drug delivery can be applied to many fields including veterinary and cosmetic fields.
5. It shows high patient compliance as it is administered in semisolid gel or cream form.
6. This drug delivery system shows better stability as compared to other conventional vesicle.
7. Avoidance of first pass metabolism.
8. Simple method of drug delivery as compared to iontophoresis, laser surgery, cryo surgery and other complicated methods.
9. Enhanced drug permeation through skin for transdermal drug delivery.

Disadvantages

1. The molecular size of the drug should be reasonable that it should be absorbed percutaneously.
2. Skin irritation or allergic reaction on contact dermatitis.
3. Product loss during transfer from alcoholic and water media.
4. Unsuccessful vesicles formation can coalesce transethosome.

OBJECTIVE

The objective of the present review about to formulate clotrimazole loaded transethosomal gel using phospholipids, ethanol and surfactants.

Drug release from these formulation is sustained and controlled manner.

It can deliver the drugs to deeper tissues without any side effects.

To reduce the frequency of administration thereby improving the patient compliance.

METHODOLOGY

Method of preparation

Cold method

Hot method

Thin film hydration method

Mechanical dispersion method

Cold method: Dissolved phospholipids in ethanol by vigorous stirring. This mixture is heated upto 300^0 c in a water bath. Water is heated up to 300 ^{0}c in a separate vessel and

added to the alcoholic mixture slowly in a fine stream. Depending on the drug solubility, it can be dissolved in water. The mixture is kept on magnetic stirrer at 700rpm during the addition of above aqueous solution to ethanolic solution. Modulation of vesicle size can be done by using probe sonicator.

Hot method: Disperse phospholipids in water by heating in water bath at 400^0 c to obtain a colloidal solution. Ethanol and glycol are mixed and heated up to 400^0 c. organic phase is added to aqueous phase. Stir for 7-10 min. Depending upon its hydrophilic /hydrophobic properties, the size of the vesicle is reduced by probe sonication.

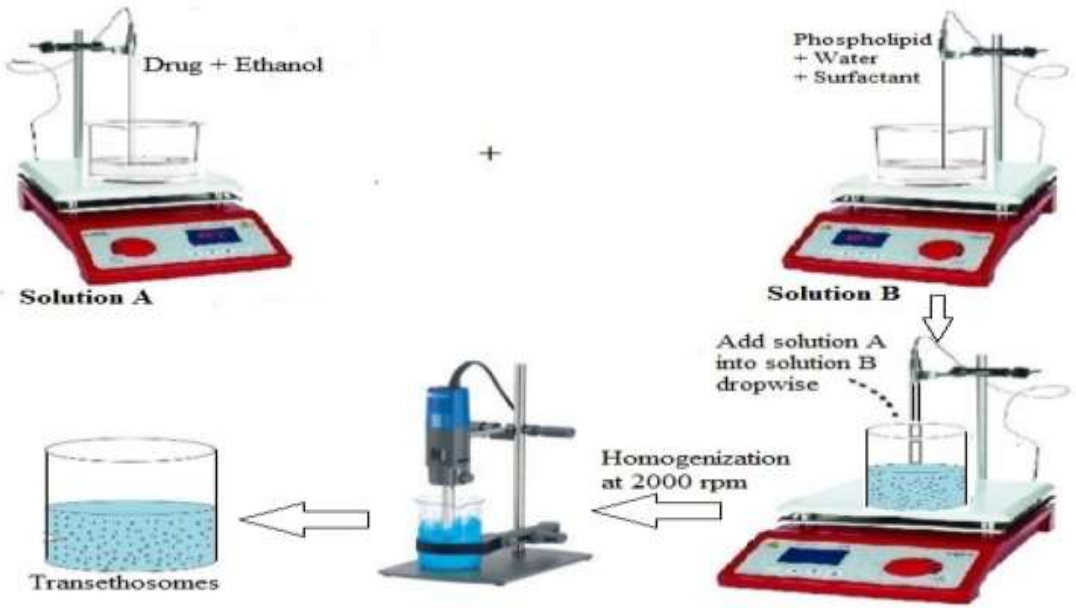
Thin film hydration method: SPC (final concentration of 36mg/ml), permeation enhancers and IM (final concentration of 0.5mg/ml) were dissolved in 25ml chloroform-methanol. The lipid mixture was deposited as a thin film in a round-bottom flask by rotatory evaporating the chloroform- methanol under reduced pressure at 35^0c, which was applied for 1h to ensure total removal of solvent traces. The lipid film was hydrated with 10ml phosphate buffer and achieved within the eluates.

