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Original Article

DESIGN AND DEVELOPMENT OF NANOSTRUCTURED LIPID CARRIER CONTAINING TRIAMCINOLONE ACETONIDE

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ABSTRACT

Objective: The aim of present study was to prepare nanostructured lipid carriers (NLCs) based Triamcinolone acetonide (TA).

Methods: Nanostructured lipid carriers (NLCs) consisted of solid lipid and liquid lipid are a new type of lipid nanoparticles, prepared by using solvent diffusion and high pressure homogenization methods, which offer the advantage of improved drug loading capacity and release properties. Glyceryl monostearate selected as the solid lipid, capmul MCM C8 as the liquid lipid, polyvinyl Alcohol (PVA) as the surfactant. NLCs dispersion was characterized by particle size analysis, zeta potential, scanning electron microscopy (SEM), differential scanning calorimetry, and an *in vitro* release study.

Results: Optimized NLCs loaded with TA were exhibited spherical shape with particle size 286.1 nm, polydispersity index 0.317, zeta potential-21.9 mV and entrapment efficiency 86.19% respectively. The result of differential scanning calorimetry (DSC) showed that drug was dispersed in NLCs in a crystalline state. *In vitro* release studies revealed that drug release of optimized batch was 8.34 % and 88.84% at 1h and 8h respectively. The release kinetics of the optimized NLCs best fitted the peppas-korsmeyer model. Furthermore, morphological investigations by SEM showed that optimized batch exhibit a spherical shape and a smooth surface.

Conclusion: Thus, the results indicated that successfully prepared TA-loaded NLCs and could potentially be exploited as a carrier with improved drug loading capacity and sustained drug release. The present results demonstrated that these systems could be a promising platform for inflammatory diseases, in particular for psoriasis topical therapy.

Keywords: Triamcinolone acetonide, Solvent diffusion method, Nanostructured lipid Carrier, Corticosteroid, Factorial design

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INTRODUCTION

Now days, nanotechnology has proved the growth of research and its applications in the area of medicine [1]. Since last decade, various techniques have been studied to formulate nanoparticulate carrier systems. Polymeric and solid lipid nanoparticles (SLNs) are two varieties of nanocarrier systems. Polymeric nanoparticles suffered with some drawbacks such as toxicity and unavailability of good techniques for production of nanoparticles at large scale. Compared to polymeric nanoparticles, SLNs gain advantages in terms of less toxicological risk because of natural origin lipids. Despite SLNs being good carriers, less capacity of drug loading and expulsion of the drug during storage, may require some good technique to overcome such problems. As an effect, nanostructured lipid carriers (NLCs) have been developed, which in some extent can avoid the afore mentioned limitations.

NLCs was second generation of SLNs having solid lipid and liquid lipid matrix that create a less ordered or imperfect structure, which helps in improving drug loading and decreasing the drug expulsion from the matrix during storage period. The development of nanocarrier systems is an approach to overcoming such problems [2, 3]. NLCs composed of a solid lipid matrix with a certain content of liquid lipid are a new generation of SLNs [4]. The advantages of these systems are, stabilize oxidation/photo-sensitive materials by incorporating poorly water soluble drugs. As dermally applied, ensure close contact with the lipid bilayer of the stratum corneum, resulting in a more efficient and deeper drug penetration into the skin layers [5, 6].

Corticosteroids are the most widely used in the treatment of dermatological disorders is related to their vasoconstrictive, anti-inflammatory, immunosuppressive and anti-proliferative effects [7, 8]. Triamcinolone acetonide (TA) is a synthetic glucocorticosteroid binds in the target cell to specific cytosolic glucocorticoid receptors and subsequently interacts with glucocorticoid receptor response elements on DNA, thereby altering gene expression [9]. NLCs are composed of biodegradable and physiological lipids as a carrier, exhibiting low systemic cytotoxicity [10-12].

In case of dermatological diseases such as psoriasis, whose triggers are situated beneath the skin, it is preferable to manage drugs topically rather than systemically due to more efficient direct action with improvement of the local access for optimum amount of drug. Topical administration also reduces the systemic burden and toxic effects of the drugs and it is considered the first line of treatment used in moderate psoriasis as it is considered safe and well accepted by the patients [13, 14]. The successful implementation of these systems for drug delivery entirely depends on their ability to go through numerous anatomical barriers, sustained release of their content and stability in the nanometer size [15, 16]. Various particulate lipid based colloidal carriers have also found application in anti-psoriatic drug delivery, in particular SLNs and NLCs [17].

Lipid based drug delivery systems are now days popular because of their potential to increase solubility and bioavailability of poorly water soluble drugs. This becomes an important tool, when it is necessary to supply the drug over a prolonged period of time, and to reduce systemic absorption, when the drug is irritating in high concentrations. NLCs have the potential to adjust the drug release over an extended period with a reduced rate of systemic absorption. The lipid film formation above the skin and the succeeding occlusion effect was described for lipid nanoparticles with reduction of transepidermal water loss caused by this effect, leads to an augment in skin hydration after dermal application of SLNs or NLCs [18-21]. NLCs systems are a promising carrier for the topical delivery of antipsoriatic drugs as revealed by improved skin permeation and reduce irritation, narrow size distribution, better bioavailability and the compatibility of the drugs.

The present investigation explored the possibility of NLCs as a unique carrier system for the topical application with regard to the modulation of release of TA. Solvent diffusion method has remarkable advantages such as use of simple equipment accessories, easiness in handling and quick manufacturing [22]. The aim of present research work was to develop stable TA loaded NLCs formulation using solvent diffusion method and to evaluate *in vitro* characteristics of prepared formulation.

