RESEARCH ARTICLE

Solid Lipid Nanoparticle-based Gel to Enhance Topical Delivery for Acne Treatment

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ABSTRACT

The aim of the present study was to investigate the treatment of acne and pimples by formulating *Terminalia chebula*-loaded solid lipid nanoparticles (SLNs) based gel. Methanolic extract of *T. chebula* is loaded into SLN by single emulsification - Solvent evaporation method followed by ultrasonication. Box-Behnken Design was employed for the optimization of the formulation. The goal was to produce SLN with the highest %entrapment efficiency while also reducing particle size. Particle size (Y1) and entrapment efficiency (Y2) were significantly affected by a few factors, including the amount of lipid, the concentration of the surfactant, and the time of the sonication, which is considered as a critical factor during the optimization of SLN. The average particle size *T. chebula*-loaded SLNs decreased with increasing concentration of surfactant. The SLNs particle size of (201 nm) with a polydispersity index of (0.342) was obtained at significant concentration of lipid and surfactant. High entrapment efficiency of SLN's 70.11% revealed the ability of SLNs to incorporate a high quantity of *T. chebula*. Further, When TC loaded SLN incorporated into carbopol 934 Gel, it showed initial burst release followed by a sustained release at 8 hour time span. Carbopol 934 increases the contact time of formulation via the topical route.

Keywords: Solid lipid nanoparticle, Box-Behnken design, Particle size, Entrapment efficiency, Topical route.

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INTRODUCTION

Acne vulgaris (AV) is a disorder of the pilosebaceous unit that causes scarring in varying degrees as well as non-inflammatory and inflammatory lesions like open, closed comedones and papules, pustules, nodules, respectively. In addition to other detrimental physiological and psychological effects on patient well-being and self-esteem, it frequently starts in the first few years of adolescence and produces pain, scarring, and disfiguring lesions, especially in young patients. Primary pathogenic mechanisms lead to acne lesions, alteration of follicular keratinization that leads to comedones; i.e., *Propionibacterium acnes* colonization of the follicles, increased and changed sebum production under the control of androgens, complicated inflammatory processes involving both innate and acquired immunity.[1]

Acne is mostly caused by the anaerobic pathogenic microbe *P. acne*s found on human skin. *P. acnes* produce a number of different enzymes, including protease, lipase, acid phosphatase and hyaluronidase, which are used to spread acne. 9.4% of humans on the earth, or around 650 million people have acne. It affects nearly 90% of teenagers and can occasionally linger into adulthood due to an increase in testosterone hormone. Females are affected somewhat more often than males are (9.8 vs. 9.0%, respectively). *P. acnes,* anaerobic pathogenic bacteria that inhabit human skin, are essential for the emergence of acne.[2]

The main cause of acne vulgaris lesions is one of the irritant-free fatty acids that are produced when lipase hydrolyzes sebum triglycerides in pilosebaceous follicles. According to earlier research, inflammation is decreased when free fatty acids are administered intradermally because they stimulate follicular epithelial growth. 40% acne patients taking lipase inhibitors, particularly those containing halopyridyl phosphorus, showed decreased levels of free fatty acids. The current study used plant extracts to study the antibacterial activity of plants like *Terminalia chebula*. *P. acnes* demonstrated the greatest bactericidal reaction to methanolic extracts of *T. chebula* (750 µg).[3] *T. chebula* is very effective in inhibiting lipase activity. The phenolic component of *T. chebula*, particularly chebulagic acid, was discovered to be a powerful inhibitor of lipase activity. The solid lipid nanoparticle was first developed in 1991. SLNs are the fastest-growing area in colloidal drug delivery systems. SLN are colloidal drug carriers that range in size from 50 to 1000 nm. Biodegradable/biocompatible solid lipids or a mixture of lipids can be used to make SLN delivery vehicles. These SLN's have gained great interest recently

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because, as colloidal drug carriers, they combine the benefits of oil emulsions, liposomes, and polymeric nanoparticles while minimizing any of their limitations. Lipids used in SLN preparation have approved status, such as generally recognized as a safe (GRAS). It comprises food additives or excipients that are acceptable for human intake due to their low toxic effects in topical cosmetic or pharmaceutical formulations. Occlusion characteristics are formed by film deposition on the skin, which can aid drug diffusion through the stratum corneum.[4]

MATERIALS AND METHODS

Materials

T. chebula was purchased from MPREX Healthcare Pvt Ltd, Pune. Tween 80 were purchased from Lobachemie, Mumbai. Poloxamer 188 as gift sample from BASF, Mumbai. All the solvents and reagents were of analytical grade. Before use, distilled water was filtered using a 0.22 µm membrane filter. Dialysis bag (Pore diameter, molecular size cut off 12 to 14 KD, 2.4 nm DM 70 membrane) was purchased from Himedia, Mumbai

Methods

Solubility study of drug in different lipids

Several lipids, including stearic acid, glyceryl monostearate, precirol ® ATO5, cetyl alcohol, compritol ® 888 ATO were examined. One gram of each lipid was heated in a vial on a magnetic stirrer at a temperature that was (5–10°C) over its melting point for this purpose. The vial was progressively filled with *T. chebula*, which was introduced in little volumes. After each addition, the mixture was agitated for 30 minutes. The drug's indicated saturation solubility in the corresponding lipid was examined for transparency loss or the development of a brown color.

The lipid with the greatest drug solubility was used to synthesize the SLN.

