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International Journal of Pharmaceutics 335 (2007) 167–175
INTERNATIONAL JOURNAL OF PHARMACEUTICS
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Pharmaceutical Nanotechnology

Development and evaluation of nitrendipine loaded solid lipid nanoparticles: Influence of wax and glyceride lipids on plasma pharmacokinetics

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Received 25 April 2006; accepted 25 October 2006
Available online 3 November 2006

Abstract

Nitrendipine is an antihypertensive drug with poor oral bioavailability ranging from 10 to 20% due to the first pass metabolism. For improving the oral bioavailability of nitrendipine, nitrendipine loaded solid lipid nanoparticles have been developed using triglyceride (tripalmitin), monoglyceride (glyceryl monostearate) and wax (cetyl palmitate). Poloxamer 188 was used as surfactant. Hot homogenization of melted lipids and aqueous phase followed by ultrasonication at temperature above the melting point of lipid was used to prepare SLN dispersions. SLN were characterized for particle size, zeta potential, entrapment efficiency and crystallinity of lipid and drug. In vitro release studies were performed in phosphate buffer of pH 6.8 using Franz diffusion cell. Pharmacokinetics of nitrendipine loaded solid lipid nanoparticles after intraduodenal administration to conscious male Wistar rats was studied. Bioavailability of nitrendipine was increased three- to four-fold after intraduodenal administration compared to that of nitrendipine suspension. The obtained results are indicative of solid lipid nanoparticles as carriers for improving the bioavailability of lipophilic drugs such as nitrendipine by minimizing first pass metabolism.

Keywords: Solid lipid nanoparticles; Nitrendipine; Lipophilic; Intraduodenal; Pharmacokinetics; Bioavailability

1. Introduction

Nitrendipine is a calcium channel blocker, which is used as an antihypertensive drug. It is practically insoluble in water and is having poor bioavailability (10–20%) because of extensive hepatic first pass metabolism resulting in the formation of four inactive metabolites (100 times less potent than nitrendipine) (Parfitt, 1999). Possible methods to avoid first pass metabolism include transdermal, buccal, rectal and parenteral routes of administration. Oral route is the most commonly used and preferred route of choice for the delivery of drugs, although several factors like pH of GIT, residence time and solubility can affect this route. Lymphatic delivery is an alternative choice to avoid first pass metabolism in peroral drug delivery (Driscoll, 2002). Enhanced lymphatic transport of drugs reduces the hepatic first-pass metabolism and improves bioavailability, because intestinal lymph vessels drain directly into thoracic duct, further in to the venous blood, thus bypassing the portal circulation (Porter and Charman, 2001). The main function of the lymphatic system is to facilitate absorption of long chain fatty acids via chylomicron formation. Two different lipid based approaches are known to enhance the lymphatic transport, which includes construction of a highly lipophilic prodrug and incorporation of drug in a lipid carrier (Charman and Porter, 1996).

Solid lipid nanoparticles (SLN) are one of the carrier systems having more advantages than other colloidal delivery systems with regard to biocompatibility and scale up (Muller et al., 2000). Reports on the use of SLN for avoiding first pass metabolism of drugs are scanty. Bargoni et al. (1998) studied the lymphatic uptake of SLN after duodenal administration to rats. Cavalli et al. (2003) reported that intraduodenally administered tobramycin loaded SLN showed sustained release and lymphatic targeting. Recently, Manjunath and Venkateswarlu (2005) also studied the pharmacokinetics and tissue distribution aspects of clozapine

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doi:10.1016/j.ijpharm.2006.11.004

loaded solid lipid nanoparticles after intraduodenal administration.

The aim of the present work is to study the improvement of bioavailability by incorporating nitrendipine, a poorly bioavailable drug in SLN prepared with different lipids such as tripalmitin (triglyceride), cetyl palmitate (wax) and glyceryl monostearate (monoglyceride). Various methods such as high-pressure homogenization, solvent emulsification/evaporation were reported in literature for preparing solid lipid nanoparticles (Mehnert and Mader, 2001). In this study, SLN were prepared by hot homogenization followed by ultra sonication method. Particle size and zeta potential measurements, crystallinity studies, in vitro release studies and bioavailability studies in rats were performed and were compared with those of nitrendipine suspension. Studies were also conducted on the effect of nature of lipid (glyceride or wax) on the in vitro and in vivo performance of SLN.

2. Materials and methods

2.1. Materials

Tripalmitin (Glyceryl tripalmitate, mp 60–62 °C), cetyl palmitate (Palmityl palmitate, hexadecyl hexadecanoate, mp 54–55 °C), and poloxamer 188 were from Sigma (St. Louis, MO, USA). Monostearin (Glyceryl monostearate, mp 56–57 °C) was from Loba Chemie (Mumbai, India). Nitrendipine was a kind gift from USV Ltd. (Mumbai, India). Felodipine (internal standard) was a kind gift from Everlight Chemicals, India. Centrisart filters (molecular weight cut off 20,000) were purchased from Sartorius, Goettingen, Germany. Chloroform, methanol, acetonitrile and dichloromethane were of HPLC grade (Merck, India.)

