Available online at www.sciencedirect.com
ScienceDirect
European Journal of Pharmaceutics and Biopharmaceutics 64 (2006) 185–192
European Journal of Pharmaceutics and Biopharmaceutics
www.elsevier.com/locate/ejpb

Research paper

Preparation and in vitro/in vivo evaluation of sustained-release metformin hydrochloride pellets

Lian-Dong Hu [a,*], Yang Liu [b], Xing Tang [c], Qian Zhang [c]

[a] College of Pharmacy, Hebei University, Baoding, PR China
[b] College of Pharmacy, Zhengzhou University, Zhengzhou, PR China
[c] College of Pharmacy, Shenyang Pharmaceutical University, Shenyang, PR China

Received 26 January 2005; accepted in revised form 27 April 2006
Available online 10 May 2006

Abstract

In this study, metformin hydrochloride (MH) sustained-release pellets were successfully prepared by centrifugal granulation. Seed cores preparation, drug layering, talc modification and coating of polymeric suspensions were carried out in a centrifugal granulator. Talc modification was performed before coating in order to overcome the high water solubility of metformin. The influence of surface modification by talc, the effects of Eudragit® types and ratios, as well as the correlation between in vitro release and in vivo absorption were investigated in detail. Experimental results indicated that talc modification made a decisive contribution to controlling the drug release by avoiding drug dumping. Three dissolution media: 0.1 M HCl, distilled water and pH 6.8 phosphate buffer were employed to determine the in vitro release behaviors of the above metformin hydrochloride pellets. The relative bioavailability of the sustained-release pellets was studied in 12 healthy volunteers after oral administration in a fast state using a commercially available immediate release tablet (Glucophage) as a reference. Following coating with a blend of Eudragit® L30D-55 and Eudragit® NE30D (1:20), at 7% or 10% coating level, respectively (referred to as F-2, F-3), the pellets acquired perfect sustained-release properties and good relative bioavailability. The C_{max}, T_{max} and relative bioavailability for F-2 and F-3 coated pellets were 1.21 μg/ml, 6 h, 97.6% and 1.65 μg/ml, 8 h, 165%, respectively. Combined use of two Eudragit® polymers with different features as coating materials produced the desired results. Restricted delivery of metformin hydrochloride to the small intestine from differently coated pellets resulted in increased relative bioavailability and a sustained release effect. The adoption of several different pH dissolution media established a better relationship between the in vitro release and in vivo absorption of the sustained-release pellets.

Keywords: Sustained-release pellets; Centrifugal granulation; Metformin hydrochloride; Eudragit® L30D-55; Eudragit® NE30D

1. Introduction

Metformin hydrochloride (MH), an antidiabetic drug, lowers both basal- and postprandial-elevated blood glucose in patients with non-insulin-dependent diabetes mellitus (NIDDM or type 2 diabetes) whose hyperglycemia cannot be satisfactorily managed by diet alone. Some high incidence of concomitant GI symptoms, such as abdominal discomfort, nausea, and diarrhea, may occur during the treatment. Gastrointestinal absorption of metformin is incomplete with an absolute bioavailability of 40–60% (under fasting conditions) [1,2] in combination with rapid elimination and 20–30% of an oral dose is recovered in faeces [3,4]. It decreases as the dose increases, suggesting some form of saturable absorption or permeability/transit time-limited absorption [2] and the negligible hepatic metabolism of metformin happened in humans [4]. Side effects and the need for twice to three times a day administration when larger doses are required can also reduce patient compliance and hinder more successful therapy. Administration of a sustained-release, once-a-day MH dosage form could reduce the dosing frequency and improve patient compliance.

* Corresponding author. Tel./fax: +86 312 5971107.
E-mail address: hldrenew@yahoo.com.cn (L.-D. Hu).

0939-6411/$ - see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.ejpb.2006.04.004

Multiple-unit sustained-release dosage forms, such as pellets, are believed to have many therapeutic advantages in comparison with the single-unit dosage forms. They can distribute in the gastrointestinal tract (GI tract) homogeneously thus maximizing drug absorption and reducing peak plasma fluctuations, minimizing the risk of local GI tract irritation and dose dumping, decreasing dosing frequency and increasing patient compliance, improving the safety and efficacy of the active ingredient.

Centrifugal granulation, an advanced method for manufacturing multiple-unit, sustained-release and drug-loaded pellets for oral administration, has numerous advantages, such as lower manufacturing costs, flexibility in operation and ease of automation compared with other pelletization techniques. Through the use of this system, seed cores were prepared first and subsequently the drug layering and coating process were carried out using the same equipment, resulting in highly spherical and multi-layered pellets with satisfactory sustained-release characteristics [5–7].

