Development and Evaluation of Antifungal Niosome of Luliconazole and Salicylic Acid

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Abstract— Objective: The main purpose of the study was to develop niosome of luliconazole and salicylic acid with increase the residence time of drugs in the skin stratum corneum layer.
Method: Antifungal niosome prepared by reverse phase evaporation method by using span 60 and 40 and cholesterol from which the best formulation was selected and characterize in term of vesicle shape, vesicle size, entrapment efficiency and in-vitro release study.
Result: The niosome size range of prepared formulation 6 µm (MLV) by using reverse phase evaporation method respectively. The entrapment efficiency of luliconazole and salicylic acid was determined by separation of entrapped drug by cold centrifugation method. The entrapped efficiency of the drug was found to be 95.19-97.48 % by using Span 40 niosomes prepared. The release of the drug was also modified and extended over a period of 420 min in all formulations
Conclusion: The niosomal formulation for luliconazole and salicylic acid with improved antifungal activity, Stability and drug bioavailability.

Indexed Terms— Luliconazole, salicylic acid, Ethanol, Reverse phase evaporation method, Span60 and 40.

I. INTRODUCTION

Niosomes are the novel drug delivery system in which both hydrophilic and hydrophobic drug is encapsulated in a vesicle. Niosomes or non- ionic surfactant are microscopic lamellar structures formed by mixture of non-ionic surfactant of alkyl or dialkyl polyglycerol ether class and cholesterol with the hydration in aqueous media.[1] They are biodegradable, biocompatible and flexible. The size of niosome are microscopic and nanometric scale. The particle size ranges from small unilamellar vesicles (SUV) were about 10-100nm, large unilamellar vesicles (LUV) 100-3000nm and multi-lamellar vesicles (MLV) greater than 5µm. In niosome, the vesicular forming amphiphile are a non-ionic surfactant such as span60, span40 and stabilized by addition of cholesterol.[2]

This study was to formulation of antifungal activity of niosome as a transdermal drug delivery system for luliconazole and salicylic acid. Luliconazole is topical antifungal agent. It is class of imidazole derivative. It is indicated for treatment of athlete's foot, jock itch, ringworm. It is poorly water-soluble drug and has high permeability. These inhibit the enzyme lanosterol-demethylase. This drug is used in patient with tinea pedis, tinea cruris and tinea corporis.[3]

Salicylic acid is a beta hydroxyl acid. It has direct activity as an anti-inflammatory agent, anti-infective agent, antifungal agent and acts as a topical antibacterial agent due to its ability to promote exfoliation.[4]

Fungal infection refers to as mycoses which are occurring and a variety of environmental and physiological conditions can provide to the development of fungal diseases. The fungal infection of the skin is one of the common dermatological problems. Antifungal agents inhibited the spread of fungi by killing fungal cell and preventing their growth.[5-7]

II. MATERIAL AND METHODS

Materials: Luliconazole was procured from IQ-GenX Pvt.Ltd. Mumbai. Salicylic acid purchased from SDFCL fine-chem Ltd. Mumbai. Span 60,40 and cholesterol was procured from Ozone International Mumbai and all other chemical reagent used of analytical grade.

Method
Reverse Phase evaporation method
Accurately weighted quantities of surfactants and cholesterol were taken to give the desired ratio and were dissolve in 50 ml of ethanol in a round bottom

flask. Then, accurately weighted amount of drug was added to the solvent (ethanol). The solvent was evaporated in a rotary flash evaporator at temperature of 60°C at 120 rpm until the smooth, dry lipid film was hydrated with 20 ml of PBS 7.0 was added and shaking on the water bath. The niosome suspension was formed at this kept at 2 to 8 °c for 24 hrs.

Table 1: Composition of niosomal suspension

Sr. No	Formulation Code	Surfactant Used	Weight taken (in gm)				Surfactant : Cholesterol Ratio
			Drugs		Surfactant	Cholesterol	
			LNZ	Salicylic acid			
1.	F1	Span 60	1	1	10	2	5:1
2.	F2		1	1	20	2	10:1
3.	F3		1	1	30	2	15:1
4.	F4	Span 40	1	1	10	2	5:1
5.	F5		1	1	20	2	10:1
6.	F6		1	1	30	2	15:1

Preliminary studies:

Infrared Spectroscopy:

The IR spectrum of pure Luliconazole and Salicylic acid API analyzed in the range 4000 to 557 cm^{-1}.

