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Research Article

FORMULATION AND EVALUATION OF OXICONAZOLE BASED NIOSOMAL GEL FOR THE EFFECTIVE FUNGAL TREATMENT

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ABSTRACT

Niosomes are significant towards drug delivery potential and increase drug efficacy as compared to other form of drug delivery. Niosomes are highly stable and cost effective towards the treatment of targeted drug delivery. Therapeutic efficacy of the drug molecules can be improved by prolonging the release rate of active drug in systemic circulation and also protecting the drug from biological environment. Niosome appears to be a well preferred drug delivery system and potential for targeted delivery in anti-cancer, antifungal agents etc. This article focuses on to formulate oxiconazole based niosome gels for antifungal infection. Different types of polymers have been used to prepare oxiconazole based niosome gels. Drug polymer compatibility study carried out by FTIR and DSC techniques. Lipid hydration method is used for the preparation of multilamellar vesicles of niosomes.TEM and SEM analysis technique is used to confirm the particle size, shape and morphology of the prepared niosome gel. Nano sized niosomes easily penetrate through skin pores and polyethylene glycol used as penetrating enhancer. Formulated niosome gels subjected for evaluation study. Dissolution study confirms that formulation N15 has shown better result in controlling oxiconazole up to 98% in 12h. The kinetic release study follows non fickian behavior. Formulation N15 subjected for ex vivo diffusion study and shows optimum release rate whereas in vivo study with commercially available oxiconazole gel reveals better results with formulated niosome gel as it transforms the infected part of the skin like a normal skin

Keywords: Antifungal; Oxiconazole; Skin Infection; Niosomes.

INTRODUCTION

The frequency and the change in growth of fungal infection have increased gradually over last few decades. Similarly the treatment procedure and surgical care as well as new drug delivery system are more potent against the fungal growth[1]. The fungal cell wall contains chitin and polysaccharides which make the outer cell very rigid and act as a barrier to prevent the penetration of the drug to the cell. Fungi cell contain ergosterol which control the efficacy of the drug and reduce its potency. Fungal infection is considered as one of the major cause of human disease[2] can leads to systemic infection in a chronic condition[3-4]. Candidiasis, cryptococcal meningitis and invasive aspergillosis are also considered as one of the life threatening fungi infection[5]. In case of eukaryotic fungi there are very less drugs are available which can kill the pathogen due to the very close evolutionary relationship with the host[6]. Novel antifungal drugs are directly targeted sterol component of cell membrane of fungi either depleting the ergosterol or inhibiting the synthesis[7]. Due to high antifungal resistance of the pathogens our research has been focused on the preparation of novel antifungal compounds. For the development of novel antifungal drug first we have to focus on the various important factors of infection and their basic mechanism, could be a key factor to develop a new antifungal drug for targeted area.

Like other imidazole antifungal, oxiconazole (OXZ) can upgraded film vulnerability to zinc, augmenting its cytotoxicity[8]. It is a biopharmaceutical gathering structure Class II quiet having low liquid dissolvability and poor essential maintenance. OXZ has an expansive fungicidal or fungistatic movement against various pathogenic organisms including candida albicans. Its antifungal action is because of the restraint of the ergosterol biosynthesis, which is basic for cell film uprightness. Due to its low water-soluble nature it shows low bioavailability which confines its antifungal productivity [9-14]. Niosome formulation could be a positive approach towards enhancing the solubility of OXZ. Thus, extraordinary strategies are utilized to upgrade the dissolvability of this ineffectively water solvent medication which incorporates utilization of surfactants, co-surfactants, co-solvents, etc[15].

Niosomes are non-phospholipids vesicular contrasting options to liposomes. They are nonionic surfactant vesicles or surfactant layer vesicles. The sizes of niosomes are infinitesimal and lie in nanometric scale. The molecule measure ranges from 10nm-100nm. Niosomes gel provides a better carrier for OXZ as it builds their dissolvability and offers a controlled discharge due to the polymers such as HPMC, Sodium alginate, chitosan and carbopol at different concentrations in the formulations. Polymer composition with OXZ at different ratios could be a useful carrier for prolonging the release rate of active drug. Niosomes gel could have the property of greaseless, better spreadability, thermally stable, stable in plasma drug concentration, more stable and compliance[16].

MATERIALS AND METHODS

Reagents and Chemicals

Oxiconazole and hydroxy propyl methylcellulose (HPMC K4M) were obtained from nice chemical laboratory, India. Chitosan and sodium alginate analytical research grade was purchased from Sigma Aldrich and used as received. Similarly carbopol and all other excipients were of analytical research grade and used as received from Shipra chemicals, India.