In this study, metformin hydrochloride pellets were prepared by centrifugal granulation in a laboratory-scale centrifugal granulator (Model BZJ-360M, Beijing Tianmin High Technology Development Co., China). In conventional sustained-release formulation, the high water solubility of metformin means that large amounts of polymer are required to control its release. In our study, the drug-loading pellets suffered surface modification by talc before coating to control in vitro release. Eudragit® L30D-55 and Eudragit® NE30D were used for coating to produce sustained-release drug-loaded pellets. In vitro dissolution tests in different dissolution media were performed to study the release properties of pellets coated with Eudragit® NE30D alone and a mixture of different ratios of Eudragit® L30D-55 and Eudragit® NE30D. The pharmacokinetics of three sustained-release MH pellet formulations and a commercial immediate-release MH tablet product (Glucophage) was compared in the fast mode. The results showed that different relative bioavailability and sustained release effects could be obtained by using different formulations. The hypothesis was that restricted delivery of metformin hydrochloride to the small intestine from pellets would result in increased relative bioavailability and a sustained plasma concentration profile. A correlation between in vitro release and in vivo absorption was evaluated, which was particularly useful for the development of such sustained-release formulations as drugs absorbed from a specific region of intestine.

2. Materials and methods

Methacrylic acid copolymers (Eudragit® L30D-55, Eudragit® NE30D) were supplied by Röhm GmbH Chemische Fabrik, Darmstadt, Germany. Talc was supplied by Yulin Talc Factory (Guangxi, China). Hydroxypropylmethylcellulose (HPMC) (Methocel E5, manufactured by Dow Chemical Co., Michigan, USA) was supplied by Coloron (Shanghai, China). MH was purchased from Beijing Shuanghe Pharmaceutical Co. Ltd. (Beijing, China). Other excipients used to prepare the pellets were standard pharmaceutical grade. Methanol was HPLC grade and other reagents were analytical reagent grade. The 0 size, hard gelatine capsules used in this paper were supplied by Suzhou Capsule Co. (Brand name: Coni-Snap, Suzhou, China). Double-distilled water was used. Metformin standard substance (99.9%) provided by Chinese National Institute for the Control of Pharmaceutical and Biological Products was used in the in vivo assays. Commercially available immediate release (IR) MH tablet (500 mg, Glucophage, Sino-American Shanghai Squibb Pharmaceuticals Ltd., China) was chosen as the reference.

2.1. Preparation of drug-loaded pellets

A laboratory-scale centrifugal granulator (Model BZJ-360M, Beijing Tianmin High Technology Development Co., China) was used for preparing both MCC seed cores and subsequent drug-layered pellets. In each experiment, 500 g microcrystalline cellulose (MCC) powder was loaded into the processing chamber and moistened by continuously spraying with distilled water. After 15 min of wetting, the MCC powder was added to the above wetted mass via a hopper, keeping all the process parameters constant. The final product was discharged after the addition of powder was complete. The size of the MCC seed cores was approximately 0.4–0.5 mm in diameter, as determined by sieving after drying at 60 °C in an oven for 2 h. The process parameters were as follows: rotational speed of plate: 200 rpm; blower rate: 10×15 L/min; air flow rate: 15 L/min; spray air pressure: 0.5 MPa; rotating rate of powder feeder: 18 rpm.

The mixed powder of drug and MCC (120 mesh) were layered onto the previously prepared MCC seed cores by simultaneously spraying of the binder solution (3% w/w aqueous solution of hydroxypropylmethyl cellulose, HPMC). The process parameters were the same as those described above.

2.2. Preparation of modified, coated pellets

For pellets modification, varying amount of talc (% w/w of the drug-loaded pellets) was layered onto the drug-loaded pellets under the same operating conditions. The resultant modified pellets were dried in an oven at 40 °C for 12 h, resulting in pellet sizes ranging approximately between 0.9 and 1.3 mm in diameter.

Eudragit® L30D-55 and Eudragit® NE30D, two types of acrylic-based aqueous polymeric dispersions, were formulated as follows to prepare sustained-release MH coated pellets:

Formulation 1 (F-1): coated with Eudragit® NE30D, resulting in 10% coat loading.
Formulation 2 (F-2): coated with Eudragit® L30D-55: Eudragit® NE30D (1:20), resulting in 7% coat loading.

Formulation 3 (F-3): coated with Eudragit® L30D-55: Eudragit® NE30D (1:20), resulting in 10% coat loading.

The coating suspensions were prepared in the following steps: (1) micronized talc (50% w/w based on dry polymer weight) was dispersed in purified water and homogenized in a high-speed disperser for 30 min, making sure that the powder was quickly wetted and no lumps formed; (2) the desired amount of Eudragit® L30D-55 and Eudragit® NE30D was mixed or Eudragit® NE30D was used alone; (3) materials prepared under (1) and (2) were mixed, stirred for 30 min and adjusted to the final volume with purified water to produce a coating suspension with polymer solid content of 15% w/w.

Five hundred grams pellets were used for each batch of coating, performed in the same centrifugal granulator by spraying the coating suspension continuously onto the pellet surface, the resultant modified pellets were dried in an oven at 40 °C for 12 h, resulting in pellet sizes ranging approximately between 0.9 and 1.3 mm in diameter. The coated pellets were filled into the 0 size hard gelatine capsules which can accommodate about 250 mg of the drug in the form of pellets for in vitro and in vivo study.

