International Journal of Pharmaceutics 383 (2010) 1–6

Contents lists available at ScienceDirect

International Journal of Pharmaceutics

journal homepage: www.elsevier.com/locate/ijpharm

Solid self-emulsifying nitrendipine pellets: Preparation and *in vitro*/*in vivo* evaluation

Zhiyuan Wang, Jin Sun, Yongjun Wang, Xiaohong Liu, Yanhua Liu, Qiang Fu, Ping Meng, Zhonggui He*

Department of Biopharmaceutics, School of Pharmacy, Shenyang Pharmaceutical University, No. 103 of Wenhua Road, Shenyang 110016, China

ARTICLE INFO

Article history:
Received 27 May 2009
Received in revised form 30 July 2009
Accepted 13 August 2009
Available online 19 August 2009

Keywords:
Solid self-emulsifying drug delivery systems
Pellets
Extrusion/spheronization
Nitrendipine
Oral bioavailability

ABSTRACT

Objective of this study is to develop and evaluate the new solid self-emulsifying (SE) pellets of poorly soluble nitrendipine (NTD). These pellets were prepared via extrusion/spheronization technique, using liquid SEDDS (NTD, Miglyol® 812, Cremophor® RH 40, Tween 80, and Transcutol® P), adsorbents (silicon dioxide and crospovidone), microcrystalline cellulose and lactose. The resulting SE pellets with 30% liquid SEDDS exhibited uniform size (800–1000 μm) and round shape, droplet size distribution following self-emulsification was nearly same to the liquid SEDDS (72 ± 16 nm and 64 ± 12 nm). The *in vitro* release was similar for the two SE formulations (over 80% within 30 min), both significantly higher than the conventional tablets (only 35% within 30 min). The oral bioavailability was evaluated for the SE pellets, liquid SEDDS and conventional tablets in fasted beagle dogs. AUC of NTD from the SE pellets showed 1.6-fold greater than the conventional tablets and no significant difference compared with the liquid SEDDS. In conclusion, our studies illustrated that extrusion/spheronization technique could be a useful large-scale producing method to prepare the solid SE pellets from liquid SEDDS, which can improve oral absorption of NTD, nearly equivalent to the liquid SEDDS, but better in the formulation stability, drugs leakage and precipitation, etc.

1. Introduction

Nowadays, an increasing number of new chemical entities and many existing drugs exhibit low solubility in water, which may lead to poor oral absorption, high intra- and inter-subject variability and lack of dose proportionality (Lipinski, 2002). Thus, for such compounds of BCS II type, the absorption rate and degree from the gastrointestinal tract (GIT) are usually controlled and limited by dissolution process (Amidon et al., 1995). To overcome the problem, various formulation strategies have been adopted including the use of cyclodextrins, nanoparticles, solid dispersions and permeation enhancers (Aungst, 1993).

In recent years, much attention has been paid to self-emulsifying drug delivery systems (SEDDS), which have shown lots of reasonable successes in improving oral bioavailability of poorly soluble drugs (Kommuru et al., 2001; Gursoy and Benita, 2004; Kang et al., 2004). SEDDS are usually composed of a mixture of oil and surfactant or cosurfactant and are capable of forming fine oil-in-water emulsions upon gentle agitation provided by the GIT motion. After oral administration, SEDDS can maintain the poorly soluble drugs dissolved in the fine oil droplets when transiting through the GIT.

However, traditional preparations of SEDDS are usually prepared in the liquid state. So the liquid SEDDS are generally enclosed by soft or hard capsules to facilitate oral administration but it produce some disadvantages, such as high production costs, low drug incompatibility and stability, drugs leakage and precipitation, capsule ageing. Then incorporation of liquid SEDDS into a solid dosage form is compelling and desirable, and some solid self-emulsifying (SE) dosage forms have been initially explored, such as SE tablet and pellets (Nazzal et al., 2002; Newton et al., 2007).

As a multiple-unit dosage form, pellets have some desired advantages, such as flexibility in designing and developing solid dosage form, reduction of intra- and inter-subject variability of drug dissolution and plasma profiles, hence improvement of the drug safety and efficacy (Abdalla and Mäder, 2007). To prepare pellets, extrusion/spheronization technique has become popular in pharmaceutical industry because it is easily large-scale, and its products have many features, including spherical shape, narrow modal size distribution, good flow properties, low friability and uniform packing characteristics. The SE pellets combine both advantages of SEDDS and pellets, and the extrusion/spheronization technique has been firstly introduced to prepared the SE pellets by Newton et al. (2001). In addition, Tuleu et al. (2004) has made a comparative bioavailability study of progesterone self-emulsifying formulations presented in a pellet and liquid form in dogs and the bioavailability of the two dosage forms was equivalent.

* Corresponding author. Tel.: +86 24 23986321; fax: +86 24 23986321.
E-mail address: hezhonggui@gmail.com (Z. He).

