Journal of Pharmaceutical Research International

33(46A): 329-341, 2021; Article no.JPRI.75554
ISSN: 2456-9119
(Past name: British Journal of Pharmaceutical Research, Past ISSN: 2231-2919, NLM ID: 101631759)

Formulation, Optimization and Pharmacodynamic Studies of Pioglitazone HCl Solid Lipid Nanoparticles

Y. Indira Muzib[1*], E. Ramya[1] and Y. R. Ambedkar[2]

[1]Institute of Pharmaceutical Technology, Sri Padmavati Mahila Visvavidyalayam Tirupati, Andhra Pradesh-517502, India.
[2]S. V. College of Veterniary Sciences, Garvidu, Vizayanagaram, Andhra Pradesh, India.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JPRI/2021/v33i46A32873
Editor(s):
(1) Dr. Rafik Karaman, Al-Quds University, Palestine.
Reviewers:
(1) Zeuko'o Menkem Elisabeth, University of Buea, Cameroon.
(2) M. Jayanthi, JIPMER, India.
Complete Peer review History: https://www.sdiarticle4.com/review-history/75554

Original Research Article

Received 06 August 2021
Accepted 13 October 2021
Published 14 October 2021

ABSTRACT

Pioglitazone HCl is an oral anti-diabetic agent used for the treatment of diabetes mellitus type II. The aim of the present work is to evaluate the pharmacodynamic activity of solid lipid nanoparticles of pioglitazone HCL prepared by using solvent injection technique and to compare with the control and test group. Among all the formulations, F5 was found to possess highest *in-vitro* drug release within 24 hrs i.e., 95.02±1.26%. The *in vivo* studies were performed using male albino rats of wistar strain (150-200g). Rats were divided in to five groups (n=6), group-I normal, group-II diabetes control, group-III placebo control, group-IV reference, group-V test group. Diabetes was induced by streptazocin (60 mg/kg) by intraperitonial route. The reference group was treated with marketed tablet of pioglitazone HCL, test groups were treated with SLNs suspended in 0.1% Tween 80 and given to animals through oral gavages. Blood samples were collected by retro-orbital puncture before treatment, and after treatment at time intervals 0, 2, 4, 6, 8, 10, 12 and 24h in anti-coagulated vials. Parameters like glucose, tri glycerides (TG), total cholesterol (TC) and HDL-C were estimated by calorimetric method. Diabetes induced rats showed elevated levels of glucose, TG, TC and reduced HDL. The oral administration of drug loaded SLNs in 0.1% Tween 80 solution

**Corresponding author: E-mail: yindira1415@gmail.com;*

showed reduced levels of glucose, TG and elevated levels of HDL-C and slightly reduced levels of TG in 24 h where as the marketed tablet showed reduced levels of glucose, TG and TC up-to 12 h and in 24thh the glucose levels get elevated. Thus the optimized SLNs showed prolonged activity.

Keywords: Solid lipid nanoparticles; pioglitazone Hcl; test group; reference group; glucose. in vivo pharmacodynamics studies.

1. INTRODUCTION

Carrier based delivery of a drug molecule to specific organ sites is one of the major thrust area in pharmaceutical sciences. So, a high potential for drug delivery has been attributed to particulate drug carriers, especially small particles such as microparticles and colloidal system in nanometer range [1]. Nanotechnology, as defined by the National Nanotechnology Initiative (NNI), is the study and use of structures roughly in the size range of 1 to 100nm. The goal of nanotechnology is the same as that of medicine, to diagnose as accurately and early as possible and to treat as effectively as possible without any side effects using controlled and targeted drug delivery approach [2]. The efficient drug delivery depends on their ability to penetrate through several anatomical barriers, sustained release of their contents and their stability in the nanometer size. However, the scarcity of biocompatible polymers with regulatory approval and their high cost have limited the wide spread application of nanoparticles to clinical medicine [3].To overcome these limitations of polymeric nanoparticles, lipids have been put forward as an alternative carrier, particularly for lipophilic pharmaceuticals. These lipid nanoparticles are known as Solid Lipid Nanoparticles(SLNs) [4]. Pioglitazone HCl is an oral anti-diabetic agent used for the treatment of diabetes mellitus type II [5]. It is the drug of choice for the preparation of SLNs since it is highly lipophilic in nature. It is formulated in the form of SLNs to increase the aqueous solubility and for sustained release over a prolonged period of time to maintain the glucose levels by preventing the hyperglycemia [6].

The present work aimed to prepare SLNs using ethyl cellulose as polymer, cholesterol as solid lipid, tween80 and span 60 as surfactants. The formulated SLNs were subjected to evaluation tests like; entrapment efficiency and *in vitro* diffusion studies and characterized by using Scanning electron microscopy (SEM), particle size by zeta seizer and polymer compatibility studies were done by using FT-IR and DSC. The optimized formulation was subjected to in *vivo* pharmacodynamics activity.

2. MATERIALS AND METHODS

2.1 Materials

Pioglitazone hydrochloride was gift sample received from Hetero drugs Pvt.Ltd, Cholesterol and Ethyl Cellulose were purchased from SD Fine–chem. Laboratories. Tween 80 and Span 60 were purchased from Merck, Mumbai, India. All the reagents used were of analytical grade.

2.2 Methods

2.2.1 Preparation of Pioglitazone hydrochloride loaded Solid lipid nanoparticles

Pioglitazone HCl Solid lipid nanoparticles were prepared by solvent injection method [7]. An accurately weighed amount of Pioglitazone hydrochloride (30 mg), cholesterol (solid lipid), ethyl cellulose (polymer) and span 60 (surfactant) were dissolved in methanol and heated upto 60±2°C (Table 1). The aqueous phase was prepared by dissolving Tween 80 in distilled by stirring which is maintained at same temperature. The organic phase containing lipid mixture was added drop wise with stirring to aqueous phase with the help of hypodermic needle. The mixture was then sonicated for 15 min to get nanoemulsion and it is lyophilized for 48h to obtain the freeze dried SLNs.

