Formulation and evaluation of captopril transdermal liposomsal gel

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Abstract- In the present study, Captopril liposomsal gel were developed in order to accomplish short half life of captopril to eliminated a first-pass metabolism of captopril in GIT. To enhances the bio-availability of drug. To increases a sustained release of drug for prolonged period of time. Firstly, developed liposomes by ether injection method and then formulated a liposomal gel by using dispersion method. For this purpose, Phospholipon-90H, cholesterol and stearylamine was chosen as excipient for formulated a liposomes. By taken different concentration of stearylamine and cholesterol applied a full factorial design. All the nine batches were evaluated with respect to the drug content and entrapment efficiency, in-vitro drug diffusion. The optimized batches were evaluated with respect to particle size, FESEM, FTIR and zeta potential. The in vitro drug released of liposomes was found to be 45.48±0.36% at 10 hrs and 54.84±2.63% up to 12 hours respectively, followed super case II transport mechanism with zero order release. The results of in-vitro revealed that captopril liposomes gives a sustained released as compared to captopril solution. The optimized sustained release batch was used to formulate captopril liposomal gel. The 1% liposomal gel was evaluated with respect to the spreadability, viscosity, drug content, ph measurement and in vitro diffusion study. Into in vitro diffusion studied data of captopril liposomal gel compared with captopril gel which showed that liposomal gel gives sustained release of drug 45.18±0.51% up to 12 hrs. Captopril gel formulation used transdermally to treat a hypertension. Also, eliminated a problem of orally administered drug caused. It also protects a drug from hepatic degradation. Increase better patient compliances.

Keywords: Captopril, Liposomes, Captopril liposomal gel, Transdermal, ether injection method, stearylamine, in-vitro diffusion study

I. INTRODUCTION

Captopril (1-[(2S)-3-mercapto-2-methyl propionyl] -1-proline), an angiotensin converting enzyme inhibitor, has been commonly used for the treatment of hypertension and congestive heart failure due to its efficiency and minimal toxicity. It is typically recommended for patients with chronic illnesses who need long-term treatment medications. The recommended dosage is 37.5-75 mg, taken three times day in divided doses. (Shanthi et al., 2010)

It has been shown to be effective in the management of severe heart failure when used in combination with continued digitalis and diuretic medication. Administration of captopril improves cardiac function through a decrease in systemic vascular resistance (afterload) and the various factors influencing left ventricular filling pressure (preload). Many positive outcomes have been seen in captopril short-term haemodynamic investigations in patients with severe congestive heart failure. When captopril (12.5 to 150 mg) is administered, cardiac output (25 to 30%), cardiac index (15 to 40%), and stroke volume index consistently rise (18 to 49 %). Systemic vascular resistance (20–45%), pulmonary vascular resistance (35–45%), total vascular resistance (25–35%), pulmonary artery pressure (15–30%), pulmonary capillary wedge pressure (25–50%), and right atrial pressure are all decreased in combination with the

effects (25 to 45 %).and right atrial pressure (25 to 45%). (Romankiewicz et al., 2012)

Limitation of captopril has a relatively short elimination half-life in plasma with estimates in man ranging from 1.6 to 1.9 h. In the presence of food there may be decrease oral in absorption of captopril by up to 25–40%. According to the previous research, the oxidation rate of captopril in dermal homogenates is significantly lower than that in intestinal homogenates because the oxidative product of captopril, i.e., captopril disulfide is difficulty absorbed from the intestine. Consequently, transdermal drug delivery system (TDDS) may be acceptable for captopril as a successful dosage form. (Pao et al., 2000)

Drug delivery system (DDS) offers the potential to improve the therapeutic index of drug concentration, the residences time in specific targets cells and minimizing the side effects. Novel drug delivery system involve delivering the potentially active drug to the site of action via a nano vehicle to enhance the pharmacokinetics properties of free drugs and cover their unfavourable features through enhancing drug pharmacokinetics and biodistribution, as well as acting as drug reservoirs. Different natural, organic and inorganic materials are used to create NPs including ceramic, polymers, metals, and lipids that generate nanoparticles like micelles and liposomes. (Nsairat et al., 2022)

Liposomes are small spheres made up of an aqueous core and one or more lipid-based outer shells that are arranged in a bilayer structure. Liposomes have been used as drug delivery systems for more than 40 years. Liposome has an ability to encapsulate both hydrophilic and lipophilic compounds, because of these properties of liposomes are considered as superior carriers.

Transdermal drug delivery system is commonly recognized as one of the most reliable, appealing as well as effective technique. Drug delivery through the skin has been an attractive as well as a challenging area for research. Over the past two decades, transdermal drug delivery had become an appealing and patient acceptable technology as it minimizes and avoids the limitations allied with conventional as well as parenteral route of drug administration such as peak and valley phenomenon that is exhibiting fluctuation in plasma drug concentration level, pain and inconvenience of injections and the limited controlled release of both.

Transdermal pharmaceutical products, whether ointments, liposomal gel, matrix formulations or reservoir systems provide the considerable advantage of a non-invasive parental route for drug therapy, avoidances of first-pass gut and hepatic metabolism, potentially minimized side effects and relative ease of drug input termination in problematic cases. The rate-controlled transdermal dosage form can provide a precise regulation of drug concentration in plasma and thus a high degree of safety and selectivity of action for some drugs. In recent years the transdermal therapeutic system has become popular for improved administration. (Pao et al., 2000)

Proposed mechanisms for the interaction of liposomes with the skin.

- Adsorption of liposomes to the skin surface; drug transfer from liposomes to skin.
- Rupture of vesicles, release of content and the penetration of the free molecules into the skin. Via are intracellular route.
- Penetration of unilamellar vesicles via the lipid-rich channels to the dermis where they slowly release their content due to disruption or degradation of liposomal membranes.
- Penetration of multilamellar vesicles via the lipid-rich channels. Multilamellar vesicles may lose one or more of their outermost lipid lamellae during the path of penetration, partial release of the encapsulated material.

Liposomes can be absorbed on to the skin surface intact before their penetration into the skin. Some liposomes can rupture on the surface of the skin. The penetration of smaller vesicles is more probable. However, it is possible that the intradermally localized uni- or oligolamellar vesicles are derived from multilamellar liposomes, which lost their outer bilayers during penetration. (Foldvari et al., 1990)

The aimed of this research work. To prepare captopril liposomal gel and characterize it's physicochemical and functional attributes for sustained and prolonged drug delivery. The objective of this research work. To provide with sustained and prolonged release of drug which lead to less dosing frequency. To enhance and improve the bioavailability of drug.

II. MATERIALS AND METHODS

A. Materials

Captopril was pucharsed from the Dhamtec Pharma and Consultants Mumbai. Phospholipon 90H was obtained from Lipoid kosmetic, Germany. Cholesterol Was obtained from Research-lab Fine Chem Industries, Mumbai. Stearylamine was obtained from National Chemicals, Vadodara.

B. Experimental Design (3^2 full factorial design)

To minimized the number of trials and attain the highest amount of information on product properties, the screening was done by applying full factorial design systematically study the joint influence of the independent variables on the dependent variables. So, in this study two factors were evaluated and each at three levels and experimental trials were performed at all nine possible combinations. For liposomes, Amount of (Cholesterol 5, 10 and 20 mg), was taken as the first independent variable (X1, mg) and amount of (Stearylamine 2.5, 5.0, 10) was selected as the second independent variables (X2, mg) for liposomes. These variables varied at three levels, i.e., low level (-1), medium level (0), and high level (+1). All the calculations data were done at (mg) milligram level. The volume of diethyl ether and chloroform in (1:1) ratio and amount of CPT (10 mg) were constant. Percent entrapment efficiency (% EE) (Y1) and percent in vitro drug release at 10 h (% DR) (Y2) were selected as dependent variables. Values of variables and batch codes are shown in table I.

