CODEN [USA]: IAJPBB ISSN : 2349-7750

INDO AMERICAN JOURNAL OF
PHARMACEUTICAL SCIENCES

SJIF Impact Factor: 7.187

https://doi.org/10.5281/zenodo.18008317

Available online at: http://www.iajps.com *Research Article*

FORMULATION AND EVALUTION OF TERBINAFINE ANTI-FUNGA NANO GEL TOPICAL DRUG DELIVERY SYSTEM

Mrs.Thurupu Sowmya* , Dr.Shoaeb Mohammad Syed

Shri Venkateshwara University, Gajraula, Amroha, Uttar Pradesh, India.

Abstract:

The aim of the present investigation was to formulate drug Terbinafine (TBH) encapsulated solid lipid nanoparticles (SLN) loaded in hyaluronic acid gel which have prolonged anti Fungal activity. The compatibility between the selected drugs and polymers has been studies using DSC and FTIR studies. The TBH-SLN have been prepared using Solvent evaporation method, Further confirmed by Box behnken design and evaluated for particle size, zeta potential, Fourier transform infrared analysis (FTIR), Scanning electron microscopy (SEM), differential scanning calorimetry (DSC), Atomic force microscopy (AFM) and X-ray powder diffraction(XRD) studies. These prepared nanoparticles are into Hyaluronic acid derivative gel (Na-Hy) and the final formulation was obtained. This Formulation is evaluated for pH, viscosity, Gelation time and temperature, Spreadibility, In bioadhesion, In vitro drug release, In vitro drug release, in vitro anti-fungal activity, In vivo irritation studies and in vivo anti-fungal activity. The results of compatibility studies between the drug and polymers revealed that both are compatible with each other without any unwanted interactions. Evaluation of TBH-SLN revealed that the nanoparticles formed have a particle size, zeta potential and EE of 264.9 nm, -27.7 mV and 70.09% respectively. The DSC and XRD studies of nanoparticles confirmed that conversion of crystalline to amorphous form of TBH. Further Morphology was also confirmed using AFM. Characterization of TBH-SLN loaded NA-Hy gel shows a optimized gel formulation with pH almost 4.2 matching vaginal pH, temperature of 320^{0}C in compatible with the standard range and with an extended and prolonged release for about 42 hrs over In vitro drug release studies. It also showed noticeable decrease in the Colony forming units (CFU) of many Fungal strains esp. Candida albicans making it a promising gel formulation, further confirmed with In vivo studies over albino wistar rats. The present study tried to have a prolonged duration of action at the site of application which ensures longer adherence of drug. The prepared novel gel formulation satisfied the properties of an ideal gel in terms of gelation time, temperature, swelling, viscosity, biodegradation, biocompatibility, controlled release, anti-fungal properties which are crucial for prolonged drug release. Hence, the present study suggests that the synergistic combination of TBH loaded as SLN.

Keywords: *Terbinafine, Solid lipid nanoparticles, Hyaluronic acid derivative, sodium hyaluronate, Gel formulation, Fungal infections, Prolonged drug release.*

Corresponding author:

Thurupu Sowmya,

Shri Venkateshwara University,
Gajraula, Amroha, Uttar Pradesh, India
Mobile- +91 9052451820
Email-sowmya092@gmail.com

QR CODE

Please cite this article in press ***Thurupu Sowmya et al., Formulation And Evalution Of Terbinafine Anti-Funga Nano Gel Topical Drug Delivery System****, Indo Am. J. P. Sci, 2025; 12(12).*

INTRODUCTION

Immuno compromised people who have fungal infections often have serious health issues with a high morbidity and death rate due to the gradual emergence of several fungal species. It is strongly linked to individuals who have diseases related to hematologic, allergenic, chronic leukopenia, and autologous transplants. Fungal infections often affect the whole body's system and may have severe consequences for the cellular system.[1] Subcutaneous mycosis is a persistent fungal infection that specifically affects the dermis and subcutaneous tissue, hence earning its name.[2] Sporotrichosis is a significant tropical illness caused by the gradual development of the fungus Sporothrix schenckii.[3]A medication need to be highly effective in preventing the spread of any fungal disease in the future while also posing no risk of severe illness. The most clear and soothing option for patients is the sole method to treat the increasing occurrence of fungal infection. Many drugs are often used as tropical medications to treat fungal infections on the skin and under the skin. Pharmaceuticals are accessible in many formulations such as creams, lotions, gels, and so on.A significant problem about patient compliance is the presence of bioavailability barriers or the limited availability of the medicine at the intended therapeutic location. Thus, considering the therapeutic importance of topical antifungal medication, the rate at which the drug is absorbed should be regulated by the formulation type in order to provide adequate therapeutic efficacy and a prolonged pharmacological impact.[4]

Solid lipid nanoparticles (SLNs) are a cutting-edge and contemporary pharmaceutical technology used for delivering new drugs (NDDS). When SLN was first identified in 1991, it represented common colloidal carriers like polymeric and micro, liposome emulsions, and nanoparticles. The current SLN approach is linked to improved drug delivery with low degradation and excellent physical stability, as well as enhanced drug permeation capacity.[6,7]Nanoparticles ranging in size from 10 to 1000 nm have potential benefits in augmenting the bioavailability of drugs. The formulation of Self-Emulsifying Drug Delivery System (SLN) is a significant factor in the current age of colloidal drug carrier systems, as it produces an alternative particle in the area of Novel Drug Delivery Systems (NDDS).[8]

Terbinafine is a modern and broad-spectrum antifungal drug that has been authorized by the FDA in the United States. Terbinafine is not suitable for topical application due to its bioavailability barrier. [9] In cases of fungal infection, it is necessary to ensure that drugs can effectively penetrate the skin and underlying tissues, allowing for large quantities of the medicine to be present at the place where it is needed for treatment.However, several Terbinafine topical medicines are available on the market that exhibit low skin permeability and shorter skin retention, resulting in improved patient compliance. Forty-nine Currently, nano formulations have seen significant development in the pharmaceutical area because to their high drug load capacity, low use of excipients, stable drug stability, reduced harmfulness, and ease of scale-up and processing. SLNs possess remarkably diverse characteristics that make them advantageous for achieving high permeability in the topical administration of medicines and enhancing prolonged retention at the site of infection. [10]

In addition to these methods, TLC bioautography involves the direct coupling of active substances on a TLC plate with enzyme reactions that can be seen. This technique may be used to directly screen for activity or activity-oriented elements. The use of TLC-based bioautography with enzymatic reactions allows us to begin developing a screening method for active chemicals based on their activity.[11]

Therefore, the current work aims to optimize and evaluate a topical gel formulation incorporating solid lipid nanoparticles (SLN) loaded with Terbinafine. In addition, the optimized formulation that has been created is assessed for its anti-fungal effectiveness against C. albicans using the TLC-Bioautography test.

Advantages

Avoid first pass metabolism

Elite any incompatibility with GI.

Highly selective towards particular site.

Improve patient adherence.

Self-medication capability.

Providing drug use with short biological half-life and small therapeutic window. Capability to stop medication easily when required.[12]

Disadvantages

Irritation of skin or dermatitis may occur to medication or excipients.

Small permeability between skins of respective medications.

Medicines of large particle size cannot be readily absorbed by skin.

Risk with allergic reactions.

It can only be used for drugs which require very limited concentrations of plasma for action. [13]

Skin

The skin is the largest organ of the human body. It considers different biological and chemical substances as obstacles to the opposite process of

Transdermal (TD) absorption. The skin plays a crucial role in regulating medication delivery by controlling the penetration and absorption of drugs via the dermis.

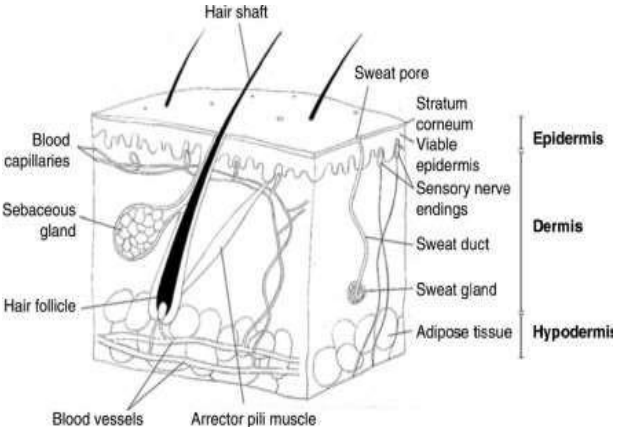
Role of skin

Provides defense against physical, chemical, and microbial assaults.

Provides protection against the penetration of ultraviolet radiation.

It controls the regulation of blood pressure (BP).[14]

Structure of Human skin



Structure of human skin.[15]

Skin is divided by three layers like as[16]

Epidermis

Dermis

Hypodermis

Epidermis

The epidermis is the outermost layer of the skin, characterized by its relatively thin and tough composition. A layer of keratinocytes makes up the epidermis. They originated from cells found in the basal layer of the inner epidermis. Fresh keratinocytes migrate seamlessly over the outer layer of the epidermis. The thickness measures 100 micrometers. Stem cells are the primary cause of skin permeability and diffusion resistance. Five epidermal layers:

Stratumbasale

Spinosum

Granulosum

Lucidum

Corneum[17]

Dermis

The dermis is often resilient; however it contains a significant number of blood vessels, lymphatic vessels, and nerve endings. The vital circulation is bound by a vast network of dermal pores, which have a complex system of horizontal branching in the dermis. This branching connects arterioles and venules to create plexuses, allowing for the passage of blood from capillaries to hair and sweat glands. Dermal capillaries facilitate the extraction of antigens from transparent, fluid substances.[18]

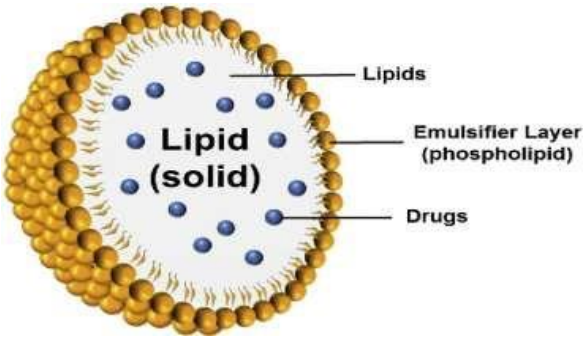
Hypodermis

The hypodermis tissue provides support to the higher layer, such as the dermis and epidermis. It should be used as a repository for fat. The hypodermis layer serves to regulate temperature, provide nutrients, and offer physical protection. It transports blood vessels and terminates nerves to the skin, and maybe includes organs that perceive pressure. In the case of topical drug delivery,

only diffusion via SC is necessary and the medication must be maintained in skin layers; in the case of Transdermal drug delivery (TDDS), pharmaceuticals must pass through all these layers and reach vital flows. [19]

Solid Lipid Nanoparticles

Solid lipid nanoparticles (SLNs) are a kind of colloidal drug carrier system composed of solid lipids dispersed in an aqueous solution containing surface-active agents. These nanoparticles typically have a particle size ranging from 50 to 1000 nm.[20] Solid Lipid Nanoparticles (SLN) provide superior alternatives to polymeric systems, colloidal systems, and other Novel Drug Delivery Systems (NDDS) due to its favorable characteristics such as excellent tolerability, strong physical stability, biodegradability, high bioavailability, protection of active pharmaceuticals from degradation, simplicity of synthesis, and low toxicity.[21, 22,23] Furthermore, the manufacturing method may be altered to facilitate the release of the desired medication, prevent the deterioration of the drug, and eliminate the use of organic solvents. This broad flexibility might be crucial for the promotion of new goods. The high quality of SLN makes it a compelling carrier system for improved medication delivery.[24]



Structure of solid lipid nanoparticles

Structure of solidlipidnanoparticles.[25]

Advantages of SLNs

Improve stability of pharmaceuticals.

