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Formulation and Evaluation of Imiquimod Loaded Nanosuspension Gel for Transdermal Delivery



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ABSTRACT

The main objective of this present study was to formulate and evaluate the nanosuspension based gel topical delivery system of imiquimod or increased solubility, permeability and sustained drug release. Imiquimod nanosuspension was prepared by media milling technique using glass beads as a milling media, poloxamer 407 as a stabilizer. Effect of various process parameters like stirring time, stirring speed and poloxamer or drug concentration, concentration of beads was optimized. Nanosuspension is optimized then incorporated into a topical gel using Carbopol 934P as a gelling agent for sustained release. The particle size results showed that decreases with increase in the surfactant and drug concentration. SEM showed that IMI nanosuspension spherical in shape. In- vitro permeation studies showed that the amount of IMI permeated through the membrane of IMI nanogel of 1% concentration (80.248 ± 0.162) (220.89 mg/cm^2) after 24 h was higher than IMI nanogel to the different concentration (pure drug, 1.5%, 2.0%) of IMI nanogel (13.207 ± 0.084 , 68.18 ± 0.162 , 55.644 ± 1.296) after 24 hrs Prepared imiquimod loaded nanosuspension gel was clear and showed good homogeneity and pH was found in the normal skin range. The drug release from imiquimod nanosuspension loaded gel was significantly prolonged by using the gelling system due to the addition of the polymer Carbopol 934P and it follows Higuchi matrix. Imiquimod nanosuspension loaded topical gel showed increased saturation solubility and sustained drug release and these results suggested that nanogel is eligible for the use as suitable nanomedicine for topical delivery of poorly soluble drugs such as IMI.

INTRODUCTION

In this paper, the imiquimod loaded nanosuspension gel is formulated and goes to improve its permeability and solubility. Imiquimod is an immunostimulant and it belongs to the clusture of nucleosides analogue's a small molecule of imidazoquinolinone family that was first synthesized as the most likely antiviral agent. Imiquimod (IMQ) drug topically used for the treatment of skin and mucosal infections, actinic keratosis or basal cell carcinoma. Its therapeutic effect is mediated by binding to Toll-like receptors 7 and 8, leading to the release of pro-inflammatory cytokines, chemokines and other mediators [1]. IMQ is a BCS class IV drug that has low solubility and low permeability in many hydrophilic and lipophilic pharmaceutical excipients. Additionally, it has been recently demonstrated that fatty acid, oily phase present in the commercial aldera® cream, they have biological activity and executes additive or synergistic action with the drug [2]. It is a chemically stable off-white crystalline powder with a high melting point (297-299°C). Given the poor solubility characteristics of the molecule, it was not possible to solubilize enough drug to form a therapeutically effective single-phase gel [3]. Imiquimod marketed cream activates the immune system, and very rarely there have been reports of pre-existing autoimmune disease flaring up. Rarely, a patient can be allergic to Imiquimod cream and develop a severe allergic rash on the body skin when taken more than normal dose [4]. A nanosuspension is a very finely colloid, biphasic, dispersed, solid drug particles present in the liquid (aqueous vehicle), and its range of size below 1 µm, without any matrix material. Surfactants and polymers are used as a stabilizer, prepared by suitable methods. Drugs compressed within nanosuspensions exist as crystalline or amorphous state. A nanosuspension improves and solve a problem of poor solubility and bioavailability. Nanosuspension formulation approach is most suitable for the compounds with high log P value, high melting point. [5-8]. Topical drug administration may be a localized drug delivery system anywhere within the body through ophthalmic, rectal, vaginal, and skin as topical routes. Skin is one of the most accessible organs of the human body for topical administration and the main course of topical drug delivery system. Numerous medicated products applied to the skin or mucosa that either enhances or restores a fundamental function of skin or pharmacologically changes action in the underlined tissues. Such products are mentioned as topical or dermatological products [9]. Gels are a substantially dilute cross-linked system, which exhibits no flow when in the steady-state. They consist of a two-component semisolid system rich in liquid. Their one characteristic feature is that the existence of continuous structure provided that solid-like properties. Gels

are the foremost material used for drug delivery formulations outstanding to its biocompatibility, network structure, and molecular stability of the incorporated bioactive agent [10-12].

MATERIALS AND METHODS

MATERIALS

The following materials were used (Grade-LR): imiquimod-API (HelioxPharma, Mumbai), Poloxamer 407 (Yarrowchem Products, Mumbai), Carbopol 934P, Potassium dihydrogen Orthophosphate, Sodium hydroxide (Spectrum Reagents and Chemicals, Cochin), Propylene glycol, Methanol (Nice Chemicals, Cochin).

A. METHOD

a) Pre-formulation studies

Pre-formulation studies are the leading phase in the development of dosage forms that can be well-defined as “investigation of physical and chemical properties of a drug substance alone and when it combines with excipients”. These physicochemical studies should focus on the applications of the novel compound that could affect the drug development and performance of an effective dosage form. A thorough understanding of the physical and chemical properties of a drug is important will ultimately provide a rationale formulation design or support the necessity for molecular modification. [13-15]

The goals of pre-formulation are:

- i. To establish the necessary physical and chemical parameters of new drug substances.
- ii. The kinetic rate profile determined.
- iii. To establish physical characteristics.
- iv. To establish compatibility with the excipient.

Therefore, pre-formulation studies are compulsory to establish the identity and physicochemical parameter of the particular drug was subjected to the following investigations:

- i. Organoleptic Properties
- ii. UV- VIS spectroscopy
- iii. FTIR spectroscopy
- iv. Melting point
- v. Partition coefficient
- vi. Solubility

Organoleptic Properties

The organoleptic studies are to study the colour, appearance, odour etc. were performed by observed visually.