0378-5173/$ – see front matter © 2009 Elsevier B.V. All rights reserved.
doi:10.1016/j.ijpharm.2009.08.014

In general, limited investigations have focused on the incorporation of liquid SEDDS into a solid dosage form until now. Hereby, we intended to develop, prepare, *in vitro* and *in vivo* evaluate the novel solid SE pellets for the oral delivery of poorly soluble drugs. Nitrendipine (NTD) was selected as the model drug, a calcium channel blocker as an antihypertensive drug, which is practically insoluble in water and has poor oral bioavailability (10–20%). In the study, we firstly prepared the liquid SEDDS containing NTD, solidified it with adsorbents, microcrystalline cellulose and lactose, and then prepared the SE pellets via the extrusion/spheronization technique. Finally, the oral bioavailability of NTD was evaluated for the SE pellets, liquid SEDDS and the commercial conventional tablets in the fasted beagle dogs.

2. Materials and methods

2.1. Materials

Nitrendipine was bought from Nanjing Pharm. Co. (Nanjing, China). Conventional NTD tablets were purchased from Shanghai Quanyu Pharm. Co. (Shanghai, China). Miglyol® 812 (MCT, caprylic/capric triglyceride) was purchased from Condea Chemie GmbH (Witten, Germany). Transcutol® P (diethylene glycol monoethyl ether) was kindly donated by Gattefossé (Saint-Priest, France). Tween 80 (polyoxyethylene-20 sorbitan monooleate) was kindly provided by Croda (U.K.). Cremophor® RH 40 (PEG-40 hydrogenated castor oil) and Kollidon® CL-SF (crospovidone) were obtained from BASF (Ludwigshafen, Germany). SYLOID® 244 FP (porous silicon dioxide) and FLOWLAC® 100 (lactose monohydrate) were gifts from Grace Davidson (Columbia, Md.) and Meggle GmbH (Wasserburg, Germany) respectively. Avicel® PH 101 (MCC, microcrystalline cellulose) was purchased from Asahi Kasei (Tokyo, Japan), and was used as a pellet forming material. HPLC-grade acetonitrile and methanol were purchased from Fisher Scientific (Pittsburgh, PA, USA). Water was prepared using an EASYPURE® II RF/UV ultrapure water system (Barnstead International Co., Boston, MA, USA). All other materials were of analytical grade and were used as received.

2.2. Methods

2.2.1. Preparation of the liquid SEDDS

Based on the pilot studies (equilibrium solubility, pseudo-ternary phase diagram and supersaturation studies), a self-emulsifying system in which the equilibrium solubility of NTD was 9.62% (w/w), containing 6.25% of the drug (w/w), could be diluted with water (1:100) without precipitation within 2 h. The composition of the optimized blank SEDDS is 30% Miglyol® 812 (oil), 60% Cremophor® RH 40 and Tween 80 (surfactant, 2:1), 10% Transcutol® P (cosurfactant). The preparation of the SEDDS involved the following steps:

- Mixing of Miglyol® 812, Cremophor® RH 40, Tween 80 and Transcutol® P at 50 °C with a magnetic stirrer.
- Dissolving NTD in the blank SEDDS with stirring until forming an isotropic mixtures.
- Cooling to room temperature and equilibrating for 24 h before use.

2.2.2. Preparation of the SE pellets

The compositions of the liquid SEDDS and the SE pellets are given in Table 1. The pellets from formulations 2, 3, 4, and 5 were produced by the following three processes: Initially, the above-mentioned SEDDS were added into SYLOID® 244 FP or the mixtures of SYLOID® 244 FP and Kollidon® CL-SF, mixing in a kneader for 10 min until the liquid SEDDS were adsorbed completely to form a powder with fine flowability. And then, the adsorbed mixtures were blended with MCC or lactose for further 5 min, followed by addition of water until a mass suitable for extrusion was obtained (water:total mixtures = 0.6:1, w/w). Eventually, the wet mass was extruded at 30 rpm in an axial screen extruder (WL 350, Wenzhou, China) equipped with a die of 1 mm thickness with 1 mm diameter circular holes. The extrudate was spheronized in 100 g quantities for 5 min at 1800 rpm on a 250 mm radial plate spheronizer (WL 350, Wenzhou, China) using a cross-hatch frictional plate of 3 mm × 3 mm pitch and 1.2 mm depth. The produced pellets were then dried for 12 h at 40 °C in a drying oven.

Additionally, for formulation 1, the same content of 30% SEDDS was directly added into MCC, mixed to form a powder with poor flowability, and then followed by addition of suitable water (water:total mixtures = 0.4:1, w/w). The extrusion, spheronization and drying were performed under the same conditions as what was mentioned above.

