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The Creation And Assessment Of Rota-Haler Capsules Containing Solid Lipid Nanoparticles Of The Asthma Medication Terbutaline Sulphate

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Abstract

Objectives: The objective of the present study was to enhance the pulmonary bioavailability of the anti-asthmatic drug Terbutaline Sulphate by incorporating the drug into solid lipid nanoparticles (SLN) and then formulating it as inhaler capsules.
Methods: This required the optimization of process parameters such as the type of solid lipid, drug-to-lipid ratios, and type and concentration of surfactants used in the formulation. SLN were prepared by the combination of two methods: hot homogenization and sonication. The prepared nanoparticles were evaluated for their particle size, zeta potential, entrapment efficacy, surface morphology, and in vitro drug release. The prepared SLN was further formulated as inhaler capsules, and evaluation tests were also conducted for the capsules. Capsules were evaluated for weight variation, content uniformity, and in vitro deposition of drug using a twin-stage impinger.
Key findings: The prepared nanoparticles were spherical in shape and within the size range (50–240 nm), as confirmed by transmission electron microscopy (TEM). SLN showed 75.7% release in a controlled manner for eight hours under the tested conditions. The selected formulation had 71% entrapment efficacy with better flow properties. The present study suggests that SLN of an anti-asthmatic drug provides a stable product with improved drug loading for enhanced bioavailability, targeted action, and sustained release of drugs in the lungs with fewer adverse effects.
Conclusion: The research effectively showed that when made as inhaler capsules, adding terbutaline sulphate to solid lipid nanoparticles (SLN) greatly increases its pulmonary bioavailability. The synthesis of spherical nanoparticles with a size range of 50–240 nm resulted from the optimisation of formulation parameters, proving their appropriateness for inhalation. The SLN demonstrated a good entrapment efficacy of 71% and a regulated release profile of 75.7% drug release over eight hours. According to these results, SLN can be a useful delivery method for medications that treat asthma, offering targeted action and prolonged release in the lungs with a low risk of side effects. This formulation strategy has the potential to enhance asthma management Therapy outcomes.

Keywords: Terbutaline sulphate, Solid lipid nanoparticles, Rota-haler capsules, Pulmonary delivery, Bioavailability, Controlled release, Asthma.

INTRODUCTION

JR. Soriano et al.'s extensive study from 1990 to 2017 found that 544. 9 million individuals globally had persistent respiratory disease, a rise of 39.8% from 1990. After cardiovascular illnesses and neoplasms as the primary causes of mortality in 2017, chronic respiratory disorders accounted for 70% of the total number of deaths. In 2017, 3914196 deaths (95%) were attributable to chronic respiratory disorders, a rise of 18% since 1990[1].

Recently, researchers have paid attention to enhancing the therapeutic efficacies of previously existing active pharmaceutical agents via various targeting approaches[2].Targeted drug delivery refers to concentrating the drug in a targeted area, which is independent of the method used for preparation and the administration route[3]. Targeting the drug to the tissue of interest is the most effective way of increasing the drug's therapeutic index and minimizing the amount of drug deposition in other tissues. Targeted delivery of drugs not only increases the efficacy of the active ingredient but also reduces the side effects that occur due to the deposition of drugs in other tissues[4]. Passive targeting is physiology-based targeting. Active targeting is of various types, which include targeting mediated by external stimuli, targeting in gene therapy, antibody-directed enzyme prodrug therapy, targeted drug delivery towards specific infections like malaria, HIV, or tuberculosis, blood-brain barrier (BBB) targeted delivery, intracellular targeting, and pulmonary targeted drug delivery[5]. The lungs are extremely incredible organs with astonishing anatomy and physiology. The air-conducting zone of the lung consists of the nose, pharynx, larynx, trachea, and bronchi in series. The respiratory zone of the lungs has a cluster of two to five alveolar sacs that open into the lumen of the alveolar duct. Alveoli are airbags that consist of type I and type II alveolar epithelium cells, while macrophages from their cell wall[6]. A pulmonary system for drug delivery is one of the innovative approach for targeting drug to its point of the action. Not only lung diseases, which include asthma, chronic obstructive pulmonary disease, emphysema, cystic fibrosis, and lung infections, can be treated by pulmonary targeting, but anticancer drugs for treating CNS cancer, anaesthetics, analgesics and peptide hormones can also be delivered by this system[7].

