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RESEARCH PAPER

Development of a Bionic System for the Simultaneous Prediction of the Release/Absorption Characteristics of Enteric-Coated Formulations

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ABSTRACT

Purpose To develop a new bionic system from an existing drug dissolution/absorption simulating system (DDASS) to simultaneously predict the release and absorption of enteric-coated formulations.

Methods In accordance with the pH-dependent characteristics of enteric-coated formulations, the modified DDASS was designed to effectively imitate the pH change process of the formulations' transfer from stomach to intestine *in vivo*. Omeprazole enteric-coated tablets were chosen as the model drug to verify the rationality and feasibility of the modified DDASS. The correlations between USP I system release and beagle dog absorption, as well as between modified DDASS elution/permeation and beagle dog absorption, were investigated by linear and nonlinear regression analyses, respectively.

Results *In vitro-in vivo* correlation between the modified DDASS elution/permeation method and beagle dog absorption was higher than between the USP I system release and beagle dog absorption in both analytical methods. The ratio of first-order permeation rate constant to first-order release rate constant was consistent with that from modified DDASS.

Conclusions The modified DDASS provided more information than the USP I system did in the evaluation of enteric-coated formulations. The proposed bionic system model could serve as a new method for improving drug effectiveness.

KEY WORDS drug dissolution/absorption simulating system (DDASS) · *in vitro-in vivo* correlations (IVIVC) · linear relationship · nonlinear relationship · omeprazole enteric-coated tablet

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ABBREVIATIONS

DC	donor compartment
DDASS	drug dissolution/absorption simulating system
DDV	drug-dissolving vessel
GI	gastrointestinal
IVIVC	*in vitro-in vivo* correlations
PAE	pH adjustment equipment
PAV	pH adjustment vessel
RC	receiver compartment

INTRODUCTION

An enteric-coated formulation is emptied from the stomach as one unit due to the enteric coating, and releases in the small bowel. Some drugs that degrade and cause irritation to the stomach or have pharmacodynamic effects in the intestine are usually produced into enteric-coated formulations to help improve the efficacy of clinical applications. The key role is enteric coatings, such as acidic polymers described by Abizer IH *et al.* These acidic polymers are intended to have low permeabilities in their unionized state in the low pH environments expected in the stomach. When they reach more neutral to alkaline environments characteristic of the intestinal milieu, enteric polymers ionize and erode to release drug from the underlying core (1). Because of the pH-dependent characteristics of enteric-coated formulations, the prediction of their release and absorption is very important for the design of formulation. The *in vivo* bioavailability test is an ideal but time-consuming evaluation method for enteric-coated formulations. It is also expensive and cannot be used as a routine measurement for each batch of product. *In vitro* dissolution tests described in the pharmacopoeia of

many countries are an important means for prescription screening and quality control but most cannot simulate the consecutive pH changes in the gastrointestinal (GI) tract or simultaneously assess the dissolution and permeation of the drug.

Several *in vitro* methods for the simultaneous measurement of dissolution and permeation have been established in the past few years, and auspicious results have been obtained. Sartorius absorption apparatus consisting of two containers separated with an artificial membrane was introduced to evaluate diffusion and absorption of drugs *in vitro* by Stricker in 1971 (2). Ginski and Polli (3) reported a continuous dissolution/Caco-2 system for predicting the dissolution and absorption of solid formulations prior to human studies. However, the method did not model the drastic pH changes in the GI tract. Kobayashi *et al.* (4) developed an *in vitro* system for the prediction of drug absorption that took into account the dissolution of solid drugs and pH changes in the GI tract. Moreover, He *et al.* (5–8) established six models with different gastric acidities and applied them in rat intestine *in vitro* system to evaluate the absorption of powders or grinded tablets. Kataoka *et al.* (9,10) invented a dissolution/permeation system in which an amount of drug corresponding to the clinical dose was applied. Motz *et al.* (11,12) used a flow-through cell to concomitantly determine dissolution and permeation. While certainly useful, these methods only allow the analysis of non-formulated compounds, not complete oral dosage forms. Li *et al.* (13) reported a drug dissolution/absorption simulating system (DDASS), which was improved with a basket installed in the drug-dissolving vessel and a two-stage filtering system. Unfortunately, the system featured several drawbacks such as the fact that enteric-coated formulations cannot disintegrate and release in the system, when evaluating the release characteristics of enteric-coated formulations. Thus, the current work aims to develop a new bionic system from this DDASS to predict the simultaneous release and absorption of enteric-coated formulations.

The proposed modified DDASS was designed to effectively imitate the *in vivo* process of an enteric-coated formulation's transfer from stomach to intestine. Omeprazole enteric-coated tablets were chosen as the model dosage form. Omeprazole is a known and well-studied substituted benzimidazole used in the treatment of gastric diseases due to its capacity to inhibit the (H^+/K^+) ATPase system in the gastric parietal cells (14). Kinetic characteristics were taken as a main line, including release kinetics, permeation kinetics, and *in vivo* pharmacokinetics. The modified DDASS is expected to be used in the development of new preparations by screening prescriptions, and in the improvement of druggability.