2.2.3. Characterization of pellets

2.2.3.1. Size distribution and shape evaluation of the pellets. Vibrating by hands for 5 min, a set of Chinese Standard Sieves (2000, 1200, 1000, 800, and 500 μm) were used for size distribution determinations of 50 g of the produced pellets. Shape evaluation was performed on pellets within the 800–1000 μm fraction using a microscope (DMBA 450 Motic Digital Microscope, China) with an optical zoom of 40×/0.17 and an eye piece of 10×/22. The subsequent characterizations were carried out on the same modal fraction of 800–1000 μm.

2.2.3.2. Emulsion droplet size determination. The size of the emulsion droplets released from the SE pellets was determined in water at 37 °C, compared to liquid SEDDS with or without the drug. Pellets (1 g) and SEDDS (0.3 ml) were gently agitated in 30 ml distilled water with a magnetic stirrer. Samples were taken after 30 min and filtered through 0.45 μm micropore filters. The droplet size of resultant emulsions was determined by Laser diffractometry (LD) using a Coulter LS 230 (Beckmann-Coulter Electronics, Germany).

2.2.3.3. Crushing strength of pellets. The crushing strength of the pellets was analyzed by diametral compression using a tablet hardness tester (YP-200B, Shanghai, China) with a 10-kg load cell and a speed of 1.0 mm/min. Ten pellets from each formulation

Table 1
Compositions of the liquid SEDDS and the SE pellets.

Liquid SEDDS		SE pellets					
Ingredients	% (w/w)	Ingredients % (w/w)	F. 1	F. 2	F. 3	F. 4	F. 5
NTD	6.25	SEDDS	30	30	30	30	30
Miglyol® 812	28.13	Kollidon® CL-SF	–	–	15	15	15
Cremophor® RH 40	37.50	SYLOID® 244 FP	–	15	15	5	5
Tween 80	18.75	Avicel® PH 101	70	55	40	50	35
Transcutol® P	9.37	FLOWLAC® 100	–	–	–	–	15

F.: formulation.

were tested. The crushing force was converted into surface tensile strength using the following equation:

$$\sigma_{f(s)} = \frac{0.4F}{\pi R^2}$$

where $\sigma_{f(s)}$ is the surface tensile strength (Pa), F is the crushing force (N), and R is the radius (m).

2.2.3.4. Disintegration test of pellets. The disintegration of the pellets was studied in deionized water at 37 °C using a disintegration apparatus (Ch.P. 2005, modified with a 0.2 mm mesh at the base of the tubes). The 0.3 mg pellet samples from each formulation were tested ($n = 3$). The endpoint was taken as the time at which no obvious particles were present on the sieve in each disintegration basket.

2.2.4. In vitro dissolution test

The dissolution test was carried out for 1 h at 100 rpm by paddle method specified in the Chinese Pharmacopoeia 2005. The dissolution medium was 200 ml hydrochloric acid solution (pH 1.2) containing 0.5% (w/v) Tween 80. The temperature of the dissolution medium was controlled at 37 ± 0.5 °C. The optimal SE pellets, liquid SEDDS and conventional tablets weighed to be equivalent to 10 mg NTD were used for the dissolution test. Five milliliters of the dissolution medium were sampled at appropriate intervals, and fresh dissolution medium was simultaneously replenished in the apparatus to maintain a constant volume. The withdrawn sample was passed through a micropore filter (0.8 μm), and the filtrate was assayed by UV spectrophotometer (UV-1801, Beijing Ruili Analytical Instrument Co., China) at 353 nm to determine the dissolved drug concentration. Each release test was carried out in triplicate.

2.2.5. In vivo bioavailability studies

2.2.5.1. Experimental design. Six healthy male beagle dogs (10–12 kg), fasted but free access to water for 12 h prior to the experiment, were used in the study. They were allocated at random to three treatment groups and administered orally hard capsules containing the optimal SE pellets and liquid SEDDS of NTD, and conventional NTD tablets in a crossover design with 1 week washout between dosing. The dose of NTD administered to the dogs was 20 mg. The animal studies were approved by the Shenyang Pharmaceutical University Animal Care and Use Committee.

Blood samples (3.0 ml) were collected from the leg veins with a heparinized tube at 0, 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, 24 and 48 h post-dosing. After collection, the plasma was obtained by the centrifugation of blood at 3000 rpm for 30 min and then kept frozen at −20 °C until analysis. The NTD concentrations of plasma were determined by ultra performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) with a lower limit of quantitation of 0.5 ng/ml.

