DESIGN, PREPARE AND IN VITRO EVALUATION OF SOLID LIPID NANOPARTICLES LOADED DEOXYCORTISONE BY USING HOMOGENISATION TECHNIQUE

BANDARU BHAVANI, G.S VALLURI*, I.NAGARAJU, G.VIJAYA KUMAR

DEPARTMENT OF PHARMACEUTICS, KGR INSTITUTE OF TECHNOLOGY & MANAGEMENT, RAMPALLY (V), KESSARA (M), RANGA REDDY DISTRICT

Corresponding Author:

G.S Valluri,
Department of Pharmaceutics,
KGR Institute of Technology and Management,
Rampally, Secunderabad,
Telangana 501301

ABSTRACT

The present study focuses on the design, preparation, and in-vitro evaluation of Deoxycortisone-loaded solid lipid nanoparticles (SLNs) formulated using the homogenization technique. Solid lipid nanoparticles offer a promising delivery system due to their biocompatibility, controlled drug release, and ability to enhance the bioavailability of lipophilic drugs. Deoxycortisone, a potent glucocorticoid with anti-inflammatory activity, was chosen as the model drug to develop a sustained and targeted delivery system. In this study, SLNs were prepared using glyceryl monostearate as the lipid matrix and a combination of surfactants Tween 80, Span 20, and Poloxamer 407 for stabilization. The formulations (F1–F8) were optimized by varying the surfactant ratios to achieve desirable particle size, zeta potential, and entrapment efficiency. The prepared SLNs were evaluated for particle size, zeta potential, drug entrapment efficiency, surface morphology, in-vitro drug release, release kinetics, and stability studies as per ICH guidelines. The optimized formulation (F7) exhibited a particle size of 132 nm, zeta potential of –23 mV, and maximum drug entrapment efficiency of 91.16%. SEM analysis confirmed spherical particles with smooth surfaces and no aggregation. In-vitro drug release studies showed an initial burst followed by sustained release over 12 hours, with formulation F7 showing the

most desirable release profile (98.55% cumulative release). Kinetic modeling indicated that the release followed zero-order kinetics and was diffusion-controlled as per the Higuchi model. Stability studies at different temperature and humidity conditions confirmed the formulation's physical and chemical stability for three months. Overall, the results demonstrated that Deoxycortisone-loaded SLNs prepared by homogenization offer a stable and efficient drug delivery system with controlled release characteristics, making it a potential carrier for topical or systemic corticosteroid therapy.

Keywords: Deoxycortisone, Solid Lipid Nanoparticles (SLNs), Homogenization, Controlled Drug Release, Entrapment Efficiency, Stability Studies.

INTRODUCTION

Solid lipid nanoparticles (SLNs) are sub-micron colloidal carriers composed of physiological lipids that remain solid at room and body temperature. SLNs combine advantages of several carrier systems the biocompatibility of liposomes and the physical stability of polymeric nanoparticles and offer controlled drug release, protection of labile drugs from degradation, and potential for improved dermal or systemic delivery. Because SLNs are manufactured from lipids generally regarded as safe (GRAS), they are particularly suitable for delivery of lipophilic drugs such as many corticosteroids.[1] In hot homogenisation, the lipid phase (containing dissolved drug) and aqueous surfactant phase are heated above the lipid melting point and emulsified; subsequent high-pressure or high-shear homogenisation produces nanosized lipid particles that crystallize on cooling.[2] Deoxycortisone, a steroidal glucocorticoid/precursor (or its pharmaceutically used derivative), possesses potent anti-inflammatory activity; therefore, developing an optimized delivery system for deoxycortisone is clinically attractive to improve local bioavailability while minimizing systemic adverse effects.[3] This study aims to design, prepare and evaluate deoxycortisone-loaded solid lipid nanoparticles using homogenisation technique. The objectives are to (1) select suitable lipid and surfactant for high drug loading and stability; (2) prepare SLNs by hot homogenisation and optimize process parameters to achieve narrow particle size distribution and high encapsulation efficiency; and (3) characterize the formulations and evaluate in-vitro release and stability to identify promising candidates for further development.[4]

MATERIALS

Deoxycortisone procured from Hetero Labs, HYD. Glyceryl monostearate, Poloxamer 407, Tween 80 and Span 20 were obtained from Synpharma Research Labs, Hyderabad. Other chemicals and the reagents used were of analytical grade.