2.2.2 Characterization of formulations

Entrapment efficiency: The freeze dried SLNs were dissolved in methanol under water bath at 65 °C for 30 min and then cooled to room temperature to preferentially precipitate the lipid [8]. The solid lipid nanoparticles along with encapsulated drug remained at the bottom of centrifuge tube and the unentrapped drug is remained in the upper supernatant layer after centrifugation at 4000 rpm for 15 min. The

amount of drug in the supernatant layer as analyzed by using UV/Visible spectrophotometer method and subtracted from the total drug content to get the amount of entrapped drug. The drug entrapment efficiencies were calculated from Equation

$$\% \text{ Entrapment efficiency} = \frac{\text{Total amount of drug} - \text{Free drug}}{\text{Total amount of drug}} \times 100$$

2.2.3 Drug-polymer compatibility studies:

a) FT-IR spectroscopy: This study was carried to find out the compatibility between drug (Pioglitazone HCl), polymer (Ethyl cellulose). Sample and KBR were taken in the ratio of 1:100 in a mortar and triturated. A small amount of triturate was taken into a pellet maker and was compressed at 10 kg/cm^2 to form a transparent pellet using a hydraulic press. The pellet was kept in a sample holder and scanned from 4000 cm^{-1} to 400 cm^{-1} in FT-IR spectrophotometer [9]. The possible interaction between Pioglitazone HCl and Ethyl cellulose was accessed by comparing FTIR spectra of pure drug, polymer and nanoparticle formulation.

b) ***Differential scanning calorimeter (DSC)****:* DSC studies were performed to understand the behavior of ethyl cellulose on application of thermal energy. DSC was performed on a DSC (METTLER STAR˚ SW 8.10) at a heating rate of 10°C/min in the temperature range of 0-250°C using empty aluminum pan as reference standard [10].

2.2.4Determination of particle surface morphology by SEM

The morphological characteristics of nanoparticles were determined by scanning electronic microscopic (SEM, Zeiss Ultra-60 (FE SEM)). Specified quantity of nanoparticles were taken and mounted directly on the SEM stub, using double sided, sticking tape and scanned in a high vacuum chamber with a focused electron beam.

2.2.5 Particle size determination:

The average particle sizes, polydispersity index and zeta potential of the lipid particulate dispersions were determined using a Zetasizer (Horiba SZ-100). The sample of dispersion was diluted to 1:9 v/v with double distilled water to ensure that the light scattering intensity was within the instrument's sensitivity range. Double distilled water was filtered through 0.45µm membrane filters prior to particle size determination.

2.2.6 *In-vitro* drug release from SLNs

The *in vitro* drug release studies were performed using the USP paddle method. The dissolution study was carried out in two dissolution media; 0.1N HCl pH 1.2, and phosphate buffer pH 6.8 and the entire system was kept at 37°±0.5° with continuous agitation speed of 50rpm [11,12]. At appropriate time intervals 5ml of release medium was removed and 5ml fresh medium was added into the system to maintain sink condition. The amount of pioglitazone in the release medium was evaluated by U.V Spectrophotometer at 269nm. The concentration of pioglitazone HCl in test samples was corrected and calculated by using the regression equation of the calibration curve.

2.2.7 Kinetic Modeling of Drug Release:

Data obtained from *in-vitro* studies were fitted to various kinetic models such as zero order, first order, Higuchi model and korsmeyer-peppas model [13].

Table 1. Composition of various formulations

S. No.	Name of the ingredients	F1	F2	F3	F4	F5	F6	F7	F8
1.	Pioglitazone hydrochloride (mg)	30	30	30	30	30	30	30	30
2.	Cholesterol (mg)	20	20	20	20	20	20	20	20
3.	Ethyl Cellulose (mg)	40	40	80	40	120	120	200	200
4.	Span 60 (mg)	20	20	20	20	20	20	20	20
5.	Tween 80 (ml)	0.53	1.3	0.53	-	0.53	1.3	0.53	1.3
6.	Methanol (ml)	3.95	3.95	3.95	3.95	3.95	3.95	3.95	3.95
7.	Distilled water (ml)	50	50	50	50	50	50	50	50

2.2.8 *In vivo*-Pharmacodynamic Studies

The drug loaded Solid lipid nanoparticles were optimized based on the performance of *in vitro* tests. The optimized drug loaded formulation was used to study *in vivo* glucose and cholesterol reducing activity on male albino rats according to the protocol (Table 1). Wistar strain of male albino rats weighing 150-200 g were used in the study (n=6). The animals were housed in standard polypropylene cages and maintained under controlled room temperature and humidity with 12h light/dark cycle [14]. All the rats were fed with commercially available rat normal pellet diet (NPD) and water and libitum, prior to the dietary manipulation. The experimental protocol was designed and approved by the Institutional Animal Ethical Committee (CPCSEA/1677/PO/Re/S /IAEC-04/2016)

Diabetes was induced by inducing Streptozocin (STZ) (60 mg/kg) by intra peritoneal route except for normal group [14]. To confirm the induction of hyperglycemia, after 24h blood samples were collected from the retro-orbital puncture. The glucose and cholesterol concentration of the blood samples was then determined using a standard enzymatic assay kit. The rats were then divided into 5 groups of 6 animals [15,16].

The blank, reference, test groups were treated with SLNs suspended in 0.1% Tween 80 and given to animals through oral gavages. Blood samples were collected by retro-orbital puncture at predetermined time intervals, viz., before treatment, and after 0, 2, 4, 6, 8, 10, 12 and 24h in anticoagulant filled vials. Plasma was separated by centrifugation at 1000rpm for 15min. The serum was quantitatively analyzed for blood glucose, triglycerides, cholesterol and HDL using calorimeter.

2.2.9 Determination of Blood Glucose:

Blood glucose levels was determined by using glucose estimation kit GOD-POD method [17].

Calculation:

Total glucose (mg/dl) **=**

$$\frac{Absorbance\ of\ Test}{Absorbance\ of\ standard} \times 100$$

Determination of Triglycerides: Triglycerides levels was determined by using the kit GPO-POD method [16].

Calculation: Triglycerides (mg/dl) **=**

$$\frac{Absorbance\ of\ Test}{Absorbance\ of\ Standard} \times 200$$

Total Cholesterol and HDL-C: The cholesterol levels were calculated by using the kit CHOD-POD method.

Calculation:

Total cholesterol (mg/dl) =

$$\frac{Absorbance\ of\ Test}{Absorbance\ of\ Standard} \times 100$$

2.3 Statistical Analysis

The data obtained from the entrapment efficiency and *in-vitro* drug release of Pioglitazone HCl SLNs were statistically analyzed with one way ANOVA ($p<0.05$) using Graph pad prism 5.0.