Detail of Instrumentation

Analytical weighing balance [ContechA224,India], pH meter[Elico LI120,India], UV visible spectrophotometer [1800, Shimadzu Corp., Japan], FTIR [Bruker Alpha, India], DSC [Q 1000 DSC System [TA Instruments, USA], Magnetic stirrer with Hotplate[Shimadzu Mumbai, India], Franz Diffusion cell Apparatus [EMFDC 06 Orchid scientific, Nasik], Scanning Electron Microscope[Hitachi, Tokyo, Japan], Transition Electron Microscope [Hitachi, Tokyo, Japan]

Formulation of Niosome gel

Lipid hydration method is used for the preparation of multilamellar vesicles of niosomes[17] shown in Table 1. Weight amount of polymers (such as HPMC, Sodium alginate, chitosan and carbopol) at different concentration has mixed with oxiconazole solution prepared in organic solvent such as ethanol in a round bottom flask. Rotary evaporator has been introduced to remove the organic solvent from the prepared sample at room temperature. A thin layer of solid mixture deposited at the bottom of the flask. The thin film was collected and hydrated with aqueous phase with gentle agitation. The obtained product is in the form of niosome and store properly for further evaluation test.

PHYSICOCHEMICAL EVALUATION

Entrapment Efficiency

The entrapment efficiency of the antifungal drug oxiconazole is determined by UV Visible spectrophotometer. This technique is used to find out the drug content in the niosome. Prepared gel has been diluted into 10 ml of methanol with proper stirring by using magnetic stirrer. A homogeneous solution has been obtained and kept for centrifuge. Centrifuge was carried out at 1200 rpm for 30 min. the supernatant liquid has been analyzed under UV Visible spectrophotometer at 296 nm with suitable dilution. The EE was calculated by formula mention below.

% Entrapment efficiency = (Amount of drug entrapped / Amount of drug added) X 100

Percentage Yield

It is calculated to determine whether the drug entrapment facing startling polymer was efficient. The product results expect close to measuring final output which should compare with the raw materials weight.

Percentage yield = (Practical yield / theoretical yield) X 100

Drug Content

Prepared gel was weighed up to 1gm and diluted with buffer sample pH 6.8 and make the volume up to 50 ml. From the solution 5ml was pipetted out in 25 ml volumetric flask, and the volume was made up utilizing phosphate buffer pH 6.8. The absorbance was estimated under UV Visible spectrophotometer at 296nm. Medication content was calculated by utilizing standard curve of the oxiconazole.

pH

The pH of various formulations niosome was evaluated by using digital pH meter. It was calibrated before use. The measurement of pH of each formulation was done in triplicate, and average were calculated.

Viscosity

Brookfield viscometer is used to measure the viscosity of the prepared gel. The gels were poured in a beaker and rotated at 50 rpm, and the corresponding reading shown on the viscometer was noted. The viscosity of the gel was obtained by Brookfield viscometer. The viscosity was measured in cps. Experiments were carried out in triplicates for all the formulations in case of niosome.

Spreadability

Prepared gel present in the form of niosome was taken for spreadability test. Approximately 350mg of gel was weighed, and then, applied on the glass plate to determine the spreadability of the gel. Another glass plate was dropped at the height of 5 cm to the previously applied glass plate. After one minute the diameter of circle was measured and the test as performed in triplicate, and average values were calculated.

SEM Analysis

Scanning electron microscopy (SEM) was conducted to analyze the surface morphology of niosomes. SEM analysis can also use to determine shape of the formulated niosomes. A drop of formulated gel was mounted on clear glass stub, air dried and visualized under SEM.

In-Vitro Drug Release

Franz diffusion cell of vertical form is used to determine in vitro drug release study. From each formulation of niosomes 3 mg of freshly prepared gel was spread on the donor side of the cellulose nitrate membrane grade 110 (each sample done in a triplicate manner). The cellulose membrane soaked with isopropyl alcohol to make it more hydrophobic. In receptor vessel 1litre of saline phosphate buffer (pH-7.4) with methanol was used. The study was carried out at 37 ±0.5°C temperature and the speed of agitator maintained as 400rpm for 12h. After a regular interval of time 5ml sample was collected and replaced with the same buffer solution. Collected samples were marked and kept it for analysis under UV Visible spectrophotometer. A suitable dilution has been made for each sample and concentration was measured at 296nm. The obtained result reveals that % of drug release at regular interval of time from the prepared gel.

Release Kinetic Studies

The release kinetic study of oxiconazole based niosome has been conducted by using dissolution profile[18]. The kinetic study were evaluated by the following equation mentioned below.

- Zero order: $M_t = M_o + K_o t$
- First order: $\ln M_t = \ln M_o + K_1 t$
- Higuchi model: $M_t = K_H \sqrt{t}$
- Korsmeyer–Peppas model: $M_t/M_o = K_k t^n$

Where M_t is the amount of drug dissolved at time t, M_o the initial amount of drug, $K_1$is the first order release constant, K_0 the zero order release constant, K_H the Higuchi rate constant, K_k the Korsmeyer–Peppas model release constant and n is the diffusional release exponent indicative of the operating release mechanism. The correlation coefficient (R^2) value was used as an indicator of the best fitting, for each of the models considered.