METHODOLOGY

Compatibility study (IR spectroscopy)

The drug-excipients compatibility was ascertained by subjecting the drug and homogenates of drug and polymer to Infrared spectrophotometric study.[5]

Method of preparation of Deoxycortisone loaded solid lipid nanoparticles:

High pressure homogenization was used to create the Deoxycortisone loaded SLN. One part methanol to one part chloroform was used to dissolve the Deoxycortisone and Glyceryl monostearate. A rotary flash evaporator was used to eliminate organic solvents. Heating to 5 °C above the melting point of the lipid melted the buried lipid layer. Tween 80 and span 20 stabilizers were dissolved in distilled water to make 30 ml, and the aqueous phase was then heated to the same temperature as the oil phase. The hot aqueous phase was introduced to the oil phase, and a high-pressure homogenization was used for 30 min. at 3000 rpm and 50 °C. The resulting coarse oil in water emulsion was subjected to a 25-minute probe sonication procedure. The heated Nano emulsion was eventually allowed to cool to room temperature and was kept refrigerated at 4 °C to produce the Deoxycortisone-loaded SLN.[6]

Table -1: Composition of Deoxycortisone for preparation of solid lipid nanoparticles

Ingredients	**F1**	**F2**	**F3**	**F4**	**F5**	**F6**	**F7**	**F8**
Deoxycortisone	10	10	10	10	10	10	10	10
Glyceryl monostearate	50	50	50	50	50	50	50	50
Poloxamer 407	20	20	20	20	20	20	20	20

Tween 80	1	1.5	2	2.5	-	-	-	-
Span 20	-	-	-	-	1	1.5	2	2.5
Methanol	10	10	10	10	10	10	10	10
Chloroform	20	20	20	20	20	20	20	20

Evaluation of Deoxycortisone loaded nanoparticles:

Particle Size and Zeta Potential

The particle size of the formulation was determined by photo correlation spectroscopy with a zeta master (Malvern Instruments, UK) equipped with the Malvern PCS software. Every sample was diluted with distilled water. The surface charge (Zeta potential) was determined by measuring the electrophoretic mobility of the nanoparticles using a Malvern zeta sizer (Malvern Instruments, UK). Samples were prepared by diluting with distilled water.[7]

SEM analysis:

The morphology of NPs was studied by a scanning electron microscope. For this purpose, the sample was lyophilized and placed on aluminum stubs and the surface was coated with a layer of gold particles using a sputter coater. The shape of the NPs was determined by scanning electron microscopy (SEM) (XL30, Philips, the Netherlands) at 15 kV and 750 mA.[8]

Drug encapsulation efficiency:

Lyophilized nanoparticles 50mg were dissolved in 100ml of phosphate buffer and the drug amount was determined by UV analysis. The encapsulation efficiency was determined as the mass ratio of entrapped Deoxycortisone in nanoparticles to the theoretical amount of the drug used in the preparation.[9] The entrapment of the Deoxycortisone nanoparticles was expressed as loading capacity.

Amount entrapped

$$\text{Entrapment Efficiency (\%)} = \frac{\text{Total drug loaded}}{\text{Total drug loaded}} \times 100$$

In-vitro drug release studies:

The release studies were carried out by franz diffusion cell. It containing 10 ml Phosphate buffer. Phosphate buffer pH 7.4 (100 ml) was placed in a 10 ml of beaker. The beaker was assembled on a magnetic stirrer and the medium was equilibrated at 37 ± 5^0C. Dialysis membrane was taken and one end of the membrane was sealed. After separation of non-entrapped Deoxycortisone dispersion was filled in the dialysis membrane and other end was closed. The dialysis membrane containing the sample was suspended in the medium. 1ml of aliquots were withdrawn at specific intervals, filtered after withdrawal and the apparatus was immediately replenished with same quantity of fresh buffer medium.[10]

Percentage of drug release was determined using the following formula.

$$\text{Perentage drug release} = \frac{\text{Da}}{\text{Dt}} \times 100$$

Where, Dt = Total amount of the drug

Da = The amount of drug released

Drug release kinetics:[11]

The models used were zero order (equation 1) First order (equation 2) and Higuchi model (equation 3) and Korsmeyerpeppas model (equation 4).

i) zero order kinetics:

$$R = Ko\ t \qquad \text{-- (1)}$$

R=cumulative percent drug

Ko=zero order rate constant

ii) First order kinetics

$\log C = \log Co - K_1 t / 2.303$ -- (2)

where C = cumulative percent drug

K_1= first order rate constant

iii) Higuchi model

$R = K_H t^{0.5}$ -- (3)

Where R = cumulative percent drug

K_H =higuchi model rate constant

iv) Korsmeyer peppas model:

$M t / M \alpha = K_k t^n$

$$\log M t / M \alpha = \log K_k + n \log t \quad \text{-- (4)}$$

where K_k =Korsmeyerpeppas rate constant

'M t / M α' is the fractional drug, n = diffusional exponent, which characterizes the mechanism of drug.

The obtained regression co-efficient (which neared 0.999) was used to understand the pattern of the drug from the nanoparticles.

Stability studies:[12]

Selected Formulation was subjected to stability studies as per ICH guidelines.

Following conditions were used for Stability Testing.

1. 25^0C/60% RH analyzed every month for period of three months.

2. 30^0C/75% RH analyzed every month for period of three months.

3. 40^0C/75% RH analyzed every month for period of three months.

RESULTS AND DISCUSSION

FT-IR Spectrum of Deoxycortisone

FT-IR Spectra of Deoxycortisone and excipients were recorded. All these peaks have appeared in formulation and physical mixture, indicating no chemical interaction between Drug and lipids. It also confirmed that the stability of drug during encapsulation process.