Preparation of T. chebula-loaded solid lipid nanoparticles

T. chebula-loaded SLN was prepared using single emulsification Solvent evaporation method followed by ultrasonication.[5] Stearic acid and *T. chebula* were dissolved in ethanol (organic phase) together. Tween 20 was used as a hydrophilic surfactant in the aqueous phase, which was likewise maintained at 80°C. Tween 80 (surfactant) and poloxamer 188 (co-surfactant) were dissolved to make the aqueous phase and poured slowly to oil phase, O/W coarse emulsion was obtained which was further sonicated by probe sonicator (PCI analytics PVT Ltd PKS 750). Solid lipid nanoparticle dispersion was obtained which was stored at airtight container.

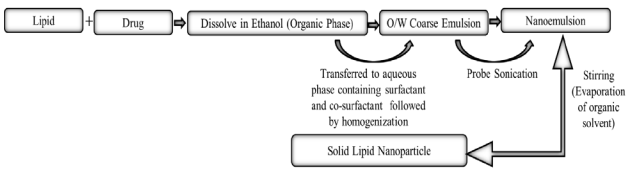

Figure 1: Preparation method of SLN used in study

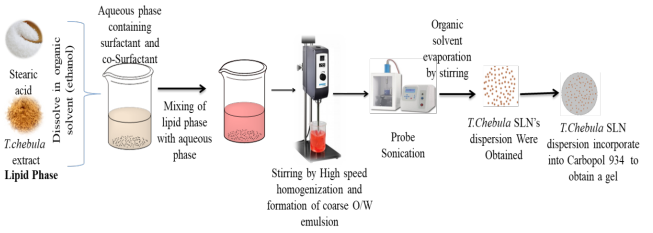

Figure 2: Detail method of preparation of SLN

Table 1: Independent variables and their levels

Independent variables	*Levels*		
	Low (-1)	*Medium (0)*	*High (+1)*
Amount of lipid (w/v)	100	150	200
Concentration of Surfactant (%)	1	2	3
Sonication Time (Min)	10	15	20

Table 2: Dependent variables and their range.

Dependent Variable (Responses)	*Constraints (In Range)*
R1- Particle Size	>500 (minimize)
R2- % Entrapment Efficiency	55-75% (maximize)

Optimization of TC SLN by Box-Behnken design

Utilizing Design Expert Software Version Design-Expert®13, the Box Behnken design was used to optimize SLN.[5] The dependent variables Y1 and Y2 were Particle size and Entrapment efficiency respectively. Amount of lipid, surfactant concentration and sonication time were the independent variables (Table 1 and 2). A total of 17 batches were developed. The effect of independent parameters on particle size and entrapment efficiency was depicted using 3D plots produced by BBD. The trial runs performed using the Box-Behnken design is shown in Table 3.

Evaluation of TC loaded Solid Lipid Nanoparticles

All 17 batches were evaluated for parameters which are listed below

Analysis of particle size and polydispersity index (PDI)

Particle size analyzer (Horiba SZ-100) was used to assess the size of the SLN particles. SLN were diluted before analysis to avoid agglomeration, and ultrasonication was used.[6] The measurement was made at 25°C and a fixed angle of 90°.

The PDI, another significant variable, describes the width or spread of the particle size distribution. The PDI value is between 0 and 1. PDI values less than 0.1 imply monodisperse particles and greater than 0.1 imply polydisperse particle size distributions.[7] Horiba SZ-100 was also used to assess PDI.

Zeta potential analysis

It is a measurement of the actual electric charge that is present on nanoparticle surfaces. Zeta potentials with larger size indicate more stable particles. The magnitude and sign of the zeta potential can be used as an additional parameter to assess surface chemical changes.[8] We have diluted the zeta potential of SLN loaded with *T. chebuala* with HORIBA SZ-100 with double distilled water.

Table 3: Optimization of TC loaded SLN in BBD with observed responses

Run	*Factor-1* *Amount of lipid (mg)*	*Factor-2* *Amount of surfactant (%w/v)*	*Factor-3* *Sonication time (min)*	*Response-1* *Particle size (nm)*	*Response-2* *Entrapment efficiency (%)*
1	200	2	20	267 ± 1.3	67.32 + 1.21
2	150	3	10	244 ± 0.9	69.00 ± 1.09
3	150	2	15	295 ± 2.1	69.23 ± 1.9
4	150	2	15	296 ± 1.09	67.24 ± 2.1
5	150	1	20	331 ± 1.8	64.54 ± 1.7
6	100	2	20	271 ± 2.09	67.11 ± 2.01
7	150	1	10	329 ± 1.02	63.09 ± 2.07
8	100	2	10	340 ± 1.04	66.6 ± 2.4
9	200	3	15	215 ± 1.1	68.93 ± 1.20
10	200	1	15	254 ± 1.3	62.38 ± 1.32
11	150	3	20	201 ± 1.2	70.11 ± 1.01
12	200	2	10	221 ± 1.5	66.09 ± 1.29
13	150	2	15	295 ± 1.3	67.23 ± 2.1
14	150	2	15	283 ± 0.4	67.2 ± 2.12
15	100	1	15	343 ± 1.32	62.6 ± 1.21
16	100	3	15	215 ± 1.23	68.93 ± 1.3
17	150	2	15	296 ± 1.32	67.13 ± 1.7

Each value is reported as mean S.D. (n=3).