MATERIALS AND METHODS

Chemicals and Drug Dosage Forms

Omeprazole (98% pure) was obtained from the National Institute for the Control of Pharmaceutical and Biological Products (Beijing, China). Omeprazole enteric-coated tablets (20 mg, Mups®) were provided by AstraZeneca Co., Ltd. (Wuxi, Jiangsu, China). HPLC-grade acetonitrile and HPLC-grade methanol were provided by Concord Co., Ltd. (Tianjin, China). Ultra-pure water was prepared using a Milli-Q Synthesis system (Millipore, Billerica, MA); MES hydrate, HEPES, and D-(+)-glucose were purchased from Sigma Co., Ltd. (USA). All other chemicals used in the present study were of analytical grade.

Animals

Male Wistar rats, weighing (280 to 320) g, were provided by the Institute of Radiation Medicine, Chinese Academy of Medical Sciences (Tianjin, China). The rats were fasted for 12 h before the experiments but had free access to water. During the experiments, the rats were anesthetised with ether. The jejunum of each rat [(2.0 to 2.5) cm] was removed. Male purebred beagle dogs, weighing (8.0 ± 0.5) kg, were obtained from the Laboratory Animal Center, Chinese PLA Academy of Military Medical Science (Beijing, China). The dogs were housed one per cage for at least 10 d before the experiments. Kennel temperature was maintained at (25 ± 2)°C. Expanded feed for dogs was purchased from KeAoXieLi Food Co., Ltd. (Beijing, China). The Academy of Military Medical Science Institutional Animal Care and Use Committee (Certificate No. SCXK-2007-004) approved the present animal study.

Modified DDASS

The existing DDASS system was modified (Fig. 1) to evaluate the drug absorption of enteric-coated formulation as previously described (13). The previous two-vessel model [drug-dissolving vessel (DDV) + pH adjustment vessel (PAV)] was improved into a single-vessel model (DDV) with the pH change of the drug-dissolving solution. As shown in Fig. 2, a time course was made for the pH change profile to prove the rationality of the modified DDASS. The pH value in DDV and the donor compartment (DC) of side-by-side diffusion chamber improved from the one established by Hidalgo *et al.* (15) were determined every 10 min until 5 h using a pH meter. Drug-dissolving solution A (pH=2.0, simulated gastric fluid) was replaced with drug-dissolving solution B (pH=6.8, simulated intestinal fluid) in the DDV at 30 min. The pH value in the DDV took approximately 10 min to increase from 2.0 to 3.5, which is the pH value at

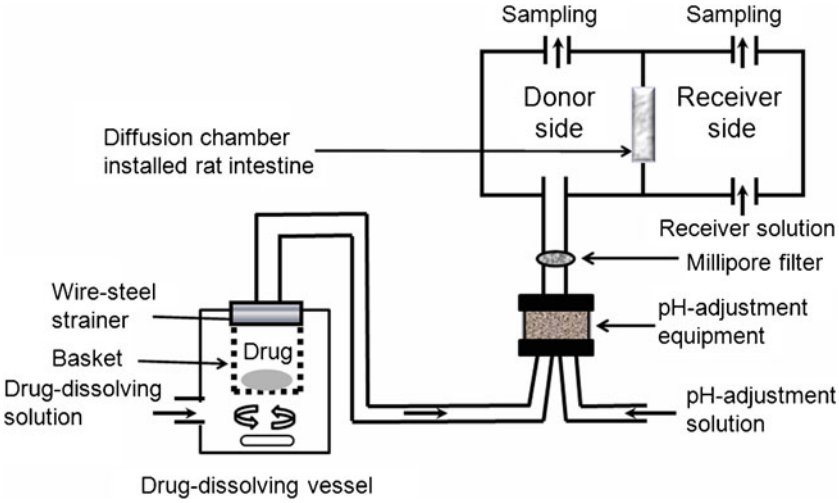

Fig. 1 Scheme of the modified drug dissolution/absorption simulating system (DDASS).

the end of the stomach or the beginning of the duodenum (16), after changing the drug dissolving solution. Thus, the complete continuous and dynamic process of pH change in the DDV from pH 2.0 to 3.5 took approximately 40 min, the fasting gastric emptying time in the human body described according to a research (17). The pH value was gradually increased to 6.8, just as in the intestinal environment. With the increased pH value, the release of the omeprazole enteric-coated tablets was accelerated and the *in vivo* process of the effective release the enteric-coated formulation's transfer from stomach to intestine could be imitated. The PAV was replaced with pH adjustment equipment (PAE). The PAE could reduce the dead volume in the previous PAV and the lag time between the drug concentration in the DC and that in the DDV. This method is more accurate and convenient for trace determination. Based on Fig. 2, the pH value in the DC maintained a steady state. This phenomenon confirmed that the PAE could fully mix the drug-dissolving solution and pH adjustment solution (pH=6.8) and avoid sudden changes in pH value, thereby maintaining the optimum condition