MATERIALS AND METHODS

Materials

Triamcinolone acetonide and Capmul MCM C8 were obtained as a gift samples from Glenmark Pharmaceuticals, Goa and Abitec Corporation, Janesville, USA respectively. Glyceryl monostearate, polyvinyl alcohol and carbopol were purchased from Loba Chemicals, Mumbai, India. All other solvents and reagents used in this work were of analytical/HPLC grade.

Methods

The sample of TA was analyzed for its nature, color and taste. The melting point was performed by using open capillary method. TA was estimated by UV spectrophotometry method. Calibration of TA was carried out by preparing different stock solution in methanol (10µg/ml to 50µg/ml) to determine concentration from absorbance.

Optimization of formulation using factorial design

A statistical model incorporating interactive and polynomial term was used to evaluate the responses.

$$Y = b_0 + b_1X_1 + b_2X_2 + b_{11}X_1^2 + b_{22}X_2^2 + b_{12}X_1X_2 \quad \text{(Eq.1)}$$

Where, Y is dependent variable, b0 is the arithmetic mean response of the 9 runs, and bi (b_1, b_2, b_{12}, b_{11} and b_{22} is the estimated coefficient for the factor X_1). The main effect (X_1 and X_2) represents the average results of changing one factor at a time from its low to high values. The interaction term (X_1X_2) show how the response changes, when 2 factors are changed simultaneously. The polynomial term (X_{12} and X_{22}) are included to investigate nonlinearity. The formulations were fabricated according to a 3^2 full factorial design, allowing the simultaneous evaluation of two formulation independent variables and their interaction.

The effect of the variables were investigated on the responses of the particle size and entrapment efficiency, were selected as dependent variables and drug and lipid concentration (X_1) and stabilizer concentration (X_2) as independent variables. The replicate experimental runs were carried out in complete randomized manner. A multilinear stepwise regression analysis was performed using microsoft excel software. The full models were used to plot two-dimension contour plots for both particle size and entrapment efficiency. All the statistical operations were carried out by PCP Disso 2000 V3 software.

Factorial design parameters and experimental conditions are the factors used drug and lipid concentration levels 35, 40 and 45 in mg and stabilizer concentration levels 240, 260 and 280 in mg. Factorial designs formulation and experimental conditions factors levels used shown in table 1 and 2.

Table 1: 3^2 Factorial designs formulation

Batch code	Coded values	
	*X_1	*X_2
B1	-1	-1
B2	-1	00
B3	-1	+1
B4	00	-1
B5	00	00
B6	00	+1
B7	+1	-1
B8	+1	00
B9	+1	+1

*X_1-Amount of drug and Capmul MCM C8, *X_2-Amount of GMS and PVA, *-1, 0,+1–Low, Medium and High amount of drug and Capmul MCM C8 and GMS and PVA

Table 2: Amount of variables in a 3^2 factorial design formulation

Coded values	Actual values	
	Amount of drug and capmul MCM C8 X_1 (mg)	Amount of GMS and PVA X_2 (mg)
-1	35	240
0	40	260
+1	45	280

Table 3: Composition of NLC batches

Ingredient	Batches code								
	B1	B2	B3	B4	B5	B6	B7	B8	B9
Drug (mg)	12	12	12	12	12	12	12	12	12
Capmul MCM C8(µl)	23	23	23	28	28	28	33	33	33
Glyceryl mono stearate (mg)	57	57	57	57	57	57	57	57	57
Polyvinyl alcohol (gm)	0.183	0.203	0.223	0.183	0.203	0.223	0.183	0.203	0.223

Preparation of NLCs

NLCs loaded with TA were developed using solvent diffusion method in aqueous system. Briefly, 228 mg mixture of capmul MCM C8 and GMS with 12.5,25 wt% lipid content, prepared by adding capmul to GMS and 12 mg of TA, total weight of 5% w/w to the drug and lipids were mixed in a solvent mixture of ethanol (6 ml) and acetone (6 ml) that is (1:1 v/v) kept in sonication bath (Spectra lab model UCB 70) for 10 min. and on water bath temperature was maintained at 60 °C. GMS was added to resultant mixture to make it clear solution of lipid and drug in organic solvent system. The aqueous phase of PVA (0.2%) used as stabilizer made up to 100 ml into which the organic mixture was added and kept on water bath, maintained temperature at 70 °C and processed under mechanical agitation of 500 rpm for 10 min. using mechanical stirrer (IKA RW 20 Digital). At room temperature TA loaded NLCs dispersion was

cooled for 20 min. The dispersion was kept on magnetic stirrer for liberation of organic solvent. Obtained dispersion was mixed by using Ultra turrex homogenizer for 10,000 rpm for 5 min. Further samples were passed through High pressure homogenizer under pressure of 1000 bar and 30 cycles. NLCs dispersion was dried in freeze drier by adding cryoprotectant (mannitol) and lyophilized, obtained a dried and free flowing powder [23, 24]. Composition of NLCs batches were shown in table 3.

Characterization of NLCs formulations

Particle size and zeta potential

The particle size distribution and zeta potential of the prepared NLCs were measured by using Horiba SZ-100 using dynamic light scattering (DLS). Particle size and particle size distribution of the NLCs determined by using particle size analyser, after dilution with water determine the diameter and mean particle size distribution. A light is passed into the cell, at 90° or 173°. Depending on both the concentration of sample and strength of the system, selects the optimum scattering angle and cell position. 1 ml of sample was injected into the zeta cell and measurements for the zeta potential analysis. Zeta potential is a measure surface charge of particles and thus it imparts the colloidal stability due to particle-particle repulsion, as particle aggregation is less to occur for charged particles having high zeta potential.