Table I: 3^2 full factorial designs: factors, factor levels and responses for CPT LP_S formulation

Factors (Independent variables)			
Factor levels used	Low (-1)	Medium (0)	High (+1)
Amount (mg) of Cholesterol (CH) (X_1, mg)	5	10	20
Amount (mg) of Stearylamine (X_2, mg)	2.5	5.0	10
Responses (Dependent variable)			
Y1 = Percent entrapment efficiency (% EE)	Minimum		Maximum
Y2 = Percent in vitro drug release (% DR)	Minimum		Maximum

Design Expert® DX 13 (Stat-Ease Inc., MN) license version software was used for the generation and evaluation of statistical experimental design.

C. Statistical analysis and optimization of formulation using RSM

Response surface modeling and evaluation of the quality of fit of the model for the current study were performed employing Design Expert® DX 13 license version software. Polynomial models including linear, interaction and quadratic terms were generated for all the response variables using multiple linear regression analysis (MLRA). A second-order Polynomial equation that describes the effect of independent factors on the response is expressed in the following forms:

Linear model: $Y = \beta_0+b_1X_1+b_2X_2$ (1)

2FI (interaction model): $Y = \beta_0 +\beta_1 X_1+\beta_2 X_2 +\beta_{12}X_1 X_2$ (2)

Quadratic model = $\beta_0+\beta_1X_1+\beta_2X_2+\beta_{12}X_1X_2+\beta_{11}X_1^2+\beta_{22}X_2^2$ (3)

Where Y is the dependent variable; β_0 is the arithmetic mean response of the nine runs and β_i (β_1; β_2; β_{12}; β_{11} and β_{22}) is the estimated coefficient for the corresponding factor X_i (X_1, X_2, X_1X_2, X_1X_1, and X_2X_2). The main effects (X_1 and X_2) represent the average result of changing one factor at a time from its low to high value. The interaction terms (X_1X_2) show how the response changes when two factors are simultaneously changed. The polynomial terms (X_1^2 and X_2^2) are included to investigate nonlinearity. The equations enable the study of the effects of each factor and their interaction over the considered responses. The polynomial equations were used to draw conclusions after considering the magnitude of coefficients and the mathematical sign they carry, i.e., positive or negative. A positive sign signifies a synergistic effect, whereas a negative sign stands for an antagonistic effect. The best fitting mathematical model was selected based on the comparisons of several statistical parameters, including the coefficient of variation (CV), the coefficient of determination (R^2), adjusted coefficient of determination (Adjusted R^2) and the predicted residual sum of square (PRESS), provided by Design Expert software. Among them, PRESS indicates how well the model fits the data and for the chosen model it should be small relative to the other models under consideration. Level of significance was considered at $p<0.05$. Mathematical

relationship in the form of polynomial equations is generated using multiple linear regression analysis (MLRA) and used to find out the relative influence of each factor on the response. Analysis of variance (ANOVA) for the responses was performed to identify a significant effect of factors on responses and the model parameters were obtained. Two-dimensional contour plots and three-dimensional response surface plots resulting from equations were obtained by the Design Expert software.

These plots are very useful in a study of the effects of two factors on the response at one time and predict the responses of dependent variables at the intermediate levels of independent variables. Subsequently, a numerical optimization technique by the desirability and graphical optimization technique by the overlay plot approach were used to generate the new formulation with the desired responses. An optimized formulation was developed by setting constraints (goals) on the dependent and independent variables. To validate the chosen experimental design, the resultant experimental values of the responses were quantitatively compared with those of the predicted values and calculated the percent relative error (PRE) by the following equation. (Tatode et al., 2018)

$$\%\ \text{Relative error} = \frac{\text{Predicted value} - \text{Experimental value}}{\text{Prdicted value}} \times 100$$

D. Preparation of Liposomes

Ether injection method

The liposomes of Captopril with phospholipon 90 H, cholesterol, and stearylamine were prepared using ether injection method. Firstly, phospholipon 90 H, cholesterol and stearylamine were co-dissolved in the mixture of diethyl ether (2.5ml) and chloroform (2.5ml) at room temperature. The resulted solution was slowly injected through 14-gauge needle into a beaker containing Captopril in 10 ml distilled water the temperature maintained during the injection was 40-60 ˚C. The difference in temperature between phases cause rapid vaporization of ether, chloroform resulting in spontaneous vesiculation and also formed milky white suspension. The liposome dispersion samples were kept at 4˚C and protected from light. The liposome dispersion was then centrifuged at 24,000 rpm, 4˚C for 3 hrs. Purified liposomes were collected by discarding supernatant. There are number of processing parameters that is drug: cholesterol: stearylamine concentration in the formulation was considered with the aim of achieve best possible preparation of captopril liposomal gel. (Ramya et al., 2021)

III. FORMULATION OF LIPOSOMAL GEL

Gel was prepared using Carbopol 934 (1.0, 1.5, 2.5, and 3%). The appropriate quantity of Carbopol 934 powder was dispersed into distilled water under constant stirring with a glass rod, taking caution to avoid the development of indispensable lumps, and allowed to hydrate for 24 h at room temperature for swelling. Topical liposome gel formulations were prepared by incorporation of liposome's containing CPT and were mixed into the carbopol gel with a mechanical stirrer (25 rpm, 2 min). The dispersion was neutralized using triethanolamine (0.5%w/w). Control gels were made under the same conditions. (Princely et al., 2018) (Thorat et al., 2020)

Table II: Formulation of CPT LP_s gel and CPT gel

Sr. No	Components	CPT LP_s gel	CPT gel
1	Equivalent to 10 mg	CPT Liposomes	CPT
2	Carbopol 940	1.0%	1.0%
3	Triethanolamine	q. s	q. s
4	PEG 400	2ml	2ml
5	Methyl paraben	q. s	q. s
6	Propyl paraben	q. s	q. s
7	Water	q. s	q. s

A. Determination of Entrapment Efficiency (EE_s)

The 2ml of liposome suspension was ultra-centrifuge at 8000 rpm for 1 hrs. At 4˚C temperature by using Remi centrifuge to separate the free drug. Supernatant containing liposomes in suspended stage and free drug at the wall of centrifugation tube. The supernatant was collected and again centrifuge at 8000 rpm at 4˚C temperature for 30 minutes. A clear solution of supernatant and pellet containing only liposomes was resuspended in distilled water until further processing. The liposomes free form unentrapped drug was soaked in 10 ml of methanol and then sonicated for 10 min. The vesicles were broken to release the drug. The absorbance of the drug was noted at 205.6nm. The

entrapment efficiency was calculated using following equation.(Patel et al., 2009)

$$\% \text{ Entrapped efficiency} \frac{\text{Entrapped drug}}{\text{Total drug added}} \times 100\text{D}$$

B. Determination of Drug Content

For drug content analysis, 10 mg drug containing captopril liposomes were dissolved in methanol and filtered. The volume was made to 10 ml with methanol. The resultant solution was suitably diluted with methanol and absorbance was measured at 205.6 nm using UV spectrophotometer (JASCO UV Spectrophotometer Model No V-630). (Afreen et al., 2021) (Alur A et al., 2017)

$$\% \text{ Drug conten} = \frac{\text{Drug conc. obtained}}{\text{Total drug added}} \times 100$$

IV. PHYSICOCHEMICAL CHARACTERIZATION OF CPT LP_S

A. Photomicroscopy:

Surface morphology of the prepared CPT LP_s was characterized using photo microscope examination. All batches of the liposomes prepared were viewed under photomicroscope. The optimized batch of liposomal dispersion was gently stirred, placed a drop of dispersion onto glass slides. The microscopic images of the sample were viewed on a microscope. (Model: DM 1802) and images were captured using an attached digital camera.

B. Zeta Potential analysis:

A dynamic light scattering (Malvern Instruments) was used to measure zeta potential of the liposomal formulation. The sample was analyzed at 25˚c with a detection angle of 90˚. The zeta potential of the prepared liposomal formulation was measured in disposable folded capillary cells. Samples should be bubble free for accurate measurement of zeta potential. All measurements were performed in triplicate. The measurements were conducted at room temperature within the equipment sensitivity range of 200 to 200mv.