2.3. Assay of the drug content

The samples were assayed by a UV spectroscopic method according to the Chinese Pharmacopoeia 2000 (CHP 2000). From each batch of the coated pellets, a certain amount (30 g) was taken and ground to fine powder. Then about 300 mg powders were accurately weighed, added to a 100-ml volumetric flask containing 70 ml of distilled water. After a 30-min ultrasonic extraction, the solution was diluted with water to 100 ml and then filtered through a 0.45 μm membrane. Precisely 0.2 ml of this solution was transferred to a 100-ml volumetric flask and water added to give a volume of 100 ml. The absorption of sample solution was measured spectrophotometrically at a wavelength of 233 nm. The intra-day accuracy of the method for metformin ranged from 98.5% to 102.1%, while the intra-day precision ranged from 0.7% to 2.4%. The inter-day accuracy ranged from 96.8% to 103.6%, while the inter-day precision ranged from 2.3% to 4.4%. The precision and accuracy of the method were both well consistent with analysis requirement and no absorption of the physical mixture of the excipients existed in 233 nm. The content of MH was calculated using the equation: $A = ECL$, where $E^{1\%}_{1\ \mathrm{cm}} = 798$.

2.4. In vitro dissolution tests

In vitro drug release was determined using a USP 24 Type2 dissolution testing apparatus (paddle method). In this, the 900 ml dissolution medium was kept at 37 ± 0.5 °C and the rotating speed was 50 rpm. 0.1 M HCl solution, purified water and pH 6.8 phosphate buffer were used as different dissolution media.

Two capsules containing the drug pellets equivalent to 500 mg MH were used in all dissolution studies. Three milliliter samples were withdrawn and replaced with an equal volume of the same fresh medium by an auto-sampler at 1, 2, 4, 6, 8, 10 and 12 h. The sample solutions were diluted and spectrophotometry was carried out at a wavelength of 233 nm. Dissolution tests were performed in triplicate.

2.5. Bioavailability studies

The bioavailability study was in accordance with GCP/GLP standards. The study followed the tenets of the Declaration of Helsinki promulgated in 1964, and the protocol was approved by an Ethics Committee on Bioavailability Studies. Twelve healthy adult male volunteers between 20 and 24 years old (mean 22.6 years, SD 0.8 years), and weighing from 58 to 75 kg (mean 63 kg, SD 7.5 kg), participated in the study after providing written informed consent. The volunteers were judged to be healthy and were not receiving any medication during the study period. Volunteers were given information on the drug and nature of the study in advance of the trial. The study was conducted according to a single-dose, randomized, four-way crossover design and the washout period was 1 week between treatments of the study. In this design, the volunteers were randomly selected to receive a single-dose, four 0-sized gelatin capsules containing coated pellets equivalent to 1000 mg MH (250 mg/capsule) or two commercial immediate-release tablets (500 mg/tablet) in the morning and after an overnight fasting (10 h) with 240 ml of water. Blood samples (4 ml) were collected into vacutainers (containing sodium heparin as an anticoagulant) at 0, 0.33, 0.66, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, and 24 h after dosing. An indwelling cannula placed in the forearm was used for drawing blood during the first 12 h. The last sample was taken by direct venipuncture. Following centrifugation, the plasma was then pipetted into polypropylene tubes and frozen immediately, stored at −20 °C until analysis.

The plasma was precipitated with acetonitrile, the supernatant was washed with dichloromethane, after centrifugation, a 20 μL of the remained supernatant was injected into HPLC. A Diamonsil™ C_{18} column was used. The mobile phase consisted of methanol and 0.05 mol L^{-1} $(NH_4)H_2PO_4$ (35:65, V:V, 0.01 mol L^{-1} sodium octanesulfonate was contained). The flow rate was 1 ml min^{-1}, and the UV detector was set at 233 nm.

2.6. Data analysis

The pharmacokinetic parameters were calculated using non-compartmental model. The area under the plasma concentration–time curve from time zero to time t (AUC_{0-t}) was calculated using the trapezoidal method. The peak concentration (C_{max}) and time of peak concentration (T_{max}) were obtained directly from the individual plasma concentration–time profile. The area from time 0 to infinity was calculated by: $AUC_{0-\infty} = AUC_{0-t} + C_t/K_e$,

where C_t is the plasma MH concentration observed at time t, K_e is the apparent MH elimination rate constant obtained from the terminal slope of the individual plasma concentration–time curves after logarithmic transformation of the plasma concentration values and application of linear regression. The relative bioavailability (F) was calculated by: $F = AUC_T/AUC_R$.

3. Results and discussion

3.1. Influence of talc modification on release

Some comparative studies were conducted to investigate whether there was a significant difference in talc modification. The drug-loaded pellets were modified using different amounts of talc (percentage based on pellet weight) during granulation and coated with Eudragit® NE30D alone to give a constant 10% coat loading. The modification influence of varying amounts of talc on drug release is shown in Fig. 1. A series of different amounts of Eudragit® NE30D were applied to the modified drug-loaded pellets, and the cumulative release profiles are shown in Fig. 2.