2.2. Preparation of nitrendipine loaded SLN

Nitrendipine (10 mg) and lipid (100–300 mg) were dissolved in 10 ml of a mixture of chloroform and methanol (1:1). The organic solvent mixture was completely evaporated at 60 °C using rotaevaporator (Heidolph, Laborota 4000, Germany). Nitrogen was blown on the lipid layer for 20 min to remove traces of vapors of organic solvents, if any. Drug embedded lipid was melted by heating at 5 °C above the melting point of lipid. The aqueous phase was prepared by dissolving Poloxamer 188 (100 mg) in double distilled water (Millipore, India) sufficient to produce 10 ml and heated to same temperature as that of molten lipid phase. Hot aqueous solution was added to molten lipid phase and homogenized (UltraTurrax T25) for 3 min at 11,000 rpm. The temperature was maintained 5 °C above the melting point of lipid throughout the homogenization process. The coarse hot o/w emulsion thus obtained was sonicated using a probe sonicator at 50 W (Vibra cell, Sonics, USA) for 20 min. The obtained nanoemulsions were cooled to room temperature and frozen in dry ice followed by freeze drying (Modulyod Freeze dryer, Thermo electron corporations, USA) for 48 h to obtain lyophilized SLN. Nitrendipine loaded SLN obtained using tripalmitin, cetyl palmitate and glyceryl monostearate were abbreviated as F-TP, F-CP and F-GMS, respectively.

2.3. Measurement of size of SLN

Size and zeta potential of all blank and drug loaded SLN samples were measured by photon correlation spectroscopy using Zetasizer 3000 HAS (Malvern Instruments, UK). All the samples were diluted with aqueous phase of the formulation to get optimum kilo counts per second (Kcps) of 50–200 for measurements. Average particle size in nanometers, polydispersity index and zeta potential were measured.

2.4. Analytical methods

Estimation of nitrendipine in phosphate buffer of pH 6.8 and in plasma samples were conducted by reversed-phase HPLC (Shimadzu 10 Ai, Japan) in a binary mode, with a photodiode array detector and a communication bus module. The analysis was performed at 238 nm with a Spherisorb ODS2, reversed-phase C_{18}, 250 mm × 4.5 mm, 5 μm column (Waters Spherisorb) maintained at 25 °C (column oven) employing a mobile phase of acetonitrile (60%) and water (40%) delivered at a flow-rate of 1.5 ml/min. The retention time of the drug was found to be 6.0 ± 0.1 min. The calibration curve was rectilinear in the concentration range of 0.1–10 μg/ml ($r^2 = 0.99$) in phosphate buffer of pH 6.8 and 10–200 ng/ml ($r^2 = 0.99$) in rat plasma. The inter- and intra-day accuracy and precision was within an R.S.D. ≤ 5%. The extraction efficacy in case of spiked plasma samples was 98.1 ± 1.8%. Data analysis and processing were done by class LC-10 software (Version 1.6).

2.5. Determination of drug content

Nitrendipine loaded SLN (0.5 ml) were diluted to 10 ml with chloroform/methanol mixture (1:1) and final dilution was made with mobile phase and total content was determined by HPLC (Shimadzu, Japan) method as described above.

2.6. Determination of entrapment efficiency

Entrapment efficiency (EE) was determined by measuring the concentration of free drug (unentrapped) in aqueous medium as reported previously (Venkateswarlu and Manjunath, 2004). The aqueous medium was separated by ultra-filtration using centrisart tubes (Sartorius, USA), which consists of filter membrane (M.wt. cut off 20,000 Da) at the base of the sample recovery chamber. About 1 ml of the formulation was placed in the outer chamber and sample recovery chamber placed on top of the sample and centrifuged at 4000 rpm for 15 min (Biofuge stratos, Heraeus). The SLN along with encapsulated drug remained in the outer chamber and aqueous phase moved into the sample recovery chamber through filter membrane. The amount of nitrendipine in the aqueous phase was estimated by HPLC method as described above and the entrapment efficiency was

calculated by the equation:

$$\mathrm{EE}(\%) = \left(\frac{\text{wt. of drug used in the formulation} - \text{wt. of drug in aqueous phase}}{\text{wt. of drug used in the formulation}} \right) \times 100$$

2.7. Stability studies

Nitrendipine loaded SLN of different lipids were stored at 25 °C for 3 months and average size and entrapment efficiency were determined.

2.8. Characterization of crystallinity by powder X-ray diffractometry (PXRD)

Powder X-ray diffractometer (Siemen's D-5000, Germany) was used for diffraction studies. PXRD studies were performed on the samples by exposing them to Cu Kα radiation (40 kV, 30 mA) and scanned from 2° to 70°, 2θ at a step size of 0.045° and step time of 0.5 s. Samples used for PXRD analysis were pure Nitrendipine, pure tripalmitin/cetyl palmitate/glyceryl monostearate and lyophilized solid lipid nanoparticles.

2.9. In vitro release of nitrendipine from SLN

In vitro release studies were performed using modified Franz diffusion cell. Dialysis membrane having pore size 2.4 nm, molecular weight cut off 12,000–14,000 was used. Membrane was soaked in double-distilled water for 12 h before mounting in a Franz diffusion cell. A volume of 1 ml of nitrendipine loaded SLN formulation was placed in the donor compartment and the receptor compartment was filled with 10 ml of dialysis medium consisting of phosphate buffer pH 6.8. An aliquot of 100 μl of sample was withdrawn from receiver compartment through side tube at time intervals of 0.5, 1, 2, 4, 6, 8, 12, 24 and 48 h. Fresh medium was replaced each time to maintain constant volume. Samples were analysed by HPLC as described above.

2.10. Animal study protocol

Male Wistar rats were obtained from National Institute of Nutrition, Hyderabad. The animals were quarantined in the animal house maintained at 20 ± 2 °C and 50–60%RH. A 12 h dark/light cycle was maintained throughout the study. Air changes (15–16 h^{-1}) were maintained with 5μ HEPA filter in a class 10,000 environment. Rats had free access to food (pellet diet supplied from M/s Petcare India Ltd., Bangalore) and water *ad libitum*. The study protocol was approved by the Institutional Animal Ethics Committee of Indian Institute of Chemical Technology, Hyderabad. Twenty-four male Wistar rats weighing 170 ± 20 g were kept for overnight fasting with free access to water. The animals were divided into four groups of six animals each and F-TP, F-CP, F-GMS and nitrendipine suspension were administered by intraduodenal route.