C. Particle size analysis:

The particle size analysis of optimized CPT LP_s formulation was carried out using dynamic light scattering method. The liposomal dispersion was examined using a particle size analyzer. The equipment was sensitive in the range of 1nm to 10um, the flexibility of positioning the sample to optimize the count rate using the associated software.

D. Field Emission Scanning Electron Microscopy (FESEM)

Approximately 5ug/ml liposomes were transferred to a cover slip, which in turn was mounted on a specimen tab. The samples were allowed to dry at room temperature. The particle size of the formulation was viewed and photographed using Field emission scanning electron microscope. The particles were coated with platinum by using vacuum evaporator and thus, the coated samples were viewed and photographed in JEOL JSM-7610F, Field Emission FESEM.

E. Fourier Transform Infrared Spectroscopy (FTIR)

Captopril, Phospholipon 90H, Cholesterol, Stearylamine, lyophilized Optimized CPT LP_S (F5) batch were further chemically characterized and compared using FTIR spectroscopy. The samples were tested using FTIR spectrophotometer. Sample preparation involved preparing potassium bromide (KBr) pellets containing ~2mg air dried samples. Thin transparent discs of KBr were compressed on a Mini Hand Press at pressure of 10 ton/Nm^2, individual samples discs were scanned within a wavelength ranging from 4000 to 400 cm^{-1} with a scan resolution of 4 cm^{-1} the obtained spectra were analyzed by the software associated with the instrument.

V. FUNCTIONAL CHARACTERIZATION

A. In-vitro diffusion study:

Procedure for dialysis membrane activation

Cut the dialysis membrane and immersed in a boiling 2% sodium bicarbonate ($NaHCO_3$) solution for 10 min and rinsed with distilled water. Then this membrane immersed in boiling water for 10 min, rinsed with fresh distilled water and kept in distilled water for 24 hours before use.

Procedure for In-vitro diffusion study:

An in vitro drug release test of the liposome suspension was performed using Franz diffusion cells with dialysis membrane. A dialysis membrane (LA393-5MT, dialysis membrane -70, average flat width 29.31 mm, average diameter 17.5 mm and capacity 2.41 ml/cm; (HIMEDIA Laboratories,

Mumbai, India), which used as diffusion barrier. The donor compartment contained a 5ml of CPT solution contained (10mg CPT) and 5ml of prepared CPT LP_s contained (5 mg CPT) while receptor compartment was filled with 20 ml phosphate buffer solution (pH 7.4, 20ml). The donor chamber was placed in such a way that it just touched the diffusion medium in the receptor chamber. The temperature was maintained constant at $37 \pm 1^{\circ}$c using magnetic stirrer (50 rpm). At predetermined time intervals, samples (5ml) were periodically withdrawn from the receptor compartment, replaced with the same amount of fresh pre-warmed buffer solution, and assayed using UV spectrophotometer (Model: V-630, JASCO International Co. Ltd., Tokyo, Japan) at 216 nm for CPT. After suitable dilution and the concentration of CPT were calculated using calibration curve. (Ramya et al., 2021)

B. Drug release kinetics

Drug release data was evaluated using the Higuchi square root, Hixson Crowell, and Peppas equations, as well as the zero-order, first-order, and Korsmeyer peppas in order to determine the correct mechanism of drug release from liposomes. The criteria for selecting the most appropriate model were chosen on the basis of goodness of fit test. The zero order kinetics describes that system in which the drug release rate is not dependents by its concentration are referred to as zero-order kinetics (equation 1). The systems in which the drug release rate is concentration dependent are referred to as first order kinetics (equation 2). According to Fickian diffusion, Higuchi defined the release of a medication from an insoluble matrix as a square root of the time-dependent process (equation 3). The Hixson Crowell cube root law describes the drug release from systems in which there is a change in the surface area and the diameter of particles present in the formulation (equation 4). When more than one type of release phenomenon may be present or when the release mechanism is unknown, Korsmeyer Peppas equation describes the release (equation 5).

$R = k_0 t$ …… (1)

$\text{Log UR} = k_1 t\ 2.303$ …… (2)

$R = k_2 t^{1/2}$ …….. (3)

$(UR)^{1/3} = k_3 t$ …… (4)

$\text{Log } R = \log k_4 + n \log t$ …… (5)

Where R and UR represent for, respectively, the released and unreleased percentages at time t. And for the rate equations of orders zero order, first order, higuchi, hixson-crowell, and peppas-korsmeyer, respectively, Ko, K_1, K_2, K_3, and K_4 are the release rate constants. (Bramhankar et al., 1995)

VI. PHYSICOCHEMICAL CHARACTERIZATION OF CPT LP_S GEL

A. Determination of pH

The pH meter was calibrated with buffered solution at 4.0, 7.0 and 9.2 before starting pH determination. The glass electrode of the pH meter was immersed into 50 ml beaker containing 10 gel and pH was noted.

B. Viscosity Measurement

Viscosity of the gel was determined by using Brookfield viscometer. Accurately weighted gel in (gm) was transferred to 100ml glass beaker. Spindle no S64 was selected and it is immersed into the gel. The viscometer was operated at various rpm until the reading gets stabilized and reading was noted in centipoises.

C. Spreadability

The Spreadability of the formulation was determined by an apparatus suggested by muttimer et al., which was suitably modified in the laboratory and used for the study. It consists of wooden block; which was provided by a pulley at one end. Two sets of glass slides of standard dimensions were taken. The CPT LP_S gel formulation was placed over one of the slides. The other slide was placed on the top of the gel, such that the gel was sandwiched between the two slides in an area occupied by a distance of 7.5 cm along the slide. 100 g weight was placed upon the upper slides so that the gel between the two slides was pressed uniformly to form a thin layer. The weight was removed and the excess of gel adhering to the slides was scrapped off. The two slides in position were fixed to a stand without slightest disturbance and in such a way that only the upper slide to slip off freely by the force of weight tied to it. A 20 g weight was tied to the upper slide carefully. The time taken for the upper slide to travel the distance of 7.5 cm and separated away from the lower slide under the influence of the weight was noted. The experiment was repeated by three times and the mean time taken for calculation. (Haneefa et al., 2010)

$S = m \times l/t$

Where,
S –Spreadability
m - Weight tied to the upper slide (20 g)
l - Length of the glass (7.5 cm)
t - Time taken in seconds.

D. Drug content

Accurately weighed 1g of gel was transferred into volumetric flask in methanol. After suitable dilution the absorbance was measured using (JASCO UV Spectrophotometer Model No V-630). at 205.6nm. (Basha et al., 2011)

$$\text{Drug content} = \frac{\text{Concentration} \times \text{Total formulation} \times \text{Dilution factor}}{1000}$$

$$\%\ \text{Drug content} = \frac{\text{Drug content}}{\text{Total drug}} \times 100$$

VII. FUNCTIONAL CHARACTERIZATION

A. In-vitro diffusion study:

Procedure for dialysis membrane activation

Cut the dialysis membrane and immersed in a boiling 2% sodium bicarbonate ($NaHCO_3$) solution for 10 min and rinse with distilled water. Then this membrane immersed in boiling water for 10 min, rinsed with fresh distilled water and kept in distilled water for 24 hours before use.

Procedure for In-vitro diffusion study:

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VIII. RESULTS AND DISCUSSION

A. Experimental design and data acquiring (3^2 full factorial design)

Experimental design and data acquiring (3^2 full factorial design) Full factorial design (3^2) was applied to optimize the CPT LP_S formulation. All nine batches of CPT were prepared according to the formulation variables as. Liposomes were obtained by the ether injection method. RSM was exploited to estimate the influence of the cholesterol and stearylamine in mg as independent variables and their interactions on the investigated responses (dependent variables; % EE and % DR). This experiment was aimed to identify considerable factor effect influencing the formulation performance and to set up to their excellent levels for the desirability of responses shown (Table III) Statistical analysis and optimization of formulation using RSM To evaluate the quantitative effects of factors (X1 and X2) and their levels low (-1), middle (0), and high (+1) on the preferred responses, the experimental values of the flux were analyzed by Design Expert® DX 13 license version software and mathematical models obtained for each response. The mathematical relationship generated using multiple linear regression analysis (MLRA) for the studied response variables (% EE and % DR at 10 h.) that were relating different response and independent variables are expressed as following polynomial equations (quadratic model).