3. RESULTS AND DISCUSSION

Pioglitazone loaded SLNs were subjected to know the effect of lipid, polymer and surfactant of the formulation on entrapment efficiency and *in-vitro* drug release to optimize the best formulation. The optimized formulation was characterized by SEM, particle size, drug-excipient compatibility studies.

3.1 Entrapment Efficiency

Among all the prepared SLNs, entrapment efficiency (EE) of Pioglitazone HCl -SLNs sample F5 was found to be 95.39% (Table 3).High incorporation was possible because lipid (cholesterol) increased the solubility of Pioglitazone HCl and span 60 as a lipid surfactant helps to solubilise the drug in to lipid which further increased entrapment of drug. The EE mainly depended on the varied concentrations of polymer (ethyl cellulose) and Tween 80 which is used as an aqueous surfactant. On increasing the amount of ethyl cellulose up to 120 mg, the EE was increased and on further increase the EE get decreased, the probable cause of this behaviour was drug expulsion from the surface of SLNs.

3.2 Particle Size and Zeta Potential

The mean particle size and zeta potential of optimized formulation was found to be 34.5±7.7 nm and -16.9 mV respectively and was shown in (Figs. 2 and 3). Particle size was found to be nanometric size with PDI lesser than 1 and zeta

potential of -16.9mV indicating the stability of the formulation.

3.3 Drug- polymer Interaction Study

The interaction study between the drug (Pioglitazone HCl) and drug loaded Solid Lipid Nanoparticles were evaluated using ATR-FT-IR and DSC analysis.

Attenuated total reflectance – Fourier transform infrared (ATR-FTIR)

The possible interactions between the drug and the excipients were studied by IR spectroscopy for pure drug pioglitazone HCl in (Fig. 4). The FT-IR spectra of polymer and their physical mixtures with drug were shown in (Fig. 5). For optimized formulation (F5) of SLNs all peaks which have been obtained as of the pure drug in the IR spectrum shown in (Fig. 6). From the physical mixtures of drug, lipid, and polymer there were no major shifting as well as no loss of any functional peaks between the spectra of drug and physical mixtures as shown in (Tables 4-6). Hence it was confirmed that there was no interaction between the drug and polymer. This finding was further supported by DSC studies.

Table 2. Experimental design for *In vivo* studies

S.no	Group no.	Purpose	Nature of contents used
1	I	Normal control	Normal pellet diet (NPD)
2	II	Diabetes control	Streptozocin (STZ)(60 mg/kg) induced rats
3	III	Placebo control	STZ with blank SLNs
4	IV	Reference	STZ with PIOGLIT (Marketed conventional tablet)
5	V	Test group	STZ with optimised formulation

Table 3. Entrapment efficiencies of different formulations of SLNs

S.No.	Formulation code	% Entrapment efficiency Mean±S.D. (n=3)
1.	F1	90.21±0.23
2.	F2	85.03±0.41
3.	F3	89.12±0.14
4.	F4	61.54±0.46
5.	F5	95.39±0.26
6.	F6	80.49±0.08
7.	F7	86.98±0.25
8.	F8	75.64±0.54

