Formulation And Evaluation Of Nanostructured Lipid Carrier Loaded Gel For Topical Delivery

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Abstract— **The present study focuses on the formulation and evaluation of a topical nanostructured lipid carrier gel containing Minoxidil, used in treatment of hypertension, hair loss, to enhance its therapeutic efficacy and provide sustained drug release. Minoxidil (MXD) loaded nanostructured lipid carriers (NLCs) was prepared by using emulsion solvent diffusion and evaporation method followed by ultra-sonication. The prepared NLCs was further characterized by SEM, particle size, zeta potential, entrapment efficiency, drug content and in-vitro diffusion study. on the basis of criteria, the drug diffused at 8 hr. the f6 batch is optimized and the optimized batch is incorporated in gel for further evaluation such as pH determination, drug content, viscosity and in-vitro diffusion study, stability. from the in-vitro diffusion study we observed that the Minoxidil release from NLCs gel by Higuchi model model indicates good linearity, which show controlled release of from Nanostructured Lipid Carriers gel.**

***Index Terms*— Nanostructured Lipid Carriers, Androgenic Alopecia, Topical drug delivery system, Gel, SEM, Entrapment efficiency etc.**

INTRODUCTION (HEADING 1)

Nanostructured Lipid Carriers (NLCs) are advanced lipid-based nano-systems developed to enhance the efficiency of drug delivery. They were introduced as a second-generation alternative to Solid Lipid Nanoparticles (SLNs), aiming to address their inherent limitations such as limited drug loading and physical instability. NLCs are composed of a mixture of solid and liquid lipids, resulting in a less crystalline matrix that accommodates higher drug incorporation and enables sustained release. [1]

These nanocarriers are typically in the size range of 50–500 nm and are stabilized using surfactants to prevent aggregation. Their internal structure facilitates improved solubility of lipophilic drugs, protection against degradation, and site-specific delivery. Based on their internal lipid organization, NLCs are categorized into three types: imperfect crystal, multiple (oil-in-fat-in-water), and amorphous.[2]

The choice of lipid (e.g., stearic acid, oleic acid) and surfactant (e.g., Poloxamer 188, Tween 80) significantly affects the formulation characteristics. Various methods are employed for their preparation, such as high-pressure homogenization (hot and cold), solvent evaporation, ultrasonication, and micro emulsion techniques. [3]

NLCs have shown potential in the treatment of androgenetic alopecia (AGA), a prevalent condition characterized by progressive hair follicle miniaturization driven by dihydrotestosterone (DHT). Topical agents like minoxidil, though commonly used, are limited by poor skin permeability and systemic absorption. NLC-based delivery systems enhance drug deposition in hair follicles, reduce systemic exposure, and improve therapeutic outcomes. [4]

Overall, NLCs offer a promising strategy for targeted and efficient dermal drug delivery, particularly in managing conditions like AGA, where enhanced follicular delivery is crucial. [5]

MATERIALS AND METHODS

MATERIALS

Minoxidil was provided as a gift sample by Psycho Remedies Pvt. Ltd – Punjab. Stearic acid, Glyceryl Monostearate, Triethanolamine, Disodium hydrogen phosphate, Potassium dihydrogen phosphate, Sodium chloride from S. D. Fine Chemicals, Mumbai. Poloxamer 188 by Signet Chemical Corporation Pvt. Ltd., Mumbai. Capryol90 supplied from Gattefosse India pvt. Ltd. Carbopol940 and methanol from Loba Pharma Chemie, Mumbai.

METHODS

PREFORMULATION STUDIES

Confirmation of Pure Drug

The received sample of drug minoxidil was standardized by carrying out the following tests.The minoxidil sample was evaluated visually for appearance and colour.[5]

Melting point

It is a criterion for purity as well as for identification. A capillary melting point apparatus was used to determine the melting point of Minoxidil. A small amount of Minoxidil was filled in capillary and the temperature at which the drug melted was noted down.[5]

UV SPECTROPHOTOMETRIC METHOD:

Determination of wavelength maxima (max)

A 10 ug/ml solution of Minoxidil was prepared in methanolic phosphate buffer pH 7.4. This solution was further scanned in the range of 200-400 nm using a UV-visible spectrophotometer.

Preparation of standard calibration curve of Minoxidil in methanolic phosphate buffer pH 7.4

Accurately weighed 100 mg of Minoxidil was dissolved in 100 ml of methanolic phosphate buffer pH 7.4 to get a stock solution of 1000 ug/ml. Aliquots of 5,10,15,20,25ug/ml were taken in a volumetric flask and the volume was up to 100 ml. Absorbances of all solutions were measured at 288 nm[6]

DSC (Differential scanning calorimetry)

A differential scanning calorimetry (DSC) study was carried out to evaluate the thermal behavior and thermo-tropic characteristics of the drug and optimized formulation (Nanostructured Lipid carrier suspension). Nearly 2.0 mg sample was sealed in an aluminum pan followed by heating at a rate of 10˚C/min over a temperature range of 40-400˚C under a nitrogen atmosphere of flow rate 10ml/min and thermo gram (STARe SW 12.10 thermal analyzer) was obtained.[7]

PRELIMINARY TRIALS FOR FORMULATION OF NANOSTRUCTURED LIPID CARRIER

Screening of Lipid and other Excipients used in Nanostructured lipid carrier Formulation