Y_1 (%EE) $=95.55+15.16X_1 + 4.48X_2 – 5.93X_1X_2-29.33X_1^2+0.6800X_2^2$ (1)

Y_2 (%DR at 10h) $= 38.23-8.29X_1-4.42X_2+2.80X_1X_2+18.96X_1^2+8.11X_2^2$ (2)

The above equations expose the quantifiable effect of the independent variables, of cholesterol and stearylamine,

Table III: Composition 3^2 full factorial design with measured responses of CPT formulation.

Batches	Variable level in coded form		Variable level in actual form		Response variables	
	X_1	X_2	Cholesterol (X_1) mg	Stearylamine (X_2) mg	Percentage entrapment efficiency (%EE) ±SD	Percentage In-vitro drug release (10h) (%DR) ±SD
F1	-1	-1	5	2.5	37.71 ± 0.78	81.33±0.32
F2	-1	0	5	5.0	48.99 ± 0.12	68.28±0.65
F3	-1	+1	5	10	64.44 ± 0.56	66.37±0.98
F4	0	-1	10	2.5	81.66 ± 1.54	56.5±2.31
F5	0	0	10	5.0	84.37±0.89	55.03±0.6
F6	0	+1	10	10	88.33±0.71	46.23±1.45
F7	+1	-1	20	2.5	86.92 ± 0.67	58.16±0.32
F8	+1	0	20	5.0	74.77 ± 1.68	51.16±1.25
F9	+1	+1	20	10	91.00±0.45	45.48±0.36

On the responses such as % EE (Y1) and in vitro % DR at 10 h (Y2) as dependent variables. The fitted polynomial equation (quadratic model) related to % EE and percent in vitro % DR used to draw a conclusion after considering the coefficient and the mathematical sign it carries. That is positive and negative. The correlation coefficient (r^2) of the quadratic model (0.9412) for response Y1 (% EE) and (0.9983) for response Y2 (% DR) was found to be significant.

B. Response 1 (Percent entrapment efficiency) (% EE)

Regression analysis of above equation (1) of response Y1 (% EE) revealed that the coefficient of β1 was positive and β2 was positive, this indicated that as CH (X_1) and stearylamine (X_2) gets increased at certain levels then %EE increased. Because stearylamine help to increase the spacing between phospholipids and cholesterol bilayer causing an increase in the volume of aqueous compartment and thus amount of hydrophilic drug more entrapped. But as

Table IV: Analysis of variance (ANOVA) table of %EE

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	2687.23	5	537.45	9.60	0.0459	significant
A-Cholesterol	1355.83	1	1355.83	24.22	0.0161	
B-Stearylamine	118.54	1	118.54	2.12	0.2416	
AB	151.23	1	151.23	2.70	0.1988	
A^2	1310.74	1	1310.74	23.42	0.0168	
B^2	0.7046	1	0.7046	0.0126	0.9178	
Residual	167.93	3	55.98			
Cor Total	2855.16	8				

we further increased the quantity of the cholesterol then %EE gets decreased. The higher concentration of cholesterol leads to rigidity in the vesicles. Which in turn decreased the % EE. The % EE of different liposomal batches was in a range of 37.71±0.78% to 91.00±0.45 %. The maximum entrapment was observed in batch F9 (table no III) with composition of CH: Stearylamine with the amount (mg) of 20:10 (+1, +1).

Factor coding is Coded**.**

Sum of squares is Type III - Partial

The Model F-value of 9.60 implies the model is significant. There is only a 4.59% chance that an F-value this large could occur due to noise.

P-values less than 0.0500 indicate model terms are significant. In this case A, A^2 are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve your model.

Table V: Parameter of selected quadratic model of %EE

Std .dev	7.48	R-squared (r^2)	0.9412
Mean	73.13	Adjusted R^2	0.9296
C.V.	10.23	Predicted R^2	0.7315
PRESS	2895.96	Adequate Precision	15.693

A negative Predicted R^2 implies that the overall mean may be a better predictor of your response than the current model. In some cases, a higher order model may also predict better.

Adeq Precision measures the signal to noise ratio. A ratio greater than 4 is desirable. Your ratio of 8.684 indicates an adequate signal. This model can be used to navigate the design space.

The relationship between the dependent and independent variables was further elucidated using contour and response surface plots. The contour (figure I) and 3D response surface plots (figure II) of % EE clearly indicated that X1 and X2 highly influenced the response 1 (%EE). The change in % EE as a function of X1 and X2 was depicted in the form of contour and response surface plots based on full factorial design. So, higher level of X1 and higher level of X2 was found to be favorable conditions for obtaining higher % EE.

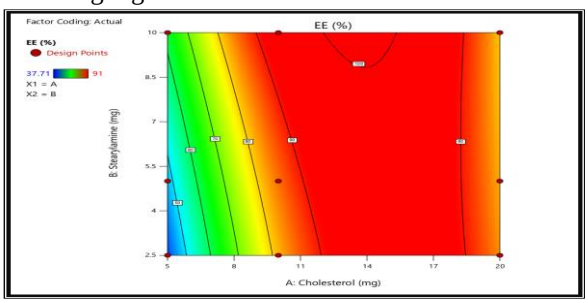

Figure I: Counter plot showing the effect of cholesterol (X1) and Stearylamine (X2) on % EE (Y1) of CPT LP_s

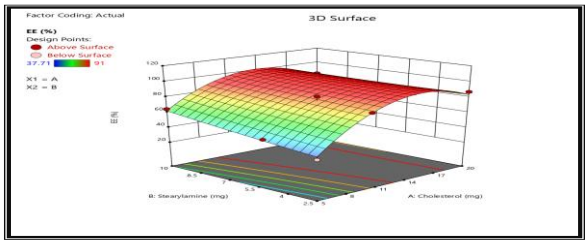

Figure II: Response surface plot showing the effect of showing the effect of cholesterol (X1) and Stearylamine (X2) on % EE (Y1) of CPT LP_s

B. Response 2 (Percent drug release at 10 h) (% DR)

The effect on drug release at 10 h (% DR) (Y2) was observed to be significant ($P<0.05$) by ANOVA and the polynomial equation (2) revealed that the coefficient of β_1 was negative and β2 was negative, this indicated that as Cholesterol (X_1) at optimum level the %DR decreased. And on optimum amount of stearylamine (X_2) which also decreased the % drug release. If we increased the amount of stearylamine the %DR increased. The release was decreased at higher levels of cholesterol. This is because cholesterol at higher levels makes the lipid bilayers more rigid and retards the release of the drug. The F9 formulation was found to 45.48±0.36% DR at 10 hrs with composition of CH: Stearylamine with the amount (mg) of 20:10. To envisage the effect of an independent factor on the response (Y2) the contour plot (figure III) and 3D-response surface plots (figure IV) of % DR at 10 h shows the curvature with a change in the factor (X1 and X2). The plot was found into which indicated that 45.48±0.36% DR at 10 hrs can be obtained for a combination higher level of X_1 and higher level of X_2 factors.

Factor coding is Coded.

Sum of squares is Type III – Partial

The Model F-value of 358.37 implies the model is significant. There is only a 0.02% chance that an F-value this large could occur due to noise.

P-values less than 0.0500 indicate model terms are significant. In this case A, B, AB, A^2, B^2 are significant model terms.