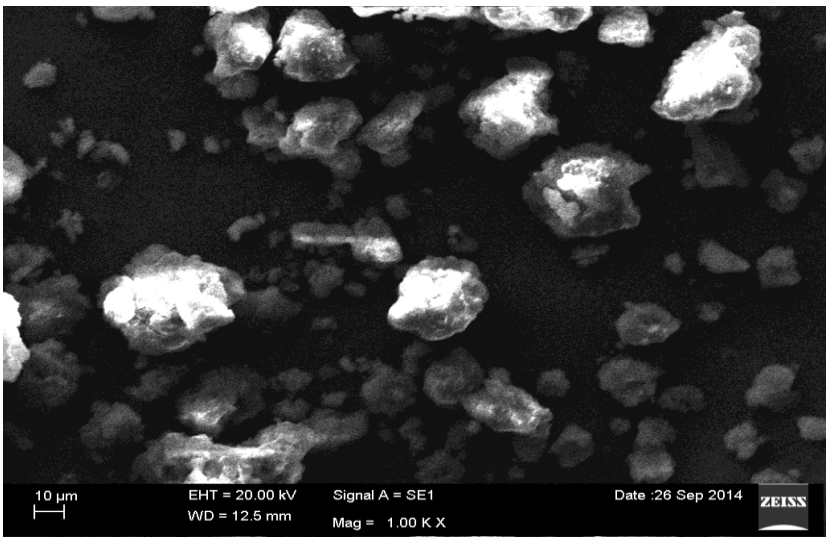

Fig. 1. Scanning Electron Microscopy of Solid Lipid Nanoparticles

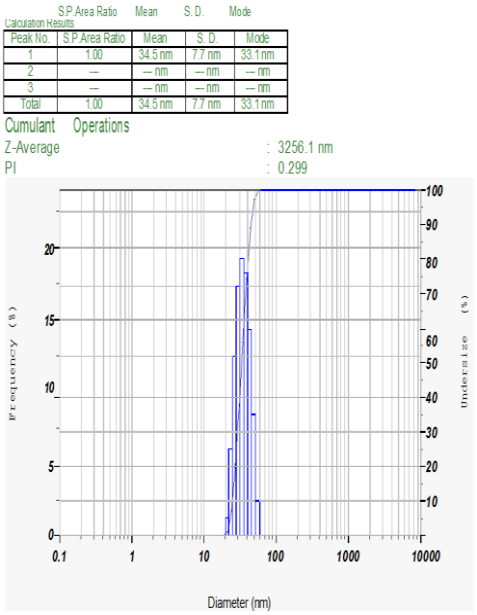

Fig. 2. Particle size of the optimized SLNs

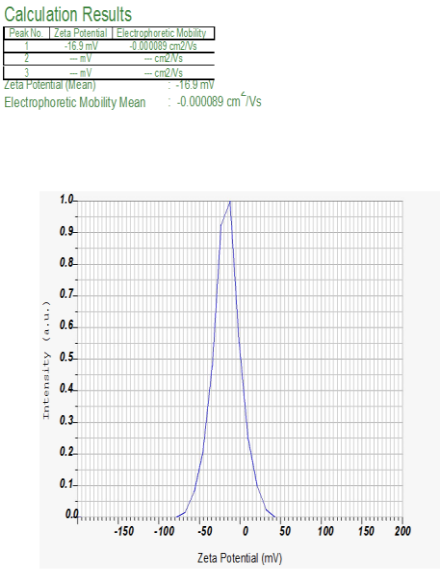

Fig. 3. Zeta potential of the optimized SLNs

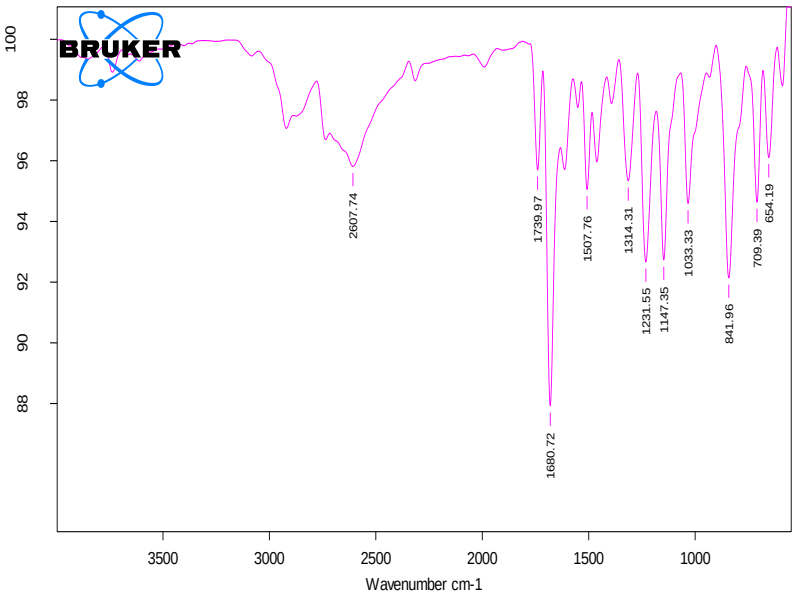

Fig. 4. FTIR spectrum of Pioglitazone HCl pure drug

Table 4. Functional Group assignment of ATR-FTIR of Pioglitazone HCl

Wave No. (cm^{-1})	Functional group
1773.97	amide C = O stretching
1680.72	C=O in amide
1231.55	C–O stretching
1507.76	C=C stretching
841.96	paradisubstituted aromatic ring
2607.74	aliphatic C-H stretching asymmetric
1033.33	aliphatic C-O-C

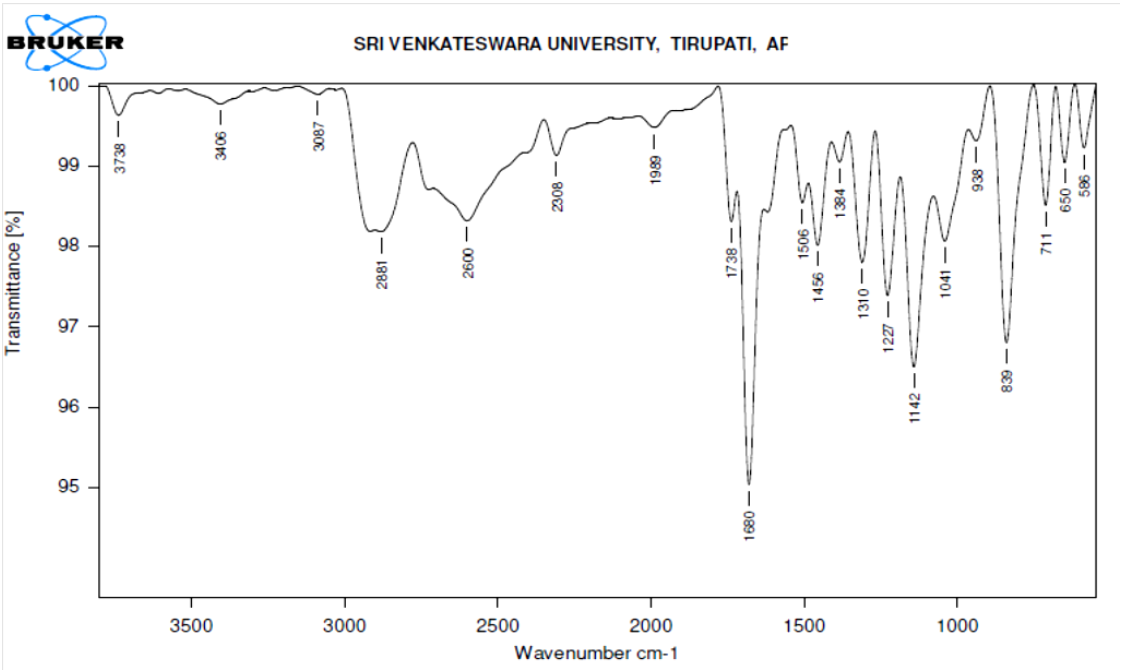

Fig. 5. FTIR spectrum of physical mixtures of polymer and Pioglitazone HCl

Table 5. Functional Group assignment of ATR-FTIR of physical mixtures of polymer and Pioglitazone HCl

Wave No. (cm^{-1})	Functional group
1738	amide C = O stretching
1680	C=O in amide
1227	C–O stretching
1506	C=C stretching
839	paradisubstituted aromatic ring
2600	aliphatic C-H stretching asymmetric
1041	aliphatic C-O-C

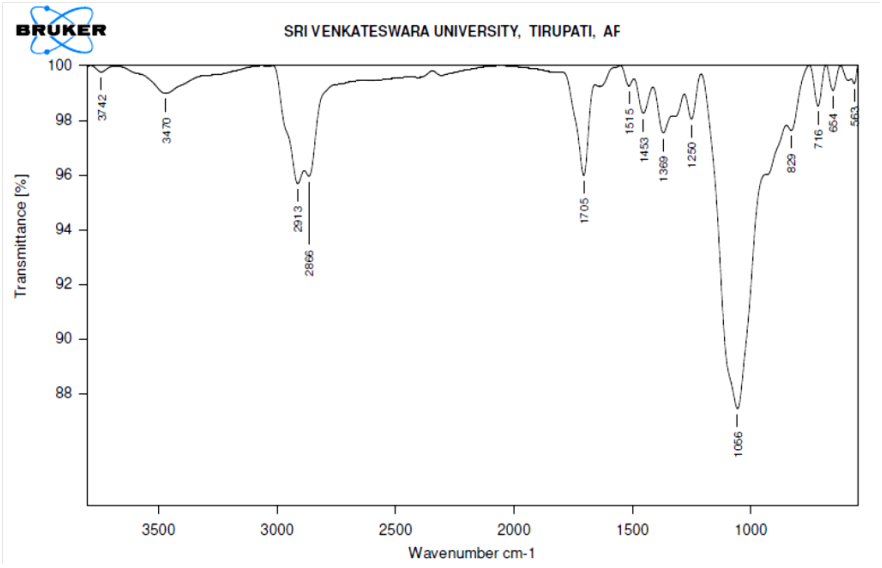

Fig. 6. FTIR spectrum of Pioglitazone HCl loaded Solid lipid nanoparticles

Table 6. Functional Group assignment of ATR-FTIR of SLNs

Wave No. (cm^{-1})	Functional group
829	Para disubstituted aromatic ring
1056	aliphatic C-O-C)
1250	C–O stretching
1515	C=C stretching
2913	aliphatic C-H stretching asymmetric
1705	amide C = O stretching

