Journal of Neonatal Surgery
ISSN(Online): 2226-0439
Vol. 14, Issue 32s (2025)
https://www.jneonatalsurg.com

OPEN ACCESS

Formulation and Evaluation of Azelnidipine Solid Lipid Nanoparticles

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Cite this paper as: Bhaskar Gugulothu , Praveena Chinthala , (2025) Formulation and Evaluation of Azelnidipine Solid Lipid Nanoparticles. *Journal of Neonatal Surgery*, 14 (32s), 5383-5389.

ABSTRACT

Introduction: Azelnidipine is a third-generation, long-acting dihydropyridine calcium channel blocker used to treat high blood pressure. The aim of the study was to prepare Azelnidipine solid lipid nanoparticles (SLN) to improve the solubility and bioavailability.

Methods: Six SLN formulations F1-F6of Azelnidipine were developed using three different lipids - Trimyristin, Tripalmitin and Tristearin at varying concentrations by the hot homogenization followed by ultrasonication method. All the formulations were characterized for their particle size (Ps), zeta potential (ZP), entrapment efficiency (EE), assay and drug release. The stability study test was conducted on optimized formulation at room temperature and refrigerated conditions for 3 months.

Results: The particle sizes of the formulations ranged from 167 ± 2.1 to 325 ± 1.3 nm, with PDI values between 0.267 ± 0.03 and 0.384 ± 0.03. Zeta potential ranged from –18.17 ± 1.1 to –23.01 ± 1.3 mV, and entrapment efficiency was between 84.21 ± 0.1% and 94.16 ± 0.1%. *In vitro* drug release studies, conducted using the dialysis method in 0.1 N HCl and pH 6.8 phosphate buffer, demonstrated a slower release in acidic medium compared to the phosphate buffer. Among all formulations, F5 prepared with Tripalmitin exhibited the highest drug release of 79.21% over 24 hours in pH 6.8 phosphate buffer. Based on particle size, entrapment efficiency, and drug release profile, F5 was selected as the optimized formulation. Stability studies confirmed that F5 remained stable for at least three months.

Conclusion: The optimized formulation (F5) demonstrated the highest zeta potential, excellent entrapment efficiency, small particle size, and sustained drug release over 24 hours, indicating that SLN are a promising delivery system for enhancing the bioavailability of Azelnidipine.

Keywords: *Azelnidipine, solid lipid nanoparticles, ultrasonication, particle size, entrapment efficiency, in-vitro release..*

1. INTRODUCTION

Solid lipid nanoparticles (SLN) are emerging as a promising drug delivery system due to their versatility, biocompatibility, and ease of formulation. They are capable of effectively encapsulating both hydrophilic and lipophilic drugs, often achieving high entrapment efficiency (EE). Composed of a solid lipid matrix, SLN facilitate drug uptake via the lymphatic system, thereby bypassing hepatic first-pass metabolism. The use of physiological lipids in their composition ensures that SLN are biodegradable, biocompatible, and exhibit minimal toxicity. Additionally, SLN offer advantages such as high drug loading capacity, long-term stability, scalability for industrial production, and cost-effective manufacturing [1-2].

Azelnidipine is a third-generation, long-acting dihydropyridine calcium channel blocker widely recommended for the treatment of hypertension. However, its therapeutic efficacy is limited by its high lipophilicity and poor solubility in the gastrointestinal tract (GIT) [3–4]. Azelnidipine lowers blood pressure by inhibiting the transmembrane influx of Ca^{2+} through voltage-dependent calcium channels in vascular smooth muscle. It was chosen as the model drug for this study due to its low oral bioavailability (22.1%), primarily attributed to poor aqueous solubility and extensive hepatic first-pass metabolism [5].

These formulation challenges, including low solubility and poor bioavailability, contribute to a delayed onset of action and limit its effectiveness as a fast-acting oral medication. The development of SLN offers a promising approach to overcome these limitations by enhancing solubility, improving bioavailability, and facilitating more efficient drug delivery..

2. MATERIALS AND METHODS

Azelnidipine was obtained as a gift sample from FDC Limited, Mumbai, India. Trimyristin (TM), Tripalmitin (TP) and Tristearin (TS) were purchased from Sigma-Aldrich Chemicals, Hyderabad, India. Egg Lecithin E-80 and Poloxamer-188 were obtained as gift samples from Hetero Labs, India. Solvents Methanol and Chloroform were of HPLC grade (Merck, Mumbai, India). Millipore Direct Q ® 3UV water was used in all the studies.