Table VI: ANOVA table of % DR at 10 hr

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	1066.65	5	213.33	358.37	0.0002	significant
A-Cholesterol	405.72	1	405.72	681.57	0.0001	
B-Stearylamine	114.99	1	114.99	193.17	0.0008	
AB	33.77	1	33.77	193.17	0.0049	
A^2	547.78	1	547.78	56.73	<0.0001	
B^2	100.13	1	100.13	920.21	0.0010	
Residual	1.79	3	0.5953	168.21		
Cor Total	1068.44	8				

Values greater than 0.1000 indicate the model terms are not significant. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve your mode.

Table VII: Parameter of selected quadratic model of %DR AT 10h

Std .dev	0.7715	R-squared (r^2)	0.9983
Mean	58.73	Adjusted R^2	0.9955
C.V.	1.31	Predicted R^2	0.9720
PRESS	29.91	Adequate Precision	55.5942

The Predicted R^2 of 0.9720 is in reasonable agreement with the Adjusted R^2 of 0.9955; i.e. the difference is less than 0.2.

Adeq Precision measures the signal to noise ratio. A ratio greater than 4 is desirable. Your ratio of 55.594 indicates an adequate signal. This model can be used to navigate the design space.

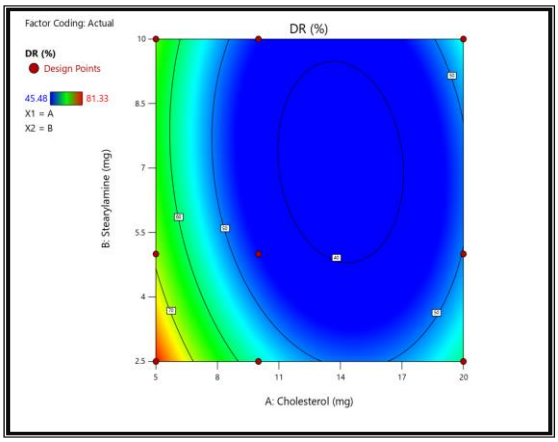

Figure III: Counter plots showing the effect of cholesterol (X1) and Stearylamine (X2) on % DR (Y2) of CPT LP_s

Table VIII: Characteristics of optimum formula

Objects	Cholesterol in mg (X_1)	Stearylamine in mg (X_2)	%EE (Y_1, %)	In vitro % DR at 10 h	Desirability	
Predicted	17.929	9.772	91.920	46.90		Selected
Actual (F9)	20	10	91.00 ± 0.45	45.48±0.36	1.000	

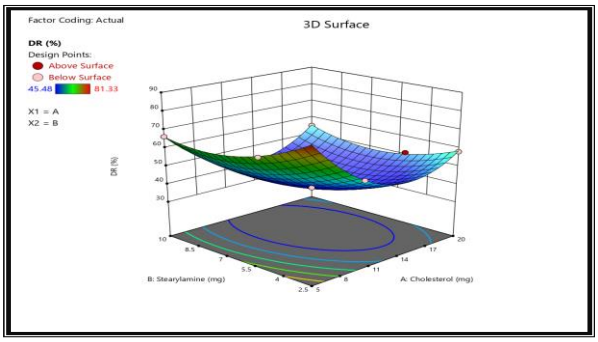

Figure VI: Response surface plot showing the effect of showing the effect of cholesterol (X1) and Stearylamine (X2) on% DR (Y2) of CPT LP_s

C. Desirability and overlay plot

The aim of pharmaceutical formulation optimization is generally to find the levels of the variable that affect the chosen responses and determine the levels of the variable from which a robust product with high-quality characteristics may be produced. All the measured responses that may affect the quality of the product were taken into consideration during the optimization procedure. The % EE and % DR at 10 h were set out in the maximum for %EE and in range %DR criteria. Each response criterion was combined (overlay plot) to obtain the optimum value (figure V). The optimization results of this research can be seen in table VIII.

Validation of RSM results

In order to evaluate the optimization capability of models generated according to the results of the RSM (3^2 full factorial design), CPT LP_S formulation was prepared using the optimal process variables settings that X_1 and X_2 were equal 20:10 (mg). The response Y_1 (% EE) and Y_2 (% DR at 10 h) obtained with predicted models and the experimental model were shown in table IX. The percent relative error was obtained using formula of (PRE). The percent relative error (PRE) for response Y_1 (% EE) and Y_2 (% DR at 10 h) were found to be (1.00) and (2.00) respectively. The maximum PRE value was (1.00). However, the values were found to be<2 %and hence it confirmed the suitability of experimental design. The results showed good agreement on preparation properties on theoretical properties. The optimization parameter of desirability was determined by regulating the optimum input variables to obtain one or more optimal parameters. The desirability value ranged between 0 and 1, where a value of 1 is perfect, i.e., the ideal parameter value. The CPT LP_S desirability plot was shown in figure VI. The optimizing desirability of CPT LP_S formulation was 1.000. This value was ideal value (1), meaning that the predicted parameters were desired parameter values. The composition of the

predicted formulations was nearly matching with F9 CPT LP_S (table VIII).

Table IX: Validation of predicted and experimental CPT LP_s batch

Response	Experimental values	Predicted values	% Relative error (PRE)
Y_1 (%EE)	91.00 ± 0.45	91.920	1.00
Y_2 (%DR at 10h)	45.48±0.36	46.90	2.00

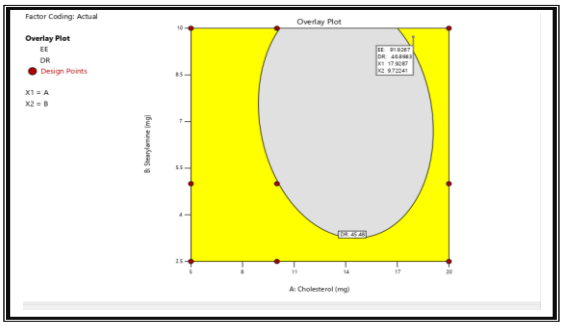

Figure V: Overlay plot of % EE and % DR at 10 h

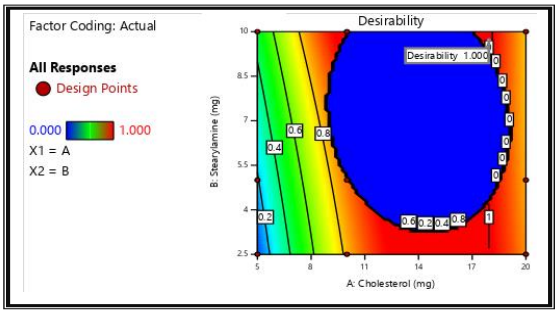

Figure VI: CPT Liposome's desirability plot

IX. ESTIMATION OF ENTRAPMENT EFFICIENCY AND DRUG CONTENT

Results of Captopril in prepared Captopril liposomes formulation are shown in table X. The entrapment efficiency and drug content of captopril in all prepared captopril liposomes formulations i.e. F1 to F9, was found to be in the range 37.71± 0.78 to 91.00 ± 0.45% w/w and 16.41± 0.89 to 39.11± 1.76 %w/w respectively. However, the F9 batch (liposomes prepared with the 1:2:1 drug: cholesterol: stearylamine ratio) exhibited highest entrapment efficiency (91.00 ± 0.45% w/w) and drug content (39.11 ± 1.76), and thus selected as optimized formulations were preferred for further assessments due to their highest entrapment efficiency and drug content and also from full factorial design.

Table X: Entrapment efficiency and Drug content of batch F1 to F9

Sr. No.	Formulation code	Entrapment Efficiency (% w/w)	Drug Content (%w/w)
1	F1	37.71 ± 0.78	16.41 ± 0.89
2	F2	48.99 ± 0.12	29.48 ± 1.79
3	F3	64.44 ± 0.56	32.85 ± 0.12
4	F4	81.66 ± 1.54	38.73 ± 0.85
5	F5	84.37 ± 0.89	34.17 ± 0.88
6	F6	88.33 ± 0.71	35.61 ± 1.54
7	F7	86.92 ± 0.67	34.80 ± 0.73
8	F8	74.77 ± 1.68	24.55 ± 0.98
9	F9	91.00 ± 0.45	39.11 ± 1.76

X. PHYSIOCHEMICAL CHARACTERIZATION OF CPT LPS

A. Photomicroscopy

The morphological characterization of prepared CAP LP_s (optimized) is shown in figure (VII). CAP LP_s appeared as a small circular/ spherical shaped at 10x magnification by using motic microscope. (model: DM-1802).