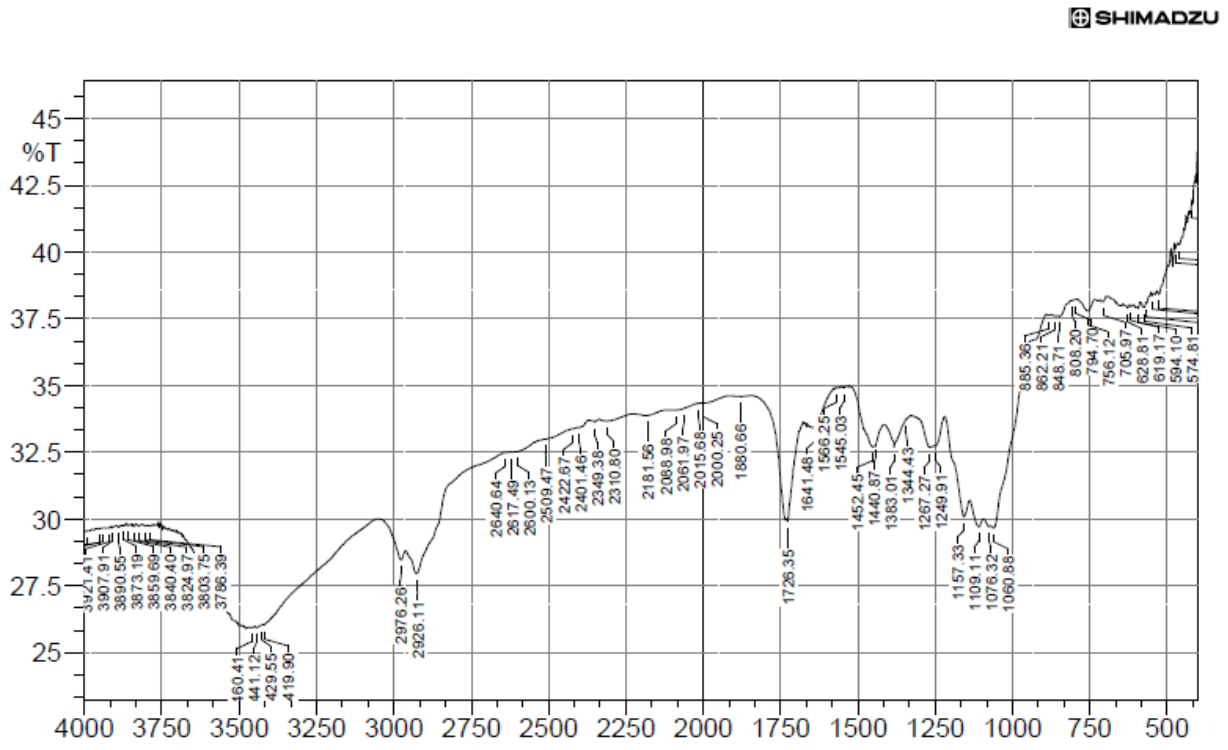

Fig-1: FTIR Studies of Deoxycortisone

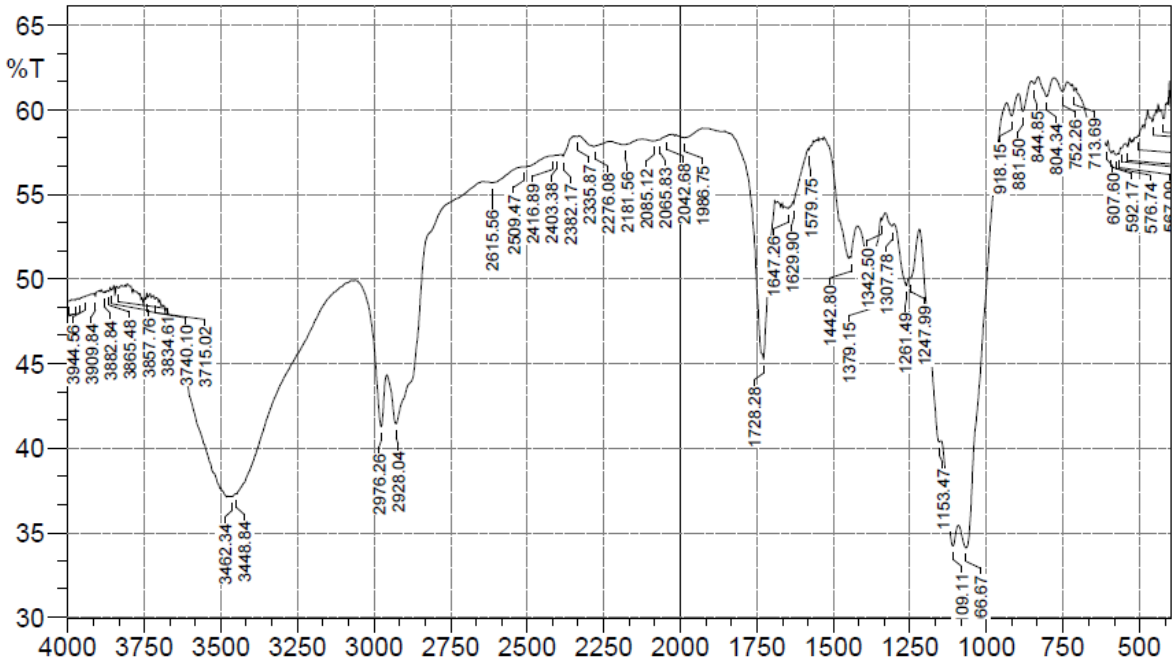

Fig-2: FTIR Studies of optimized formulation

EVALUATION PARAMETERS

Particle size:

With an increase in lipid concentration, the particle size increased. based on entrapment effectiveness and particle size distribution.