Colour: Small quantity of drug is placed on the butter paper and observed at illuminated place.

Odour: Smelled small amount of drug and identify its odour. [16,17]

Solubility

In different solvents like water, ethanol, methanol, acetone and dichloromethane, the solubility of imiquimod active ingredient was observed. [18]

Melting Point

By capillary diffusion method the melting point of the imiquimod drug is determined the method followed from USP. [18]

Determination of maximum absorption of the imiquimod drug by UV-Spectrophotometer

Absorption maxima (λ_{max}) of imiquimod drug were determined by UV Spectrophotometer (Shimadzu Pharma. Spec1800). [19]

Preparation of imiquimod Calibration Curve in 0.1N HCl

The stock solution was prepared by dissolving 10mg drug in 10ml (1000 μ g/ml) of 0.1N HCl buffer. [20]

Partition coefficient of imiquimod

Partition coefficient (oil/water) is a measure of a drug's lipophilicity and an indication of the drug's ability to cross cell membranes. The partition coefficient study was performed by using the shake flask method. [21,22]

Drug and excipient compatibility

Fourier transform infrared spectroscopy were performed for the identification of compounds. FT-IR Spectroscopy of the pure drug (Imiquimod) and the polymer was done. Various peaks in FT-IR Spectrum were interpreted for the identification of different groups present in the structure of pure drug (Imiquimod), & its mixture. FT-IR Spectroscopy can also be used to investigate and predict any physicochemical interactions between different components. [23,24]

b) Preparation of imiquimod loaded nanosuspension

Nanosuspension was formulated by using media milling technique using glass beads as the milling agent. In 10 ml glass vial, 5 g of glass beads having a diameter between 0.2– 0.4mm were added along with 3mL distilled water. Pluronic F 127 (Poloxamer 407) and active pharmaceutical ingredient imiquimod (50mg/ml) were incorporated and micronized using a magnetic stirrer for 12 h at 1200 rpm. [25,26]

c) Screening of formulation and other various parameters

Screening of various parameters is very important for the formation of nanosuspension by media milling method. A preliminary screening study was first approved to rank the significance of four major parameters like surfactants and check their wettability. Depends on their wettability the surfactant is selected according to their particle size. Then further screening of surfactant with their different concentration, and examined particle size. All parameters finalized according to their particle size.

d) In vitro characterization of imiquimod loaded nanosuspension

Particle Size

It is the most important parameter in the evaluation of the suspensions as it is having straight effect on the solubility and the physical stability of the formulation. The mean particle size

and the width of particle size can be determined by Photon Correlation Spectroscopy (PCS), electrophoretic mobility, respectively, using a Malvern Zeta Sizer instrument. The prepared nanosuspension was 100 times dilute, then added into the sample cell, put into the sample holder unit and the measurement was administered. All the samples were examined in triplicate.

Zeta Potential

The particle charge importance in the study of the steadiness of the suspensions. The zeta potential of more than $\pm 40\text{mV}$ will be required for the stability of the dispersions. For electrostatically stabilized nanosuspension a minimum zeta potential of $\pm 30\text{mV}$ is required and just in occurrence of combined steric and electrostatic stabilization it should be the lowest of $\pm 20\text{mV}$ zeta potential is required. The prepared nanosuspension was diluted 100 times, added into the sample cell, put into the sample holder unit and measurement was carried out. All samples were examined in triplicate. [27]

Saturation Solubility

Nanosuspension increases the saturation solubility. An increase in solubility that occurs with relatively low particle size reduction may be mainly due to a change in surface tension leading to increased saturation solubility; therefore, it is necessary to investigate the saturation solubility of a nanosuspension. Saturation solubilities (SS) of bulk imiquimod powder and nanosuspension formulation were measured in distilled water. For bulk imiquimod excess of the drug was added to water and magnetically stirred for 12 h. Thereafter, both samples were centrifuged at 15 000rpm for 10 min. The supernatant layer was measured using a UV Spectrophotometer. [27]

Scanning electron microscopy

The morphology of coarse nanosuspension of imiquimod was determined by scanning electron microscopy, respectively. The nanosuspension (freeze-dried) was kept in the sampling unit as a thin film and photographs were taken at various magnifications using a scanning electron microscope. [28]

e) Preparation of imiquimod loaded nanosuspension gel

Process: The nanosuspension of imiquimod was generally prepared by media milling method [24]. Take 5 ml nanosuspension and added different gelling substances 50mg (1%) like Carbopol- 934, Carbopol-980, Hydroxy Propyl methylcellulose E15, Hydroxy Propyl methylcellulose E50 and kept aside for 1 hour for swelling continuous stirring at a speed of 800 rpm using a mechanical agitator. The dispersion was neutralized using triethanolamine until pH 6.0 to 8.0. [29]

The gel could stand overnight to remove entrapped air.

f) Screening of different type of gelling agent for the preparation of nanosuspension gel

The optimization of different gelling agents and analyzed it. The finalized gelling agent screened with different concentration to formulate nanosuspension gel.