Ex-Vivo Diffusion Studies for Best Formulation

A male healthy albino rats weighing 150-180 g were sacrificed[19] for abdominal skin. The best formulation in the form of gel was applied on the animal skin. 3mg of gel of oxiconazole drug was applied through donor compartment on the animal skin. Similarly marketed gel (MR) of oxiconazole of 3mg was taken and applied through the donor compartment on other diffusion cell. In both the diffusion cells, reservoir compartment was filled with 10 mL of methanol and 40 mL phosphate buffer solution (pH 7.4) at 37±0.5°C at 400 rpm/min for 12 hrs. Samples were withdrawn from reservoir compartment at regular interval of time and the amount of drug release was determined by UV Visible spectrophotometer at 296nm after suitable dilution. Each time the reservoir compartment was replenished with the same quantity of fresh phosphate buffer solution (pH 7.4). Oxiconazole drug content was estimated prepared in gel form and similarly marketed gel was calculated and reported. All the studies were conducted according to the ethical guidelines of CPCSEA after obtaining necessary clearance from the committee (Approval No: 1688/PO/E/2013/CPCSEA).

In-Vivo Studies for Best Formulation

Preparation of Microorganism

Sabouraud dextrose agar media is used for the growth of the fungus such as Candida albicans by culture for 48h at 30°C. The cells were collected and hydrated with sterile saline to obtain a final microorganism (concentration of 107 CFU/mL) which is used as a causative organism for skin infection in animal[20].

Preparation of the Animal

The rats were divided into 4 groups of 6 animals each.
Group 1: Serves as negative control without fungal infection
Group 2: Serves as positive control induced by fungal infection
Group-3: Serves as test group treated with niosome gel (best formulation)
Group-4: Serves as standard with marketed drug (Oxistaj cream 1%)

All rats were kept under observation during the experiment to make sure any type of clinical features due to fungal infection. Symptoms can be identified as rashes, redness of skin, white discharge, cracking of skin and pimples filled with puss were observed and noted as evidence for skin infection. As time passed the evidence has been recorded in the form of photos and compared between different groups of rats and observed the activity of our best formulation (Niosome based gel) and its control on fungal skin infection. Blood samples were collected in a blood collection tube containing K2EDTA as an anticoagulant, from the tail end of each group of rats. The collected blood samples were subjected for centrifuge at 4°C at 5000 rpm for 5 minutes to separate plasma. The collected plasma stored properly for further investigation of pharmacokinetic parameters such as Cmax, Tmax, AUC 0-t Ka, KE and Vd.

RESULTS AND DISCUSSION

FT-IR study

The drug-polymer mixtures were taken, and their agreeableness schedule was performed. This is to set up that other suspenseful therapeutically active cure has not passed through any physicochemical change after it has been subjected to the processing steps during formulation. This may be accustomed on anticipating out the following studies like FTIR. The FTIR spectra of oxiconazole and oxiconazole with HPMC were shown in the Figure 1 and 2.

The FTIR spectral analysis report reveals that oxiconazole has shown its characteristic peaks without any shifting and broadening with the combination of HPMC polymer (similar results obtained with other polymers). From the above results it is concluded that the absorption peaks of oxiconazole remain unchanged in drug-polymer admixture which indicates that there is not any prominent chemical reaction between oxiconazole and the polymers used in the formulation of niosome gel.

DSC study

DSC techniques were used to study the compatibility on the active drug such as oxiconazole, different polymers and their compositions. DSC curve of the pure drugs was compared with1:1 ratio physical mixtures. Thermal sphere of the blends i.e. melting point, the absence of a substantial shift in sudden liquefying point or absence in the display coming from new exothermic/endothermic peak in the blend indicated agreeableness in the middle medicate as well as polymers. Moreover, slight changes in the peak shape, height and width could be the indication of incompatibility. DSC curve of pure oxiconazole, polymers and the complex of drug and polymers has been represented in Figure 3

DSC results reveal that there are no sharp endothermic peaks were detected in furtherance of drug polymers mixture confirms that the polymers used in formulation is compatible with oxiconazole.

EVALUATION STUDY OF NIOSOMES GEL

Drug Entrapment Efficiency

Table 2 shows the drug entrapment efficiency of niosomes containing oxiconazole as an antifungal drug. It provides the data that how much volume of drug entrapped in the prepared formulations. It was observed that 78% to 95% of drugs were entrapped. A larger than involvement was once noticed in spite of formulations upon reducing the particle magnitude tense net appear area of the particles increases. Furthermore thus, a most area appear on the part of medicate entrapment, though in pursuance of longer particles with shorter transpire shows a better result of entrapment rather than adhesive medication. The greater fluidity of ethanol also played an important role to increase the entrapment efficiency. Due to the above reason the amount of ethanol kept constant in each formulation.

Percentage Yield

In the course of the formulation of niosomes the proportion give way executed afterwards the complete deal with used to be one hundred. The percentage of yield is a relation between practical yield and theoretical yield. The percentage yields from the formulations lies in-between 70% to 95%. All these observations values are displayed in Table 2.

Drug Content

The percentage drug content of all the formulations was found to be in the range of 86% – 96% shown in Table 2. The highest drug content was found in the optimized formulation N15 containing 1:2 ratio of oxiconazole with CP.

pH

Skin compatibility is the major requirement for a good topical formulation. It was found that the pH of all the niosome and niosome gel formulations was in the range of 6.51–7.34 that suits the skin pH indicating the skin compatibility shown in Table 2.