2.2.5.2. Quantitative analysis of NTD in plasma. A selective, rapid and sensitive UPLC-MS/MS method was developed for the quantification of NTD in dog plasma. With propranolole hydrochloride as an internal standard, sample pretreatment utilized a simple liquid–liquid extraction. A 100 μL methanol solution of internal standard (100 ng/ml) was added to 0.5 ml plasma. After vortex-mixing for 30 s, 3 ml diethyl ether was added and vortexed for 3 min. After centrifugation at 3000 rpm for 10 min, the organic layer was collected and dried under nitrogen gas at 40 °C. The residue was reconstituted with a 200 μl water and acetonitrile (1:1). The separation was carried out on an ACQUITY UPLC™ BEH C18 column (50 mm × 2.1 mm, 1.7 μm; Waters Co., Milford, MA, USA) with water (containing 0.1% formic acid) and acetonitrile as the mobile phase under gradient conditions at a flow rate of 0.2 ml/min.

The detection was performed by a Waters Tandem Quadrupole (TQ) Detector (Waters, USA). The mass spectrometer was operated with an electrospray ionization (ESI) interface in positive ionization mode and with multiple-reaction monitoring mode. The selected reaction monitoring (SRM) of NTD and the internal standard were m/z 361.0 → 315.0 and m/z 260.2 → 116.1, respectively. The concentration of NTD was determined by standard linear calibration curve in the concentration range of 0.5–500 ng/ml.

2.2.5.3. Pharmacokinetic data analysis. The peak plasma concentration (C_{max}) and the time for their occurrence (T_{max}) were obtained directly from the individual plasma concentration versus time profiles. The area under the concentration–time curve (AUC) was estimated according to the linear trapezoidal rule. The experimental results were analyzed by a two-sample t-test, with the level of statistic significance set at $P < 0.05$. The relative bioavailability (F) of SE pellets and liquid SEDDS to the conventional tablets (reference) was calculated using the following equation:

$$F = \frac{AUC_{test}}{AUC_{reference}} \times 100\%$$

3. Results and discussions

3.1. Preparation of the liquid SEDDS

To formulate and prepare SEDDS, there were some basic guidelines needed to conform: safety, compatibility, drug solubility, efficient self-emulsification efficiency and droplet size, etc. (Kommuru et al., 2001; Zhang et al., 2008). In the current study, the liquid SEDDS were prepared and optimized based on the pilot studies, such as equilibrium solubility, pseudo-ternary phase diagram and supersaturation studies (data not shown).

As for the optimized SEDDS (Table 1), Miglyol® 812 was used as the oil. Besides the higher solubility of NTD, Miglyol® 812 could form a stable emulsion when diluted with water and had less possibility of drug precipitation. SEDDS containing single Cremophor® RH 40 as surfactant could form a gel-like structure region by dilution, useful for maintaining the supersaturation state. But the gel-like structure required a longer time to disperse, so the mixture of Cremophor® RH 40 and Tween 80 (2:1) were utilized as surfactants. Transcutol® P was selected as the cosurfactant, due to higher drug solubility and good capability to form stable emulsion. The SEDDS systems containing 6.25% of NTD (w/w), were efficiently emulsified within 3 min when in contact with water at 37 °C and no phase separation was observed.

3.2. Preparation of the SE pellets

To prepare satisfactory SE pellets by extrusion/spheronization, MCC was used as a forming material for its ability to retain water during the producing process. In our pilot studies, we found that more amount of MCC was added in the formulation, more easily the pellets were formed. Moreover, considering drug loading and formation of pellets, the amount of liquid SEDDS added was fixed at 30%. However, without the aid of adsorbents, the pellets containing 30% SEDDS appeared poor physical properties (poor flowability, easy agglomeration and low hardness), even when 70% MCC was used (formulation 1). Podczeck (2008) has reported the pellets can be prepared with the use of colloidal silica itself by extrusion/spheronization. So SYLOID® 244 FP was used as adsorbent to adsorb the liquid SEDDS to form a fine flowable powder (the qualitative ratio of SEDDS to SYLOID® 244 FP was 2:1), and then pellets with good physical properties could be successfully prepared. But as for formulation 2, without the aid of disintegrant, the pellets could not disintegrate within 2 h, which would influence the drug

Table 2
Percentage of the optimal SE pellets in each size range (μm).

Size range (μm)	Percentage of pellets (%)
>2000	0
2000–1200	6.01
1200–1000	20.52
1000–800	51.61
800–500	21.86

release. Therefore, Kollidon® CL-SF was added as a disintegrant and it also could be combined with SYLOID® 244 FP as adsorbent to form a fine flowable powder (the qualitative ratio of SEDDS to Kollidon® CL-SF was 1:1). Additionally, incorporation of lactose into the formulation could be useful to improve the appearance of the pellets and might be expected to favor the disintegrating performance. Table 1 gives the compositions of the different SE pellets.

3.3. Characterization of SE pellets

3.3.1. Size distribution and shape evaluation of the pellets

Details of the percentage of pellets in the sieve fraction of the definite formulation 5 are presented in Table 2. The size of modal fraction was 800–1000 μm and the production of the modal pellets was about 52%. The shape of pellets was evaluated by a digital microscope and is shown in Fig. 1. From the microgram, the pellets have good physical properties and without any apparent agglomeration.