2.10.1. Intraduodenal administration

Rats were anaesthetized with diethyl ether and a small incision was made at abdomen of anaesthetized rats. Duodenum was located and SLN formulations were slowly administered directly into the duodenum with a syringe. The incision was sutured carefully. For comparison, nitrendipine was administered as 1% gum acacia suspension to fourth group. Blood samples (0.3 ml) were collected from the tail vein in EDTA coated tubes at 1, 2, 4, 6, 8, 12, 18, 24, 36, 48 and 72 h after drug administration. The blood samples were centrifuged (5000 rpm, 15 min) and plasma was collected and stored at −70 °C until analysis.

2.10.2. Plasma sample preparation

For determination of nitrendipine, 100 μl of plasma was taken in a 5 ml test tube and to this 20 μl of internal standard (Felodipine, 10 μg/ml) was added and vortexed for 2 min. The samples were made alkaline by addition of 100 μl of 1N NaOH and vortexed for 2 min. To this sample, 780 μl of Dichloromethane was added and vortexed for 5 min and then centrifuged at 4000 rpm for 15 min. The organic layer was separated and evaporated using Automatic Nitrogen evaporator (TURBO VAP® LV, Zymark, USA). The residue was reconstituted with mobile phase and a volume of 100 μl of sample was injected into HPLC for analysis.

2.11. Pharmacokinetic analysis

Non-compartmental analysis with WinNonlin software (Version 1.1) was used to estimate the pharmacokinetic parameters of nitrendipine. Maximum plasma concentration (C_{max}) and time to reach C_{max} (T_{max}) were taken directly from the observed concentration versus time profiles. Area under the curve (AUC) and area under the first moment curve (AUMC) were calculated using linear trapezoidal rule and MRT was determined by dividing AUMC by AUC. Relative Bioavailability of SLN formulations was calculated using the formula:

$$\mathrm{Fr} = \frac{\mathrm{AUC_{SLN}}}{\mathrm{AUC_{Susp}}}$$

2.12. Statistical analysis

The data on entrapment efficiency and the cumulative percent drug released from different SLN formulations were analysed using ANOVA followed by Tukey's range test. Significance was evaluated at p-value of 0.05. The pharmacokinetic parameters such as AUC, MRT, $t_{1/2}$ and C_{max} obtained in animal studies for different formulations of F-CP, F-TP and F-GMS and control suspension were analysed using one-way ANOVA. The difference in each nanoparticle formulation with suspension formulation was evaluated using Dunnett's multiple comparison procedure. Significance was evaluated at p-value of 0.05.

3. Results and discussions

3.1. Preparation of nitrendipine loaded SLN

Homogenization followed by ultrasonication is a reliable, simple and reproducible method for preparing SLN (Venkateswarlu and Manjunath, 2004). The solvent mixture of chloroform and methanol (1:1) was found to be effective in homogenously dispersing the drug in the lipid phase. Rota evaporation at 60 °C followed by nitrogen purging ensured the complete removal of the traces of organic solvents. Homogenization of the lipid phase with hot aqueous poloxamer solution for 3 min was sufficient to produce a coarse emulsion with average particle size between 3.12 and 3.23 μm. Further increase in homogenization time did not show any significant decrease in the particle size (3.11–3.20 μm) Thus a homogenization time of 3 min was selected for all the formulations and further reduction of size was proceeded with sonication. Sonicating the coarse emulsion for 20 min resulted in particles between 110 and 140 nm with narrow size distribution. In order to optimize the lipid to drug ratio, different amounts of all three lipids (100, 200 and 300 mg) were tried with fixed amount of drug (10 mg). At lower lipid contents (100 and 200 mg), micro particles and drug deposition at the bottom indicating the poor encapsulation efficiency. However, when the lipid content was increased to 300 mg, no deposition occurred in all the three lipids Based on these results, the lipid to drug ratio of 300:10 was used for further studies.

3.2. Measurement of size

To obtain stable and smaller SLN, poloxamer concentration was varied from 0.5% to 1.5% and their effect on particle size was measured (Fig. 1). It is evident from Fig. 1 that a poloxamer concentration of 1% was effective in producing smaller size SLN in case of all the three lipids (110.6–115.4 nm for F-TP, 116.4–122.3 nm for F-CP and 132.6–137.4 nm for F-GMS). Further increase in poloxamer concentration to 1.5% did not reduce the particle size. These results clearly suggest that an optimum concentration of 1% poloxamer was sufficient to cover the surface of nanoparticles effectively and prevent agglomeration during the homogenization process. High concentration of surfactant (1.5%) was avoided to prevent decrease in the entrapment efficiency and also toxic effects associated with surfactants (Muller et al., 2000). Among the glycerides used, TP produced least size SLN when compared with GMS. Similar trend was observed for the etoposide loaded SLN (Harivardhan Reddy and Murthy, 2005).