Mechanical dispersion method: Lipid and surfactant is taken in clean, dry round bottom flask. Lipid mixture is dissolved in a solvent mixture of chloroform and ethanol mixture. A thin film of lipid is obtained using rotary evaporator above the lipid transition temperature. It is kept overnight under vacuum to remove traces of organic solvent. The deposited film is hydrated with 10 % v/v ethanol in phosphate buffer pH 6.5 by rotation at 60 rpm. Drug is added to the formulation. The vesicles are sonicated for desired size.

Formulation of transethosomes

By using cold method

The cold method was used for the preparation of clotrimazole transethosomes. A solution of phosphotidylcholine and edge activator in ethanol at 30℃ act as organic phase. The aqueous phase was heated at 30℃ and then delivered to the organic phase drop wise with constant stirring at 700 rpm with the help of magnetic stirrer. The suspension will be cooled down at room temperature of 25℃. Finally the suspension will be homogenized at 2000 rpm and stored in a refrigerator for further study. This method was found to give more stable transethosomes.



Preparation of gel base

Carbopol 934 (%w/v) was accurately weighed and dispersed into double distilled water (80ml) in a beaker. This solution was stirred continuously at 800 rpm for 1 hour and then 10ml of propylene glycol was added to this solution. Volume of gel was adjusted to 100ml and then sonicated for 10 min on bath sonicator to remove air bubbles. Final pH of the gel base was adjusted to 6.8. Transethosomal preparation corresponding to 1.0% w/w of clotrimazole was incorporated into the gel base to get the desired concentration of drug in gel base.

Evaluation of transethosomes

1. **Size of the particle and surface charge:** Size of the particle can be determined using laser scattering particle size distribution analyser. Zeta potential analyser the surface charge can be determined.

2. **Transmission electron microscopy (TEM):** Visualization of the vesicles can be done using TEM. Conventional negative staining is performed using 1% PTA(Phosphotungistic acid), dried later is visualized.

3. **Determination of entrapment efficiency:** Ultracentrifugation technique is used to determine the entrapment efficiency of transethosomes. Ultracentrifugation is performed at 1500rpm for 60 min at 4°c. Sediment and supernatant liquid is separated and the amount of sediment was determined and drug entrapment efficiency was calculated using the equation:

% Entrapment efficiency = Amount entrapped API x 100

4. **Surface morphology study:** Transethosomes are made up of lipids. Different type of lipids influence the surface morphology/ shape of the particles. SEM is used in determining the surface morphology.

5. **Interaction study by using DSC and FTIR:** DSC is used to conduct study for the interaction between lipid and drug. Mettler DSC can be used in determination of the transition temperature(Tm) of the vesicular lipid system. The transition temperature is measured using aluminium crucibles at a heating rate 10°/min within a temperature ranging between 200-300°c. FTIR technique can also be used for conduction the interaction study.

6. **Drug content:** UV spectrophotometer can be utilized in studying the drug content present in the transethosomes. HPLC can be used for quantification.

7. **Stability of ethosome:** The ability of transethosomal formulations to retain the drug is checked by keeping the preparations at different temperatures, i.e. 25±2°C(room temperature), 37±2°C and 45±2°C for different periods of time. The stability of ethosomes can also be determined quantitatively by monitoring size and morphology of the vesicles using DLS and TEM.

8. **Drug content:** The % drug content of transethosomal preparation was determined by using following formula

%drug content = Sample absorbance /standard absorbance.