Luliconazole:

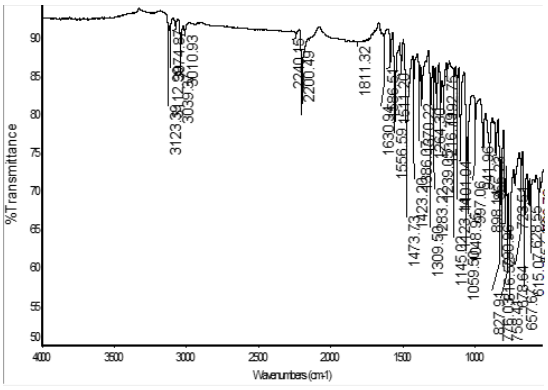

Figure 1: IR Spectra of Luliconazole

Salicylic acid:

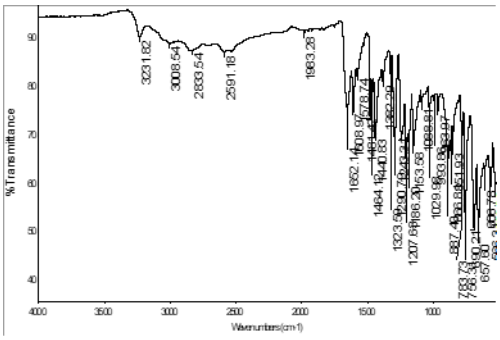

Figure 2: IR Spectra of Salicylic acid

Evaluation of Niosome[8-10]

Vesicle shape and Morphology

Shape of niosomal formulation and drug loaded niosomal formulation was determined by electronic microscope and results was shown in figure 3.

Vesicle size of niosomes

Vesicle size of formulation was determined by Nanoparticle analyzer (HORIBA).1ml of niosomal formulation (F5) diluted with ethanol for 10 ml and kept for 15 min sonication. Then the sample was analyzed and particle size determined.

Entrapment Efficiency

Entrapment efficiencies of niosomal formulation was determined by cold centrifugation method. Where the niosomal dispersion was centrifuged at 7000 rpm for 50 min. The clear supernatant layer was separated and diluted using methanol. The determined the entrapped drug spectrophotometrically. The percentage of

entrapment efficiency was calculated by using following equation

Entrapment Efficiency= Amount of drug taken – amount of drug in supernatant/ amount of drug*100

In-vitro drug release study

In-vitro drug release from niosomal formulation by using Franz diffusion cell and semipermeable cellophane membrane. Added niosomal dispersion equivalent to 20 mg into the diffusion tube having diameter 2.5cm which acted as donor compartment and covered with cellophane membrane. The glass tube was placed in a beaker containing 20 ml methanol and 80 ml of phosphate buffer 7.0 which acts as receptor compartment. The temperature was maintained at 37 ± 0.5 °C and stirred at 100 rpm speed using magnetic stirrer. Samples were withdrawn from the receptor compartment at specified time interval. Each time immediately after the removal of sample, the medium was replaced with fresh 7.0 phosphate buffer. The sample were analyzed at 297 nm in Uv-visible spectrophotometer.

III. RESULT AND DISCUSSION

Niosome of Luliconazole and salicylic acid were prepared by Reverse phase evaporation method using cholesterol and span 60 and 40. Niosome were evaluated for vesicle shape, size determination, Entrapment efficiency and in-vitro drug release study.

Vesicle shape

Optical microscope for the selected formulation F5 was carried out. The result was shown in figure3.

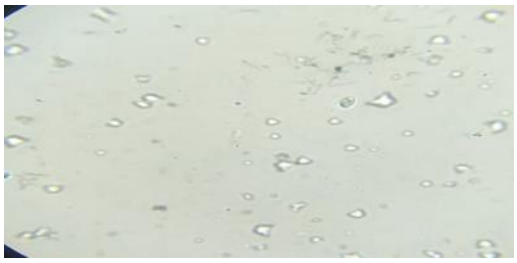

Figure 3: Optical images of niosomal formulation

Vesicle Size

The mean vesicular diameter of formulation was evaluated by using HORIBA Nanoparticle analyzer. The formulation, F5 (with span 40) showed a mean vesicular diameter of 1.1 nm (Figure 4)