3.3.2. Droplet size determination

Droplet size distribution following self-emulsification is a critical factor to evaluate self-emulsifying systems *in vitro*. However, the influence of emulsion droplet size in the *in vivo* performance of the formulation is not yet clear. Nielsen et al. (2008) has demonstrated bioavailability enhancement of probucol through the reduction in the emulsion droplet size. The possible explanation to the enhanced absorption could be that the smaller the droplet size, the larger the interfacial surface area, which facilitate and improve drug absorption. Moreover, some authors have paid much attention to the drug solubilization capacity in the lipid formulations when dilution and digestion in the GIT, which would influence the subsequent dispersion and absorption of drug (Gao et al., 2003; Kossena et al., 2004; Porter et al., 2004).

In our study, the droplet size was determined by means of laser diffractometry and expressed as the average value. The droplet size distribution of the different SEDDS formulations is shown in Fig. 2. The average dispersing droplet sizes of blank SEDDS, liquid SEDDS

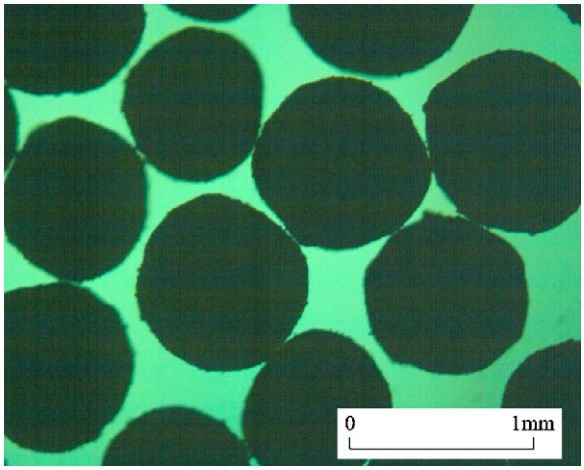

Fig. 1. Microscopic picture of the optimal SE pellets.

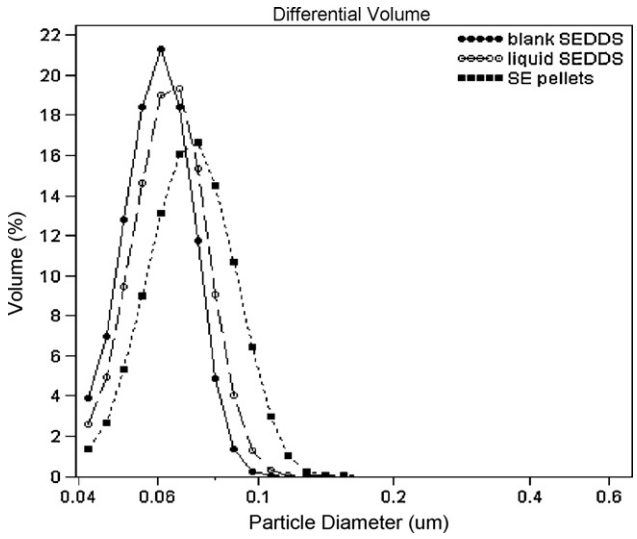

Fig. 2. The droplet size distribution of the reconstitution emulsions from blank SEDDS (●), liquid SEDDS (○) and the optimal SE pellets (■).

with drug and SE pellets of the formulation 5 were 61 ± 10 nm, 64 ± 12 nm and 72 ± 16 nm respectively. The result indicated a microemulsion system (droplet size less than 100 nm) could be formed at the definite proportion of the lipid excipients in SEDDS (Table 1). Moreover, the mean droplet size increased slightly for the drug-loaded formulations, maybe due to very small amounts of drug precipitated by dilution with time.

3.3.3. Crushing strength and disintegration

The results of crushing strength and disintegration test are shown in Table 3. Crushing strength (Newton et al., 2001) of the SE pellets was significantly affected by the amount of liquid SEDDS and physical adsorbents. Increasing the amounts of SEDDS of the pellets would weaken the interactions within the pellets and decrease their hardness. Nevertheless, if the SEDDS were not adsorbed by the physical adsorbents, the $\sigma_{f(s)}$ of the produced pellets (formulation 1) with 30% liquid SEDDS was 0.63 MPa, far lower than that of the pellets (formulations 2, 3, 4, and 5) with adsorbed SEDDS (about 5 MPa). Moreover, with the aid of physical adsorbents the preparation process and appearance of SE pellets were improved to a larger extent.