Percentage entrapment efficiency

A 5 ml sample of NLCs dispersion was centrifuged at 8,000 rpm at 10 °C for 20 min. by using cooling centrifuge. The supernatant was separated, and then resuspended the NLCs lipid layer in methanol and centrifuged. This washing process was repeated two times to guarantee that the free drug was no longer present in the voids between the NLCs. The collected NLCs residue was lysed with absolute methanol. TA in the supernatant was determined by UV spectrophotometric method at specified wavelength. The % entrapment efficiency was calculated by using formula 1.

$$\% \text{ Entrapment efficiency} = \frac{Tp - Tf}{Tp} \text{ X } 100 [1]$$

Where, Tp is the total amount of drug, Tf amount of drug in supernatant

***In vitro* release study**

In vitro release of TA from NLCs was evaluated by the dialysis diffusion technique. The diffusion medium was used phosphate buffer pH 7.4. NLCs equivalent to 1 mg of TA was dissolved in buffer, placed in the dialysis bag and sealed at both the ends. The dialysis bag was immersed in 70 ml of the receptor compartment (beaker), which was stirred at 50 rpm and maintained temperature at 37±2 °C. The receptor compartment was covered to prevent the evaporation of medium. A 5 ml sample was withdrawn at every 15 min. interval for first 1h and 1h interval for next 8h. The same volume was replaced by dissolution medium in the flask to maintain a constant volume. The samples were filtered, suitably diluted and % cumulative drug release determined by UV spectroscopy at λ max of 238 nm.

Fourier transform infrared spectroscopy (FTIR)

FTIR spectrum shows the fundamental peaks corresponding to the chemical nature of the drug and excipients. FTIR studies were carried out in order to determine the possible interaction among drug and excipients used. IR absorption spectrum of TA was determined by fourier transform infrared spectrophotometer Perkin Elmer (Brooker) at resolution of 2-1 cm. Spectra were recorded over the wave number 400-4000 cm-1. Infrared spectrums of pure drug and optimized batches were recorded. From the spectrum analysis the compatibility of ingredients in the formulations were determined. The infrared spectrum of TA was recorded and determine the chemical component, which present at specified wavelength.

Differential scanning calorimetry (DSC)

DSC studies were carried out using (Mettle-Toledo DSC821 instrument). Indium and zinc standards were used to calibrate the DSC temperature and enthalpy scale. Freeze dried of NLCs optimized batch and pure drug were hermetically sealed in aluminium crucible and heated at 10 °C/min over a temperature range of 0-450 °C. An inert atmosphere was maintained by purging with nitrogen at a flow rate of 50 ml/min. An empty aluminium pan was used as standard reference. Thermograms of pure TA, physical mixture and NLCs formulation were obtained using DSC.

Powder X-ray diffraction (P-XRD)

The XRD patterns were recorded on X-ray diffract meter (PW 1729, Philips, Netherlands). Samples were irradiated with monochromatized Cu-Ka radiation (1.542A˚) and analyzed from 2 °-100° 2θ at a scan speed of 0.1° 2θ/sec using 1.524 A˚ radiations. The spectra of XRD are recorded and analyzed.

Scanning electron microscopy (SEM)

SEM analysis was carried out by using ESEM (QUANTA-200-3D, FEI, USA) at 20.0KV in environmental mode for identification and morphology of NLCs. Thin film of test was set up on carbon covered copper network by simply dropping a one drop of test on framework, test was expelled by channel paper/tissue paper and allow drying a sample for overnight. Before taking an image, coat the grid by gold coating, and observe the size and morphology of particle.

Stability study

Stability studies were carried out according to ICH guidelines Q1A (R^2). The stability of NLCs was calculated by storing the sample for a period of 30 d at 40±2 °C/75±5% temperature and relative humidity respectively. Samples were removed at interval of 1 mo and examined for particle size, zeta potential, and % entrapment efficiency.

RESULTS AND DISCUSSION

The sample of TA was amorphous powder having white color, odorless and bitter in taste. The melting point was observed in the range of 292 °C-294 °C. The standard solution of TA was scanned through 200-400 nm wavelength on Shimadzu UV-1800 spectrophotometer. The TA absorption maximum was found to be 238 nm. The obtained NLCs have been assessed for particle size analysis, zeta potential, entrapment efficiency and solid state characteristic by XPRD, DSC, FTIR and SEM analysis.

Selection of surfactant and its concentration

Liquid lipid was selected based on the maximum solubility of the drug in different solvent. Lipids were used for the study, capmul MCM C8, isopropyl myristate, oleic acid, labrafil ILM 1944 CS and labrafec CC. Selection of best surfactant from tween 20, tween 80, span 20, span 80 and PVA on the basis of its particle size and polydispersity index. For each surfactant of 1.0% concentrations was prepared and fixed stirring speed at 1000 rpm. The concentrations of surfactants were optimized according to the resultant particle size and polydispersity index of each batch. When surfactant tween 20 was used, particle size and PDI observed 396.4±0.95 nm and 0.325±0.002 as compared to tween 80, 425.95±5.05 nm and 0.355±0.04 and PVA 286±6.35 nm and 0.317±0.03 respectively. PVA was used as surfactants for stabilization of nanolipids more effectively with smallest particle size and narrowest particle size distribution, observed as compared to tween 80 and tween 20. Hence PVA was selected as a surfactant for further development of nanolipid carrier formulation with smallest particle size.

