Review Article

Ethosomes: A Novel Approach towards Transdermal Drug Delivery

Abhishek Chandel[1*], Vishal Patil[1], Rohit Goyal[2], Hitesh Dhamija[1] and Bharat Parashar[1]

[1]Department of Pharmacy, Manav Bharti University, Solan, Himachal Pradesh, India
[2]School of Pharmaceutical Sciences, Shoolini University, Solan, Himachal Pradesh, India

ABSTRACT

Skin is the main target of topical and transdermal preparations. Major aim of transdermal drug delivery system is to cross the stratum corneum. Now-a-days we better know vesicles have importance in cellular communication. Ethosomal carriers are systems containing soft vesicles, composed of hydroalcoholic or hydro/glycolic phospholipid in which the concentration of alcohols is relatively high. The high concentration of ethanol brings increase in fluidity of lipids hence increase in permeability of the skin and improves the drug penetration. Ethosomal formulation may contain many drugs such as acyclovir, salbutamol, Insulin, cyclosporine, fluconazole, minodixil, etc. These are prepared by hot method and cold methods. The size of Ethosomal formulation can be decreased by sonication and extrusion method. The high concentration of ethanol makes the ethosomes unique and useful for transcellular delivery, delivery of hormones, anti-arthritis, anti-HIV etc. These are one of the best formulations developed.

Keywords: Ethosomes, Carriers, vesicles, Transdermal drug delivery.

INTRODUCTION

A number of techniques and efforts have been targeted to weaken and disrupt the highly organized intercellular lipids in an attempt to improve drug transport across the whole skin or to increase the permeation of drugs across this skin barrier. The vesicles have been well known for their important in cellular communication and particle transportation for many years. Researchers have known the properties of vesicular structures for better use in drug delivery, they incorporated drugs within their cavities that would allow for tagging the vesicle for cell specificity[1,2]. The soft, malleable vesicles adapt for superior delivery of active agents. Ethosomes are lipid vesicles containing phospholipids, alcohol (ethanol and isopropyl alcohol) in relatively high concentration and water[3]. The size of Ethosomes can be modulated to range anywhere from 30nm to a few microns. Ethosomes provides a number of important benefits including improving the drug efficacy, enhancing patient compliance and comfort and reducing the total cost of treatment[4]. Ethosomes are mainly used for the delivery of drugs through transdermal route. The transdermal delivery is one of the most significant routes of drug administration. The main factor which limits the application of transdermal route for drug delivery is the permeation of drugs through the skin. Human skin has selective permeability for drugs, only the lipophilic drugs having molecular weight < 500 dalton can pass through it. To overcome the stratum corneum barrier, various mechanisms have been investigated, including use of chemical or physical enhancers, such as sonophoresis, iontophoresis, etc. Liposomes, niosomes, transferosomes and ethosomes also have the potential of overcoming the skin barrier and have been reported to enhance permeability of drug through the stratum corneum barrier[5,6]. Ethosomes were designed to

enhance the delivery of drugs into the deep layers of the skin and through the skin. Depending on the formulation, delivery can be targeted for local delivery or for systemic use. Thus comparing to the other transdermal drug delivery systems, it is very important and with research point of view, an interesting system.

ROUTES OF PENETRATION

Human skin comes into contact with sebum, cellular debris, microorganisms and other materials, which somewhat affect the permeation of vesicles. The penetrant permeates by three potential pathways to the viable tissue: (i) through hair follicles with associated sebaceous glands, (ii) via sweat ducts, or (iii) across continuous stratum corneum between these appendages. These pathways are important for ions and large polar molecules that struggle to cross intact stratum corneum.

ETHOSOMES

"Ethosomes are ethanolic liposomes"

Ethosomes can be defined as non-invasive delivery carriers that enable drugs to reach deep into the skin layers and/or the systemic circulation. These are soft, malleable vesicles tailored for enhanced delivery of active agents. The vesicles have been well known for their importance in cellular communication and particle transportation for many years. Vesicles would also allow controlling the release rate of drug over an extended time, keeping the drug shielded from immune response or other removal systems and thus be able to release just the right amount of drug and keep that concentration constant for longer periods of time. One of the major advances in vesicle research was the finding of a vesicle derivative, known as an ethosomes [9, 10]. Ethosomal carriers are systems containing soft vesicles, ethanol at relatively high concentration and water. It was found that ethosomes penetrate the skin and allow enhanced delivery of various compounds to the deep strata of the skin or to the systemic circulation.