SLNs (Solid Lipid Nanoparticles) are a very ensuring approach for the pulmonary drug delivery. They are made up of phospholipids, which are part of the lung surfactant layer; therefore, they are proven to be biocompatible carriers, and hence they do not induce any immune mechanisms[8]. They encapsulate the drug molecule and give a sustained release of the drug, which results in a reduction of dosing frequency and patient compliance[9,10]. Namrata Soni, Neetu Soni et al. concluded that Gemcitabine an anticancer drug molecule, produces enhanced retention in the adenocarcinoma cells when given as mannosylated solid lipid nanoparticles formulation[11].

Dry powder inhalers are one of the most commonly used delivery devices for pulmonary drug administration due to their simple structure and enhanced stability of the active pharmaceutical ingredient in the solid form[12].

Terbutaline Sulphate is a phenylethanolaminesubstituted by 2-(tert-butylamino)-1-hydroxyethyl group at the position 5th. It is a bronchodilator and highly selective agonist with little cardiac side effects[13]. Terbutaline appears tostimulate beta receptors in the bronchial, vascular and uterine smooth muscles more than beta receptors in the heart[14].

MATERIAL AND METHODS

Terbutaline Sulphate was purchased from Amneal Pharmaceuticals Co India Pvt. Ltd., Bavla, Ahmedabad. Glyceryl monostearate (GMS), disodium hydrogen phosphate, sodium chloride, and potassium dihydrogen phosphate were purchased from Loba Chem. Pvt. Ltd., Mumbai. Poloxamer 188 was taken from Yarrow Chem. Pvt. Ltd., Mumbai and soya lecithin from Tokyo Chemical Industry Co. Ltd.All of the rest of the reagents were analytical-grade and used exactly as directed. Deionized, distilled water was used completely.

METHOD FOR THE PREPARATION

SLN has been created by employing the hot homogenization method. The aqueous phase has been formed by mixing 20mg of drug in the distilled water containing an aqueous phase surfactant (poloxamer 188) at the same temperature as the oil phase. Glyceryl Monostearate polymer was accurately weighed in

concentrations 5 to 9% and melted at temperature 5°C over its melting temperature. The heated aqueous phase was added to melted polymer slowly along with constant stirring at high speed and 60°C temperature for 40 minutes to get homogenous dispersion. The prepared dispersion was further subjected to sonication for about 15 minutes to get the homogeneous product. This product was then lyophilized to get the stabilized dry powder.

EVALUATION OF SLN OF TERBUTALINE SULPHATE

Drug entrapment efficacy (EE %) determination[15,16]

One milliliter of each drug loaded sample of SLN dispersion was centrifuged for 40 minutes at 5000 rpm to segregate lipid or aqueous phases. The supernatant was diluted with water and analyzed on U.V. Spectrophotometer at 276nm. The concentration of drug in supernatant liquid was calculated as follows:

$$EE\% = \frac{Wa - Ws}{Wa} \times 100$$

Where,
Wa = Amount of Terbutaline Sulphate added to the formulation
Ws = Analyzed weight of drug in the supernatant

Measurement of particle size

Zeta Sizer was used to calculate the particle size of solid lipid nanoparticles formulation. At 25^0C, aqueous dispersions were diluted to proper scattering intensity and the average hydrodynamic SLN diameter was determined using zeta sizer dynamic light scattering measurements (Malvern instruments)[17,18].

Measurement of zeta potential

The dispersed state of Terbutaline Sulphate loaded SLNs was investigated for the zeta potential utilising a particle size analyzer (Malvern instruments). The measurements were carried out at the 90º angle in an automated mode by keeping the equilibrium time of 120 seconds.

Transmission Electron Microscope (TEM)

The surface morphology of the SLN formulation was investigate by Transmission Electron Microscopy. As compared to the SEM, TEM is a microscopic method for determining the morphology and 3D structure of the product. This method involves passing an electron beam through a thin specimen and once the beam interacts with the specimen, it creates an image on an imaging device that may be seen on photographic film. It is used to determine the physical characteristics of SLN. The sample was stained with Phosphotungstic acid (PTA) 2% for 5 minutes then excess PTA was removed, spread on a gold grid and examined[19].

In- vitro release study of Terbutaline Sulphate from SLN

Investigations through in vitro releases have been conducted employing the Franz diffusion cell containing a dialysis membrane that comprising a 2.4 nm pore size. After being submerged within SILF (simulated interstitial lung fluid), the dialysis membrane within Franz diffusion cell filled at the pH 7.4 over 12 hours. The donor compartment consisted with SLN formulation, whereas the SILF resided within receptor compartment. sample had been taken through receiver compartment via a side tube at periodic intervals. The fresh medium (SILF) was implemented so as to hold the receiver compartment's volume at a constant level. The samples were assessed employing a UV spectrophotometric strategy using 276 nm wavelength[20].