Preparation of Azelnidipine-loaded solid lipid nanoparticles

A total of six Azelnidipine-loaded SLN were prepared using Trimyristin (TM), Tripalmitin (TP), and Tristearin at varying concentrations by the hot homogenization followed by ultrasonication method. The composition of the various formulations is presented in Table 1.

Azelnidipine, the selected lipid (TM, TP, or TS), and egg lecithin were dissolved in 15 mL of a 1:1 mixture of Chloroform and Methanol. The organic solvents were completely evaporated using a flash evaporator, yielding a drug-embedded lipid layer, which was then melted by heating to 5°C above the melting point of the lipid [6].

Separately, the aqueous phase was prepared by dissolving Poloxamer 188 in double-distilled water and heating it to the same temperature as the molten lipid phase. The hot aqueous phase was added to the lipid phase, and the mixture was homogenized at 12,000 rpm for 4 minutes using a high-speed homogenizer. The resulting coarse oil-in-water emulsion was then ultrasonicated using a 12T probe sonicator for 20 minutes. Finally, the emulsion was allowed to cool to room temperature to form the Azelnidipine-loaded SLN.

Table 1: Composition of Azelnidipine-loaded SLN

Ingredients (mg)	F1	F2	F3	F4	F5	F6
Azelnidipine	08	08	08	08	08	08
Trimyristin	100	200	-	-	-	-
Tristearin	-	-	100	200	-	-
Tripalmitin	-	-	-	-	100	200
Poloxamer-188	120	120	120	120	120	120
Egg lecithin	80	80	80	80	80	80
Solvent (ml) (1:1) (Chloroform: Methanol)	15	15	15	15	15	15
Purified water	15	15	15	15	15	15

Characterization of Azelnidipine-loaded SLN

Measurement of Particle size (PS), Polydispersity index (PDI) and Zeta potential (ZP)

The particle size (PS), polydispersity index (PDI), and zeta potential (ZP) of the SLN were determined using a Malvern Zetasizer. For analysis, 100 µL of the SLN formulation was diluted with 5 mL of double-distilled water and used for measurement [7].

Determination of entrapment efficiency

Entrapment efficiency (EE) was determined by measuring the concentration of free (unentrapped) drug in the aqueous medium. Ultra-filtration was carried out using Centrisart tubes equipped with a filter membrane having a molecular weight cut-off of 20,000 Da at the base of the sample recovery chamber. Approximately 2.5 mL of the SLN formulation was placed in the outer chamber, and the sample recovery chamber was positioned on top. The system was centrifuged at 20,000 rpm for one hour. During centrifugation, the SLN along with the encapsulated drug remained in the outer chamber, while the aqueous phase containing the unentrapped drug passed through the membrane into the recovery chamber. The concentration of Azelnidipine in the aqueous phase was quantified using a UV-Visible spectrophotometer at 255 nm. Entrapment efficiency was then calculated based on the amount of unentrapped drug.

Assay

To determine drug content 100µl of the SLN formulation was taken and dissolved in Chloroform and Methanol mixture (1:1). Then further dilutions were made with the above mixture and calculated the amount of Azelnidipine present in formulations by UV method[8].

In vitro drug release studies

In vitro drug release studies were performed using dialysis method. Dialysis membrane with a pore size 2.4 nm was used for the release studies. Before conducting drug release studies, dialysis membrane was soaked overnight in double distilled water. Hydrochloric acid (0.1N) and phosphate buffer pH 6.8 were used as release media. The experimental unit had donor and receptor compartments. Donor compartment consisted of a boiling tube which was cut open at one end and tied with dialysis membrane at the other end into which 1 ml of SLN dispersion was taken for release study. Receptor compartment consisted of a 250 ml beaker which was filled with 100 ml of release medium and the temperature was maintained at 37 ± 0.5º C. At 0.5, 1, 2, 3, 4, 6, 8, 10, 12 and 24 hour time points, 2 ml samples were withdrawn from receiver compartment and replaced with the same volume of release medium. The collected samples were suitably diluted and analyzed by UV-Visible spectrophotometer (SL-150, ELICO, and India) at 255 nm [9].

Stability studies

Optimized Azelnidipine-loaded SLN formulation F5 were stored at room temperature, 25 ± 2°C and refrigerated temperature, 4°C for three months. The physical characters such as average particle size, ZP, assay and entrapment efficiency were determined at specific periods i.e., 1 day, 1 month, 2 months and 3 months.