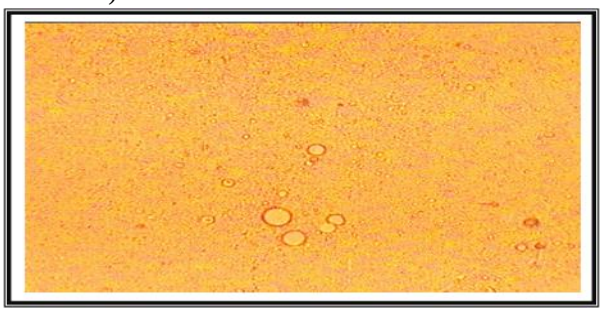

Figure VII: The microscope characterization of optimized CAP LP_s

B. Field emission scanning electron microscope (FESEM)

FESEM images, microscopic evaluation showed that most of the vesicles were spherical shape as shown in Figure VIII. For better skin permeation the vesicle size must be of 100-400 nm. The diameter (nm) of optimized CPT LP_s (batch F9) was found to be 48.4 to 72.0 nm

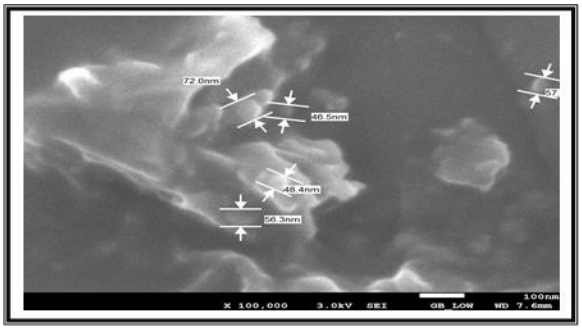

Figure VIII: The Field emission scanning microscope image of (F_9 Batch)

C. Zeta potential

Zeta potential (ζ) is the overall charge a lipid vesicle acquires in a particular medium. It is a measure of the magnitude of repulsion or attraction between particles in general and lipid vesicles in particular. Zeta potential is also useful in controlling the aggregation, fusion and precipitation of nanoliposomes, which are important factors affecting the stability of nanoliposomal formulations. The zeta potential values in the range of -30 mV to +30 mV are generally considered to have sufficient repulsive force to attain better physical colloidal stability. The zeta potential value for optimized CPT LP_s (F_9 Batch) + 31.5 ± 0.41 mV which found within the desired range as above mentioned. Therefore, suitable zeta potential value demonstrates the suitability of optimized LPs formulation for transdermal administration accompanied with an excellent physical stability.

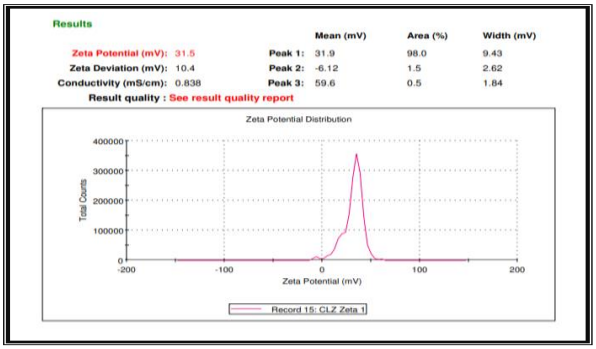

Figure IX: Zeta potential of optimized CPT LP_s (F_9 Batch)

D. Particle size

The particle size is valuable predictors of effective distribution. According to Ravi Kumar et al., 2012 the particle size 1 to 10µm (1-10,000 nm), indicating narrow size distribution. Such particles sizes and narrow size distributions were considered favorable for transdermal administration. In the present study the formulation prepared by ether injection method displayed particle size at about 817.0 ± 0.45nm with polydispersity index (PDI) i.e. 0.290 ± 0.08 for optimized CPT LP_s (batch F9) is appropriate for transdermal drug delivery. The low PDI indicates narrow range of particle size distribution.

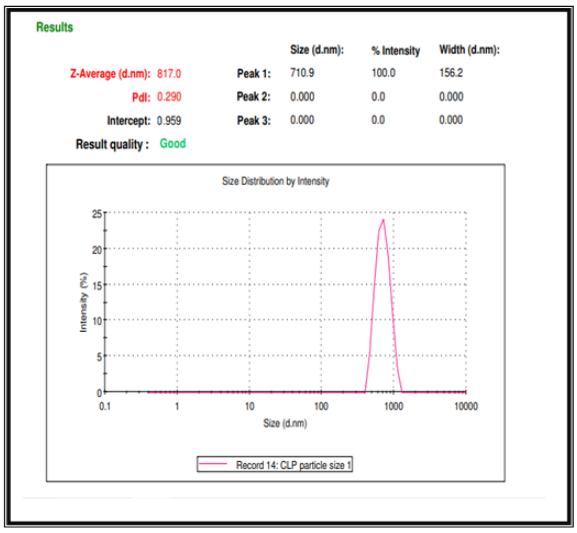

Figure X: Particle size of optimized CAP LP_s (F_9 Batch)

E. Fourier transforms infrared spectroscopy (FT-IR)

The spectra obtained from the FTIR analysis of Captopril, Phospholipon-90H, Cholesterol, Stearylamine, Optimized CPT LP_s (batch F9) are shown in Figure XI to XII. The IR Spectra did not show any significant difference from those obtained for their optimized CAP LP_s (F_5 Batch). This obtained result indicates that there was no positive evidence for the interaction between captopril and the utilized materials. These results clearly indicated that usefulness of utilized materials for the preparation of liposomes of captopril. These observations were compared with those earlier (Singh et al., 2012).

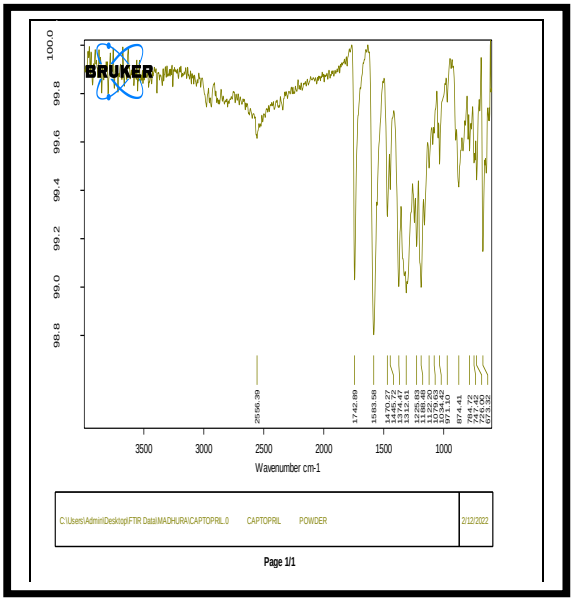

Figure XI: FTIR spectra of captopril

Table XI: FTIR interpretation of captopril

Sr. No.	Types of vibration	Intensity and region (cm^{-1})
1.	O-H Bending	1445.72
2.	S-H Stretching	2556.39
3.	C=O Stretching	1742.89
4.	C-N Stretching	1374.47
5.	S-H Bending	1188.48
6.	C-H Bending	874.41