Viscosity

The viscosity ranged between 450 and 580 cps shown in Table 2. Low viscosity was found for the E1 and N1 formulation containing low molecular weight and lower concentration of CH, 1:1 ratio of oxiconazole and CH polymer. CH is low viscosity grade of polymer as compare to other polymers such as HPMC, CP, SA.

Spreadability

The healing effect of formulation depends on its spreading coefficient. The value of spreadability of all niosomes gel formulations ranged from 4 to 5.3 g cm/s shown in Table 2. Spreadability depends on the viscosity and gelling property of the polymers used in the formulation. The formulation N15 having highest viscosity 580.04cps has high spreading coefficient of 5.3 g cm/s, and the formulation N1 has lesser spreading coefficient of 4 g cm/s as its viscosity is 450.09 cps.

SEM Analysis

The front design as well as shape of oxiconazole loaded niosome gel has been analysed by using SEM. Formulation N15 shows smooth surface observed in the image reveals complete removal of the solvent from the formulation, and it also indicates particles size ranges from 50 nm to 150nm. The SEM image of formulation N15 has found spherical in shape and observe as separated entity represent in Figure 4.

In-Vitro Drug Release

The dissolution investigation was performed in a triplicate way by utilizing the diffusion medium Phosphate buffer with the pH 7.4. The percentage of drug release for all formulations of niosomes prepared gel ranged from 90% to 98% at the end of 12 h. Maximum drug release in a sustained manner was observed in the formulation N15 after 12 h. The reason for maximum release may be due to the concentration and the viscosity grade of polymers. High viscosity grade of polymers or having gelling nature of polymer could be a useful property for topical formulation to retain the drug molecule for long time and provide a steady plasma drug concentration. HPMC[21] and CP both have gelling property and shown better controlled release as compare to CH polymer. In between HPMC and CP, CP shows better viscosity than HPMC. CP did not disintegrate rapidly due to higher viscosity grade, which could be a barrier for aqueous buffer solution and can easily sustain the release of the active drug. In some cases, the excess viscosity of CP hold the active drug and shows incomplete release. Reports confirm that 1:2 ratio of drug and polymer respectively used for the controlled drug delivery system. CH is a natural and low viscosity grade of polymer [22] which could not able to control the release rate of antifungal drug for optimum time period. Due to this reason CH based formulations having less control on oxiconazole drug release. In few formulations it was observed that if concentration increases drug release decreases that means drug molecule has been retarded in the formulation and the final percentage of drug release decreases. Oxiconazole % of drug release from all the formulations (N1 to N16) in the form of niosomes gel shown in Table 3 and Figure 5.

Kinetic Studies for Niosomes Gel

Keeping in mind the end goal to decide the correct system of medication discharge from the formulation, the in-vitro dissolution studies was assessed by zero order, first order, Higuchi, and Peppa's equations. The standard of picking the most proper model was in accordance with highest R^2 value as the best fit. The results are shown in Table 4. The free up illustration data was determined from peppa's plot shown non-fickian release that implies release rates happened by diffusion release of the gels. If n value is less than 0.5, it shows fickian diffusion release, and if n value is between 0.5 and 0.89, it follows nonfickian (anomalous) behavior, i.e., drug release is both diffusion and erosion-controlled mechanism observed in niosomes formulations N1 to N16.

Ex-Vivo Diffusion Studies for Best Formulation

The ex *vivo* study shows that the percentage of drug released from the niosome formulated gel from the formulation N15 was 94.5% drug release after 12 h. This could be due to the niosome based formulation. SEM analysis also reveals that particle size is in nano range. Niosome in nano range as a gel formulation N15 control the release rate for longer period of time as compare to other formulations due to the addition of different type of polymers. The study confirmed significant difference in the drug release and drug content. Figure 6 showed that niosome gel based formulation when compared with marketed cream (Oxistaj cream 1%). The amount of drug released from marketed cream was 93.9% which is less as compare to niosome gel i.e. 94.5% shown in Table 5. Transdermal flux value for niosome gel found to be 158.91 µg/cm^2/hr and marketed cream showed 141.22 µg/cm^2/hr. This data confirms that niosome gel N15 reside at targeted site for a longer period of time as compare to marketed product and improved patient compliance was observed. Similarly, the percentage of drug retention in niosome showed better result as compare to the marketed cream. The results obtained for *ex vivo* studies are closely equal to the values of *in vitro* studies. From the study it concluded that oxiconazole gel in the form of niosomes combined with hydrophilic polymer carbopol, exhibit better control in drug release and also form a good consistency of gel for topical application in fungal infection.

In-Vivo Studies for Best Formulation

Candida albicans commonly used for the evaluation of antifungal activity[23]. Animals used for the application should be healthy and free from inflammation cracking, edema, puss formation etc. once animal induced for fungal infection animal checked for occurrence of changes happen such as edema, patches, inflammation, color change of the skin, redness and skin irritation etc. Infected animals were treated by oxistaj cream (1%) and observed that edema, redness and inflammation slowly disappeared except some white spot and scars. The application of oxiconazole based niosome gel showed better results as it transforms the infected part of the skin like a normal skin with slight redness, the study represents in figure 7. The in vivo pharmacokinetic parameters like Cmax, Tmax, AUC $_{0\text{-}t}$Ka, K_E, $V_d$were found to be comparable with marketed Oxistaj cream 1% reported in Table 6.