The imiquimod loaded nanosuspension gel was formulated by varying different concentrations (1.0, 1.5, 2.0 %) of Carbopol-934 (gelling agent) to analyze their better permeation through skin.

g) Evaluation of nanosuspension gel:

Appearance of gel

The prepared nanosuspension gel visually observed by placed in the petri dish and visual inspection under black and white background and it was graded as follows: turbid, clear, very clear. [30]

Percentage drug content

To ensure uniform formulation of the gel. Drug content of the gels was determined by dissolving an accurately weighed quantity of gel (about 1 gm) added in 5ml nanosuspension gel of different concentration (1%, 1.5, 2.0 %) and diluted in 25 ml methanol, sonicate for 10 mins and fill it into Eppendorf, centrifuged for 15 mins. After that 1ml upper supernant layer taken and makeup volume upto10ml with methanol. Then further and further diluted and analyzed under UV spectrophotometry analysis for imiquimod at 226 nm. Drug content was determined from the standard curve of imiquimod. [31]

Percentage detection of pH

The pH of nanosuspension gel was determined by using a digital pH meter at room temperature. Dissolve 1g of gel in 100 ml of distilled water and sonicate it for 5 mins then the electrode was then dipped into gel formulation and constant reading was noted in triplicate. [32]

In- vitro permeation studies

The in- vitro permeation studies were performed using Franz diffusion cells for studying the dissolution release of various formulation of gel through a dialysis membrane-70 with an effective diffusion area of 1.77 cm² and receptor volume of 20 ml. 1g of gel sample was taken in dialysis membrane. The diffusion studies carried out at 37 ± 1°C using 250 ml of phosphate buffer (pH 7.4) as the dissolution medium. A standard dialysis membrane (soaked in 20% ethanol for 24 hours before use) was fixed to one end of the cylinder with the aid of an adhesive to result in permeation cell. One gram of gel was taken in the cell (acceptor compartment) and the phosphate buffer 7.4 filled in the donor compartment of the beaker. Sample 1 ml of pure drug, 1, 1.5, 2.0 % of nanogel filled in the donor compartment was taken at different interval of time (0.25, 0.5, 1, 2, 3, 4, 6, 8, 10, 12, 24 hr) and assayed for nanogel of imiquimod at 226 nm. The sample was withdrawing, and this was replaced with a phosphate buffer. The quantity of imiquimod released at various intervals of time was calculated and plotted against time. [33]

Stability Studies

The 5gm nanosuspension gel is placed at different temperatures for 6 months and detects its pH and solubility and drug content after a regular interval of time (monthly for 6 months).

Drug release kinetic studies

In the present study, raw data obtained from in vitro release studies were examined, wherein data was formfitting to diverse equations and kinetics model to calculate the percent drug release and release kinetics of imiquimod loaded nanosuspension gel. The kinetic models used were a Zero-order equation, First-order, Higuchi's model and Korsmeyer-Peppas equation.

Zero-order kinetic

A zero-order release would be predicted by the subsequent equation.

$$A_t = A_0 - K_0t \quad (1)$$

Where:

A_t = Drug release at time 't'

A_0 = Initial drug concentration

K_0 = rate constant (hr^{-1})

When the values of the data are plotted as cumulative percent drug release versus time, if the plot is linear then the information of data obeys zero-order release kinetics, with a slope equivalent to K_0 .

First-order kinetic

A first-order release would be predicted by the resulting equation.

$$\text{Log } C = \text{Log } C_0 - K_t/2.303 \quad (2)$$

Where:

C = Amount of drug remained at (t) time

C_0 = Initial amount of drug

K = First-order rate constant (hr^{-1})

When the data values are plotted as log cumulative percent drug remaining versus time yields a straight line, indicating that the release surveys first-order kinetics. The constant 'K' often obtained by multiplying 2.303 with slope values.

Higuchi's model

Drug release from the matrix devices by diffusion has been described by following Higuchi's classical diffusion equation:

$$Q = [D\varepsilon/\tau (2A - \varepsilon C_s)] C_s t^{1/2} \quad (3)$$

Where:

Q= Amount of drug released at time 't'

D= Diffusion coefficient of the drug within the matrix

A = Total amount of drug in the volume of matrix

C_s = The solubility of drug in the diffusion medium

ε = Porosity of the matrix

τ = Tortuosity

t = Time (hrs) at which 'Q' amount of drug is released.

Equation may be simplified if one may let that Dε, C_s and A are constant. Then the equation becomes:

$$Q=K.t^2 \quad (4)$$

When the data plotted according to the equation i.e., cumulative drug released versus square root of time, yields a straight line, indicating that the drug was released by diffusion mechanism. The slope is equal to 'K'.

Korsmeyer-Peppas's Model

The release rates from controlled release polymeric matrices can be pronounced by the equation:

$$Q = K_1 t^n \quad (5)$$

Where:

Q = Percentage of drug released at the (t) time

K = Kinetic constant incorporating structural and geometric characteristics and 'n' is the diffusion exponent indicative of the release mechanism.

For Fickian release, $n = 0.45$ while anomalous (Non-Fickian) transport, n ranges between 0.45 and 0.89 and for zero-order release, $n = 0.89$.

RESULT AND DISCUSSION

A. Pre-formulation studies

Pre-formulation studies are an integral part of the entire drug development process. It is the study of the physical and chemical properties of drug previous compounding process. In the simplest case, these pre-formulation surveys may merely confirm that there are no substantial barriers to the compound's development. These studies are requisite protocol for the development of safe, effective, suitable and stable dosage form. The obtained drug sample was recognized by various analytical techniques such as UV spectroscopy, IR spectroscopy, melting point, solubility.

Organoleptic properties

The drug is odourless, white to off white crystalline in nature. It is in powder form.

Melting point

The melting point was found to be range; hence drug sample was free from any type of impurities. Melting point is observed within 293-295°C and their reported melting point is 293-294°C.

Compatibility Studies

The observed major peaks in the spectrum were also detected in the spectrum of drug with the polymer Poloxamer- 407 was within the characteristic peaks range.