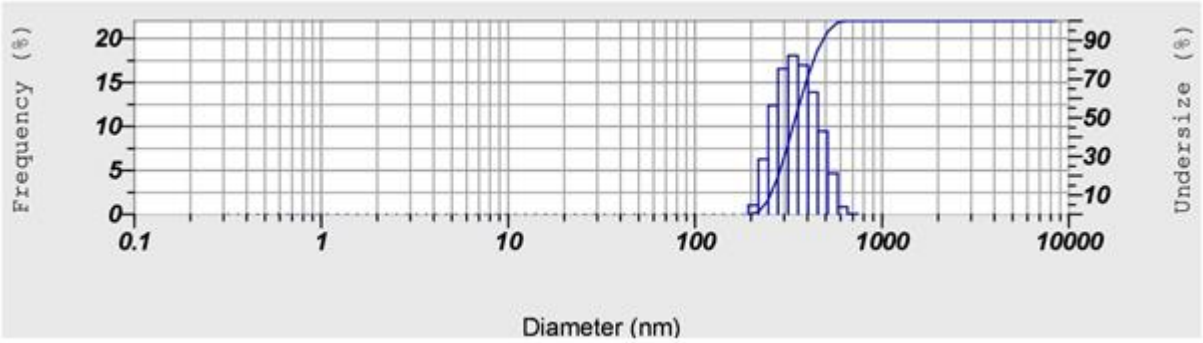

Fig-3: Particle size analysis of Optimized formulation

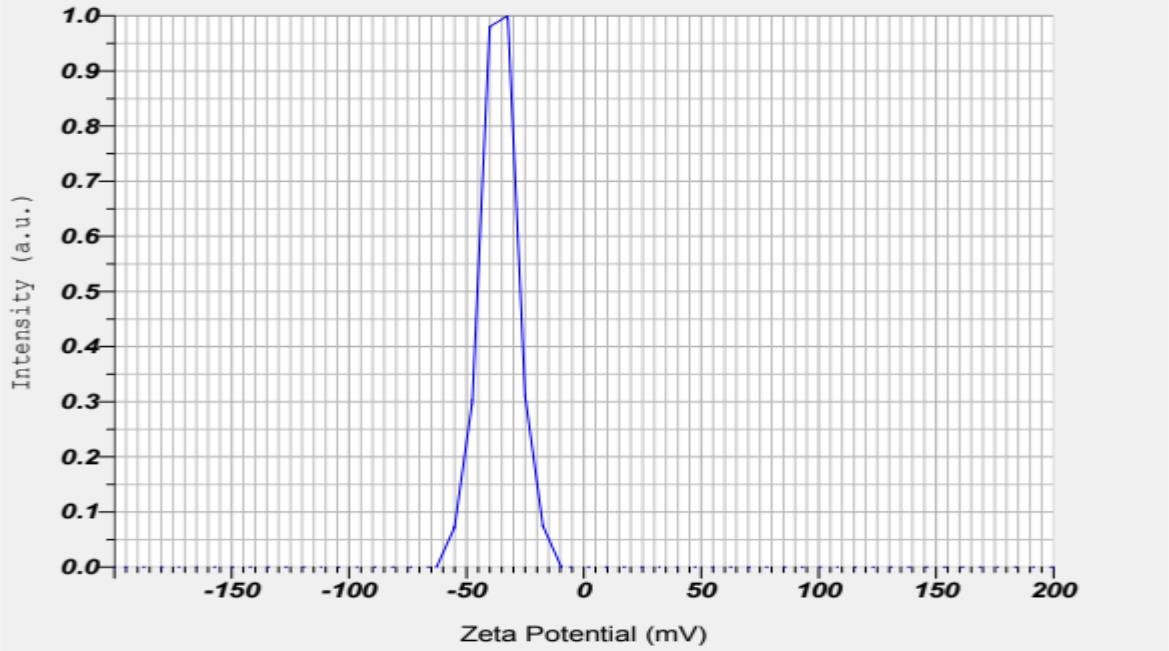

Fig-4: Zeta potential of Optimized formulation

Drug entrapment efficiency:

Optimizing the lipid concentration to be used in the creation of solid lipid nanoparticles was the first step of the work plan. Based on the particle size and entrapment effectiveness of the discovered solid lipid nanoparticles, the lipid content was optimized.