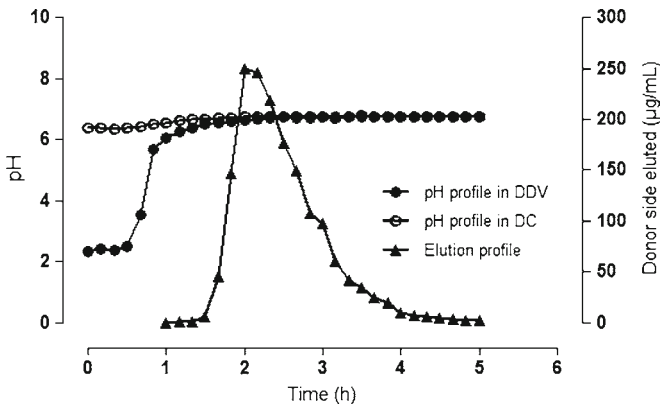

Fig. 2 Time course of pH change and elution profile in modified DDASS.

of rat intestines for survival and ensuring that the drugs continuously channel into the acceptor solution (pH= 7.4), which imitates plasma.

In this system, an enteric-coated formulation was added into a DDV. The dissolved drug was transferred to the PAE and mixed with pH adjustment solution. The drug solution was transferred to the DC and constantly permeated the acceptor solution in the receiver compartment (RC) across the rat intestine. A wire-steel strainer was added on the top of the DDV as a first filtering system to prevent the escape of undissolved particles from the DDV. A Millipore filter was installed between the PAE and side-by-side diffusion chamber as a second filtering system to purify the drug solution further. The flow rate of each solution (0.5 mL/min) was controlled using a peristaltic pump. The volume of DDV was fixed (10 mL). These were justified in a previous study by the co-authors (5). The composition of the flowing solution was improved from that in the two-vessel model (13). In drug-dissolving solution A, NaCl was changed from 61.4 mM to 135.0 mM, HCl was changed from 13.6 mM to 5.0 mM, and 10.0 mM MES was added. Drug-dissolving solution B was added as a new composition; this solution included 135.0 mM NaCl, 50.0 mM D-Glucose, 2.52 M $CaCl_2$, 0.81 mM $MgSO_4$, and 10.0 mM MES. In the pH adjustment solution, NaCl was changed from 199.2 mM to 135.0 mM, and MES was changed from 40.0 mM to 70.0 mM. The pH value of drug-dissolving solution B and the pH adjustment solution were adjusted to 6.8 with Tris. Other components and the acceptor solution were unchanged.

Elution and Permeation Test in the Modified DDASS

For omeprazole enteric-coated tablets, the eluted solution in the DC was collected every 10 min over a period of 5 h. In the

study for the permeation properties of omeprazole enteric-coated tablets, a jejunum removed from a rat was mounted between the DC and RC. Under bubbling conditions with O_2-CO_2 (95:5) mixture gas, the solution eluted into the DC and RC were collected every 10 min during a period of 5 h. Each release/permeation profile was obtained from three trials.

Release Test by USP I System (Basket)

The release profile for the omeprazole enteric-coated tablets was evaluated with the USP I system (basket) at 100 rpm. The release medium (900 mL), which included 0.1 M hydrochloric acid (pH 1.8) and 0.05 M pH 6.8 phosphate buffer, was deaerated and maintained at 37.0°C±0.5°C. At the beginning, 0.1 M hydrochloric acid was given. After 2 h, the release medium was changed into 0.05 M pH 6.8 phosphate buffer. At (0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, and 4.5) h, 2 mL samples were withdrawn and filtered through 0.45 μm Millipore filter®. An equivalent volume of fresh medium was replaced after each sampling. Each release profile was obtained from six replications.

Pharmacokinetic Studies of Omeprazole Enteric-Coated Tablets in Beagle Dogs

According to a single-dose oral administration, omeprazole enteric-coated tablets were assessed in five beagle dogs. The dogs were fasted for at least 12 h before the experiment, with free access to water and food 4 h after drug administration. Blood samples (1 mL) were collected from the cephalic vein of the front leg at (0, 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 5, 6, 8, 10, and 12) h. These samples were immediately centrifuged at 3500 r/min for 10 min and stored at –20°C until analysis.

In a glass centrifuge tube, 25 μL of 4.5 μg/mL carbamazepine (the internal standard) was added to an aliquot of plasma sample (500 μL). After vortex mixing for 30 s, 4 mL dichloromethane was added and vortexed for 5 min. The organic layer was separated by centrifugation at 3500 r/min for 10 min and evaporated to dryness under a gentle stream of nitrogen at 40°C. The residue was dissolved with 100 μL methanol and centrifuged before injection.