3.4 Differential Scanning Calorimeter

The differential scanning calorimeter was carried out for the pioglitazone HCl SLNs, to study the compatibility or any interaction of drug and polymer after the formation of SLNs and is shown in (Figs. 7 and 8). DSC thermogram of pure drug exhibited a sharp endothermic peak at 190.84°C corresponding to its melting point and the peak obtained for the pioglitazone HCl SLNs is at 186.9°C for the drug. Thus there was no significant change in the position of peak of the drug in the Pioglitazone HCl SLNs, but there is change in the relative intensities of the peak. It may be due to reduced drug crystallinity and also indicates that the drug is only physically entrapped in the polymer matrix and there is no interaction between drug and polymers.

3.5 In-vitro Dissolution of Formulated Solid Lipid Nanoparticles

The *in vitro* release results of Pioglitazone HCl loaded SLNs formulations are shown in (Fig. 9).In vitro drug release study of Pioglitazone HCl loaded SLNs showed the sustained release behaviour. Almost all formulation dissolution profiles showed biphasic behaviour consisting of initial burst release followed by a slow release phase.Among all the formulations, F5 was found to possess highest *in-vitro* % drug release within 24h i.e., 95.02%. The release profiles of these SLN resemble the drug enriched core model. The initial burst release can be attributed to the presence of free drug in the external phase and drug adsorbed on the surface of particles while the slow release was due to drug encapsulated within lipid matrix and also an outer phase of increased viscosity. The increase in polymer concentration had significant effect on the drug release which prolonged the release from SLNs [17,18]. The increase in amount of polymer concentration appeared to substantially decrease the release in all formulations. This may be attributed to the slower rate of drug diffusion from these SLNs into the dissolution mediums due to increased viscosity of the polymeric outer membrane. Thus, with the use of the Pioglitazone HCl loaded SLNs, it is possible to achieve the loading dose of the drug due to initial burst release and followed by maintenance dose due to the sustained release.