Table-8: Evaluation Studies of Prepared solid lipid nanoparticles: Entrapment Efficiency and Particle size

Batch No	**Particle size (nm)**	**Zeta potential (mV)**	**Entrapment Efficiency (%)**
F1	149	-26	76.93
F2	151	-22	81.20
F3	148	-28	79.86

F4	143	-25	82.35
F5	157	-29	80.19
F6	137	-27	83.98
F7	132	-23	91.16
F8	146	-25	85.49

Surface morphology:

According to scanning electron microscopy (SEM), the solid lipid nanoparticles were round, smooth, and free of any aggregation.

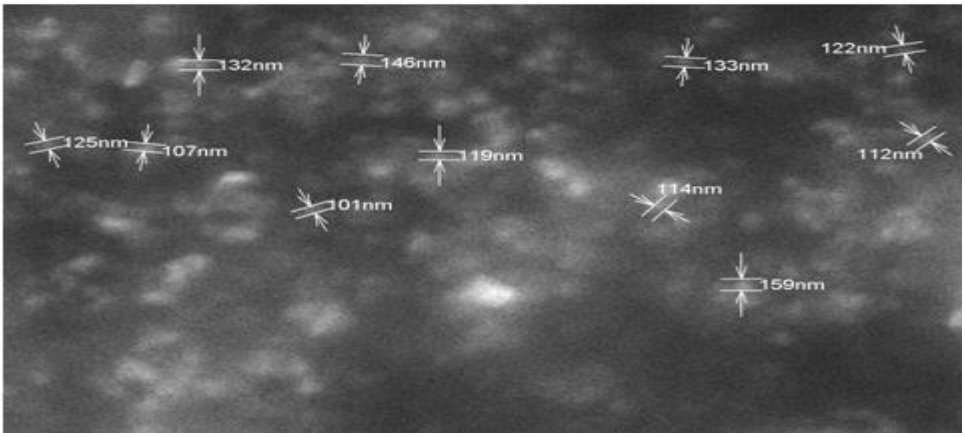

Fig-5: SEM analysis of Optimized Solid lipid nanoparticle

In vitro drug release studies

Using a dialysis membrane and a pH 7.4 buffer, the in vitro diffusion investigations were carried out for eight hours. This resulted from the drug's release from the surface of the solid lipid nanoparticles. Later, for 8 hours, a consistent and gradual medication release was seen. The lipid and surfactant ratio in the F7 formulation was shown to be the most effective one.

Table-9: *In vitro* drug release profiles of SLN (F1-F8)

Time	F1	F2	F3	F4	F5	F6	F7	F8

0	0	0	0	0	0	0	0	0
1	18.63	19.67	16.39	17.84	16.89	15.97	19.68	18.69
2	26.35	22.35	21.14	22.50	21.20	20.33	26.98	27.56
3	37.10	36.79	35.95	36.93	33.65	36.49	39.67	35.67
4	55.69	55.51	54.72	49.86	45.37	44.20	48.55	47.82
6	69.86	66.39	68.18	62.10	60.10	59.86	63.59	65.86
8	78.91	75.52	74.96	72.15	71.25	78.97	79.82	76.15
10	84.13	82.15	83.29	80.22	81.64	83.58	85.22	86.98
12	93.69	94.63	95.56	92.39	94.58	96.36	98.55	97.25