In Vitro–In Vivo Correlation (IVIVC)

Linear Regression

A key goal in the pharmaceutical development of dosage forms was a good understanding of the *in vitro* and *in vivo* performance of the dosage forms. The FDA defines the IVIVC as a predictive mathematical model describing the relationship between an *in vitro* property of a dosage form and a relevant *in vivo* response (18).

The percentage fraction absorbed (F_a) of omeprazole enteric-coated tablets was calculated by the Wagner–Nelson Eq. 1 for a single compartment model (19).

$$F_a = \frac{C_t + k_{10} \times AUC_{0-t}}{k_{10} \times AUC_{0-\inf}} \times 100\% \quad (1)$$

Where C_t is the concentration at time point t, k_{10} is the elimination rate of the dosage form, $AUC_{0\text{-}t}$ is the area under the curve from zero to time t, and $AUC_{0\text{-inf}}$ is area under the curve from zero to infinity. A regression equation was fitted to the percentage of release (F_d) and F_a at corresponding time points by the least square method. The regression coefficient was the index of correlation between *in vitro* release and *in vivo* absorption.

Nonlinear Regression

IVIVC degrees range from "low" to "high" in a gradual, continuous fashion, rather consisting of only the currently perceived options of "yes, a correlation exists" and "no, a correlation does not exist (20)." Thus, the nonlinear relationship was thought to be more reasonable and objective. The degree of correlation was found to be dependent upon two factors: the relative rates of release and permeation, and the fraction of dose absorbed. The release and absorption course of oral dosage forms must comply with mass balance. If the release characteristic *in vitro* was the same as that *in vivo*, Eq. 2 could be obtained:

$$F_a = \frac{1}{f_a}\left(1 - \frac{\alpha}{\alpha - 1}(1 - F_d) + \frac{1}{\alpha - 1}(1 - F_d)^{\alpha}\right) \quad (2)$$

Where f_a is the fraction of the dose absorbed at $t=\infty$ and α ($\alpha=k_p/k_d$) is the ratio of the first-order permeation rate constant (k_p) to the first-order release rate constant (k_d). For $\alpha>>1$, the permeation rate constant is much greater than the release rate constant, such that the rate-limiting step in drug absorption is release. For $\alpha<<1$, the permeation rate constant is much smaller than the release rate constant, such that the rate-limiting step in drug absorption is intestinal permeation. For $\alpha\approx1$, the permeation rate constant and release rate constant are approximately equal, such that the rate-limiting steps in drug absorption are both release and permeation. The nonlinear regression method was also used to analyze the value of α *in vitro* between the modified DDASS release and permeation to assess the absorption characteristics of formulations.

HPLC Analysis of *In Vitro* Samples

The concentrations of omeprazole samples *in vitro* were determined by HPLC. A MS-C_{18} column (5 μm, 4.6 mm×150 mm, Waters, Ireland) with an analytical

TC-C_{18} guard column (5 μm, 4.6 mm×12.5 mm, Agilent, USA) was maintained at 30°C. The analytical mobile phase consisted of water and methanol in a 57:43 (v/v) ratio. The flow rate was 1 mL/min. The injection volume was 10 μL, and omeprazole was detected by absorbance at 302 nm.

HPLC Analysis of *In Vivo* Samples

HPLC was used to determine the *in vivo* samples. A MS-C_{18} column (5 μm, 4.6 mm×150 mm, Waters) with an analytical TC-C_{18} guard column (5 μm, 4.6 mm×12.5 mm, Agilent) was applied. The column temperature was maintained at 30°C. The analytical mobile phase consisted of water, acetonitrile, and methanol in a 55:5:40 (v/v/v) ratios. The flow rate was 1 mL/min. The injection volume was 30 μL, and omeprazole was detected by absorbance at 302 nm.

Data Analysis and Statistics

Pharmacokinetic parameters in the beagle dogs were determined using Phoenix WinNonlin version 6.1 (Pharsight Co., Ltd, USA). The appropriate compartmental model was optimised by adopting Akaike's information criterion (21). Here, t_{max} was the time of maximum observed concentration, C_{max} was the maximum observed concentration at t_{max}, and $t_{1/2}$ was the drug's half-life. The nonlinear relationship curve was fitted using Matlab version 4.0. The difference in correlation coefficients (r) between the USP I (basket) and the modified DDASS for five dogs' data by linear or nonlinear regression was analyzed using a *T*-test in SPSS 11.5. The *p*-value was considered statistically significant when less than 0.05.

RESULTS

Release Kinetic Characteristics of Omeprazole Enteric-Coated Tablets

The release trait of omeprazole enteric-coated tablets was obtained by the USP I system (basket). Omeprazole enteric-coated tablets did not release until 2 h in 0.1 N hydrochloric acid (pH 1.8). Changing the release medium into 0.05 M pH 6.8 phosphate buffer after 2 h produced an almost instantaneous release (n=6) of 63.31%±5.15% at 0.5 h; 76.89%±5.87% at 0.75 h; 85.92%±5.22% at 1 h; 91.25%±8.96% at 1.5 h; 95.90%±10.47% at 2 h; and 99.39%±9.90% at 2.5 h, respectively. Release within 0.75 h was over 75%, which meets USP Reference standards. Therefore, omeprazole enteric-coated tablets legitimately demonstrated a delayed-release trait affected by pH value.