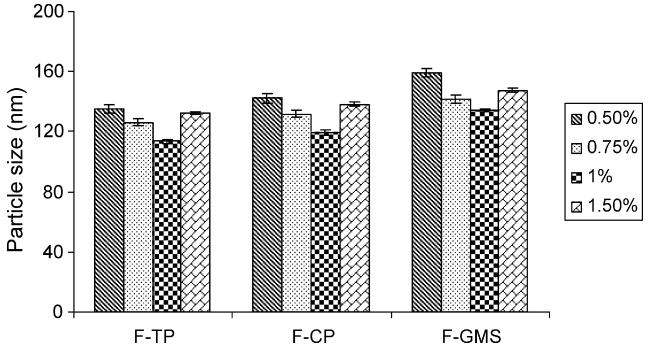


Fig. 1. Effect of poloxamer 188 concentration (0.5%, 0.75%, 1.0% and1.5%) on particle size of nitrendipine SLN of different lipids (F-TP, F-CP and F-GMS) (mean ± S.D., $n = 3$).

3.3. Total content and entrapment efficiency

The drug content of the SLN was highest for the formulation F-TP (1.01 mg/ml) followed by F-CP (0.92 mg/ml) and least for F-GMS (0.513 mg/ml). Entrapment efficiencies of F-TP, F-CP and F-GMS were shown in Table 1. Formulation F-GMS prepared with glyceryl monostearate has shown less drug-loading capacity compared with those of F-TP and F-CP prepared with tripalmitin and cetyl palmitate respectively

Table 1
Particle size, zeta potential and entrapment efficiency of different lipid SLN formulations (mean ± S.D., $n = 3$)

Formulation	Particle size (nm)		Zeta potential (mv)	Entrapment efficiency (%)
	Mean size (nm)	Poly dispersity index		
F-TP	112.6 ± 1.19	0.242 ± 0.03	−19.3 ± 0.3	97.53 ± 0.11
F-CP	121.6 ± 1.75	0.258 ± 0.02	−20.6 ± 1.6	94.67 ± 0.21[$]
F-GMS	136.2 ± 1.34	0.295 ± 0.05	−22.3 ± 0.2	91.8 ± 0.36[*]

[*] Statistical significance with F-TP and F-CP ($p < 0.05$).
[$] Statistical significance with F-TP ($p < 0.05$).

Table 2
Effect of storage time (at 25 °C) on particle size and entrapment efficiency of SLN (mean ± S.D., $n = 3$)

Formulation	Particle size (nm)				Entrapment efficiency (%)			
	Zero day	1 month	2 months	3 months	Zero day	1 month	2 months	3 months
F-TP	109.3 ± 1.2	116.9 ± 1.5	130.5 ± 2.3	142.6 ± 2.1	97.52 ± 0.64	97.21 ± 0.51	96.23 ± 0.25	94.12 ± 0.35
F-CP	123.2 ± 3.4	124.9 ± 1.9	130.6 ± 2.5	140.3 ± 2.4	94.66 ± 0.36	94.28 ± 0.56	93.87 ± 0.36	92.59 ± 0.34
F-GMS	140.3 ± 2.8	149.3 ± 2.3	167.4 ± 3.2	179.4 ± 3.1	91.8 ± 0.58	90.35 ± 0.72	89.72 ± 0.46	87.53 ± 0.61

($p < 0.05$). The entrapment efficiency of the SLN for nitrendipine was in the order of F-TP > F-CP > F-GMS. The higher drug content and entrapment efficiency with TP is attributed to the high hydrophobicity due to the long chain fatty acids attached to the triglyceride resulting in increased accommodation of lipophilic drugs (Jenning and Gohla, 2000).

3.4. Stability studies

In all SLN formulations, storage at 25 °C resulted in increased particle size. Increase in size was from 109.3 to 142.6 nm with F-TP, from 123.2 to 140.3 nm with F-CP and from 140.3 to 179.4 nm with F-GMS. SLN prepared with wax (CP) exhibited good long-term stability while glyceride SLN (both TP and GMS) showed enhanced increase in particle size (Table 2). Within glycerides, SLN prepared with TP showed better physical stability than GMS. Increased amount of partial glycerides like monoglycerides (0% for TP, 50% for GMS) was responsible for this physical destabilization. These results are in agreement with those reported by Jenning and Gohla (2000). Decrease in entrapment efficiency on long-term storage (3 months) was 3.48% with F-TP, 2.18% with F-CP and 4.65% with F-GMS (Table 2). SLN prepared with wax (CP) had shown excellent stability with low expulsion (2.18%). Lipids of less ordered crystal lattices such as GMS, though favored drug inclusion, were unstable due to the presence of monoglycerides. The presence of surfactant in the SLN led to reduced crystallinity and slow transition of the lipid particles