In the case of non-talc modification, as the coat loading increased, the drug release decreased slightly (data was not shown). Even at a higher coating amount, good release results were not obtained. Fig. 2 indicates that talc modification played an important role in the control of drug release. At 10% talc modification, the percentage of drug released decreased rapidly when the coating amount increased. This result may be due to the higher solubility of metformin hydrochloride in aqueous solution. When conducting the coating operation with an aqueous coating solution, the drug on the pellet surface would dissolve fast and diffuse into the film. Therefore a dumping effect would occur in dissolution test because the drug in the film acted as pore-forming agent. Because of the strong hydrophobic effect, the talc powder covering on the surface of the pellets prevented the drug from diffusing into the film, and thus

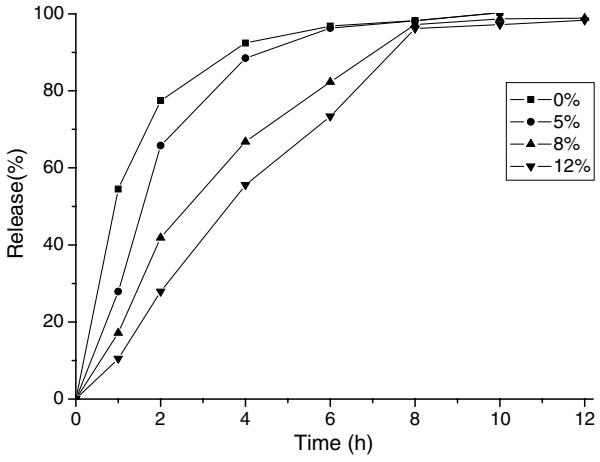

Fig. 1. Effects of talc amounts on the drug dissolution in distilled water.

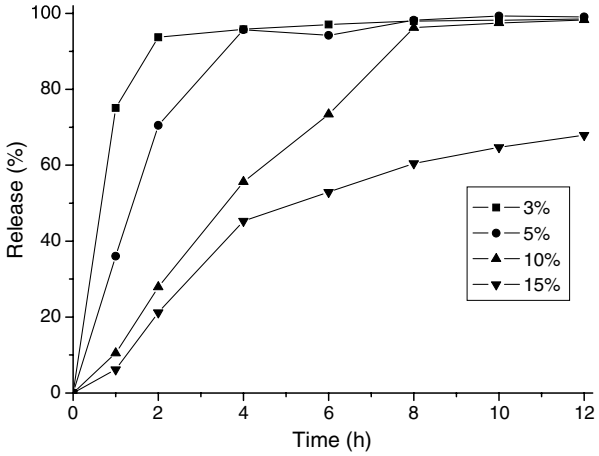

Fig. 2. Effects of coating amounts of Eudragit® NE30D on the release of talc-modified pellets in distilled water.

significantly modified the release velocity. From the experimental results, it was concluded that 10% talc modification produced a satisfactory control effect. So in the following coating operations, drug-loaded pellets with 10% talc modification were used.

3.2. Influence of coating formulations on release

The profiles of MH released from sustained-release pellets in three dissolution media and under different coating formulations are shown in Fig. 3.

When comparisons were made among the three formulations, it was found that there were some differences in the release of MH. In 0.1 M HCl, the release rate of MH from F-3 was remarkably slower than those from F-1 and F-2. The accumulated release percentages of MH from F-1, F-2 and F-3 were 67.3%, 66.0% and 26.6% by 4 h, 83.4%, 96.1% and 55.2% after 6 h, respectively. In purified water, the release rates of MH from F-1 and F-3 showed no significant difference, while the rate was faster from F-2. The accumulated release percentages of MH from F-1, F-2 and F-3 were 25.4%, 45.8% and 27.6% after 2 h, 52.4%, 90.4% and 55.1% after 4 h, respectively. In pH 6.8 phosphate buffer, the release rates of MH from F-2 and F-3 were markedly faster than that from F-1, but there was no significant difference between F-2 and F-3. The accumulated release percentages of MH from F-1, F-2 and F-3 were 22.3%, 55.4% and 61.4% after 2 h, 48.3%, 92.4% and 98.3% after 4 h respectively.

The release results also demonstrated that the release rate of MH from F-1, i.e. the pellets coated with 10% Eudragit® NE30D, is not significantly affected by the pH of the dissolution media used. Whereas the pH of dissolution media had a significant influence on the release of MH from F-2 and F-3, i.e. pellets coated separately with a 7% and 10% loading of the mixture of Eudragit® L30D-55 and Eudragit® NE30D (1:20). The drug release rate increased as the pH values increased, as was particularly high for F-3.