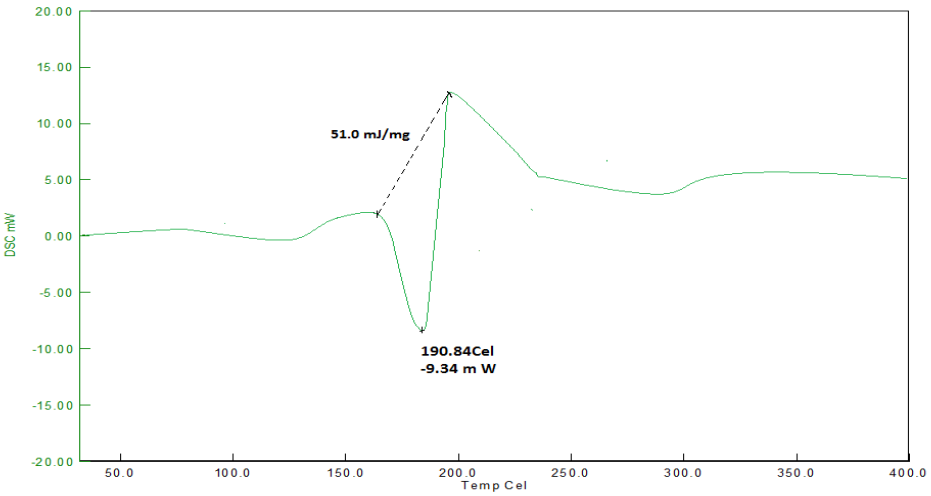

Fig. 7. DSC thermograms of Pioglitazone HCl

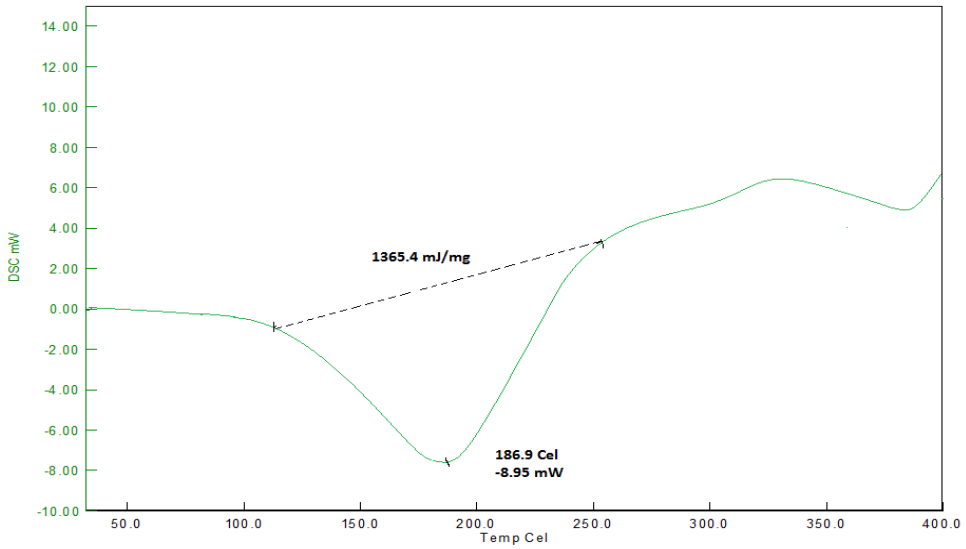

Fig. 8. DSC thermograms of Pioglitazone HCl loaded Solid lipid nanoparticles

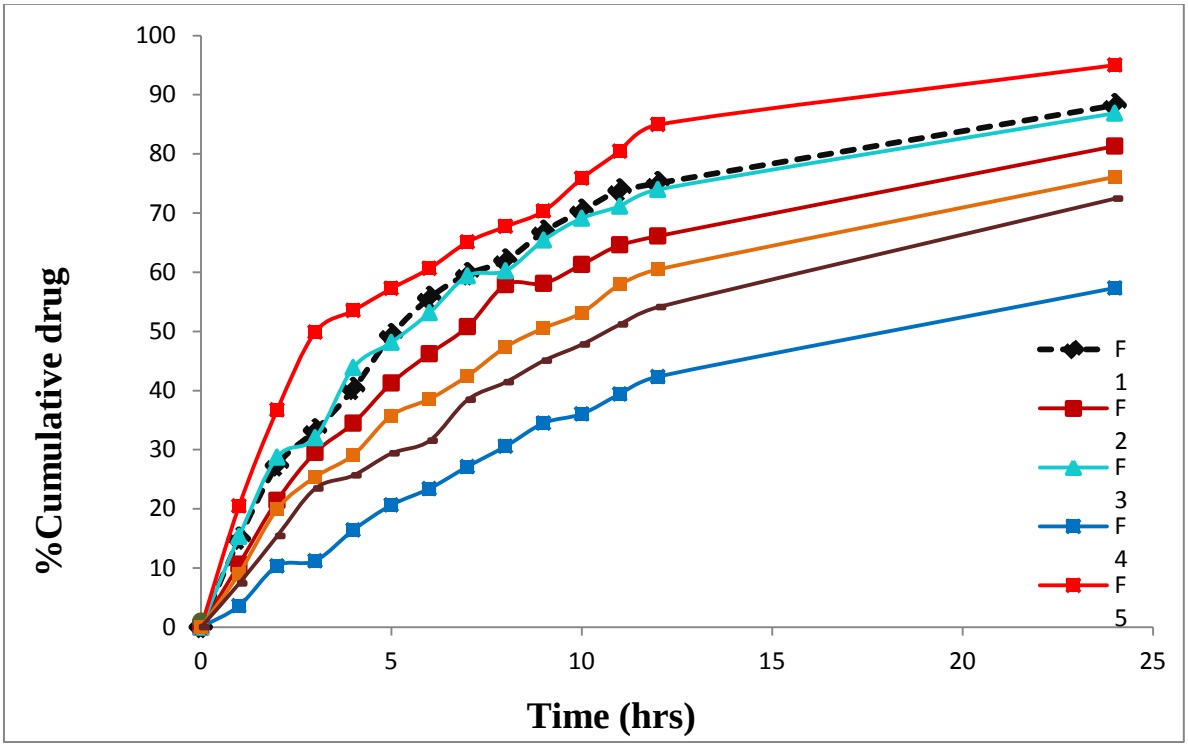

Fig. 9. *In-vitro* dissolution profile of pioglitazone HCL solid lipid nanoparticles

Therefore, F5 formulation was considered as the optimized formulation based on entrapment efficiency and *in-vitro* drug release.

3.6 Kinetic Modeling of Drug Release

The data was analyzed by zero order, first order, Higuchi and Peppas equation models, their plots and 'R^{2}' values were shown in (Table 7). When the release data were analyzed as per zero and first order models, the 'r' values were relatively higher in first order model with all the formulations of SLNs indicating that the drug release from all these formulations followed first order kinetics. Pioglitazone HCl release data also obeyed Higuchi and peppas equation models with '$R^{2,}$' values greater than 0.943. When percent release was plotted against √time, linear

regressions with 'R^{2}' > 0.943 were observed with all the SLNs formulations prepared indicating that the drug release from all these formulations was diffusion controlledfrom the pores on the particle surface. As per peppas equation, the release exponent 'n' was found in the range 0.4693 to 0.8801 indicating non-fickian diffusion as the release mechanism from all the SLNs formulations.

3.7 In-vivo -Pharmacodynamic Studies

The Pharmacodyanamic studies were carried in male wistar albino rats with F5 formulation by comparing placebo,reference and test formulations**.** The Glucose, TC, TG and HDL-C were estimated in plasma samples of different groups as given in (Figs. 10- 13).

Table 7. Kinetics of Drug Release of pioglitazone HCL solid lipid nanoparticles

Formulations Code	Zero order R^2 value	First order R^2 value	Higuchi's R^2 value	KorsmeyerPeppas	
				R^2 value	n value
F1	0.7769	0.9977	0.9656	0.9598	0.5875
F2	0.8068	0.9955	0.9653	0.9572	0.6497
F3	0.7850	0.9940	0.9630	0.9590	0.5578
F4	0.9070	0.9857	0.9641	0.9693	0.8801
F5	0.7728	0.9918	0.9432	0.9383	0.4693
F6	0.8649	0.9948	0.9668	0.9549	0.6141
F7	0.8044	0.9807	0.9804	0.9684	0.6645
F8	0.9007	0.9805	0.9827	0.9736	0.7104

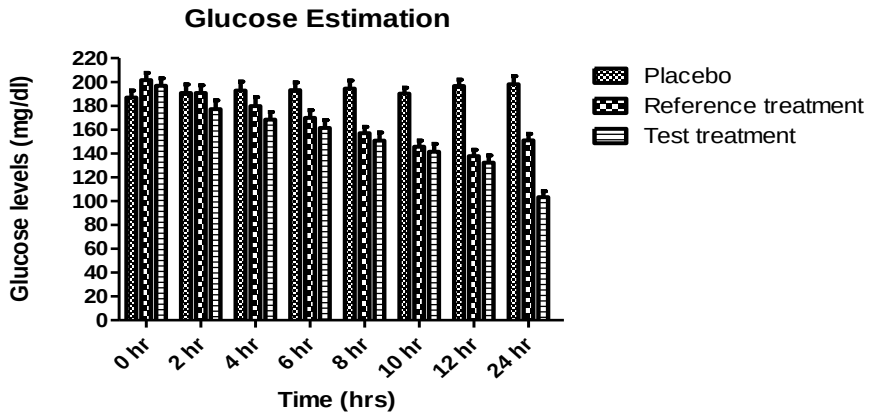

Fig. 10. Glucose levels (mg/dl)