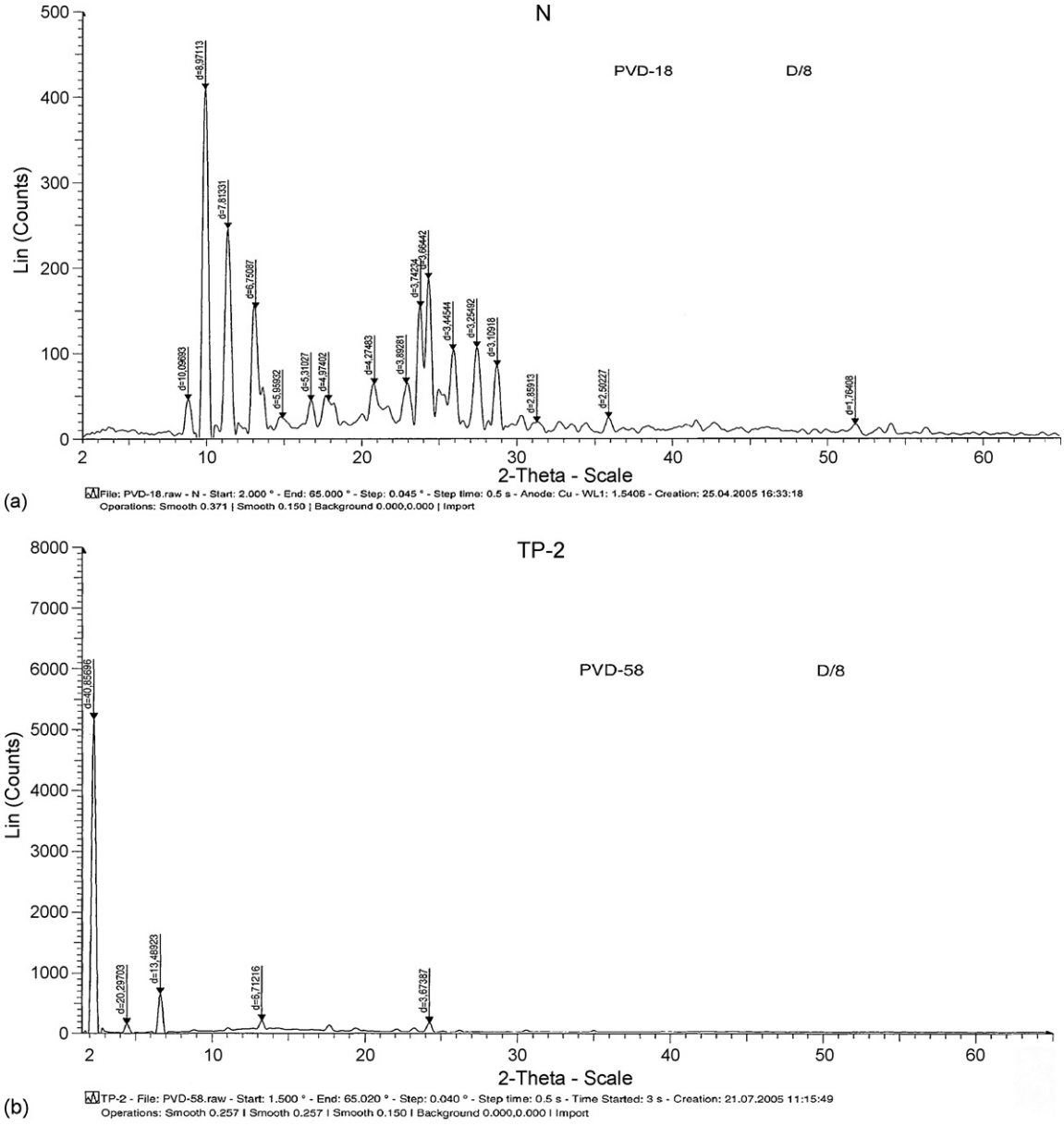
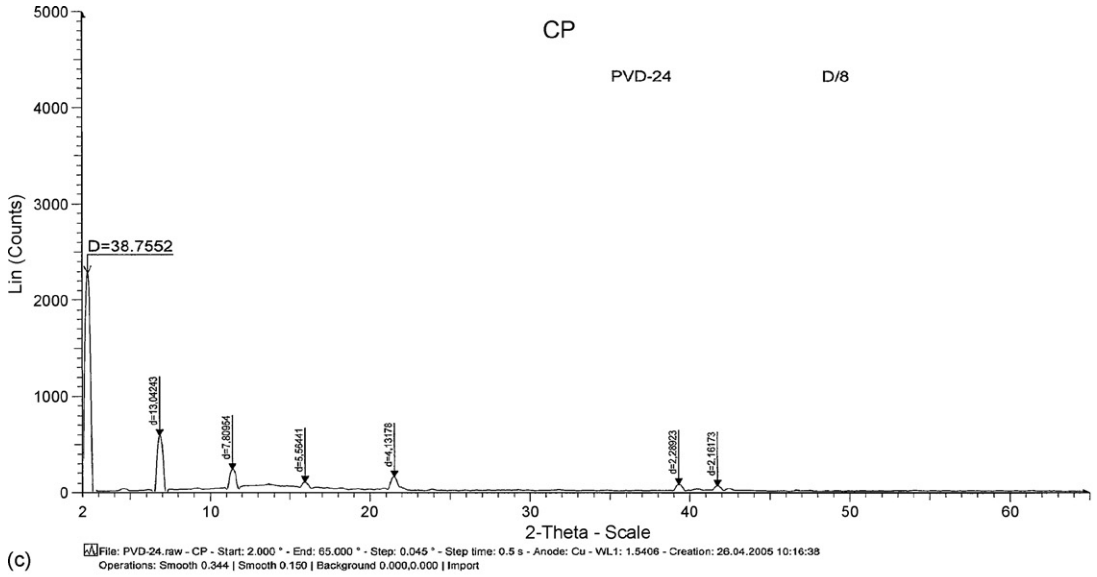


Fig. 2. P-XRD patterns of drug, lipid and lyophilized formulations. (a) P-XRD pattern of nitrendipine; (b) P-XRD pattern of tripalmitin; (c) P-XRD pattern of cetyl palmitate; (d) P-XRD pattern of glyceryl monostearate; (e) P-XRD pattern of F-TP (tripalmitin SLN); (f) P-XRD pattern of F-CP (cetyl palmitate SLN); (g) P-XRD pattern of F-GMS (glyceryl monostearate SLN).