Excellent biocompatibility.

Avoidance of organicsolvent.[74]

Long-term stability.

Improve bioavailability of trapped bioactive compounds.

Easy to scale up and sterilize.

Control and targetdrugrelease.[27]

Low toxicity.

Biodegradable and biocompatible

Disadvantages of SLNs

Poor drugloadingcapacity.[28]

Less compatibility for hydrophilic drugs

Unsure gelation tendency.

Gels

Gels are characterized as a "semisolid system in which the liquid phase is restricted inside a polymer matrix, resulting in a significant level of physical and chemical interconnections."

Properties of gels

Gellingagentsareusedforpharmaceuticalformulations/cosmeticformulations.

It should be safe, inert and does not react with different ingredients in formulation

Inclusion of gelling agent in formed will produce logical solid-like consistency during storage, which can be simply broken down when individual to shear forces are created by pressing container, vortexing bottle or during topical uses.

Preventing microbial strike should be possible with anti-microbial.

Not to be messy.

As with ophthalmic gel, it should be sterile.

Gels classified on the basis of nature of solvent uses, colloidal phases, rheological properties and physical nature.

From on colloidal phases: They are classified into

Inorganic(two phase system)

Organic(single phase system)

From on nature of solvent

Hydrogels(water based):

Organic Gels (with an on-aqueous solvent)

Aerogels

From on rheological properties

They are classified into,

Plastic gels

Pseudo plastic gels

Thixotropic gels.

From on physical nature

Elastic gels.

Rigid gels.[29]

Evaluation of gel.[30]

Appearance

pH

Viscosity

Spreadability

Drug content

Aim and objective

The main aimof this study focuses on developing and assessing a Terbinafine anti-fungal nano gel for a topical medication delivery system utilizing the Quality by Design (QbD) methodology.

Materials and Methods

Reagents and chemicals

Terbinafine received as gift sample from SMS Pharmaceuticals, INDIA, Carbopol 934 purchased from sigma Aldrich, Italy, Stearic acid purchased from Fisher Scientific India Pvt.Ltd, Ethanol purchased from Merck, India, n-octanol purchased from SD Fine-chem. Ltd, Mumbai, Methanol purchased from Fisher Scientific India Pvt.Ltd, Poloxomer188 purchased from Central Drug House (P)Ltd, Sodium Hydroxide, Potassium Di hydrogen orthophosphate, and Disodium hydrogen orthophosphate purchased from Thomas Baker, New Delhi.

Pre formulation studies

Determination of the absorption maximum of Terbinafine in ethanol

The absorption maximum of Terbinafine determined as per standard protocol with some modification. In brief, stock solution of Terbinafinedevelopedatconcentrationof1mg/ml in methanol. Further, it followed by serial dilution to get concentration of Terbinafine as 2, 4, 6, 8, 10µg/ml, and then it proceeded to UV spectrophotometric analysis at λmax of 299nm. Qualification is taken in triplicate and obtained datawereanalyzedstatistically.[31]

Determination of Aqueous Solubility

The determination of aqueous solubility of Terbinafine estimated through Saturation shake - flask method. An optimum amount of Terbinafine dissolved in distilled water and acetate buffer pH 5.5 then followed by vortex and centrifugation at 50 rpm at 37 °C for 48 hrs, resulting solution filtered and analyzed spectrophotometrically at 299 nm. Qualification is taken in triplicate.[32]

Determination of lipophilicity

Lipophilicity of Terbinafine determined through traditional shake flask method as described in protocol with some modification. In brief, an optimum uniform amount of Terbinafine poured inside three different volumetric flasks and then measured quantity of lipids such as Stearic acid, prectrol, dynasan114 placed to each flask simultaneously. Resulting heterogeneous mixture proceeded to vortex and then centrifugation at 50 rpm at 37 °C for 48hrs. Supernatant are separate and filtered using syringe filter of 0.22µm. Filtrate then analyzed spectrophotometrically at 299 nm.[32]

The further partition coefficient of Terbinafine determined using n-octanol and water partition system. Measured amount of Terbinafine placed inside conical flask containing measured volumes of an n-octanol and aqueous buffer solution. flask shaken with uniform time interval for 48h to attain equilibrium and then resulting mixture placed to separating flask with final shaking and kept remains undisturbed to be separated inside two layers. Targeted measurement proceeded to be analyzed spectrophotometrically at 299 nm. Resulting values of both phases were determined in form of $\log_{10}$ P of ratio calculated. All qualification is taken in triplicate. [33]

Fourier Transform Infrared (FTIR) Spectroscopy

The spectral analysis is for Terbinafine and Stearic acid performed by Win-IR, Bio-Rad FTS spectrophotometer. Individual sample assorted with potassium bromide and later proceeds for spectroscopic observation under range of 4000 to 400 cm^{-1}.[34]

Preparations of Terbinafine loaded solid lipid nanoparticles (SLN)

The SLN prepared using referenced protocol of solvent diffusion method with some modification. Briefly, known amount of Terbinafine and Stearic acid placed inside 5 ml of ethanol and heated at 60±3.0°Con water bath. Obtained solution placed inside 5ml of aqueous poloxamer 188 solutions at 4 – 8°C under magnetic stirring at 2000 rpm with help of syringe. SLN formed instantly and recovered by centrifugation at 2000 rpm for 30 min at 4°C.Obtained heterogeneous mixture further proceeded to high-pressure homogenization via APV2000 homogenizer at 1200 bars. Obtained mixture placed to be stable at room temperature, which turns to clear nanocrystals by recrystallization of dispersed lipid.[35]

Evaluation of SLN

Evaluation of entrapment efficiency

The EE of SLN loaded with Terbinafine estimated through described method with some modification. In brief, prepared SLN dried at room temperature then 5 mg of dried SLN were dissolved in10 ml HPLC grade ethanol and further proceeds by filtration through syringe filter of 0.22 µm capacity. Concentration of Terbinafine determined spectrophotometrically at 299 nm. [52] The qualification taken in triplicate and based on percentage entrapment, best one selected for further evaluation. Entrapment efficiency has been determined according to following equation:

EE%=W(Addeddrug)−W(freedrug)/W(Addeddrug) x 100

Where, $W_{(addeddrug)}$ is quantity of drug added during preparation of SLN, $W_{(freedrug)}$ is quantity of free drug measured in supernatant after centrifugation.

Physico chemical property

Physicochemical Properties of SLN dispersions were characterized as color, odor, pH, and solubility of SLNF6 in aqueous medium.[35,36]

Particle size and zeta potential

The average particle size and zeta potential were determined as per described protocol with some modification. Analysis performed at room temperature by zeta potential/ particle size analyzer. SLN F6 diluted with phosphate-buffered saline and pH of solution stabilized at 7.4 and then sample proceeded for analysis.[37]

Optical microscopy

Optical microscopic analysis of optimized formulation SLN F6 analyzed with help of digital light optical microscope equipped with fluorescent lamp (LabomedLX-400) at 100x magnification. It aimed to determine whether Terbinafine SLN is effectively localized with homogenous and uniform texture with in SLN dispersion.[38]

FTIR spectra of SLNF6

The spectral is analysis for SLN F6 performed by Win-IR, Bio-Rad FTS spectrophotometer. Individual sample assorted with potassium bromide and later proceeds for spectroscopicalobservationunderrangeof4000 to 400 cm^{-1}. [39]

Preparation of gel

The gel developed as per referenced protocol with slight modification. Briefly, Carbopol 934Pplaced in defined quantity of distilled water while constant stirring at 600rpm and followed by adding of methyl paraben sodium (0.02% w/v) and propyl paraben sodium (0.1% w/v) and remained undisturbed with continuous stirring for 30 min. Prepared gel base set aside for 24hrs.Next,SLNF6disseminatedwithmeasuredquantityofpropyleneglycol(5%w/w)and1%ethanol(20%w/w)andfaraheaditaddedtocarbopolgelbaseswithcontinuousshakingat 1000 rpm and followed by churning for 30min. Tri-ethanol amine (TEA) subjected to final stage to maintain pH (5.5 - 6.5) for drug stabilization and stirred thoroughly to obtain cleargel.[40]

The same procedure applied to get four formulations having varying amount of Carbopol and aim is associated to prepare different forms of gel is to obtain best homogeneous and uniform texture with stable physicochemical reliability in respect of % release of leading moiety. Different formulations of SLN gel are enlisted as in Table;

Preparation of different formulations of solid lipid nanoparticles containing gel

Formulation code	Carbopol934%(w/v)
G1	0.5
G2	1
G3	1.5
G4	2

Characterization of gel

Determination of pH

ThepHofgelevaluatedasperstandardprotocolwithhelpofdigitalpHmeter.Glasselectrode of pH meter immersed in optimized SLN gel formulation and revolved to determine pH of gel. [40]

Determination of viscosity

The viscosity of gel evaluated as per standard protocol with some modification as follows. In brief, obtained SLN gel evaluated based on physical appearance and then viscosity of SLN gel evaluated through Brookfield Viscometer**.** [41]

6.1.1 Determination of the entrapment efficiency

The % EE of different prepared batches of gel estimated by quantitating free mass drug in diffused phase of gel solution after centrifugation. In brief, 1g of gel diffused with ethanol and vortexes for 5 minutes to ensure proper extraction of drugs in ethanol. Then, obtained mixture proceeded for centrifugation at 15000 rpm for 60 minutes at 4 °C temperature. Supernatant collected from centrifuged mixture and allowed to an alyze for quantitative analysis spectrophotometrically at 299 nm**.**[42] The EE percentage calculated from equationas follows;

$$EE\%=W_{(Added\ drug)}-W_{(free\ drug)}/W_{(Added\ drug)}x\ 100$$

Where, $W_{(initialdrug)}$ is mass of drug added initially, $W_{(freedrug)}$ is mass of free drug detected in supernatant after centrifugation.