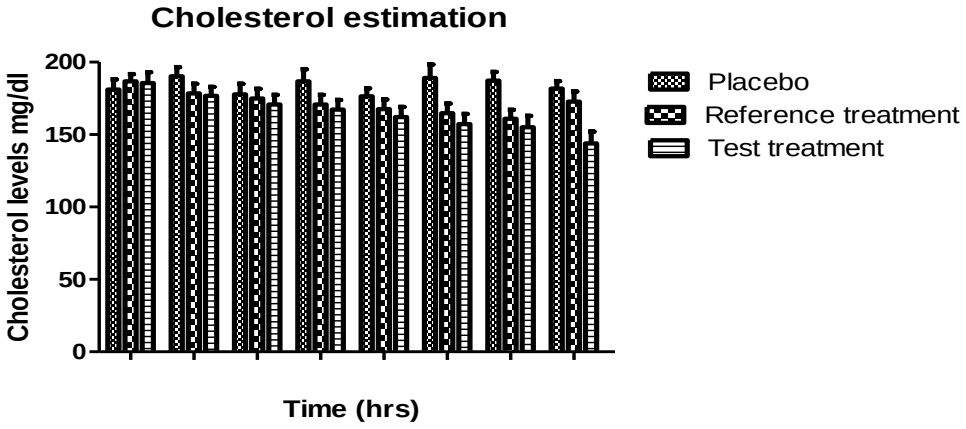

Fig. 11. Total Cholesterol (TC) (mg/dl)

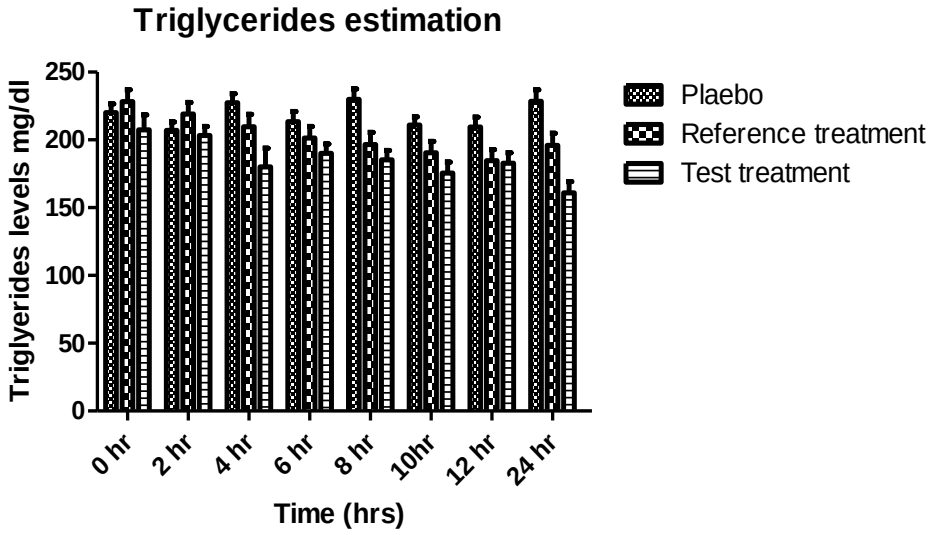

Fig. 12. Triglycerides (TG) (mg/dl)

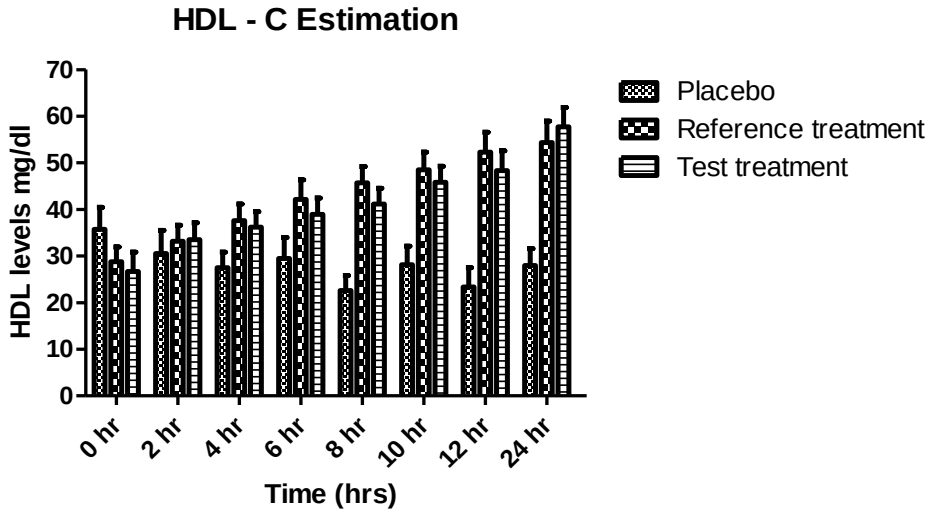

Fig. 13. HDL-C (mg/dl)

Hypoglycemic activity of pioglitazone HCl causes reduction in plasma glucose levels and this action is due to increase in both insulin-stimulated glucose uptake in peripheral tissues and insulin sensitivity in hepatic and adipose tissues, thereby lowering plasma glucose [19]. At the same time, pioglitazone HCl SLNs also shows beneficial effects on lipid metabolism by significant reduction in TG levels and elevation of the HDL-C levels and also slight decrease in TC levels.

In the present Study, diabetes induced rats showed elevated levels of glucose, TG, TC and reduced HDL. The oral administration of drug loaded SLNs in 0.1% Tween 80 solution showed reduced levels of glucose, TG and elevated levels of HDL-C and slightly reduced levels of TG in 24 h where as the marketed tablet showed reduced levels of glucose, TG and TC up-to 12 h and in 24^{th}hr the glucose levels get elevated. Thus the optimized SLNs showed prolonged activity.

On the other hand, the diabetic rats treated with blank SLNs showed equivalent levels as in diabetic group i.e., no reduction in glucose, TG and TC levels. So, the results proved that the SLNs without drug didn't show any possible effects.

The statistically significant (P<0.05) decrease in glucose, TG, TC and elevated levels of HDL-C levels were found in reference and test groups indicating that the Pioglitazone HCL was released from conventional marketed tablet and prepared SLNs and showed hypoglycemic activity.

Though there was no significant difference in Pharmacodyanamic effects with reference and test formulation upto 12 hrs, but on prolonged use possess more advantages with extended release drug delivery system compared to the conventional release drug formulation in respect to dose, side effects etc.