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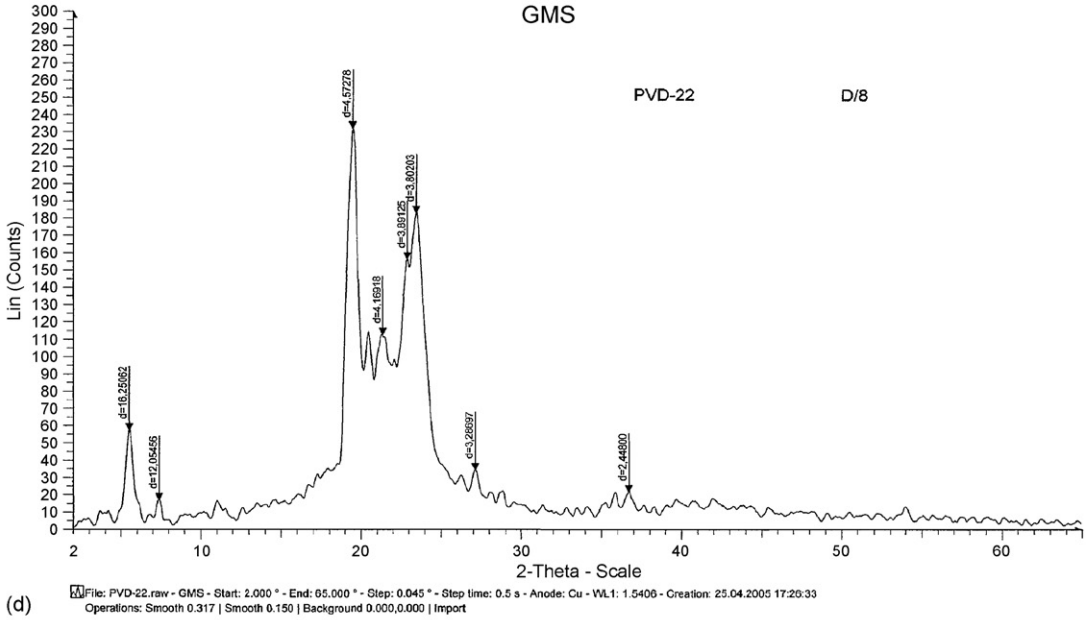

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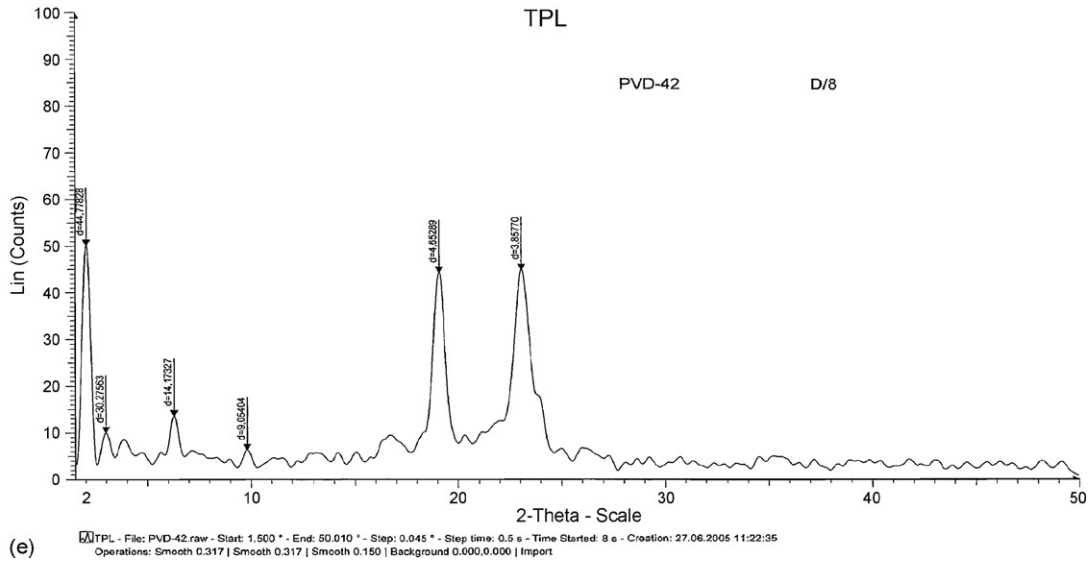

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Fig. 2. (*Continued*)