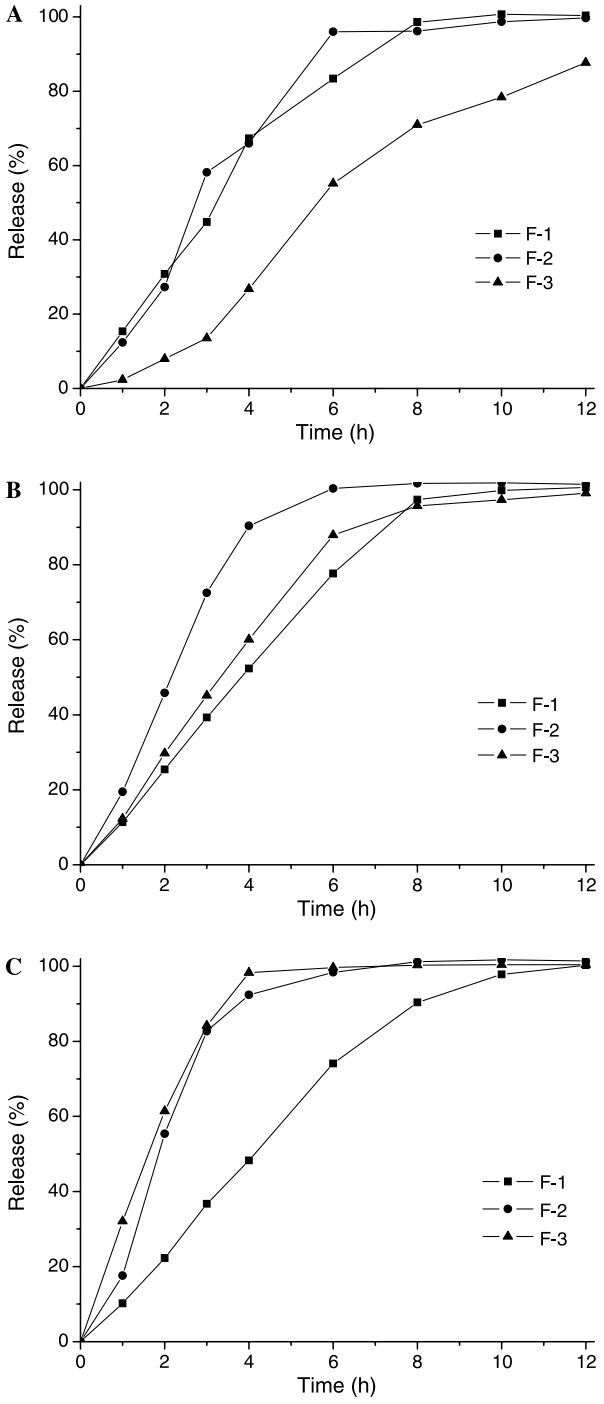

Fig. 3. Comparison of drug dissolution profiles for pellets coated with three formulation in different media: (A) 0.1 M HCl, (B) distilled water, (C) pH 6.8 PBS.

Comparing the release results between F-1 and F-3 which had the same coat loading, it was found that Eudragit® L30D-55 had a marked effect on drug release. Comparing the release results between F-2 and F-3, which had the same coating material, it was found that the coat loading of the mixture of Eudragit® L30D-55 and Eudragit® NE30D (1:20) had a significant influence on the drug release rate from these pellets.

This result was due to the different nature of these two coating materials. Eudragit® NE30D, a polymer composed of methyl methacrylate and ethyl acrylate monomers in a ratio of 2:1, has a low glass transition temperature (T_g) at −8 °C and a minimum film formation temperature at 5 °C. Polymeric films prepared from Eudragit® NE30D are soft and flexible [8]. Because the permeability of the film is pH-independent, Eudragit® NE30D has been used widely in sustained-release film coatings [9,10], granulations [11,12] and as a component of transdermal films [13]. Eudragit® L30D-55, a methacrylic acid copolymer, has been widely used in enteric coating formulations. Films prepared from it are insoluble in gastric fluid but are readily soluble in aqueous media with a pH above 5.5. The blends of acrylic polymers have been successfully used to modify various film properties [14–18]. In our study, Eudragit® L30D-55 was added to a Eudragit® NE30D dispersion. The dissolution of Eudragit® L30D-55 at intestinal pH values would result in a higher drug release rate in the intestine.

3.3. Bioavailability

The in vivo pharmacokinetic behavior of pellets coated with different formulations was investigated. Fig. 4 shows mean plasma concentration–time curves after administration of three types of sustained-release MH pellets and IR MH tablets. Their bioavailability parameters of them are listed in Table 1. The differences in bioavailability were very significant.

The plasma level of IR MH tablets rose quickly and the maximum concentration (2.38 μg/ml) was reached 3 h after administration. There was a marked fall in plasma concentration between 3 and 12 h. For pellets coated with F-1, the maximum concentration (0.92 μg/ml) was reached 4 h after administration, but it was lower than that of the IR MH tablets and also lower than those of pellets coated with F-2 and F-3, showing that incomplete absorption occurred

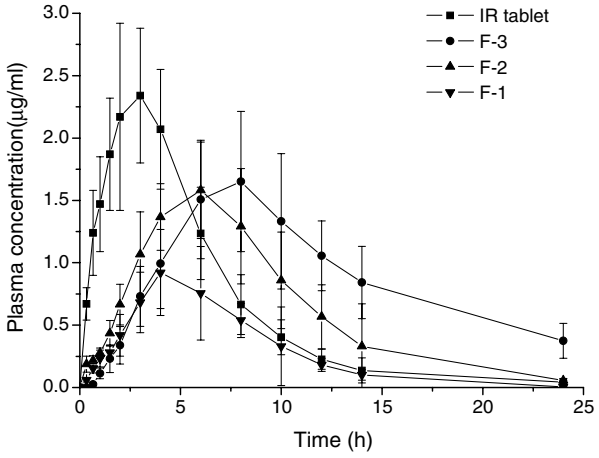

Fig. 4. Mean plasma metoformin concentration–time profiles of three sustained MH pellets and IR MH tablets.