Determination of maximum absorption by UV Spectroscopy of imiquimod

UV-VIS spectroscopy is mainly used for quantitative analysis and serves as a useful auxiliary tool for structural elucidation of various drugs to obtain specific information on the chromophore's part of the molecules in solution when exposed to light in the visible/ultraviolet region of the spectrum absorbing light of particular wavelength depending on the type of electronic transition associated with the absorption. The UV spectrum is generally recorded as a plot of absorbance versus wavelength.

A double beam UV-visible spectrophotometer was used for quantitative analysis of the drug. A 100 µg/ml solution of Imiquimod in 0.1N HCl buffer was scanned in the range of 200-400 nm. The result of UV spectrum of Imiquimod is shown in Fig.1.

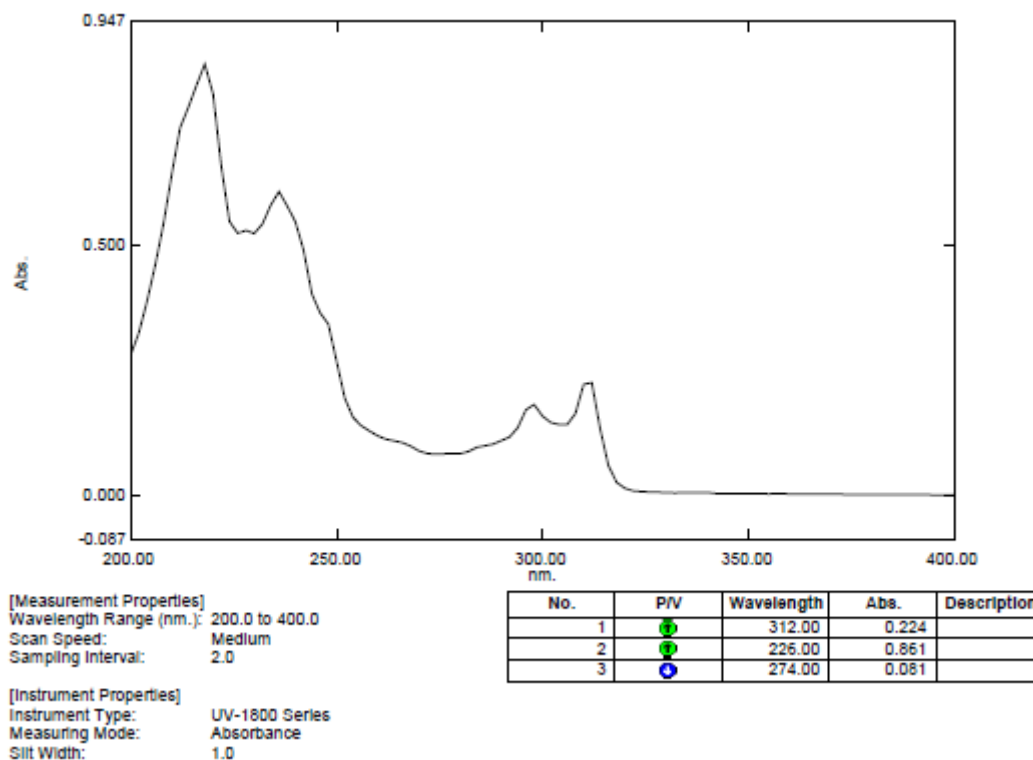


Fig. no. 1 UV spectrum of imiquimod in 0.1N HCl

Discussion: The maximum wavelength of Imiquimod was observed at 226 nm.

Preparation of imiquimod Standard Calibration Curve in 0.1N HCl buffer

The standard stock solution of imiquimod (10 mg/ml) was prepared in 0.1N HCl buffer. This solution was diluted with 0.1N HCl buffer to obtain apposite dilution examined spectrophotometrically at 226 nm.