Spreadability

The Spreadability of gel determined as per described protocol with some modification. In brief, 500 mg of optimized formulation put on acrylic plate at middle center and second plate concentrically situated above it. Width of circle in which gel spread measured as primary width. An approx 500 g weight applied on above plate for few mins. Spreadability of gel estimated as per rises in diameter due to dissemination of gels and obtained diameter of disseminated gel noted.[43]

***In-vitro* drug release and kinetics study**

The drug release and kinetics profiling of optimized formulation (SLN G3) gel were evaluated by *in-vitro* drug release profiling methods using dialysis bag technique.[36] 1g of gel sample accurately weighed and placed to cellulose dialysis membrane. Membrane tied with thread and placed to flask containing 50 ml ethanol and phosphate buffer solution. Container placed to magnetic stirrer at 37 °C with constant stirring at 50 rpm. Thereafter, 1 ml of sample withdrawn at regular intervals of 0.25, 0.5, 1, 2, 3, 4, 6, 8, 12, and 24, and withdrawn amount replenished with dissolution media at same time withdrawn. Released mass of SLN entrappedTerbinafinequantitatedspectrophotometrically at 299nminrespectofblank.Eachmeasurement is taken in triplicate. *In-vitro* drug release profiles of prepared Terbinafine loaded SLN gel formulation evaluated statistically by various kinetic models named as zero-order, first-order, and Higuchi and Korsemeyer–Peppas model. Kinetics models were determined statistically to enlighten mechanism of drug release profiling. The high regression coefficient value considered to be much effective for initialization and acceptance of kinetics orders.

Fourier transform infrared spectroscopy

The spectral analysis is SLN G3 performed by Win-IR, Bio-Rad FTS spectrophotometer. Individual sample assorted with potassium bromide and later proceeds for spectroscopical observation under range of 4000 to 400 cm^{-1}. [44]

Scanning Electron Microscopy

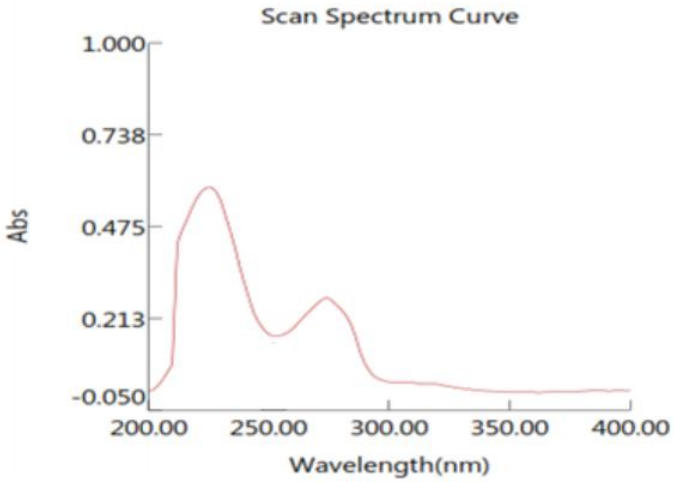
The morphological analysis is SLN G3 examined by SEM using standard protocol with some modification**.** Little sample of SLN gel placed on glass stub and vacuum dried. After that, stub having sample sited to SEM chamber coated with gold-palladium and then sample observed microscopically at an accelerating voltage of 10 kV.[40]

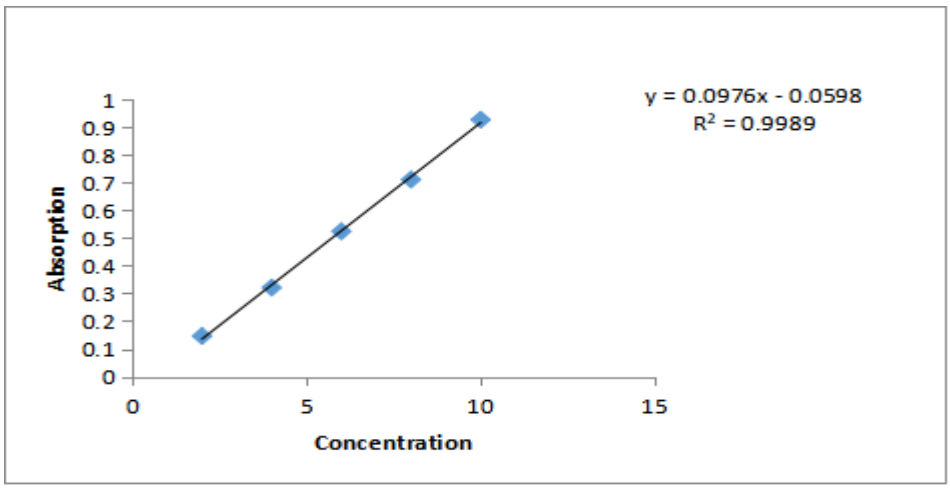
RESULTS AND DISCUSSION:

Pre formulation study of drug

Determination of the absorption maximum of Terbinafine in ethanol

The potential drug absorption calculated as per standard protocol through absorption maximum of Terbinafine at 283 nm λmax against concentration 2-10 µg/ml. regression equation and coefficient were found to be 0.0664x-0.0478 and 0.998 respectively. Associated aim to determine Terbinafine absorption maxima and method validation is for qualitative and quantitative analysis.[48]





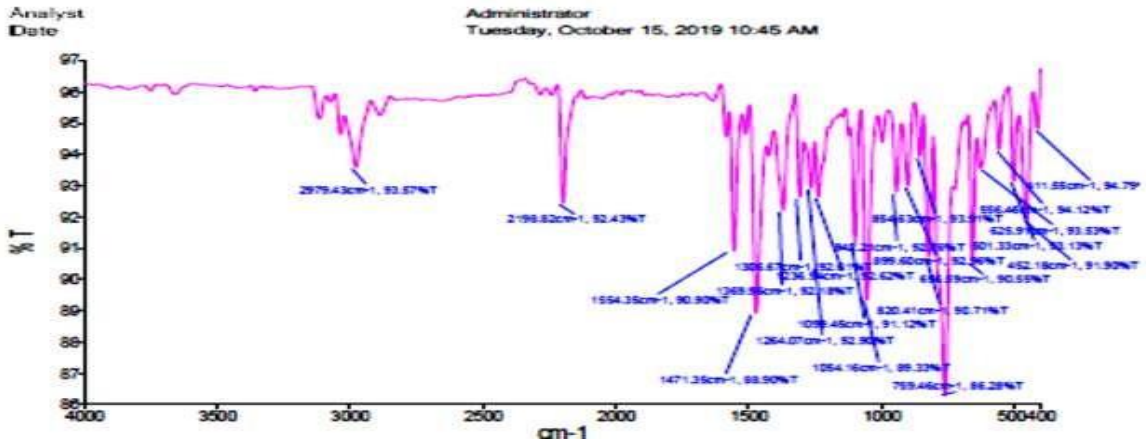
Absorption maxima of Terbinafine and regression coefficient against the different concentration of Terbinafine (µg/ml)

Physicochemical studies of Terbinafine were conducted to evaluate physicochemical properties of drug. Studies conducted to evaluate Terbinafine hydrophilic and lipophilic compatibility. Result shows that Terbinafine having poor solubility potential with water which found to be 0.00585 ± 0.293 mg/ml, and solubility of Terbinafine in stearic acid, prectrol, dynasan 114 obtained 23.754±0.47, 18.314± 0.85, 22.875±0.32 mg/ml. Besides, non-aqueous solubility obtained 17.984 ± 0.52 mg/ml for Terbinafine in n-octanol. $\log_{10}$Pvalue of Terbinafine in stearic acid, prectrol, dynasan 114, and n-octanol obtained as 3.98,3.30,3.87, and 3.65 simultaneously.

The FTIR analysis performed of Terbinafine and Stearic acid for better compatibility analysis of leading moiety before and after formulation. FTIR spectra of Terbinafine is shown in Figure 7; Table 6. principal IR absorption peaks of Terbinafine at 2979.43 cm^{-1}(C-H stretch), 2198.82 cm^{-1} (C≡N stretch), 1554.35 cm^{-1} (C-H aromatics stretch), 1471.35cm^{-1} (C=C-C aromatic ring stretch), 820.41 cm^{-1} (para C–H distribution) and 759.46cm^{-1}(C-Cl stretch) were all detected in spectra of Terbinafine. These detected principal peaks confirmed purity and authenticity of Terbinafine as similar to referenced report.