Determination of entrapment efficiency

The entrapment efficiency of *T. chebula* in SLN was examined using dialysis. Two ends of a dialysis bag with the SLN formulation (2 mLH) inside were knotted. The bag was submerged in 100 mL of a 30:70 methanol and distilled water solution at 37.5°C and stirred using a magnetic mixer at 80 to 100 rpm. 5 mL of the sample was taken out of the beaker after 30 minutes, filtered and tested using a UV spectrophotometer at 280 nm (Shimadzu, UV-1800).[9] The drug used for analysis was expected to be free, and the formula below was used to calculate the entrapment efficiency.

$$\text{Entrapment Efficiency} = \frac{\text{Amount of drug added} - \text{Amount of drug in supernatant}}{\text{Amount of drug added}} \times 100$$

Scanning electron microscopy

Using a scanning electron microscope, the morphological characteristics of SLN were identified. The signal that reveals details about the sample's chemical composition and surface morphology (texture).[10] A single drop of the sample was applied on a cover slip, and any extra moisture was dried out at room temperature. At the Savitribai Phule Pune University (SPPU's) Central Instrumentation Facility (CIF), we have performed SEM.

X-ray diffraction

A German-made XRD called the Pan Analytical X'Pert Pro model was used to measure the crystalline nature of the sample. The process was carried out on the sample (optimized SLN's) using a Cu K radiation source (λ = 0.15418 nm) at room temperature. A voltage and current of 45 kV and 40 mA were used to scan at 2θ range 5°-90° 243.[5]

Gel Preparation

The required amount of carbopol 934 was weighed out and mixed with a small amount of distilled water to make an aqueous dispersion. This mixture was hydrated for 4 to 6 hours to form SLN gel.[11] 40 mg drug of *T. chebula* dispersion was added to SLN dispersion with propylene glycol 400, EDTA, methylparaben, and propylparaben. A mechanical stirrer running at 1200 rpm was used to add triethanolamine to the aforementioned dispersion. Up till the carbopol was thoroughly mixed, stirring was done. The gel was left to stand for the next day to liberate any trapped air.

Characterization Study of Gel

Determination of physical appearance and homogeneity

Visual observation was done of the gel's physical characteristics and homogeneity.

Determination of viscocity

The viscosity of the gel was measured using Brookfield viscometer (Brookfield, RVDV pro II, USA) at 37°C using T-bar spindle with helipad attachment at 5, 10, 20, 50 and 100 rpm.[12]

Determination of pH

By placing the electrode in your sample and starting to read. Once your electrode has been inserted into the sample, hit the

measure button and let the electrode sit in the sample for one to two minutes settle on a pH value.

Determination of Spreadability study of SLN gel

Using a fabricated spreadability device, the gel's spreadability was assessed. Two glass slides of 7.5 x 2.5 cm each made up the device. One glass slide was fastened to the wooden board, and the other was mobile and attached to a thread that crossed a pulley to support weight. The two glass slides were separated by 0.5 g of gel sample.[12] In order to release the trapped air between the slides and create a homogenous gel coating, a 100 gm weight was kept on the upper slide and left there for one to two minutes. After the weight was removed, a 20 gm pull was applied to the top slide.[13] The top slide's travel time (in seconds) over a previously set distance was recorded. Following that, spreadability was examined using below formula:

$$S = M \times \frac{L}{t}$$

Where S-spreadability, L-length of glass slide, t-time taken, and M-weight which is tied on upper slide.

Determination of Drug Content

By properly weighing a sample of gel (approximately 400 mg) and dissolving it in a little amount of water (about 100 mL), the drug content of the gels was ascertained. Following that, the solutions were filtered and spectrophotometrically measured at 280 nm.[14]

***In-vitro* drug diffusion SLN Based gel**

Using modified Franz diffusion cells*, in-vitro* diffusion studies were performed. These cells consist of an acceptor and donor compartment, dialysis membrane 70, sample device, magnetic stirring, donor compartment, and thermostatic water bath. The releasing membrane's surface area was 3.14 cm^2. Approximately 45 mL of phosphate buffer saline (PBS), pH 7.4, were used as the receptor compartment, which was agitated by a magnetic bar at 700 rpm to avoid concentration variations only in the donor medium and to reduce stationary layers. In the donor compartment, each SLN-based Gel had 1-mg of drug. The solution on the receptor compartment was kept at 37°C with a 0.5°C error throughout the testing. 5 mL of the sample solution was removed from the receiving chamber after a predetermined time, and the equal amounts of medium were added again.[15] At 280 nm, the samples were examined in triplicate spectrophotometrically.

Antimicrobial Study

Using the agar well diffusion method, the bactericidal activity of *T. chebula* gel and *T. chebula* loaded solid lipid nanoparticles at the same concentration was determined *in-vitro*. For *Staphylococcus aureus* and *Escherichia coli*, respectively, nutrient agar and MacConkey agar are utilized. In an autoclave, the medium was prepared and sterilised for 20 minutes at 121°C.[16] Sterilized media were added to sterile petri plates and given time to set up aseptically. The microbial inoculum on the agar plate inoculated the topmost layer. The antimicrobial extract solution (SLN dispersion and gel) is then added to the well after a 6 mm-diameter hole is aseptically made with a sterile cork borer or tip and a volume (20–100 mL) of the solution at the required concentration. The prepared gel sample and solid lipid nanoparticle were filled into bore at 37 ± 0.5°C for 48 hours under aerobic conditions. Following that, agar plates are incubated at the appropriate temperature based on the test microorganism. In the agar medium, the antimicrobial agent (TC SLN dispersion and TC SLN gel) diffuses and prevents the growth of the tested microbial strain. The zone of inhibition was measured.[17]