Composition

The ethosomes are composed of hydroalcoholic or hydro/glycolic phospholipid in which the concentration of alcohol is relatively high. Typically, ethosomes may contain phospholipids with various chemical structures like phosphatidylcholine (PC), hydrogenated PC, phosphatidic acid (PA), phosphatidylserine (PS), phosphatidyl ethanolamine (PE), phosphatidylglycerol (PPG), phosphatidylinositol (PI), hydrogenated PC, alcohol (ethanol or isopropyl alcohol), water and propylene glycol (or other glycols). Such a composition enables delivery of high concentration of active ingredients through skin. Drug delivery can be modulated by altering alcohol: water or alcohol-polyol: water ratio. Some preferred phospholipids are soya phospholipids such as Phospholipon 90 (PL-90). It is usually employed in a range of 0.5-10% w/w. Cholesterol at concentrations ranging between 0.1-1% can also be added to the preparation. Examples of alcohols, which can be used, include ethanol and isopropyl alcohol. Among glycols, propylene glycol and Transcutol are generally used. In addition, non-ionic surfactants (PEG-alkyl ethers) can be combined with the phospholipids in these preparations.

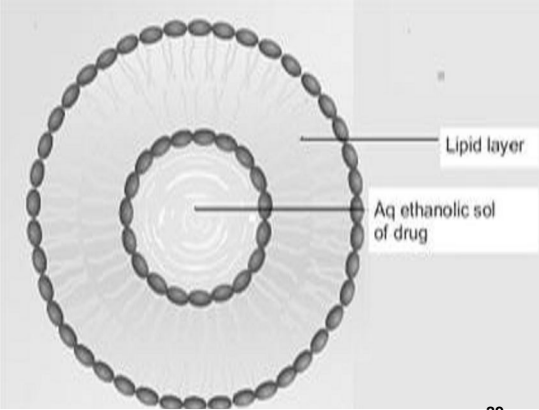

Fig. 1: Proposed diagram of ethosomes[29]

Cationic lipids like cocoamide, POE alkyl amines, dodecylamine, cetrimide etc. can be added too. The concentration of alcohol in the final product may range from 20 to 50%. The concentration of the non-aqueous phase (alcohol and glycol combination) may range between 22 to 70%[12].

MECHANISM OF DRUG PENETRATION

It is thought that the first part of the mechanism is due to the 'ethanol effect' whereby intercalation of the ethanol into intercellular lipids increasing lipid fluidity and decreases the density of the lipid multilayer. This is followed by the 'ethosome effect', which includes inter lipid penetration and permeation by the opening of new pathways due to the malleability and fusion of ethosomes with skin lipids. Absorption of ethosomes is still not clear.

The drug absorption probably occurs in following two phases:

1. Ethanol effect
2. Ethosomes effect

ETHANOL EFFECT

Ethanol is major ingredient and acts as a penetration enhancer during the skin. The mechanism of its penetration enhancing effect is well known. Ethanol interacts with lipid molecules in the polar hard group region, resulting in a reducing the rigidity of the stratum corneum lipids, increasing their fluidity. The intercalation of ethanol into the polar head group environment can result in an increase in the membrane permeability. In addition to the effect of ethanol on stratum corneum structure, the ethosome itself may interact with the stratum corneum barrier[7].

ETHOSOMES EFFECT

Increased cell membrane lipid fluidity caused by the ethanol of ethosomes results increased skin permeability. In the case of ethosomes encapsulating drugs, the higher positive zeta potential imparted by the drug can improve skin attachment of the vesicles. While encapsulated drug in classic liposomes remained primarily at the surface of the skin the Ethosomal system was showed to be highly efficient carrier for enhanced drug delivery through the skin due to increased fluidity of the lipids.