FTIR interpretation of Terbinafine

Characteristics Peaks	Reported(cm^{-1})	Observed(cm^{-1})
C-H stretch	2850 -3000	2979.43
C≡N Stretch	2100 -2400	2198.82
C=C aromatic stretch	1450 -1650	1554.35
C=C-C Aromatic ring stretch	1510 -1450	1471.35
Para C-H distribution	860 -800	820.41
C-Cl stretch	600 -800	759.46

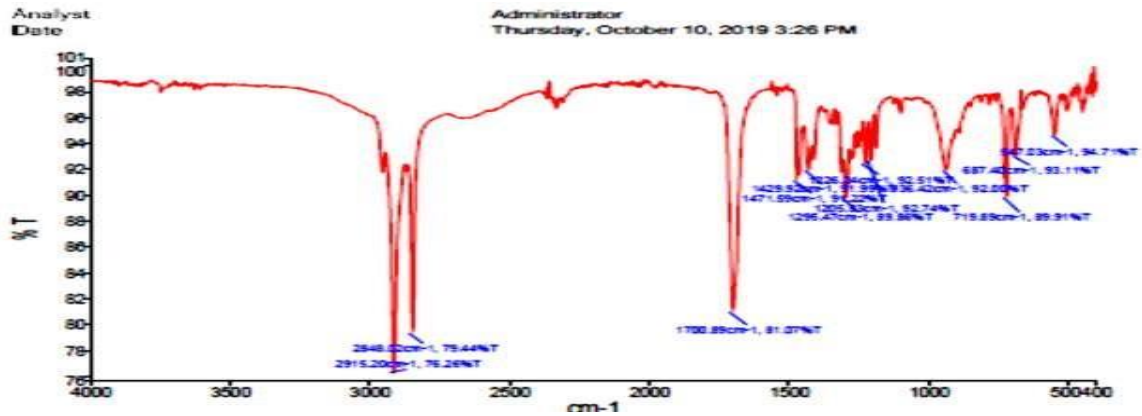


FTIR spectrum of Terbinafine

The FTIR spectra are interpretation of Stearic acid shown in Figure 9 and Table7. Principal IR absorption peaks of Stearic acid at 2915.20cm^{-1}(C–H stretch alkanes), 2848.02cm^{-1}(C-Hstretchaldehyde),1700.89cm^{-1}(C=Ostretchsaturated),1471.59cm^{-1}(C-Cstretch),1295.47cm^{-1}(C-Ostretch,aromaticaster),936.42cm^{-1}(O-Hbend),719.89cm^{-1}(C=C bend) and 547.03 cm^{-1} (C-I stretch) were all detected in spectra of Stearic acid. These detected principal peaks confirmed purity and authenticity of Stearic acid as similar to reported data**.**

FTIR interpretation Stearic acid

Characteristics Peaks	**Reported(cm^{-1})**	**Observed(cm^{-1})**
C–H stretch alkanes	2850 -3000	2915.20
C-H stretch aldehyde	2800 -2860	2848.02
C=O stretch saturated	1700 – 1730	1700.89
C-C stretch	1400 -1500	1471.59
C-O stretch, aromatic aster	1250 -1310	1295.47
O-H bend	910 – 950	936.42
C=C bend	665 -730	719.89
C-I stretch	500 -600	547.03



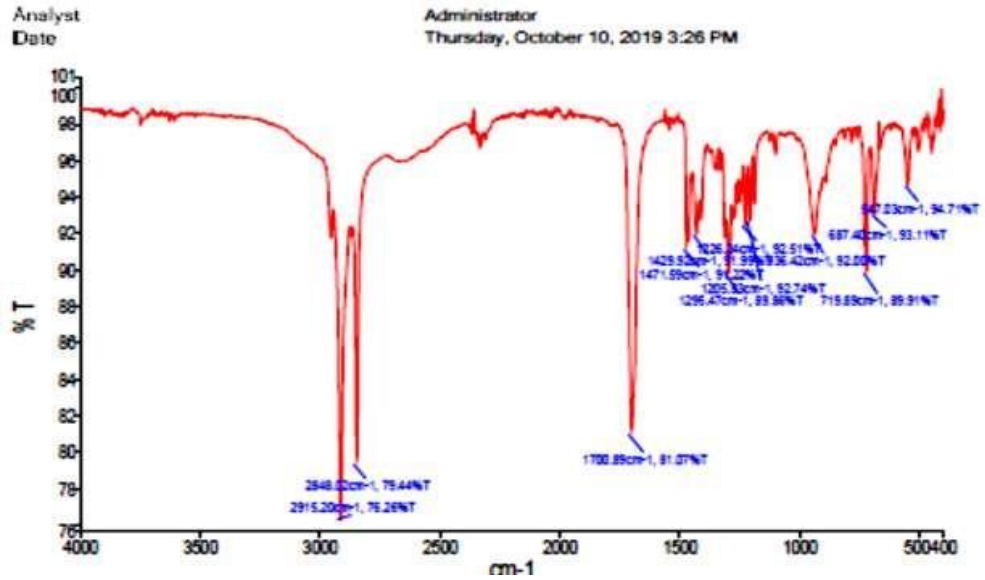
FTIR spectrum of Stearic acid

Development of the method for Terbinafine loaded solid lipid nanoparticles (SLN)

The method deals with various modified nano-precipitation methods for optimization of SLN in respect of EE of Terbinafine at both treatment segments, i.e. nano-precipitation and cooling sonication probe. Temperature controlled by 4°C and 25°C in both segments. Instant adding of organic phase to aqueous phase conserved at 4°C which gives immediate precipitation due to hyphenation with anti-solvent. Temperature controlled at initial phase of nano-precipitation which helped to achieve homogeneity. High-pressure homogenization supports to get uniform homogeneity by decreasing larger crystals size and bead milling aggregation.[87]Further, in optimization of SLN, method archived step by step with alternate changes in concentration of Stearic acid and poloxomer 188 (w/v) ranging from 0.5-2%.

All prepared groups of SLN were

Characteristics Peaks	Reported(cm^{-1})	Observed(cm^{-1})
C–H stretch alkanes	2850 -3000	2915.20
C-H stretch aldehyde	2800 -2860	2848.02
C=O stretch saturated	1700 – 1730	1700.89
C-C stretch	1400 -1500	1471.59
C-O stretch, aromatic aster	1250 -1310	1295.47
O-H bend	910 – 950	936.42
C=C bend	665 -730	719.89
C-I stretch	500 -600	547.03

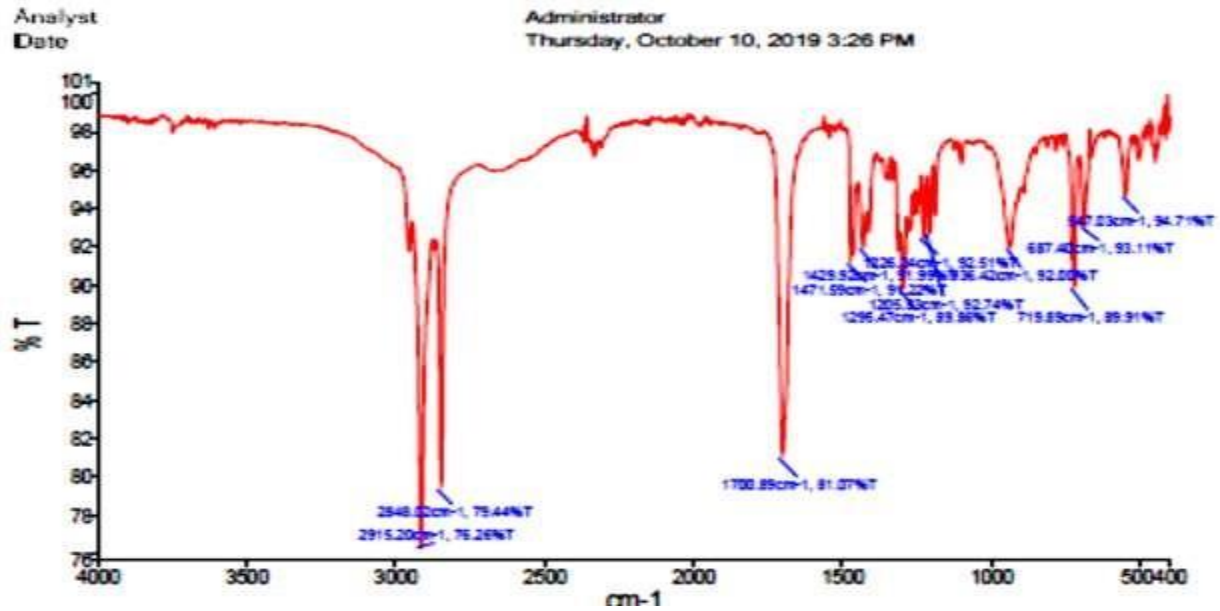


FTIR spectrum of Stearic acid

Development of the method for Terbinafine loaded solid lipid nanoparticles (SLN)

The method deals with various modified nano-precipitation methods for optimization of SLN in respect of EE of Terbinafine at both treatment segments, i.e. nano-precipitation and cooling sonication probe. Temperature controlled by 4°C and 25°C in both segments. Instant adding of organic phase to aqueous phase conserved at 4°C which gives immediate precipitation due to hyphenation with anti-solvent. Temperature controlled at initial phase of nano-precipitation which helped to achieve homogeneity. High-pressure homogenization supports to get uniform homogeneity by decreasing larger crystals size and bead milling aggregation.[87]Further, in optimization of SLN, method archived step by step with alternate changes in concentration of Stearic acid and poloxomer 188 (w/v) ranging from 0.5-2%. All prepared groups of SLN were

Characteristics Peaks	Reported(cm^{-1})	Observed(cm^{-1})
C–H stretch alkanes	2850 -3000	2915.20
C-H stretch aldehyde	2800 -2860	2848.02
C=O stretch saturated	1700 – 1730	1700.89
C-C stretch	1400 -1500	1471.59
C-O stretch, aromatic aster	1250 -1310	1295.47
O-H bend	910 – 950	936.42
C=C bend	665 -730	719.89
C-I stretch	500 -600	547.03



FTIR spectrum of Stearic acid

Development of the method for Terbinafine loaded solid lipid nanoparticles (SLN)

The method deals with various modified nano-precipitation methods for optimization of SLN in respect of EE of Terbinafine at both treatment segments, i.e. nano-precipitation and cooling sonication probe. Temperature controlled by 4°C and 25°C in both segments. Instant adding of organic phase to aqueous phase conserved at 4°C which gives immediate precipitation due to hyphenation with anti-solvent. Temperature controlled at initial phase of nano-precipitation which helped to achieve homogeneity. High-pressure homogenization supports to get uniform homogeneity by decreasing larger crystals size and bead milling aggregation.[87]Further, in optimization of SLN, method archived step by step with alternate changes in concentration of Stearic acid and poloxomer 188 (w/v) ranging from 0.5-2%. All prepared groups of SLN were coded successfully and proceed to quantitate percent entrapment of active moiety spectrophotometrically at 299nm. Obtained data were evaluated statistically. SLN which deal with high entrapment of Terbinafine chosen as optimized SLN and proceed for further evaluation.