The time course of omeprazole enteric-coated tablet elution into the DC of the modified DDASS is shown in Fig. 3a, and the corresponding cumulative elution profile is given in Fig. 3b. The release of omeprazole enteric-coated tablets was accelerated from approximately 50 min. The omeprazole enteric-coated tablets yielded a cumulative elution of 76.08%±7.78% (n=3) in the modified DDASS; the USP I system (basket) achieved a similar level of 76.89%±5.87%. However, the release course of omeprazole enteric-coated tablets in the modified DDASS demonstrated a step-by-step change characterised by rapid release in the USP I system (basket).

Characteristics of Permeation Kinetics of Omeprazole Enteric-Coated Tablets

Figure 3a and b respectively illustrate the time course of permeation and cumulative permeation profiles of omeprazole enteric-coated tablets in the RC of the modified DDASS. The nonlinear relationship curve was fitted using Matlab version 4.0. Here, f_a=60%, refers to the bioavailability of omeprazole enteric-coated tablets, which was cited from its instruction (http://www.drugs.com/uk/losec-mups-tablets-10mg-20mg-40mg-spc-5082.html). F_d from modified DDASS release and the percentage of permeation (F_p) from modified DDASS permeation were obtained, as shown in Fig. 3b. By fitting, we got values of r_{mean} and α_{mean}, shown in Fig. 4 and Table I. A better correlation coefficient was gained, with $r_{mean}(r_{mean} = 0.9963 \pm 0.003) > r_{19,0.001}(r_{19,0.001} = 0.6652)$ cited from the table of the correlation coefficient thresholds (the sample size n is 21). With α_{mean} (α_{mean}=1.558±0.491) close to 1, the permeation rate constant and the release rate constant were approximately equal, such that the rate-limiting steps in drug absorption were both release and permeation. However, the limit of release rate was dominant, owing to the fact that α_{mean} was slightly greater than 1. When α_{mean} was substituted into Eq. 2, Eq. 3 was generated. The equation could give a quantified relationship between release and permeation behaviors by describing all the variables. This might help us predict the process of drugs *in vivo* better using the modified DDASS.

$$F_p = \frac{1}{f_a}\left(1 - \frac{\alpha_{mean}}{\alpha_{mean}-1}(1-F_d) + \frac{1}{\alpha_{mean}-1}(1-F_d)^{\alpha_{mean}}\right) \quad (3)$$

Here, f_a=0.6, refers to the bioavailability of omeprazole enteric-coated tablets; $\alpha_{mean} = 1.558 \pm 0.491$, stands for the ratio of the first-order permeation rate constant (k_p) to the first-order release rate constant (k_d).

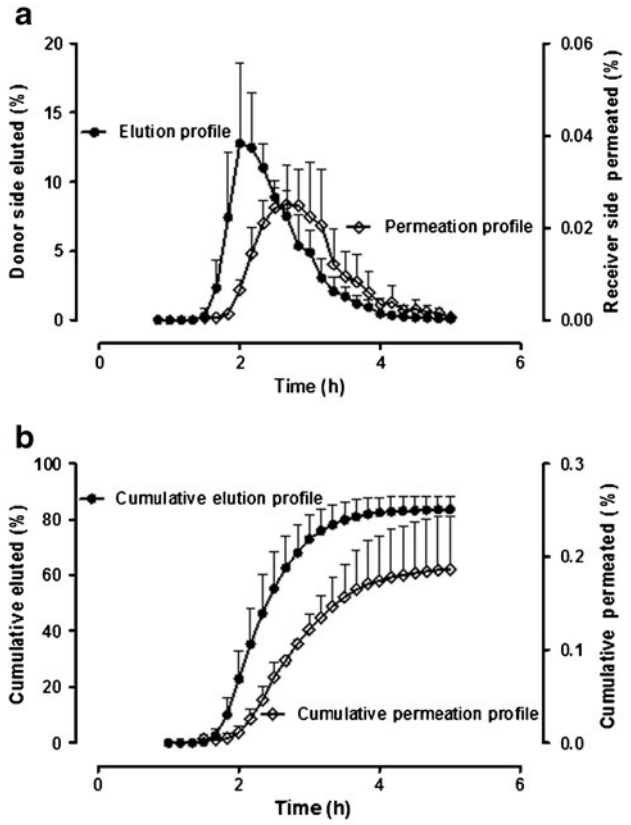

Fig. 3 (**a**) Time course of elution into the donor compartment and permeation to the receiver compartment from omeprazole enteric-coated tablets in the modified DDASS. (**b**) Corresponding cumulative elution profile and cumulative permeation profile. Each point represents the mean ± SD of three experiments.