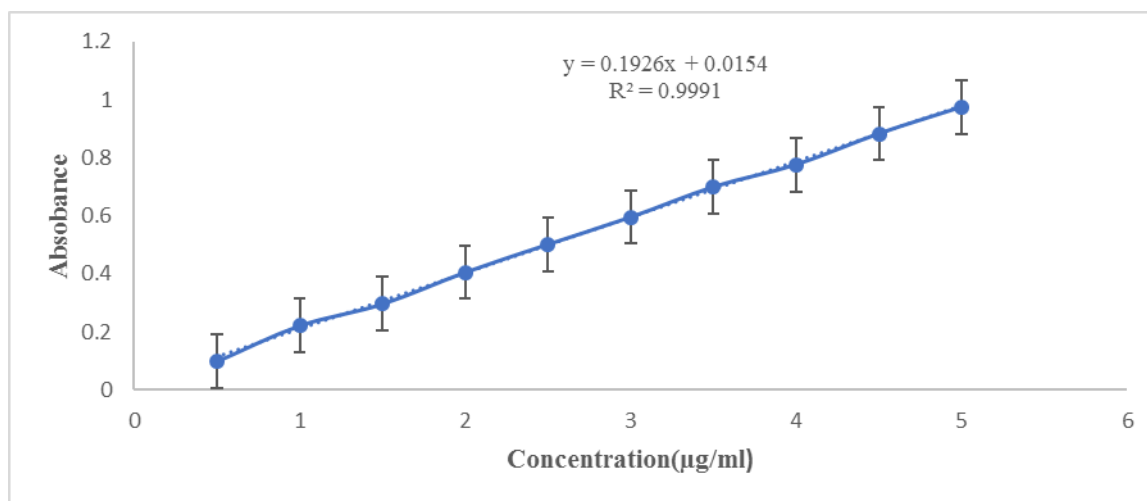


Fig. no. 2 Standard calibration curve of imiquimod

Solubility studies

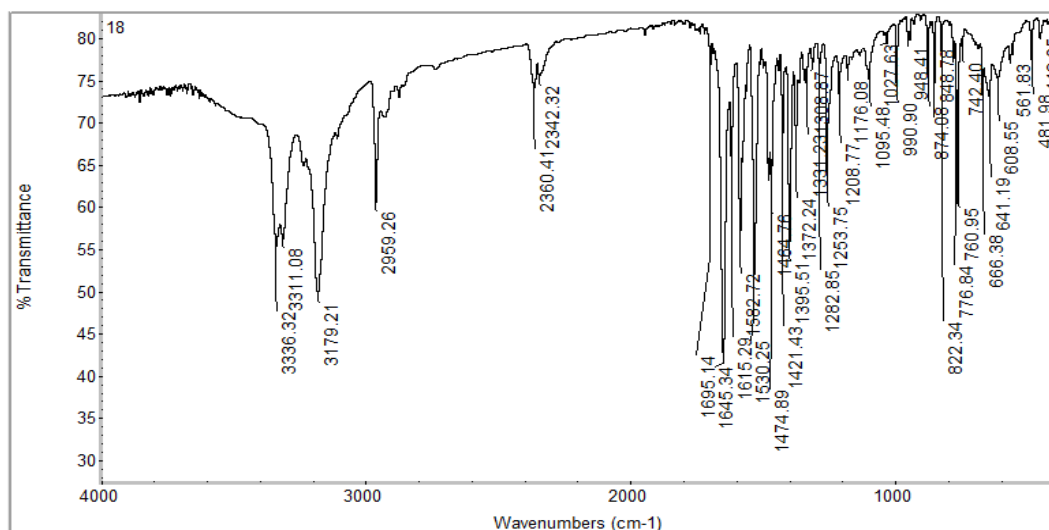
The spontaneous interaction of two or more substances to make a uniform molecular dispersion is named as solubility. The drug imiquimod was more soluble in 0.1N HCl<DCM< Acetone< Diethyl ether< Methanol<Ethanol< Water< Chloroform< Phosphate buffer 6.8< Phosphate buffer 7.4.

Partition coefficient

Partition coefficient study was performed by using shake flask method. The partition coefficient of Imiquimod in n- Octanol: Water was found to be 2.194 ± 0.0030 . This indicates that the imiquimod drug is lipophilic in nature.

FTIR analysis of pure drug

FT-IR analysis measures the selective absorption of light by the vibration modes of specific chemical bonds in the sample. The FT-IR spectrum of imiquimod is shown in Fig. 2.



Analysed By:

Checked By:

Thu Sep 05 15:41:33 2019 (GMT+05:30)

Fig. no. 3 FTIR spectrum of imiquimod

Discussion: The observed FT-IR spectrum confirmed and identified the presence of functional groups and the purity of the drug.

FTIR Analysis of physical mixture

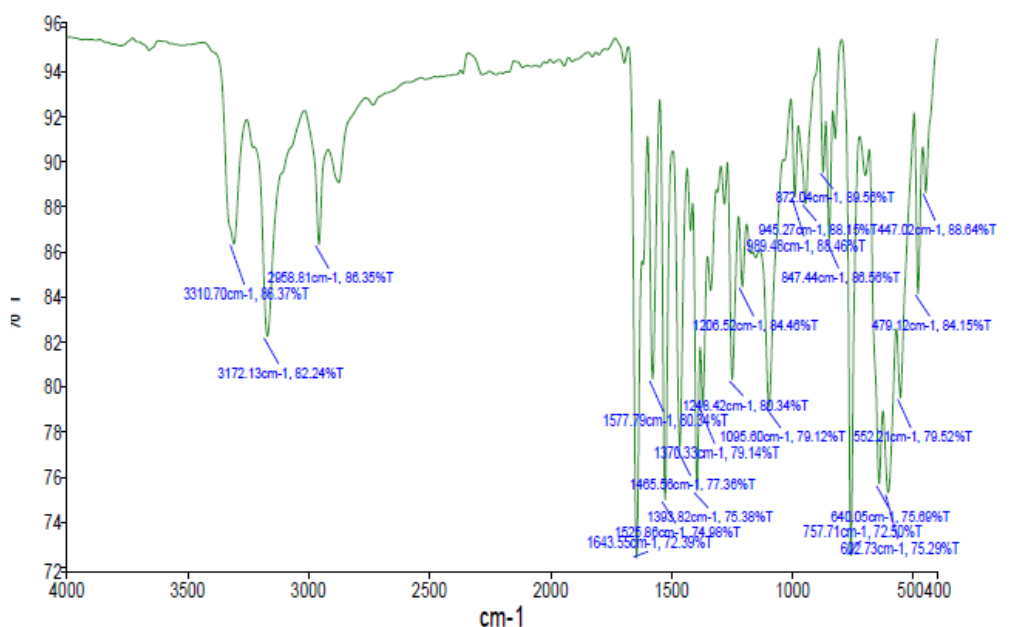


Fig. no. 4 FTIR spectrum of imiquimod and poloxamer 407(physical mixture) Preparation and screening of nanosuspension