Screening of Lipid -Lipid is most important parameter for formulation of Nanostructured lipid carrier. So Screening was done in between Screening was done in between stearic acid, glyceryl monostearate, phospholipon-90, capryol-90, oleic acid etc. by solubility method. Method: An excess amount of Minoxidil was added to each glass vials containing 1gm of the selected molten solid lipids. After the mixture was sonicated using a bath sonicator (PCI, Mumbai) for 5 min in order to facilitate proper mixing of Minoxidil with the vehicles. Drug was added upto super-saturation from. Then the mixture was centrifuged at 3000 rpm for 15 min using a centrifuge (Remi Mumbai, India). The 0.1ml supernatant was then pipette out and diluted in 10 ml volumetric flask upto the mark with methanolic phosphate buffer 7.4. Absorbance reading for the same was taken using a Shimadzu UV- 1800 UV-Visible Spectrophotometer against a blank solution at a wavelength of 288 nm. Same procedure were performed for liquid lipids an excess amount of minoxidil was added to glass vials containing 1ml of liquid lipids. Solubility of Minoxidil in different excipients is described in table No. 13 like, oleic acid, stearic acid, glyceryl monostearate, Poloxamer 188, Span 20, Phospholipon90h, Tween 80, PEG 200, Tween 20, Tween 60, poloxamer 188.[7]

Formulation of Nanostructured Lipid Carrier [9]

Minoxidil loaded nanostructured lipid carriers (MXD-NLCs) were prepared by an emulsion solvent diffusion and evaporation method followed by ultrasonication. In brief, 0.25g of MXD was dispersed in the liquid lipid then added to molten solid lipid. Following that, of acetone and ethanol mixture (1:1) was added to the lipids maintained in a water bath at 80°C until complete lipids get dissolve in the organic phase. The latter was dispersed in an aqueous solution containing Poloxamer 188 at 80°C and mixed using a magnetic stirrer rotating at 1,000 rpm. The resulting pre-emulsion was then ultrasonicates for 3 minutes to produce an oil/water nanoemulsion that was cooled down at room temperature while stirring at 500 rpm until evaporation of the organic solvent to form MXD-NLC dispersion of 10ml. [7]

EVALUATION OF NANOSTRUCTURED LIPID CARRIER

Appearance: All formulations were visually inspected for their appearance.

Scanning Electron Microscopy

A scanning electron microscope (SEM) is a type of electron microscope that produces images of a sample by scanning it with a focused beam of electrons. The electrons interact with atoms in the sample, producing various signals that can be detected and that contain information about the sample's surface topography and composition.[8]

Drug Entrapment Efficiency

The Nanostructured lipid carrier suspension was ultra-centrifuged at 7000 rpm for 1hr by using ultra centrifuge to separate the free drug. A clear solution of supernatant of NLCs were obtained. 1ml solution from the supernatant was collected and absorbance of the drug was noted at 288 nm. The entrapment efficiency was then calculated using following equation. %Entrapment efficiency(EE)= amount of drug in NLCs/initial amount of drug X 100 [9]

Drug Content

Drug content of Nanostructured lipid carrier was determine by using 1 ml of sample diluted upto10 ml with distilled water followed by 5 min Ultrasonication, then filtered the above solution, and took 1 ml of that solution and further dilute upto 10 ml with distilled water and absorbance of the drug was noted at 288 nm. The drug content was determined by the following equation – Drug Content = (practical value /Theoretical value) X 100[8]

Particle Size

Nanostructured lipid carrier diameter can be determined using Malvern zetasizer.[10]

Zeta Potential Determination

Charge drug loaded vesicles surface was determined using Malvern. Analysis time was kept for 60 s and average zeta potential and charge on Nanostructured lipid carrier was determined.[11]

In Vitro Diffusion study of Nanostructured lipid carrier suspension

The in vitro diffusion study was carried out using a Franz diffusion cell with egg membrane. The membrane with effective diffusion area of 2.0 cm² was mounted between the donor and receptor compartments of the diffusion cell. The volume of receptor medium was 110ml of PBS (pH 7.4) thermo stated at 37 ± 1 °C, which was continuously stirred at 100 rpm throughout the experiment. 1ml of MXD was placed in the donor compartment and the receptor compartment was filled with dialysis medium (7.4 Methanolic PBS buffer). Required quantity (1ml) of the medium was withdrawn at specific time periods (1, 2, 3, 4, 6, 8 hr) and the same volume of media was replaced in the receptor compartment to maintain a constant volume. The withdrawn samples were filtered and then 1ml filtrate was made up to volume with 10 ml of methanolic buffer. The samples were analysed for drug release by measuring the absorbance at 288 nm using a UV/ visible spectrophotometer (Shimadzu Corporation, Japan). The experiments for all formulations were conducted triplicate and average value recorded.[12]

PREPARATION OF GEL

The preparation of gel for the incorporation of Nanostructured lipid carrier containing minoxidil was select on the basis of literature review.

Preparation of Nanostructured lipid carrier Gel

To make the optimized Nanostructured lipid carrier suspension suitable for topical applications, it was further incorporated into Carbopol1940 hydrogel (0.5% w/w). Carbopol940 was chosen to prepare the NLCs gel due to its hydrophilic nature and bio-adhesive properties. This may result in increased residence time of a drug at the absorption site by interacting with the skin. The aqueous gel base of Carbopol1940 was prepared, and the optimized NLCs suspension was gradually mixed with continuous stirring to obtain a Nanoostructured lipid carrier-based hydrogel.[7]

Evaluation of MXD-loaded NLCs Gel Organoleptic Characteristic

After gel preparation, the formulation was determined by visual examination for appearance, consistency, and phase separation.[7]

Measurement of pH

1 g of gel was dispersed in 20 ml of distilled water, and a digital pH meter was used to determine the pH value. The measurement was performed three times and mean +- SD was calculated.[5]

Spreadability

A weight quantity of 1g gel was placed within a circle of 1cm diameter pre-marked on a glass plate over which a second glass plate was placed. A weight of 500gm was allowed to rest on the upper glass plate for 5min. The increase in diameter due to spreading of glass was noted.[5]

The spreadability of formulation was done in triplicate and the mean value was calculated. Spreadability = M.L / T Where, M – weight tied to upper glass slide, L – Length of glass slide, T – Time taken to separate slides.