These results indicated that the optimized formulation (F5) could function as a prolonged release hypoglycemic drug delivery system to prevent hyperglycemia by lowering the plasma glucose levels for extended period of time without maintenance dose.

4. CONCLUSION

Pioglitazone HCl SLNs successfully optimized to achieve increased solubility of Pioglitazone as well as the higher and sustained release. The obtained SLNs showed nanosized homogeneous particles with high entrapment efficiency and low recrystallization properties. The *in-vivo* studies showed the significant decrease in glucose levels on oral administration of Pioglitazone HCl SLNs for 24 hrs. This work facilitates the use of SLNs as a novel delivery system to enhance the solubility of pioglitazone HCl and could be effective in sustaining drug release for a prolonged period. The SLNs acts as efficient carrier for pioglitazone HCl for an antidiabetic activity that can increase the therapeutic efficacy of pioglitazone HCl in the oral treatment of type II diabetes. *In-vivo* experimentation was conducted for the optimized formulation and the levels of glucose and lipid profile (triglycerides, total cholesterol, HDL) were estimated. From the results it was found that there was decrease in the levels of glucose which proved that nanoparticles through oral route released the drug up to 24 h and showed the hypoglycemic activity. This could be due to slow release of drug from lipid particles and maintained peak plasma concentration over a prolonged period.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT

It is not applicable.

ETHICAL APPROVAL

As per university standard written ethical approval has been collected and preserved by the author(s).

ACKNOWLEDGEMENT

Authors of this research articles are thankful to Institue of Pharmaceutical Technology,Sri Padmavati Mahilavisva Vidyalayam, Tirupati, DST PURSE centre Sri Venkateswara University, Tirupati for providing FTIR,Zeta sizer, SEM facilities.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. The Royal Society. Nanoscience and nanotechnologies: Opportunities and uncertainties, London: Royal Society. 2004;4.
2. Mishra B, Bhavesh B Patel, Sanjay Tiwari. Colloidal nanocarriers: A review on formulation technology, types and applications toward targeted drug delivery. Nanomedicine. 2010;6(1):9-24.
3. Helgason T, Awad TS, Kristbergsson K, McClements DJ, Weiss J. Effect of surfactant surface coverage on formation of solid lipid nanoparticles (SLN J Colloid Interface Sci. 2009;334(1):75-81
4. Muller RH, Mader K, Gohla S. Solid lipid nanoparticles (SLN) for controlled drug delivery -a review of the state of the art. Eur. Jour of Pharma and Biopharmaceutics. 2000;50:161-177.

5. Muller RH, Radtke M, Wissing SA. Solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC) in cosmetic and dermatological preparations. Adv Drug Delivery Rev. 2002;54:S131-155.
6. Ye J, Wang Q, Zhou X, Zhang N. Injectable actarit-loaded solid lipid nanoparticles as passive targeting therapeutic agents for rheumatoid arthritis. Int Jou of Pharm.2008;352:273–279.
7. A.R.Gardouh, Shadeed Gad, Hassan M. Ghonaim, and Mamdouh M. Ghorab Design and Characterization of Glyceryl Monostearate Solid Lipid Nanoparticles Prepared by High Shear Homogenization. Jou Pharm Res Int. 2013;3(3):326-346.
8. Ebrahimi HA, Javadzadeh Y, Jalali MB, Hamidi M. Preparation and in vitro characterization of Repaglinide-loaded Solid Lipid Nanoparticles. Research in Pharmaceutical Sciences. 2012; 7(5):281
9. Faruksha AU, Vectrichelvan T. Formulation, characterization and optimization of pioglitazone hydrochloride nanoparticles by solvent displacement method using 3 2 factorial design. International Journal of PharmTech Research. 2013;5:754-66.
10. Nair R, Kumar KA, Vishnupriya K, Badivaddin TM, Sevukarajan M. Preparation and characterization of rizatriptan loaded solid lipid nanoparticles (SLNs). J. Biomed. Sci. Res. 2011;3(2): 392-6.
11. Verma S, Kumar A, Malik VK, Kumar V. Compritrol ATO 888 based solid lipid nanoparticles of cefixime: formulation and evaluation. Der Pharmacia Sinica. 2013;4(3):8-13.
12. Dash S, Murthy PN, Nath L, Chowdhury P. Kinetic modeling on drug release from controlled drug delivery systems. Acta Pol Pharm. 2010;67(3):217-23.
13. Srinivasan K, Viswanad B, Asrat L, Kaul CL, Ramarao P. Combination of high-fat diet-fed and low-dose streptozotocin-treated rat: a model for type 2 diabetes and Pharmacological Screening. Pharmacological Research. 2005;52(4): 313-20.
14. Hilly M, Adams ML, Nelson SC. A study of digit fusion in the mouse embryo. Clin Exp Allergy. 2002;32(4):489-98.
15. Sharma N, Rana S, Shivkumar HG, Sharma RK. Solid lipid nanoparticles as a carrier of matformin for transdermal delivery. International Journal of Drug Delivery. 2013;5(2):137.
16. Sinha VR, Srivastava S, Goel H, Jindal V. Solid Lipid Nanoparticles (SLN'S)–Trends and Implications in Drug Targeting. International Journal of Advances in Pharmaceutical Sciences. 2011;1(3):232-238.
17. Boddupalli BM, Masana P, Anisetti RN, Kallem SV, Madipoju B. Formulation and evaluation of pioglitazone loaded bovine serum albumin nanoparticles along with piperine, Drug Invent. 2013; 212–215.
18. Radheshyam Tiwari, Kamla Pathak Nanostructured lipid carrier versus solid lipid nanoparticles of simvastatin: Comparative analysis of characteristics, pharmacokinetics and tissue uptake. International Journal of Pharmaceutics. 2011;232–243.
19. Junji Matsui,Yasuo Terauchi,Naoto Kubota,Iseki Takamoto,Kazuhiro Eto,Tokuyuki Yamashita, Kajuro Komeda,Toshimasa Yamauchi,Junji Kamon,Shunbun Kita,Mitsuhiko Noda and Takashi Kadowaki Pioglitazone Reduces Islet Triglyceride Content and Restores Impaired Glucose-Stimulated Insulin Secretion in Heterozygous Peroxisome Proliferator–Activated Receptor-γ–Deficient Mice on a High-Fat Diet.Diabetes. 2004;53(11):2844-2854.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle4.com/review-history/75554