Table 1: Formulation of oxiconazole containing niosomes gel (%)

Formulation	Drug (%)	CH (%)	HPMC (%)	SA (%)	CP (%)	Soya lecithin (%)	Ethanol (%)	Polyethylene glycol (%)	Distilled water(q.s)
N1	1	1	-	-	-	3	45	2	q.s
N2	1	1.5	-	-	-	3	45	2	q.s
N3	1	2	-	-	-	3	45	2	q.s
N4	1	2.5	-	-	-	3	45	2	q.s
N5	1	-	1	-	-	3	45	2	q.s
N6	1	-	1.5	-	-	3	45	2	q.s
N7	1	-	2	-	-	3	45	2	q.s
N8	1	-	2.5	-	-	3	45	2	q.s
N9	1	-	-	1	-	3	45	2	q.s
N10	1	-	-	1.5	-	3	45	2	q.s
N11	1	-	-	2	-	3	45	2	q.s
N12	1	-	-	2.5	-	3	45	2	q.s
N13	1	-	-	-	1	3	45	2	q.s
N14	1	-	-	-	1.5	3	45	2	q.s
N15	1	-	-	-	2	3	45	2	q.s
N16	1	-	-	-	2.5	3	45	2	q.s

Table 2: Evaluation study of oxiconazole containing niosomes gel

Formulation	Drug entrapment efficiency(%)	Percentage yield (%)	Drug content (%)	pH	Viscosity(cps)	Spreadability (g cm/s)
N1	78.12±0.92	87.52±1.81	86.81±1.82	6.31±0.03	450.09±1.08	4±0.19
N2	80.19±1.63	88.21±1.28	88.21±1.94	6.51±0.09	469.86±12.24	4.24±0.24
N3	83.90±1.96	90.01±1.38	89.81±1.07	6.68±0.34	487.01±12.27	4.52±0.01
N4	89.70±1.78	92.11±1.17	91.32±1.17	6.91±0.01	518.66±11.09	4.71±0.37
N5	89.97±1.99	92.71±1.03	88.21±1.73	6.8±0.31	528.88±10.39	4.48±0.19
N6	91.91±2.72	89.71±0.73	89.73±0.19	6.09±0.47	542.72±10.16	4.83±0.27
N7	93.03±1.42	78.91±1.01	93.18±1.74	6.73±0.02	571.31±12.06	4.86±0.89
N8	94.02±1.01	72.81±1.28	94.38±1.09	6.82±0.14	576.47±12.03	5.2±0.37
N9	84.32±1.27	94.08±1.15	87.77±2.18	7.1±0.03	489.09±13.83	4.81±0.17
N10	86.73±1.07	92.11±1.03	90.72±1.81	7.16±0.22	530.71±10.23	4.91±0.07
N11	92.91±1.43	88.71±1.73	92.81±1.32	7.21±0.04	547.63±10.81	5.04±0.81
N12	93.80±1.01	70.3±1.91	94.16±1.38	7.34±0.91	574.16±11.19	5.11±0.11
N13	90.51±1.91	77.26±0.38	88.71±1.37	6.75±0.92	529.71±11.81	4.83±0.28
N14	92.92±1.82	87.73±1.82	93.09±1.32	6.92±0.01	569.39±9.91	5.14±0.85
N15	95.01±1.14	95.05±1.79	95.72±0.21	7.31±0.01	580.04±10.18	5.3±0.01
N16	94.81±1.32	73.83±2.71	94.09±0.82	7.17±0.61	578.99±10.07	5.3±0.07

Table 3: In vitro dissolution profile for oxiconazole containing niosomes gel (Formulations N1-N16)

Formulation	60 min (1h)	120 min (2h)	180 min (3h)	240 min (4h)	300 min (5h)	360 min (6h)	420 min (7h)	480 min (8h)	540 min (9h)	600 min (10h)	660 min (11h)	720 min (12h)
N1	33.59	49.76	76.32	91.32	-	-	-	-	-	-	-	-
N2	29.62	43.90	69.84	81.53	94.81	-	-	-	-	-	-	-
N3	25.96	39.40	57.03	71.13	85.94	91.26	-	-	-	-	-	-
N4	21.85	33.46	49.28	67.96	81.44	90.91	-	-	-	-	-	-
N5	29.58	42..80	67.98	86.54	97.10	-	-	-	-	-	-	-
N6	27.21	38.32	56.33	69.31	82.16	93.27	-	-	-	-	-	-
N7	26.34	35.76	53.26	66.39	78.91	85.21	92.11	-	-	-	-	-
N8	24.72	30.41	47.21	58.99	67.21	78.33	89.23	96.35	-	-	-	-
N9	41.39	56.57	79.33	90.32	-	-	-	-	-	-	-	-
N10	37.33	51.79	69.36	81.55	92.31	-	-	-	-	-	-	-
N11	34.11	49.27	61.71	78.33	89.21	96.32	-	-	-	-	-	-
N12	29.22	41.27	56.26	69.43	82.39	90.28	93.21	-	-	-	-	-
N13	21.72	29.23	42.18	55.32	69.19	77.21	87.35	95.01	-	-	-	-
N14	19.71	26.31	38.32	47.72	61.45	73.26	80.18	87.36	93.32	96.27	-	-
N15	15.32	22.37	32.37	45.78	57.98	65.33	72.32	83.76	89.91	93.32	96.29	98.38
N16	13.45	19.37	27.31	35.87	41.33	49.37	58.36	67.35	75.31	81.38	87.91	90.27