Drug content

The drug content was determined by dissolving 1 g of gel in 10 ml of phosphate buffer. The solution was then filtered through Whatman filter paper, and the filtrate was analyzed using a UV spectrophotometer.[13]

Viscosity

Viscosity of formulation was determined using Brookfield viscometer with spindle at room temperature and at a 10, 20, 50, 100 rpm.[14]

In vitro diffusion study

The in vitro diffusion study was carried out using a Franz diffusion cell with egg membrane. The membrane with effective diffusion area of 2.0 cm² was mounted between the donor and receptor compartments of the diffusion cell. The volume of receptor medium was 20ml of PBS (pH 7.4) thermo stated at 37 ± 1 °C, which was continuously stirred at 100 rpm throughout the experiment. 1gm of nanostructured loaded gel was applied on membrane and placed between the compartments. At 1, 2, 3, 4, 5, and 6 hr. intervals 1ml of the solution in the receptor compartment was removed and replaced immediately with an equal volume of fresh buffer. Samples were analyzed using UV spectrophotometer and the data was recorded.[15]

In vitro drug release kinetics study

In vitro diffusion study has been recognized as an important element in drug development. To analysis the mechanism for the drug release and release rate kinetics of the formulated dosage form, the data obtained from conducted studies was fitted into Zero order, First order, Higuchi matrix, Korsmeyer-Peppas. In this by comparing the r-values obtained, the best-fit model was selected.[5]

Stability studies

Stability of a drug has been defined as the ability of a particular formulation, in a specific container, to remain within its physical, chemical, therapeutic and toxicologicalspecifications. This testing should be performed on the constituted or diluted product through the proposed in-use period on primary batches as part of the formal stability studies at initial and final time points. In the present study, stability studies were carried out at 2-8°C, Room Temp and at 40°C for a specific time period up to 25days for the optimized formulation. The Nanostructured Lipid Carrier Loaded gel preparation was kept in sealed vials (10 mlcapacity). Samples were withdrawn periodically and analyzed for Drug Entrapment and MXD loaded NLCs gel formulation also check visually for change in colour of NLCs. [11]

RESULTS AND DISCUSSION

PREFORMULATION STUDIES

Confirmation of Pure Drug

Minoxidil was supplied by Psycho Remedies Pvt. Ltd. was a crystalline powder, which is almost white in colour. Hence it was confirmed that received sample was Minoxidil.

MELTING POINT DETERMINATION:

The melting point of the Minoxidil drug sample was found to be 240°C- 257°C which is within the reported range of 248°C-250°C. It complies with the purity of the drug sample.

SPECTROPHOTOMETRIC METHOD FOR THE ESTIMATION OF MINOXIDIL:

Standard calibration curve of minoxidil in methanolic phosphate buffer 7.4:

Determination of maximum wavelength (λmax)

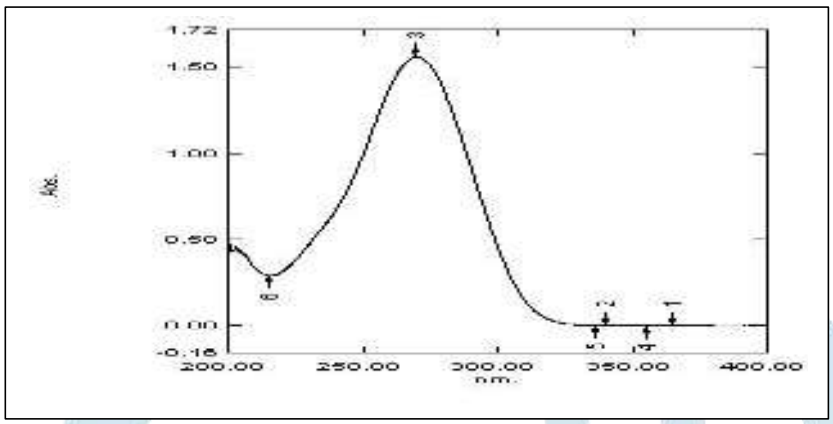

Figure No. 1- UV Spectrum of Minoxidil in methanolic phosphate buffer 7.4

After scanning 100µg/ml solutions, only one peak at 288 nm was observed and considered as λmax. From the standard curve, it was observed that the drug obeys Beer's law in concentration range of 5-25 µg/ml in methanolic pbs 7.4. Drug shown good linearity with regression of coefficient (r^2 = 0.9938) and equation for this line obtained was found to be (y = 0.0278x + 0.0607) which is used for the calculation of entrapment efficiency.