Table 1
The pharmacokinetic parameters of different MH formulations ($n = 12$)

Parameters	IRGlucophage (Reference)	F-1	F-2	F-3
C_{max} (μg/ml)	2.385 ± 0.406	0.922 ± 0.129	1.621 ± 0.337	1.651 ± 0.154
$AUC_{0\to24}$ (μg h/ml)	15.35 ± 2.83	6.91 ± 0.34	14.94 ± 2.52	21.29 ± 2.32
$AUC_{0\to\infty}$ (μg h/ml)	15.62 ± 3.00	6.93 ± 0.43	15.22 ± 2.59	25.77 ± 2.08
$t_{1/2}$ (h)	2.89 ± 0.90	2.13 ± 0.13	3.47 ± 0.42	8.284 ± 0.064
MRT (h)	5.21 ± 0.54	6.68 ± 0.45	8.26 ± 0.51	14.85 ± 0.21
T_{max} (h)	2.72 ± 0.46	4.17 ± 0.92	5.56 ± 0.86	7.33 ± 0.97
K_e (h^{-1})	0.267 ± 0.10	0.327 ± 0.02	0.203 ± 0.029	0.084 ± 0.003
F (%)	–	46.4 ± 11	97.6 ± 14	165 ± 10

Note: IR: IR MH tablets. F-1, F-2, F-3: pellets coated with F-1,F-2 and F-3, respectively.

in vivo. For pellets coated with F-2, maximum concentration (1.62 μg/ml) was reached 6 h after administration, while the fall in drug concentration occurred at a lower rate than that of IR MH tablets. For pellets coated with F-3, it was very interesting to investigate the changes in plasma level because maximum concentration (1.65 μg/ml) was reached 8 h after administration and the drug concentration fell slowly even at 24 h after administration, when the drug concentration was 0.37 μg/ml. Compared with IR MH tablets, the relative bioavailability judged from the $AUC_{0-\infty}$ was found to be 46.4%, 97.6% and 165%, respectively, for pellets coated with F-1, F-2 and F-3.

3.4. In vitro–in vivo evaluation

In vitro dissolution tests were performed to study the release behaviors of different formulations in different dissolution media and to establish a correlation between in vitro release and in vivo absorption for the modified-release pellets.

Many studies have reported that metformin is incompletely absorbed from the GI tract [19]. Tucker et al. showed that absorption of the drug from the GI tract occurred mainly within 6 h after taking an immediate-release tablet [2]. Many researchers had reported that the bioavailability of sustained-release preparations of metformin is reduced compared with that from the immediate-release tablets. This indicated that absorption of metformin hydrochloride is confined to the small intestine [20]. Marathe et al. showed that metformin was well absorbed throughout the intestine but rapidly decreased in the colon [21]. We have studied the intestinal absorption kinetics of metformin hydrochloride in rats in situ. Our results showed that the absorption rate constants (K_a) of metformin hydrochloride were $0.0657 \pm 0.0050\ h^{-1}$, $0.0381 \pm 0.0112\ h^{-1}$ and $0.0157 \pm 0.0057\ h^{-1}$, respectively, in the duodenum, jejunum and ileum, while no absorption was found in colon. From the literature and our research, it appears that metformin hydrochloride is a drug that exhibits site specificity absorption in the intestine, because the absorption rate decreases rapidly from the duodenum to the colon. The fact that the bioavailability of sustained-release preparations of metformin was reduced compared with that of the immediate-release tablets might be because of this. It was thought that two aspects need to be carefully considered when evaluating the in vivo behavior of the drug, i.e. the factors influencing its in vivo absorption and the formulation factors influencing its in vitro release. The in vitro release behavior and the transfer mechanism of pellets are important when developing a sustained-release dosage form for such a kind of drug. So, we concluded that the absorption mechanism of the sustained-release pellets combined the above two aspects.

In the current study, the bioavailabilities of sustained-release MH pellets coated with three formulations and a commercial immediate-release MH tablet product were compared in the fast mode. A comparison of the results showed that different degrees of sustained effect were obtained. After oral administration in the fast state, pellets are widely distributed in the stomach and then passed into the duodenum. Although gastrointestinal (GI) transit time of pellets was difficult to estimate, we presumed the in vivo transit behavior of three pellets formulations for the same volunteer in three administration period was similar, such as the gastric emptying time, the pellets distribution amount at the same site of GI tract at same time point after every administration period and the absorption ability of GI tract, the intrasubject difference could be excluded. Therefore, it could be predicted that the different drug-release velocities from pellets of different coating formulations result in the differences in bioavailability and sustained-release effects. So in order to investigate the relationship between in vitro release and in vivo absorption for the modified-release pellets, several different pH dissolution media were employed to study drug release.

It has been reported in many papers that the pH values in the stomach ranged from 1.2 to 5.0, whereas the pH values in the duodenum, jejunum and ileum are 6.63 ± 0.53, 7.41 ± 0.36 and 7.49 ± 0.46, respectively [22–24]. For pellets, the average gastric emptying time was 2 h, and the mean cecum arrival time is about 5–7 h after oral administration [25–27]. For the comparison of in vivo behaviors of immediate-release tablet and sustained-release pellets, we must pay much attention to the absorption site specificity of MH and the active saturable absorption process in the region of the GI tract [28]. Because of the absorption site specificity of metformin hydrochloride in the intestine, three dissolution media, 0.1 M HCl, distilled water and

phosphate buffer (pH 6.8), were used to mimic the in vitro release behaviors of MH from pellets in order to determine the in vivo absorption mechanism.