Table 4: Release kinetics of oxiconazole containing niosomes gel (Formulations N1-N16)

Formulation	R^2 Values					
	Zero order plots	First order plots	Higuchi plots	Korsmeyer-peppas plots		Order of release
				R^2	Diffusional exponent (n)	
N1	0.927	0.782	0.899	0.997	0.421	Diffusion
N2	0.933	0.879	0.876	0.988	0.439	Diffusion
N3	0.783	0.989	0.998	0.966		Diffusion
N4	0.911	0.971	0.891	0.993	0.937	Diffusion & Erosion
N5	0.875	0.847	0.908	0.985	0.7.32	Diffusion & Erosion
N6	0.971	0.781	0.971	0.998	0.89	Diffusion & Erosion
N7	0.892	0.878	0.981	0.996	0.78	Diffusion & Erosion
N8	0.871	0.981	0.991	0.998	0.821	Diffusion & Erosion
N9	0.827	0.981	0.979	0.991	0.321	Diffusion
N10	0.874	0.989	0.879	0.998	0.97	Diffusion & Erosion
N11	0.991	0.971	0.893	0.997	0.738	Diffusion & Erosion
N12	0.875	.977	0.879	0.983	0.82	Diffusion & Erosion
N13	0.978	0.997	0.871	0.998	0.728	Diffusion & Erosion
N14	0.891	0.899	0.921	0.999	0.891	Diffusion & Erosion
N15	0.926	0.971	0.998	0.998	0.881	Diffusion & Erosion
N16	0.776	0.877	0.971	0.993	0.731	Diffusion & Erosion

Table 5: Ex vivo diffusion studies for best formulation N15 and marketed cream

Time in hr	Sqrt time	Log Time	Market Sample		N15	
			% CDR	Log % CDR	% CDR	Log % CDR
1	1	0.1	5.1	0.707	6.5	0.812
2	1.414	0.301	11.3	1.053	14.2	1.152
3	1.732	0.477	12.4	1.093	19.2	1.283
4	2	0.602	20.3	1.307	29.2	1.465
5	2.236	0.698	29.5	1.469	35.7	1.552
6	2.449	0.778	38.7	1.587	47.2	1.673
7	2.645	0.845	48.8	1.688	56.4	1.751
8	2.828	0.903	57.7	1.761	65.5	1.816
9	3	0.954	67.6	1.829	70.6	1.848
10	3.162	1	78.4	1.894	87.8	1.943
11	3.316	1.041	83.7	1.922	91.1	1.959
12	3.475	1.079	93.9	1.972	94.5	1.974

Table 6: In vivo pharmacokinetic parameters for best formulation N15 and marketed cream

Parameters	Marketed formulation (Oxistaj cream 1% w/v)	Test Formulation (N15)
K_E (hr-1)	16.21± 0.19	14.18 ± 0.22
Ka (hr-1)	52.18 ± 0.27	48.21 ± 0.30
Cl_T (lit/hr)	20.38 ± 0.38	17.37 ± 0.32
V_d (lit)	2.41 ± 2.23	2.39 ± 0.19
Cmax (µg/mL)	95.12 ± 2.23	91.00 ± 3.35
Tmax (hr)	4	4
AUC 0-12 (µg.h/mL)	223.18±2.13	234.32 ± 2.10