Preparation and characterization of NLC

NLCs loaded with TA were developed using solvent diffusion method in aqueous system.

Particle size and polydispersity index

The formulated NLCs sizes were analyzed using DLS, results revealed that presence of NLCs mean diameter of optimized batch was about 286.1 nm, maximum NLCs lie between size ranges of 200-500 nm with an average particle size was 286.1 nm with polydispersity index 0.317. Particle size of optimized batch shown in fig. 1.

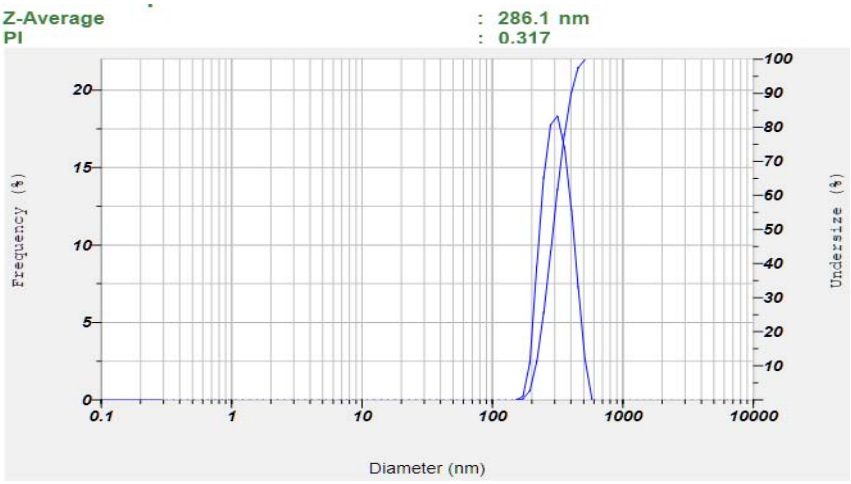

Fig. 1: Particle size of optimized batch

Zeta potential

Zeta potential of the NLCs was determined by using zeta sizer. NLCs with zeta potential±30mV were considered as highly stable. The zeta potential of optimized batch B6 was obtained-21.9 mV which correspond to stability of formulation. The zeta potential in the range of-14.30 to-21.9 mV was obtained of all prepared batches. The results of zeta potential of all batches and optimized batch were shown in table 4 and fig. 2. This finding is in agreement with that published by Salunkhe SS *et al.* [1].

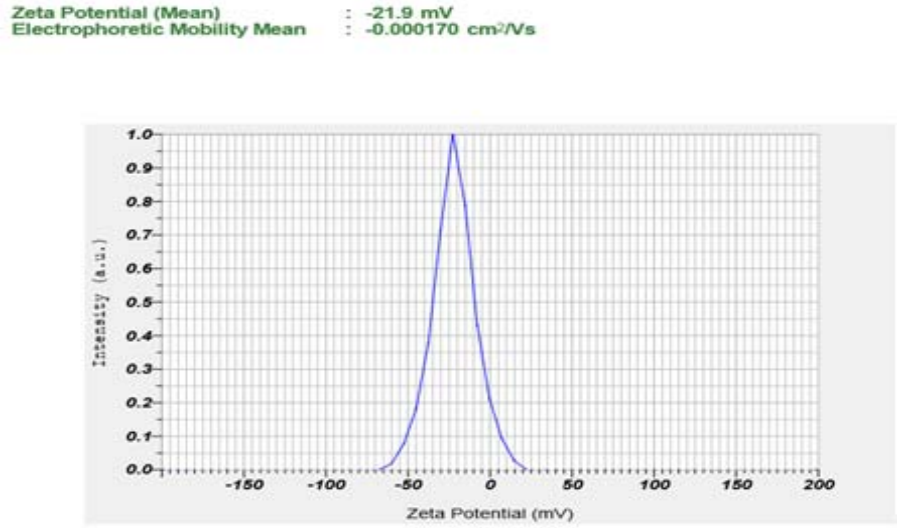

Fig. 2: Zeta potential of optimized batch

Entrapment efficiency

The % entrapment efficiency of NLCs was found to be in the range of 75.2% to 86.19 %. The entrapment efficiency of the optimized batch B6 was 86.19% because of the perfect ratio of lipid volume in the vesicles as compared to other batches. Thus, it shows the effect of capmul, PVA and edge activators ratios on entrapment efficiency. The % entrapment efficiency of all batches were shown in table 4. This finding is in agreement with that published by Salunkhe SS *et al.* [1].

Table 4: Optimized batches of NLC formulation

Batches code	X1	X2	Particle size	Entrapment efficiency	Zeta Potential
B1	-1	-1	389	83.06	-14.54
B2	-1	0	370	82.07	-15.01
B3	-1	+1	360	81.03	-17.45
B4	0	-1	386	82.16	-14.30
B5	0	0	350	84.45	-18.26
B6	0	+1	286	86.19	-21.91
B7	+1	-1	410	78.03	-15.20
B8	+1	0	394	76.01	-16.89
B9	+1	+1	362	75.02	-18.25

Development of polynomial equations

The experimental design and parameters shown in table 1 and 2 for factorial formulations B1 to B9, polynomial equations for two dependent variable particle size and entrapment efficiency derived by using PCP Disso 2000 V.3 software. The response surface plots of effects of TA and capmul, PVA on particle size and entrapment efficiency are shown in fig. 3 and 4 respectively. The multiple regression equation relating the response particle size and entrapment efficiency to transformed factors were given in equations 2 and 3 respectively.