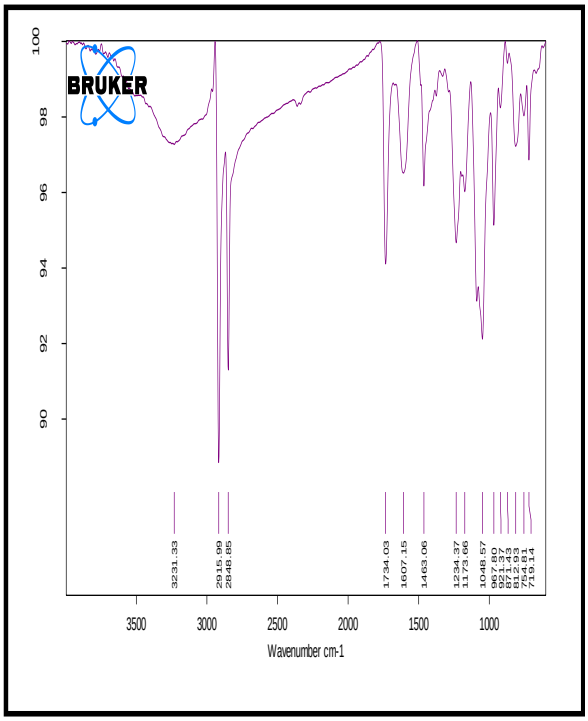

Figure XII: FTIR spectra of optimized CAP LP_s (F_9 Batch)

Table XII: FTIR interpretation of optimized CAP LP_s (F_9 Batch)

Sr. No.	Types of vibration	Intensity and region (cm^{-1})
1.	C=O Stretching	1734.03
2.	C-N Stretching	1048.57
3.	S-H Bending	1173.66
4.	C-H Bending	871

XI. FUNCTIONAL CHARACTERIZATION OF CAP LPS

A. In-vitro diffusion studies

The results of the comparative diffusion profiles of CPT and CPT LP_s formulations that is batch F1 to F9 in phosphate buffer saline (7.4) are shown in figure. XIII to XVI The release profiles of CPT appeared up to 94.09±1.03 at 30 minutes. This fastest amount of release in diffusion media could be due to short half life of CPT. Prepared CPT LP_s formulations were showed an sustained release of drug for prolonged period of time. CPT LP_s formulations i.e. batch F5, F6, F7, F8 and F9 demonstrated a sustained release of drug as compared to CPT. Amongst these formulations Batch F9 showed an excellent sustained release rate at about 54.84±2.63% up to 12 hrs and 45.48±0.36 at 10 hrs. CPT LP_s formulation showed a sustained release due to addition of cholesterol and stearylamine which has been shown to increase liposome stability and also increased retention time of drug by modulating phospholipid packing.

B. Drug Release kinetics

In vitro release data was fitted to kinetic models like, zero order, higuchi, Hixson crowell and Korsemeyer–Peppas to elucidate drug release pattern and captopril release mechanism from liposomes. In this investigation, in vitro release of drug from all formulations followed zero-order model with R^2 values of 0.997 of F3 and R^2 value of optimized batch F9 was found to be 0.991. Which showed drug release is not dependent on initial concentration of drug. To confirm drug release phenomenon, release data was evaluated by Korsemeyer–Peppas equation. Additionally, the release exponent value (n) was estimated to be 1.430. “n” value was found be higher than 1. Thus, indicating super case II transport mechanism. It was found to be erosion and relaxation mechanism of drug release. According to the rate constant of zero order the optimized batch F9 shows lower rate constant value that is 5.195 as compared to other batch. According to this the optimized batch was found to be a sustained release with less rate of constant. Hence captopril liposomes were significantly follows sustained release. This zero order is useful for transdermal drug delivery.

Table XIII: In-vitro diffusion profile of drug

Time (hrs)	Drug
0	0±0
0.1h	21.99±0.26
0.2h	76.64±0.52
0.5h	94.09±1.03

Table XIV: In-vitro diffusion profile of F9 batch

Time (hrs)	F9 Batch
0	0±0
1h	0.1±0.34
2h	1.47±0.42
3h	5.27±0.78
4h	12.6±1.51
6h	20.87±2.36
8h	31.21±1.25
10h	45.48±0.36
12h	54.84±2.63

Table XV: In vitro diffusion profiles of CPT liposomes formulation (F1-F9)

Time (hr)	FORMULATION CODE								
	F1	F2	F3	F4	F5	F6	F7	F8	F9
0	0±0	0±0	0±0	0±0	0±0	0±0	0±0	0±0	0±0
1	8.38±0.35	2.68±0.34	2.21±0.2	1.3± 0.14	0.9±0.45	1.31±0.15	0.07±0.01	1.72±0.01	0.1±0.12
2	10.65±2.15	6.08±0.42	7.78±1.90	6.72±1.71	5.79±0.5	2.52±1.36	6.13±1.22	7.02±1.51	1.47±0.42
3	21.46±1.32	18.37±1.71	19.44±1.13	8.65±0.46	7.18±0.40	6.83±1.34	14.53±0.51	14.5±0.78	5.27±0.78
4	30.01±2.22	25.29±1.51	25.39±2.40	15.47±1.25	16.79±1.31	14.13±2.25	23.99±0.64	22.1±0.81	12.6±1.51
6	44.6±2.36	36.28±0.81	38.55±2.13	23.68±0.26	27.62±0.73	21.75±1.2	35.77±1.88	31.38±1.5	20.87±2.36
8	63.16±0.16	51.63±2.36	52.18±0.38	41.83±2.03	42.68±1.39	31.67±0.65	45.78±1.17	39.34±2.36	31.21±1.25
10	81.33±0.32	68.28±0.65	66.37±0.98	56.5±2.31	55.03±0.6	46.23±1.45	58.16±0.32	51.61±1.25	45.48±0.36
12	90.56±1.92	87.76±1.25	81.54±1.21	78.34±0.67	57.38±0.52	63.81±0.34	74.05±1.94	78.42±0.98	54.84±2.63

Table XVI: Kinetics study for the release profile of CPT

Formulation code	Zero order kinetics		First order kinetics		Hixson crowell		Higuchi model		Korsmeyer Peppas	
	R	K	R	K	R	K	R	K	R	n
F1	0.968	12.72	0.858	0.310	0.947	0.220	0.969	35.91	0.977	1.044
F2	0.993	7.613	0.887	0.165	0.930	0.260	0.959	34.10	0.979	1.404
F3	0.997	7.134	0.948	0.140	0.906	0.250	0.98	32.22	0.971	1.404
F4	0.973	6.827	0.880	0.122	0.968	0.267	0.908	30.06	0.985	1.560
F5	0.980	5.661	0.978	0.082	0.915	0.252	0.962	25.55	0.971	1.62
F6	0.977	5.596	0.921	0.082	0.964	0.258	0.917	24.71	0.984	1.617
F7	0.995	6.544	0.961	0.112	0.818	0.282	0.981	29.62	0.82	1.380
F8	0.968	6.326	0.855	0.115	0.921	0.242	0.926	28.20	0.971	1.422
F9	0.991	5.195	0.969	0.071	0.917	0.286	0.945	23.13	0.944	1.430

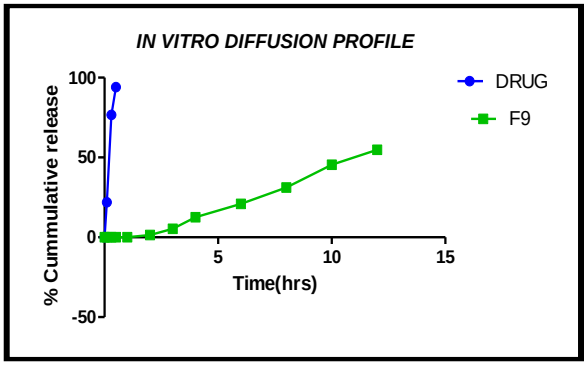

Figure XIII: In-vitro diffusion profile of drug and optimized batch F9

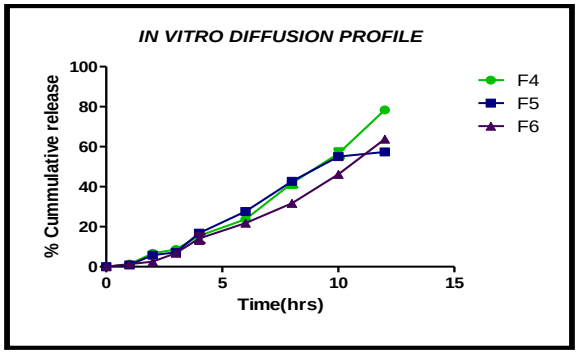

Figure XV: In-vitro diffusion profile of The CPT release from CPT LPs formulation (batch F4 to F6). Values are mean ± Std. dev. (n=3)