Metformin is highly soluble, which usually results in rapid dissolution of drug from immediate-release tablet in the stomach, but metformin is poorly absorbed from the stomach (about 10% over a 4-h period) [29], when a great quantity of drug transit through the stomach to the intestine, because the drug amount is much at the same time, the absorption is saturable and the drug cannot be absorbed completely in the upper site of intestine with some drug excreted in faeces.

For pellets coated with F-1, because Eudragit® NE30D aqueous dispersion produced an insoluble film, which swelled throughout the entire gastrointestinal tract, the in vitro release was very similar under the three different pH conditions. It is thought that in vivo drug release from this kind of pellet takes place at a constant velocity, the pellets release some drug in the stomach with a little absorption, when transited to intestine, the released drug cannot be absorbed completely in the jejunum and ileum due to the active saturable absorption, furthermore, the surplus part of drug in the pellets cannot release rapidly in the intestine, after 5–7 h when the pellets are transferred to the colon, much of the still remained unreleased or was transited to colon without absorption and thus the bioavailability is relatively low.

For pellets coated with F-2 and F-3, Eudragit® L30D-55 was added to Eudragit® NE30D dispersion to achieve a higher drug release rate in the intestine because of the higher porosity and permeability of these films after dissolution of the enteric polymer at intestinal pH values. From the in vitro dissolution tests, the velocities of F-2 and F-3 coated pellets were faster in pH 6.8 PBS than at other pH values and there was no difference between them. So, it appears that when the pellets are transferred from the stomach into intestine, because of the permeability increase of the coated film, drug release increases greatly for pellets coated with F-2 and F-3. A higher C_{max} and bioavailability could be achieved compared with F-1 coated pellets.

The F-2 released fast both in HCl and pH 6.8 PBS, which means a rapid release of drug in the stomach and intestine, at a certain time much of drug was transit to duodenum, the released drug may exceed the absorption ability of the intestine, like the immediate release tablet resulting in some drug transit to colon without absorption.

In 0.1 M HCl and purified water, the velocity of F-3 coated pellets was significantly lower than that of F-2 coated pellets. When the pellets remained in the stomach, the in vivo drug release from F-3 coated pellets was lower than that from F-2. When the pellets transit from stomach to duodenum, and jejunum, the drug released from stomach can be completely absorbed and the unreleased part of F-3 coated pellets can release rapidly at the absorption site of intestine. Due to in vivo transition of the pellets being a continuous process, with a gradual drug release not exceeding the absorption ability of the intestine, it resulted in a prolonged input of the drug to the main absorption sites located in the small intestine. Enhanced bioavailability was also achieved when compared with F-2 coated pellets or IR MH tablet. The relative bioavailability of F-3 coated pellets was found to be as high as 165% in comparison with IR MH tablet.

Combining the in vitro dissolution tests with the in vivo absorption mechanism, restricted delivery of metformin hydrochloride to the small intestine from pellets was hypothesized to result in increased bioavailability and a sustained plasma concentration profile.

4. Conclusion

Because of high solubility of metformin hydrochloride, the drug-loaded pellets were subjected to surface modification by talc before coating. The results suggest that talc modification effectively controls drug release and avoids drug dumping. After using Eudragit® NE30D alone and a blend of Eudragit® L30D-55/Eudragit® NE30D (1:20) for coating, three kinds of sustained-release pellets with different formulations were obtained. The in vivo bioavailability showed varying sustained-release characteristics for the coated pellets when compared with IR MH tablets. When coated with a blend of Eudragit® L30D-55 and Eudragit® NE30D (1:20) to a loading weight of 7% or 10%, pellets exhibited excellent sustained-release effects and high relative bioavailability. Because of the absorption site specificity of metformin hydrochloride in the intestine, three dissolution media, 0.1 M HCl, distilled water and phosphate buffer (pH 6.8), were used to mimic the in vitro release behaviors of MH from pellets in order to determine the in vivo absorption mechanism for the sustained-release pellets and restricted delivery of metformin hydrochloride to the small intestine from pellets was hypothesized to result in the increased bioavailability and sustained plasma concentration profile.

References

[1] L.S. Hermann, A. Melander, Biguanides: basic aspects and clinical use, in: K.G.M.M. Alberti, R.A. DeFronzo, H. Keen, et al. (Eds.), International Textbook of Diabetes Mellitus, Wiley, New York, 1992, pp. 773–795.
[2] G.T. Tucker, C. Casey, P.J. Phillips, Metformin kinetics in healthy subjects and in patients with diabetes mellitus, Br. J. Clin. Pharmacol. 12 (2) (1981) 235–246.
[3] C.J. Dunn, D.H. Peters, Metformin: a review of its pharmacological properties and therapeutic use in noninsulin-dependent diabetes mellitus, Drugs 49 (1995) 721–749.
[4] A.J. Scheen, Clinical pharmacokinetics of metformin, Clin. Pharmacokinet. 30 (5) (1996) 359–371.
[5] B. Gajdos, Rotary granulators – evaluation of process technology for pellet production using factorial design, Drugs Made Ger. 27 (1984) 30–36.
[6] I. Ghebre-Sellassie, R. Gordon, M.B. Fawzi, R.U. Nesbitt, Evaluation of a high-speed pelletization process and equipment, Drug Dev. Ind. Pharm. 11 (1985) 1523–1541.
[7] M. Niskanen, Powder layering and coating in a centrifugal granulator: Effect of binder-solution concentration, powder particle size and