PS $=347.233+6.8417X_1-31.5000X_2+39.1750X_1^2-9.8500X_2^2-6.0500X_1X_2$ (Eq.2)

(R^2= 0.88161)

%EE $= 84.2827-2.8022X_1-0.1683X_2-5.0155X_1^2-0.0240X_2^2-0.2820X_1X_2$ (Eq.3)

(R^2= 0.87479)

The polynomial equation can be used to draw conclusion after considering the magnitude of coefficient and the mathematical sign it carries, (i.e., positive or negative).

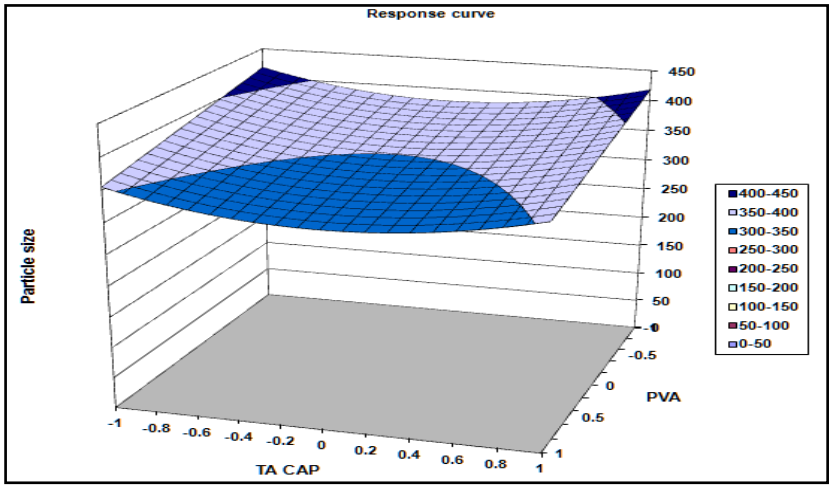

Fig. 3: Response surface plot for particle size

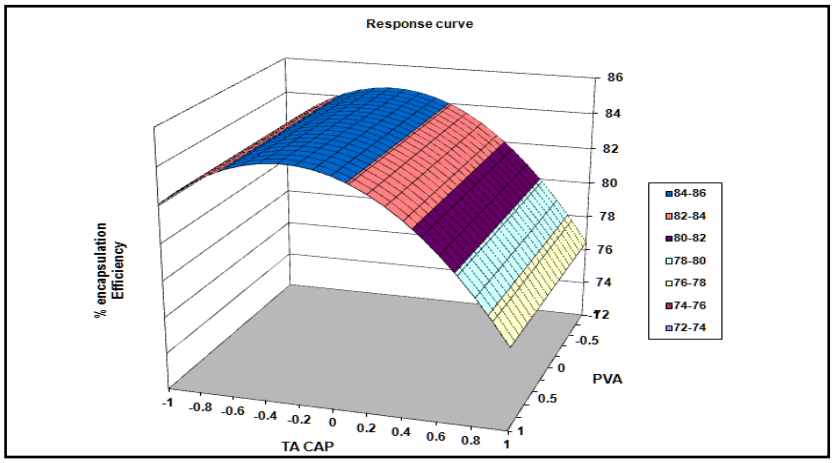

Fig. 4: Response surface plot for entrapment efficiency

In equation 2, negative sign for coefficient of X_2 indicate that the particle size of NLCs increases, when concentration of PVA was decreased and positive sign for coefficient of X_1 indicate the positive effects of drug and capmul concentration on particle size. In equation 3, negative sign for coefficient of X_1 indicate that % entrapment decreases, when concentration of drug and capmul increases and negative sign for coefficient of X_2 indicate that % entrapment increases, when concentration of PVA decreases.

The closeness of predicted and observed values of particle size and % entrapment indicates the validity of derived equations for dependent variable. The data clearly indicate that the particle size and entrapment efficiency were strongly dependent on the selected independent variables. The values of the correlation coefficient indicate good fit.

The response surface plots illustrate that as concentration of drug and capmul increases, the value of dependent variable, particle size decreases and concentration of PVA increases the values of dependent variable, particle size decreases. Similarly, the response surface plots for % drug entrapment shows negative effects of independent variable, drug and capmul concentration and PVA concentration. The data demonstrate that both drug, capmul (X_1) and PVA (X_2) affect the drug release, particle size and entrapment efficiency. It concluded that the high levels of X_1 and X_2 favor the drug release and hence particle size was less as compared to the low levels of X_1 and X_2. Findings are in accord with those concluded in many relevant works [10, 12].

***In vitro* drug release study**

The cumulative percentage drug release of batches B1 to B9 were in the range of 72.59 to 88.84% for 8 h. The percentage release of optimized batch was found to be 8.34 % and 88.84 % at 1 h and 8 h respectively. *In vitro* drug release study of TA loaded NLC was indicated sustained release for 8 h. The % cumulative drug release of drug of all batches were shown in fig. 5. This finding is in agreement with that published by Dixit CM *et al.* [20].