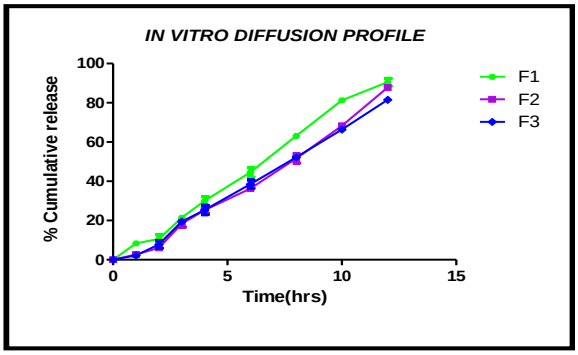

Figure XIV: In-vitro diffusion profile of The CPT release from CPT LPs formulation (batch F1 to F3). Values are mean ± Std. dev. (n=3)

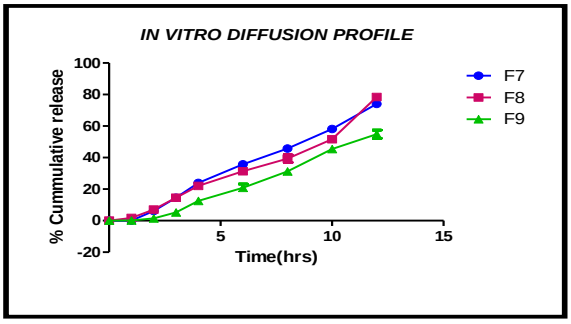

Figure XVI: In-vitro diffusion profile of The CPT release from CPT LPs formulation (batch F7 to F9). Values are mean ± Std. dev. (n=3)

XII. PHYSIOCHEMICAL CHARACTERIZATION of CAP LPS GEL

A. pH measurement

The gel obtained were examined and characterized for the parameters shown in table XVII. pH of all the formulation was found in the range of 6.5 to 7. The pH of both formulations was found near to the skin pH value.

Table XVII: pH measurement of CPT gel and CPT LP_s gel

Sr.No.	Formulation	Colour	PH
1	Captopril Gel	White	6.6
2	Captopril Liposomal Gel	Milkywhite	6.2

B. Viscosity Measurement

Viscosity of captopril gel and captopril liposomal gel was determined at different (RPM) speed. The results are given are as follows as. Viscosity of both the formulations was found to be using brookfield viscometer at spindle no. 64 in 10, 20, 30, 60,100rpm. The viscosity of the all gel formulations ranged from 3756-34389cps shown in table XVIII and XIX. The viscosity of the formulation decreased on increasing the shear rate. From the results obtained it was observed that in CPT gel and CPT LP_s gel viscosity decreased by increasing a shear rate. And at 100 rpm viscosity of CPT LP_s gel was found to be less as compared to CPT gel.

Table XVIII: Viscosity measurement of captopril gel

Sr.No.	Speed (RPM)	Spindle	CP	%
1	10	64	34389	57.3%
2	20	64	22000	73.6%
3	30	64	17200	86%
4	60	64	8740	87%
5	100	64	5586	93.1%

Table XIX: Viscosity measurement of captopril liposomal gel

Sr. No.	Speed (RPM)	Spindle	CP	%
1	10	64	29040	48.4
2	20	64	14970	49.9
3	30	64	10760	53.8
4	60	64	5490	54.9
5	100	64	3756	62.6

C. Spreadability Measurement

Spreadability of both the formulations was found in the range of 8.49 to 11.33(gm.cm/sec) respectively. The Spreadability of captopril liposomal gel was found to be good range as compared to captopril gel.

Table XX: Spreadability measurement of captopril gel and captopril liposomal gel

Sr. No.	Formulation	Spreadability (gm.cm/sec)
1	Captopril Gel	8.49
2	Captopril Liposomal Gel	11.33

D. Drug Content

The drug content value of captopril liposomal gel was found to be more as compared to captopril gel. In captopril liposomal gel due to stearylamine drug entrapped increases and hence drug content of liposomal gel was also found to be more.

Table XXI: Drug content of captopril gel and captopril liposomal gel

Sr.No	Formulation	Drug content %
1.	Captopril Gel	41.56±0.36
2.	Captopril Liposomal Gel	62.3±0.12

E. In vitro diffusion studies of CPT LP_S Gel

In-vitro drug release study was carried out the using the Franz diffusion cell in pH 7.4 phosphate buffer saline. In-vitro release profile of CPT gel and CPT LP_s gel was monitored for 12 hrs. The release profile of CPT from CPT gel was found to be 90.77±0.75 % at 4hrs. It was found to be fastest release from plain gel as compared to liposomal gel. The release profile of CPT from CPT liposomal gel was found to be 45.18±0.51% at 12hrs. As compared to CPT gel it was found to be sustained release of drug from captopril liposomal gel formulation because carbopol and cholesterol composition in formulation give a sustained of drug.

Table XXII: In-vitro diffusion profile of The CPT release from CPT gel and CPT liposomal gel.

Sr. No.	Time (hr.)	% Release for CPT GEL ± SD	% Release for CPT LP_s GEL ± SD
1	0	0.00±0.00	0.00±0.00
2	0.5	23.35±0.78	1.69±0.65
3	1.0	43.90±1.98	5.26±0.12
4	2.0	82.55±2.12	9.10±1.13
5	4.0	90.77±0.75	14.23±0.98
6	6.0		25.36±0.46
7	8.0		30.36±0.25
8	10.0		37.23±0.69
9	12.0		45.18±0.51

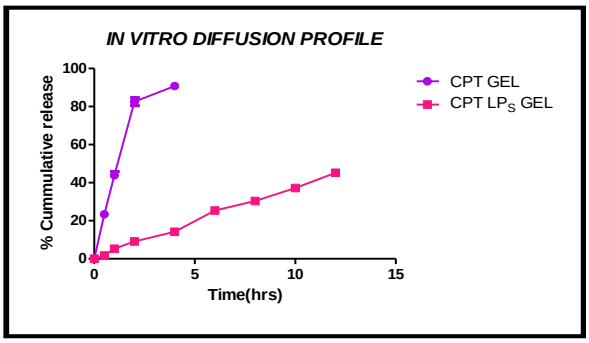

Figure XVII: In-vitro diffusion profile of The CPT release from CPT gel and CPT liposomal formulation. Values are mean ± Std. dev. (n=3)

CONCLUSION

Hence, from the present experimental data, it can be concluded that the proposed objectives of the project are achieved. The particle size, FESEM and zeta potential showed the captopril liposomes are physical stable and value of zeta potential revealed that more cationic charged present on the surface of liposomes due to the incorporation of stearylamine. It is help the liposomes to diffuse more readily into the skin because the presences of negative charged on the skin layer. Particle size value of captopril liposome was almost closed to the transdermal size ranged of liposomes. The FESEM showed that vesicles are almost in spherical and uniform size. The FTIR studied showed that there was no possible interaction found to be in drug and optimized formulation. The in vitro diffusion study of captopril liposomes formulation showed sustained release of captopril than captopril solution until 12 hrs. The captopril liposomes gel was showed better spreadability, viscosity, and ph and drug content values as compared to the captopril gel. The in vitro diffusion study of captopril liposomal gel formulation gives a prolonged or sustained release of drug as compared to captopril gel until 12 hrs. Therefore, it can be concluded that the captopril liposomal gel gives a sustained release of captopril. And transdermal application of gel it is good for patient compliances and also eliminated the problem of GIT degradation of drug.

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