Preparation of different Terbinafine solid lipid nanoparticles

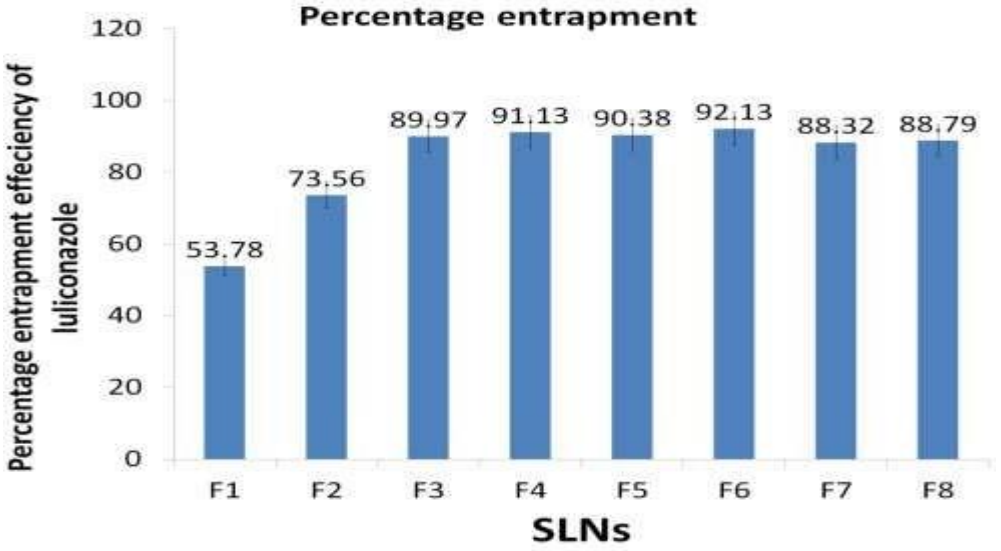
		Different concentration of leading reagents for the formation of SLN	
SLN code	**Terbinafine%(w/v)**	**Stearic acid %(w/v)**	**Poloxomer188%(w/v)**
F1	1	0.5	1
F2	1	0.7	1
F3	1	1	1
F4	1	2	1
F5	1	1	0.5
F6	1	1	0.7
F7	1	1	1.5
F8	1	1	2

Evaluation of SLN

Evaluation of entrapment efficiency of SLN

Initially, in pre-formulation studies, Terbinafine characterized physico chemically and spectroscopically. After successful formation of different batch s of nanoparticles, percentage EE of Terbinafine determined. Percentage of EE evaluated spectrophotometrically at 299 nm. Thereafter results reveal that SLN F6 and SLN F1 have highest and lowest % EE of Terbinafine loaded SLN by 92.13% ± 0.975 and 53.78%±1.052 w/w respectively. Similarly, study cited by Ige et al. reported maximum %EE by 90–95% w/w.[39]Therefore, based on percent drug entrapment, SLNF6 selected as an optimized SLN and proceed for further evaluation includes physicochemical properties and gel formation.

Percentage entrapment efficiency of Terbinafine in SLN



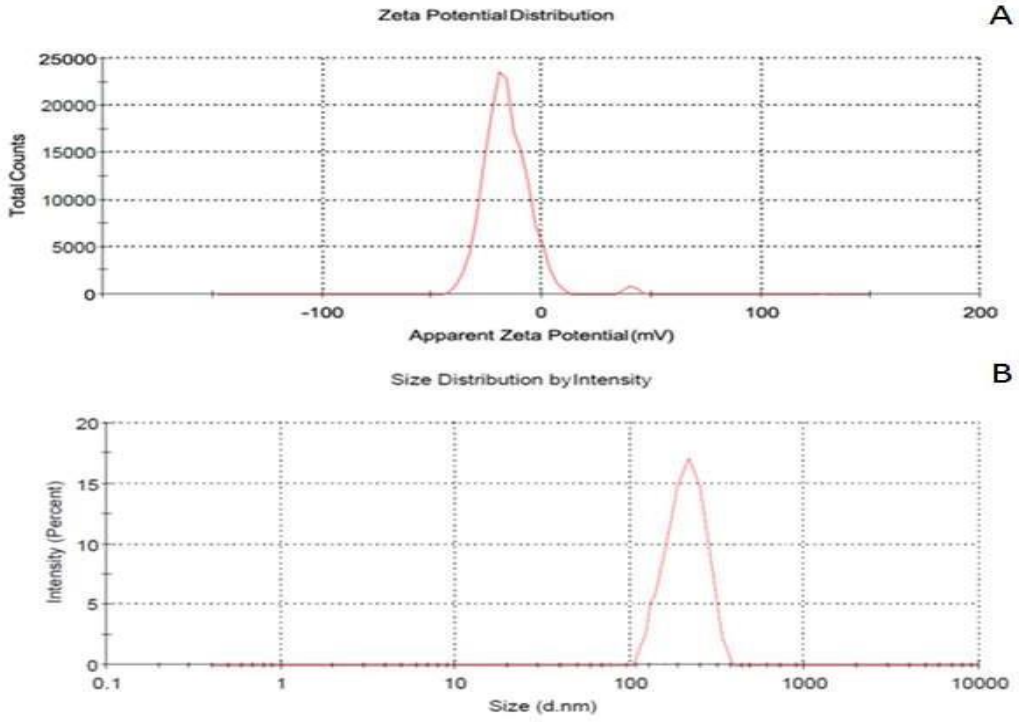
Physicochemical property

The SLN F6 evaluated based on their physicochemical characteristics such as color, odor, pH stability, and aqueous solubility. Physicochemical results reveal that SLN has white transparent color with homogeneous and uniform texture, aromatic odor, better stability at pH, and water solubility found 0.01819±0.035mg/ml,i.e. much enough than Terbinafine solubility.

Zeta potential and particle size and size distribution identification

The particle size analysis and zeta potential measurement of Terbinafine SLN were identified successfully using nano ZS90 zeta sizer system. Zeta potential is one of important parameters used to forecast physical stability of nanoparticles. Stability of nanoparticles system depends on high zeta potential value which points toward better stability of nano system since it could deliver deterring force between nanoparticles.[81] Asshown in Fig. 11, SLN shows quite high value of zeta potential by ~18.8 mV and states to high stability of nano system. In particle size analysis, SLN unveiled with mean particle diameter by ~344.3 nm, uni modal size distribution, poly dispersity index (PDI) by 0.168, intercept value 0.98 and 92% peak intensity. PDI is parameter that represents dissemination factor with low aggregation of nanoparticles when PDI value would be <0.5.

Zeta potential, particle size and size distribution of Terbinafine SLN F6



Optical microscopy

Optical microscopy of optimized preparation i.e. SLN F6 defined with help of digital lightoptical microscope at 100x magnification and observation shows that Terbinafine SLN iseffectively localized with homogenous and uniform texture within SLN dispersion.Itstates that only particles with mean diameter higher than 2.5 µm which were visualizedclearly against microscopy resolution power. Moreover, SLN preparation has even no self-assembledstructuresobserved.Micellarstructureswerenotobservedduringobservationof optical microscopy. Optical microscopy images of Terbinafine loaded SLN F6 shown in figure12

Optical microscopy images of Terbinafine loaded SLN F6

Drug-excipients comparability study by FTIR

FTIR analysis of SLN F6 performed to determine possible interaction between drug and drug additives. spectral data reveal principal absorption peaks of Terbinafine at 2955.75cm^{-1} for C-H stretching, 2523 & 2647 cm^{-1} for S-H stretching, 2201.52 cm^{-1} for C≡Nstretching,1556.90cm^{-1}forC=N stretching,1471.88$cm^{-1}$for C=C aromatic ring stretching and 720.33and 1101.29 cm^{-1} for C-Cl stretching. Whereas, principal absorption peaks of stearic acid were found at 2914.97cm^{-1}& 2848.05 cm^{-1} in high-frequency region attributed to -CH_2- band asymmetric and symmetric stretching vibrations, whereas and1698.03$cm^{-1}$for-COOH stretching is attributed in low-frequency region. Spectral analysis of optimized SLN confirmed that there are no more changes in Terbinafine after successful formation of SLN. Spectral data strongly supports referenced values as reported.

FTIR interpretation of SLN F6

Characteristics Peaks	Reported(cm^{-1})	Observed(cm^{-1})
C-H stretch	2850 -3000	2955.75 2914.97 2848.05
C≡N Stretch	2100 -2400	2201.52
C=C alkene stretch	1650 -2000	1698.03
C=C Aromatic stretch	1450 -1650	1463.82
C-Cl stretch	550 -850	609.29

Optimization and evaluation of SLN gel

The topical gel containing SLN loaded with Terbinafine prepared successfully by stirring method using carbopol 934 as gelling agent. Method of preparation of different SLN gel found unpretentious and robust. Initially, all four different preparations of SLN gel coded as G1, G2, G3, and G4, were evaluated to quantitate percent entrapment of Terbinafine spectrophotometrically at 299 nm. Resulting data shows that SLN G3 with 1.5% Carbopol w/w showing highest percentage of drug entrapment with 91.39% ± 0.187. There after optimized formulation is further evaluated to access physiochemical parameters includes visual appearance, pH, viscosity and Spreadability. Resulting data reveals that viscosity of G3 gel as found to be 369 cP, similar to gel viscosity as reported by Janaetal and pH found to be 6.12±0.255.[41]further in Spreadability evaluation, Spreadability factor of prepared SLN gel found to be 4.5 and it states that prepared gel produces excellent Spreadability as an ideal topical formulation. Spreadability is one of important physical properties of any topical formulation from patient's compliance point of view.

Visual appearance of SLN G3 gel

FTIR spectra of SLN F6

Optimization and evaluation of SLN gel

The topical gel containing SLN loaded with Terbinafine prepared successfully by stirring method using carbopol 934 as gelling agent. Method of preparation of different SLN gel found unpretentious and robust. Initially, all four different preparations of SLN gel coded as G1, G2, G3, and G4, were evaluated to quantitate percent entrapment of Terbinafine spectrophotometrically at 299 nm. Resulting data shows that SLN G3 with 1.5% Carbopol w/w showing highest percentage of drug entrapment with 91.39% ± 0.187. There after optimized formulation is further evaluated to access physiochemical parameters includes visual appearance, pH, viscosity and Spreadability. Resulting data reveals that viscosity of G3 gel as found to be 369 cP, similar to gel viscosity as reported by Janaetal and pH found to be 6.12±0.255.[41]further in Spreadability evaluation, Spreadability factor of prepared SLN gel found to be 4.5 and it states that prepared gel produces excellent Spreadability as an ideal topical formulation. Spreadability is one of important physical properties of any topical formulation from patient's compliance point of view.