Lyophilization of Azelnidipine-loaded SLN

Lyophilization technique was used to enhance the stability of SLN. The SLN containing 10% w/v maltose were prepared and kept in deep freezer at - 40ºC for overnight. The frozen samples were then transferred into freeze-dryer. Vacuum was applied and sample was subjected to various drying phases for about 48 h. to get powdered lyophilized product [10].

Solid state characterization

Drug-excipient compatibility studies by differential scanning calorimeter (DSC)

DSC study was conducted for pure drug, trimyristin (TM), tripalmitin (TP) and tristearin (TS), physical mixtures (PM in 1:1 ratio) were performed. The instrument was calibrated with indium. All the samples (10 mg) were heated in aluminum pans using dry nitrogen as the effluent gas. The analysis was performed within a heating range of 20–200º C and at a rate of 20º C/min.

Morphology by scanning electron microscopy (SEM)

Scanning Electron Microscope was conducted to study the morphology of SLN. Freeze dried Azelnidipine-loaded SLN were taken and suitably diluted with double distilled water (1 in 100). A drop of nanoparticle formulation was placed on sample holder and air dried. Then the sample was observed at accelerating voltage of 15000 volts.

3. RESULTS AND DISCUSSION

In this study, total of six formulations were developed by hot homogenization followed by ultrasonication method by using three lipids namely, trimyristin, tripalmitin and tristearin each with two different concentrations. The homogenization time and sonication time used in the formulation development was 4 and 20 min respectively after various trials.

Determination of Particle size, Polydispersity index and Zeta Potential of the formulations

Particle size, PDI and ZP of different prepared batches is presented in Table 2. The particle size of all the prepared formulations were found in the range of167 ± 2.1 nm to 325 ± 1.3 nm. The PDI was ranging from 0.26 ± 0.03 to 0.38 ± 0.03, indicating the narrow size distribution. Zeta potential of a nanoformulation is an important factor as the measurement of the ZP allows predictions about the storage stability of SLN. The zeta potential values of all formulations were found to be in between-18.17 ± 1.1 mV to -23.01 ± 1.3 mV, indicated the stability of all the formulations. The optimum zeta potential values i.e., -15 mV to -30 mV indicates good stability of the formulation [11].

4. DETERMINATION OF ASSAY AND ENTRAPMENT EFFICIENCY

Assay was conducted for all the SLN formulations and found to be ranging from 84.3 ± 0.2% to 98.2 ± 0.03%.Among all the formulations, F5 formulation shown highest drug content i.e., 98.2%.

Entrapment efficiency of all the developed formulations was found in the range of 84.21 ± 0.1% to 94.16 ± 0.1 % (Table 2). This high entrapment efficiency of drug could be resulted due to high lipophilicity of drug [12]. Formulation F5 has shown highest drug entrapment efficiency compared to other formulations.

Table 2: Characterization of Azelnidipine-loaded SLN

Formulation code	PS (nm)	PDI	ZP (mV)	Assay (%)	EE (%)

F1	219±1.2	0.321±0.02	-18.17±1.1	95.7±0.1	89.86±0.2
F2	245±2.3	0.384±0.03	-21.15±1.7	92.1±0.1	91.13±0.4
F3	279±1.1	0.325±0.04	-20.21±2.1	88.8±0.3	84.21±0.1
F4	325±1.3	0.332±0.01	-21.35±1.5	84.3±0.2	87.33±0.2
F5	167±2.1	0.267±0.03	-23.01±1.3	98.2±0.03	94.16±0.1
F6	175±1.4	0.289±0.02	-22.03±1.1	94.7±0.4	89.21±0.3

(Mean ±SD)

***In vitro* drug release studies**

In vitro drug release studies were conducted to evaluate the release profile of Azelnidipine from SLN formulations (F1–F6) using the dialysis method in 0.1 N hydrochloric acid and pH 6.8 phosphate buffer.

The cumulative percentage of drug release in 0.1 N HCl was found to be 39.01%, 30.00%, 21.23%, 19.04%, 33.20%, and 27.19% for formulations F1 to F6, respectively. In pH 6.8 phosphate buffers, the corresponding release values were 52.14%, 49.62%, 41.31%, 38.73%, 79.21%, and 59.32% (Figure 1).