EVALUATION OF TS-SLN LOADED CAPSULES

Weight variation

20 capsules were weighed individually using digital analytical balance, then opened the capsule without losing any part of shell and removed as completely as possible. Weighed the shell, the weight of the contents was the difference between weighing. The entire procedure was repeated with further 19 capsules. Determined the average weight of capsule contents[21,22].

Table 1. IP, BP and phEur limits for uniformity of weight

Average weight of capsule contents (mg)	Percentage deviation (%)
Less than 300	±10%
More than 300	±7.5%

Content uniformity

Total 30 capsules were selected and 10 were assayed individually for the specified potency range provided in standard procedure in I.P. If 9 capsules out of these 10 capsules fall under the 85-110% limit and one capsule fall under 75-125% limit then test will be considered as qualified otherwise test should be performed on 20 more capsules. Out of these 30 capsules 27 must fall within limit of 85-125% to qualify the test.

In vitro deposition study using Twin Stage Impinger

Terbutaline Sulphate formulations were delivered from Rota-haler to twin impinger. The upper (I) and lower (II) impingement stages of the twin impinger filled with 7ml and 30 ml deionized water, respectively. Air was drawn through the device with flow rate of the 60±0.5 L min^{-1}, set at the to the glass throat pre-separator with the DPI in series. The primed device was positioned in an airtight adaptor in the throat and the vacuum pump switched on for 60s. Washings were taken from the device, stage I and stage II, made up the volume and the quantity of Terbutaline Sulphate in each determined by UV analysis at 276nm. The procedure was repeated for a total of four capsules for each formulation[22].

RESULT AND DISCUSSION:

% Entrapment Efficacy

Concentration of Poloxamer 188 highly effects the formulation. Entrapment efficacy of solid lipid nanoparticles increases with increase in the concentration of surfactant.

Table 2. % Drug Entrapment Efficacy of TS loaded SLN

Formulation Code	% Entrapment Efficacy
F1	41.4±1.2%
F2	54.5±1.1%
F3	63±0.9%
F4	**71±0.7%**
F5	69±1.4%

Based on % Entrapment efficacy F4 was selected for the further evaluation.

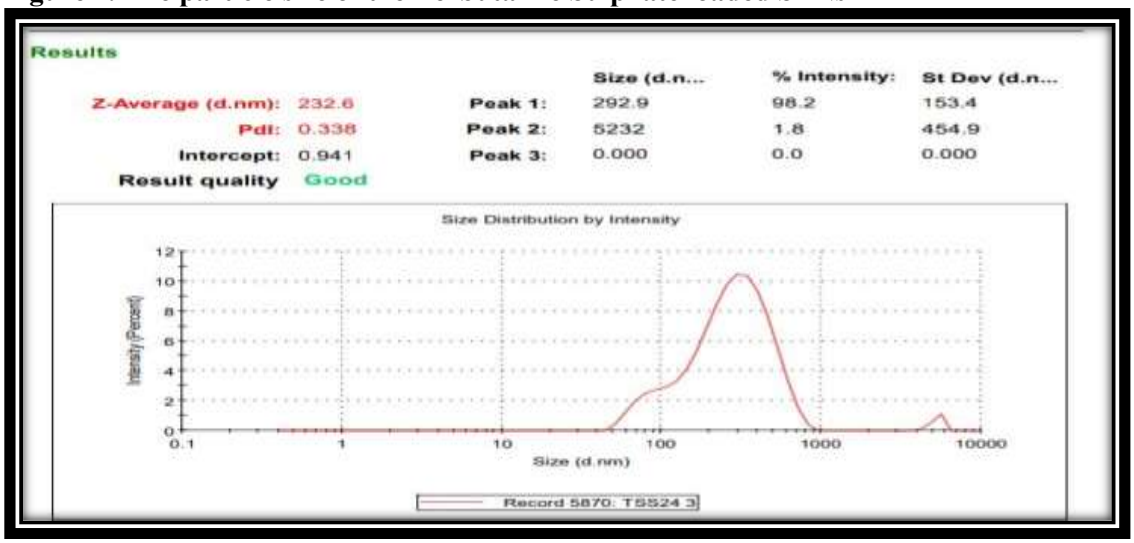
Flow properties

The powder has shown better angle of response which was 26±2.0°C representing its free flow, carr's index of 13.5±0.12 g/cm^3. Hausner's ratio was 1.15 ± 2.31as tapped and bulk densities were 23.24 and 20.10 g/cm^{3}respectively, which proved its acceptability as a carrier.