At the same time, 0.19% of the cumulative permeation from Fig. 3b, more than 0.03% (5), was thought to be absorbed almost completely; Omeprazole is insoluble in water (22,23). This predicted relationship generally agreed with the observed results, indicating that the modified DDASS provides more information for the evaluation of enteric-coated formulations than the USP I system (basket).

Pharmacokinetic Characteristics of Omeprazole Enteric-Coated Tablets in Beagle Dogs

Figure 5 shows the individual/mean plasma concentrations of omeprazole after a single oral administration of omeprazole enteric-coated tablets to beagle dogs. In the individual profiles, there are some deviations about lag time mainly due to individual differences of gastric emptying. The data fit well to a one-compartment open-system pharmacokinetic model by Phoenix WinNonlin version 6.1. The pharmacokinetic parameters are summarized in Table II. The omeprazole enteric-coated tablets showed a delayed-release trait.

IVIVC

Linear Regression

The optimisation of one compartment model was judged by Akaike's information criterion for omeprazole enteric-coated tablets after validation. F_a of the omeprazole enteric-coated tablets in dogs was calculated by the Wagner–Nelson Eq. 1. Here, k_{10} and AUC_{0-inf} were obtained from Table II, C_t and $AUC_{0\text{-}t}$ were obtained from Fig. 5, F_d of the USP I system (basket) was obtained from above section, and F_p of the modified DDASS was obtained from Fig. 3b. The linear regression equations and the correlation coefficients (r) between F_d and F_a, as well as that between F_p and F_a, were summarized (Table III). By a statistical comparison, modified DDASS permeation and dog absorption showed a better level of IVIVC with a statistically significantly higher correlation coefficient, $r_{mean}(r_{mean} = 0.9117 \pm 0.0327) > r_{5,0.01}(r_{5,0.01} = 0.8745)$, compared with the correlation between USP I system (basket) release and dog absorption ($p<0.05$) (Table III).

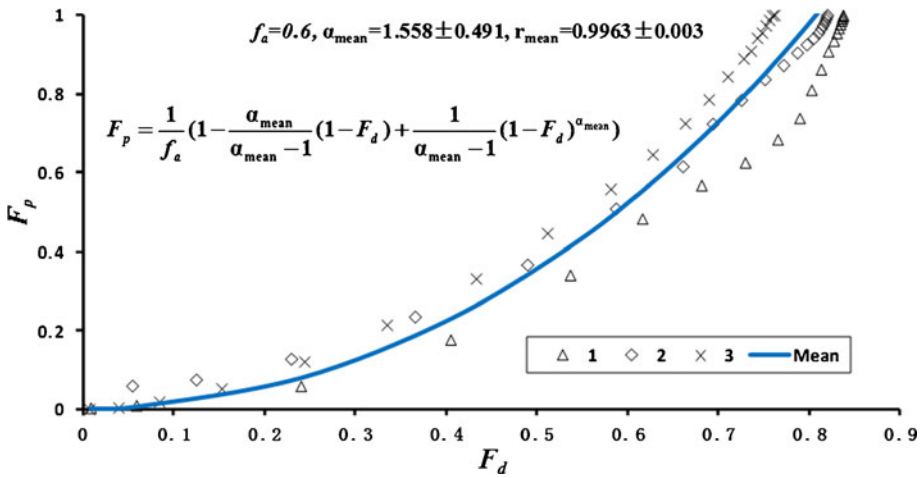

Fig. 4 Profile of nonlinear relationship between percentage of release (F_d) from modified DDASS release and percentage of permeation (F_p) from modified DDASS permeation. 1, 2, 3 represented three parallel trials and Mean represented a curve of statistical trend.

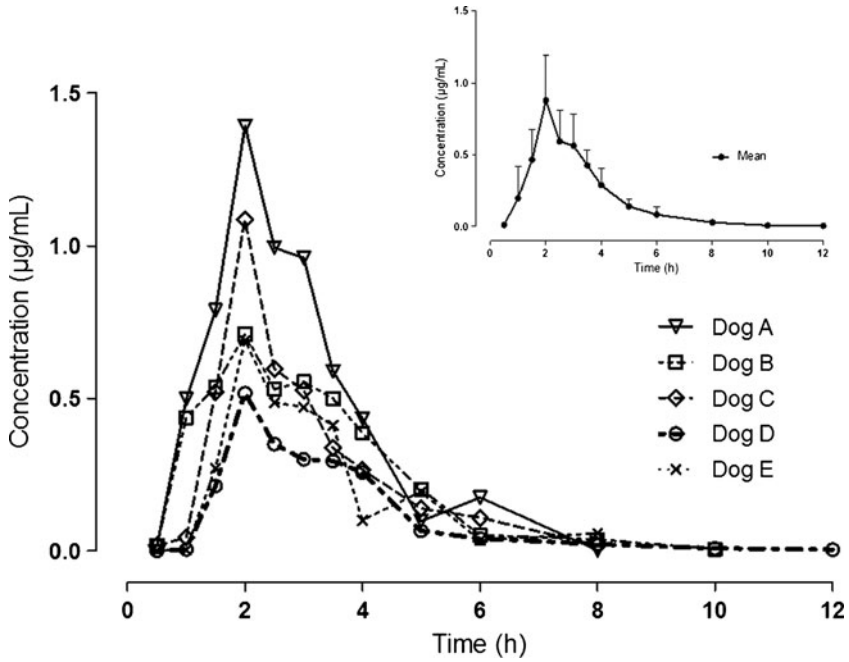

Fig. 5 Profile of individual plasma concentration after oral administration of omeprazole enteric-coated tablets to each beagle dog. The inserted plot shows the profile of mean plasma concentration after oral administration of omeprazole enteric-coated tablets to beagle dogs (Each point represents the mean ± SEM of five experiments).