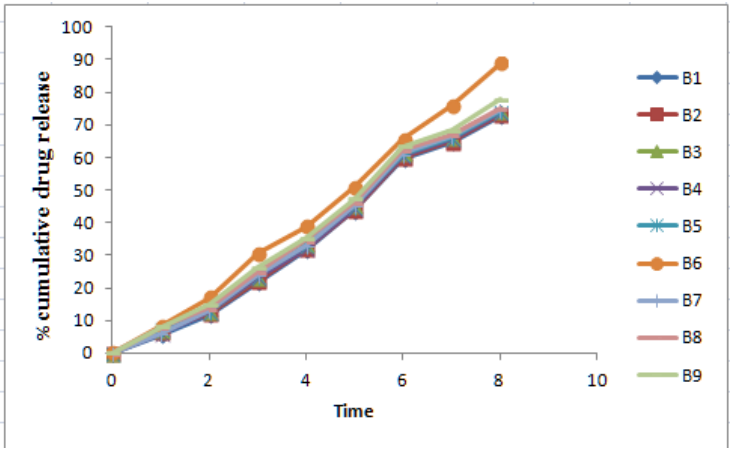

Fig. 5: % Cumulative drug release of drug

FTIR spectroscopy study

The FTIR study was carried out to confirm the compatibility between selected drug and excipients. Spectrums of TA were showed characteristics peaks belonging to measure functional groups such as principle peaks at wave numbers 1710 cm^{-1}, 3343 $cm^{-1}$and 1660 cm^{-1}. The observed peaks and possible functional groups for TA, physical mixture and NLCs optimized batches were shown in fig. 6, fig. 7 and fig. 8 respectively. The major IR peaks observed in TA were 1710 (1710-1720) (C=O), 3343 (3300-3400) (O-H), 1660 (1660-1820) (C=O).

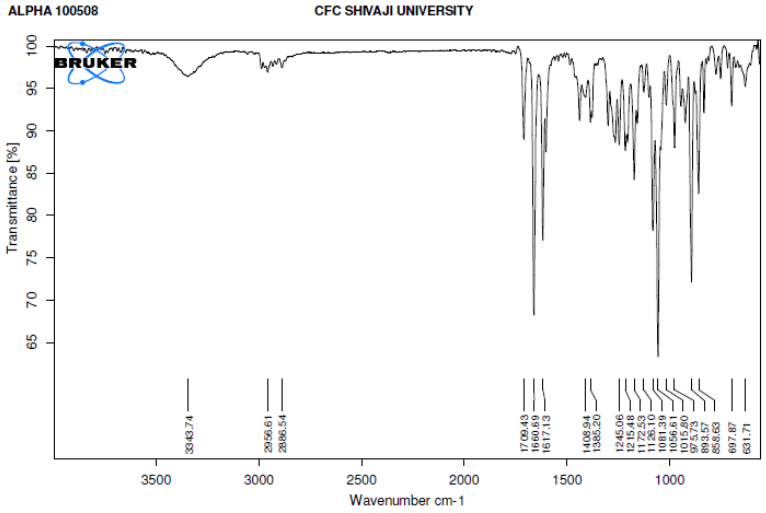

Fig. 6: FTIR spectral analysis of TA

The major IR peaks of physical mixture (PVA+drug) were observed, 3396 (3550-3200) (O-H),1708(1750-1680)(C=O),1662(1690-1630)(C=O),1611(1700-1500)(C=C)1278,1215(1320-1210)(C=O), 1079(1200-1025)(C-N), 1079,1056(1200-1025)(C-N)$^{cm-1}$.

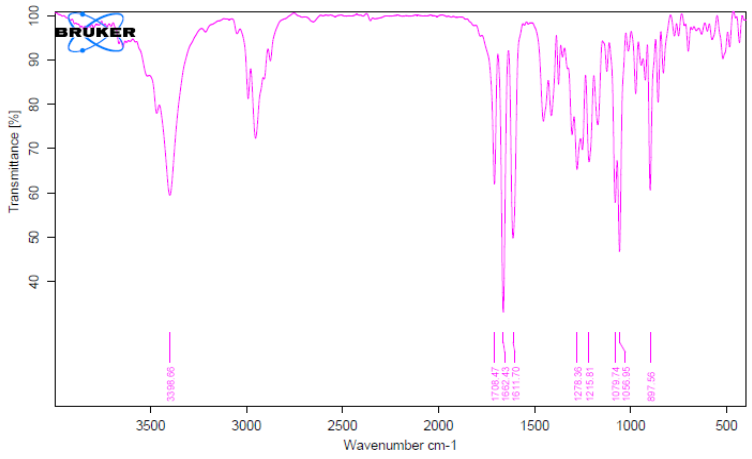

Fig. 7: FTIR spectral analysis of physical mixture

The major IR peaks of NLCs were observed, 3265 (3550-3200) (O-H), 2936 (2950-2800) (C-H)1460(1600-1450) (C=C),1088, 1022(1260-1000)(C-O),831,890(890)(C-H),and 718(780-690)(C-H) cm^{-1}.

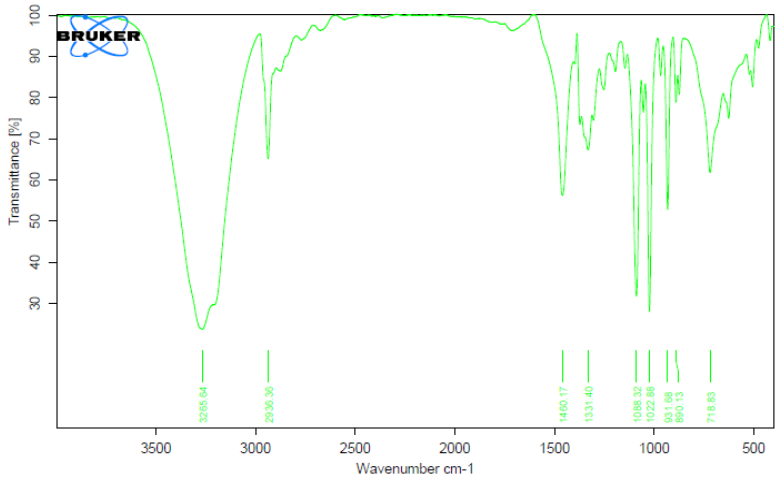

Fig. 8: FTIR spectral analysis of NLCs

FTIR results reveals that the fundamental peaks of the TA were retained in the optimized batch B6 formulation. Results showed that no major changes as well as no loss of functional peaks. This indicates that absence of chemical interaction or any changes between TA and excipients used in the formulation.