Table No. 1: Optimization parameters for preparation of imiquimod nanosuspension

Criteria	Level	Particle Size	PDI*
Surfactant	Poloxamer 407	345.3	0.201
	Tween 80	554	0.485
	Sodium lauryl sulphate	496	0.366
Concentration of surfactant (%)	0.2% (w/v)	326.1	0.512
	0.4% (w/v)	448.2	0.365
	0.6% (w/v)	655	0.455
	0.8% (w/v)	745	0.403
Size of beads	0.2-0.4nm		
Concentration of beads	80% (w/v)	698	0.322
	100% (w/v)	346.6	0.275
	120% (w/v)	445	0.362
Concentration of drug	3.7%	347.1	0.266
	5%	566	0.245
Stirring time (hr)	6	784	0.312
	8	556	0.235
	12	355.2	0.271
	16	425	0.242
	18	884	0.312

To find an appropriate range for each parameter, a set of preliminary screening was conducted. Various formulation parameters such as type of surfactant, the concentration of surfactant, concentration of milling medium (beads), the drug concentration and stirring time were optimized in the preliminary stages. Preliminary parameters were optimized by varying one parameter at a time while keeping others constant and observing their effect on particle size. Surfactant plays a vital role in stabilization and wetting of particles. When the new surface area is formed, it produces very high free energy, which leads to the agglomeration of particles and thus leads to rise particle size. The agglomeration of particles can be avoided by adding the surfactant which reduces the interfacial tension and thus surface free energy. Various surfactants were tried for selecting a suitable stabilizer for the nanosuspension formulation. Minimum particle size and low PDI were obtained with Poloxamer 407.

It was observed that particle size reduced, and the concentration of beads was increased 80%, 100%, 120% w/v. Therefore, 100% w/v of small beads (mm) were selected as the milling agent. Minimum particle size (nm) was obtained at 3.7% w/w drug concentration as compared to 5% w/w concentration (nm).

Stirring time was also varied to obtain minimum particle size. It was observed that particle size decreased while stirring and then further increased indicating that stirring time was a critical parameter affecting the size of the nanosuspension. Therefore, stirring time was selected in the range of 12 hrs.

Table No. 2: Final formulation of imiquimod nanosuspension

Final formulation	Particle size (nm) *	PDI*
19	312.3	0.235

B. Evaluation of imiquimod loaded nanosuspension

Particle size

Mean particle size of the imiquimod loaded nanosuspension nanoparticles was observed 312.3 for F 19. PDI is the measure of homogeneity of dispersion and usually ranges from 0 to 1. The value close to zero indicates a homogenous dispersion. Narrow size distribution is essential in preventing particle growth and for maintaining stability of nanosuspensions. Formulation 19 had the least PDI values (0.235) and hence were considered as optimum formulation.

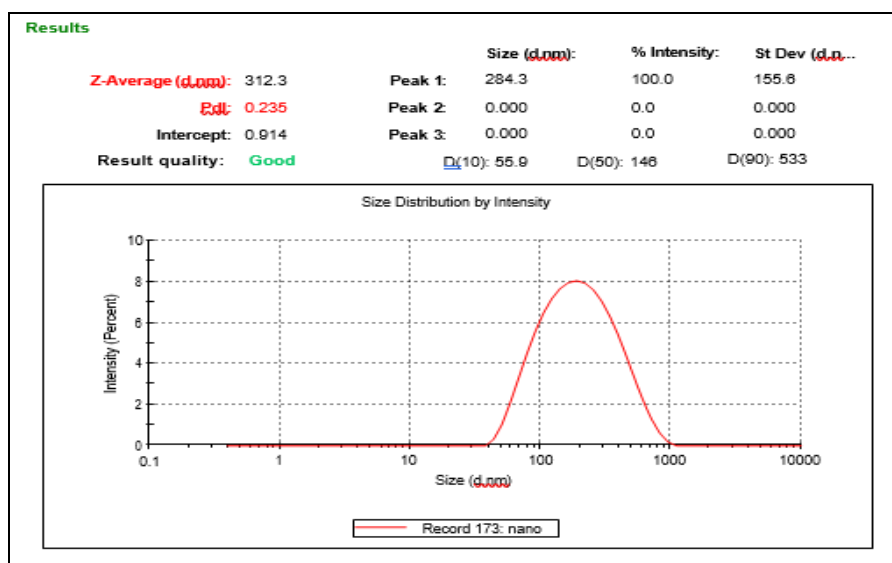


Fig. no. 5 Particle Size of nanosuspension

Zeta potential

Zeta potential specifies the degree of repulsion between likewise charged particles in the dispersion. Zeta potential final formulation F-19 was found to be 23.0 ± 6.23 mV respectively. The presence of reasonably high zeta potential will confer stability, i.e. it will lead to the repulsion between the particles and reduces the chances of formation of agglomeration. Besides, Poloxamer 407 is used as a stabilizer which provides steric stabilization.