Measurement of particle size

Terbutaline Sulphate loaded SLN were investigated for the mean size of the and polydispersivity index (PDI) photon correlation using Malvern Zeta Sizer. Sample was suitably diluted with distilled water before examination. The size of the particle and the PDI of the SLN formulation was found be as showed as following.

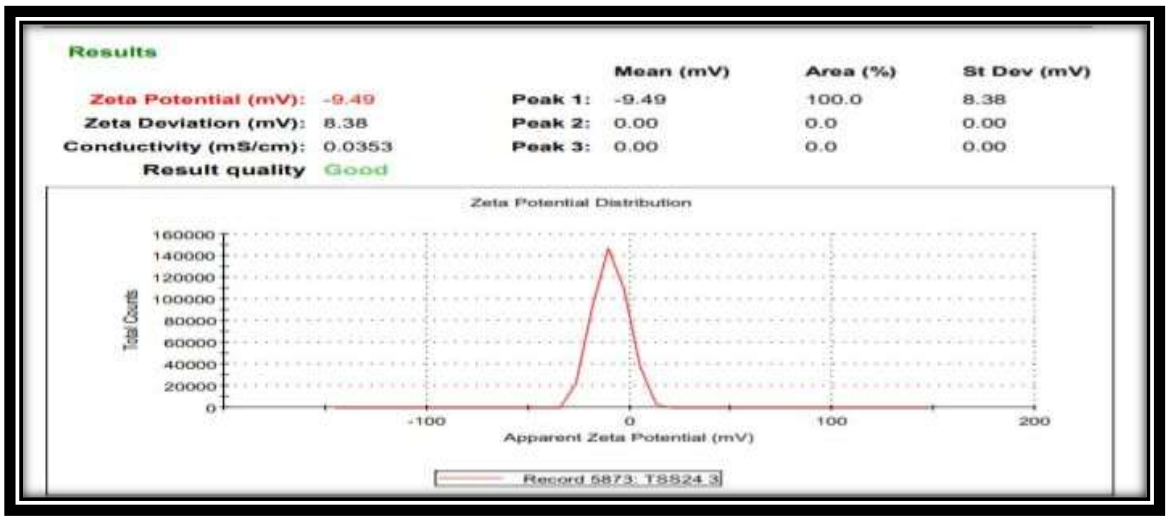
Figure 1. The particle size of the Terbutaline Sulphate loaded SLNs



Measurement of the Zeta Potential

Zeta potential is charge on surface of a particle in a particular liquid media. This value of surface charge is beneficial for figuring out how particles in suspension interact with other particles.. Zeta potential was measured using technique of the electrophoretic light scattering where particle motion was detected in an applied electric field using Malvern Zeta Sizer.

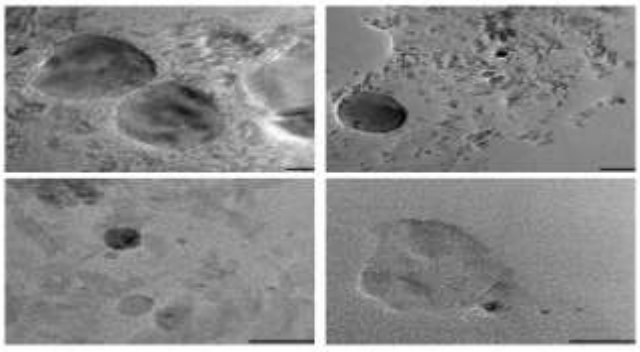
Figure 2. Zeta potential of Terbutaline Sulphate loaded Solid lipid nanoparticles



Transmission Electron Microscopy

Particle morphology was observed using HR-TEM in Punjab University, Chandigarh. A transmission electron microscope with a secondary electron detector was used to obtain digital images of the sample at the accelerating voltage 120 KV. From this, I concluded that the SLN formulation exhibited regular morphology with particle size range betweem10-100nm.