However, the disintegration time of SE pellets would affect the drug release. Tuleu et al. (2004) has used a low quantity of ethanol to increase the porosity of the SE pellets, which can aid in the disintegration of pellets with subsequent improved release of the drug. In the study, Kollidon® CL-SF was chose as disintegrant. To the formulations 2 and 3, incorporation of Kollidon® CL-SF had less influence on the crushing strength of the produced pellets ($\sigma_{f(s)}$ = 4.91 and 5.12 MPa) but highly affected the disintegration time (more than 2 h vs. 25.66 min). Moreover, decreasing the quantity of SYLOID® 244 FP and increasing the relative quantity of MCC would be useful to the disintegration of the pellets (F. 4 vs. F. 3). But 5% colloidal silica was needed to adsorb the SEDDS and would also be benefi-

Table 3
Crushing force *F*, tensile strength $\sigma_{f(s)}$ and disintegration time of the SE pellets.

Formulation number	*F* (N)	$\sigma_{f(s)}$ (MPa)	Disintegration time (min)
1	4.90 ± 0.98	0.63 ± 0.13	30.33 ± 4.56
2	38.56 ± 3.01	4.91 ± 0.38	>120
3	40.19 ± 1.65	5.12 ± 0.21	25.66 ± 3.23
4	37.21 ± 2.43	4.74 ± 0.31	15.41 ± 2.60
5	37.89 ± 1.50	4.83 ± 0.19	15.25 ± 2.32

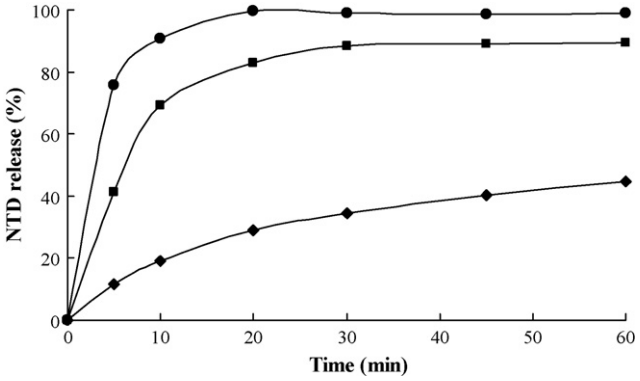

Fig. 3. *In vitro* dissolution profiles of the optimal SE pellets (■), liquid SEDDS (●) and conventional tablets (♦).

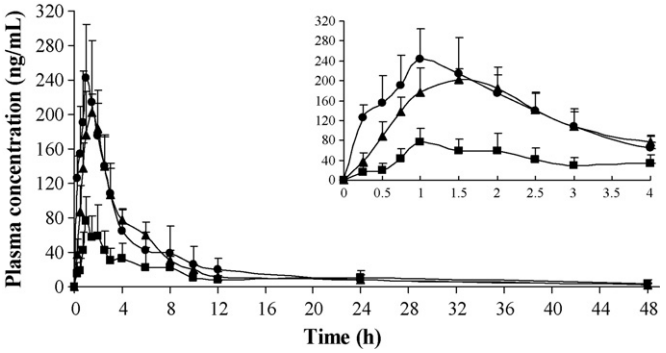

Fig. 4. Mean plasma concentration versus time curves of NTD after oral administration of the optimal SE pellets, liquid SEDDS and conventional tablets to beagle dogs. (▲) SE pellets, (●) liquid SEDDS, (■) conventional NTD tablets.

cial to the flowability of the mixed powder. The addition of lactose had less effect on disintegration of the pellets (F. 5 vs. F. 4), but which would be useful to improve the appearance of the prepared pellets. The disintegration time of the pellets of formulation 1 was 30.33 min, but their crushing strength was the lowest. These results clearly demonstrated that with increasing crushing strength of SE pellets, incorporation of disintegrants would be helpful to the disintegration process and the relative quantity of SYLOID® 244 FP and MCC would also affect the disintegration time. Therefore, in the consideration of the appearance, hardness and disintegration of the SE pellets, the optimal formulation 5 was used in the following characterization tests and *in vivo* evaluation.

3.4. In vitro dissolution test

As shown in Fig. 3, the release performance of NTD from SEDDS formulations is significantly improved, compared with the conventional tablets. However, the release rate and extent of liquid SEDDS are both slightly higher than that of the optimal SE pellets. Two reasons may probably explain the result. Firstly, there is a disintegration process for the SE pellets, which will delay the first step of drug release. Secondly, the excipients such as SYLOID® 244 FP will have a relatively strong interaction with the adsorbed SEDDS, probably impairing the release rate and extent of NTD.

Moreover in recent years, considering the specificity of SEDDS, some authors considered that the conventional *in vitro* dissolution method might not better predict the release behavior of the SEDDS formulations in the GIT, because the solubilization of SEDDS might be lost more or less when dilution and digestion by GIT fluid and enzymes, leading to drug precipitation. So some *in vitro* dispersion and digestion models that are more reflective of the GIT environment have been developed (Porter and Charman, 2001; Ljusberg-Wahren et al., 2005; Dahan and Hoffman, 2006, 2007). With these methods, *in vitro* dissolution might be further correlated with *in vivo* performance of the SEDDS. But the absorption mechanism for oral solid SEDDS formulations is more complicated than liquid SEDDS, and it may not be feasible to create a single *in vitro* dissolution environment simulating the physiological conditions. In our study a general dissolution test was only used to show the advantage of SEDDS formulations compared to the conventional tablets. For the *in vivo* fate of this SEDDS formulations will be in our further study aided with suitable *in vitro* evaluation methods.