DSC of Minoxidil Pure Drug

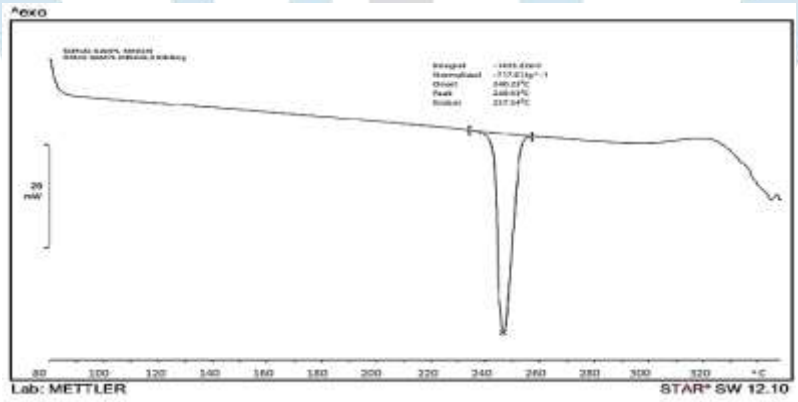

Figure No. 2- DSC of Minoxidil Pure

The most noticeable thing about the output from the sample of MXD the apparent presence of one peak. The accepted melting point of this drug is 240°C 257°C and in DSC study intense peak shown at 249.63°C show the melting point of drug and purityof drug sample.

PRELIMINARY TRIALS FOR FORMULATION OF NANOSTRUCTURED LIPID CARRIER (NLCs):

SCREENING OF LIPIDS:

Lipid is main component of formulation of NLCs and here the lipids were screened on the basis of solubility of drug in lipid and result are following maximum solubility shows in stearic acid shows 31.26mg /1gm as a solid lipid and liquid lipid capryol90 shows the maximum solubility 21.91mg/ml.

Table No. 1: Solubility of Minoxidil in melted solid/ liquid Lipid

Sr. No.	Solid Lipids	Solubility
1.	Stearic acid	31.26 mg/1 gm lipid
2.	Glyceryl monostearate	29.11 mg/1 gm lipid
3.	Phospholipon 90h	22.99 mg/1 gm lipid
	Liquid Lipids	
1.	Capryol90	21.91mg/ml
2.	Oleic acid	19.33mg/ml
3.	Soyabean oil	16.88mg/ml

Discussion: To develop a Nanostructured lipid carrier of Minoxidil for topical administration, it should possess good solubility in the components of the system, to avoid any drug precipitationupon dilution. The solubility of Minoxidil in various lipids.

FORMULATION STUDY:

Formulation of Nanostructured lipid carriers (NLCs) using emulsion solvent diffusion and evaporation method followed by ultrasonication method

Figure No. 3: Photograph of Nanostrucctured Lipid Carrier

EVALUATION OF NANOSTRUCTURED LIPID CARRIERS:

Appearance:

Formulation appear as milky white. NLCs suspension have smooth texture with no visible grittiness or coarseness.

Scanning Electron Microscopy

The determination of shape and surface morphology was done by scanning electron microscope JEOL-5400, Japan. SEM analysis of the samples revealed that all Nanostructured lipid carrier prepared were spherical in shape. Scanning electron photographs of Nanostructured Lipid Carrier were shown in Figure18

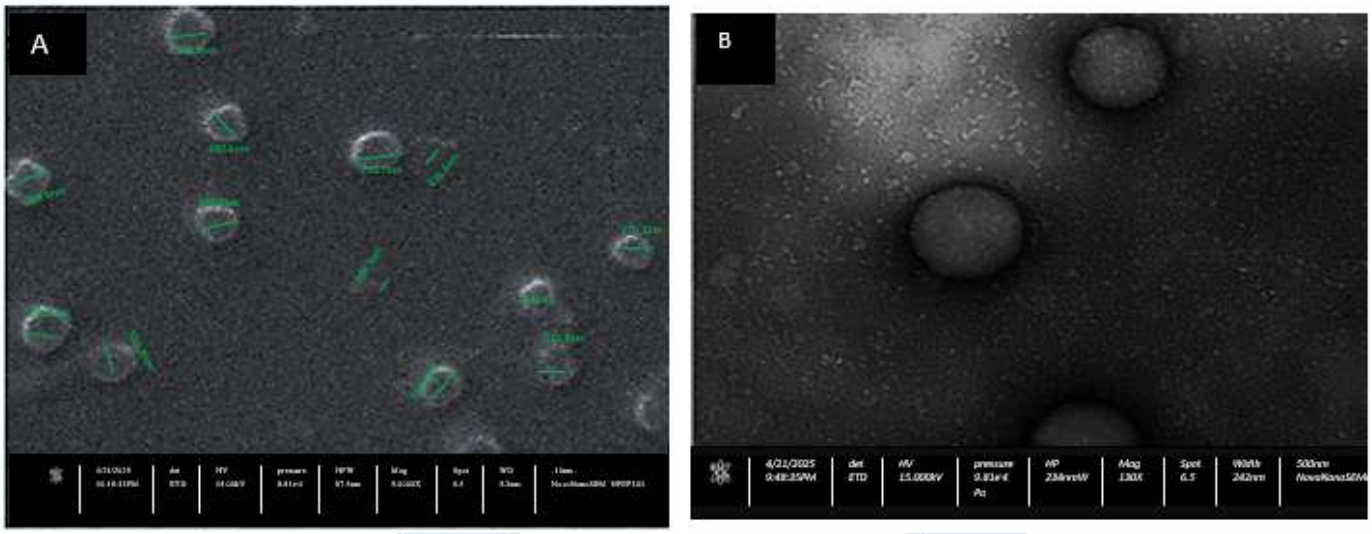

Figure No. 4: Scanning Electron Microscope Photographs of The Nanostructured Lipid Carrier

Drug entrapment efficiency and drug content:

Table no. 2 Results Of Percent Drug Entrapment Efficiency and drug content

Batch No.	Batches Name	Entrapment Efficiency (%)	Drug content (%)
1	MXD- 1	79.58±0.24	40.68± 0.34
2	MXD- 2	80.29±0.19	42.10± 0.25
3	MXD- 3	88.06±0.22	47.46± 0.15
4	MXD- 4	89.66±0.26	45.63± 0.42
5	MXD- 5	90.38±0.46	66.32± 0.21
6	MXD- 6	91.54±0.36	89.23± 0.23
7	MXD- 7	78.3±0.20	42.33±0.18
8	MXD- 8	81.74±0.14	58.90±0.22
9	MXD-9	83.32±0.35	59.6±0.19

Discussion: From the above observation the F-3, F-5, F-6, F-8 and F-9 show the highest %entrapment efficiency and %drug content. So in vitro diffusion study of F-3, F-5, F-6, F-8 and F-9 batches are carried out and selection of the final batches.

Particle Size Analysis:

Average particle size of Nanostructured lipid carrier was determined by MALVARN ZETASIZER.

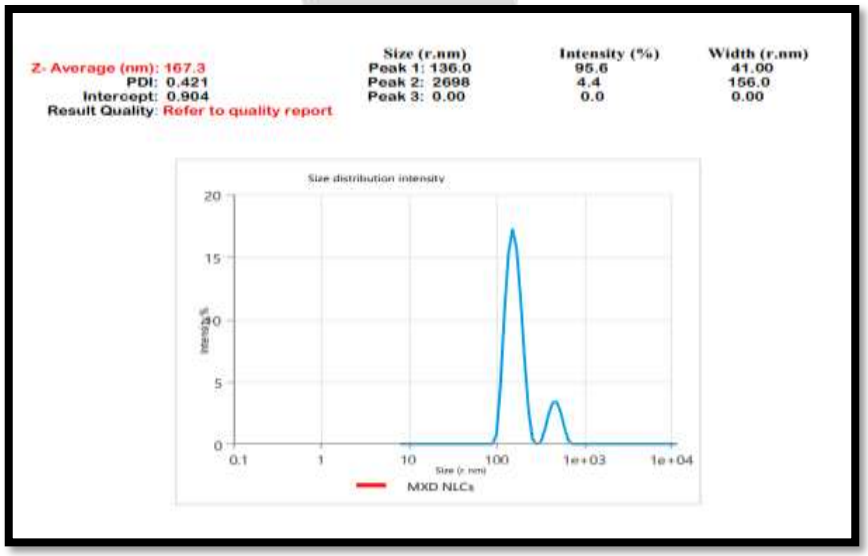

Figure No. 5: Particle size of optimize formulation (MXD-6) of Nanostructured Lipid Carrier

Discussion: This graph illustrate the distribution of particle sizes within a sample. The x-axis represents the size of particles, typically ranging from nanometers to micrometers, while the y-axis shows the frequency or proportion of particles within each size range.The average particle size of optimize formulation MXD-6 was found to be 167.3nm.

ZETA POTENTIAL OF NANOSTRUCTURED LIPID CARRIER:

Zeta potential of Nanostructured lipid Carrier was determined by MALVARNZETASIZER. Figure 27 illustrate Zeta potential for optimized batch of Naostructured Lipid Carrier was -21.65 mV indicating presence of optimum charge on the surface of formulations to prevent aggregation during their shelf life.

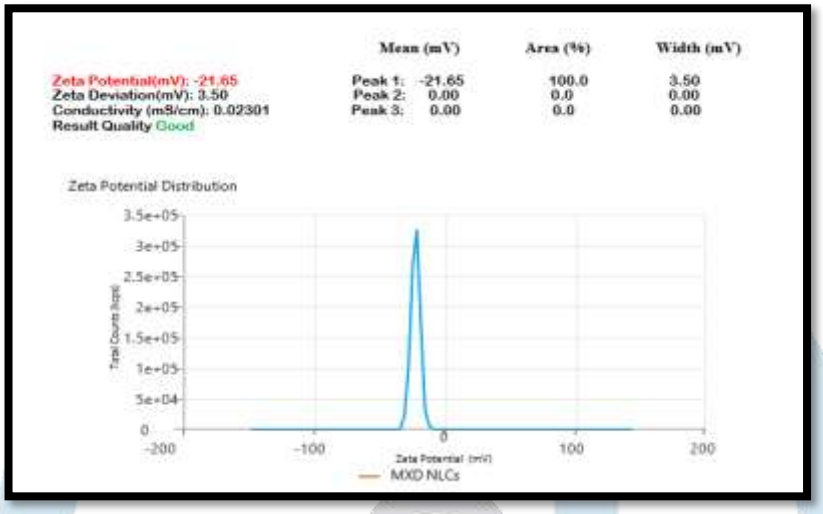

Figure No. 6: Zeta potential of optimize formulation (MXD-6) of Nanostructured Lipid Carrier

IN VITRO DRUG DIFFUSION STUDY OF NLCS SUSPENSION:

Table No. 3: In Vitro Drug Diffused Study of NLCs suspension

Sr. No.	Time (Hr)	% Cumulative Drug Diffused								
		MXD 1	MXD 2	MXD 3	MXD 4	MXD 5	MXD 6	MXD 7	MXD 8	MXD 9
1	0	0	0	0	0	0	0	0	0	0
2	1	12.3 ±0.33	23.00 ± 0.15	25.59 ± 0.26	12.64± 0.28	42.28± 0.35	33.36± 0.37	33.64± 0.23	39.69± 0.32	13.21± 0.33
3	2	19.26 ± 0.13	32.72 ± 0.32	27.60 ± 0.17	33.93± 0.16	46.02± 0.22	49.47± 0.32	41.13± 0.44	43.72± 0.43	33.36± 0.22
4	3	22.13 ± 0.23	41.41 ± 0.21	32.49± 0.22	41.70± 0.43	48.03± 0.43	53.21± 0.45	46.31± 0.26	46.59± 0.44	41.99± 0.18
5	4	32.40 ± 0.43	45.74 ± 0.16	40.26 ± 0.35	46.88± 0.32	49.76± 0.33	55.51± 0.22	48.32± 0.23	48.61± 0.18	46.59± 0.21
6	5	40.55 ± 0.22	50.66± 0.33	44.58± 0.23	48.61± 0.33	52.35± 0.21	70.19± 0.32	50.05± 0.33	50.33± 0.17	48.89± 0.32
7	6	57.36 ± 0.16	66.36 ± 0.13	52.35 ± 0.33	50.33± 0.34	51.77± 0.21	78.82± 0.17	51.77± 0.45	66.57± 0.12	50.49± 0.18
8	7	63.78 ± 0.42	69.96 ± 0.23	63.61 ± 0.12	52.35± 0.14	67.31± 0.32	81.70± 0.14	66.56± 0.21	69.08± 0.22	52.35± 0.27
9	8	68.36 ± 032	73.64 ± 0.12	71.55 ± 0.21	64.44 ± 0.22	70.19 ± 0.33	84.58 ± 0.45	81.12 ± 0.23	75.35 ± 0.33	63.66 ± 0.32

N=3

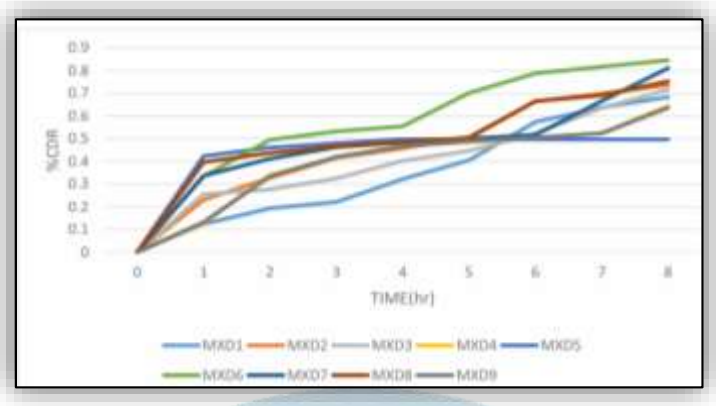

Figure No. 7 % Cumulative Drug Diffusion of Batch MXD-1 To MXD-9 at Different TimeIntervals

Table No. 20 presents the results of the in vitro drug release study for formulations MXD-1 to MXD-9, at various time intervals. The cumulative drug release percentages are recorded over time to assess the release profile of the formulations. As time progresses, the cumulative drug release increases for all formulations. Formulations MXD-1 to MXD-9 exhibit cumulative drug release percentagesat different time intervals. From the In-vitro Release study of all formulations (MXD-1 To MXD-9), formulation MXD-6 release around 84.58 % of drugat the end of 8 hours for a sustained release. Therefore, the MXD-6 formulation chosenas the optimize formulation from all nine batches.

Preparation gel:

The preparation of gel was achieved by gradual addition of gelling agent 0.5% Carbopol 940 by mechanical mixing at 500 rpm for 30 min. A 7ml NLCs suspension equivalent to 175mg drug was subsequently introduced gradually into the polymer gel. The ultimate amount was adjust to 50ml utilizing distilled water. The gel that were produced were subsequently stored at ambient temperature in order to facilitate subsequent investigation.

Table no. 4. Preparation of MXD loaded NLCs gel (MXDG-6)

Sr. no.	Excipients	Quantity(%)
1	Carbopol940	1.0%
2	Glycerol	2.5%
3	Methylparaben	0.2%
4	Triethanolamine	q.s.
5	Water(q.s)	Upto 50ml

Evaluation of MXD loaded NLCs gel(MXDG-6):

Table no. 5 Evaluation parameter of gel and results:

Sr. no.	Evaluation Parameter	Results
1	Colour	Milky white
2	Clarity	Opaque
3	Texture	Smooth and homogenous
4	Consistency	Thick, semi-solid
5	pH	7.1
6	Spreadability	8.26 g.cm/sec
7	Viscosity	2061.9
8	Drug content(%)	87.4%

Figure no. 8. Minoxidil loaded NLCs Gel

In vitro drug diffusion study of MXD loaded Nanostructured lipid carrier gel (MXDG-6)

In vitro diffusion studies were performed using Franz diffusion cells. The egg membrane was placed between receptor and donor compartments. The in vitro diffusion study in saline phosphate buffer pH 7.4 was carried out using an egg membrane according to the procedure explained in the section of the chapter.

Figure no. 9. In vitro diffusion study by using franz diffusion cell

Table no.6. %cumulative amount of drug diffused vs time(hr)

Sr.no.	Time(hr)	%CDD
1	0hr	0
2	1hr	22.82 ± 0.46
3	2hr	28.57 ± 0.88
4	3hr	31.45 ± 0.46
5	4hr	37.20 ± 0.60
6	5hr	42.96 ± 0.13
7	6hr	48.71 ± 0.42
8	7hr	51.59 ± 0.21
9	8hr	57.35 ± 0.33

Discussion: In vitro drug diffusion study perform by using the franz diffusion cell with the MXD loaded Nanostructured lipid carrier gel (MXDG-6) show the 57.35± 0.33 cumulative amount of drug diffused upto 8 hr.