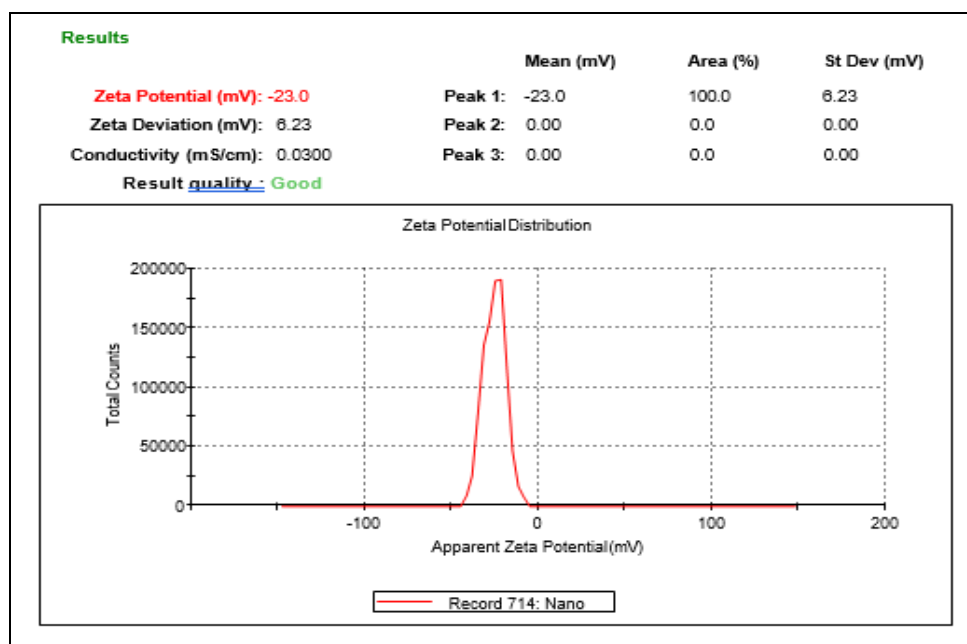


Fig. no. 6 Zeta potential of nanosuspension

Saturation Solubility

The SS of pure drug and NS were found to be 0.05006 ± 0.0003 mg/ml and 1.53014 ± 1.9914 mg/ml respectively which shows rise in saturation solubility due to nanosizing. This increase in saturation solubility is attributed to decrease in particle size, increased permeability. Moreover, the reduction in size of real drug from micron to nanosized particles may form defects in lattice space changes in space lattice energy or dislocations. Such changes in the energy of space lattice leads to rise in surface free energy which successively increases saturation solubility.

Scanning electron microscopy

SEM images of optimized nanosuspension show that the nanocrystals are spherical in shape not of uniform size as manifest by the high value of the polydispersity index.

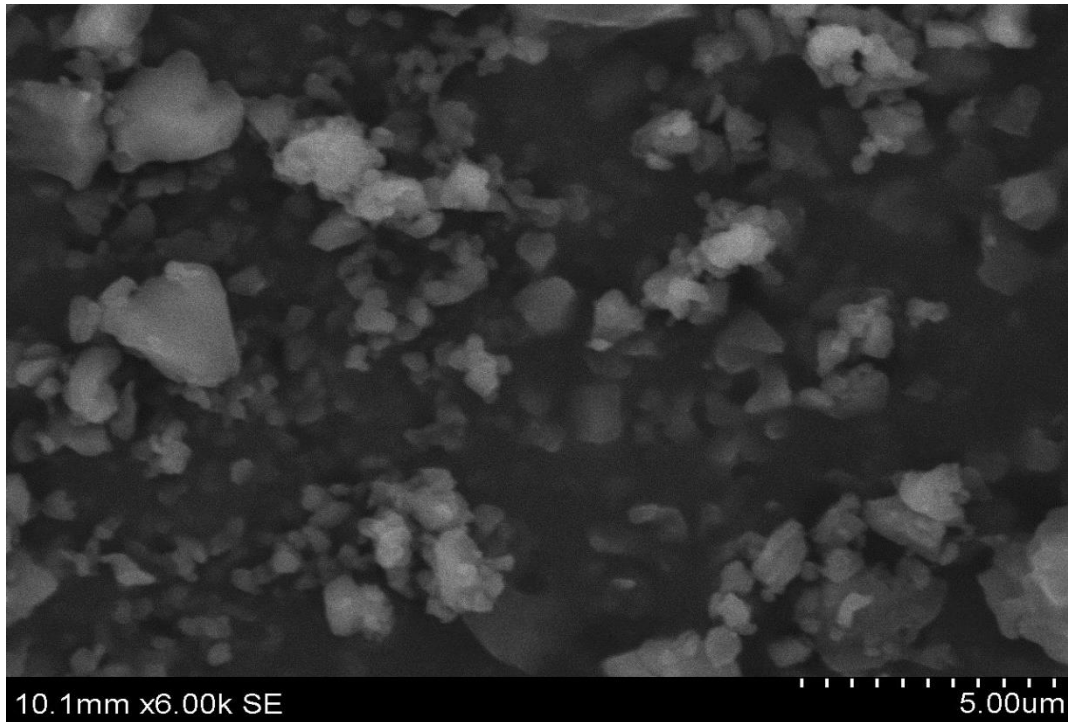


Fig. no. 7 Scanning electron microscopy of nanosuspension

Evaluation of imiquimod loaded nanosuspension gel

Physical Appearance

The appearance of nanogel formulation found to be homogeneous white gel preparations.

Percentage of drug content

The drug content of the imiquimod loaded nanosuspension was found to be for F24, F25, F-26 is 95.165, 94.814, 93.645 %.

Percentage detection of pH

The percentage pH measurement of imiquimod loaded nanosuspension gel was found to be 7.01 ± 0.11 .