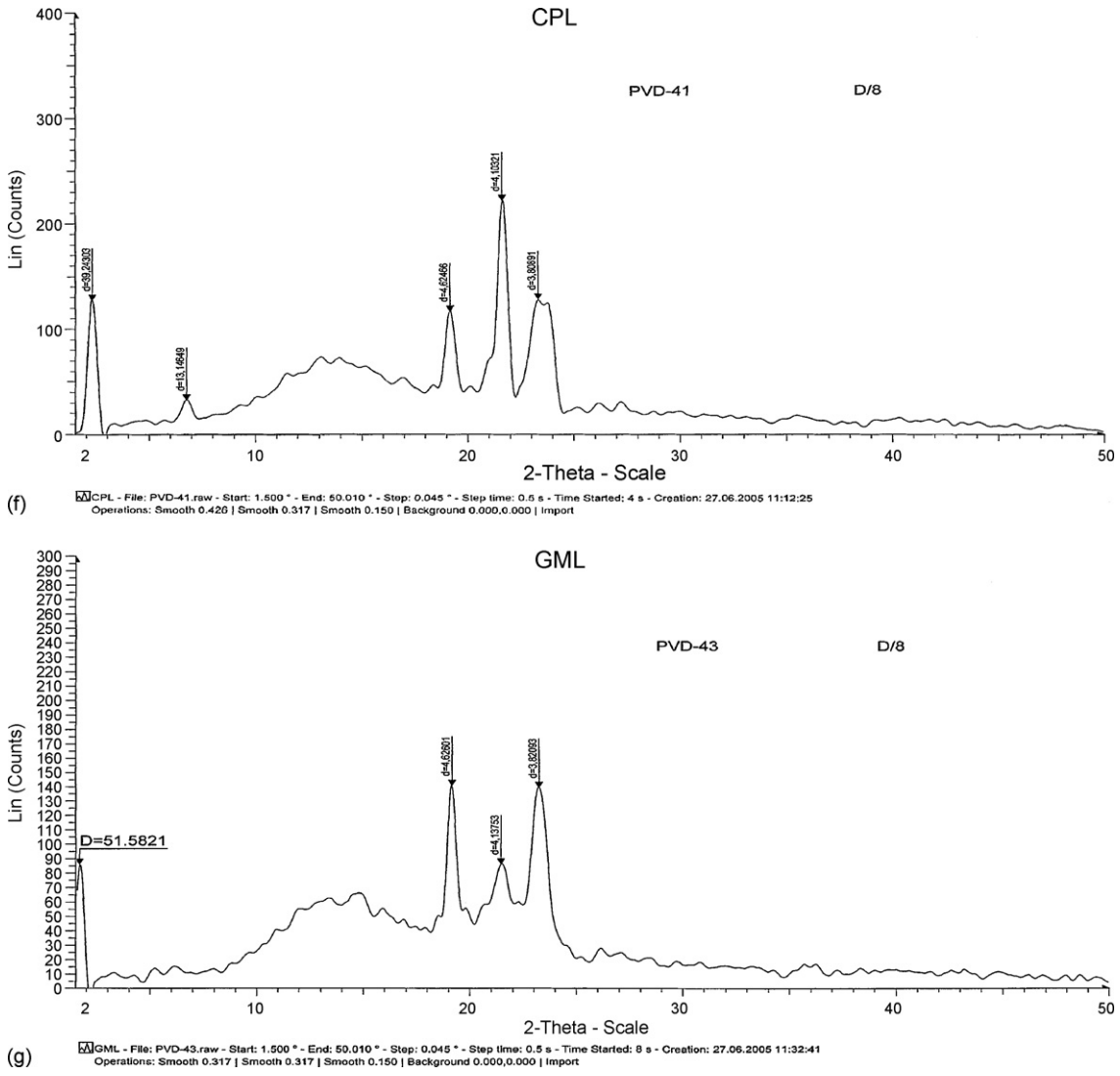

Fig. 2. (*Continued*).

thereby influencing the entrapment efficiency on long term storage.

3.5. Powder X-ray diffractometry

P-XRD patterns of nitrendipine have given sharp peaks at 2θ-scattered angles of 10.5, 11.5 and 13.5, these were indicating crystalline nature of drug (Fig. 2a). The crystallinity of pure lipids is shown in the P-XRD patterns (Fig. 2b–d). The crystalline peaks of nitrendipine were absent in the lyophilized SLN samples (Fig. 2e–g) indicating that the drug was not in crystalline form. Intensity of pure lipid peaks was also decreased in the lyophilized SLN samples submitted to XRD analysis. This reduced intensity indicates the decreased crystallinity of lipid in the SLN formulations.

3.6. In vitro release of nitrendipine from SLN

Fig. 3 shows the cumulative percent release of nitrendipine from F-TP, F-CP and F-GMS. Percentage of nitrendipine released from SLN formulations up to 48 h was 31.12% with F-TP, 26.7% with F-CP and 37.8% with F-GMS. Interestingly, the particle size had no influence on the in vitro release of nitrendipine as F-GMS with larger particle size (136.2 nm) showed high

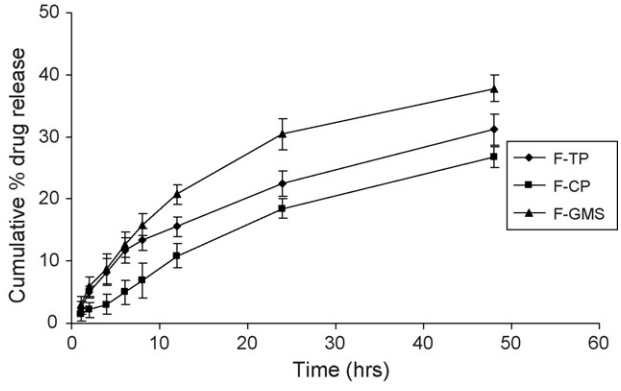

Fig. 3. Effect of lipid (tripalmitin (TP), cetyl palmitate (CP) and glyceryl monostearate (GMS)) on in vitro release of nitrendipine from SLN (mean ± S.D., $n = 3$).

Table 3
Pharmacokinetic parameters (mean ± S.D., $n = 6$) of nitrendipine after intraduodenal administration of nitrendipine suspension and SLN of different lipids (tripalmitin, cetyl palmitate and glyceryl monostearate)

Parameter	F-CP	F-TP	F-GMS	Suspension
T_{max} (h)	2	1.667 ± 0.058	2	2
C_{max} (μg/ml)	0.91 ± 0.06*	0.82 ± 0.07*	0.92 ± 0.045*	0.32 ± 0.01
K (h^{-1})	0.04 ± 0.01	0.035 ± 0.009	0.027 ± 0.005	0.03 ± 0.01
$t_{1/2}$ (h)	21.6 ± 1.8*	22.5 ± 1.7*	23.1 ± 0.9	15.02 ± 1.5
$AUC_{(0-\infty)}$ (μg/ml h)	5.23 ± 0.1*,†,‡	6.24 ± 0.11*	6.65 ± 0.24*	1.69 ± 0.13
MRT (h)	21.9 ± 1.45*	26.5 ± 2.05*	21.9 ± 1.9*	16.2 ± 1.1*
F (%)	309.46	369.23	393.49	100

† Statistical significance with F-TP, $p < 0.05$.
‡ Statistical significance with F-GMS, $p < 0.05$.
* Statistical significance with control, $p < 0.05$.