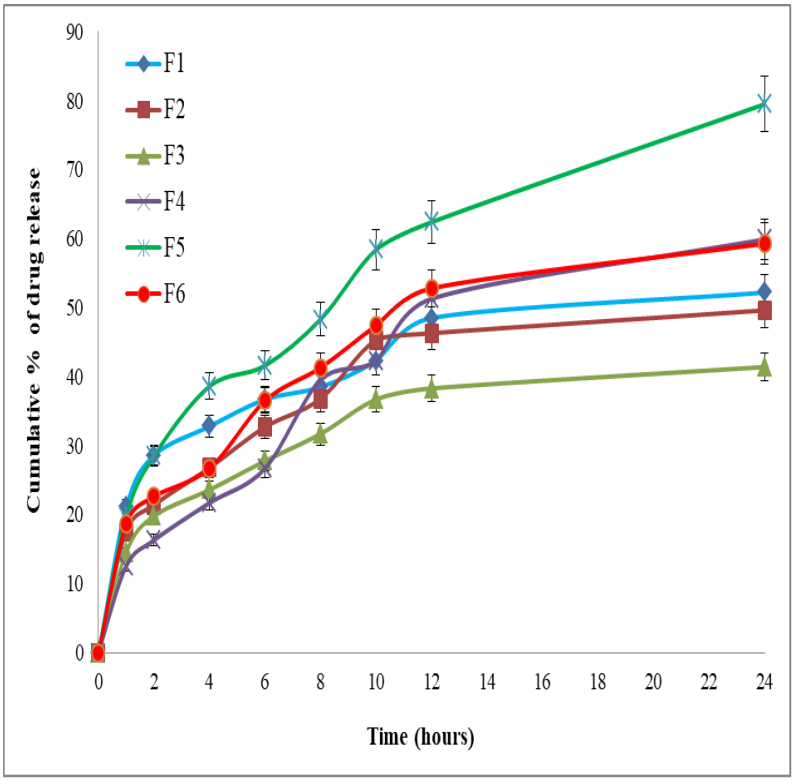

Fig 1: Cumulative % drug released profiles of Azelnidipine SLN formulations in pH 6.8 buffer (F1-F6)

Drug release from SLN was comparatively slower in 0.1 N HCl than in phosphate buffer, which may be attributed to the differential solubility of Azelnidipine in these media [13–14]. Among all formulations, F5 demonstrated the highest cumulative drug release of 79.21% in pH 6.8 phosphate buffer over 24 hours. In addition to superior drug release, F5 also exhibited favorable characteristics, including optimal particle size, narrow PDI, high zeta potential, and excellent entrapment efficiency. Based on these results, formulation F5 was identified as the optimized formulation and selected for further evaluation.

The DSC thermogram was conducted for pure drug and for physical mixture. Pure drug Azelnidipine showed a sharp endothermic peak at 195 ºC, whereas Trimyristin, Tripalmitin and Tristearin showed drug peaks at 56 ºC, 64 ºC and 72 ºC respectively (Figure 2: A-E).