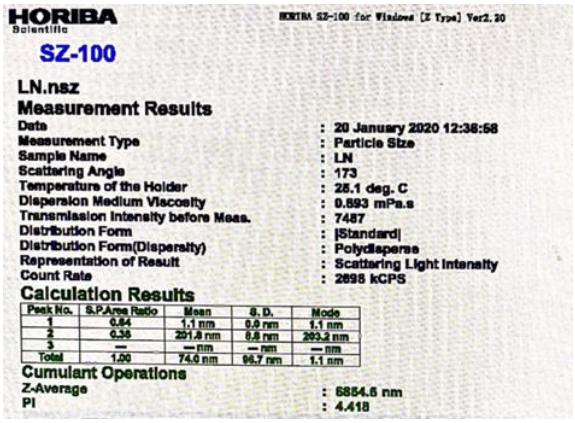

Figure 4: Mean vesicular diameter of formulation F5 (Span 40)

Entrapment Efficiency:

The prepared niosome Formulations F1, F2, F3, F4, F5 and F6 were evaluated for entrapment efficiency (table 2). The percentage of entrapment efficiency of luliconazole and salicylic acid was found to be 97.48 and 95.23%.

Table 2: Percentage of entrapment efficiency of niosomal formulations

Sr.No.	Formulation code	% Entrapment Efficiency	
		Luliconazole	Salicylic acid
1	F1	28.15	27.12
2	F2	44.73	47.31
3	F3	93.56	94.89
4	F4	31.59	49.31
5	F5	97.48	95.19
6	F6	67.51	55.94

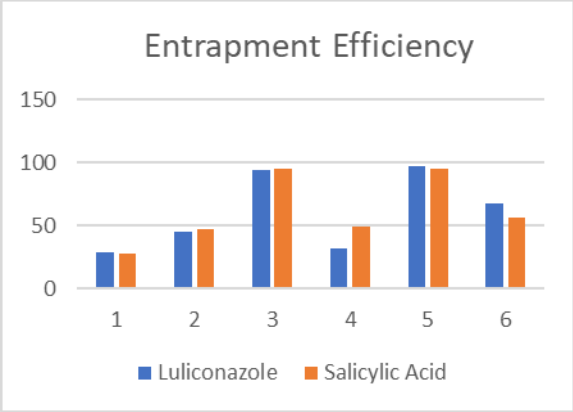

Figure 5: Percentage entrapment efficiency of niosome formulations

In-vitro Drug release study

The drug release from the niosome was determined by using Franz diffusion cell shown in figure 6. The percentage drug release, after 420 min was found to be 98.33 to 99.67%. The result of in vitro drug release study of niosome are given in (Table 6).

Table 6: Drug release profile of niosomal formulation

Time (min)	Luliconazole	Salicylic Acid
0	0.00	0.00
30	19.07	15.67
60	31.38	22.10
120	41.25	35.69
180	50.09	46.79
240	62.34	56.56
300	77.71	69.02
360	88.91	88.35
420	98.33	99.67

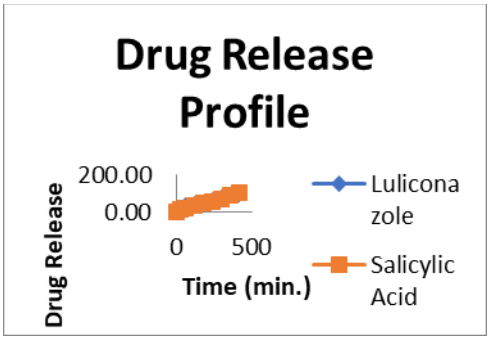

Figure 6: Percentage drug release profile of niosomal formulation

IV. CONCLUSION

This study indicates the all formulations prepared by reverse phase evaporation method. As niosome are made of non-ionic surfactant so these are more stable, safe by using Span 60, Span 40 and cholesterol. F5 formulation was found to be best vesicle size (6 µm), entrapment efficiency (95.19-97.48 %) and in-vitro release study (98.33-99.67 %) at the end of 420 min. The properties of niosomes are affected by methods of preparation, drug properties, amount, structure and type of surfactant used, cholesterol content.

V. ACKNOWLEDGEMENT

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