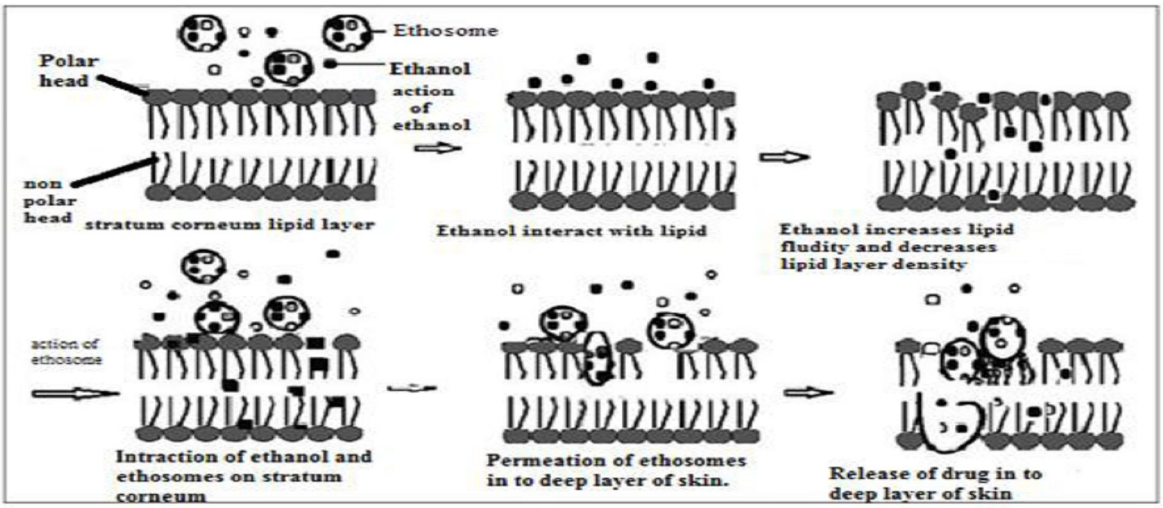

Fig. 2: Mechanism of Action of Ethosomes[29]

Table 1: List of various drug molecules used in Ethosomal drug delivery[3,8]

Drug	Applications	Advantages
Acyclovir	Treatment of Herpetic infection.	Improved drug delivery.
Zidovudine	Treatment of AIDS.	Improved transdermal flux.
Trihexypenidyl HCl	Treatment of Parkinsonian Syndrome.	Increased drug entrapment Efficiency, reduced side effect &constant systemic levels.
Erythromycin	Efficient healing of S. aureus – Induced deep dermal infections.	Improved drug penetration and Systemic effect.
Insulin	Treatment of Diabetes	Improved therapeutic efficacy of drug.
Testosterone	Treatment of male hypogonodism	Enhance skin permeation.
Cannabidol	Prevents inflammation and edema	Significant accumulation of the drug in the skin.
Minodixil	Hair growth promotion effect	Higher skin retention.
Bacitracin	Treatment of dermal infections	Reduced drug toxicity.
Salbutamol	Anti-asthmatic Bronchodilator	Enhanced drug delivery through skin with Ethosomes.
Cyclosporine ·	Treatment of Inflammatory skin disease	GIT degradation, Poor oral absorption and bioavailability.

Table 2: Different Additives Employed In Formulation of Ethosomes[2]

Class	Example	Uses
Phospholipid	Soya phosphatidyl choline Egg phosphatidyl choline Dipalmityl phosphatidyl choline Distearyl phosphatidyl choline	Vesicles forming component.
Polyglycol	Propylene glycol Transcutol RTM	As a skin penetration enhancer.
Alcohol	Ethanol Isopropyl alcohol	For providing the softness for vesicle membrane As a penetration enhancer
Cholesterol	Cholesterol	For providing the stability to vesicle membrane.
Dye	Rhodamine-123,Rhodamine red Fluorescene Isothiocynate (FITC) 6- Carboxy fluorescence	For characterization study.
Vehicle	Carbopol Ð934	As a gel former.

METHOD OF PREPARTION

There are two methods which can be used for the formulation and preparation of ethosomes. These methods are very simple and convenient and do not involve any sophisticated instrument or complicated process. Ethosomes can be formulated by following two methods.

Hot method - In this method, phospholipid is dispersed in water by heating in a water bath at 400ºC until a colloidal solution is obtained. In a separate vessel properly mix ethanol and propylene glycol and heat up to 400^0C. Add the organic phase into the aqueous phase. Dissolve the drug in water or ethanol depending on its solubility. The vesicle size of ethosomal formulation can be decreased to the desire extent using probe sonication or extrusion method[9,10].

Cold method - This is the most common and widely used method for the ethosomal preparation. Dissolve phospholipid, drug and other lipid materials in ethanol in a

covered vessel at room temperature with vigorous stirring. Add propylene glycol or other polyol during stirring. Heat the mixture up to 300ºC in a water bath. Heat the water up to 300ºC in a separate vessel and add to the mixture and then stir it for 5 min in a covered vessel. The vesicle size of ethosomal formulation can be decreased to desire extend using sonication[11] or extrusion method[12]. Finally, the formulation should be properly stored under refrigeration.