Stability Study

In accordance with ICH recommendations, the optimized formulation (SLN dispersion and TC-loaded SLN gel) was kept in a tightly closed container at 40 ± 2°C and 75 ± 5% RH for 3 months in a stability chamber that ICH verified. Change in physical appearance, entrapment efficiency, particle size, and ease of redispersibility are examined for SLN dispersion and change in physical appearance for TC-loaded SLN gel

RESULT AND DISCUSSION

Solubility Study

The most important requirement for component screening is the solubility of least soluble drugs. In solid lipid nanoparticle, preservation of the drug in solubilized form depends on the drug's increased solubility in lipid. Based on solubility studies stearic acid was chosen as lipid matrix. Based on the stability of dispersion surfactant and co-surfactant, Tween 80 and poloxamer 188 were selected, respectively (Table 10).

Optimization and Validation

Maximum entrapment efficiency, physical formulation stability, and a low particle size in the nanoscale range were the main factors for choosing an optimised batch. In order to verify the efficacy of the optimised formulation, a second batch of the identical composition with the predicted response was prepared using the formulation with 136 mg of stearic acid, 3% of tween 80 as a surfactant, and a 20-minute sonication time. It was discovered that the experimental outcomes of the optimised formulation were in good accordance with the value predicted (Figure 4, 5 and Table 3 to 6).

Independent Variables Effect on Particle Size

Given polynomial equation illustrates how a variable affects a particle:

$$\text{Particle size (Y1)} = +293.00 - 26.50*A - 47.75*B - 8.00C + 22.25*AB + 28.75*AC - 11.25*BC - 18.88*A^2 - 17.38*B^2 + 0.6250*C^2$$

The coded equation can be used to calculate the relative significance of the components by comparing the factor coefficients. According to the equation above, the amount of lipid (F-value - 65.65) has a less significant impact than surfactant concentration (F-value -213.16) and sonication time (F-value -5.98). Particle size decreases when independent variables like lipid amount, surfactant concentration, and sonication time increase. Particle size changes when two

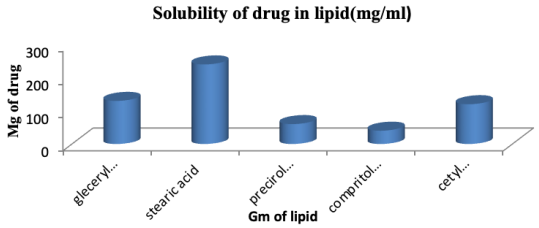

Figure 3: Solubility of drug in lipid (mg/mL)

variables are changed at once. The interaction terms AB, AC and BC and squared terms A2, B2 and C2 show this. When the lipid content increases while the surfactant concentration remains constant, the particle size increases because there isn't enough surfactant. As a result, the surfactant's ability to emulsify becomes less effective, leading to increased particle size and particle aggregation. A smaller emulsion droplet is formed when the interfacial tension between the aqueous and lipid phases is effectively reduced as a result of a high surfactant concentration. An optimum concentration of surfactant is desired to effectively stabilize the SLN particles by preventing particle aggregation. Particle-particle interactions are caused by sonication because the system's higher kinetic energy results in smaller particles. Figure 4 shows 3D design of an optimized batch that illustrates how different variables affect particle size.

Independent Variables Effect on %Entrapment efficiency

The entrapment efficiency of the various batches ranged from 62.60 to 70.11%. Higher entrapment efficiency is preferred for the best possible drug delivery to the target site. The polynomial equation below shows how the variable affects entrapment efficacy:

$$\text{Entrapment Efficiency (Y2)} = +67.61 - 0.0487 * A + 3.03 * B + 0.5375 * C + 0.0875 * AB + 0.1800 * AC - 0.0850 * BC - 0.9168 * A^2 - 1.01 * B^2 + 0.0908 * C^2$$

According to the aforementioned equation, the amount of lipid ratio (f-value of 0.8529) negatively impacts entrapment's effectiveness. Due to the average space being available for drug entrapment into the lipid matrix, the SLN's entrapment efficiency improves as lipid concentration falls. According to the equation, the surfactant had a favorable impact on entrapment effectiveness (F-value of 142.88), although its overall impact was biphasic. Due to the drug's increased solubility in readily available lipid and the decrease in drug partition in the aqueous phase, the initial rise in surfactant concentration results in an improvement of entrapment efficiency. The efficiency of entrapment was positively impacted by sonication time. Figure 4 shows 3D design of an optimized batch that illustrates how different variables affect entrapment efficiency.