DRUG RELEASE KINETICS

Table No.7: Drug Release Kinetics of Formulation MXDG-6

Sr. No.	Time(T) (hr)	Sq. rt. of T	Log T	% Cum. Drug Released	% Cum. Drug Retained	Log %Cum. Drug Released	Log %Cum. Drug Retained
1	0	0	0	0	0	0	0
2	1	1.0	0	22.82	77.18	1.35	1.88
3	2	1.41	0.30	28.57	71.49	1.45	1.85
4	3	1.73	0.47	31.45	68.55	1.49	1.83
5	4	2.0	0.60	37.20	62.55	1.57	1.79
6	5	2.23	0.69	42.96	57.04	1.63	1.75
7	6	2.64	0.77	48.71	51.29	1.68	1.71
8	7	2.64	0.84	51.59	48.41	1.71	1.68
9	8	2.82	0.90	57.35	42.65	1.75	1.62

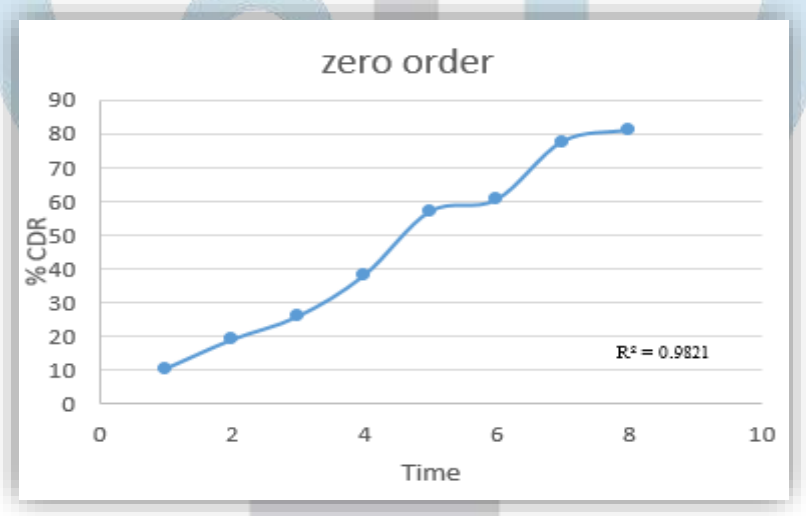

Figure No. 10: Plot of % Cum. Drug Released Vs. Time (Zero Order Kinetics) For Formulation MXDG-6

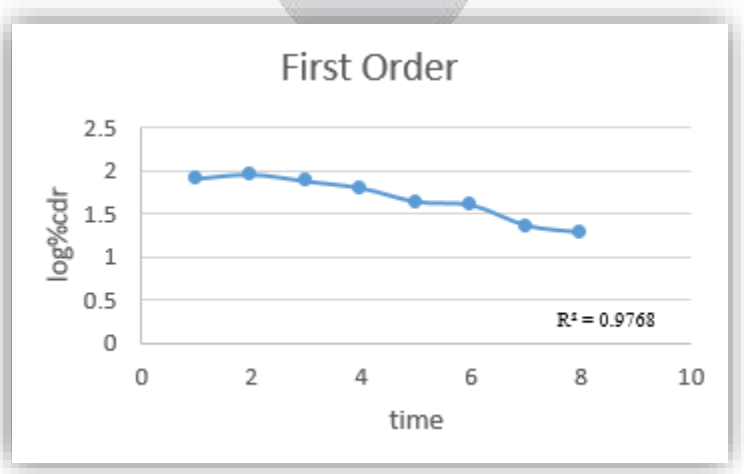

Figure No. 11: Plot of Log % Cum. Drug Retained Vs. Time (First Order Kinetics) For Formulation MXDG-6

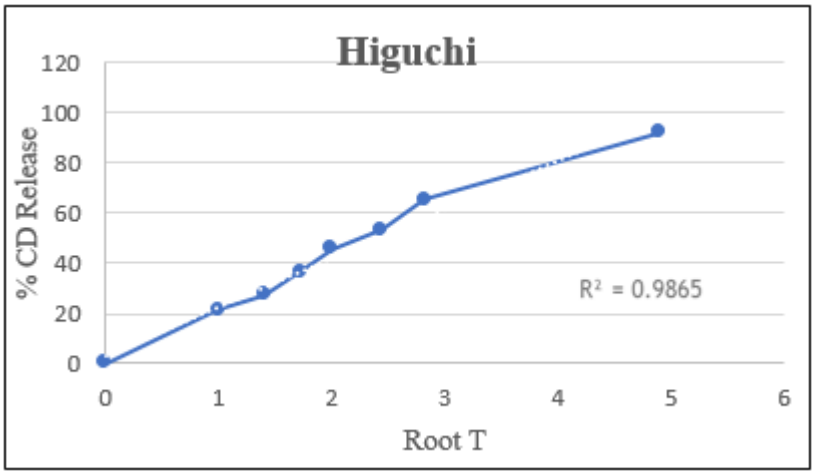

Figure No. 12: Plot of % Cum. Drug Released Vs. Sq. rt. of Time (Higuchi Model) For Formulation MXDG-6