FTIR of nanosuspension gel

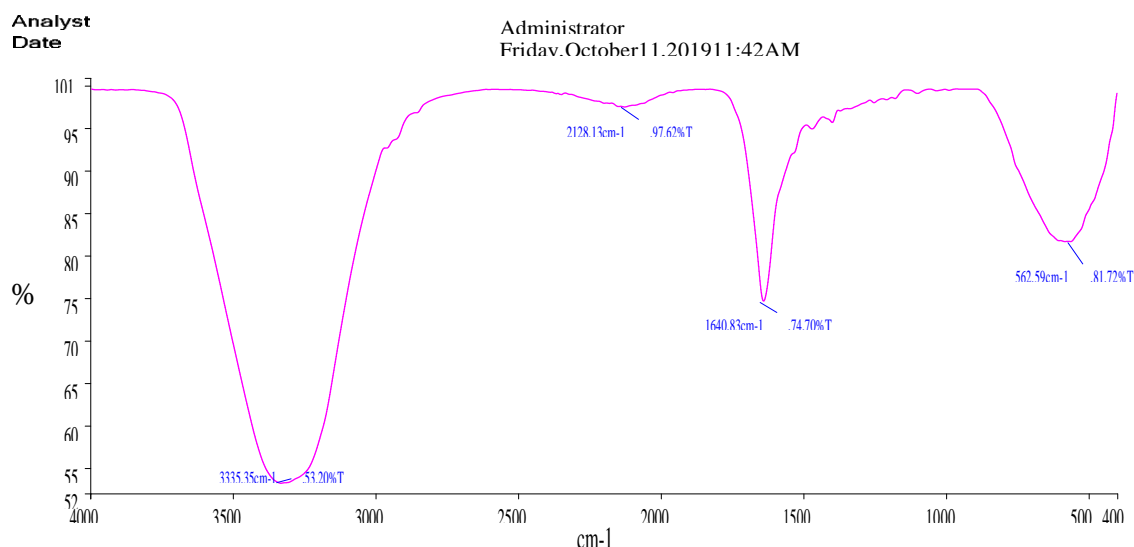


Fig. no. 8 FTIR of nanosuspension gel

In-vitro Permeation Studies

In-vitro permeation studies were used to find the amount of drug being released into system/medium at a period. Formulation containing different concentration of gelling agent (Carbopol- 934) nanosuspension gel showed good characteristic fast drug release in comparison to pure imiquimod suspension. In the case of F-24 (1%), F-25 (1.5%) formulation, about 80.248 ± 0.162 , 68.18 ± 0.162 of the drug was released in the medium within 24 hours apart from it, in- vitro release of pure drug suspension showed 13.207 ± 0.084 drug release within 24 hrs.

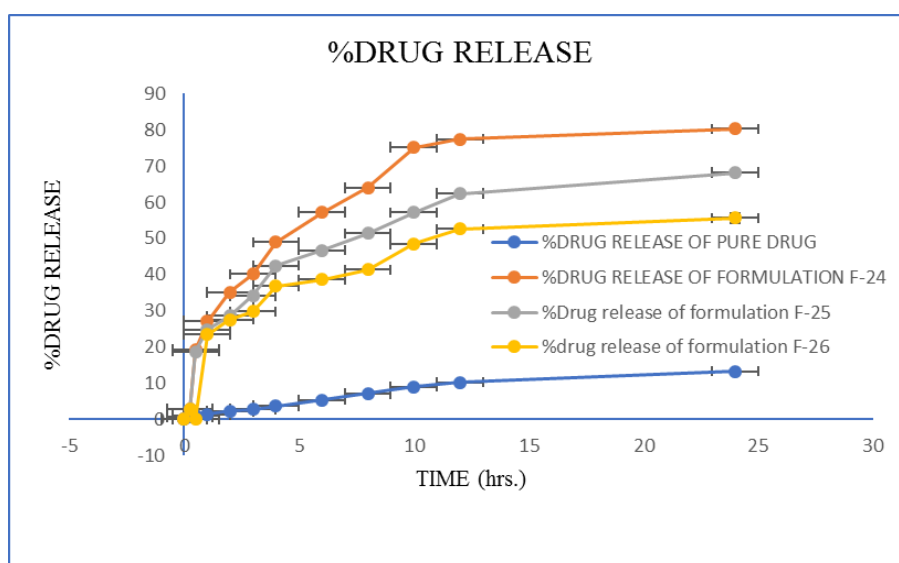


Fig. no. 9 Comparative drug release profile

Stability studies

Drug content and drug release profile of the formulation after a month for 6 months determined at different temperature but still stable at a cool temperature. There is no considerable change in these parameters.

Table No. 3: Stability studies OF F-24 imiquimod nanosuspension gel

Formulation	Before stability study		After stability study (avg. of six months)	
	Drug content	% Drug release	Drug content	% Drug release
Imiquimod loaded nanosuspension gel	93.878	80.248	92.881	79.783

In-vitro release Kinetic

The data obtained for in-vitro release was fitted in the equation for the zero-order, first-order, Higuchi and Korsmeyer Peppas's models. The interpretation of values of data maintain the worth on the value of the resulting regression coefficients. Calculated regression coefficients for zero order, first order and Higuchi model and Korsmeyer was found that the in vitro drug release of F-24 was best explained by Higuchi models as the plot showed the highest linearity. The value of R^2 found to be 0.8937 highest for the Higuchi models for sustained release.

CONCLUSION

Imiquimod nanosuspension was successfully prepared by media milling method. This method of manufacturing was found to be very simple and has scale-up feasibility. Prepared nanosuspensions were estimated by solubility and in vitro drug release. The drug release profiles from the nanosuspension formulations was studied, and the results showed that drug release increased with an increase in the surfactant concentration with high rotation speed. The optimum formulation was prepared and performed various evaluations such as particle size and zeta potential, morphological characterization by SEM, percentage in vitro drug release studies. Optimized formulation of nanosuspension was chosen for formulation of topical gel. Prepared imiquimod loaded nanosuspension gel was clear and presented good homogeneity and the range of pH was found normal. Drug release profile of imiquimod nanosuspension gel was compared with pure imiquimod gel. Comparison of imiquimod

nanogel with pure imiquimod nanogel showed drug release from imiquimod nanosuspension loaded gel was significantly prolonged by using the gelling system due to the addition of the polymer Carbopol 934P. Drug release profile concludes that the mechanism of drug release from imiquimod nanosuspension gel was non-Fickian and it follows Higuchi matrix.

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