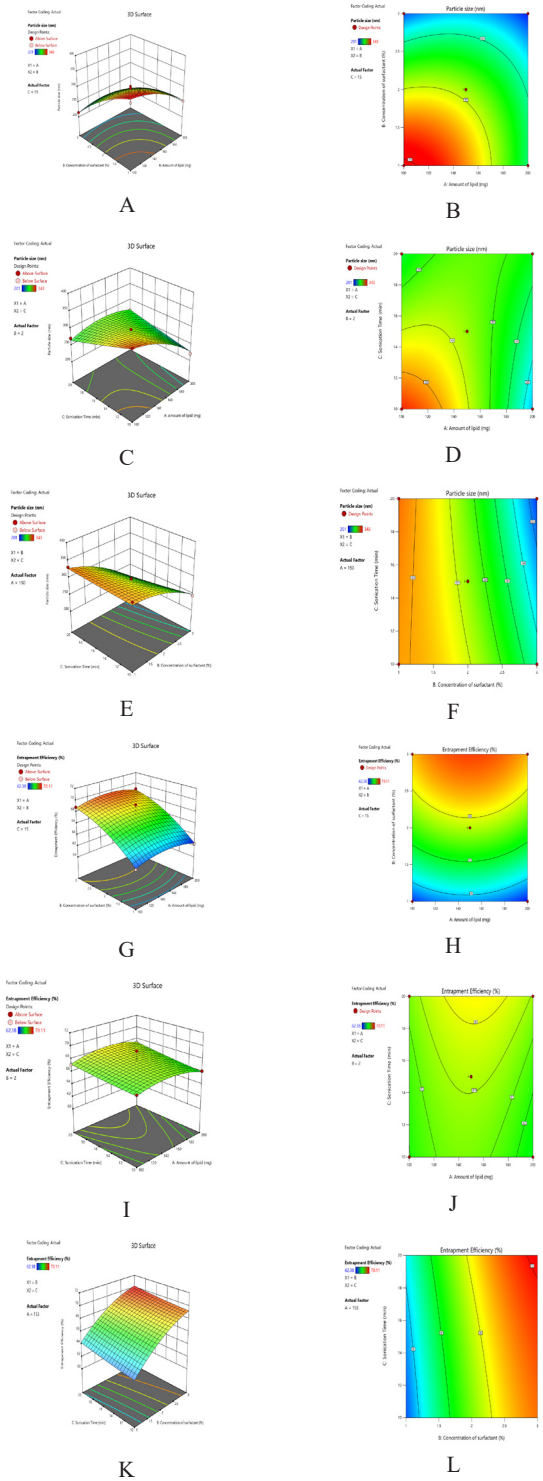

Figure 4: A,C,E,G,I,K -3D response plot B,D,F,H,I,J- Contour plot of depicting the effect of concentration of surfactant, amount of lipid and Sonication time on Particle size and % Entrapment efficiency respectively

Evaluation of Optimized Formulation

Particle size

Dynamic light scattering (DLS) analysis was used to determine the SLN particle size (HORIBA SZ-100). Figure 6. shows the particle size distribution of the optimized SLN. The polydispersity index, which was found to be 0.342, indicates that the particle distribution is polydisperse. Particle sizes for batches B1 through B17 range from 101 to 395 nm. According to Figure 6, the particle size of the optimized batch was discovered to be 201 nm.

Table 3: Fit Statistic of Quadratic Model

(Quadratic Model)	*R2*	*Adjusted R2*	*Predicted R2*	*Inference*
Particle Size	0.9822	0.9594	0.7696	The Predicted R^2 - 0.7696 and the Adjusted R^2 - 0.9594 are reasonably in agreement i.e., difference <0.2.
Entrapment Efficiency (%)	0.9590	0.9064	0.8881	The Predicted R^2 - 0.8881 and the Adjusted R^2 - 0.9064 are reasonably in agreement i.e., difference <0.2.

R^2 = Regression value

Table 4: Result of ANOVA of quadratic model for solid lipid nanoparticle

Response	R1 Particle size	R2 Entrapment Efficiency%
Model	Quadratic Model	Quadratic Model
Some of square	33096.06	88.11
Degrees of freedom	9	9
Mean squares	3677.34	9.79
F-Value	3.95	11.74
P-value (prob>F)	0.0420 Significant	0.0019 Significant
R^2 value	0.9822	0.9836
Lack of fit F-Value	0.0769	0.21
Lack of Fit P value	0.3592 (Non- Significant)	0.8820 (Non- Significant)

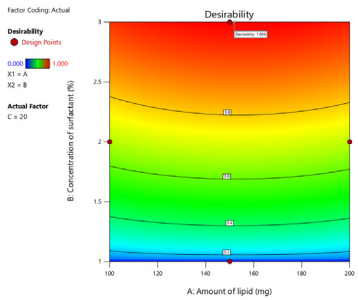

Figure 5: Desirability plot for the software generated solution. Desirability value is 1 which validates the optimized model and process.

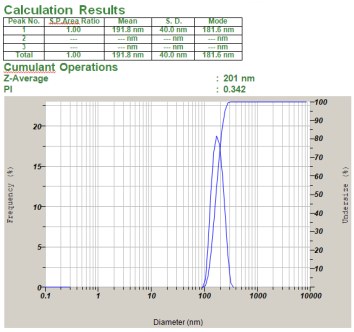

Figure 6: Particle size analysis (optimized batch)

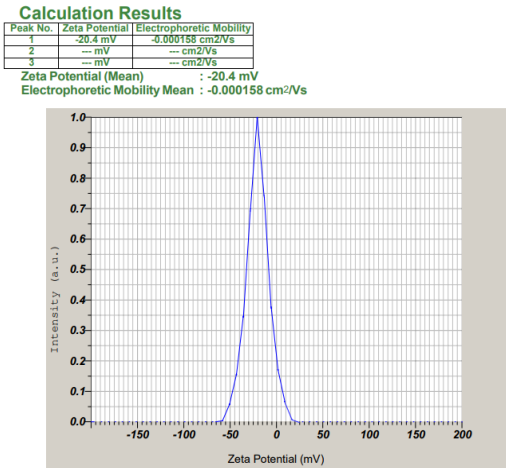

Figure 7: Zeta potential analysis of optimized formulation

Zeta potential

Understanding the stability of nanoparticles in aqueous solutions requires understanding the surface charge potential, which is indicated by the zeta potential. The SLN's zeta potential was found to be -20.4 mv. It was claimed that the surface of the synthesized nanoparticles was negatively charged. By bringing the same charges together, the positive or negative charge on a nanoparticle's surface gives it stability and prevents it from aggregating (Figure 7).