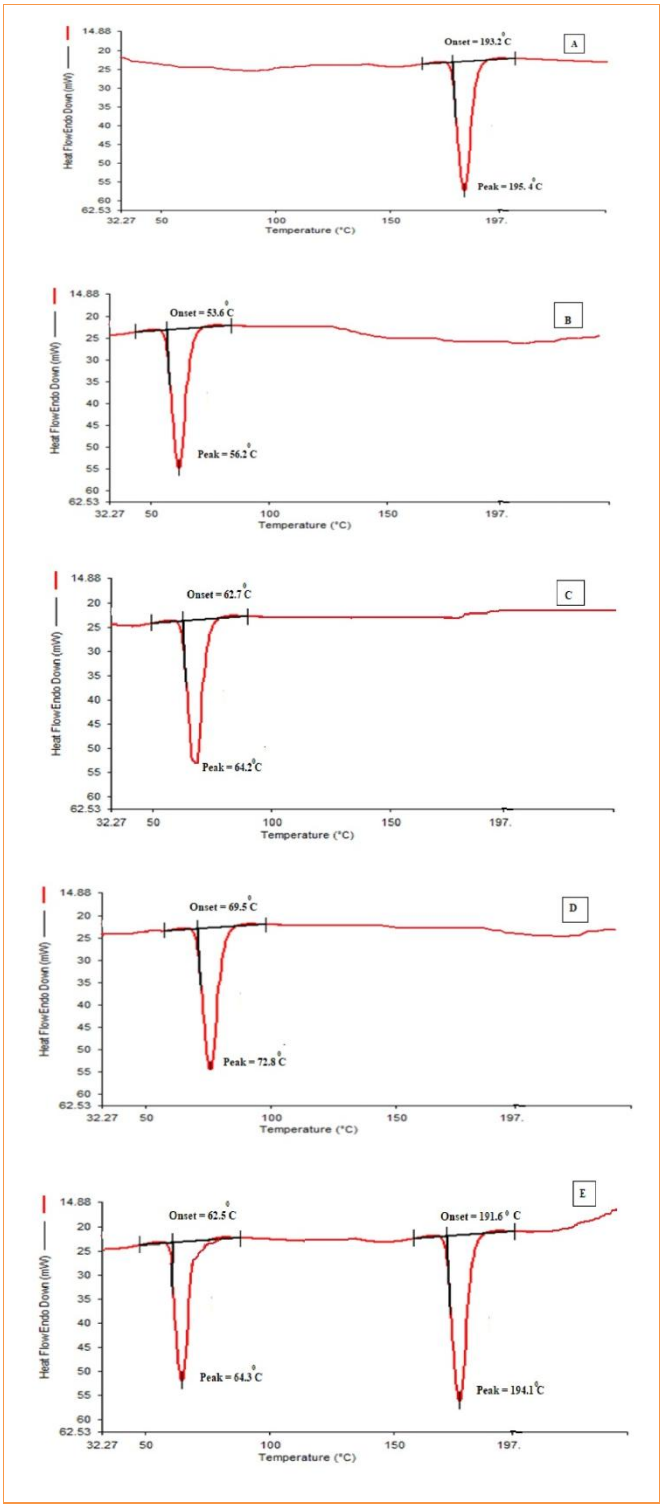

Fig 2: A) DSC thermogram for pure drug (Azelnidipine); B) DSC thermogram of Trimyristin; C) DSC thermogram of Tripalmitin; D) DSC thermogram of Tristearin; E) DSC thermogram of Physical mixture.

SEM studies were conducted for optimized formulation F5 revealed that the particles possessed smooth surface and spherical in shape (Figure 3).

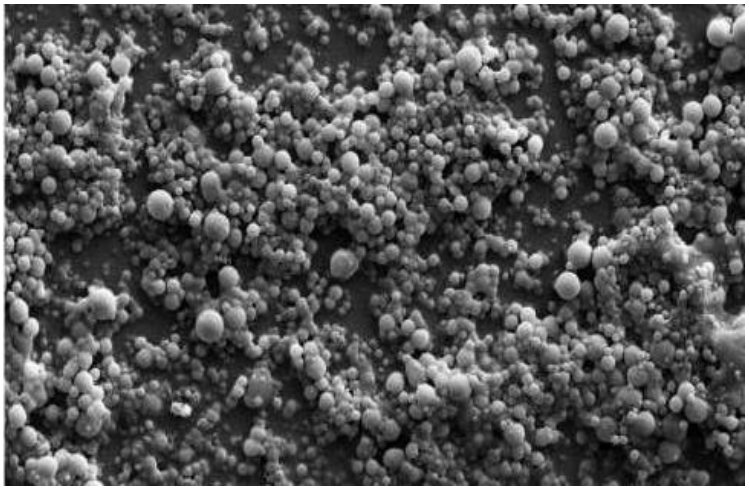

Fig 3: SEM image of Azelnidipine lyophilized formulation (F5)

Stability Studies

The optimized Azelnidipine-loaded SLN formulation (F5) was evaluated for stability over three months under room temperature (25 ± 2°C) and refrigerated (4°C) conditions. At both storage conditions, particle size remained stable (~167 nm and ~166 nm, respectively) with no significant aggregation.

Table 3: Stability studies of optimized formulation (F5)

Storage condition	Duration (months)	PS (nm)	ZP (mV)	Assay (%)	EE (%)
Room temperature	0	167±2.1	-23.01±1.3	98.2±0.03	94.16±0.1
	1	167±1.7	-23.01±1.4	98.1±0.02	94.01±0.2
	2	167±1.6	-23.02±1.2	98.1±0.03	94.01±0.3
	3	167±1.3	-23.01±1.1	97.6±0.01	93.15±0.1
Refrigerator temperature	0	167±2.1	-23.01±1.3	98.2±0.03	94.16±0.1
	1	166±1.9	-23.21±1.2	98.2±0.02	94.15±0.3
	2	166±1.8	-22.13±1.1	98.2±0.03	94.16±0.2
	3	166±1.5	-22.09±1.2	98.2±0.02	94.16±0.2

(Mean ± SD)

Zeta potential showed minimal variation, remaining around –23 mV. Drug content exhibited a slight decrease at room temperature (from 98.2 ± 0.03% to 97.6 ± 0.01%) but remained constant under refrigeration. Entrapment efficiency showed minor reduction at room temperature (from 94.16 ± 0.1% to 93.15 ± 0.1%), while it remained unchanged under cold storage. These results indicate that formulation F5 remained physically and chemically stable under both conditions for at least three months.