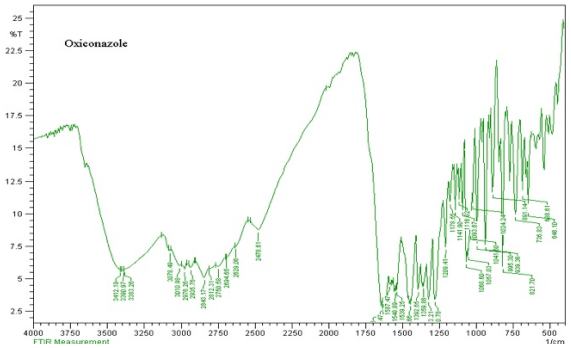

Figure 1: FTIR Spectra of pure Oxiconazole

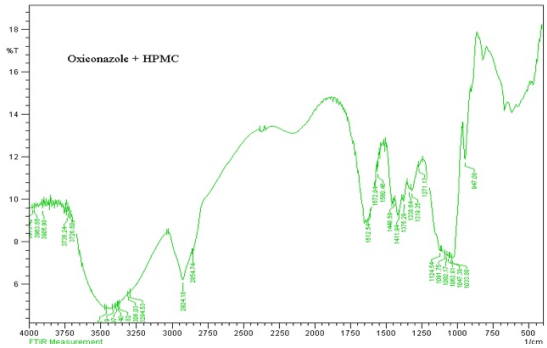

Figure 2: FTIR Spectra of Oxiconazole with HPMC

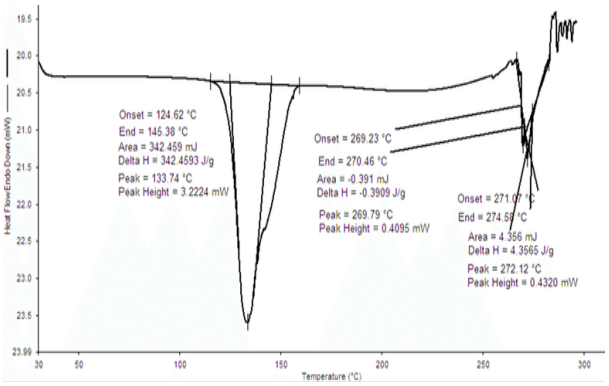

Figure 3: DSC Study of Oxiconazole with HPMC

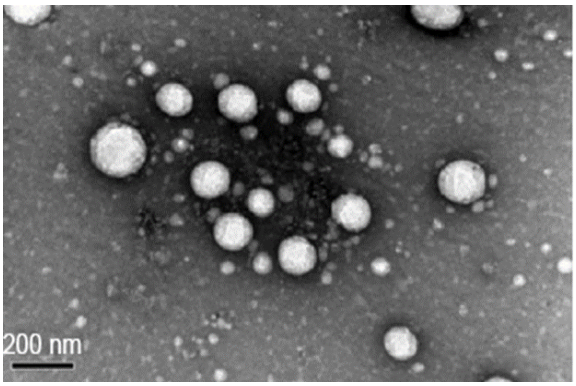

Figure 4: SEM image of Formulation N15 (Oxiconazole as Niosome gel)

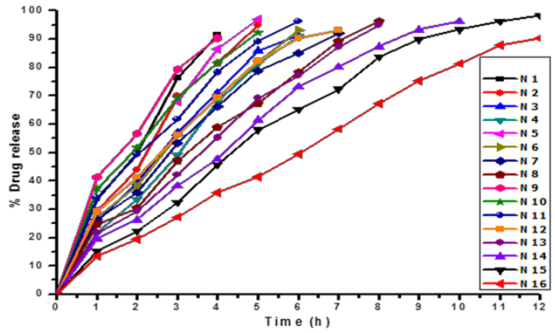

Figure 5: In vitro drug release of oxiconazole containing niosomes gel formulations N1 to N16

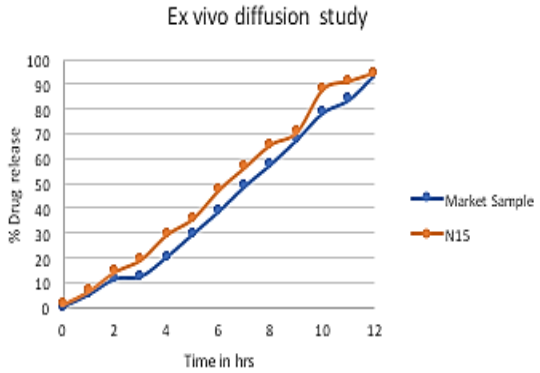

Figure 6: Ex vivo diffusion studies for best formulation N15 and marketed cream

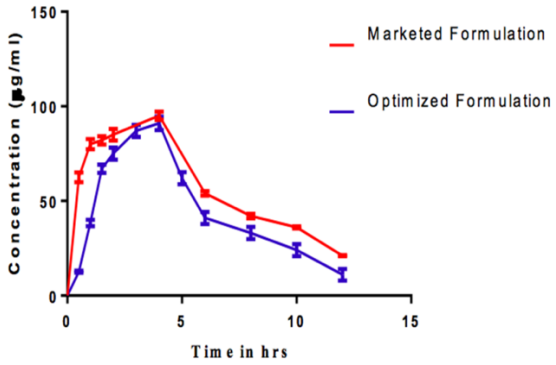

Figure 7: In vivo study for best formulation N15 and marketed cream (Oxistaj cream 1%)

CONCLUSION

Oxiconazole drug prepared in the form of niosomes as a gel for the topical skin infection. The whole processed formulation was once investigated with considerable test FTIR, DSC become undertaken with the intention to determine the compatibility between drug and polymers. Different type of polymers is used which is natural, semisynthetic and containing gelling nature is useful for control the release rate and spreadability of prepared gels. The *In vitro* Franz's diffusion program of studies conducted for all the formulations for niosomes and found that formulations N15 showed optimum drug release control 98% at 12h and release kinetic follows non fickian diffusion. Obtained ex vivo results conclude that the marketed gel has shown less percentage of drug release as compare to the prepared formulation niosome based gel N15. From the *In vivo* program of studies adept by prepared formulation N15 showed better results as it transforms the infected part of the skin like a normal skin with slight redness as compare to marketed Oxistaj cream 1%. The in vivo pharmacokinetic parameters like Cmax, Tmax, AUC 0-t Ka, KE, Vd were found to be comparable with marketed Oxistaj cream 1%.