Applications

When compared to liposomes, ethosomes and nanoethosomes; transethosomes are more effective. They distribute drugs 65% mor effectively than liposomes do because they can pass through more layers of human skin with ease. The effectiveness of these vesicular systems is being investigated using a small number of bioactive compounds.

Delivery of NSAIDs (Non-steroidal Anti-inflammatory Drugs)

NSAIDs taken orally are linked to gastrointestinal adverse effects. Transethosomal formulation of ketorolac tromethamine demonstrated improved penetration. Piroxicam transethosomal gel recently demonstrated greater stability and elasticity compared to other

deformable vesicle systems. They gave humans ethosomes that had been ammonium glycyrrhizinate- entrapped. The formulation with 45% ethanol and a lower proportion of lecithin produced the better results. The in vitro study produced improved tolerability and percutaneous permeability.

Delivery of hormones

Hormone administration orally has been associated with a number of problems, including high first-pass metabolism, poor oral bioavailability, and a range of dose-dependent side effects. Touitou et al., compared a commercially available testosterone transdermal patch (Testoderm® patch, Alza Corporation, California) to the skin penetration capacity of testosterone ethosomes through rabbit pinna skin. The ethosomal formulation's testosterone skin penetration was around 30 times greater than that of a commercially available transdermal patch. Also, it was discovered that the ethosomal system's AUC and Cmax were larger than those of Testoderm®28.

Delivery of antibiotics

The therapeutic efficacy of antibiotics can be increased more effectively when applied topically. The use of oral medication in the past has had a number of adverse consequences, including allergic reactions. Concerns about restricte permeability to subdermal tissues and deeper skin layers, which are frequent with conventional external preparations, can be avoided with ethosomes. Ethosomes rapidly penetrate the epidermis, carrying several medications to the skin's deeper layers and squelching infection at its root. For this objective, Godin and Touitou developed an ethosomal formulation for cutaneous and intracellular administration containing bacitracin and erythromycin. The results of this study showed that an ethosomal antibiotic formulation could be very powerful and circumvent the shortcomings of conventional treatment.

Delivery of antifungal drugs

Terbinafine, amphotericin B, and ketoconazole-containing transethosomes had better penetration. Voriconazole transethosomes demonstrated skin penetration and deposition when compared to regular liposomes, deformable liposomes, and ethosomes.

Delivery of Anti-parkinsonism agent

Trihexyphenidyl hydrochloride (THP), a psychoactive drug, was made into an ethosomal formulation by Dayan and Touitou, and they compared it to conventional liposomal

formulations. Parkinson's disease is managed with THP, an M1 muscarinic receptor antagonist. The results indicated that the ethosomal-THP formulation had a greater capability for skin penetration and may be used to more effectively manage Parkinson's disease.

Delivery of anticancer drugs

While treating cutaneous melanoma, Lei et al., conducted experiments using dual drug loading and transethosomal formulation. They settled on two drugs, dacarbazine and tretinoin, because they worked better together than the other formulations and had less cytotoxicity. Dual loaded transethosomes showed improved antitumor efficacy as compared to a single loaded drug. Skin penetration can be increased, they found. Shaji et al. discovered that encapsulating 5-Fluorouracil into a transethosomal gel led to improved deformability, higher skin penetration, and deeper skin targeting as compared to ethosomes.

CONCLUSION

For the treatment of fungal infections like candidiasis, antifungal class of drug is used. As compared to other drug, clotrimazole having less side effect and excellent pharmacokinetic profile. Oral form may show various disadvantages. Therefore, the development of a novel carrier system for transdermal delivery is required to minimize these disadvantages. The clotrimazole loaded transethosomes showed high stability, prolonged release of the drug, and excellent in-vitro skin permeation to the deeper skin layers. Based on this it can be considered that transethosomal gel may be a promising carrier to deliver drug through topical route.

They provide safety, efficacy and more patient compliance hence are more superior to any other vesicular system. Transethosomes have become promising carriers not only for topical treatment of local but also for systemic disorders. They can be explored in the future for delivery of various drugs through transdermal delivery. Formulation of transethosome in the form of vesicles in gel may improve their viscosity and hence increase their residence time on the site of action.

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