5. CONCLUSION

The study successfully developed and optimized Azelnidipine loaded SLN to overcome the limitations of poor solubility and low oral bioavailability associated with Azelnidipine. Among the formulations, F5 formulated with Tripalmitin—demonstrated optimal characteristics, including high entrapment efficiency, nanoscale particle size, and sustained drug release over 24 hours. The formulation remained physically stable under both room temperature and refrigerated conditions for three months. These findings indicate that SLN, particularly the optimized F5 formulation, offer a promising delivery system for enhancing the therapeutic efficacy.

ACKNOWLEDGEMENTS

All the authors are thankful to Chaitanya (Deemed to be University) for providing facilities.

CONFLIT OF INTEREST

There is no conflict of interest

REFERENCES

[1] Mukherjee S, Ray S, Thakur RS. Solid lipid nano particles: A modern formulation approach in drug delivery system. Indian J Pharm Sci. 2009; 71: 349–58.

[2] Ekambaram P, Abdul HA, Priyanka K. Solid lipid nano particles: a review. Chem. Commun, 2012; 2(1): 80-102.

[3] Carretero OA, Oparil S. Essential hypertension. Part I: Definition and etiology. Circulation. 2010; 101(3): 329-35.

[4] Chen BL, Zhang YZ, Luo JQ, Zhang W. Clinical use of azelnidipine in the treatment of hypertension in Chinese patients. Therapeutics and clinical risk management. 2015; 24: 309-18.

[5] Xiao Y, Hu G. The effects of azelnidipine and amlodipine in treatment of mild to moderate hypertension: a systematic review. Int J Clin Exp Med. 2017; 10(7): 11273-8.

[6] Thatipamula RP, Palem CR, Gannu R.Formulation and in vitro characterization of domperidone loaded solid lipid nano particles and nano structured lipid carriers. Daru, 2011; 19: 23–32.

[7] Imran B, ud Din F, Ali Z, Fatima A, Khan MW, Kim DW, Malik M, Sohail S, Batool S, Jawad M, and Shabbir K. Statistically designed dexibuprofen loaded solid lipid nanoparticles for enhanced oral bioavailability. J.Drug Deliv.Sci.Tech.2022; 77:103904.

[8] Amira A Rashad, S.Nageeb El-Helaly, Randa T Abd El Rehim, Omaima N El-Gazayerly Core-in-cup/liquisol dual tackling effect on azelnidipine buccoadhesive tablet micromeritics, in vitro release, and Muco adhesive strength. Acta Pharma., 2019; 69: 1-18.

[9] El-Assal MI, Samuel D. Optimization of rivastigmine chitosan nanoparticles for neuro degenerative alzheimer; in vitro and ex vivo characterizations. Int J Pharm Pharm Sci. 2022; 14(1):17-27.

[10] Elbahwy IA, Ibrahim HM, Ismael HR, Kasem AA. Enhancing bioavailability and controlling the release of glibenclamide from optimized solid lipid nanoparticles. J Drug Deliv Sci Technol. 2017; 38:78-89.

[11] Nagaraj B, Tirumalesh C, Dinesh S, Narendar D. Zotepine loaded lipid nanoparticles for oral delivery: development, characterization, and in vivo pharmacokinetic studies. Futur J Pharm Sci. 2020; 6(1):1.

[12] Vaidya A, Jain S, Jain A, Jain A. Simvastatin-loaded pegylated solid lipid nanoparticles: lipid functionalization to improve blood circulation. Bio Nano Science. 2020; 10(3):773-82.

[13] Kushwaha AK, Vuddanda PR, Karunanidhi P, Singh SK, Singh S. Development and evaluation of solid lipid nanoparticles of Raloxifene hydrochloride for enhanced bioavailability. Bio Med Res Int. 2013; 20: 539-549.

[14] Shaveta S, Singh J, Afzal M, Kaur R, Imam SS, Alruwaili NK. Development of solid lipid nano particle as carrier of Pioglitazone for amplification of oral efficacy: formulation design optimization, in vitro characterization and in vivo biological evaluation. J Drug Deliv Sci Technol. 2020; 57: 126-132.