3.5. Bioavailability studies

Bioavailability studies of the two deferent SEDDS formulations of NTD compared with conventional tablets were investigated following oral administration of 20 mg drug to six healthy beagle dogs. The profiles of the mean plasma concentrations of NTD vs. time and the main pharmacokinetic parameters are shown in Fig. 4 and Table 4, respectively.

Fig. 4 indicated significantly greater improvement of drug absorption for the two SE formulations than the commercial conventional tablets. The results showed that C_{max} of liquid SEDDS and the optimal SE pellets was 3.2- and 2.4-fold higher than that of the conventional tablets. Additionally, T_{max} of the two SE formulations was all shorter than that of the conventional tablet, suggesting that SE technique could improve drug release and absorption in GIT. However, T_{max} of this SE pellets was relatively delayed compared to liquid SEDDS (1.31 h vs. 0.94 h), consistent with the dissolution performances. The relative bioavailabilities of the SE pellets and liquid SEDDS of NTD compared with conventional NTD tablets were 159.87% and 192.64%, respectively. Although the AUC of SE pellets was slightly lower than that of liquid SEDDS, there was no significantly statistical difference between the two formulations. It indicated that the adsorption of NTD was evidently improved after it was dispersed in solid SE formulations as a dissolved state.

Therefore, for poorly soluble drugs such as NTD, SEDDS can improve their solubility and maintain them as a dissolved form. Once the SEDDS enters the GIT, the spontaneous formation of an emulsion will improve drug release significantly and be beneficial to enhance absorption. Furthermore, the solid SE pellets, combining the advantages of SEDDS and pellets, will enlarge the application scope of the advanced pharmaceutical SEDDS technology.

Table 4
Relative bioavailability and pharmacokinetic parameters of NTD SE pellets, liquid SEDDS and conventional tablets administered orally to the beagle dogs (mean ± S.D., $n = 6$).

Parameters	Conventional tablets	liquid SEDDS	SE pellets
T_{max} (h)	1.62 ± 0.47	0.94 ± 0.43	1.31 ± 0.80
C_{max} (ng/ml)	97.6 ± 15.91	308.15 ± 33.19[a]	232.30 ± 67.71[a]
AUC_{0-t} (ng h/ml)	595.18 ± 270.11	1146.56 ± 223.89[a]	951.50 ± 265.79[a]
$AUC_{0-\infty}$ (ng h/ml)	624.29 ± 356.39	1243.18 ± 207.71[a]	1082.92 ± 343.69[a]
Relative bioavailability (%)	–	192.64	159.87

[a] Statistically higher than conventional tablets ($P < 0.05$).

4. Conclusion

These current results demonstrated that the SE NTD pellets with 30% of the liquid SEDDS were successfully developed by the use of extrusion/spheronization technique. The resulting SE pellets had a uniform size, a spherical shape and suitable hardness. Moreover, the self-emulsifying properties were still preserved in the pellets. Following self-emulsification in water the droplet size distribution of the SE pellets was nearly same to the liquid SEDDS, and the *in vitro* dissolution performance was similar for the liquid SEDDS and SE pellets both significantly higher than the conventional tablets. The oral bioavailability of NTD from the SE pellets was much greater than the conventional tablets and no significant difference compared with the liquid SEDDS. Therefore, the extrusion/spheronization technique is a useful alternative to prepare the solid SE pellets from the liquid SEDDS and the SE pellets could improve oral absorption of poorly soluble drug such as NTD.

Furthermore, the formation of the SE pellets is strongly dependent on the pellet compositions. To produce pellets with good quality, there exists a compromise between the least amount of MCC and the largest amount of liquid SEDDS. Using physical adsorbents to adsorb liquid SEDDS before producing pellets, the preparation process and physical characteristics of the SE pellets will be improved to a large extent. However, some attention should be paid to the retarding effect of physical adsorbents on the release of liquid from pellets, and then the oral absorption performance *in vivo*.

Acknowledgement

We are grateful for financial support from the National Basic Research Program of China (973 Program), No. 2009CB930300.

References

Abdalla, A., Mäder, K., 2007. Preparation and characterization of a selfemulsifying pellet formulation. Eur. J. Pharm. Biopharm. 66, 220–226.

Amidon, G.L., Lennernas, H., Shah, V.P., Crison, J.R., 1995. A theoretical basis for a biopharmaceutic drug classification: the correlation of in vitro drug product dissolution and in vivo bioavailability. Pharm. Res. 12, 413–420.