Entrapment efficiency (%)

To deliver the ideal dose of drug to the target site, entrapment efficiency is desired. The amount of surfactant and the duration of the sonication had a significant effect on the entrapment efficiency. Entrapment efficiency for the improved batch was determined to be 70.11%, indicating that the drug is trapped inside the lipid matrix.

Scanning electron microscopy

FESEM images clearly show the presence of synthesized nanoparticles. The nanoparticles are spherical in shape. Nanoparticles are aggregated due to the spreading and drying technique on coverslip and few individual particles are observed. The synthesized nanoparticle is in range of 140.8 to 192.8 nm (Optimized Batch)

Table 5 : Selected optimized batch

Sr.No	*Amount of lipid*	*Concentration of surfactant*	*Sonication Time*	*Particle size*	*Entrap-ment Efficiency*	*Desirability*	*Inference*
1	136.00	3.000	20.000	201.25	70.03	1.000	Selected

Table 6: Predicted and actual values of optimized batch by design expert software

Response	*Predicted values*	*Observed values*
Particle Size	201.00	201
Entrapment efficiency (%)	70.03	70.11

X-Ray Diffractometry

X intercept	*Y count*
20.054	234.705
20.034	233.105

Powder diffraction experiments were performed to further study the interactions between the components of the formulations principle peaks of *T. chebula* loaded SLN was observed at 2θ angle 20.054, 20.034, 21.188, indicating the amorphous nature of the drug, it is due to we don't get a sharp peak. SLN has a complex lipid and drug structure mixture, and we have successfully synthesized SLN.

Evaluation of T. *chebula*-loaded Solid Lipid Nanoparticle-Based Gel

Appearance and clarity

The SLN formulation is faint brown and translucent

Drug content and pH

Drug content of *T. chebula*-loaded SLN gel was found to be 86.92 ± 0.55%.

Viscosity

Observed viscosities at 100, 50, 20,10,5 RPM are shown in Table 7 & Figure 10.

Spreadability

The spreadability of the synthesized SLN gel formulation was determined to be 12.48 0.29 gcm/sec, indicating easy spreadability. The produced gel's thickness and spreading time were inversely related to the gel's spreadability. Good spreadability is preferred to apply the antibacterial gel to the affected area and ensure patient compliance.

In-vitro drug diffusion study

The immediate drug release from the SLN gel formulation was higher because there was more unentrapped drug in the SLN dispersion and more drug on the SLN's outermost shell. After the initial burst, it was discovered that the drug release was sustained, releasing 92 ± 98% of *T. chebula* in *T. chebula* in 8 hours. Terminalia chebula is trapped in SLN and then diffused in a viscous gel matrix in SLN gel. As a result, SLN gel provides two barriers, SLN matrix and gel matrix for drug release by diffusion, maintaining the release of the drug (Table 8 and Figure 10).

Antimicrobial study

The *T. chebula*-loaded solid lipid nanoparticle showed significant effect on microbes *S. aureus* and *E. coli* having zone of inhibition 18.9 ± 0.18 mm and 19.7 ± 0.21 mm, respectively (Table 9 and Figure 11, 12). The zone of inhibition of *S. aureus*

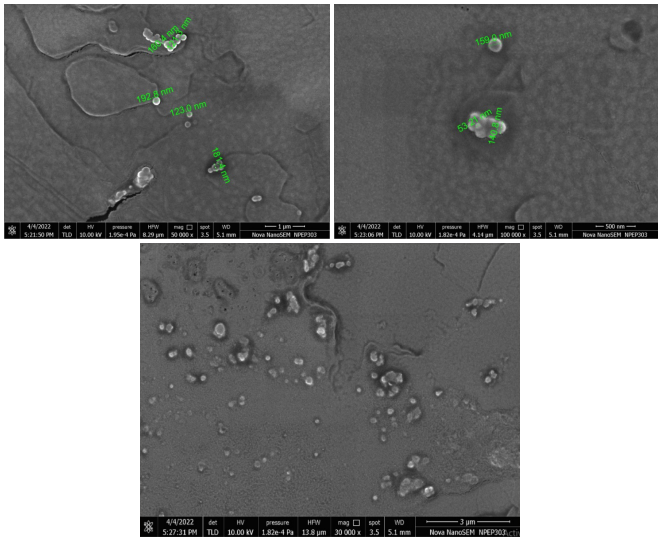

Figure 8: FE-SEM images of solid lipid nanoparticle

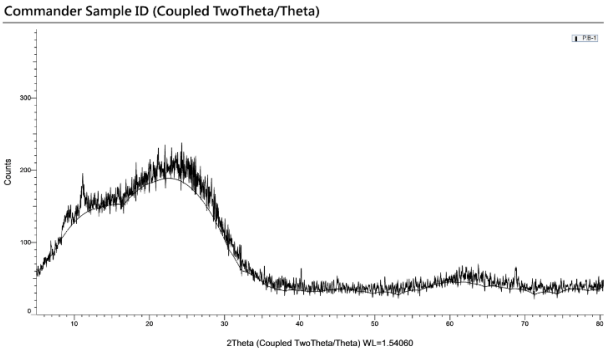

Figure 9: X-Ray Diffraction pattern of *T. chebula* loaded SLN

Table 7: Viscosity of SLN gel formulation

rpm	SLN gel
100	17200 ± 121
50	23600 ± 2.11
20	28000 ± 2.09
10	35530 ± 1.82
5	51390 ± 2.32

Each value is reported as mean S.D. (n=3).