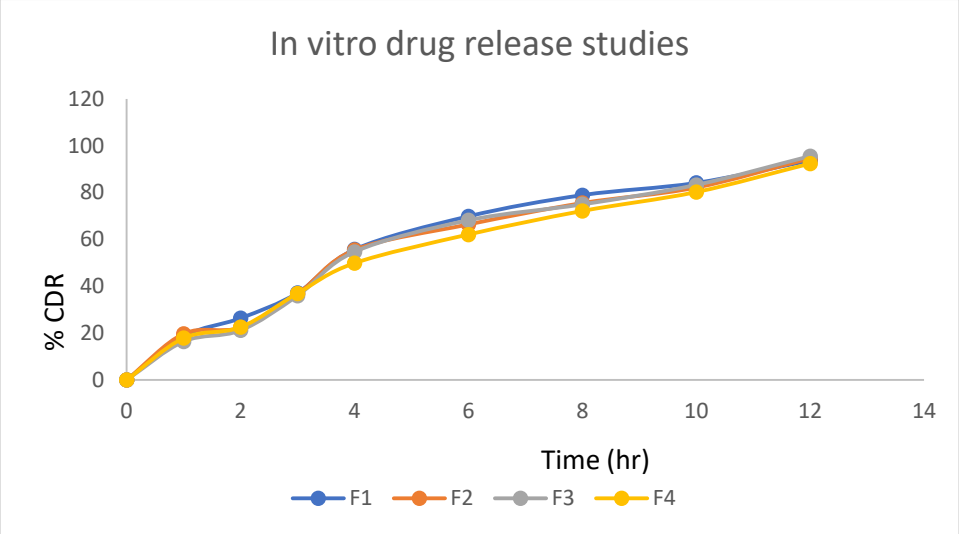

Fig-6: Drug release for (F1-F4) formulations

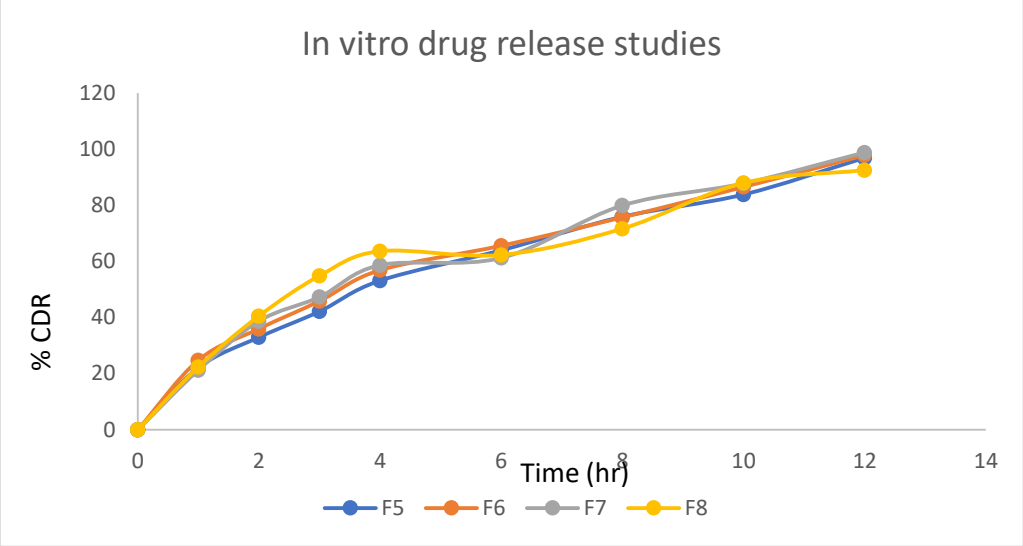

Fig-7: Drug release for (F5-F8) formulations

Kinetic modelling of drug release

All the 8 formulation of prepared Deoxycortisone SLN were subjected to in vitro release studies these studies were carried out using diffusion apparatus.

The results obtaining in vitro release studies were plotted in different model of data treatment as follows:

1. Cumulative percent drug released vs. time (zero order rate kinetics)
2. Log cumulative percent drug retained vs. time (First Order rate Kinetics)
3. Cumulative percent drug released vs. square root of time (Higuchi's Classical Diffusion Equation)
4. Log of cumulative % release Vs log time (Peppas Exponential Equation)