STABILITY STUDIES

A) Stability studies for Ethosomal cream and gel

Percent entrapment

The optimized ethosomal formulation was kept in sealed vials (10 ml) at 5±3ºC and at 25±2ºC for 1, 2 and 3 months to study the effect of different storage conditions on percent entrapment.

B) Stability studies of ethosomal gel

1. Physical Appearance: Optimized gel was kept for 1, 2 and 3 months under 5ºC ± 3ºC as well as 25ºC ± 2 ºC temperature conditions to study the effect of storage conditions on their physical appearance.

2. Content Uniformity of Gel

The uniformity of drug content in ethosomal gel formulation was evaluated in triplicate. For this investigation ethosomal gel (1.0g) was kept in a beaker containing 1000 ml of phosphate buffer pH (7.4) containing SLS 2.5%w/v for 48 h on magnetic stirrer. Solution was filtered and analyzed by UV spectrophotometer at λmax 290nm.

Table 3: Methods for the Characterization of Ethosomal Formulation[13]

Parameters	Methods	References
Vesicle size and size distribution	transmission electron microscopy (TEM) scanning electron microscopy (SEM)	[14,15]
Entrapment efficiency	Mini column centrifugation method Fluorescence spectrophotometry	[16]
Vesicle shape (morphology)	Transmission electron microscopy Scanning electron microscopy	[17, 18]
Vesicle Skin interaction study	Confocal laser scanning microscopy Fluorescence microscopy Transmission electron microscopy Eosin-Hematoxylin staining	[19, 20]
Phospholipid-ethanol interaction	^{31}P NMR Differential scanning calorimeter	[21, 22].
Degree of deformability	Extrusion method	[23, 24]
Zeta potential	Zeta meter	[25]
Turbidity	Nephalometer	[25]
In vitro drug release study	Franz diffusion cell with artificial or biological membrane, Dialysis bag diffusion	[25-26]
Drug deposition study	Franz diffusion cell	[23, 24]
Stability study	Dynamic light scattering method	[27]

THERAPEUTIC APPLICATION OF ETHOSOMES

1. In the treatment herpetic infection- 5% acyclovir ethosomal preparation compared to the 5 % acyclovir cream showed significant improvements in treatment of herpetic infections.

2. Transcellular Delivery- **Ethosomes** as compared to the marketed formulation suggested ethosomes to be an attractive clinical alternative for anti-HIV therapy[28].

3. Ethosomes are used in pilosabeceous targeting. Ethosomes, the high ethanol containing vesicles are able to penetrate the deeper layers of the

skin and hence appear to be vesicles of choice for transdermal drug delivery of hydrophilic and impermeable drugs through the skin.

4. Transdermal Delivery of Hormones. Oral administration of hormones is associated with problems like high first pass metabolism, low oral bioavailability and several dose dependent side effects. The risk of failure of treatment is known to increase with each pill missed.

5. Delivery of Anti-Arthritis Drug Topical delivery of anti-arthritis drug is a better option for its site-specific delivery and overcomes the problem associated with conventional oral therapy.

CONCLUSION

Transdermal route is promising alternative to drug delivery for systemic effect. Ethosomes has initiated a new area in vesicular research for transdermal drug delivery. Ethosomes are soft, malleable vesicles and possible carrier for transportation of drugs. Ethosomes are characterized by simplicity in their preparation, safety and efficacy and can be tailored for enhanced skin permeation of active drugs. Ethosomes have been found to be much more efficient at delivering drug to the skin, than either liposomes or hydro-alcoholic solution. Ethosomes have been tested to encapsulate hydrophilic drugs, cationic drugs, proteins and peptides. Ethosomal carrier opens new challenges and opportunities for the development of novel improved therapies.