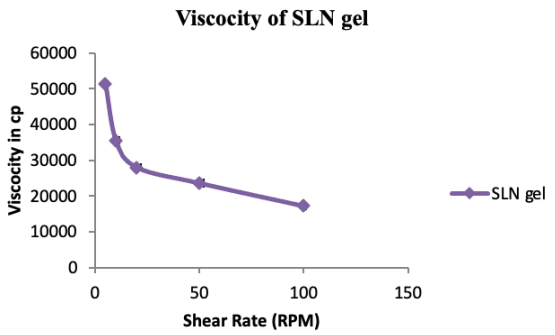

Figure 10: Viscocity of SLN gel

Table 8: Drug release through dialysis membrane of *T. chebula*-loaded SLN gel

Formulation	*Time*	*TC loaded SLN gel*	*TC loaded SLN dispersion*
TC loaded solid lipid nanoparticle gel	0 minutes	0	0
	15 minutes	33.43 ± 2.12	41.58 ± 1.3
	30 minutes	64.5 ± 2.01	75.53 ± 1.43
	60 minutes	75.31 ± 2.13	83.71 ± 1.32
	120 minutes	81.74 ± 2.11	89.54 ± 1.23
	240 minutes	84.81 ± 1.02	93.36 ± 2.12
	480 minutes	89.24 ± 1.09	96.79 ± 2.32

Each value is reported as mean S.D. (n=3)

Table 9: Zone of Inhibition shown by *T. chebula*-loaded SLN and gel

S.No	*Formulation*	*Zone of inhibition*	
		E. coli	*S. aureus*
1	A. SLN Dispersion	18.9 ± 0.18 mm	19.7 ± 0.21 mm
2	B. *T. chebula* loaded SLN gel	15.6 ± 0.25 mm	17.7 ± 0.17 mm
3	C. Control	nil	nil

Each value is reported as mean S.D. (n=3)

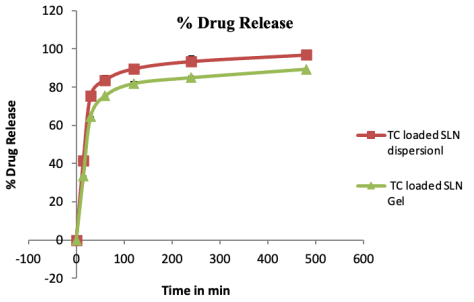

Figure 11: *In-vitro* Drug Release of *T. chebula*-loaded SLN gel

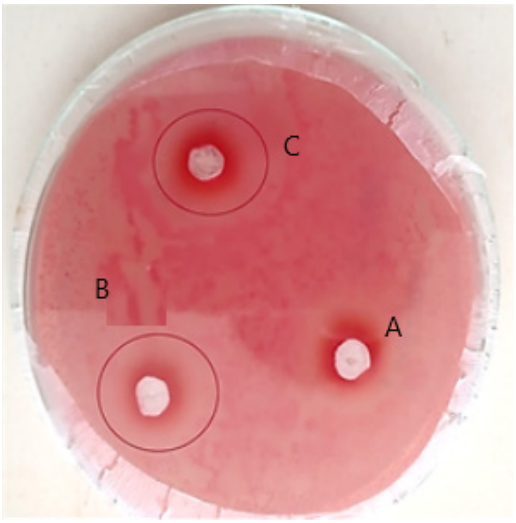

Figure 12: Zone of inhibition shown by A. Control B. SLN dispersion C. SLN gel for *E.coli*

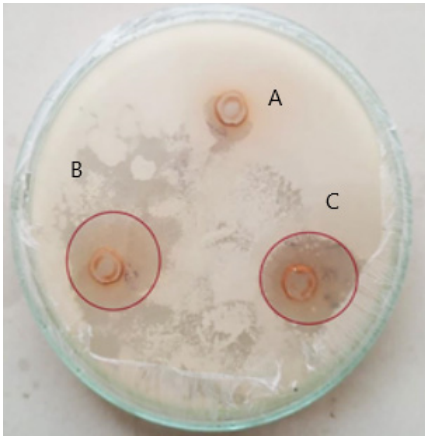

Figure 13: Zone of inhibition shown by A. Control B. SLN dispersion C. SLN gel for *S. aureus*

Table 10: Accelerated stability study as per ICH Guideline

Time in months	*Evaluation parameters (40 ± 2°C and 75 ± 5%RH)*				
	Change In Physical appearance	*Entrapment efficiency*	*Particle Size*	*Ease of Redispersibility*	*Change in Physical Appearance*
	TC loaded SLN	*TC loaded SLN*	*TC loaded SLN*	*TC loaded SLN*	*TC loaded SLN gel*
0 month	No	70.11 ± 0.23%	201 ± 0.06 nm	+	No
1st month	No	69.59 ± 0.29%	204 ± 0.5 nm	+	No
2nd month	No	69.09 ± 0.46%	212 ± 0.67nm	+	No
3rd month	No	68.79 ± 0.32%	219 ± 0.74 nm	+	No

and *e. coli* by *T. chebula*-loaded SLN-based gel were found 15.6 ± 0.25 and 17.7 ± 0.17 mm, respectively.