Nonlinear Regression

With the calculated F_d and F_a from above section, Matlab version 4.0 was used to fit the nonlinear relationship curve by Eq. 2. Here, f_a=60%, refers to the bioavailability of omeprazole enteric-coated tablets, which was cited from its instruction (http://www.drugs.com/uk/losec-mups-tablets-10mg-20mg-40mg-spc-5082.html). By fitting, we got values of r_{mean} and α_{mean}. A significant nonlinear relationship was obtained between F_d from modified DDASS release and F_a from dog absorption, $r_{mean}(r_{mean} = 0.8968 \pm 0.055) > r_{5,\,0.01}(r_{5,\,0.01} = 0.8745)$. With α_{mean} (α_{mean}=2.0638 ± 0.726) close to 1, the permeation rate constant and release rate constant were approximately equal, such that the rate-limiting steps in drug absorption were release and permeation. However, the limit of release rate was dominant, owing to the fact that α_{mean} was slightly greater than 1. The results were consistent with that between F_d and F_p obtained from the modified DDASS, with α_{mean} (α_{mean}= 1.558 ± 0.491) close to 1. This demonstrates that the release and absorption process of omeprazole enteric-coated tablets *in vivo* could be predicted reasonably using the modified DDASS, and that nonlinear regression could give more parameters than linear regression for the modified DDASS. By a statistical comparison, a significantly higher goodness of fit ($p < 0.05$) could be obtained for the omeprazole enteric-coated tablets between the modified DDASS and the dogs (r_{mean}=0.8968 ± 0.055) than between the USP I system (basket) and the dogs (r_{mean}=0.7805 ± 0.057).

DISCUSSION

Modification Study on DDASS

The DDASS is considered a new method developed by He *et al.* (5–8) that may be used to evaluate the release and permeation characteristics of oral solid dosage forms. This new model could be used to continuously and dynamically conduct real-time co-evaluation of the dissolution/permeation of oral solid drug preparations and their relationship. However, with further research, the DDASS was found to exhibit difficulties in evaluating enteric-coated formulations. Enteric-coated formulations remained in the

Table I Parameters of Nonlinear Regression Between Percentage of Release (F_d) from Modified DDASS Release and Percentage of Permeation (F_p) from Modified DDASS Permeation (n=3). In following equations, f_a = 0.6, refer to the bioavailability of omeprazole enteric-coated tablets

Trial	Equation	α	r
1	$F_p = \frac{1}{f_a}\left(1 - \frac{\alpha}{\alpha-1}(1-F_d) + \frac{1}{\alpha-1}(1-F_d)^{\alpha}\right)$	1.132	0.9947
2		1.448	0.9950
3		2.095	0.9992
Means ± SD	$F_p = \frac{1}{f_a}\left(1 - \frac{\alpha_{mean}}{\alpha_{mean}-1}(1-F_d) + \frac{1}{\alpha_{mean}-1}(1-F_d)^{\alpha_{mean}}\right)$	1.558 ± 0.491	0.9963 ± 0.003

Table II Kinetic Parameters of Omeprazole Enteric-coated Tablets *In Vivo* ($n=5$)

	t_{max} (h)	C_{max} (μg/mL)	AUC_{0-inf} (μg/mL*h)	$t_{1/2}$ (h)	k_{10} (1/h)
Dog A	2.00	1.39	3.31	0.68	1.02
Dog B	2.00	0.71	2.31	1.07	0.65
Dog C	2.00	1.09	2.12	1.13	0.61
Dog D	2.00	0.52	1.24	1.86	0.37
Dog E	2.00	0.69	1.62	1.49	0.46
Means ± SD	2.00 ± 0.00	0.88 ± 0.35	2.12 ± 0.79	1.25 ± 0.45	0.62 ± 0.25