REFERENCES

1. Schreier H and Bouwstra JA. Liposomes and niosomes as topical drug carriers: dermal and transdermal drug delivery. J Control Rel. 1994;30:1– 15.
2. Anitha P, Ramkanth S, Uma Sankari K, Alagusundaram M, Gnanapraksah K and Devaki Devi P. Ethosomes - A noninvasive vesicular carrier for transdermal drug delivery. Int. J. Rev. Life. Sci 2011;1(1):17-24.
3. Akiladevi D and sachinandan basak. Ethosomes a noninvasive approach for transdermal drug delivery. IJCPR 2010; 2: 1-5.
4. Pavan K,Radhika PR and Sivakumar T. Ethosomes-A Priority in Transdermal Drug Delivery. International Journal of Advances in Pharmaceutical Sciences 2010;1: 2-3.
5. Merdan VM, Alhaique F and Touitou E. Vesicular carriers for topical delivery. Acta Techno. Legis Medicament 1998; 12:1-6.
6. Upadhyay N, Mandal S, Bhatia L, Shailesh S and Chauhan P. A Review on Ethosomes: An Emerging Approach for Drug Delivery through the Skin. RRST-Pharmaceutics 2011; 3(7):1.
7. Touitou E, Dayan N, Bergelson L, Godin B and Eliaz M. Ethosomes-novel vesicular carriers for enhanced delivery: characterization and skin penetration properties. J. Control. Release2000; 65:403-418.
8. Tiwari RK, Chauhan NS and Yogesh s. ethosomes: a potential carries for transdermal drug delivery, International Journal of Drug Development & Research. 2010; 2[4]:2-4.
9. Manosroi A, Jantrawut P, Khositsuntiwong N, Manosroi W and Manosroi J. Novel Elastic Nanovesicles for Cosmeceutical and Pharmaceutical Applications. Chiang Mai. J. Sci. 2009; 36(2): 168-178.
10. Touitou E, Dayan N, Bergelson L, Godin B and Eliaz M. Ethosomes-novel vesicular carriers for enhanced delivery: characterization and skin penetration properties. J. Control. Release 2000; 65:403-418.
11. Jain S, Umamaheshwari RB, Bhadra D and Jain NK. Ind J Pharm. Sci 2004; 66(1):72-81.
12. Verma DD and Fahr A. Synergistic penetration effect of ethanol and phospholipids on the topical delivery of

Cyclosporine A. J. Control Release. 2004; 97: 55-66.
13. Touitou E, Dayan N, Bergelson L, Godin B and Eliaz M. Ethosomes - novel vesicular carriers for enhanced delivery: characterization and skin penetration proper-ties. J Control Rel 2000; 65: 403-18.
14. Bhalaria MK, Naik S and Misra AN. Ethosomes: A novel delivery system for antifungal drugs in the treatment of topical fungal diseases, Indian Journal of Experimental Biology 2009; 47: 368-375.
15. Touitou E and Nava Dayan. Carriers for skin delivery of Trihexyphenidyl HCl: ethosomes vs liposomes. Biomaterials. 2000;21:1879-1885.
16. Fry DW, White JC and Goldman ID. J. Anal. Biochem. 1978; 90:809.
17. Jain S,Umamaheswari RB, Tripathi, P and Jain NK. Ind. J. Pharm. Sci. 2003; 65(3): 223-231.
18. Touitou E, Godin B and Weiss C. Drug Develop Research 2000; 50: 406-415.
19. Simonetti O, Hoogstraate J, Bodde HE. Arch. Dermatol. Res.1995;287, 465.
20. Honeywell-Nguyen, PL, Graaff D, Anko M, Groenink HW and Bouwstra JA. Biochim. Biophys. Acta. 2002:1573, 130-138.
21. Touitou E, Dayan N, Bergelson L, Godin B and Eliaz M. J Control Release. 2000; 65: 403-418.
22. Maghraby E, William AC GMM and Barry BW. Int J Pharm 2000; 196: 63-74.
23. Jain S, Jain P and Jain NK. Drug Dev. Ind Pharm. 2003;29(90):1013-1026
24. Jain S, Jain N, Bhadra D, Tiwary AK and Jain NK. Current Drug Delivery 2005;2(3): 222-233.
25. Dayan N and Touitou E. Biomaterials 2000; 21: 1879-1885.
26. Jain S, Umamaheshwari RB, Bhadra D and Jain NK. Ind. J. Pharm. Sci. 2004; 66(1):72-81.
27. Touitou E, Dayan N, Bergelson L,Godin B and Eliaz M. J. Control. Release 2000; 65: 403-418.
28. Jain S. Vesicular approaches for transdermal delivery of bioactive agent. PhD thesis Dr. H.S. Gour University Sagar India 2005
29. Verma P and Pathak k. Therapeutic and cosmoceutical potentials of ethosomes: An overview JAPTR 2010;(1):2-3