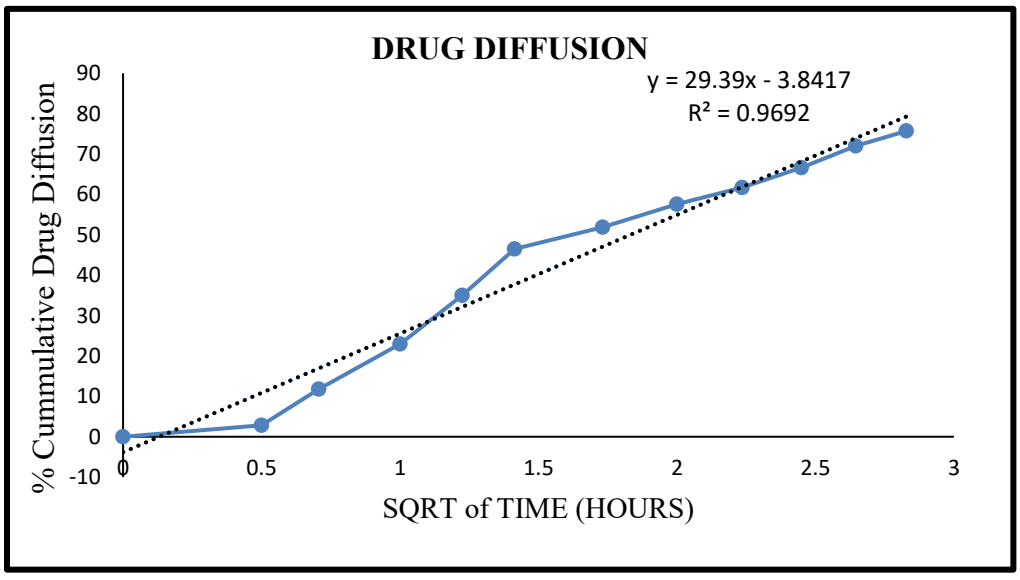
Figure 3. TEM images of Terbutaline Sulphate loaded solid lipid nanoparticles



In- vitro Diffusion Studies

In vitro diffusion of drug SLNs was performed at 100 rpm and 37±0.5℃ in SILF (Simualted Interstitial Lung Fluid), pH 7.4. When the cumulative drug diffusion of TS-SLN and a conventional drug were evaluated, SLN demonstrated improved release over time. Thus, it was determined that solid lipid nanoparticles had better drug diffusion than the standard drug, which lacked lipid. In vitro diffusion curves of Drug-loaded nanoparticles revealed a biphasic drug release pattern, with drug burst release at the beginning and sustained release at a constant rate thereafter. Initially, the drug embedded in the surfactant layer was released and showed burst release; after that, the drug diffused slowly from the polymer coating.

Figure 4. in vitro drug release of Solid lipid nanoparticles



Weight variation: Test was performed by weighing 20 capsules individually. Determined the average weight of capsule contents.

Table 3. Data of weight variation test

S.no.	Formulation code	Weight of content of individual capsule	% deviation
1	C1	101.2	-0.14
2	C2	102.0	0.64
3	C3	101.8	0.44
4	C4	101.3	-0.04
5	C5	97.8	-3.5
6	C6	99.7	-1.62
7	C7	102.4	1.03
8	C8	101.2	-0.14
9	C9	98.9	-2.4
10	C10	102.2	0.83
11	C11	103.3	1.92
12	C12	101.5	0.14
13	C13	102.0	0.64
14	C14	102.6	1.23
15	C15	101.3	-0.04
16	C16	98.6	-2.7
17	C17	103.4	2
18	C18	101.6	0.24
19	C19	103.2	1.82
20	C20	101.0	-0.34

Average weight of capsule = 100.96
Upper limit (% deviation) = 1.92
Lower limit (% deviation) = -2.7

Content Uniformity

Table 4. Data of Content Uniformity Test

S.no.	Formulation code	Labelled amount of drug (µg) (x)	Actual amount of drug in each capsule (µg) (y)	% drug content (y/x)*100
1	C1	1000	964	96.4
2	C2	1000	963	96.3
3	C3	1000	966	96.6
4	C4	1000	969	96.9
5	C5	1000	972	97.2
6	C6	1000	971	97.1
7	C7	1000	965	96.5

8	C8	1000	959	95.9
9	C9	1000	967	96.7
10	C10	1000	963	96.3

In vitro deposition study using twin stage impinger

The quantity of the deposited in second stage of the impinger was considered as a fine particle dose (FPD). The recovered dose (RD) is the quantity of the drug present in stage 1 and the stage 2 of the impinger, inhaler device and the capsule shell. Respirable fraction (RF) was the ratio of FPD and RD and expressed in the percentage.

Table 5. Data of in vitro deposition study

Data	Values
Fine particle dose (FPD)	56.21±0.02µg
Recorded Dose (RD)	109.02±0.03µg
Respirable Fraction	51.55±0.02%

Data expressed as mean ± SD (n=3)

The respirable fraction of the F4 formulation was 51.550.02% as comparable to 45.32±0.05% for commercial DPI.

SUMMARY

Terbutaline sulphate-loaded solid lipid nanoparticles were effectively formulated. Following the final formulation's characterisation, it was determined that the product's improved flow characteristics allowed it to improve medication deposition in the lungs. Reduced dosage and increased anti-asthmatic action of the Rota-haler of Terbutaline Sulphate loaded Solid Lipid Nanoparticles helped to mitigate dose associated side effects.

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