DDV, and could not disintegrate to imitate the process of the enteric-coated formulation's transfer from stomach to intestine for release, making it difficult to predict *in vivo*. There was a dead volume in the PAV that not only resulted in a lag time between the drug concentration in the DC and DDV but also reduced the drug's concentration for effective analysis. Determination became harder to perform. To solve these problems, the DDASS was developed and further improved. The two-vessel model was changed to a single-vessel model. The pH value of the drug-dissolving solution was changed from 2.0 to 6.8 at a certain time point. This step could effectively reflect the enteric-coated formulation's transfer in the GI tract. The drug-dissolving solution B at pH 7.8 could also be chosen to assess colon preparation. The PAV was replaced with the PAE. The PAE could reduce the dead volume and allow the drug-dissolving solution and pH adjustment solution to fully mix. To verify the rationality and feasibility of the modified DDASS, omeprazole enteric-coated tablets were chosen as a model dosage form to study *in vitro* release characteristics. Appraisal of the IVIVC between the USP I system/modified DDASS and beagle dogs was carried out and the results of the two *in vitro* models were compared. The results showed that a better goodness of fit could be obtained for the omeprazole enteric-coated tablets between the modified DDASS and dogs than between the USP I system (basket) and dogs, and the goodness was higher than the threshold limit value. This finding demonstrates the rationality of the modified DDASS for enteric-coated formulations, and the feasibility of the modified DDASS as a research method on the *in vitro* release characteristics of enteric-coated formulations. He *et al.* declare four patents (24–27) from the proposed model, of which two have been authorised (24,25). These studies are now under investigation and will be the subject of future reports.

Analysis of IVIVC Using the Linear and Nonlinear Regression Methods

IVIVC can be used in the development of new pharmaceuticals to reduce the number of human studies during formulation development. IVIVC can also be employed to establish release specifications, as well as support and validate the use of the release methods. In the present study, the correlations between conventional method release and beagle dog absorption and between modified DDASS release/permeation characteristics and beagle dog absorption were investigated by linear and nonlinear regression analysis methods, respectively. Linear regression analysis showed a higher correlation between modified DDASS permeation and dog absorption than between USP I system (basket) release and dog absorption. Linear correlation is currently a widely used method. Evaluation of the IVIVC is mainly based on the linear correlation between F_a *in vivo* and F_d *in vitro*. During nonlinear regression analysis, a better IVIVC was obtained between the modified DDASS release and dog

Table III Linear Regression Comparison Between the USP I (Basket) and the Modified DDASS for the Dog Data ($n=5$)

	USP I system (basket) *vs.* Dogs		Modified DDASS *vs.* Dogs	
	Equation	r	Equation	r
Dog A	$F_a=2.0727\ F_d - 1.396$	0.8918	$F_a=0.8463\ F_p + 0.159$	0.9624
Dog B	$F_a=2.1950\ F_d - 1.4581$	0.9091	$F_a=0.7799\ F_p + 0.2584$	0.9256
Dog C	$F_a=2.7602\ F_d - 1.8934$	0.7940	$F_a=0.9268\ F_p + 0.1861$	0.8925
Dog D	$F_a=3.342\ F_d - 2.3216$	0.7827	$F_a=1.2186\ F_p + 0.1884$	0.8963
Dog E	$F_a=2.9245\ F_d - 2.0412$	0.7803	$F_a=1.0731\ F_p + 0.1547$	0.8817
Means ± SD	$F_a=(2.6589 \pm 0.5259)\ F_d - (1.8221 \pm 0.3926)$	0.8316 ± 0.0634	$F_a=(0.9689 \pm 0.1773)\ F_p + (0.1893 \pm 0.0415)$	0.9117 ± 0.0327[a]

[a] Significant difference from correlation coefficients (r) between the USP I (basket) and the Modified DDASS for five dogs' data at $p < 0.05$

absorption than between conventional method release and dog absorption. In addition, the value of α was consistent with that between F_d from modified DDASS release and F_p from the modified DDASS permeation. Nonlinear relationship (28–30) is very scarce in the literature, but it is seen as more appropriate (20). IVIVC should be in degrees that range from "low" to "high" in a gradual, continuous fashion, rather than consisting of only the currently perceived options of "yes, a correlation exists" and "no, a correlation does not exist." The degrees of IVIVC depended on the relative rates of release and intestinal permeation and on the fraction of dose absorbed. This is because drug release and permeability of oral solid dosage forms are the rate-limiting steps *in vivo* absorption, which affect bioavailability. Consequently, nonlinear regression could give more objective results and parameters than linear regression for the modified DDASS.

CONCLUSION

The modified DDASS could not only simulate the physiological pH state of the GI tract but also effectively imitate the *in vivo* process of the formulation's transfer from stomach to intestine for release. It overcomes the drawback of the original DDASS, which cannot be used to evaluate enteric-coated formulations. Application of the modified DDASS could be extended to other types of solid preparations. The modified DDASS can offer more parameters than conventional release devices for screening prescriptions, especially when based on the nonlinear regression analysis. Better linear/nonlinear IVIVC for omeprazole enteric-coated tablets was established between modified DDASS elution and dog absorption than between USP I system (basket) release and dog absorption. Thus, the proposed system can be a new method for predicting the *in vivo* characteristics of enteric-coated formulations. The proposed bionic system model can serve as a new method for designing more-effective enteric-coated formulations and has the potential to decrease research and development costs while improving drug effectiveness.

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