***Invitro* drug release and kinetics study**

Statistical models are commonly used to forecast release mechanism and compare release profile. *in-vitro* release profile of drug performed in prepared buffer system using dialysis bag technique for 24 hrs. de solvation percentages of Terbinafine from SLN are increased in proportion of time as illustrated in Figure 15 and Table 10. Pieces of evidence of release profiles show that developed SLN is proficient to release drug in regulated manner. Slow release of leading moiety from most SLN form is based on homogeneous drug entrapment throughout systems.[62] Ekambaram et al. state same concept and claimed that controlled drug desolvation profile can be attained when drug is uniformly distributed in lipid matrix. Poloxamer 407 having immense efficacy against drug release rate from SLN then Cremophor RH40 because of its higher HLB value than cremophor RH 40.[20] Besides, Poloxamer 407 has high exterior Spreadability so that it eases effects of interfacial tension between SLN and dissolution medium. It also reduces accumulation of drug particles and increases drug dissolution rate. Moreover, lipid mass in SLN can control size of nanoparticles and increase drug desolvation strength. Thickness of lipid surrounded nanoparticle increases length of drug disassociation resulting prolonged effect of drug release.

Percentage drug release profile of G3 and control gel

Sr.no.	Time in hours	Percentage drug release of G3	Percentage drug release of control gel
1	0	0	0
2	0.25	7.375±0.153	1.923± 0.011
3	0.5	14.002±0.185	2.052± 0.155
4	1	22.064±0.102	3.042± 0.158
5	2	32.289±0.173	3.182± 0.162
6	3	40.622±0.165	5.094± 0.122
7	4	47.048±0.151	7.815± 0.205
8	6	55.582±0.163	8.706± 0.215
9	8	62.309±0.134	9.387± 0.118
10	12	69.939±0.115	9.035± 0.205
11	24	79.578±0.213	9.773± 0.158

***Invitro* drug release and kinetics study**

Statistical models are commonly used to forecast release mechanism and compare release profile. *in-vitro* release profile of drug performed in prepared buffer system using dialysis bag technique for 24 hrs. de solvation percentages of Terbinafine from SLN are increased in proportion of time as illustrated in Figure 15 and Table 10. Pieces of evidence of release profiles show that developed SLN is proficient to release drug in regulated manner. Slow release of leading moiety from most SLN form is based on homogeneous drug entrapment throughout systems.[62] Ekambaram et al. state same concept and claimed that controlled drug desolvation profile can be attained when drug is uniformly distributed in lipid matrix. Poloxamer 407 having immense efficacy against drug release rate from SLN then Cremophor RH40 because of its higher HLB value than cremophor RH 40.[20] Besides, Poloxamer 407 has high exterior Spreadability so that it eases effects of interfacial tension between SLN and dissolution medium. It also reduces accumulation of drug particles and increases drug dissolution rate. Moreover, lipid mass in SLN can control size of nanoparticles and increase drug desolvation strength. Thickness of lipid surrounded nanoparticle increases length of drug disassociation resulting prolonged effect of drugrelease.

Percentage drug release profile of G3 and control gel

Sr.no.	Time in hours	Percentage drug release of G3	Percentage drug release of control gel
1	0	0	0
2	0.25	7.375±0.153	1.923± 0.011
3	0.5	14.002±0.185	2.052± 0.155
4	1	22.064±0.102	3.042± 0.158
5	2	32.289±0.173	3.182± 0.162
6	3	40.622±0.165	5.094± 0.122
7	4	47.048±0.151	7.815± 0.205
8	6	55.582±0.163	8.706± 0.215
9	8	62.309±0.134	9.387± 0.118
10	12	69.939±0.115	9.035± 0.205
11	24	79.578±0.213	9.773± 0.158

Invitro drug release and kinetics study

Statistical models are commonly used to forecast release mechanism and compare release profile. *in-vitro* release profile of drug performed in prepared buffer system using dialysis bag technique for 24 hrs. de solvation percentages of Terbinafine from SLN are increased in proportion of time as illustrated in Figure 15 and Table 10. Pieces of evidence of release profiles show that developed SLN is proficient to release drug in regulated manner. Slow release of leading moiety from most SLN form is based on homogeneous drug entrapment throughout systems.[62] Ekambaram et al. state same concept and claimed that controlled drug desolvation profile can be attained when drug is uniformly distributed in lipid matrix. Poloxamer 407 having immense efficacy against drug release rate from SLN then Cremophor RH40 because of its higher HLB value than cremophor RH 40.[20] Besides, Poloxamer 407 has high exterior Spreadability so that it eases effects of interfacial tension between SLN and dissolution medium. It also reduces accumulation of drug particles and increases drug dissolution rate. Moreover, lipid mass in SLN can control size of nanoparticles and increase drug desolvation strength. Thickness of lipid surrounded nanoparticle increases length of drug disassociation resulting prolonged effect of drugrelease.

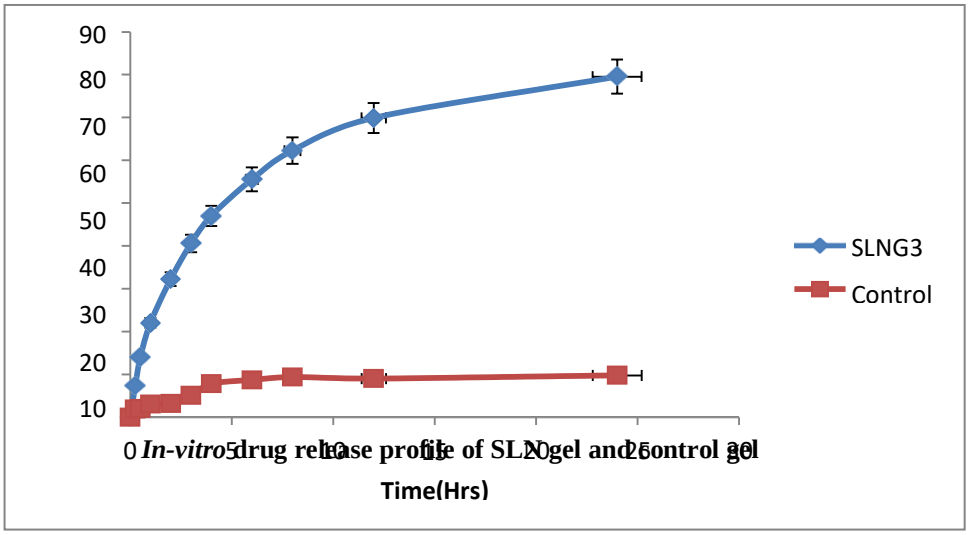
Percentage drug release profile of G3 and control gel

Sr.no.	Time in hours	Percentage drug release of G3	Percentage drug release of control gel
1	0	0	0
2	0.25	7.375±0.153	1.923± 0.011
3	0.5	14.002±0.185	2.052± 0.155
4	1	22.064±0.102	3.042± 0.158
5	2	32.289±0.173	3.182± 0.162
6	3	40.622±0.165	5.094± 0.122
7	4	47.048±0.151	7.815± 0.205
8	6	55.582±0.163	8.706± 0.215
9	8	62.309±0.134	9.387± 0.118
10	12	69.939±0.115	9.035± 0.205
11	24	79.578±0.213	9.773± 0.158

***In-vitro* drug release profile of SLN gel and control gel**

Furthermore, an *in-vitro* drug release profile for optimized formulation applied to various kinetic models (zero-order, first-order, Higuchi, and Krosmayer Peppas model). To state kinetics profiling of drug release, obtained data were analyzed statistically in respect of rate constant and highest correlation. Best-fitted line found in all models except little suitability in zero-order equation. Resulting data described is semination of drug in controlled or regular manner from homogenous matrix systems and it states why drug disseminates at slower rate. Observations concluded that SLNG3 is far efficient as potential topical formulation for sustained drug delivery. This finding is almost similar to virtuous covenant as per previous shreds of evidence.

Furthermore, an *in-vitro* drug release profile for optimized formulation applied to various kinetic models (zero-order, first-order, Higuchi, and Krosmayer Peppas model). To state kinetics profiling of drug release, obtained data were analyzed statistically in respect of rate constant and highest correlation. Best-fitted line found in all models except little suitability in zero-order equation. Resulting data described is semination of drug in controlled or regular manner from homogenous matrix systems and it states why drug disseminates at slower rate. Observations concluded that SLNG3 is far efficient as potential topical formulation for sustained drug delivery. This finding is almost similar to virtuous covenant as per previous shreds of evidence.



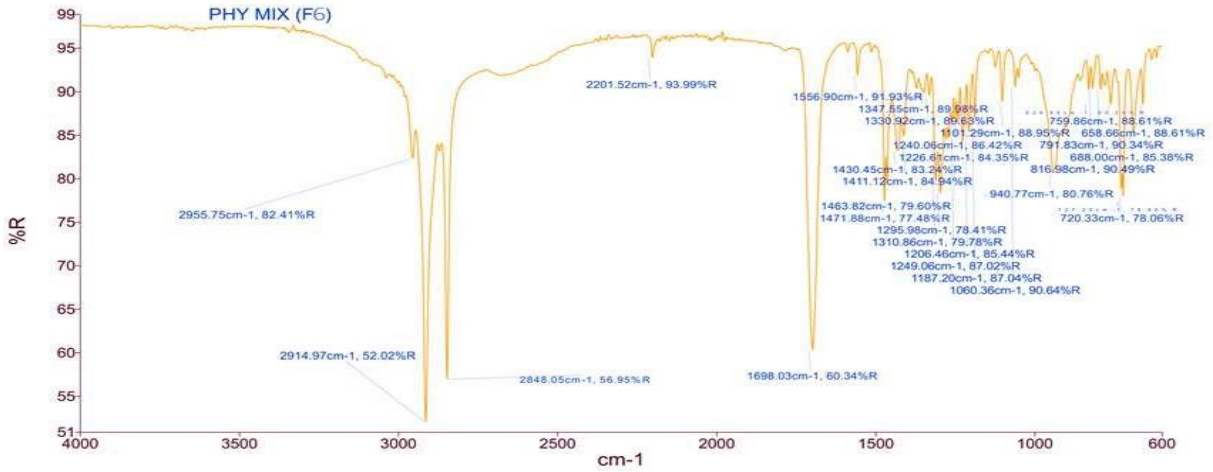
***In-vitro* drug release profile of SLN gel and control gel**