X-ray diffraction study

The diffraction spectrum of TA showed that the drug was crystalline nature indicated by numerous, relative sharp and distinct peak at diffraction angle 2θ shown in fig. 9. The results indicated that XRD spectrum of NLCs batch shown reduced peak intensity it leads to reduced crystallinity. It revealed that the there was no such interaction observed in excipients and polymer used. In XRD analysis all peak intensity was observed in decreasing order, therefore the NLCs formulation shows decrease in crystallinity. The overlain of diffraction spectrum of XRD of A) drug B) physical mixture C) NLCs shown in fig. 9

Differential scanning calorimetry (DSC)

The DSC curve of TA shows a broad endotherm indicating single, sharp melting endotherm peak at 191.6 °C, which corresponded to its intrinsic melting point indicating it's crystalline nature. Thermogram of physical mixture showed almost the same peaks of drug at 191.6 °C. However, no sharp endotherm was seen at 161.6 °C and 170 °C for NLCs optimized batch B6, it suggesting that TA in NLCs was molecularly dispersed as a less crystalline form, it's melting point was decreased indicating reduced crystallinity due to dilution effect of the polymers. DSC results were support of the XRD analysis, which also shows decrease in drug crystallinity in NLCs formulation. The overlain of drug, physical mixture, NLCs optimized batch B6 were shown in fig. 10. Findings are in accord with those concluded in many relevant works [1, 12].

Scanning electron microscope (SEM)

The results of SEM can be revealed that TA showed needle shaped large crystals, indicating crystalline nature. However, the prepared NLCs optimized batch B6 had a drastic change in morphology, nearly spherical shape with nonporous and smooth surface observed under 2500X and 18kv, so no drug crystals were present in NLCs formulation. The SEM of pure TA and optimized batch B6 shown in fig. 11 and 12 respectively. Thus, prepared NLCs confirm stability in their structure and shape. Findings are in accord with those concluded in many relevant works [4, 10, 24].