Aungst, B.J., 1993. Novel formulation strategies for improving oral bioavailability of drugs with poor membrane permeation or presystemic metabolism. J. Pharm. Sci. 82, 979–986.

Dahan, A., Hoffman, A., 2006. Use of a dynamic in vitro lipolysis model to rationalize oral formulation development for poor water soluble drugs: correlation with in vivo data and the relationship to intra-enterocyte processes in rats. Pharm. Res. 23, 2165–2174.

Dahan, A., Hoffman, A., 2007. The effect of different lipid based formulations on the oral absorption of lipophilic drugs: the ability of in vitro lipolysis and consecutive ex vivo intestinal permeability data to predict in vivo bioavailability in rats. Eur. J. Pharm. Biopharm. 67, 96–105.

Gao, P., Rush, B.D., Pfund, W.P., Huang, T., Bauer, J.M., Morozowich, W., Kuo, M.S., Hageman, M.J., 2003. Development of a supersaturable SEDDS (S-SEDDS) formulation of paclitaxel with improved oral bioavailability. J. Pharm. Sci. 92, 2386–2398.

Gursoy, R.N., Benita, S., 2004. Self-emulsifying drug delivery systems (SEDDS) for improved oral delivery of lipophilic drugs. Biomed. Pharmacother. 58, 173–182.

Kang, B.K., Lee, J.S., Chon, S.K., Jeong, S.Y., Yuk, S.H., Khang, G., Lee, H.B., Cho, S.H., 2004. Development of self-microemulsifying drug delivery systems (SMEDDS) for oral bioavailability enhancement of simvastatin in beagle dogs. Int. J. Pharm. 274, 65–73.

Kommuru, T.R., Gurley, B., Khan, M.A., Reddy, I.K., 2001. Self-emulsifying drug delivery systems (SEDDS) of coenzymeQ10: formulation development and bioavailability assessment. Int. J. Pharm. 212, 233–246.

Kossena, G.A., Charman, W.N., Boyd, B.J., Dunstan, D.E., Porter, C.J.H., 2004. Probing drug solubilization patterns in the gastrointestinal tract after administration of lipid-based delivery systems: a phase diagram approach. J. Pharm. Sci. 93, 332–348.

Lipinski, C.A., 2002. Poor aqueous solubility—an industry wide problem in drug discovery. Am. Pharm. Rev. 5, 82–85.

Ljusberg-Wahren, H., Nielsen, F.S., Brogard, M., Troedsson, E., Müllertz, A., 2005. Enzymatic characterization of lipid-based drug delivery systems. Int. J. Pharm. 298, 328–332.

Nazzal, S., Nutan, M., Palamakula, A., Shah, R., Zaghloul, A.A., Khan, M.A., 2002. Optimization of a self-nanoemulsified tablet dosage form of Ubiquinone using response surface methodology: effect of formulation ingredients. Int. J. Pharm. 240, 103–114.

Newton, J.M., Petersson, J., Podczeck, F., Clarke, A., Booth, S., 2001. The influence of formulation variables on the properties of pellets containing a self-emulsifying mixture. J. Pharm. Sci. 90, 987–995.

Newton, J.M., Pinto, M.R., Podczeck, F., 2007. The preparation of pellets containing a surfactant or a mixture of mono- and di-gylcerides by extrusion/spheronization. Eur. J. Pharm. Sci. 30, 333–342.

Nielsen, F.S., Petersen, K.B., Müllertz, A., 2008. Bioavailability of probucol from lipid and surfactant based formulations in minipigs: Influence of droplet size and dietary state. Eur. J. Pharm. Biopharm. 69, 553–562.

Podczeck, F., 2008. A novel aid for the preparation of pellets by extrusion/spheronization. Pharm. Technol. Eur. 20, 26–31.

Porter, C.J.H., Charman, W.N., 2001. In vitro assessment of oral lipid based formulation. Adv. Drug. Deliv. Rev. 50, S127–S147.

Porter, C.J.H., Kaukonen, A.M., Taillardat-Bertschinger, A., Boyd, B.J., O'Connor, J.M., Edwards, G.A., Charman, W.N., 2004. Use of in vitro lipid digestion data to explain the in vivo performance of triglyceride-based oral lipid formulations of poorly water-soluble drugs: studies with halfantrine. J. Pharm. Sci. 93, 1110–1121.

Tuleu, C., Newton, M., Rose, J., Euler, D., Saklatvala, R., Clarke, A., Booth, S., 2004. Comparative bioavailability study in dogs of a self-emulsifying formulation of Progesterone presented in a pellet and liquid form compared with an aqueous suspension of Progesterone. J. Pharm. Sci. 93, 1495–1502.

Zhang, P., Liu, Y., Feng, N., Xu, J., 2008. Preparation and evaluation of self-microemulsifying drug delivery system of oridonin. Int. J. Pharm. 355, 269–276.