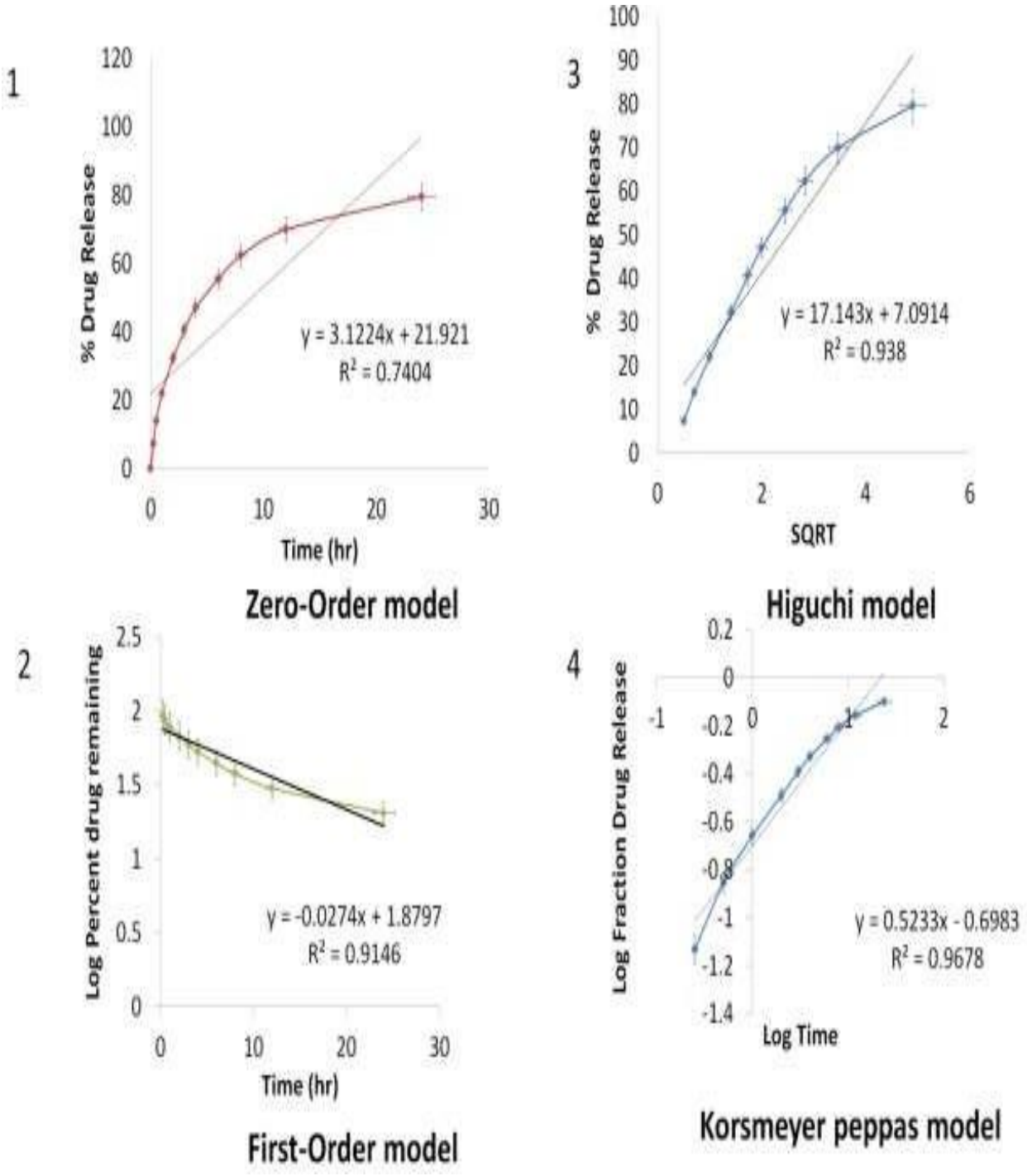
Furthermore, an *in-vitro* drug release profile for optimized formulation applied to various kinetic models (zero-order, first-order, Higuchi, and Krosmayer Peppas model). To state kinetics profiling of drug release, obtained data were analyzed statistically in respect of rate constant and highest correlation. Best-fitted line found in all models except little suitability in zero-order equation. Resulting data described is semination of drug in controlled or regular manner from homogenous matrix systems and it states why drug disseminates at slower rate. Observations concluded that SLNG3 is far efficient as potential topical formulation for sustained drug delivery. This finding is almost similar to virtuous covenant as per previous shreds of evidence. Graphical representation of kinetics order of SLNG3gel shown in Figure16[84]

Optimization and evaluation of SLN gel

The topical gel containing SLN loaded with Terbinafine prepared successfully by stirring method using carbopol 934 as gelling agent. Method of preparation of different SLN gel found unpretentious and robust. Initially, all four different preparations of SLN gel coded as G1, G2, G3, and G4, were evaluated to quantitate percent entrapment of Terbinafine spectrophotometrically at 299 nm. Resulting data shows that SLN G3 with 1.5% Carbopol w/w showing highest percentage of drug entrapment with 91.39% ± 0.187. There after optimized formulation is further evaluated to access physiochemical parameters includes visual appearance, pH, viscosity and Spreadability. Resulting data reveals that viscosity of G3 gel as found to be 369 cP, similar to gel viscosity as reported by Janaetal and pH found to be 6.12±0.255.[41]further in Spreadability evaluation, Spreadability factor of prepared SLN gel found to be 4.5 and it states that prepared gel produces excellent Spreadability as an ideal topical formulation. Spreadability is one of important physical properties of any topical formulation from patient's compliance point of view.

FTIR spectra of SLN F6

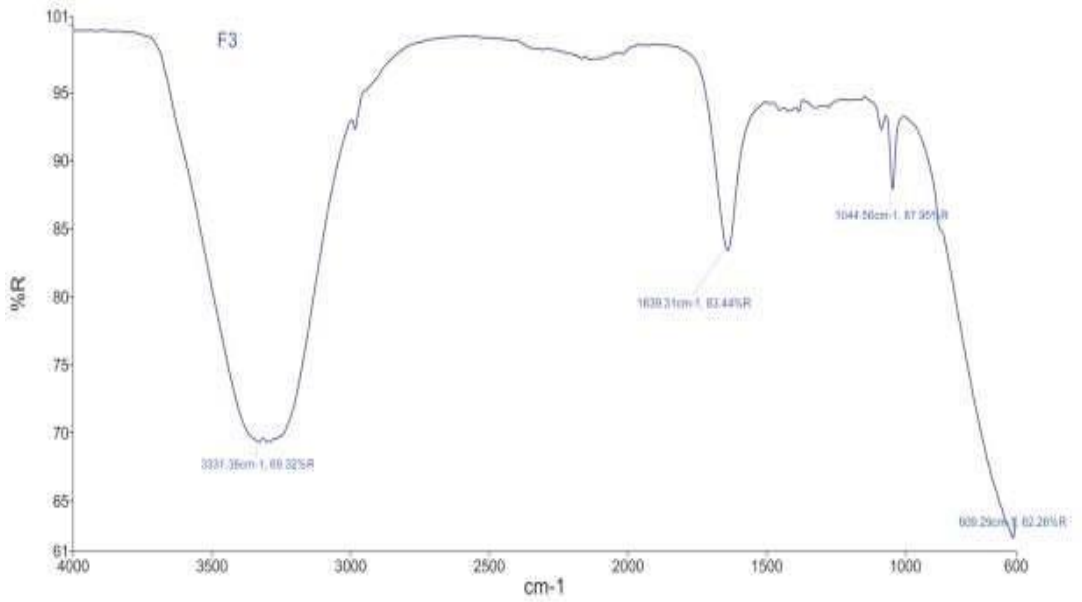




kinetics order of SLN G3 gel

FTIR spectral analysis of SLN G3gel

The FTIR spectral analysis of SLN gel G3 performed successfully to determine possible interaction between drug and drug additives and obtained spectral data matched with spectral data of Terbinafine and Stearic acid. Findings of spectral analysis show principal absorption peaks at 3331.36cm^{-1} for N–H stretching, 2961.88 cm^{-1} for C-H stretching,2193.49cm^{-1}for C≡N stretching, 609.26 & 1044.56cm^{-1} for C-Cl stretching for Terbinafine. Whereas, principal absorption peaks of Stearic acid were described to2932.49cm^{-1} & 2863.16 cm^{-1} in high-frequency region attributed to -CH_2- band asymmetric and symmetric stretching vibrations, and 1639.31 cm^{-1} for -COOH stretching is attributed in low-frequency region. Spectral analysis of optimized formulation G3 reveals that no more possible interaction between drug and drug additives even after successive formation of topical gel. Hence it can be said that spectra show purity and authenticity of SLN G3gel**.**



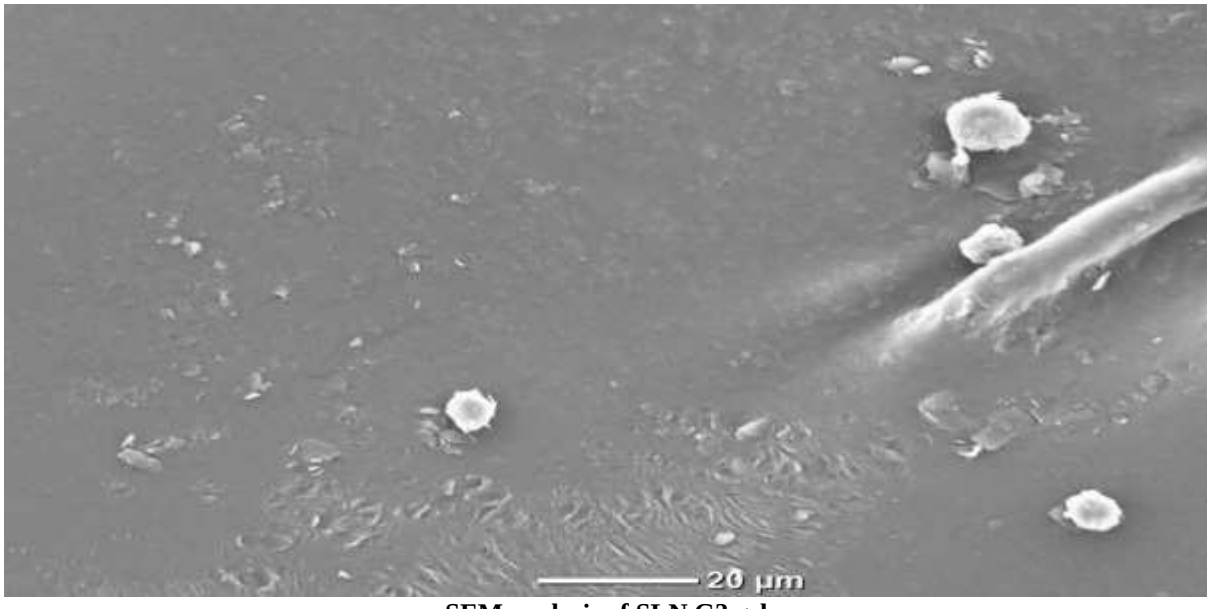
FTIR spectra of SLN gel G3

FTIR interpretation of SLN G3 gel

Characteristics Peaks	Reported(cm^{-1})	Observed(cm^{-1})
N-H stretch	3300 – 2400	3331.36
C=C stretch	1638 – 1648	1639.31
CO-O–CO stretch	1040 – 1050	1044. 56
C–Cl stretch	550 – 850	609. 26