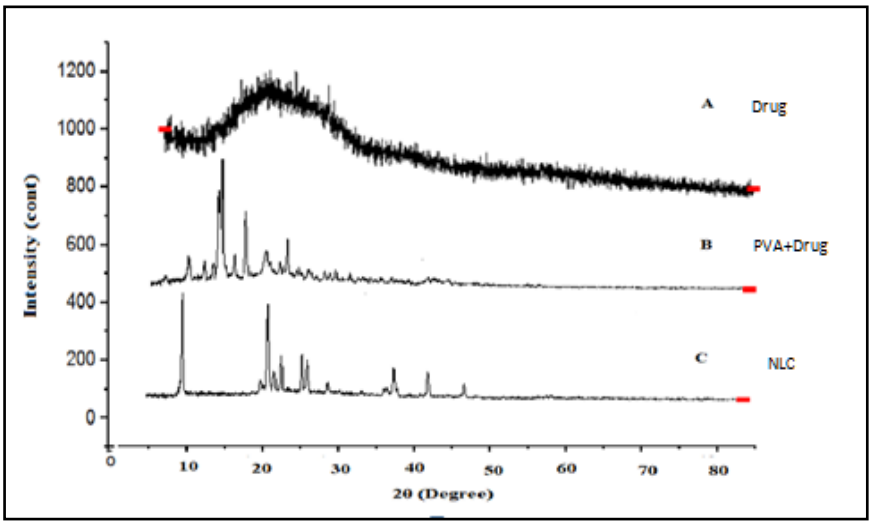

Fig. 9: Overlain XRD of A) Drug B) Physical mixture C) NLC

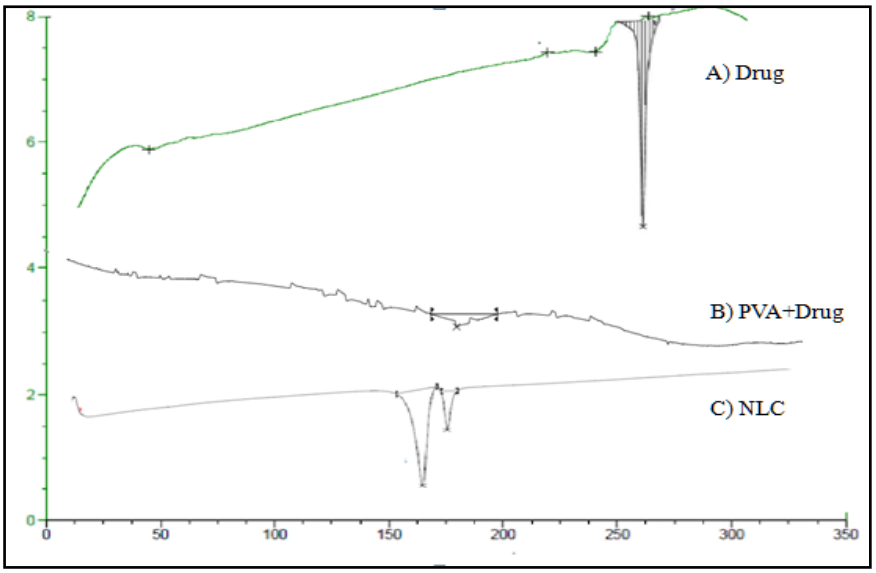

Fig. 10: Overlain of drug, physical mixture, NLC

Fig. 11: Scanning electron microscopy of TA

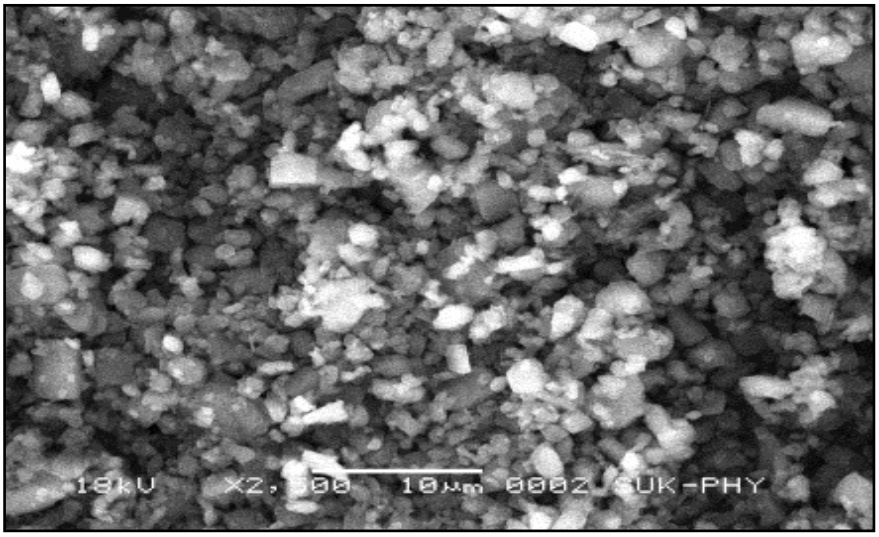

Fig. 12: Scanning electron microscopy of NLC optimized batch

Stability study

Stability studies of TA-NLCs was evaluated based upon of physical properties obtained. Over a period of 30 d, under specified conditions of temperature and relative humidity, the results reveals that no morphological changes observed but particle size, PDI was found to be increased. However, zeta potential remains almost unchanged for TA-NLC indicating stability of the structure in formulation. Thus, it concludes that the drug does not undergo degradation on storage. Stability study of optimized batch B6 for particle size shown in fig. 13. Findings are in accord with those concluded in many relevant works [4, 10].

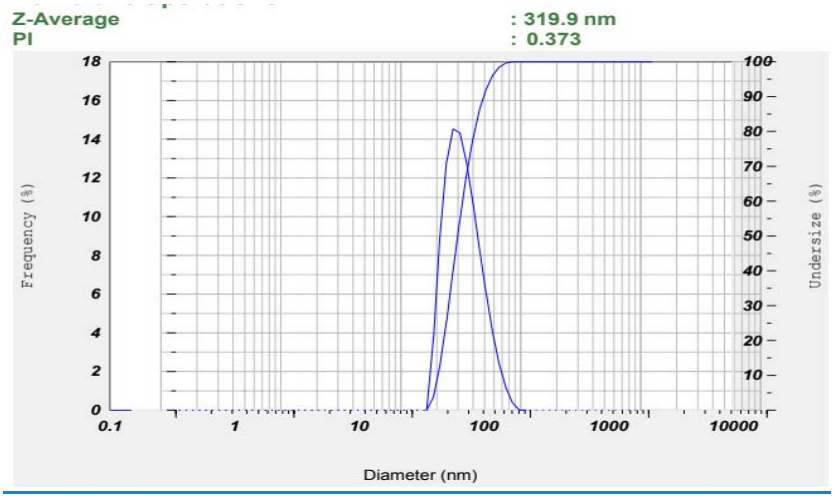

Fig. 13: Stability study of optimized batch for particle size

CONCLUSION

The present study was to prepared NLCs formulation loaded with TA by solvent diffusion method using a 3^2 full factorial design. Compared to SLNs, the NLCs exhibit high entrapment efficiency with sustained release of drug up to the period of 8h. It showed improved drug loading capacity and a good ability to reduce the drug expulsion on storage. For optimized batch B6, particle size, % entrapment efficiency and zeta potential were 286.1 nm, 86.19 and-21.9 mV respectively. The % cumulative release of optimized batch B6 was found to be 8.34 % and 88.84 % at 1 h and 8h respectively.

DSC and XRD studies confirm the transformation of crystaline nature of drug into amorphous that plays an important role in enhancement of absorption rate followed by bioavailability. SEM study confirms nanosized discrete spherical shape with smooth surface area. Optimized formulation at room condition shows extremely stable formulation for the period of one month that support the fact that dried lyophilized nanocarriers may remain stable for longer period of time. It was concluded that the development of NLCs formulation could potentially be exploited as a carrier with improved drug loading capacity and controlled drug release properties. Thus, TA loaded NLCs formulation can be beneficial in treatment of psoriasis.

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AUTHORS CONTRIBUTIONS

The author designed and performed the experiment, analyzed data and prepared the manuscript. All authors played an equal role in completing this research work.

CONFLICT OF INTERESTS

The authors declared no conflict of interest.

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