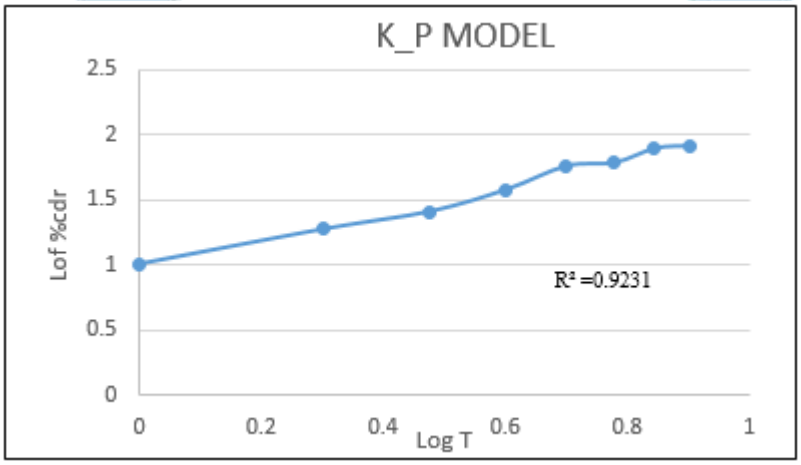

Figure No. 13: Plot of Log % Cum. Drug Released Vs. Log of Time (Korsmeyer -Peppas Model) For Formulation MXDG-6

Table No. 8: Regression Coefficient of MXDG – 6

NLCs loaded gel formulation	Regression Coefficient (R^2) values			
	Zero order	First order	Higuchi Model	Korsemeyer peppas
MXD gel -6	0.9821	0.9768	0.9865	0.9231

Drug release study shows that the release of drug from the NLCs by higuchi model model indicates good linearity R2 = 0.9865 followed by zero order and first order model. The release exponent "n" was found to be 0.65 which appears to indicates the anamolous (non- fickian) diffusion indicating drug release is controlled by more than one process, i.e. superposition of both phenomenon, the diffusion controlled as well as swelling controlled release.

STABILITY STUDY:

The stability studies of optimum formulation revealed that there is no significant reduction in drug entrapment efficiency and Physical appearance was observed over period of 1month da. The results are shown in Table 19 and 20.

Table No 9: Stability Study of Nanostructured Lipid Carrier

Sr. No	days	Drug entrapment efficiency (%)		
		2-8 °C	Room Temp	40±2° C
1	0	78.09	78.09	78.09
2	5	77.97	77.05	76.80
3	10	77.80	76.55	74.58
4	15	77.69	75.97	72.47
5	30	76.48	74.4	68.37

Table No 10: Stability Study of Nanostructured Lipid Carrier

Sr. No	days	Physical appearance		
		2-8 °C	Room Temp	40±2° C
1	0	Milky White	Milky White	Milky White
2	5	Milky White	Milky White	Milky White
3	10	Milky White	Milky White	Milky White
4	15	Milky White	Milky White	Milky White
5	30	Milky White	Milky White	Faint yellow

Here, 1month stability study of Nanostructured Lipid Carrier was conducted with respect to the Nanostructured Lipid Carrier ability to retain an entrapped drug during a defined time period. storage conditions, i.e. at refrigeration condition (2 - 8 ºC) at room temperature (25±2ºC) and at 40ºC table 19 and 20 shows Nanostructured Lipid Carrier were relatively stable at refrigerated storage condition. The drug leakage percent amounts of original entrapped in Nanostructured Lipid Carrier are very small (< 5%) at 2–8ºC and have no significant difference after 30days compared with immediately after preparation. The results of drug retention studies show higher drug leakage at higher temperature. This may be due to the higher fluidity of lipid bilayers at higher temperature, resulting in higher drug leakage.

After 1 month only 40±2ºC storage condition shows only faint yellow colour of the formulation of Nanostructured Lipid Carrier. 2-8ºC storage condition and room temperature shows no significant change in physical appearance of NLCs.

CONCLUSION

Minoxidil-loaded Nanostructured Lipid Carriers (NLCs) were successfully formulated using the emulsion solvent diffusion and evaporation method, followed by ultrasonication to achieve nano sized particles. During the initial screening of lipids, stearic acid (solid lipid) and Capryol 90 (liquid lipid) demonstrated the highest drug entrapment efficiency and were therefore selected as the lipid components for NLC formulation. The optimized NLC formulation exhibited a particle size of 167.3 nm, indicating successful nanoscale dispersion. Drug entrapment efficiency across all batches ranged from 78.3% to 91.54%, with batch MXD-6 showing the highest entrapment (91.54%) and thus selected as the optimized batch for further gel formulation. DSC thermograms further supported the purity and stability of the drug within the formulation. SEM analysis revealed the spherical morphology and smooth surface of the NLCs, while zeta potential measurement for the optimized batch was –21.65 mV, indicating sufficient surface charge to maintain colloidal stability and prevent aggregation during storage. In vitro drug release studies using a Franz diffusion cell demonstrated maximum cumulative drug release of 84.58% in batch MXD-6, confirming its potential as an optimized formulation. The concentration ratio of solid lipid, liquid lipid, and surfactant significantly influenced the drug release behavior. The release kinetics followed the Higuchi model, indicating a diffusion-controlled mechanism, and was consistent with zero-order release kinetics. The release pattern exhibited non Fickian (anomalous) diffusion, suggesting a combination of diffusion and erosion mechanisms. Stability study of NLCs gel was perform at different temp. condition. It show less than 1% decreasment of drug entrapment in 2-80 C storage condition for 1month and in 400C after 1month. In conclusion, the optimized Minoxidil-loaded NLC

gel formulation exhibits excellent physicochemical properties, high drug entrapment, sustained release, and stability—making it a promising candidate for effective topical treatment of androgenic alopecia.

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