REFERENCES

1. Fleming RV, Walsh TJ, Anaissie EJ. Emerging and less common fungal pathogens. Infectious Disease Clinics. 2002; 16(4):915-933.
2. Shapiro RS, Robbins N, Cowen LE. Regulatory circuitry governing fungal development, drug resistance, and disease. Microbiology and molecular biology reviews.2011; 75(2):213-267.
3. Cannon RD, Lamping E, Holmes AR, Niimi K, Baret PV, Keniya MV, Tanabe K, Niimi M, Goffeau A, Monk BC. Efflux-mediated antifungal drug resistance. Clinical microbiology reviews. 2009; 22(2):291-321.

4. Brown GD, Denning DW, Gow NA, Levitz SM, Netea MG, White TC. Hidden killers: human fungal infections. Science translational medicine. 2012; 4(165):165rv13-165rv13.
5. Gugnani HC, Ideyi C, Gugnani MK. Oxiconazole in the treatment of tropical dermatomycoses. Current therapeutic research. 1993; 54(1):122-125.
6. Heitman J. Microbial pathogens in the fungal kingdom. Fungal biology reviews. 2011; 25(1):48-60.
7. White TC, Marr KA, Bowden RA. Clinical, cellular, and molecular factors that contribute to antifungal drug resistance. Clinical microbiology reviews. 1998; 11(2):382-402.
8. Lopez-Martinez R. Candidosis, a new challenge. Clinics in dermatology. 2010; 28(2):178-84.
9. Polak A. Antifungal activity of four antifungal drugs in the cutaneous retention time test. Sabouraudia: Journal of Medical and Veterinary Mycology. 1984; 22(6):501-503.
10. Ramelet AA, Walker-Nasir E. One daily application of oxiconazole cream is sufficient for treating dermatomycoses. Dermatology. 1987; 175(6):293-295.
11. Gouveia DC, da Silva Jones C. Oxiconazole in the treatment of vaginal candidiasis: single dose versus 3-day treatment with econazole. Pharmatherapeutica. 1984; 3(10):682-685.
12. Gip L. Comparison of Oxiconazole (Ro 13–8996) and Econazole in Dermatomycoses: Vergleich von Oxiconazol (Ro 13–8996) und EconazolbeiDermatomykosen. Mycoses. 1984; 27(6):295-302.
13. Wagner W, Reckers-Czaschka R. Oxiconazole in dermatomycosis--a double-blind, randomized therapy compared with bifonazole. Mykosen. 1987; 30(10):484-492.
14. Sharma G, Jain S, Tiwari AK, Kaur GU. Once daily bioadhesive vaginal clotrimazole tablets: design and evaluation. ActaPharmaceutica-Zagreb-. 2006; 56(3):337-345.
15. Heitman J. Microbial pathogens in the fungal kingdom. Fungal biology reviews. 2011; 25(1):48-60.
16. Cannon RD, Lamping E, Holmes AR, Niimi K, Baret PV, Keniya MV, Tanabe K, Niimi M, Goffeau A, Monk BC. Efflux-mediated antifungal drug resistance. Clinical microbiology reviews. 2009; 22(2):291-321.
17. Azmin MN, Florence AT, Handjani-Vila RM, Stuart JF, Vanlerberghe G, Whittaker JS. The effect of non-ionic surfactant vesicle (niosome) entrapment on the absorption and distribution of methotrexate in mice. Journal of Pharmacy and Pharmacology. 1985; 37(4):237-242.
18. Iizhar SA, Ahmed MA, Arief M. Formulation and Characterization of MucoadhesiveBuccal Films of TrimetazidineDihydrochloride. Latin american journal of pharmacy. 2015; 34(8):1585-1593.
19. Bibi Marjan Razavia, Ali Alyasina, Hossein Hosseinzadeha, Mohsen Imenshahidib. Saffron Induced Relaxation in Isolated Rat Aorta via Endothelium Dependent and Independent Mechanisms: Iranian Journal of Pharmaceutical Research. 2018; 17(3):1018-1025.
20. Abdellatif MM, Khalil IA, Khalil MA. Sertaconazole nitrate loaded nanovesicular systems for targeting skin fungal infection: In-vitro, ex-vivo and in-vivo evaluation. International journal of pharmaceutics. 2017; 527(1-2):1-11.
21. Haque SE, Sheela A. Design and in vitro evaluation of interpolymer complex bound metformin sustained release tablet. Journal of Applied Polymer Science. 2014; 131(21).
22. Haque SE, Sheela A. Development of polymer-bound fast-dissolving metformin buccal film with disintegrants. International journal of nanomedicine. 2015; 10:199-205.
23. Guo F, Wang J, Ma M, Tan F, Li N. Skin targeted lipid vesicles as novel nano-carrier of ketoconazole: characterization, in vitro and in vivo evaluation. Journal of Materials Science: Materials in Medicine. 2015; 26(4):175.

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