release (37.8%). The release of a drug from the SLN can be influenced by nature of the lipid matrix, surfactant concentration and production parameters (Muller et al., 2000). Since the surfactant concentration was optimized at 1% in the present investigation, the drug release profile was affected by other parameters such as lipid nature, solubility of the drug in lipid and partition coefficient. All the three formulations exhibited a controlled release with <40% drug released upto 48 h. The release profiles of these SLN resemble the drug enriched core model (Wissing et al., 2004). In such a model, the drug enriched core is surrounded by a practically drug-free lipid shell. Due to the increased diffusional distance and hindering effects by the surrounding solid lipid shell, the drug has a sustained release profile. Similar sustained release of clozapine was observed from TP-SLN prepared by the homogenization followed by ultrasonication method (Venkateswarlu and Manjunath, 2004). It was clear from the release profile (Fig. 3) that wax lipid (F-CP) had more pronounced sustained release than the glycerides. The in vitro release data correlates well with the observed stability data where the wax SLN had excellent long-term stability with good drug retaining capacity (Table 2). Among the glycerides, TP had shown slow release than GMS which can be attributed to the hydrophobic long chain fatty acids of the triglyceride that retain lipophilic drugs.

3.7. Pharmacokinetic parameters

In this study, four different formulations (F-TP, F-CP, F-GMS and nitrendipine suspension) were given to rats as intraduodenal administration. Plasma concentration–time curves of nitrendipine after intraduodenal administration are shown in Fig. 4. Peak plasma concentration (C_{max}) for nitrendipine suspension observed was 0.32 ± 0.01 μg/ml at 2 h. There were insignificant ($p > 0.05$) differences in T_{max} values for F-CP and F-GMS except for F-TP, where T_{max} was found to be 1.67 h. All SLN formulations of nitrendipine displayed significantly ($p < 0.05$) higher values of C_{max} compared to that of suspension formulation. Peak plasma concentrations were in the following order: F-TP > F-GMS > F-CP > suspension. The $AUC_{(0-\infty)}$ of all three SLN formulations (6.65 ± 0.24 μg-h/ml with F-GMS, 6.24 ± 0.11 μg-h/ml with F-TP and 5.23 ± 0.1 μg-h/ml with F-CP) were significantly ($p < 0.05$) higher than that of nitrendipine

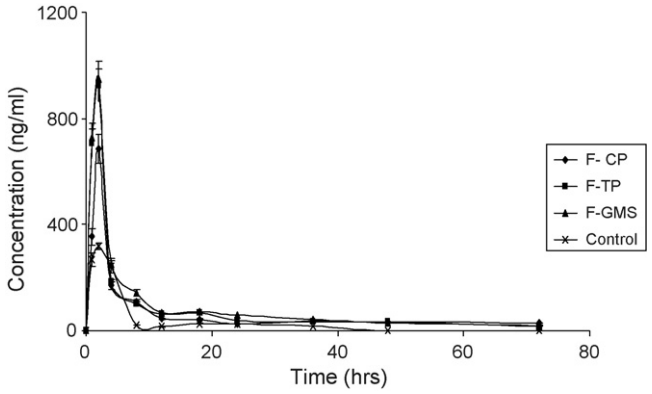

Fig. 4. Mean plasma concentration of nitrendipine–time curves after intraduodenal administration of nitrendipine suspension and SLN of tripalmitin, cetyl palmitate and glyceryl monostearate ($n = 6$).

suspension (1.69 ± 0.13 μg-h/ml). Increase in $AUC_{(0-\infty)}$ for SLN might be due to the avoidance of first pass metabolism by lymphatic transport because poor bioavailability of nitrendipine was mainly because of first pass metabolism. Increase in $AUC_{(0-\infty)}$ suggests that solid lipid nanoparticles of nitrendipine were taken up by Peyer's patch of intestine (Mansbach and Nevin, 1998; Cavalli et al., 2003). Lipid nature, fatty acid chain length and hydrophobicity will influence the lymphatic uptake (Nordskog et al., 2001; Holm et al., 2001; Ros, 2000). In vitro release data of nitrendipine loaded SLN formulations also showed more release with F-GMS than other two SLN formulations. Esterification of glycerol by long chain fatty acids is responsible for high hydrophobicity of glycerides; this influences the lymphatic uptake of F-TP and F-GMS. The higher chain length of fatty acid present in GMS is responsible for more lymphatic uptake. Mean residence time (MRT) was also increased significantly ($p < 0.05$) when compared with control suspension (Table 3).

4. Conclusion

In the present study, nitrendipine loaded SLN were prepared by homogenization followed by ultrasonication method using different lipids. The method resulted in consistent production of smaller size nanoparticles in the range of 110–138 nm with narrow size distribution and good entrapment efficiency. The sur-

factant (poloxamer) concentration was optimized at 1% based on the particle size and entrapment efficiency. The stability data and in vitro release profile indicated controlled release of the drug and excellent physical long-term stability in cetyl palmitate SLN. However the plasma pharmacokinetics after intraduodenal administration of SLN and nitrendipine suspension to rats has shown an increased $AUC_{(0-\infty)}$ for glyceride SLN (TP and GMS) followed by wax SLN (CP). Enhanced bioavailability of nitrendipine (3.09–3.93 times) was observed with all the SLN formulations. The current investigation illustrates the effect of lipid nature on the entrapment efficiency, in vitro release and bioavailability of lipophilic drug.

Acknowledgements

Authors wish to thank Dr. J.S. Yadav, Director, IICT for providing necessary facilities and encouragement. Authors also thank Dr. K. Ravikumar, IICT for P-XRD studies.

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