Scanning electron microscopy

Most of vesicles is well specified, spherical, and discreet having large internal aqueous space. Low density of nanoparticles is shown in SEM analysis which may lead due to factor of dilution of nano suspension before preparing SEM photographs. SEM studies reveal that Terbinafine loaded SLN in gel had spherical shape with smooth surface.



SEM analysis of SLN G3 gel

CONCLUSION:

The purpose of topical drug delivery system is to allow therapeutic quantity of drug to correct place in body and to achieve and sustain desired effect of drug for while. In present investigation, we have designed solid lipid nanoparticles (SLN) loaded with Terbinafine to enhance skin permeation and controlled drug release at targeted site and incorporate them in topical gel of Carbopol 934with good skin retention time. Physicochemical property of prepared gel determined as per standards protocol to overcome compliance after

patient use. Even spectro scopical analysis reveals no chemical interactivity between Terbinafine and excipients. Microscopic examination (optical microscopy and scanning electron microscopy) of gel showed uniform distribution of SLN inside gel with good order of kinetics of drug release. Hence, it can be concluded that SLN gel provides controlled release of drug and these systems can be good source as drug carriers for lipophilic drugs, bioavailability enhancer for poorly water-soluble drugs by nanoparticles, drug delivery system.

REFERENCES:

1.Badiee, Parisa,and Zahra Hashemizadeh."Opportunistic invasive fungal infections: diagnosis & clinical management." *The Indian journal of medical research* 139.2 (2014):195.

2.Bonifaz, Alexandro,and AndrésTirado-Sánchez."Cutaneous disseminated and extracutaneous sporotrichosis: current status of a complex disease." *Journal of Fungi* 3.1(2017):6.

3.Mukherjee, S., S. Ray, and R. S. Thakur. "Solid lipid nanoparticles: a modern formulation approach in drug delivery system." *Indian journal of pharmaceutical sciences* 71.4 (2009):349.

4.Muller, R. "Solid lipid nanoparticles (SLN) for controlled drug delivery â€“a review of the state of the art."*Eur. J. Pharm. Biopharm* 50.1(2000):161-177.

6.MaliAD.Anupdatedreviewontransdermaldrugdeliverysystems.Skin.2015;8(9).

7.Ng KW, Lau WM. Skin deep: the basics of human skin structure and drug penetration. In Percutaneous penetration enhancers chemical methods in penetration enhancement 2015(pp.3-11). Springer, Berlin, Heidelberg.

8.Muller, R. "Solid lipid nanoparticles (SLN) for controlled drug delivery â€“a review of the state of the art."*Eur. J. Pharm. Biopharm* 50.1(2000):161-177.

9.Kumar M, Shanthi N, Mahato AK, Soni S, Rajnikanth PS. Preparation of Terbinafine nanocrystals loaded hydrogel for improvement of dissolution and antifungal activity. Heliyon.2019 May1;5(5):e01688.

10.Liu W, Hu M, Liu W, Xue C, Xu H, Yang X. Investigation of the carbopol gel of solidlipid nanoparticles for the transdermal iontophoretic delivery of triamcinolone acetonideacetate.Internationaljournal of pharmaceutics.2008Nov19;364(1):135-41.

11.Bräm S, Wolfram E, Recent Advances in Effect-directed Enzyme Assays based on Thin-layer Chromatography, Phytochemical Analysis, 2017;28(2):74-86.

13.Kaur J, Kaur J, Jaiswal S, Gupta GD. Recent advances in topical drug delivery system. Pharmaceutical Research. 2016;6(07).

14.Tiya boonchai W, Tungpradit W, Plian bang chang P. Formulation and characterization of curcuminoids loaded solid lipid nano particles. International Journal of Pharmaceutics.2007Jun7;337 (1-2):299-306.

15.Paliwal, R., Rai, S., Vaidya, B., Khatri, K., Goyal, A. K., Mishra, N., et al. (2009). Effect of lipid core material on characteristics of solid lipid nanoparticles designed for oral. Nanomedicine Nanotechnology,Biol.Med.doi:10.1016/j

16.Vidlářová,Lucie, etal."Nano crystals for dermal penetration enhancement–effect of concentration and underlying mechanisms using curcumin as model." *European Journal of Pharmaceutics and Biopharmaceutics* 104 (2016):216-225.

17.Pople PV, Singh KK. Development and evaluation of topical formulation containing solid lipid nanoparticles of vitamin A.Aaps Pharmscitech. 2006Dec1;7(4):E63-9

19.Naseri N, Valizadeh H, Zakeri-Milani P, Solid lipid nanoparticles and nano structured lipid carriers: Structure preparation and application, Advanced Pharmaceutical Bulletin, 2015;5(3):305–313.

21.Hu FQ, Hong Y, Yuan H. Preparation and characterization of solid lipid nanoparticles containing peptide. International journal of pharmaceutics.2004Apr1;273(1-2):29-35.

22.Kang KW, Chun MK, Kim O, Subedi RK, Ahn SG, Yoon JH, Choi HK. Doxorubicin-loaded solid lipid nanoparticles so overcome multi drug resistance in cancer therapy. Nanomedicine: Nanotechnology, Biologyand Medicine.2010Apr 1;6(2):210-3.

23.Kumar M, Shanthi N, Mahato AK, Soni S, Rajnikanth PS. Preparation of Terbinafine nanocrystals loaded hydrogel for improvement of dissolution and antifungal activity. Heliyon.2019 May1;5(5):e01688.

24.Abdel bary G,Fahmy RH.Diazepam-loaded solid lipid nanoparticles: design and characterization. Aaps Pharmsci tech. 2009Mar1;10 (1):211-9.

25.https://www.semanticscholar.org/paper/Recent-advances-in-oral-delivery-of-drugs-and-using/b74f5b81ade6cd18889a5b063a54f8342328ab65/figure/0

20.Ekambaram P, Sathali AA, Priyanka K. Solid lipid nanoparticles: a review. Sci Rev ChemCommun.2012 Feb;2(1):80-102

28.Lakshmi CV, Bengalorkar GM, Kumar VS. Clinical efficacy of topical terbinafine versus topical Terbinafine in treatment of tinea corporis/tineacruris patients. Journal of Pharmaceutical Research International. 2013 Aug 24:1001-14.

29.Basha BN, Prakasam K,Goli D. Formulation and evaluation of gel containing fluconazole-antifungal agent. Int J Drug Dev Res. 2011 Oct;3(4):109-28

30.Chandira RM, Pradeep PA, Bhowmik D, Chiranjib JB, Tripathi KK, Sampath Kumar KP. Design, development and formulation of antiacne dermatological gel. J Chem Pharm Res.2010;2(1):401-14.
31.Khanna,D.,and S.Bharti."Terbinafine for the treatment of fungal infections: An evidence-based review. Core Evid. 2014;9:113–24."
32.Desai, Noopur J., andDilip G. Maheshwari. "UV spectrophotometric method for the estimation of Terbinafine in marketed formulation (lotion)." *Pharma Science Monitor* 5.2(2014): 48-54.
33.Di,Li, and Edward H.Kerns. *Drug-like properties: concepts, structure design and methods from ADME to toxicity optimization*. Academic press,2015.
34.Hosseini, Seyed Mostafa, et al. "Doxycycline-encapsulated solid lipid nanoparticles as promising tool against Brucella melitensis enclosed in macrophage: a pharmacodynamicsstudyonJ774A.1cell line."*Antimicrobial Resistance & Infection Control*8.1(2019):62.
35.Lakshmi CV, Bengalorkar GM, Kumar VS. Clinical efficacy of topical terbinafine versus topical Terbinafine in treatment of tinea corporis/tineacruris patients. Journal of Pharmaceutical Research International. 2013 Aug 24:1001-14.
36.El-Housiny, Shaimaa, et al. "Fluconazole-loaded solid lipid nanoparticles topical gel for treatment of pityriasisversi color: formulation and clinical study." *Drug delivery* 25.1(2018): 78-90.
37.Naseri N, Valizadeh H, Zakeri-Milani P, Solid lipid nanoparticles and nano structured lipid carriers: Structure preparation and application, Advanced Pharmaceutical Bulletin, 2015;5(3):305–313.
39.Harde H, Agrawal AK, Katariya M, Kale D, Jain S. Development of a topical adapalene-solid lipid nano particle loaded gel with enhanced efficacy and improved skin tolerability. RSC Advances. 2015;5(55):43917-29.
41.Pawar PM, SolankiKP, MandaliVA. Recent Advancements in Transdermal Drug Delivery System. International Journal of Pharmaceutical and Clinical Research.2018;10 (3):65-73.
42.Makri,Nikoleta,etal."First case report of cutaneous sporotrichosis (Sporothrixspecies)in a cat in the UK." *Journal of Feline Medicine and Surgery Open Reports* 6.1 (2020):2055116920906001.
44.Tyagi RK, Chandra A, Singh D, Rahman MA. Transdermal drug delivery system (TDDS):an overview. International journal of pharmaceutical sciences and research.2011 Jun1;2 (6):1379-88.