The result indicate that the SLN formulation has better antimicrobial effect against many causative agents reported

Stability study

The result of stability study shows that TC-loaded SLN dispersion and TC-loaded SLN gel did not show major changes in physical appearance, entrapment efficiency, particle size, and ease of re-dispersibility.

CONCLUSION

The current study is carried out for determining the rationality of phytopharmaceutical *T. chebula*-loaded SLN gel as an antibacterial agent. The optimized SLN formulation, which contains tween 80 and stearic acid, exhibits low particle

size, high drug entrapment, and sustained drug release. This study's findings demonstrated that plants contain bioactive compounds, which are associated with antibacterial characteristics in plants. *T. chebula* gel shows a significant antibacterial effect against causative pathogens like *S. aureus*, *E. coli*. A Box-Behnken Design was used to systematically optimise synthesized drug-loaded SLNs. The findings of this study suggested that a topical formulation for the treatment of acne could be replaced by SLN gel that has been loaded with *T. chebula*.

REFERENCES

1. Ak M. A comprehensive review of acne vulgaris. J. Clin. Pharm. 2019;1(1):17-45.
2. Patil V, Bandivadekar A, Debjani D. Inhibition of *Propionibacterium acnes* lipase by extracts of Indian medicinal plants. International journal of cosmetic science. 2012 Jun;34(3):234-9.
3. Saising J, Voravuthikunchai SP. Anti *Propionibacterium acnes* activity of rhodomyrtone, an effective compound from Rhodomyrtus tomentosa (Aiton) Hassk. leaves. Anaerobe. 2012 Aug 1;18(4):400-4.
4. Das G, Kim DY, Fan C, Gutiérrez-Grijalva EP, Heredia JB, Nissapatorn V, Mitsuwan W, Pereira ML, Nawaz M, Siyadatpanah A, Norouzi R. Plants of the genus Terminalia: an insight on its biological potentials, pre-clinical and clinical studies. Frontiers in pharmacology. 2020 Oct 8;11:561248.
5. Yasir M, Chauhan I, Zafar A, Verma M, Noorulla KM, Tura AJ, Alruwaili NK, Haji MJ, Puri D, Gobena WG, Dalecha DD. Buspirone loaded solid lipid nanoparticles for amplification of nose to brain efficacy: Formulation development, optimization by Box-Behnken design, in-vitro characterization and in-vivo biological evaluation. Journal of Drug Delivery Science and Technology. 2021 Feb 1;61:102164.
6. Kumar VV, Chandrasekar D, Ramakrishna S, Kishan V, Rao YM, Diwan PV. Development and evaluation of nitrendipine loaded solid lipid nanoparticles: influence of wax and glyceride lipids on plasma pharmacokinetics. International journal of pharmaceutics. 2007 Apr 20;335(1-2):167-75.
7. Tantra R, Schulze P, Quincey P. Effect of nanoparticle concentration on zeta-potential measurement results and reproducibility. Particuology. 2010 Jun 1;8(3):279-85.
8. Honary S, Zahir F. Effect of zeta potential on the properties of nano-drug delivery systems-a review (Part 2). Tropical journal of pharmaceutical research. 2013 May 9;12(2):265-73.
9. Deshkar SS, Bhalerao SG, Jadhav MS, Shirolkar SV. Formulation and optimization of topical solid lipid nanoparticles based gel of dapsone using design of experiment. Pharmaceutical nanotechnology. 2018 Dec 1;6(4):264-75.
10. Rodrigues LR, Jose J. Exploring the photo protective potential of solid lipid nanoparticle-based sunscreen cream containing Aloe vera. Environmental Science and Pollution Research. 2020 Jun;27(17):20876-88.
11. Deshkar SS, Palve VK. Formulation and development of thermosensitive cyclodextrin-based in situ gel of voriconazole for vaginal delivery. Journal of Drug Delivery Science and Technology. 2019 Feb 1;49:277-85.
12. Salem HF, Kharshoum RM, Abou-Taleb HA, Farouk HO, Zaki RM. Fabrication and appraisal of simvastatin via tailored niosomal nanovesicles for transdermal delivery enhancement: In vitro and in vivo assessment. Pharmaceutics. 2021 Jan 21;13(2):138.
13. Jain N, Verma A, Jain N. Formulation and investigation of pilocarpine hydrochloride niosomal gels for the treatment of glaucoma: intraocular pressure measurement in white albino rabbits. Drug delivery. 2020 Jan 1;27(1):888-99.
14. Jana S, Manna S, Nayak AK, Sen KK, Basu SK. Carbopol Gel containing chitosan-egg albumin nanoparticles for transdermal aceclofenac delivery. Colloids and surfaces B: Biointerfaces. 2014 Feb 1;114:36-44.
15. Kesharwani R, Sachan A, Singh S, Patel D. Formulation and evaluation of solid lipid nanoparticle (SLN) based topical gel of etoricoxib. Journal of Applied Pharmaceutical Science. 2016 Oct 29;6(10):124-31.
16. Ahuja C, Kaur H, Sharma R. Antibacterial activity of *Terminalia chebula* fruit by agar well diffusion method. Journal of Chemistry, Environmental Sciences and its Applications. 2015 Mar 30;1(2):67-72.
17. Mostafa MG, Rahman M, Karim MM. Antimicrobial activity of *Terminalia chebula*. Int. J Med Arom. Plan. 2011;1(2):175-9.