coating amount on pellets properties, Doctor's Thesis, University of Helsinki, 2001, pp. 9–11.
[8] K. Lehmann, Chemistry and application properties of polymethacrylate coating systems, in: J.W. McGinity (Ed.), Aqueous Polymeric Coatings for Pharmaceutical Dosage Forms, second ed., Marcel Dekker, New York, Basel, HongKong, 1997, pp. 101–176.
[9] R. Bodmeier, O. Paeratakul, Evaluation of drugcontaining polymer films prepared from aqueous latexes, Pharm. Res. 6 (1989) 725–730.
[10] S. Watano, K. Ando, K. Miyanami, Y. Ii, S. Sasatani, Preparation of core particles for aqueous film coating using agitation fluidized bed, Chem. Pharm. Bull. 45 (1997) 2039–2042.
[11] A. Trojak, R. Pisek, O. Planinsek, S. Srcic, Granulation of model drugs with different binders in the fluid bed granulator and prediction of optimum binder with spreading coefficient values, Farmacevtski. Vestnik. 50 (1999) 365–366.
[12] T. Niskanen, J. Yliruusi, M. Niskanen, O. Kontro, Granulation of potassium chloride in instrumented fluidized bed granulator. part 1. Effect of flow rate, Acta Pharm. Fenn. 99 (1990) 13–22.
[13] C. Valenta, R. Biebel, In vitro release study of transdermal delivery systems of progesterone, Drug Dev. Ind. Pharm. 24 (1998) 187–191.
[14] A.A. Deshpande, N.H. Shah, C.T. Rhodes, W. Malick, Evaluation of film used in development of a novel controlled-release system for gastric retention, Int. J. Pharm. 159 (1997) 255–258.
[15] K. Lehmann, Preparation and use of latex dispersions from redispersible anionic acrylic resins, Acta Pharm. Technol. 31 (1985) 96–106.
[16] K. Lehmann, D. Dreher, Mixtures of aqueous polymethacrylate dispersions for drug coating, Drugs Made In Germany 31 (1988) 101–102.
[17] K. Amighi, J. Timmermans, J. Puigdevall, E. Baltes, A.J. Moës, Peroral sustained-release film-coated pellets as a means to overcome physicochemical and biological drug-related problems. I. In vitro development and evaluation, Drug Dev. Ind. Pharm. 24 (1998) 509–515.
[18] K. Amighi, A.J. Moës, Evaluation of thermal and film forming properties of acrylic aqueous polymer blends: application to the formulation of sustained-release film-coated theophylline pellets, Drug Dev. Ind. Pharm. 21 (1995) 2355–2369.
[19] D.H. Dunn, Metformin: a review of its pharmacological properties and therapeutic use in non-insulin-dependent diabetes mellitus, Drugs 49 (1995) 721–749.
[20] P.J. Pentikainen, Bioavailability of metformin: comparison of solution, rapidly dissolving tablet, and three sustained release products, Int. J. Clin. Pharm. Ther. Toxicol. 24 (1986) 213–220.
[21] P.H. Marathe, Y. Wen, J. Norton, D.S. Greene, T.F. Barbhaiya, I.R. Wilding, Effect of altered gastric emptying and gastrointestinal motility on metformin absorption, Br. J. Clin. Pharmacol. 50 (2000) 325–332.
[22] D.F. Evans, G. Pye, R. Bramley, A.G. Clark, T.J. Dyson, J.D. Hardcastle, Measurement of gastrointestinal pH profiles in normal ambulant human subjects, Gut 29 (1988) 1035–1041.
[23] P.J. Watts, L. IIIum, Colonic Drug Delivery, Drug Dev. Ind. Pharm. 23 (1997) 893–913.
[24] J.E. Devereux, J.M. Newton, M.B. Short, The influence of density on the gastrointestinal transit of pellets, J. Pharm. Pharmacol. 42 (1990) 500–501.
[25] K.H. Yuen, A.A. Deshmukh, J.M. Newton, M. Short, R. Melchor, Gastrointestinal transit and absorption of theophylline from a multiparticulate controlled release formulation, Int. J. Pharm. 97 (1993) 61–77.
[26] G.M. Clarke, J.M. Newton, M.B. Short, Comparative gastrointestinal transit of pellet systems of varying density, Int. J. Pharm. 114 (1995) 1–11.
[27] F. Podczeck, J.M. Newton, K.H. Yuen, The description of the gastrointestinal transit of pellets assessed by gamma scintigraphy using statistical moments, Pharm. Res. 12 (1995) 376–379.
[28] N.C. Sambol, J. Chiang, M. O'Conner, Pharmacokinetics and pharmacodynamics of metformin in healthy subjects and patients with noninsulin dependent diabetes mellitus, J. Clin. Pharmacol. 36 (1996) 1012–1021.
[29] N. Vidon, S. Chaussade, M. Noel, C. Franchisseur, B. Huchet, J.J. Bernier, Metformin in the digestive tract, Diabetes Res. Clin. Pract. 4 (1988) 223–229.