Zero order kinetics

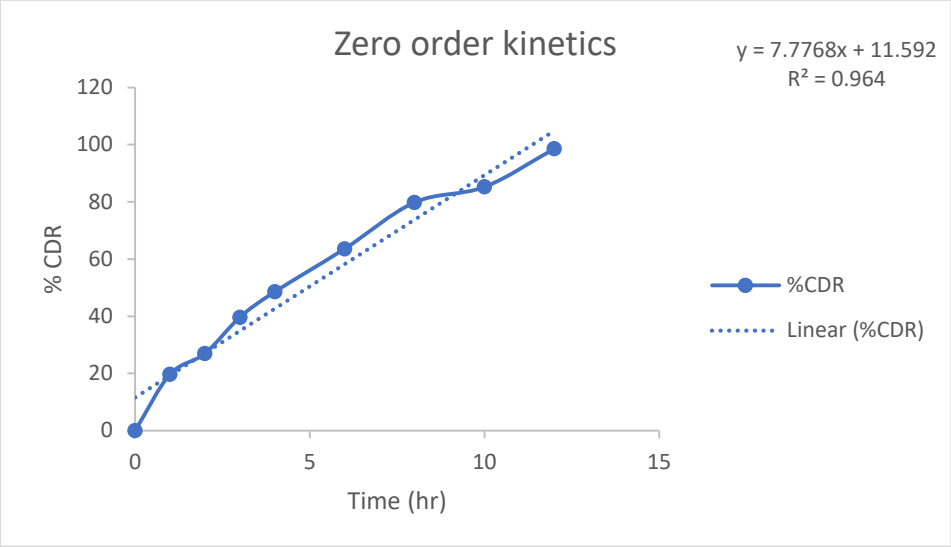

Fig-8: Zero order kinetics of optimized formulation

First order kinetics

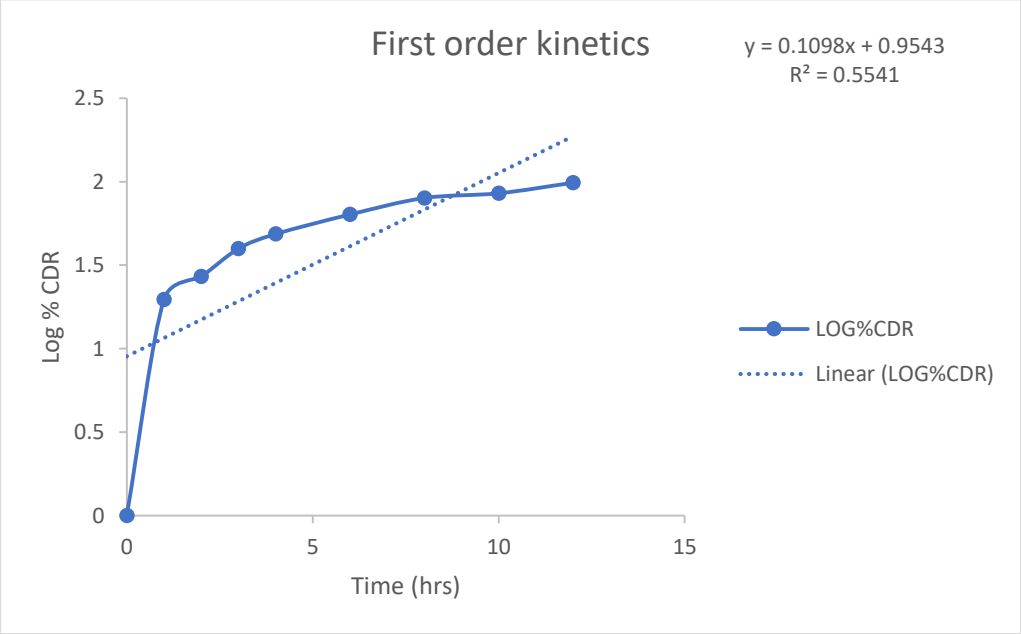

Fig-9: First order kinetics of optimized formulation

Higuchi model

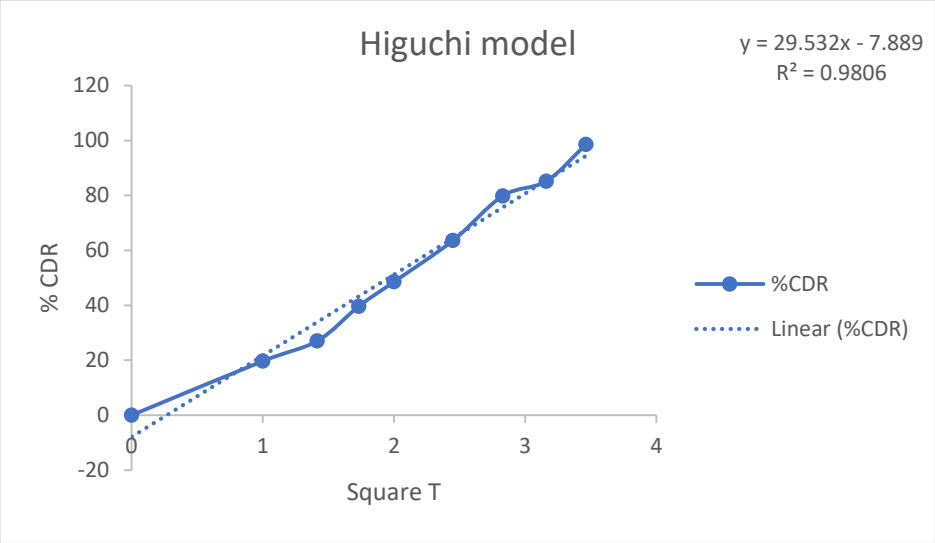

Fig-10: Higuchi model of optimized formulation

Korsmeyer peppas

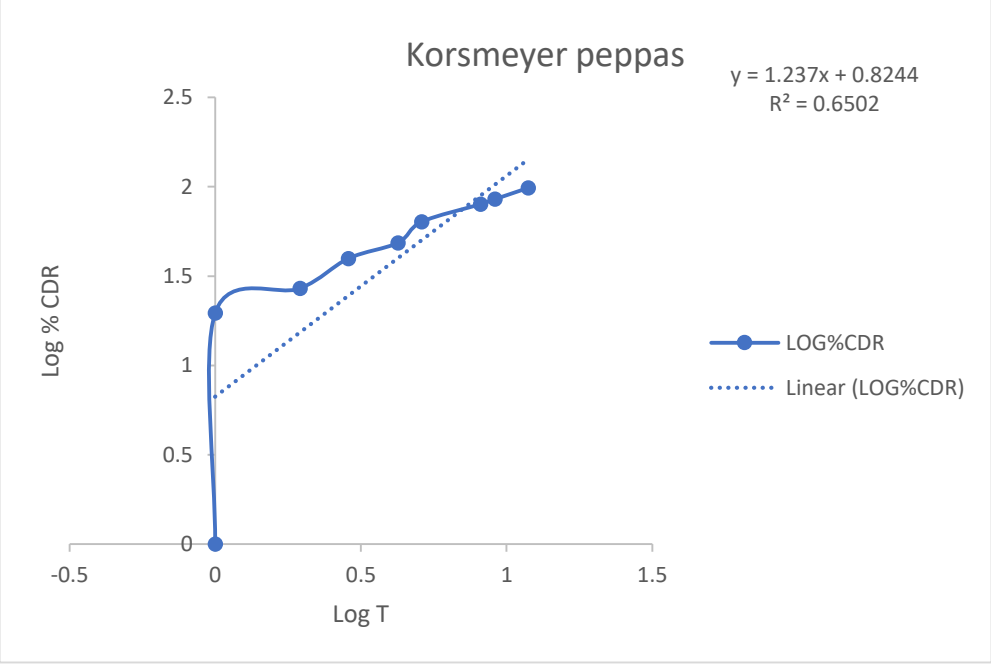

Fig-11: Korsmeyer peppas of optimized formulation

The values of in vitro release were attempted to fit into various mathematical models. Plots of zero order, first order, Higuchi matrix and Peppas.

Regression values are higher with Zero order release kinetics. Therefore, all the Deoxycortisone SLN follows Zero order release kinetics. The table indicates that r^2 values are higher for Higuchi's model compared for all the Deoxycortisone SLN. Hence Deoxycortisone release from all the solid lipid nanoparticles followed diffusion rate-controlled mechanism.

Stability studies:

After three months, the physical and chemical characteristics of the solid lipid nanoparticles of formulation F-7 had not significantly changed. The parameters quantified at various times were displayed.

Table-10: Results of stability studies of optimized formulation F-7

Formulation Code	Parameters	Initial	1st Month	2nd Month	3rd Month	Limits as per Specifications
F-7	25^0C/60%RH	98.55	97.56	96.35	95.85	Not less than
F-7	30^0C/75% RH	98.55	97.21	96.20	95.48	Not less than
F-7	40^0C/75% RH	98.55	97.26	96.13	95.36	Not less than

CONCLUSION

The current study suggested a unique Deoxycorticosterone solid lipid nanoparticle formulation for regulated release. Investigation into the solid lipid nanoparticles' production, characterization, and in-vitro release was done. The numerous formulations with varied drug-lipid and surfactant ratios were analyzed and improved. A drug encapsulation effectiveness of up to 91.16 % has been attained in this

study. Deoxycorticosterone solid lipid nanoparticles containing soy lecithin were created using the Homogenization method, then the particle size was decreased by sonication. formulations using solid lipid nanoparticles performed well in terms of medication content and encapsulation effectiveness. This shows that the formulation procedure was suitable and reproducible in nature, and it provided a good yield. The formulation with the best encapsulation efficiency was (F-7) It was discovered that the percentage of encapsulation efficiency rose along with the soy lecithin concentration. According to the method described, permeation studies with dialysis membrane were conducted. The in vitro drug release profiles of all the formulations indicated an initial burst effect, followed by a gradual drug release. The formulations demonstrated good drug release from the lipid. These solid lipid nanoparticles contained more Deoxycorticosterone and released it more quickly.

REFERENCES

1. Mehnert, W., & Mäder, K. (2001). Solid lipid nanoparticles: production, characterization and applications. Advanced Drug Delivery Reviews.
2. Müller, R. H., Mäder, K., & Gohla, S. (2000). Solid lipid nanoparticles (SLN) for controlled drug delivery — a review of the state of the art. European Journal of Pharmaceutics and Biopharmaceutics.
3. Wissing, S. A., & Müller, R. H. (2003). The influence of the surfactant and the production method on the physical stability of solid lipid nanoparticles (SLN). International Journal of Pharmaceutics.
4. Freitas, C., & Müller, R. H. (1998). Effect of process parameters on the production of solid lipid nanoparticles by high pressure homogenization. Journal of Controlled Release.
5. Doktorovová, S., Silva, A. M., & Souto, E. B. (2014). Lipid nanoparticles — production, characterization and applications in drug delivery. Handbook of Experimental Pharmacology (Chapter on lipid nanoparticles).
6. Pardeike, J., Hommoss, A., & Müller, R. H. (2009). Lipid nanoparticles (SLN, NLC) in cosmetic and pharmaceutical dermal products. International Journal of Pharmaceutics.
7. Müller, R. H., Keck, C. M. (2004). Challenges and solutions for the delivery of biotech drugs — a review of drug nanocrystal and lipid nanoparticle technologies. Journal of Biotechnology.

8. Patel, A. R., & Zhang, J. (2012). High pressure homogenization: principles and applications for nanoparticles. Pharmaceutical Technology (review).
9. Rabinarayan Parhi, Preparation and Characterization of Solid Lipid Nanoparticles-A Review Current Drug Discovery Technologies, Volume 9 , Issue 1 , 2012 DOI : 10.2174/157016312799304552.
10. Taraka sunil kumar k, mohan varma m, and ravi prakash p. "development and optimization of enzalutamide-loaded solid lipid nanoparticles using box–behnken design". Asian journal of pharmaceutical and clinical research, vol. 12, no. 6, june 2019, pp. 67-76, doi:10.22159/ajpcr. 2019.v12i6.31617.
11. Chen H, Chang X, Du D, Liu, W, Liu J, Weng T, Yang Y, Xu H, Yang X. Podophyllotoxin-loaded solid lipid nanoparticles for epidermal targeting J. Control. Rel. 2006; 110: 296-306.
12. Cavalli R, Caputo O, Marengo E, Pattarino F, Gasco MR. The effects of components of micro emulsions on both size and crystalline structure of solid lipid nanoparticles (SLN) containing a series of model molecules. Pharmacies, 1998; 53: 392-396.